USING STUDENT DIFFICULTIES TO IDENTIFY AND MODEL FACTORS INFLUENCING THE ABILITY TO INTERPRET EXTERNAL REPRESENTATIONS OF IgG-ANTIGEN BINDING

By

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B.Sc. Hons. (Natal)

Submitted in fulfilment of the
academic requirements for the degree of
Doctor of Philosophy
in
Biochemistry
in the
School of Biochemistry, Genetics, Microbiology and Plant Pathology
University of KwaZulu-Natal
Pietermaritzburg

February 2005

For my Father, Mother and Brother

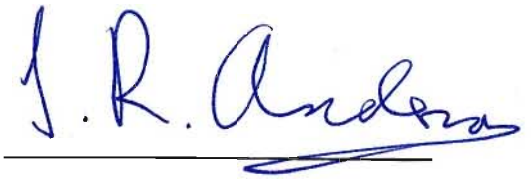
Without your inspiration, guidance, kindness and love
this journey would have been impossible.
I have followed your example.

PREFACE

The research described in this thesis was carried out in the Science Education Research Group (SERG), Discipline of Biochemistry, School of Biochemistry, Genetics, Microbiology and Plant Pathology, University of KwaZulu-Natal, (Pietermaritzburg Campus), from January 2000 to February 2005 under the supervision of Professor Trevor R. Anderson and co-supervision of Professor Diane J. Grayson (University of South Africa, Pretoria).

These studies represent original work by the author and have not otherwise been submitted in any other form for any degree or diploma to any other University. Where use has been made of the work of others, it has been duly acknowledged in the text.

Konrad J. Schönborn (Candidate)



Prof. Trevor R. Anderson (Supervisor)

February 2005

ABSTRACT

Scientific external representations (ERs), such as diagrams, images, pictures, graphs and animations are considered to be powerful teaching and learning tools, because they assist learners in constructing mental models of phenomena, which allows for the comprehension and integration of scientific concepts. Sometimes, however, students experience difficulties with the interpretation of ERs, which has a negative effect on their learning of science, including biochemistry. Unfortunately, many educators are not aware of such student difficulties and make the wrong assumption that what they, as experts, consider to be an educationally sound ER will necessarily promote sound learning and understanding among novices. On the contrary, research has shown that learners who engage in the molecular biosciences can experience considerable problems interpreting, visualising, reasoning and learning with ERs of biochemical structures and processes, which are both abstract and often represented by confusing computer-generated symbols and man-made markings.

The aim of this study was three-fold. Firstly, to identify and classify students' conceptual and reasoning difficulties with a selection of textbook ERs representing IgG structure and function. Secondly, to use these difficulties to identify sources of the difficulties and, therefore, factors influencing students' ability to interpret the ERs. Thirdly, to develop a model of these factors and investigate the practical applications of the model, including guidelines for improving ER design and the teaching and learning with ERs. The study was conducted at the University of KwaZulu-Natal, South Africa and involved a total of 166 second and third-year biochemistry students. The research aims were addressed using a post-positivistic approach consisting of inductive and qualitative research methods. Data was collected from students by means of written probes, audio- and video-taped clinical interviews, and student-generated diagrams.

Analysis of the data revealed three general categories of student difficulties, with the interpretation of three textbook ERs depicting antibody structure and interaction with antigen, termed the process-type (**P**), the structural-type (**S**) and DNA-related (**D**) difficulties. Included in the three general categories of difficulty were seventeen sub-categories that were each classified on the four-level research framework of Grayson *et al.* (2001) according to

how much information we had about the nature of each difficulty and, therefore, whether they required further research. The incidences of the classified difficulties ranged from 3 to 70%, across the student populations and across all three ERs. Based on the evidence of the difficulties, potential sources of the classified difficulties were isolated. Consideration of the nature of the sources of the exposed difficulties indicated that at least three factors play a major role in students' ability to interpret ERs in biochemistry. The three factors are: students' ability to *reason* with an ER and with their own conceptual knowledge (**R**), students' understanding (or lack thereof) of the *concepts* of relevance to the ER (**C**), and the *mode* in which the desired phenomenon is represented by the ER (**M**).

A novel three-phase single interview technique (3P-SIT) was designed to explicitly investigate the nature of the above three factors. Application of 3P-SIT to a range of abstract to realistic ERs of antibody structure and interaction with antigen revealed that the instrument was extremely useful for generating data corresponding to the three factors. In addition, analysis of the 3P-SIT data showed evidence for the influence of one factor on another during students' ER interpretation, leading to the identification of a further four interactive factors, namely the reasoning-mode (**R-M**), reasoning conceptual (**R-C**), conceptual-mode (**C-M**) and conceptual-reasoning-mode (**C-R-M**) factors. The Justi and Gilbert (2002) modelling process was employed to develop a model of the seven identified factors. Empirical data generated using 3P-SIT allowed the formulation and validation of operational definitions for the seven factors and the expression of the model as a Venn diagram.

Consideration of the implications of the model, yielded at least seven practical applications of the model, including its use for: establishing whether sound or unsound interpretation, learning and visualisation of an ER has occurred; identifying the nature and source of any difficulties; determining which of the factors of the model are positively or negatively influencing interpretation; establishing what approaches to ER design and teaching and learning with ERs will optimise the interpretation and learning process; and, generally framing and guiding researchers', educators' and authors' thinking about the nature of students' difficulties with the interpretation of both static and animated ERs in any scientific context. In addition, the study demonstrated how each factor of the expressed model can be used to inform the design of strategies for remediating or preventing students' difficulties with the interpretation of scientific ERs, a target for future research.

ACKNOWLEDGEMENTS

Recognition is due to the following individuals and institutions for making this research possible:

My supervisor and mentor; Prof. Trevor R. Anderson, for your absolute commitment, belief in, and dedication to, this research. Your patience, open-door policy and countless extra hours spent on academic discussion, review and critiquing of manuscripts is gratefully and humbly appreciated. Your quest for perfection and excellence and, your creativity, work ethic and tenacious spirit of endeavour are truly awe-inspiring. It has been a privilege to interact with an academic of your calibre. *Knock back the frontiers.*

My co-supervisor; Prof Diane J. Grayson, for your succinct and constructive inputs to the work and for stimulating me to, "think outside the box". Thank you for allowing me the opportunity to share inspiring interaction with such an acclaimed academic.

The academic staff and student colleagues in the Department of Biochemistry; for instilling an interest in the submicroscopic world. Thanks to Prof. J.P. Dean Goldring for providing lecture time for student data collection and for your encouragement and, to Prof. Theresa H. T. Coetzer for your expert opinion on the nature of representation in immunology.

The academic staff and student colleagues in the Department of Psychology; for instilling a fascination in the behavioural sciences during my undergraduate studies.

My fiancée and confidant; Christina Holmes, your belief in me and your complete unselfishness, unconditional love and compassion have been astonishing. Thank you for putting up with the numerous "all-nighters" and mood swings. My sincerest gratitude for your unwavering support and commitment to this goal, for which I shall be forever indebted.

Fellow (ex-)student and friend; Tracy Hull, for all the encouragement and stimulating academic debate. Your humour and friendship made the sometimes-frustrating months bearable.

Members of the Science Education Research Group (SERG); for the fruitful interaction, academic analysis and for all your encouragement. Keep the faith.

My friends and extended family; who have been by my side throughout the many years of study. Drinks are (finally) on me.

The Staff and Boys of St. Charles' College; for affording me the opportunity to practice the art of science education at your great institution.

The National Research Foundation (NRF) (GUN Number 2053218) and University Research Fund (URF); for funding the work presented in this thesis. Special thanks to the Dean of Science and Agriculture (University of KwaZulu-Natal) for awarding a Special Doctoral Research Grant and, to the American Society of Biochemistry and Molecular Biology (ASBMB) for awarding a travel fellowship.

The following educational sources for permission to reproduce the external representations contained in this thesis: ER A from Bohinski, R.C., 1987; ER C from Silverton *et al.*, 1977 and adapted in Bohinski, R.C., 1987: both reproduced by permission of Pearson Education, Inc., Upper Saddle River, NJ 07458; ER D from Silverton *et al.*, 1977 and adapted in Stryer, L., 1995; ER E from Valentine, R.C. and Green, N.M., 1967 and reproduced in Roitt, I.M., 1997: reproduced by permission of Academic Press Ltd, London, UK; ER F from Amit, A.G. *et al.*, 1986, and reprinted in Roitt, I.M., 1997: reproduced by permissions of American Association for the Advancement of Science and Blackwell Science, Inc., respectively; ER G reproduced with permission of Jackson, J.G, University of KwaZulu-Natal, South Africa; ERs in Fig 3.1 were obtained from the following sources: (a) and (g) www.ntri.tamuk.edu/ immunology/ab-structure.html; (b) *The Antibody Resource Page* www.antibodyresource. com/); (c) www.biology.arizona.edu/immunology/tutorials/antibody/structure.html; (d) rex.nci.nih.gov/ behindthenews/uis/immunesystframe.htm ; (e) and (f) www.path.cam.ac.uk/ ~mrc7/igs/ mikeimages.html ; (h) edcenter.med.cornell.edu/CUMC_PathNotes/ Immunopathology/Immuno_01.html ; (i) Hames *et al.*, 2000; (j) Stryer, 1995. Venn graphics for Fig. 6.2 created at and reproduced with permission of the online *Create-A-Venn system* at www.venndiagram.com)

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LIST OF ABBREVIATIONS[1]

[ ]	Words included between square brackets in the transcript text are inserted by the researcher for the purposes of adding meaning to a student's otherwise uncompleted word or phrase, for adjusting an immediately previous word or phrase for the purposes of scientific or grammatical clarity, or for providing accompanying descriptions of physical and external behaviour
[...]	Ellipse between square brackets to designate an excluded section of transcript text
...	Ellipse used in the transcript text to designate a sudden change in thought, slight pause, or verbal interruption
α	Alpha
β	Beta
3P-SIT	Three-phase single interview technique
A	Refers to "region A" of Fig 5.2 E (electron micrograph)
a	Refers to "frame a" of Fig 5.2 F (space-filling display)
Ab	Antibody
Ab-Ag	Antibody-antigen complex
Abs	Absorbance (ELISA graph)
Ag	Antigen
Ags	Antigens
AI	Artificial intelligence theory
ax.	X or Y axis of a Cartesian plane
B	Refers to "region B" of Fig 5.2 E (electron micrograph)
b	Refers to "frame b" of Fig 5.2 F (space-filling display)
b. site	Antigen binding site on antibody
beg.	Begin(s)
bot.	At the bottom

[1] Included in the list of abbreviations are those used by the author in the interview transcript text to denote and describe tacit physical gestures, drawing behaviours and additional verbal outputs of students during data collection. The abbreviations serve as a nomenclature used by the author as a means with which to present data corresponding to students' observable and explicit behaviours.

C	Constant amino acid region of an IgG antibody
C	Conceptual factor
CDR	Complementarity-determining region(s)
C-M	Conceptual-mode factor
C-R-M	Conceptual-reasoning-mode factor
c	Refers to "frame c" of Fig 5.2 F (space-filling display)
DCT	Dual-coding theory
DNA	Deoxyribonucleic acid
DNP	Dinitrophenyl
ELISA	Enzyme-linked immunosorbent assay
ER(s)	External representation(s)
Fab	Fragment antigen binding
Fc	Fragment crystallisable
Fig.	Figure
Figs	Figures
γ	Gamma
gen.	Generate(s)
Gln	Glutamine
grad.	Gradient
H	Heavy chain
I	Interviewer
Ig	Immunoglobulin
IgG	Immunoglobulin G
inserts	Used to describe the physical insertion, addition or modification of a diagrammatic marking when a student produced a diagram
IP	Information-processing theory
κ	Kappa
L	Light chain
log	Logarithm
lt	On the left or, on left hand side
lyso.	Lysozyme
M	Representation mode factor
mid.	Middle
pers. comm.	Personal communication

R	Reasoning factor
R-C	Reasoning-conceptual factor
R-M	Reasoning-mode factor
RNA	Ribonucleic acid
rt	On the right or, on right hand side
S	Student
SGD(s)	Student-generated diagram(s)
top	At the top
V	Variable amino acid region
wk	Week number (ELISA graph)

1 INTRODUCTION AND AIMS

Research into students' conceptual and reasoning difficulties with the learning of science has been an important focus in science education for several decades (Anderson and McKenzie, 2002). In this regard, extensive studies have revealed that if such difficulties are not addressed they can seriously hinder students' learning and understanding of science (Treagust *et al.*, 1996). A large number of student difficulties have been reported in physics (e.g. Harrison *et al.*, 1999), chemistry (e.g. Huddle and Pillay, 1996; Garnett *et al.*, 1995) and biology (e.g. Odom, 1995; Marek, 1986). In biochemistry, however, only a few such difficulties (e.g. Talbot, 2001) have been identified by formal research (e.g. Hull *et al.*, 2002; Anderson *et al.*, 1999; Anderson and Grayson, 1994; Fisher, 1985).

The teaching and learning of science often involves the use of external representations (ERs) such as diagrams, photographs, images, pictures, graphs and computer-based visuals. Although research in various disciplines has shown that ERs can be valuable learning tools for communicating, clarifying and integrating scientific concepts (e.g. Peña and Quílez, 2001), for the mental representation of text (Schnotz, 1993a), and for the construction of useful mental models of abstract phenomena (Winn, 1991; Hurt, 1987), this is certainly not always the case. Research has also shown that it is wrong to assume that the use of ERs for teaching and learning in science will always lead to improved understanding and desired learning outcomes (Lowe, 2003; Cheng *et al.*, 2001). This is because some ERs have been shown to actually cause numerous alternative conceptions (e.g. Ametller and Pintó, 2002), and incorrect ways of reasoning (Mayer *et al.*, 1995), that are resistant to change and very difficult to correct through conventional teaching methods (Wheeler and Hill, 1990; Hill, 1988).

Besides the sometimes-poor nature of ER design, another source of the above conceptual and reasoning difficulties is the failure of educators, textbook authors, and students alike, to acknowledge that the interpretation of ERs is a highly cognitively demanding task (Lewalter, 2003; Lowe, 1996), which needs to be explicitly learnt and taught (e.g. Petre and Green, 1993). In addition, studies in the field suggest that reasoning and conceptual difficulties associated with ERs stem largely from the graphical language that is used within ERs to

convey scientific ideas (e.g. Pintó and Ametller, 2002). Unlike linguistic and verbal representations (e.g. spoken English or written Spanish), not all sciences contain a standardised graphical language that can be applied exclusively to all ERs used in a particular discipline (e.g. Blackwell, 2001). In contrast to Mathematics and Physics, most other abstract sciences make use of multiple ERs and symbolism to communicate a *single* phenomenon. Such ERs often contain numerous markings, signs and symbols that can be both abstract and idiosyncratic. For this reason, the viewer of the ER has to sometimes contend with ER markings that are beyond their current or past experience (e.g. Henderson, 1999). Thus, it is not surprising that a student's background knowledge will also play a role when reading and interpreting an ER (e.g. Lowe, 1996). Roth (2002) and Cheng *et al.* (2001) refer to the influence of conceptual knowledge on the interpretation of scientific ERs as a "chicken-and-egg" dilemma. One needs to possess at least some related conceptual knowledge in order to understand an ER while at the same time one needs to understand the markings used by the ER in order to acquire that conceptual knowledge. Furthermore, in relation to the former, the literature also suggests that the cognitive mechanisms of visualisation that the viewer employs affect the way ERs are interpreted and if reasoning is erroneous, difficulties can be induced (e.g. van Dusen *et al.*, 1999). However, there appears to be only a few studies that have attempted to understand the cognitive processes associated with the interpretation of scientific ERs and only a few that have a direct application to science education (e.g. Lowe, 2003; Blackwell *et al.*, 2001; Scaife and Rogers, 1996; Zhang and Norman, 1994; Larkin and Simon, 1987). In the present study, this area of learning is further investigated.

Although extensive literature exists on the general use of, and difficulties with, ERs in scientific fields such as astronomy, geography, physics and biology (e.g. Lowe, 2003, 1999; Sanders, 2002; Stylianidou *et al.*, 2002; Peña and Quílez, 2001; Henderson, 1999; Mayer, 1989b; Johsua, 1984), very few reports have been published on the effectiveness of ERs in the field of biochemistry, the focus of the present study. Besides the dearth of knowledge in this area, the decision to explore ERs in the learning and teaching of biochemistry was motivated by the following three points. Firstly, instructors have often naively dismissed student difficulties with the interpretation of scientific ERs as being due to "poor diagrams", or "poor learners", without any confirmation by research. In this regard, very little *empirical* research has been undertaken on student interpretation of ERs used in biochemistry, nor on the role of ERs in the teaching and learning of this subject. Nevertheless, some examples of the work that has been undertaken thus far are as follows. Recently, Hull (2003) investigated students'

use of ERs in the visualisation of biochemical processes. In addition, Seufert (2003) has studied students' learning from multiple representations that showed the biochemical relevance of iron and vitamin C in human metabolism. Their results illustrated that multiple representations can serve many learning functions provided they are not "overloaded" with information, in which case learning is greatly reduced (e.g. Mayer, 2003). Furthermore, Nerdel *et al.* (2003) have explored students' understanding of animated ERs that show dynamic biological processes associated with cell membranes. Stewart (1981) has commented on the role of ERs in biochemistry texts, while Nuñez de Castro and Alonso (1997) have explored how the presentation of energy ERs for enzyme-catalysed reactions in textbooks may cause confusion in that they are often very simplified depictions that exclude essential chemical steps. Moreover, Menger *et al.* (1998) have shown that the portrayal of micelles in texts is not always accurate and students receive a distorted view of reality when a micelle is presented as "spokes of a wheel". Finally, Crossley *et al.* (1996) have exposed certain reasoning difficulties with ERs depicting the electron transport chain in the mitochondrion. The authors indicated that difficulties with the concept of uncoupling and coupling in oxidative phosphorylation might be attributed to the depiction of the mechanism in textbooks. For instance, some ERs show no apparent link between the oxidation of $FADH_2$ and NADH molecules and the simultaneous phosphorylation of ADP molecules. Paralleling work in the biochemistry domain, but in a more chemistry-weighted context (which often applies to biochemistry) Lewis (1980) has discussed how the use of potential energy diagrams can act as conceptual tools in the study of electron transfer reactions. In addition, Treptow (1980) has examined methods for graphically illustrating Le Chatelier's principle and Borrell and Dixon (1984) have considered the use of electrode potential diagrams as a way to represent biochemical electron-transfer reactions in photosynthesis.

The second reason for exploring ERs in the learning and teaching of biochemistry was motivated by urgent calls (e.g. Flores *et al.*, 2003; Kindfield, 1993/1994[2]) for more ER research into students' learning of biologically applied subjects, a poorly understood and largely uncharted domain of ER research. Recently, disciplines in the molecular and cellular biosciences have experienced an onslaught of visual media ranging from modern text-base mediums, that are accompanied by their CD-ROM counterparts, to electronic textbooks that are available as Internet and software resources (e.g. Richardson and Richardson, 2002). In

[2] Reference correctly cited

this domain, there has truly been an ER "explosion" and with it, have come numerous potential learning and visualisation difficulties for students. Modern biochemical education makes extensive use of colourful, attractive and aesthetically pleasing ERs that are considered by experts to be very useful as teaching and learning vehicles. Typical examples are ERs such as electron micrographs, space-filling representations, "ball-and-stick" representations, computer-based displays, Cartesian graphs and other schematic visuals. However, can we always be sure that these ERs enhance the construction of knowledge and deliver the expected learning outcomes? For instance, extensive research in science education has proven that there are large differences between the manner in which experts and novices learn from and use ERs (e.g. Kozma, 2003; Lowe, 1996). In this regard experts, in addition to having greater conceptual knowledge and experience, make use of far more superior cognitive mechanisms to organise and integrate ER information than do students (e.g. Koedinger and Anderson, 1990; Egan and Schwartz, 1979). Therefore, it is wrong for biochemistry instructors to simply assume that an ER, which proves useful for them, will necessarily be useful for a student.

The third reason for studying ERs used for teaching and learning biochemistry was that much ER research has focused on ERs that are highly abstract in nature. In a biochemical context, almost all the thinking related to biochemical phenomena takes place at the *submicroscopic* level (e.g. Hoffman and Laszlo, 1991). Hence, in order to understand its discourse, biochemists have to rely heavily on ERs in an attempt to capture "reality" and thus, define their science. Therefore, learners who engage in the molecular biosciences are required to visualise biochemical structures and processes, which are both abstract and often represented by unfamiliar computer-generated symbols and man-made markings. It goes without saying that learners have to contend with these ERs, and the associated graphical symbolism, during the formulation of biochemical concepts, and are therefore, required to have at least some degree of visual literacy (e.g. Roth, 2002). However, in the science of biochemistry, acquiring these skills is challenging since ERs used to convey a particular biochemical concept rarely exist in isolation and hence, the *way* in which students interpret them is of crucial pedagogical importance (e.g. Cheng *et al.*, 2001).

Thus in lieu of the above rationale, the author considers it vitally important to perform a thorough investigation of student difficulties with the interpretation of ERs particularly, but not exclusively, in the area of biochemistry. In addition, it is also important to trace the

possible sources of student difficulties with ER-processing and to use this knowledge to suggest guidelines for the improvement of ER design and for teaching, learning and researching with ERs. Such guidelines could be used to facilitate the formulation of strategies for remediating (correcting) the difficulties. This will enable educational practitioners and students to make the most effective use of ERs as visualisation tools for the teaching and learning of science, including in the largely unexplored area of biochemistry education. In so doing it is hoped that overall, such research will make an important contribution towards the largely unexplored area of biochemistry education.

The research presented in this thesis, therefore, aims to contribute to improving the use of ERs in the learning and teaching of biochemistry as well as in science in general. Towards achieving this overall goal, the present project addresses the following research questions:

1. What types of difficulties do students have with ERs used in the teaching and learning of biochemistry?

2. What are the sources of such difficulties and, therefore, what are the factors affecting students' ability to interpret ERs?

3. How might we obtain empirical data to further investigate the nature of the factors affecting students' ability to interpret ERs?

4. Can the factors be incorporated into an appropriate model?

5. How might we obtain empirical data to confirm the validity of the model?

6. What practical applications will the model have and will it be generalisable to all ERs in biochemistry and science?

7. What guidelines can be suggested for teaching and learning with ERs?

8. What guidelines can be suggested for improving ER design?

The above research questions are addressed in seven chapters constituting this thesis. Chapter 2 constitutes a major review of the multidisciplinary studies done on the use of ERs for the teaching and learning of science. Chapter 3 presents an overview of the general methodological approaches used in this study. Details of the specific methods pertaining to each study are presented in the respective results chapters 4, 5 and 6.

To assist the reader in navigating through this thesis, an overview of the research approach used to address the above research questions is presented in Fig. 1.1 below. As shown in the diagram, chapter 4 reports on the use of the research framework of Grayson *et al.* (2001) to identify and classify student difficulties with the interpretation of three selected ERs of IgG structure and function. Discussion is also given to potential sources of the identified difficulties with the interpretation of the three ERs, which inform the proposal of factors that affect student's ability to interpret ERs in biochemistry. Chapter 5 describes the development of a novel clinical interviewing technique, which we named 3P-SIT (Fig. 1.1), and which was specifically designed to generate empirical data corresponding to the above factors. Chapter 6 then describes the use of the modelling framework of Justi and Gilbert (2002) together with the 3P-SIT data-gathering instrument, to design, develop and express a model of factors (Fig. 1.1) that influence a student's ability to interpret, visualise and learn from ERs in biochemistry. Chapter 6 also discusses how the data generated from 3P-SIT can be used to empirically test the expressed model (Fig. 1.1) in order to assess its validity so that it can be defined operationally (Fig. 1.1). The latter includes investigating the nature of the relationship between the factors constituting the model so that feedback into the design of the model (Fig. 1.1) can be obtained. Chapter 7 serves as a general discussion of the work presented in this thesis and considers the implications of the findings for improving learning and teaching with ERs and for ER design. This chapter also considers further avenues for this type of research in science education.

In summary, Chapter 2 presents a review of the literature pertinent to the current study while Chapter 3 discusses the methodology employed to answer the above research questions. Following this, Chapter 4 addresses research questions 1 and 2, Chapter 5 addresses research question 3 and Chapter 6 addresses research questions 4 and 5. Finally, Chapter 7 brings the findings of the thesis together by addressing research questions 6, 7 and 8.

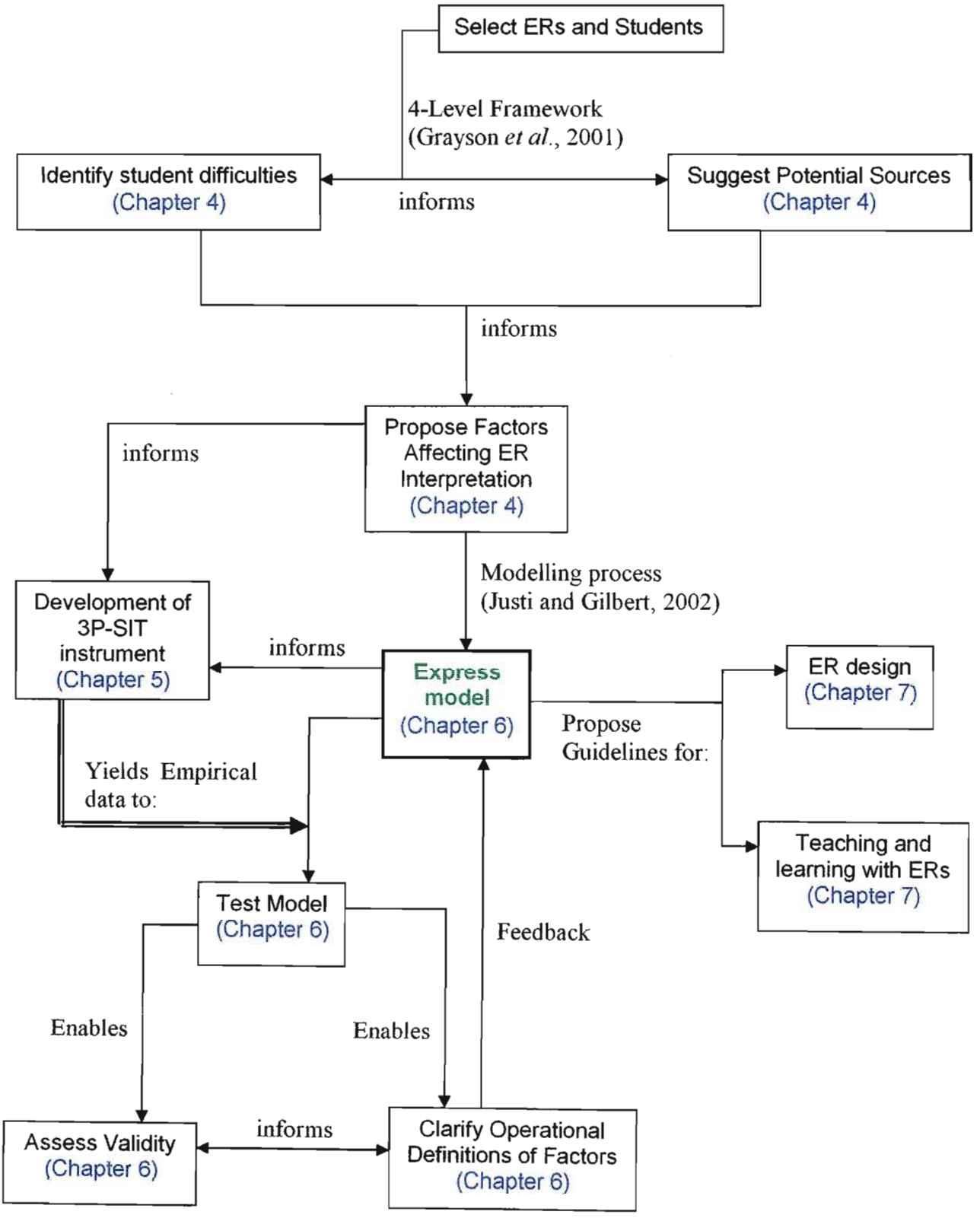


Figure 1.1 Overview of the research approach used to address the research questions

2 LITERATURE REVIEW

2.1 Introduction

Research on external representations (ERs) in science has been an ongoing effort for about the last sixty years. The field was waning until Larkin and Simon published their influential paper in 1987 entitled, "Why a diagram is (sometimes) worth ten thousand words". Their findings fuelled renewed interest in the field characterised by a large volume of recent research. What has been of interest to the observer is the production of research influenced by a range of fields that include philosophy, educational psychology, cognitive psychology, science education and cognitive science. There have also been recent theoretical and practical implications for computer science, artificial intelligence and human-computer interaction.

This chapter provides an account of popular research on how individuals learn from scientific representations that are *external* to the mind such as diagrams, pictures, animations and multimedia. External representations exist in the external world (e.g. on a page or computer screen) and can be discriminated from internal representations, which exist in the mind as "mental models", "mental images" or "mental representations". In this regard and according to the ideas of constructivism (von Glasersfeld, 1989), humans construct knowledge of the world by constructing internal models of the world (Johnson-Laird, 1983). Through the processes of meaningful (Mayer *et al.*, 1995) and generative learning (Osborne and Wittrock, 1983), individuals learn from *external representations* through an active process in which they make sense of the external information *themselves* by constructing internal mental representations (e.g. Mayer, 2003; Kosslyn, 1987, 1985; see section 3.3.2 later). This process is in contrast (e.g. Anderson *et al.*, 2000; Grayson, 1995) with traditional views that see learners passively internalising external information directly. For learners to understand and make sense of the information presented in any external visual display, the information has to be internally processed through already existing conceptions (Stylianidou *et al.*, 2002, Ward and Wandersee, 2002).

In keeping with the spirit of previous writers of similar reviews (e.g. Winn, 1991; Alesandrini, 1984; Levie and Lentz, 1982; Gropper, 1963), this review considers the multidisciplinary

nature of the field. The overall objective is to distil key findings from the research by drawing commonalities and differences from research outputs. This was not an easy endeavour, as research contributions abound from several areas of science and the literature is expansive. Without attempting to examine all the work ever published, an overview of the research believed to have the most application for learning and teaching with external representations in science is presented.

2.2 The nature of external representations in science

ERs consist of physical and written symbols that portray phenomena in the external world (e.g. Lohse *et al.*, 1991; Paivio, 1986). ERs contain spatial relationships and can be distinguished from internal representations, which are an archetype of the mind (Zhang and Norman, 1994). Commentary on the *nature* of ERs focuses on *differences* between their forms, with a quest to understand underlying cognition (e.g. Blackwell, 2001; Stenning and Oberlander, 1995; Glenberg and McDaniel, 1992). ERs occur in different representational *modes*, making one representation of a particular phenomenon different from another. One clear distinction is between a *linguistic* or *propositional* mode and a *graphical* or *pictorial* mode. *Pictorial* representations are associated with pictures, images and diagrams, while *linguistic* representations are sets of sentences (e.g. Shimojima, 2001). Linguistic representations are single "sequences" which correspond to natural language, while diagrammatic representations on the other hand, are indexed by "location in a plane" (Larkin and Simon, 1987, p. 65). Shimojima (2001) has referred to a useful continuum with which to understand differences between representational modes. A diagrammatic or pictorial ER *resembles* what it represents. That is to say, it is *isomorphic* with the target (the entity or idea that is represented) (see Stenning and Lemon, 2001). It is implied therefore, that when isomorphism increases, the ER becomes more pictorial, while a decrease in isomorphism causes an ER to become more propositional.

It follows, that pictorial ERs are not linguistic in the way that speech and written text are (e.g. Blackwell, 2001). Pictorial representations use spatial properties such as location, topology and geometry to convey information (e.g. Cheng *et al.*, 2001), which is explicit and present at one, two-dimensional location (Larkin and Simon, 1987). At the same time as verbal representations are sequential, the "graphical language" contained within a diagram is

simultaneous because all symbols that are conveyed, as well as their *relationships,* can be considered at the same time (e.g. Laseau, 1980).

Science education resources are abundant with external images that include amongst others; photographs, micrographs, pictures, diagrams, illustrations, drawings, models, analogies, maps, plans, graphs, icons, static visuals, dynamic visuals, animated visuals, multimedia and virtual reality environments. In this thesis, "external representation" or "ER" is used to refer to *any*, or *combinations* of these images. Even though use of the term "ER" refers to visual displays that contain graphical, diagrammatic or pictorial elements *rather* than textual components, in some instances, both graphical and textual elements will together be referred to as an "ER". This is in cases where captions (figure legends), textual adjuncts (additions) like labels and/or numerical symbols accompany the pictorial element of an ER. A desire to use the term ER as a label to include *all* graphical displays used in science education is born out by the following. Firstly, a variety of visual displays are referred to. Given the terminological diversity, and the objective of this review, it is plausible to encompass all pictorial forms used in science education under one banner. Secondly, even though some ERs that shall be discussed contain a minimal proportion of textual or numerical symbols, Fry (1981) has stated that even in these cases, "basic transmittal of information is nonverbal" (p. 388).

2.3 Visual literacy and science education

According to Lowe (1988b) and Seels (1994), to be "literate" in written language means that one is able to *read* and *write* language. Like verbal literacy, numerical literacy involves the reading and writing of numbers (e.g. Boardman, 1976) and, both literacies are governed by formal rules for their *reading* and *writing*. As recent writers have expressed the educational need for learners' to *read* as well as *write* ERs (e.g. Lowe, 2000), the idea of "literacy" has been extended to include *visual literacy* (e.g. Ametller and Pintó, 2002; Szabo *et al.*, 1981). Calls for a *visual literacy* can be attributed to the fact that today's world is very much a visual one (e.g. Roth, 2002; Pyle, 1999; Lowe, 1996, 1991; Hardin, 1993; Bennett and Flach, 1992). Now, more than ever before humans are interacting with an array of ERs (e.g. Scaife and Rogers, 1996; Cox and Brna, 1995). As a result, science education is also being exposed to an ever-increasing collection of ERs (Gillespie, 1993). If learners are to process visual

information in the sciences efficiently, Roth (2002) and Lowe (2000) both suggest that visual literacy should be considered a vital component of learning and teaching.

Multiple authors have attempted to formalise a definition for *visual literacy*. For example, Braden and Hortin (1982) describe it as the ability to understand and use both external and internal (mental) images, including the ability to think, learn and express oneself in terms of images (p. 41), while Szabo *et al.* (1981) have ascribed visual literacy to the use of visual materials to improve learning. In addition to definitions pertaining to visual literacy per se, Tierney *et al.* (1990) define the ability to read and interpret ERs as *graphical literacy*, while Boardman (1976) defines *graphicacy* as the skills a learner needs in order to develop conceptions of space. In whatever manner we define it, encompassed within visual literacy is the idea of *visual thinking*, viewed by Seels (1994) as the ability to *visualise* through images. When knowledge is constructed from the interpretation of visual information in science learning (Seels, 1994), visual thinking parallels *visual learning*. If people are to make sense of visual information in science therefore, they are required to be *visually literate* or, at least familiar with the *visual language* used to portray the information. An inability to communicate within this language may result in erroneous ideas being passed to ER readers (e.g. Guri-Rozenblit, 1988). Lowe (1987) and Fry (1981) have pointed out that visual literacy should also include the ability to construct ERs (e.g. draw diagrams). In this regard, it is advocated that learners' abilities within the visual medium be seen as a significant component of science curricula (e.g. Lowe, 1987; Reid *et al.*, 1983; Reid and Miller, 1980; Boardman, 1976).

Commentary has cautioned that there is a negative effect on student performance when they do not possess visual literacy skills (e.g. Pintó and Ametller, 2002). The general consensus is that when visual literacy is neglected, students show learning difficulties, especially in cases where interpretation of ERs of abstract scientific concepts is required (e.g. Hill, 1990). This is an important point because modern science has seen a dramatic increase in the use of abstract ERs particularly as we gain more understanding of the sub-microscopic world (e.g. atomic structure) as well as the macroscopic world (e.g. size of the universe). However, viewers may have problems with scientific ERs that are highly abstract in nature, because they may be unsure of what particular areas need to be processed on the ER and, may lack the procedural knowledge necessary for interpretation (e.g. Lowe, 1997). It seems therefore, that visual literacy has become more important than ever for science education.

2.4 Cognitive mechanisms responsible for ER interpretation

When a viewer *reads* an ER, external information is perceived, leading to the construction of internal information, existing in the mind (e.g. Zhang and Norman, 1994). The internal information is said to take the form of a "mental representation" or "mental image" (e.g. Fleming, 1977). Ultimately, the *way* this image is constructed determines how the ER will be *understood* (Lowe, 1993a). During this visual processing, *cognition* between interactions of the eye and cerebral cortex of the brain organises information in the mind (e.g. Ward and Wandersee, 2002; van Dusen *et al.*, 1999). McCormick *et al.* (1987) and Lohse *et al.* (1991) define this *visualisation* process as the cognitive mechanisms by which humans perceive, interpret, use and communicate visual information. Visualisation allows for mental representations to be formulated, often referred to as visual or mental *imagery* (e.g. Baker and Hill, 1983; Anderson, 1978). According to Denis (1989), *imagery* has great potential for learning because mental images can be *transformed*, effectively mimicking transformations in the real world (e.g. Gordin *et al.*, 1994; McIntyre and Reed, 1976). In order to provide an account of individuals' mental representation and processing of scientific ERs, three popular theoretical foundations are outlined.

Firstly, *visual information processing* theory (e.g. Kosslyn, 1989, 1987, 1985) suggests that visual processing is controlled by three components: *perception*, *short-term memory* and *long-term memory* (e.g. Spoehr and Lehmkuhle, 1982). Perception is a process related to the sensory modality of vision and is responsible for the organisation of patterns, colours and shapes (Kosslyn, 1989, 1985). Perceptual information "fades away" easily unless attention is paid long enough for it to be temporarily stored in short-term (e.g. Mayer and Anderson, 1992), or working memory (e.g. Baddeley, 1992). Even though working memory has a *limited* capacity, as only about seven items of information can be stored at any one time (Mayer, 2003; Kosslyn, 1989), it is where cognitive operations such as learning and reasoning occur. In addition to perception, information can also be inputted to working memory from long-term memory (Kosslyn, 1985). Long-term memory is what gives meaning to visual stimuli that are perceived and contains information that has been encoded from short-term memory (e.g. Kosslyn, 1989; Mayer, 1989a). Long-term memory is where previous experiences, propositions, schemata, models and knowledge are stored (e.g. Taconis *et al.*, 2001; Gillespie, 1993; Johnson-Laird, 1983). When interpreting ERs for instance, long-term

memory stores the information related to the *meaning* of the graphical conventions as well as the procedural and science conceptual knowledge required for interpretation (e.g. Kosslyn, 1985).

Secondly, Paivio's (1986, 1971) *dual-coding theory* (DCT) suggests that two functionally distinct processes code external information. A verbal system processes textual and verbal information, leading to the construction of *verbal* mental representations while a visual mode processes pictorial information such as colour, size and pattern, leading to the construction of pictorial or *image-based* mental representations (e.g. van Dusen *et al.*, 1999; Mayer *et al.*, 1995). A *dual* processing occurs when the *simultaneous* and one-to-one *mapping* between internal representations builds *referential connections* (Clark and Paivio, 1991), resulting in the formulation of a *mental model* (e.g. Mayer *et al.*, 1995; Winn *et al.*, 1991). Mental models can be stored in long-term memory for future use, or retrieved by working memory during problem solving (e.g. Mayer, 2003). Based on DCT, Mayer's (2003) *theory for multimedia learning* suggests that learning is improved when referential connections between verbal and pictorial representations and prior knowledge are *promoted* (e.g. Lewalter, 2003; Mayer and Sims, 1994). Mayer's (2003) theory proposes a cognitive framework with which to explain how individuals learn from multimedia (ERs that present verbal *together* with pictorial information simultaneously). According to this theory (Mayer, 2003; Mayer and Sims, 1994) each processing system (verbal and visual) has a limited processing capacity. For example, information is not captured into working memory unless *attention* is paid to it. Also, because working memory can only hold approximately seven items of information at any one time, learning occurs only when referential connections are made during an active and integrated process (Mayer, 2003; Mayer and Sims, 1994).

Thirdly, *information-processing* (IP) theorists describe human cognition in terms of algorithmic procedures responsible for processing external information (e.g. Cheng *et al.*, 2001; Larkin and Simon, 1987). Information can be processed in the form of *expressions*, *assemblies* and *symbols*. With respect to ERs, *symbols* equate to the external markings of the ER such as shape, colour and size (Cheng *et al.*, 2001). In addition, an ER is an *expression* in that it contains an *assembly* of *symbols* that represent a certain target (e.g. object, entity or idea) (e.g. Cheng *et al.*, 2001; Stenning and Lemon, 2001). During ER interpretation, humans engage in an algorithmic search for expressions or "states of knowledge" in order to achieve a "goal", defined by the original task. The strength of IP theory is that it allows certain

predictions to be made, such as predicting that processing static ERs is computationally different to processing animated ERs (e.g. Lewalter, 2003). With regard to IP theory, there have been recent implications for *artificial intelligence* (AI), where theorists mimic human processing on computers (artificial agents) so as to replicate or automate human reasoning (e.g. Olivier, 2001; Bowen, 1994). Ultimately, computational frameworks applied to artificial agents can be contrasted with human processing (natural agents) (e.g. Olivier, 2001).

One agreement between the above three theoretical accounts is that the human visual system is able to perceive and process visual information enormously quickly (e.g. Bennett and Flach, 1992). An advantage of this for reading ERs is that *meaningful* mental image formation is assisted because concepts are made overt, due to the spatial nature of ERs (e.g. Winn, 1987). This allows prior knowledge to be rapidly activated when necessary (e.g. Lowe, 1999; Gillespie, 1993). In terms of learning from ERs, a feature of the above accounts is that viewers can obtain information through two sources: from the ER as well as through already existing mental (internal) representations (Lowe, 1999, 1994). These mental representations are often referred to as *mental models* (e.g. Gobert and Clement, 1999; Cox and Brna, 1995; Denis, 1989).

A mental model is a multifaceted and complex entity. Johnson-Laird (1983), the most respected proponent of mental models, has described them as being responsible for, "the higher processes of cognition..." (p. 446), and that they, "play a central and unifying role in representing objects... the way the world is... they enable individuals to make inferences and predications, to understand phenomena" (p. 397). In agreement with a constructivist paradigm (section 2.1), Johnson-Laird (1983) says that mental models are constructed from internal representations that exist as symbolic notations in the mind and that they, "contain tokens that correspond to entities in the world..." (p. 422).

In addition to Johnson-Laird (1983), Schnotz (1993b) describes mental models as, "internal quasi-objects, which represent the respective subject matter by analogy on the basis of common structural properties," (p. 248). Kindfield (1993/1994) further suggests that, when learners formulate mental models that correlate favourably with accepted scientific models, they are, in effect, in the process of constructing conceptual understanding. Essentially, "mental models are situational representations that an individual constructs as the need arises" and, "provide a basis for thinking about the represented situation" (Lowe, 1999, p. 226).

Importantly, mental models should not be thought of as static and rigid entities. Instead, Hegarty (1992) and Mayer and Gallini (1990) suggest that when humans make predictions or inferences, mental models possess dynamic components in that they can be *run* in order to complete a task and therefore, can be up-dated and modified.

In the literature, mental models have been discussed with great reference to the interpretation of scientific ERs. Since mental models are thought to possess "spatial" properties (e.g. Winn *et al.*, 1991; Kosslyn, 1981), they are more powerful when encoded from external information that is well organised (e.g. Ward and Wandersee, 2002). Well-structured ERs help students build meaningful mental models that can be managed effectively within working memory (e.g. Glenberg and Langston, 1992; Mayer, 1989b). The role of mental models in the interpretation of scientific ERs will remain an important pedagogic component of this thesis.

2.5 The nature of reasoning with ERs in science

When humans use ERs such as diagrams to make inferences, they engage in *diagrammatic* (e.g. Anderson and Armen, 1998) or *ER reasoning* (e.g. Cox and Brna, 1995). Formal accounts of ER reasoning in science can be found in contexts such as solving geometry proofs (e.g. Mousavi *et al.*, 1995; Koedinger and Anderson, 1990), interpreting pulley systems in physics (e.g. Hegarty, 1992; Larkin and Simon, 1987), and interpreting the kinship represented by family trees (e.g. Olivier, 2001; Winn *et al.*, 1991). Reasoning with ERs in science is complicated and relies on the use of mental models as well as on the ER itself. Modern opinion (e.g. Glasgow, pers. comm.) suggests that when reasoning with ERs in science, the role of *both* internal representations (in the mind) as well as external representations in the world (e.g. on the page or screen) must be taken into account. The relationship between external and internal representations during ER reasoning processes is discussed below.

Cox and Brna (1995) and Zhang and Norman (1994) have suggested that explanations for ER-reasoning have traditionally focused on the functions of *internal representations* alone, without considering the cognitive role of the external representation itself. It is argued that the *interplay* between *both* internal representations and the external representation should be seen as *one system* (Scaife and Rogers, 1995). Literature has begun to consider this internal-

external relationship seriously (e.g. Brna *et al.*, 2001), an approach that Scaife and Rogers (1995) term *external cognition.* The study of external cognition aims to define, " properties of the internal *and* external structures" (p. 188), and refers to, "the *totality* of the relationship between external representation, internal representation and their interaction" (p. 189).

The *representational system* has two efficacies associated with it (Stenning and Lemon, 2001; Cox and Brna, 1995). *Computational* efficacy is concerned with how individuals draw inferences from the representational system and *expressive* efficacy is concerned with the semantic properties of the ER (the "meaning" contained within the ER). Cox and Brna (1995) also suggest that selection of the most appropriate ER with which to reason goes a long way to defining how effectively a problem will be solved (e.g. Bodner and Domin, 2000). Effective *ER reasoners* are able to transfer their skills from one ER context to another, as is the case with individuals who employ *multiple representations* of a situation (e.g. Brna *et al.*, 2001; van Someren et al., 1998; Kozma and Russell, 1997; Dufresne *et al.*, 1997; Moore and Skinner, 1985; Hayes and Readence, 1983).

With reference to the representational system, Scaife and Rogers (1995) have outlined three characteristics of external cognition. Firstly, *computational offloading* refers to how a particular ER can *decrease* the amount of cognitive effort required to read the information (e.g. Cheng *et al.*, 2001). Secondly, *re-representation* is concerned with how different representational modes of the same idea (i.e. multiple representations) make processing easier or more difficult (e.g. Cheng *et al.*, 2001; Zhang and Norman, 1994). Thirdly, *graphical constraining* is concerned with how ER markings limit the range of interpretations that can be generated from the ER (e.g. Cheng *et al.*, 2001; Stenning and Oberlander, 1995).

External cognition principles can be extended to include Zhang and Norman's (1994) theory of *distributed cognition.* In a similar stance, ER processing is considered to be, "distributed across the internal mind and the external environment" (p. 87) and that the, "representational system of a distributed task can be considered as a set, with some members internal and some external" (p. 89). One component of this theoretical framework is the idea of a *representational effect,* which suggests that different representational modes that represent the *same* idea (e.g. multiple ERs) can induce different interpretations. Furthermore, they argue that a "representation" should be defined as an *abstract* entity made up of internal *and* external representations that function *together.* The theoretical tenets described above are a

foundation from which to interpret the following research findings on the use of ERs in science education.

2.6 Use of different types of ERs for learning and teaching in science

Researchers such as Schnotz and Lowe (2003), Peña and Quílez (2001), Henderson (1999), Mayer *et al.* (1995), Lowe (1994a, 1989, 1986) and Hurt (1987) have stressed that the prevalence of ERs in science instruction does not always lead to a favourable understanding of concepts. Not much attention is paid to the information-carrying properties of ERs, or to what ERs actually *do* for viewers (e.g. Moore *et al.*, 1993; Duchastel and Waller, 1979). Consequently, ERs often seem to serve little instructional purpose, and are sometimes included for aesthetic purposes alone (e.g. Mayer *et al.*, 1995; Schnotz, 1993a; Kindfield, 1993/1994, 1992; Lowe, 1991; Holliday, 1990, 1973).

Given the observations above, various researchers argue that studying the role of ERs in science education is of extreme pedagogical importance (e.g. Roth, 2002; Mayer, 1997). This is particularly suggested because so many educators make claims that all ERs will automatically benefit learners, claims that are naïve, and often based on intuition alone, rather than on any theoretical grounds (e.g. Cheng *et al.*, 2001; Guri-Rozenblit, 1988). For example, one claim is that the role of ERs in expository (explanative) text is "transparent" to the reader (Lowe, 1994b, 1991) in that ERs are seen as self-explanatory tools that *always* aid understanding, due to their mere presence within textbook pages or on the screen (e.g. Gobert and Clement, 1999; Bernard, 1990). Furthermore, Goldman (2003) and Lowe and Schnotz (2003) have indicated that together with recent technological developments, other general assumptions about the usefulness of ERs have emerged. For instance, Scaife and Rogers (1996) have discussed the following unwarranted claims: 3-D representations are better than 2-D representations, solid modelling is better than wire-frame modelling in chemistry, coloured ERs are better than black and white ERs and animated ERs are more effective than static ERs. Given these sweeping assumptions, in contrast with investigations concerned with the interpretation of text, little is known about students' use of ERs as learning aids (e.g. Mayer, 1997; Winn, 1993) and thus such issues require urgent investigation.

It is also important to research how humans process scientific ERs, as ER-processing is a cognitively demanding exercise that is not as easy as often thought (e.g. Henderson, 1999; Lowe, 1989; Weidenmann, 1988). If interpreted erroneously, ERs have the potential to induce conceptual and reasoning difficulties (e.g. Ametller and Pintó, 2002; Stylianidou *et al.*, 2002; Cheng *et al.*, 2001; Sumfleth and Telgenbüscher, 2001; Wheeler and Hill, 1990). Indeed, Treagust *et al.* (2002) have suggested that the potential of learners to generate difficulties makes sense, especially when one considers that ER information has to be processed through each individual's unique understanding. As a result, students may struggle to *filter* the relevant information presented in an ER and to effectively link it to their current knowledge (e.g. Wandersee, 1994). Therefore, not *all* ERs are effective for learning (e.g. Mayer and Gallini, 1990; Hill, 1990) and research needs to be done to establish the extent of learning and the nature of difficulties.

In this review, prominent studies on students' interpretation of ERs in science will be discussed in seven parts, each part corresponding to a different *type* of ER used in science education. Studies that deal with students' interpretation of static ERs that convey *structural* phenomena, such as chemical and biological structures, are dealt with first. Work on static ERs that infer *spatial* phenomena such as ERs portraying rotations of chemical structures and cross-sections of biological specimens are discussed second. Thirdly, static ERs that portray *dynamic* phenomena that are *physical* in nature, such as, weather patterns, phases of the moon, lightning, mechanics, hydraulic pumps, braking systems, and plate tectonics are dealt with third. Investigations on static ERs that infer *dynamic* phenomena that are *abstract* in nature such as subcellular processes, energy, optics and electric circuits are presented fourth. Fifthly, research on static ERs that are *graphic-word* in nature such as flow diagrams, food webs, kinship trees and ERs that contain arrow symbolism are examined. Studies that have considered the use of *animated* ERs in science education, are discussed sixth. Finally, investigations on *multimedia* ERs in science education are dealt with. In addition to the research findings, attention is also given to the possible sources of students' difficulties.

2.6.1 Learning and teaching with static ERs that portray structural phenomena

In the 1960-1980's, Francis Dwyer studied students' interpretation of ERs representing the structure of the human heart. In one example, Dwyer (1967) investigated students' interpretation of ERs of the heart across a *visual realism continuum*. By studying the static ERs across such a continuum, it was found that a useful account of the effectiveness of different ERs of the *same* phenomenon could be formulated. For instance, realistic pictures of the heart were found to be most effective compared to other representations in meeting desired learning outcomes, a result replicated in further studies (e.g. Dwyer, 1969). An explanation for this was that realistic ERs contain more pragmatic detail and, therefore, learners are able to encode information more naturally (Dwyer, 1969). Additional findings suggested that not *all* ERs are effective for promoting understanding and some ERs are more efficient than others (e.g. Lohse *et al.*, 1991; Dwyer, 1970). A further study (Joseph and Dwyer, 1984) used a similar continuum approach and investigated students' interpretation of static ERs portraying an *integration* of abstract and realistic information. Realistic ERs were integrated with abstract ERs by merging a line drawing of one half of the heart with a photograph of the other half. It was discovered that increased levels of prior knowledge supported learning with the realistic part of the ER favourably, while students with lower prior knowledge levels found the abstract half more beneficial. In relation to this, Dwyer (1975) has shown that students with low prior knowledge levels need to spend more *time* on interpreting realistic ERs of the heart.

Dwyer (1968) has also revealed that the learning effects of ERs depend on many criteria. For example, learners that have not been exposed to many ERs lack the procedural skills necessary for interpretation. Also, an ER may have such an extrinsic impact on a learner, that it causes distraction from the underlying content contained in the ER (Dwyer, 1968). In other writings, Dwyer (1970) has expressed that it is important for learners, and educators alike, to identify which graphical ER components best facilitate learning. For instance, Dwyer (1972, 1970) has shown that students preferred coloured ERs of drawings of the heart rather than their monochrome counterparts (see de Lange, 1999). Dwyer (1972) has correlated the use of colour with increased motivation in learners and has concluded that the use of colour is an important instructional variable in science education. In support of this, Reid and Miller

(1980) have observed that a learner's attention to a static ER is very much influenced by the use of colour and have revealed differing learning outcomes with different colour use. Interestingly, colour was sometimes found to be a distracter in that it restricted learners' "scanning" processes, causing them to be directed to insignificant graphical features.

Continuing with biology, Reid and Beveridge (1986) investigated students' interpretation of static ERs portraying biological structures such as cells, tissues, teeth, skulls, insects and the mammalian heart. The general performance of school learners was shown to share a positive correlation with their ER processing skills (Reid and Beveridge, 1990; Reid *et al.*, 1986). More specifically, findings revealed that learners with different ER processing abilities employed different strategies when learning from illustrated text. For example, less successful learners needed more time to integrate the information presented in ERs while Reid and Beveridge (1990) have implied that learners are often unaware of how to *use* ERs appropriately. Subsequently, Reid (1990a, b) defined a *picture superiority effect*, which suggests that ERs are automatically seen to facilitate learning from text because ERs are always considered suitable representations of the concept. In this regard, it is cautioned that ERs are often naively and incorrectly seen by experts to be superior learning devices that *always* yield the intended understanding. In support of Reid's effect, Soyibo (1994) has found that, when required to draw physical specimens from direct observation, secondary school biology students reverted to externalising the associated textbook ER, instead of drawing what they observed.

In terms of students' interpretation of static ERs of structures in a chemistry domain, Noh and Scharmann (1997) investigated students understanding of ERs depicting matter. The research revealed that questions presented together with ERs portraying the molecular level helped students construct more scientifically correct conceptions of matter and was an effective means for improving students' conceptual understanding in chemistry. Additionally, a study by Pavlinic *et al.* (2001) has suggested that chemistry learners should be presented with the opportunity of 'moving between' different ERs of the same chemical structure, be it at the macroscopic, microscopic, submicroscopic or symbolic level. The authors found that a *multiple representations approach* was directly related to improved understanding of chemical ERs (see Barke, 1993). It was also found that factors such as 3-D-shape, colour and interactivity where important criteria for the refinement of students' ideas. On this score, Sumfleth and Telgenbüscher (2001) have advised that factors such as, learners' personal

views as to whether an ER is relevant, social context, attitudes and personal learning styles also affect the way static ERs of chemical structures are interpreted (e.g. Wheeler and Hill, 1990).

2.6.2 Learning and teaching with static ERs that portray spatial phenomena

Exercising spatial cognition is necessary for processing scientific ERs (e.g. Lord, 1990). This is especially true for domains such as chemistry, biochemistry, physics and astronomy, where students are required to visualise spatial configurations of 3-D objects (e.g. Richardson and Richardson, 2002; Seddon and Shubber., 1984). To interpret static ERs that portray spatial properties, students' have to mentally manipulate 2-D ERs into their 3-D analogues (e.g. Shubbar, 1990; Pribyl and Bodner, 1987). Not only do students have to understand the *spatial relationships* represented in the static ERs (e.g. width, depth and height) but they also have to visualise how the ER would *transform* upon rotation or change in view (Shubbar, 1990). Even though much work has focused on 3-D visual thinking in chemistry (e.g. Tuckey and Selvaratnam, 1993; Tuckey *et al.*, 1991; Pribyl and Bodner, 1987; Baker and Talley, 1974), what has not often been considered is that spatial aptitude also applies to other disciplines such as biology. In these cases, students are also required to spatially visualise structures such as cut surfaces of tissue cross-sections, or interpret ERs such as Cartesian graphs (Lord, 1990).

In the context of interpreting static ERs portraying spatial properties in chemistry however, work by Shubbar (1990) has shown that a large proportion of students find spatial operations such as rotations, reflections and inversions difficult. Shubbar (1990) has demonstrated that the difficulties emanate, in part, from the lack of student understanding of the artistic means used to represent spatial features. In the study, an experimental group of students observed changes in the rotation of physical 3-D molecular models by viewing the shadows the models cast upon rotation, while a control group was not exposed to the viewing. Afterwards, both groups performed a post-test where they had to choose a static ER (from four possible options) that best represented the effect of a rotation about one of either the X, Y or Z axes. The post-test data revealed that the experimental group were better at visualising the rotations than the control group. Interestingly, Shubber (1990) noted that there were no significant

differences between the "shadows" and "no-shadow" forms of display or between "high and low" rotation speeds on students' interpretation of the static 2-D ERs.

In agreement with Shubber' s (1990) findings, work by Tuckey and Selvaratnam (1993) has found that student difficulties with spatial ERs in chemistry are often due to learners' misinterpretation of the depth information provided in 2-D ERs (e.g. Seddon and Shubber., 1984). Interestingly, they have also suggested that forming part of students' spatial difficulties are misunderstandings of the text used to describe the ERs. For example, the authors found that students struggled with the semantics of phrases such as, "rotation about the X-axis". Together with others (e.g. Tuckey *et al.*, 1991), the study advocates that spatial skills are critical to understanding chemistry and therefore, student proficiency should first be tested before entering chemistry courses. Seddon and Shubber (1984) investigated students' visualisation of chemical structures by presenting 2-D ERs of different stages of a particular rotation. In agreement with results presented in section 2.6.1, it was shown that when ERs were monochrome, no significant learning occurred, while multi-coloured ERs yielded significant learning (e.g. Winn, 1991). It is suggested that during learning, students should be explicitly guided as how to compare different 2-D ERs that portray 3-D space in chemistry.

With respect to the studies above, Tuckey and Selvaratnam (1993) have advised that there are at least three levels of cognitive complexity associated with the visualisation of chemical structures. The proficiency shown to be the easiest is the *transformation* of the 2-D ER into its 3-D representation. Perceiving the *orientation* of the structure in space is considered more difficult. The most demanding however is *rotating* the structure in the mind's eye. In this regard, research shows that students with better visualisation skills are better at solving chemistry problems in general. Baker and Talley (1974) have shown this to be true for inorganic chemistry, while Pribyl and Bodner (1987) have found a positive correlation between spatial ability and achievement in organic chemistry. Overall, it is argued that spatial learning be viewed as an *active* process that does not just benefit learning in chemistry, but facilitates the learning of other scientific subjects as well (e.g. Barke, 1993; Lord, 1990).

In terms of interpreting static ERs that show spatial relationships in biology, Lord (1990) assessed 250 undergraduates' visualisation of 2-D ERs showing cut surfaces of 3-D cross-sections. Spatial orientation tasks required subjects to, "mentally envision an object within its surroundings" and spatial visualisation tasks required students to "mentally manipulate" the

image (Lord, 1990). The skills were integrated into biology-specific questions and students had to consider an object's symmetry and depth, and view plant and tissue specimens under a microscope in order to respond to the questions. In agreement with findings in chemistry, results confirmed that students who initially showed high visual-spatial ability performed best in subsequent tests. Students, who were found to initially have poor visual-spatial skills, showed improved understanding after receiving visual-spatial training. In a link to this work, Constable *et al.* (1988) has also investigated high school students' understanding of sectional drawings in biology textbooks. Findings revealed that students' struggled to interpret the cut surfaces of ERs representing alveoli, spirogyra, hydra, blastula, fish and the uterus. Among other factors, as was the case in chemistry, difficulties were found to be related to the graphical means in which the ERs were represented. As an implication, understanding pictorial conventions is necessary if spatial interpretation is to be at all beneficial.

Similarly to Lord (1990) and Constable *et al.* (1988), Sanders (1995) established that a large proportion of students struggle to interpret *depth cues*. Depth cues are ER markings that provide information about an object's 3-D space (Coon, 2001) and are used in biology textbook ERs to represent 3-D biological specimens as 2-D longitudinal- and cross- sections. Statistical analyses (Sanders, 2002, 2001) showed a strong correlation between students' difficulties with depth cues and their low spatial visualisation ability. Participants found spatial visualisation of biological cross- and longitudinal sections of ERs displaying hydra, the throat of a fish, spirogyra and flatworm extremely demanding. In general, studies dealing with the spatial interpretation of static biology ERs (e.g. Sanders, 2001, 1995; Lord, 1990; Constable *et al.*, 1988) suggest that students find it challenging to mentally transform and manipulate 2-D ERs that represent the third dimension. The research above goes a long way towards confirming Reid's (1990b) *picture superiority effect* by demonstrating that students do not always interpret an ER's conventions and visual markings as textbook authors intend and as teachers assume.

2.6.3 Learning and teaching with static ERs that portray dynamic phenomena that are physical in nature

Since the 1980's, Richard Lowe has published remarkable findings on students' interpretation of static ERs that represent the dynamic and physical ideas of meteorology. In doing so,

Lowe (1996, 1994a, 1993a, 1993b, 1989) has extensively studied ER-processing differences between experts (meteorologists) and novices (non-meteorologists). Early research (Lowe, 1989) asked experts and novices to complete the markings on a meteorological ER while viewing an incomplete version. Here, participants had to rely solely on their existing mental models of meteorological phenomena: those constructed from newspapers and television in the case of novices and from experience in the case of experts. Lowe (1989) discovered that the two groups found particular ER features more salient than others, and experts inspected the chart in a fundamentally different manner to novices, indicating crucial qualitative differences between experts and novice's mental representation of weather map ERs.

Findings from a subsequent study confirmed the results above when Lowe (1993a) showed clear differences between *where* novices and experts focused their attention, and the way information was *searched* for on the ER. Novices tended to view the ER in a simple east-to-west manner, in accordance with explicit visuo-spatial markings on the map such as shape, position and topography. Experts on the other hand, viewed the map in a much more complex way: in a north-to-south manner, in accordance with the actual meteorological concepts implied by the graphical markings (Lowe, 1993a). Hence, experts built up their understanding in a step-wise fashion that depended on the conceptual relevance of the markings.

Further findings (Lowe, 1993b) have shown that experts construct mental representations that are more *semantically based*, while novices' mental representations are based largely on the visuo-spatial characteristics of the ER (see Bennett and Flach, 1992), which causes novices' mental representations to be very unorganised. For instance, the study showed that novices often discarded subtle ER markings instead of interpreting them as being of importance to the context of the weather map (Lowe, 1993b). In subsequent writings, Lowe (1994a) explains that an ER has many *levels of structure* and that students are often unaware of this and concentrate on superficial elements of the ER. Hence, students "miss" features, which even though subtle, are important for gaining the intended meaning.

In addition to the studies above, an important feature of Lowe's work has been establishing the extent to which an individual's existing knowledge affects ER processing. In this regard, Lowe (1996) has stated that even though an expert possesses a larger *knowledge base* than a novice, this on its own, *cannot* account for processing differences. Lowe (1994a, 1993a) has

emphasised that experts are not superior ER processors just because they "know more", it is also because mentally, they represent information differently to novices. With respect to Lowe's findings, Chi *et al.* (1981) considered this exact question. Novices (beginning physics students) and experts (experienced physicists) were asked to sort a number of mechanics problems into categories. Novices grouped problems that involved similar surface features (e.g. inclined planes), while experts grouped the problems according to the particular physics principles needed to solve them. Novices tended to focus on the surface structure of the problems, while experts focussed on the problems' deeper structure. Chi *et al.* (1981) suggest that experts bring a lot of *procedural knowledge* to the problem, while novices lack the abstract procedural knowledge needed to solve the problem (e.g. Egan and Schwartz, 1979).

Based on the work above, it can be argued that the mental representation that a learner constructs from an ER has a direct bearing on how the ER will be understood (e.g. Lowe, 1993a; 1989; Chi *et al.*, 1981), with both background and procedural knowledge playing roles (e.g. Lowe, 1996; Winn, 1993). As stated by Cheng *et al.* (2001), during interpretation of an ER, perception of the graphical markings is also *modulated* by learners' knowledge of what the markings *mean* (e.g. Ametller and Pintó, 2002). If this modulation is unsuccessful, an *over* reliance on the ER markings occurs, which can cause difficulties. Furthermore, Lowe (1996, 1993a, 1989) has suggested that experts organise their domain-specific knowledge hierarchically. It is thought that this arrangement allows for the relative importance of each graphical feature to be easily identified and processed. Consequently, the processing goals of novice and expert viewers become very different (Lowe, 1989). It follows, that experts are able to *chunk* information from the ER into meaningful wholes, something that novices struggle to do (e.g. Lowe, 1989; Egan and Schwartz, 1979).

Apart from Lowe's significant research on students' interpretation of static ERs that show dynamic phenomena that are physical in nature, Peña and Quílez (2001) investigated 78 students' interpretation of ERs that represented different phases of the moon. Upon analysis of data obtained through drawing outputs, the study showed that students' found it an immense challenge to communicate their ideas through diagrams. A further compounding factor was that the quality of their diagrams as tools for explanation was found to be poor. As noted in section 2.6.1, it was also revealed that students often drew phases of the moon diagrams similar to "standardised" textbook ERs (e.g. Soyibo, 1994). Since students continuously referred to "accepted" ERs as a means of explanation, they used their ERs in a

superficial way, characterised by memory rather than on a deeper understanding of what the ER represented (e.g. Kozma, 2003). In relation to the former, Yair *et al.* (2003) have suggested that astronomy learners in general, have the potential to construct misconceptions even when viewing rich and detailed ERs. Since astronomy ERs are often complex, learners deviate from the intended leaning objectives. As discussed with respect to the spatial visualisation of objects (section 2.6.2), engagement in astronomy requires specialised cognition such as 3-D ability as well as an understanding of geometrical dynamics, which makes ER processing that much more demanding (e.g. Yair *et al.*, 2003). With regard to studying learners' use of their generated ERs as a means of communication, Gobert and Clement (1999) investigated students' mental model construction and related conceptual understanding in the domain of plate tectonics. Through the use of student-generated ERs, the authors found that diagramming allowed students to construct rich mental models. They found subsequently, that these mental models caused students to make better inferences to the conceptual nature of plate tectonics and allowed for deeper text processing (e.g. Waddill *et al.*, 1988).

Richard Mayer is another worker who has thoroughly investigated students' understanding of static ERs that portray dynamic physical processes. In one study, Mayer *et al.* (1995) examined subjects' interpretation of static annotated ERs that showed how lightning worked. Annotated ERs were found to help students *signal* which images and words were relevant for learning. In addition, annotated ERs helped subjects organise information and provided appropriate cues for linking visual and verbal representations. Similar work (Mayer *et al.*, 1996) investigated subjects' interpretation of the process of lightning through the use of ERs and textual captions. Results suggested that a verbal summary alone was not as effective as a *multimodal* summary: one that contained both ERs *and* text within the same proximity. An inference from the work is that multimodal ERs can be beneficial because they place low cognitive loads on working memory. Multimodal ERs are examined in detail in section 2.6.7.

In addition to the above, an earlier study by Mayer and Gallini (1990) explored students' interpretation of ERs that represented the functions of a braking and pump system. The mechanical systems were presented to subjects in three forms. In one form, the ERs were presented as "steps" where a picture was accompanied by a textual annotation, explaining how brakes and pumps *worked*. In another form, ERs were presented as "parts" where textual labels pointed to pictures of the mechanical *parts* involved in the systems. The last form

combined the two former presentations as "steps-and-parts" ERs. Results from the study implied that during learning with ERs, *both* the text and graphics should match the proposed instructional goal. The study concluded that, "a diagram is worth ten thousand words" (p. 725) when the text present in an ER can be understood, when the effectiveness of an ER is considered in terms of learners' interpretations, when ERs are explanatory, and when a learner does not have any prior knowledge. A previous study (Mayer, 1989b) on students' interpretation of ERs representing hydraulic brake mechanisms, found that students who interpreted labelled ERs performed better than those who interpreted only pictures or only text. The data demonstrated that ERs, which contain suitable textual adjuncts, help learners focus their attention, which aids the construction of useful mental models.

2.6.4 Learning and teaching with static ERs that portray dynamic phenomena that are abstract in nature

Students can show difficulties when reasoning about processes that cannot be observed directly (e.g. Hull, 2003; Lowe, 1996; Mayer *et al.*, 1995). Research on learners' interpretation of static ERs that portray dynamic phenomena that are abstract in nature has shown this to be true. When portraying *subcellular* processes for example, ERs are utilised to represent the biological situation. In these cases, learners have to read symbolic markings that represent abstract processes; which requires certain skill (e.g. Egan and Schwartz, 1979). However, although there has been a dramatic increase in the number and complexity of such ERs used in science teaching, instructors continue to ignore the fact that ERs displaying abstract concepts contribute to students' learning difficulties (e.g. Kindfield, 1993/1994, 1992). For example, in one study investigating the above, Kindfield (1993/1994) considered how individuals with varying domain-specific knowledge used ERs to reason about meiosis. Data was collected in the form of think-aloud interview sessions. As discussed with respect to static ERs showing physical as opposed to abstract phenomena (section 2.6.3), Kindfield (1993/1994) also found significant differences between the manner in which advanced participants used their generated diagrams to solve problems in comparison to less advanced participants. In particular, Kindfield (1993/1994) observed that less advanced participants only used a maximum of two different representations to portray replicated chromosomes, while more advanced participants used a variety of diagrams. With more advanced participants, the entire chromosome wasn't always represented; writing down only the allele

letter(s) corresponding to a chromosome often sufficed (Kindfield, 1993/1994). Less advanced participants however, generated literal representations of the chromosomes and included irrelevant structural detail. The study also found that more advanced participants adjusted their generated diagrams with ease, depending on what the problem required at a particular time. More advanced participants removed irrelevant detail and "fine-tuned" their diagrams as problem solving proceeded, using their diagrams in distinguishable and systematic ways. Kindfield (1993/1994) suggests that the fine-tuned diagrams helped advanced participants formulate problem-solving strategies that in effect, mirrored the cognitive mechanisms that were used to arrive at a solution. Overall, Kindfield (1993/1994, 1992) concluded that individuals' domain-specific knowledge of meiosis shared a close relationship with the way generated diagrams are used to solve problems: more advanced participants made use of *diagram-related reasoning behaviours*, behaviours which novices lacked. Kindfield (1993/1994) has postulated therefore, that when individuals generate understanding of abstract processes in science, a *coevolution* of pictorial skill and conceptual understanding occurs.

In a review of four studies conducted on learners' interpretation of static ERs portraying dynamic and abstract ideas, such as optics and energy, Pintó and Ametller (2002) established that students often interpret ERs showing these phenomena in a *narrative* manner, resulting in the formation of irrelevant ideas. Through a 'story-like' interpretation, learners attach a time variable to such ERs, when no time dimension is implied. Furthermore, the authors suggest that when such ERs are unfamiliar, learners turn to everyday conceptions to 'make up' for missing background knowledge and fail to appreciate the metaphorical function of ERs (e.g. Levin *et al.*, 1987). Contributing to this narrative problem is the fact that English-speaking students, unlike Jewish or Arabic-speaking students, tend to read ERs in a left-to-right manner (Lowe, 1993a; Winn, 1993); causing even further problems when complex ERs are viewed. Other work by Stylianidou *et al.* (2002) with 104 pupils, on their understanding of textbook ERs portraying energy, found that students' struggle to interpret ERs that portray ideas that are conceptually demanding, particularly those that are abstract.

Exploration of static ERs portraying abstract concepts has also been carried out on students' interpretation of electric circuit ERs. In one study, Egan and Schwartz (1979) showed that interpreting these symbolic ERs requires certain perceptual skill. In particular, Egan and Schwartz (1979) found that experts could internalise a large amount of graphical information

very efficiently. This process, termed *perceptual chunking*, helped experts make meaningful links to the appropriate conceptual understanding, something that novices battled to do. Chunking allowed experts to group their perceptions of the circuit ERs into functional units, in a similar way to how a Chess Master is able to recall specific chess positions. Novices however, seemed to have fewer chunking units at their disposal (Egan and Schwartz, 1979).

In regard to other work on circuit ERs, Hill (1990) has suggested that students' often interpret circuits as the *reality* rather than as symbolic abstractions of a scientific idea. Similarly, Johsua (1984) found that students' interpreted circuit ERs as a "system of pipes" (p. 275), where the passage of current was seen as being similar to a "fluid" with little cognisance given to underlying concepts such as potential difference. Additionally, Johsua (1984) revealed a *topological effect*, where students interpreted different ERs of the *same* electric circuit in varying ways. Finally, Winn's (1991, 1988) studies have found that students' ability to process circuit ERs depended very much on the amount of detail in the ERs. In agreement with Dwyer's (1972, 1970) work in the context of static ERs portraying structural phenomena (section 2.6.1), it was found that when levels of detail were increased, students paid more attention to the detail, rather than to the holistic message conveyed by the ER.

2.6.5 Learning and teaching with static ERs that are graphic-word in nature

William Holliday (e.g. 1977) has referred to static ERs that contain graphical components *as well as* textual components as *picture-word* or *block-word* ERs. Picture-word ERs have textual adjuncts associated to the picture(s), while block-word ERs contain verbal information that is placed within "block", or other regular shapes (e.g. Winn, 1980). For this chapter, Holliday's designation is extended, and the term *graphic-word* is used to include ERs such as family trees, flow diagrams, food webs, and ERs that contain arrow symbolism.

In 1977, Holliday *et al.* investigated high school students' cognitive responses to flow diagrams in biology (also see Holliday, 1975b). One finding was that learners considered flow diagrams to be *manageable* ERs because they were immediately exposed to the "big" picture. In an explanation of this, Holliday *et al.* (1977) have referred to the tenets of Gestalt psychology. The Gestalt paradigm suggests that, "the whole is greater than the sum of its

parts" and emphasises that learning from ERs therefore, should be considered in terms of the perception of whole units, rather than on the individual parts making up the unit (e.g. Coon, 2001). For this reason, Holliday *et al.* (1977) imply that the *whole* ER rather than its component parts should be presented to students whenever possible. However, since learners are often unaware of how to *use* flow diagrams appropriately, Holliday (1976) has cautioned that for learning, these ERs aren't *always* superior to text.

In addition to Holliday's work, William Winn has studied students' problem solving with graphic-word ERs that represent kinship relationships (family trees). In one study, Winn *et al.* (1991) postulated that when students interpret ERs that represent concepts spatially, then viewers' processing demands are substantially reduced. The research showed that visual objects that were in close proximity to each other (e.g. separate family names and the lines linking the names) were perceived as belonging to the "same group". In support of this finding, Winn *et al.* (1991) suggest that a triangle is indeed interpreted *as a triangle* and not as three separate lines. Through the same argument, Winn *et al.* (1991) found that for a family tree ER, it was easy for subjects to perceive hierarchical structures quickly, which made problem-solving more efficient. The results also demonstrated that the computation required to interpret an ER can be reduced significantly when the spatial arrangement of concepts carry meaning (e.g. Olivier *et al.*, 2001; Winn *et al.*, 1991; Larkin and Simon, 1987).

A different study by Griffiths and Grant (1985) revealed four misconceptions related to students' interpretation of food web ERs. Firstly, some students thought that a change in size of one population would only affect the size of another population when the two populations were directly related (i.e. through predator and prey). Similar *localised* (rather than global) reasoning has been discussed by Cohen *et al.* (1983) in physics and by Anderson *et al.* (1999) in biochemistry. Secondly, some students thought that populations, which were "higher" in terms of their spatial arrangement, were always predators of the populations "below" them. Thirdly, some students did not acknowledge that a change in size of a prey population would affect the size of the predator population. Lastly, some students' thought that if the size of a single population was changed, then all other populations would be altered by the same degree.

Remaining in a biological domain, Soyibo (1994) studied 11 290 graphic-word ERs present in 12 O-level biology textbooks. Three major labelling mistakes were revealed, namely the

scientifically incorrect labelling of drawings, the labelling of *single* structures in their *plural* form and, lines that linked structures to labels, pointing to empty spaces. Soyibo (1994) has claimed that due to these errors, students are presented with inaccurate external models, which hamper their understanding of biological functions. Soyibo (1994) has also stated that those teachers, who realise that erroneous labelling does occur, may find it challenging to convince students of such problems, because textbooks are often viewed as error free.

Schollum (1983) has conducted research on graphic-word ERs in science textbooks that contain *arrow* symbolism. In this study, fourteen-year-old students' understanding of ERs portraying food chains, matter, forces and the earth's gravitational field was gathered. Two findings were that arrows were interpreted in ways that textbook authors would not expect and, arrow "conventions" across ERs and textbooks were used inconsistently. Inconsistency was made clear when at least six different uses for arrows were revealed: as labels, for measurement, as forces, to show relationships, to show changes and, to show sequences (Schollum, 1983; also see Henderson, 1999). Furthermore, Schollum (1983) found that students often interpret ERs in a manner that parallels their prior, everyday views and has suggested that science instructors be made aware of the extreme variation in arrow use across science textbooks.

In relation to ERs containing arrow symbolism, other workers have reported detailed findings. Ametller and Pintó (2002) found that when secondary students interpreted ERs containing arrows to represent energy, instead of interpreting the arrows as indicating a *transfer* of energy, they were interpreted as energy somehow escaping from an object. In addition, *broader* arrows were interpreted as having a *larger* amount of energy. The same study found that *different* interpretations were stimulated by identical arrow markings. Du Plessis *et al.* (2003) examined high school biology students' interpretation of arrow symbolism contained in ERs of the cardiac cycle and thermoregulation. Perceptual difficulties, arising out of erroneous search strategies within the ER, reasoning difficulties, emanating from poor ER processing skills and conceptual difficulties, originating from limited prior knowledge, emerged from the data. The work suggested that difficulties are enhanced when ERs are of poor quality, especially when ERs have not been designed in accordance with any meaningful design principles. The authors also suggested that the diversity of arrow use and the lack of standardisation across scientific ERs will continue to contribute to many student difficulties.

2.6.6 Learning and teaching with animated ERs

Modern science education is witnessing a sharp increase in the use of dynamic and simulated computer-based ERs (e.g. Lewalter, 2003; Scaife and Rogers, 1996). As a result, workers have begun investigating the role of animated ERs in science learning in earnest (e.g. Lowe, 2003, 1999; Kozma, 2003; Nerdel *et al.*, 2003). Animated ERs differ from static ERs in that they exhibit *transitory* information such as *form* and *position changes* (Lowe, 2003; Cheng *et al.*, 2001) and viewers of animated ERs are presented with information that static ERs cannot offer. As a result, during interpretation of static versus animated ERs, different cognitive demands are placed on viewers (e.g. Lewalter, 2003; Lowe, 2003).

One concern in recent literature is that due to the nature of their presentation, many educators simply assume that animated visuals are more powerful learning tools than their static counterparts (e.g. Schnotz and Lowe, 2003; Scaife and Rogers, 1996). However, research has shown that learning from animations may not always be beneficial (e.g. Lewalter, 2003). Lowe (2003) has provided two possible reasons for this. In what he terms *overwhelming*, processing an animated ER is extremely demanding. The fact that the ER information is dynamic and aesthetically pleasing does not always mean that learning is effective. This is because the animation will place greater cognitive load on the viewer than in the case of static ERs (e.g. Lowe, 1999). It follows, in what is termed *underwhelming*, that the viewer may decrease their level of engagement with the visual, due to its highly dynamic and aesthetic appearance.

Lowe's (2003) recent research, aims to aid students' interpretation of static weather map ERs through the use of animated ERs. He has postulated that dynamic ERs could be used to provide novices with the necessary domain-specific knowledge required to interpret static weather maps. The literature refers to this process as *bootstrapping* (e.g. Roth, 2002; Cheng *et al.*, 2001), a situation similar to a "chicken-and-egg" dilemma: without at least some content knowledge a learner is unlikely to interpret a scientific ER adequately but obtaining this knowledge requires ER interpretation. In an attempt to solve this tautology, Lowe's (2003) designed animations aimed to actively bootstrap novices into experts' ways of reading ERs so that novices could *model* expert thinking. Upon analysis of the data generated from Lowe's (2003) study, he found that novices extracted information from animated weather

maps by concentrating on the *perceptual salience* of the display. Animated features of high "perceptual salience" (more transitory or more graphically vivid) were read most often, while features showing low salience were generally neglected, despite being important for successful interpretation (Lowe, 2003). This finding is in line with Lowe's earlier work (e.g. 1993a, 1993b), which found that when interpreting ERs, learners engage in a highly *selective* approach, defined by the search for graphical markings that are more prominent (e.g. Ametller and Pintó, 2002; Cheng *et al.*, 2001). As a result, a *perceptual effect* (Lowe, 2003) comes into play: if students concentrate on the salient visual information rather than on the underlying relevance of the graphics, superficial mental models are formulated. Interestingly, Lowe (2003) concluded that even though dynamic weather changes can be animated, no significant interpretation differences between animated and static ERs were discovered.

Of concern to teaching with animated ERs, Lowe (2003) suggests that misconceptions are induced when viewers are unable to *control* the animation, such as being able to manipulate the speed of presentation. Associated to this control is the necessary instructional guidance, considered imperative for learning with animated ERs (e.g. Duchastel, 1988). Overall, Lowe (2003) points out that there is a danger brewing. Even though there are tremendous prospects for animation as a learning medium, users should be guided in how to use animated ERs proficiently and should avoid using animated ERs just for the sake of using them. Rather, educators should be sure of their potential learning outcomes as well as their design.

In a different study, Lewalter (2003) examined 60 students' interpretation of static versus animated computer-based ERs of ideas in astrophysics. Like Lowe (2003), despite the apparent learning advantages of dynamic ERs, no statistically significant superiority of dynamic ERs over static ERs was obtained. The findings suggest that learning from static and animated ERs can, in certain cases, be equally effective. In agreement with the field in general, Lewalter (2003) advocates that the learning support offered by an ER is very much dependent on the cognitive strategies that the viewer employs. Thus, this research also suggests that viewers of animated ERs require facilitative guidance for interpretation to be favourable. In agreement with this stance, Duchastel (1988) has pointed out that the potential benefits of animated ERs must be determined in terms of the style and design of presentation.

2.6.7 Learning and teaching with multimedia ERs

The term *multimedia* is a buzzword in our technological age. Formally, the term refers to, "the combination of multiple technical resources for the purpose of presenting information represented in multiple formats..." (Schnotz and Lowe, 2003, p. 117). Examples of *multiple formats* include the *combination* of text, static ERs, animated ERs, video ERs or sounds. When two or more formats are presented *simultaneously*, then communication is no longer a single medium, but a *multimodal* medium or, a multimedia (e.g. Seufert, 2003; Mayer, 1997). Therefore, multimedia can be book-based or computer-based. Many of the inroads that have been made into learning from multimedia can be largely attributed to the work of Richard Mayer. Even though Mayer's work (e.g. 2003) has been prolific in the area of cognitive psychology, multimedia in science education research is still very young. In general, research on multimedia ERs has been concerned with how best to *combine* information so as to ensure the greatest learning benefit (e.g. Mayer, 1997). As with animated ERs, this concern has arisen due to the need to reduce the cognitive load placed on viewers of multimedia (e.g. Mayer *et al.*, 1996).

In one study, Mayer and Sims (1994) investigated learners' interpretation of the human respiratory system. Learners viewed computer animations while concurrently listening to a narration. It was found that multimedia ERs helped learners with low prior knowledge to transfer what they had learnt to new problem-solving domains, especially when verbal and pictorial representations were presented together, rather than separate. In a similar study, Mayer and Anderson (1992) investigated students' interpretation of multimedia showing how a bicycle tyre pump and vehicle braking system functioned. Animation on its own did not improve learning; only when coupled with narration, did learning improve statistically. It was confirmed that constructing meaningful connections between visual and verbal modes is crucial if multimedia learning is to be at all significant (e.g. Mayer and Anderson, 1991).

Mayer's *theory for multimedia learning* (section 2.4) identifies four aspects central to multimedia learning (Mayer, 2003; Mayer *et al.*, 1996, 1995; Mayer and Anderson, 1992, 1991). Firstly, the *multimedia effect* suggests that deeper learning takes place when ERs (e.g. pictures, diagrams and animations) and words (e.g. text or spoken) are *combined* rather than when they are presented in isolation. Secondly, the *coherence effect* suggests that learning is

increased when irrelevant information is reduced. Thirdly, the *spatial contiguity effect* suggests that learning is enhanced when words are placed in close proximity to pictures. Finally, the *personalization effect* proposes that students construct more useful mental models when accompanying text is presented in a conversational manner.

Although Mayer (e.g. 2003, 1997) suggests that the potential of multimedia learning is enormous, multimedia learning does not *always* lead to favourable understanding. It is pivotal that designers produce information that promotes efficient *mapping* between verbal and pictorial modes. To do so, information should be combined in a coordinated manner, one that matches current learning theories (Mayer, 2003, 1997; Mayer and Anderson, 1992, 1991). Current learning models view individuals as being *actively* engaged in making sense of information, rather than absorbing information *passively* (e.g. Mayer, 2003; Osborne and Wittrock, 1983). Although this is deemed crucial, Mayer (1997) has commented that, "the potential for computer-based aids to learning remains high, although the current contribution of technology to pedagogic innovation is frustratingly low." (p. 17).

In the form of various electronic resources, science educators in the molecular and cellular biosciences are increasing their use of multimedia ERs (e.g. Flores *et al.*, 2003). It is assumed that multimedia provides students with an always-effective way of presenting 3-D structure-function relationships of molecules. In perhaps an extension of Reid's (1990b) picture superiority effect (sections 2.6.1 and 2.6.2), consider the following. Richardson and Richardson (2002), famous for their development of *ribbon* ERs to depict 3-D protein structure in biochemistry, have warned that, "...there is little experimental data on either the absolute or the relative effectiveness of these materials [multimedia] for teaching 3-D literacy and only minimal guidance about the best ways to use them..." (p. 21). It appears, as argued for static and animated ERs that, the use of multimedia tools might not always lead to the desired learning outcomes in the molecular sciences. As discussed previously, factors such as students' 3-D visualisation skills (section 2.6.2), their prior knowledge (e.g. Brna *et al.*, 2001) and the nature of the multimedia itself (e.g. Duchastel, 1988) have to be carefully considered.

Work by Seufert (2003) on multiple ERs, portraying the biochemical relevance of iron and vitamin C in human metabolism, indicated that often during viewing, learners did not construct appropriate mental representations. The work found that students with low prior knowledge tended to memorise the ERs, rather than expend any effort on actually processing

the ERs (e.g. Mayer and Sims, 1994). Similar to work on animated ERs (section 2.6.6), the study suggested that learners with low prior knowledge levels must be *supported* when learning from multiple ERs. Furthermore, the study has called for more research into the relationship between external and internal representations during learning (Seufert, 2003; Nerdel *et al.*, 2003), an important feature of distributed cognition, outlined in section 2.1.

Studies by Kozma (Kozma, 2003; Kozma and Russell, 1997) investigated novice and expert understanding of multimedia ERs of chemistry phenomena. By representing chemical reactions through video, graphs, animations, molecular models and symbolic equations, student data was collected. One finding was that students' construction of understanding in chemistry is an immense challenge because molecular phenomena cannot be experienced directly (e.g. section 2.6.4; Hoffmann and Laszlo, 1991). In addition, since chemistry is often communicated through symbolic graphical markings, understanding chemical ERs is made even more complex. In support of findings discussed in sections 2.6.3 and 2.6.6, novices focused on surface features of the multiple ERs to generate meaning. Interestingly, *experts* were found to also rely on surface features of the ERs, but were able to organise their interpretations based on the necessary underlying conceptual knowledge. However, experts' showed a more *transformational* use of surface ER features across different representation modes. Thus, experts are capable of moving across ERs with "fluidity" and their understanding is shared *across* multiple ERs. Kozma and Russell (1997) and Kozma (2003) have referred to this as *representational competence* and suggest that experts extract "clusters" of information as meaningful groups. Elsewhere in this review (section 2.6.3 and 2.6.4), a comparable process has been referred to as perceptual "chunking" (e.g. Koedinger and Anderson, 1990; Egan and Schwartz, 1979).

2.7 Summary

A synthesis of the field's findings on the use of different types of ERs for teaching and learning in science has been offered in this review chapter. Based on the discussion and analysis, the following salient points have emerged as being representative of the popular literature. These points will be carried forward into the rest of the thesis to where appropriate, facilitate discussion and interpretation of the results.

1. The cognitive processing required for reading text is *different* to the processing required to read ERs.
2. Not *all* ERs are effective for learning: some ERs are better than others and some ERs are more difficult to process than others.
3. Educators and authors often view ERs as unfaltering learning tools that *always* convey the intended understanding and learners often view scientific ERs to be *error free*.
4. In general, academic performance in science shares a close *relationship* with ER-reasoning skills and those students proficient in *spatial visualisation* often interpret other scientific ERs effectively.
5. Generally, realistic ERs are *easier* to interpret than abstract ERs.
6. Students with poor ER-reasoning skills have to spend more *time* reading ERs because they respond to ERs in *different* ways and differ in their visual literacy proficiencies.
7. A high degree of skill is required to interpret ERs that represent *abstract* phenomena.
8. Students that have not been exposed to a variety of ERs, lack the *procedural* skills needed for interpretation; skills, which develop over time.
9. Learners often interpret ERs literally as "the reality" and "the truth", rather than as *representations* of the reality and therefore, are unaware of an ER's limitations.
10. Learners struggle to *translate* between different ERs of the same scientific idea.
11. *No* correlation exists between an increase in the amount of detail on an ER and an increase in understanding. In some cases, excessive ER detail has a negative effect on ER processing. However, there is a correlation between the amount of detail on an ER and the ability to *memorise* the ER.
12. *Colour* aids ER interpretation because it helps learners discriminate between graphical features and to refine ideas. Learners prefer coloured ERs but an overuse of colour can cause misdirection.
13. Difficulties with scientific ERs are often due to a lack of understanding of, as well as an inability to decode the artistic, graphical, or symbolic markings on the ER.
14. Many "universal" conventions used in ERs have shown not only to be idiosyncratic, but also inconsistent *across,* as well as *within,* scientific ERs.
15. Sometimes, ERs with a large aesthetic impact cause learners to be *distracted* from the underlying meaning implied by the ER.
16. Difficulties are enhanced when ERs are poorly *designed.*
17. The fact that experts bring more conceptual knowledge to an ER than novices *cannot* on its own, explain processing differences.

18. When searching ERs, novices and experts employ strategies that are *distinct* from one another because experts and novices have different processing goals.
19. Novices often pay more attention to markings that stand out, and ignore those that are less salient, resulting in processing that is superficial.
20. Novices often interpret ERs *literally*, rather than in relation to underlying conceptual knowledge that is implied by the ER.
21. Experts are able to internalise and organise a large group of markings *at once*. This perceptual chunking process is often absent in novices.
22. Experts' mental models are more *semantically* based, while novices often construct unorganised mental models, based largely on the visuo-spatial features of an ER.
23. Experts organise the knowledge obtained from an ER in a structured, hierarchical and *integrated* manner.
24. A problem facing science students is similar to a "chicken-and-egg" dilemma: to interpret an ER effectively, certain content knowledge is required. But, to acquire the content knowledge, one needs to engage in ER interpretation.
25. Learners often interpret abstract ERs in a *narrative* and story-like manner.
26. As the case with text, English learners read ERs in a left-to-right manner, which hinders the processing of more spatially complex ERs.
27. Some learners engage in *localised reasoning* when reading ERs. In this case, more attention is given to only one area of the ER resulting in a failure to appreciate the holistic nature of the ER.
28. Students find it challenging to generate their *own* ERs of scientific ideas and struggle to use their generated ERs as tools for explanation.
29. When generating their own ERs, students often revert to externalising "accepted" or standardised ERs and revert to *memory* rather than to their own interpretations.
30. Experts *adjust* their generated ERs as the need arises and as the task requires. Novices insert irrelevant detail into their generated ERs, of no direct significance to the task.
31. When learning scientific ideas that are abstract, a *co-evolution* of ER-processing skills and construction of conceptual understanding occurs.
32. Graphic-word ERs that represent concepts in a *spatial* manner decrease the cognitive load placed on the viewer. By arranging graphical features in close proximity to one another, the amount of required *search* and computation is reduced.
33. Static ERs and animated ERs each place *unique* cognitive demands on viewers.

34. Educators have placed a lot of faith in animated ERs as infallible learning tools. This is based on *intuition* alone rather than on any theoretical grounds.
35. Learning with animated ERs does not *always* lead to favourable learning outcomes.
36. Little empirical proof exists to show that animated ERs are *superior* to static ERs for learning. In some instances, both have been shown to be equally beneficial.
37. As with static ERs, when interpreting animated ERs, novices rely heavily on markings that stand out, rather than on the underlying relevance of the markings.
38. Multimedia ERs are meaningless to students who cannot *map* between pictorial and textual representation modes.
39. Multimedia ERs are effective when they are designed appropriately, when the cognitive load placed on the viewer is reduced, when irrelevant information is eradicated, when pictorial and textual elements are in close proximity and when text is presented in a conversational manner.
40. Implications for learning from ERs should be considered in terms of *current* learning models, which imply that meaningful learning is an active rather than passive process.

The following Chapter presents the methods employed in the current thesis to answer the research questions provided in Chapter 1.

3 METHODS

3.1 Introduction

This chapter presents the overall theoretical and methodological framework employed to address the proposed research questions (Chapter 1). It also outlines and discusses the general methodology used to gather data and considers the nature, strengths and limitations of the methods. Details of the methods used in each study comprising the thesis are given in the relevant results Chapters 4-6.

As pointed out in Chapter 1, students' alternative conceptions, misconceptions, pre-conceptions and reasoning difficulties can hinder, and often prevent, beneficial learning and teaching of science (Grayson, 2004; Kuiper, 1994; Hasweh, 1988; Treagust, 1988). One reason for this is that such difficulties tend to be resistant to change (von Aufschnaiter and von Aufschnaiter, 2003; Ausubel, 1968). Another reason is that these difficulties are often *part* of an individual's conceptual make-up and therefore seem completely logical to learners (e.g. Fisher, 1985; Osborne and Wittrock, 1983). Thus to identify and explicitly study these learning difficulties we required an overall theoretical framework in which to operate. Furthermore, we needed to decide on the nature of the methods that can be employed to gather data pertinent to the proposed research objectives (Chapter 1). Moreover, we needed to consider the validity and reliability of the methods chosen for this project. All these issues are addressed in this chapter in sections 3.2, 3.3 and 3.4, respectively.

3.2 Student and course context

The research reported in this thesis was done from 2000 to 2004 at the University of KwaZulu-Natal. A total of 166 second and third year undergraduate biochemistry students' participated in the research. To enter the biochemistry curriculum, which commences at the second year of a science degree, all the students who participated in the study would have had to pass full first-year courses in Chemistry, in Mathematics or Physics and, in one of Biosciences, Zoology or Botany. Therefore, all students choosing to study biochemistry would have entered the second year with a prior knowledge corresponding to these

prerequisite courses. Second-year students who pass the full one-year biochemistry course may choose to major in the subject as part of the third and final year of their science degrees. The students who participated in the study were from diverse educational backgrounds ranging from rural to private high-school environments. Not all participants possessed English as their first language with some students' having English as a second or even third language. In such cases Zulu and/or Xhosa was the first and/or second language of the student. Both males and females represented the group of participants.

The studies investigated students' interpretation of ERs that are used in the teaching and learning of biochemistry. Six ERs were used in the study and constituted multiple representations of the structure of immunoglobulin G (IgG) and its primary interaction with antigen, and fell on an abstract to real continuum. The six different ERs used in the study will be introduced in each relevant results chapter, where applicable. The 166 students who participated in the study consisted of the following general groups. One hundred and thirty of the total participants were second-year biochemistry students who had completed a module on immunology as part of the second year biochemistry course in 2000 and 21 were third-year students who had studied the same course the previous year in 1999. In both years, the immunology module made use of the same course notes, the same prescribed textbooks and the same instructor lectured the module. All of these students responded to written probes in the year 2000. In addition, 10 second-year students and 6 students who had all completed at least one module of biochemistry at the third-year level were interviewed at the end of 2000. A further nine third-year biochemistry majors participated in clinical interviews at the end of 2001.

With regard to the student and course context of this research, an important point is raised. The science of biochemistry has classed a host of immunoglobulin (Ig) molecules including IgA, M, E, D and G. During probing of students' conceptual understanding of antibody structure and interaction with antigen in the current study, *all* participants called upon the structure of *immunoglobulin G* as their basis for describing the term "antibody" to the researcher. IgG is the most basic structure of all antibody molecules in humans and as a result, is the molecule that is used by textbooks and instructors to introduce students to concepts surrounding antibody structure and binding. Indeed, biochemistry textbooks that contain a section on immunology usually begin with a discussion of IgG molecules, before proceeding with more complex antibody structures (e.g. Hames and Hooper, 2000; Stryer,

1995; Lehninger *et al.*, 1993; Mathews and van Holde, 1990). Furthermore, in confirmation of the former, it was found that the antibody diagrams, which participants themselves generated when required, corresponded to the basic structural features of IgG. Therefore, a control variable was set up, in that the author could be certain as to what antibody structure students were expressing during probing of their understanding. Hence, valid and reliable comparative analyses of students' responses could be made against the accepted scientific knowledge of IgG structure and function, which is provided next in section 3.3.1.

The student and course context of the study informed the structure of the theoretical framework employed by the thesis. In the next section, we present the theoretical framework used to frame the research questions (Chapter 1).

3.3 Theoretical framework

The theoretical framework that structured this study is discussed in sections 3.3.1 and 3.3.2, respectively. Firstly, the biochemistry context of the study is outlined with respect to the propositional knowledge represented by multiple external representations of the structure of IgG and its interaction with antigen. Secondly, the science education context of the study is framed by presenting an applicable learning theory that the researcher used as a basis for explaining how individuals learn new knowledge and integrate already existing knowledge.

3.3.1 Biochemistry context

Concepts surrounding antibody structure and its interaction with antigen formed the biochemistry context of this thesis. The nature of visual representation of the propositional (scientific) knowledge that represents these biochemical concepts is discussed in this section. Such knowledge is essential for the studies performed in this thesis.

Biochemistry is a science that is often investigated within the sub-microscopic environment. Since we cannot physically see this environment, scientists use physical and chemical data to construct theories, hypotheses and models in an attempt to explain these abstract phenomena. These constructs in turn, if accepted by the community of biochemists, govern how we subsequently interpret, and reason about, the nature of the sub-microscopic environment and,

therefore, what we include in educational resources (e.g. textbooks and computer software) and teach to students in order to promote their understanding of the subject matter.

For a holistic understanding of biochemistry, one is required to move between macroscopic, microscopic and symbolic models of phenomena (e.g. Pavlinic *et al.*, 2001). In addition, one is required to visualise, and translate between, abstract (e.g. graphical plots), symbolic (e.g. formulae), molecular (e.g. space-filling models) and realistic (e.g. electron micrographs) levels. Thus biochemistry, like chemistry, is a "mix of empirical observation and abstract reasoning", and a variety of external representations or "models of reality" (Hoffmann and Laszlo, 1991) that often consist of different levels of abstraction (e.g. Knight, 2003; Sumfleth and Telgenbüscher, 2001).

Abstract phenomena such as protein molecules are represented in a number of different ways including 2-D ERs, 3-D physical models, and as various computer-generated ERs. These *modes of representation* are intended to assist students to construct mental models of how we currently believe a particular protein molecule looks in reality. For example, current understanding of the structure of immunoglobulin G (IgG) and its interaction with antigen can be reflected by a range of ERs. Possible ERs include electron micrographs showing the general shape of an antibody-hapten complex, crystallographic ERs of antibody fragments, stylised ball-and-stick ERs, or colorimetric indicator systems, all of which assist us in "seeing" antibody-antigen binding. The manner in which the concepts are represented may depend on the pedagogical aim of the ER, on the technology used to generate the ER, or on the particular mode in which the representation is externally generated. Since there is a variety of "models of reality" (Hoffmann and Laszlo, 1991) in biochemistry for depicting knowledge such as the structure of antibody molecules, the *way* scientists/authors represent these phenomena visually will play a role in determining *how* knowledge will be acquired and communicated amongst learners. This will remain so until the currently accepted model is adjusted, modified or discarded.

A selection of ten typical representations available to the community of biochemists for depicting the concept of "antibody" and/or "antibody-antigen binding" is shown in Fig. 3.1 (a-j) below.

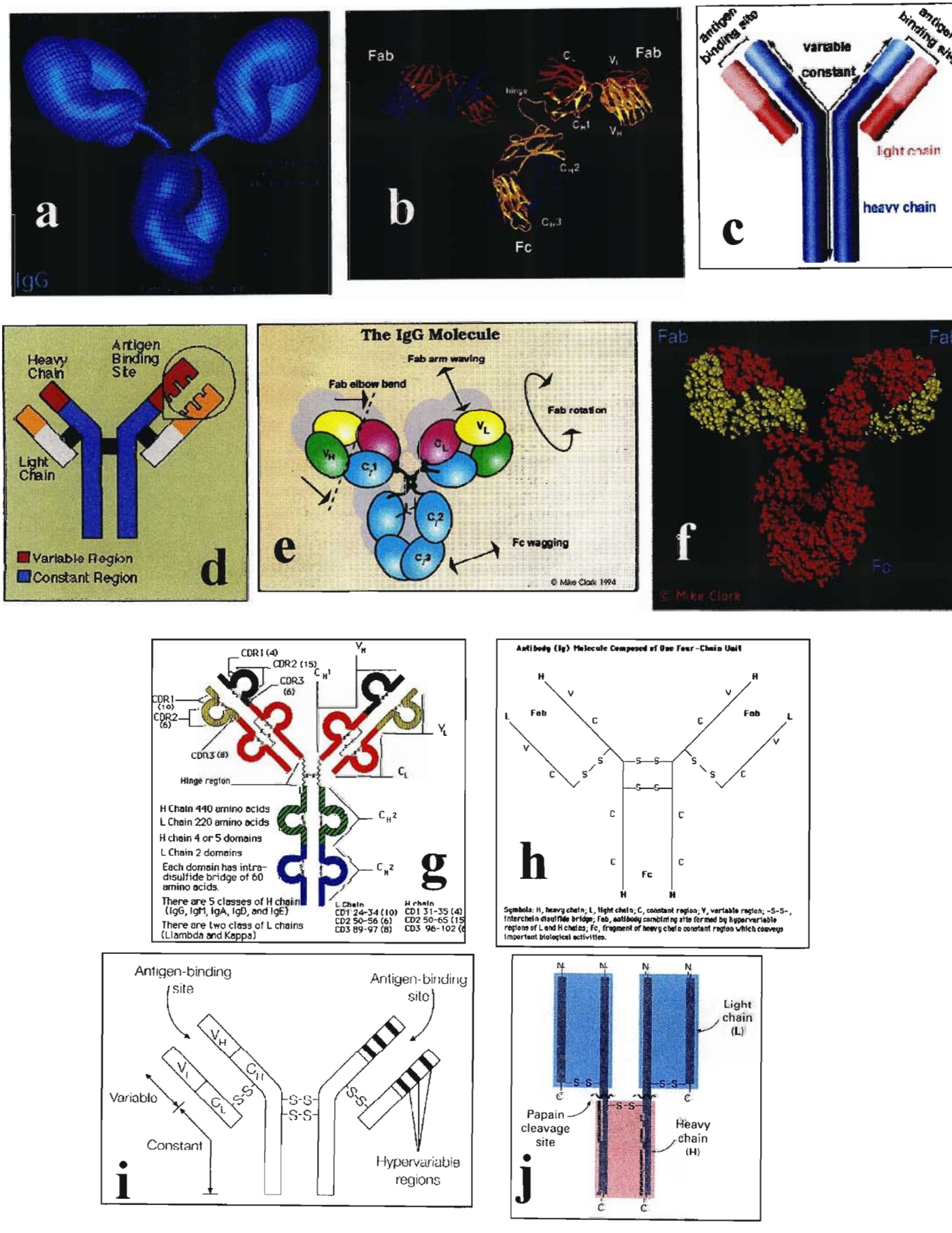


Figure 3.1 Ten examples of ERs (a-j) that depict antibody structure and interaction with antigen

All the ERs (Fig. 3.1) serve as typical examples of the scientific (propositional) knowledge used by scientists, authors and designers to convey the structure of an IgG molecule and its interaction with antigen, the biochemistry context of this thesis. Other ERs that represent the same concept that were used in this thesis as the basis for the reported investigation are discussed in results chapters 4-6. The antibody most familiar to undergraduate students (section 3.2) is that of immunoglobulin G (IgG) (Fig 3.1). The IgG class represented in Fig 3.1 can be further divided into four subclasses, namely, IgG_1, IgG_2, IgG_3 and IgG_4 (Hames and Hooper, 2000), which show only minor differences in structure (Roitt and Delves, 2001; Roitt, 1997). For the purpose of this thesis, the term "antibody" will refer to the structure of IgG unless stated otherwise.

The basic structure of IgG is a four-peptide unit - two identical heavy and two identical light polypeptide chains (e.g. Fig 3.1c, d, g, h, i and j) held together by interchain disulfide bonds (e.g. Fig 3.1d, g, h, i, and j) (Roitt, 1997; Campbell and Smith, 2000). The general shape of an antibody is often described as a "Y" (e.g. Fig. 3.1a, c, d, g, h and i). Each light (L) chain consists of approximately 220 amino acids and each heavy (H) chain of approximately 440 amino acids. Carbohydrate residues are attached to the heavy chains of the molecule, shown in purple on Fig. 3.1f. Due to this structural characteristic, the IgG molecule is often referred to as a glycoprotein. Each light chain and each heavy chain consists of a variable (V) region and a constant (C) region (e.g. Fig. 3.1c, d, h and i) (Hames and Hooper, 2000). The variable regions differ in amino acid composition across all IgG molecules whereas the amino acid composition of the constant regions remains more or less the same. The N-termini of the two heavy and light chains are situated at the variable end and the C-termini of the two heavy and light chains at the constant end (e.g. Fig. 3.1 j) (Hames and Hooper, 2000). Variability in the variable region is largely localised in three hypervariable regions, shown in Fig 3.1i (Hames and Hooper, 2000). The enzyme papain can split the antibody into three fragments (Fig. 3.1j); two identical Fab (Fragment antigen binding) fragments, each with a single and identical antigen binding site and one Fc (Fragment crystallizes) fragment that cannot bind antigen (Hames and Hooper, 2000; Roitt, 1997). The location of the Fab and Fc regions on an antibody molecule is represented in Fig 3.1a, b, e, f and h. In addition to interchain disulfide bonds between light and heavy chains, intrachain disulfide bonds form loops within the light and heavy polypeptide chains. These loops constitute the hypervariability of IgG and are termed complementarity-determining regions (CDRs) (Fig 3.1g). Furthermore, the CDR loops fold to form β-pleated sheet globular domains (Fig 3.1b) (Roitt, 1997). A complete

antibody molecule consists of twelve domains; each light chain folds into two domains, one domain in the variable region and another in the constant region (e.g Fig 3.1b and g). Each heavy chain folds into four domains, one domain in the variable region and three in the constant region (e.g. Fig 3.1b, e, g) (Hames and Hooper, 2000). Each antibody molecule has two antigen binding sites (Fig. 3.1). The variable domain of a light chain and variable domain of a heavy chain each form one of two antigen-binding sites (e.g. Fig. 3.1c, d and i). Therefore, the antibody molecule is *bivalent* and can bind a maximum of two antigen molecules, one at each antibody binding-site (Hames and Hooper, 2000).

An antigen is a molecule that may interact with the hypervariable regions (Fig. 3.1g) of the heavy and light chains (Campbell and Smith, 2000). The antigen binding sites are explicitly indicated on Fig 3.1c, d and i. When an antigen binds to an antibody (primary interaction), a cellular immune response may be initiated, which results in the degradation of the antigen molecule. The part of the antigen that makes contact with the antibody is termed the epitope. The parts of the hypervariable regions of the antibody that make contact with the epitope are termed the paratope (Roitt and Delves, 2001). Primary interaction between paratope and epitope is specific, spatially complementary and non-covalent (Roitt, 1997).

In addition to the structural characteristics of IgG, a degree of flexibility is associated with the IgG molecule (Roitt, 1997; Brekke *et al.*, 1995). This is characterised by a hinge region located at the intersection of the first (C_H1) and second (C_H2) domains of the constant portion of the heavy chain (e.g. Fig. 3.1b and g) (Brekke *et al.*, 1995). Potential flexibility (e.g. Fig 3.1e) of the IgG molecule may include 'waving', 'rotation' or 'elbow bending' of the Fab arms and/or 'wagging' of the Fc region (Roitt, 1997; Brekke *et al.*, 1995). The flexibility of the antibody molecule allows for optimal binding to antigen and other effectors of the human immune system (Roitt and Delves, 2001).

As pointed out earlier, the IgG molecule is often presented and described in textbooks as having a characteristic "Y" shape. However, the molecule is sometimes represented as a "T" shape (Fig. 3.1b and f) or in an "upright" conformation (Fig. 3.1j) in ERs. The differences in presentation may depend largely on the original position the molecule was in when it was captured in time during crystallographic analysis (e.g. Silverton *et al.*, 1977), or, on how authors or ER designers decide to depict it. As mentioned, the antibody molecule is not a static entity in *vivo*, the "arms" of the "Y" are in constant motion, as is the Fc "tail" due to the

flexibility of the hinge region (Martin, pers. comm.). Due to this flexibility, only a few researchers (e.g. Harris *et al.*, 1997) have managed to solve the structure of an entire intact antibody, including the hinge region. Therefore, the literature often depicts the molecule as a T-shape (e.g. Silverton *et al.*, 1977; Harris *et al.*, 1997), Y-shape (e.g. Fig 3.1c) or as an upright shape (e.g. Fig. 3.1j). It is common practice for separate fragments of the immunoglobulin G molecule to be solved individually first, and then, for the researchers to "put the molecule together" to represent an entire IgG molecule (Martin, pers. comm.). Similarly, it is common for one group of researchers to solve say the Fab portion, and then use previous data from other studies to represent the entire molecule (e.g. Davies and Padlan, 1990). Due to these laboratory methods, many ERs of IgG are represented in textbooks as containing a deleted hinge region.

In further elaboration of the biochemistry context of this thesis, crystallographic studies have shown that certain features of IgG (e.g. two heavy and two light chains, constant and variable regions, twelve structural domains and bivalency) are generic to all IgG structures. This finding remains constant even if the results from crystallography (e.g. amino acid composition) may have varied slightly amongst studies (e.g. Janeway, Martin, Landry, Pincus and Smith, pers. comm.). This is widely accepted amongst workers in the field and is supported by the following extracts from correspondence with some prominent workers in the field of bioinformatics and immunology:

> Researcher: I've noticed that almost all of the diagrams I've encountered; especially in textbooks, are based on the x-ray crystallographic study of Silverton *et al.* (1977). My question is: is this structure still the basis for diagram design in current textbooks? I've seen adaptations of the former in Stryer (1995), Lehninger (1993) and Campell (2000) to name a few. I've also seen that with recent studies, the schematic, line-type representations of the IgG antibody have remained constant, even when you compare them to diagrams in textbooks of the late seventies and early eighties.
>
> Martin: X-ray crystallography is a method for viewing a protein structure. This can be done at different resolutions (levels of detail), but essentially (providing no major mistakes were made in solving the structure - which is rare but has been known to happen), then a structure from 30 years ago should be just a good as one solved now.
>
> Janeway: Yes, the original structure is from Sliverton *et al.*, as you surmise. Until a new technique with higher resolution comes along, we will be stuck with this one.
>
> Landry: It may no longer be the basis because there are many new crystal structures available; however the relevant features are the same in all IgG structures.

From the extracts above, it can be deduced that any collective pool of antibody ERs will share a high degree of structural commonality. If this is the case, then ERs that convey this information will certainly contain common graphical features as well. This is clear in Fig 3.1 in which all the presented ERs share at least one visual feature generic to all antibody structures. For instance, all the ERs show IgG as a four-chain (two heavy and two light) unit and each show two possible binding areas for antigen. The ERs may also differ in other respects. For example, Fig. 3.1b, e and g all show the twelve structural domains of IgG where the other ERs (Fig. 3.1) do not and, Fig 3.1 a, b, e and f all give some idea of the volume that is occupied by the molecular components constituting the overall shape of the molecule, where the others do not. Visual representation of whichever structural feature of IgG in an ER is a function of a biochemist's analysis or a function of what the textbook author or ER designer wishes to make salient for whatever instructional purpose. Fig 3.1 serves as an example of ERs that are included as part of lecture notes, textbooks, tutorial packages, teaching aids, learning aids or as part of research papers. It is evident that the ERs (Fig 3.1) are diverse in terms of their visual representation and contain varying degrees of graphical and symbolic information.

As pointed out in Chapter 2, ERs that contain symbolic information have to be interpreted according to a certain convention for learners to construct a scientifically acceptable mental model (e.g. Kosslyn, 1989). In other words, the ERs have to be interpreted in the same way by different people on each occasion, if any agreement between interpretations is going to be established. Even though there are diverse ERs available to teachers and learners (e.g. Fig 3.1), there seems to be little systematic and standardised means for defining the visual ER "conventions" used to represent antibody structure in the science of biochemistry. As has been shown for the learning of science in general, understanding an ER requires an understanding of the conventions used (e.g. Henderson, 1999). Sometimes, many of the conventions used are not universal or consistent across ERs within the same class. Given that the nature of an ER is often a function of what the designer intends to convey or the educational objective of the ER (e.g. Petre and Green, 1993; Fleming, 1967), it would be fair to suggest that many of the depictions presented in Fig 3.1 consist of "conventions" that are rather idiosyncratic in nature. Nevertheless, some of these "conventions" have become accepted as "universal conventions" in their own right. As part of further exposing the biochemistry context of this thesis, these "conventions" shall be discussed next.

Hoffmann and Laszlo (1991) have discussed the issue of idiosyncratic graphical features in ERs in a chemistry context. These authors say that often convention and realism are mixed "in the most innocent manner" and that the representation of a chemical structure is "ideology-laden". Therefore, to represent them graphically, a "reunification of the theoretical and the experimental" is required. By presenting the reader with some of the structural representations available for the compound *camphor*, from symbolic to ball-and-stick and space-filling types, Hoffmann and Laszlo (1991) raise the question, "Which of the representations is right? Which *is* the molecule?" In an answer to the question posed, they suggest that, "all are, and none is". The point exemplified by their discussion is to affirm that each representation is just a model, useful in certain instances but not in others. Like in chemistry, representation in biochemistry is similar. There are many ERs available to depict a single phenomenon such as antibody structure, each one serving its own purpose. In this regard, two probable examples of accepted and "universal" conventions in biochemistry are the space-filling model (e.g. Fig. 3.1f) to depict atomic and molecular volume (e.g. McKee and McKee, 1996; Amit *et al.*, 1986) and the "ribbon" representation (e.g. Fig. 3.1b) to depict folding of polypeptide chains. It would be fair to suggest that the space-filling (Fig. 3.1f) and ribbon (Fig 3.1b) conventions have become standardised features of modern ERs used in biochemistry (e.g. Richardson and Richardson, 2002). However, in lieu of other diverse and non-standardised "conventions" used to represent concepts in biochemical ERs (e.g. Fig. 3.1a and g), the author posed a question to the Chairman of the Nomenclature Committee of the IUBMB to obtain clarity on this issue. The exchange that occurred was as follows (Cammack, pers. comm.):

> Researcher: ...Part of my work has been concerned with analysing textbooks, web pages, course notes, teaching, and learning aids that incorporate the representation of immunoglobulin molecules. Is there a standardised or accepted format for representing biochemical structures diagrammatically, other than the normal symbolic notations? I know that physics have certain rules for drawing vectors, pulley-systems, momentum diagrams etc. From analysing the diagrammatic representations used in biochemistry, things like ball-and-stick models, space-filling models, ribbon diagrams, backbone models etc. form the basis for representing protein structures. Are there any rules or laws stipulating how structures should be drawn, especially when authors depict stylised representations that are sometimes idiosyncratic in nature?
>
> Chairman: ...Dr. Moss has forwarded your message to me. He did not know of any conventions for the representations of molecular structures in biochemistry, and I have not heard of them either. There are, as you say, many different representations, depending on the different types of software used to generate them, based on two- and three-dimensional formats. The type of representation, and the aspect of the molecule in the picture, are usually chosen on the basis of the type of information that the diagram is intended to convey. Two-dimensional Chemdraw-type programs are used for chemical formulae and mechanisms. Programs such as Rasmol and molscript provide a sort of standard representation for three-dimensional structures. Ball-and-stick or wireframe are used for chain conformations; spacefill

> for looking at the overall dimensions or surface of proteins; and there are many more. The journals such as Structure or Nature Structural Biology have their own conventions, but these relate mostly to the file transfer formats. So I cannot give any firm advice. Are there cases that you know of, where more consistent representations of structures would be helpful? We are always interested to hear of such cases, and if necessary take advice.

Based on the above, there appear to be no formal rules or standards that govern the visual representation of protein structure in biochemistry, let alone the structure of IgG molecules. Given that the molecular features of many ERs would be recognised by trained biochemist instantly, it appears that many idiosyncratic "conventions" (e.g. Fig 3.1) used in biochemical ERs are not conventions at all. As Cammack (pers. comm.) states above, "the type of representation, and the aspect of the molecule depicted in the ER is usually chosen on the basis of the type of information that the diagram is intended to convey". Even though this is a clear statement, it appears that this process is put into practice automatically by teachers, textbooks authors and ER designers without any serious consideration of the effects on student learning. Surely then, should this not cause potential problems for learners who are expected to interpret these ERs without fault? This is a major question addressed in the present thesis.

In summary, in section 3.2.1 we have presented the propositional (scientific) knowledge constituting concepts surrounding antibody structure and interaction with antigen. Describing the nature of this knowledge is crucial to the present study that deals with students' interpretations of ERs that represent these concepts. The diverse and sometimes idiosyncratic nature of visual representation in biochemistry has also been emphasised. In the next section, we consider the science education context of the theoretical framework employed in this study.

3.3.2 Science Education context

Much of the progress made in understanding how individuals learn can be attributed to the Swiss psychologist, Jean Piaget. In his theory of cognitive development (Piaget, 1952), he proposed that all individuals pass through four distinct stages of development (e.g. Coon, 2001; Bukatko and Daehler, 1992). During the *sensorimotor stage* (0-2 years), the child's development is characterised by non-verbal manifestations while s/he begins to make connections between sensory and motor inputs. The *preoperational stage* (2-7 years) sees the

child beginning to make use of language and other symbolic inferences. At this stage, the child's thought processes remain egocentric. However, when the child is able to make use of concepts, such as conservation of mass and volume, and these concepts remain concrete, then the child has passed into the *concrete operational stage* (7-11 years). A child's ability to engage in abstract and theoretical thinking marks the final stage of cognitive development, the *formal operations stage* (11 years and beyond). After further experience and construction of knowledge in years to come, this stage allows the individual to engage in deductive, inductive or hypothetical reasoning processes (e.g. Coon, 2001; Bukatko and Daehler, 1992), the stage expected of the students who participated in the present study.

Encompassed within Piaget's theory is the postulate that cognitive development occurs through two general processes. Firstly, *assimilation* describes the use of existing knowledge (schemes) in a novel situation, or the process of integrating new information into existing knowledge. *Accommodation* is concerned with the process of adjusting one's existing knowledge in a novel situation (e.g. Coon, 2001). Through assimilation and accommodation, an individual reaches a greater *equilibrium*, which is described as the "balance" between his/her knowledge structures (e.g. Bukatko and Daehler, 1992). These two processes have become the cornerstones for a popular cognitive theory that describes how it is that people are able to "learn" and, serve as one component of the theoretical framework implemented in this thesis.

In a development of Piaget's theory for cognitive psychology, but applied specifically to an educational context, Bruner (1986, 1960) proposed an epistemology to describe how individuals "learn" new information and "use" existing information. In this regard, Bruner (1986, 1960) has suggested that the process of learning should be viewed as an *active*, rather than a *passive* process. It is this *active* process that is responsible for the *construction* of new concepts that are based on already existing knowledge and experience. According to Bruner (1986), the learner uses his/her *cognitive structure*, which consists of sets of unique schema and mental models to select and transform knowledge. Initiated by Piaget, the above viewpoints, which form the basis of our thinking in this study, have become known as the post-modern learning theory of *constructivism* (e.g. Gall *et al.*, 1996). Von Glasersfeld (2003, 1989, 1983), perhaps the most respected constructivist in modern times, suggests that knowledge in the world cannot merely be transferred from the instructor to the learner. Instead, each individual's knowledge exists due to the unique organisation of his or her own

conceptual structure. Constructivist-learning theory suggests therefore, that the learner has to *assimilate* (e.g. Bukatko and Daehler, 1992; Dean and Enemoh, 1983) or *accommodate* (e.g. Ward and Wandersee, 2002; Bukatko and Daehler, 1992) the information that is perceived. Therefore, generating conceptual understanding can never be a passive process but rather, is a unique *product* (von Glasersfeld, 1989) of a learner's conceptual organisation, experiences and social reality (e.g. Gall *et al.*, 1996).

In relation to the sentiments expressed by the constructivist movement, Wittrock (1974) has proposed a *generative* theory of learning. The theory suggests that children develop their own scientific ideas, based on everyday experience, even before they are formally "taught" science. It is these previously constructed naïve ideas and views that affect the way individuals learn *new* scientific concepts and therefore, have a direct bearing on the processes of accommodation and assimilation described above. The theory of generative learning (Osborne and Wittrock, 1983) postulates that the brain actively constructs unique interpretations rather than passively absorbs information (e.g. von Glasersveld, 2003, 1983; Anderson *et al.*, 2000). It follows, according to the theory, that the process of *generation* is concerned with generating *meaningful* learning through comprehension that, "organizes the information selected from the experience in a way that makes sense to us, that fits our logic, or real world experiences, or both" (Osborne and Wittrock, p. 493). Therefore, according to the theory, learning science is seen as a creative process where new ides have to be integrated into already existing ways of reasoning and existing knowledge (e.g. Osborne and Wittrock, 1983). With reference to constructivism and generative learning, Mayer (e.g. 2003, 1993) has identified four cognitive processes that drive meaningful learning. The four processes are the *selection* of relevant information, the *organisation* of the information into a coherent structure, the *integration* of the information into existing knowledge and finally, the *encoding* of the information into long-term memory.

According to the constructivist movement, each individual constructs knowledge that is unique and based on an individual's prior knowledge, experiences and social reality. Therefore, during learning, since the construction of new knowledge is a unique product for each individual, a particular individual could construct knowledge that that does not correlate with currently accepted propositional (scientific) knowledge. As a result, this newly constructed knowledge may take the form of *alternative conceptions* (e.g. Driver, 1989), which are conceptual structures that are not consistent with current scientific worldviews. In

addition, a student may show particular *reasoning difficulties* (e.g. Arons, 1990) when employing their constructed knowledge in different scientific contexts (e.g. Grayson *et al.*, 2001; Cohen *et al.*, 1983). Under the banner of constructivism, a large volume of research has identified students' alternative conceptions and learning difficulties in science. Examples of such studies can be found in physics (Harrison *et al.*, 1999; Pfundt and Duit, 1994), chemistry (Birk and Kurtz, 1999; Boo, 1998; Garnett *et al.*, 1995), biology (Flores *et al.*, 2003; Sanders, 1993; Lazarowitz and Penso, 1992; Boyes and Stanisstreet, 1991; Griffiths and Grant, 1985), and to a lesser degree, astronomy (Stahly *et al.*, 1999; Jones and Lynch, 1987). As pointed out in Chapter 1, diagnosing students' difficulties with the learning of biochemistry has received very limited attention (e.g. Anderson and Grayson, 1994; Fisher, 1985).

Based on the above examples of research conducted within a constructivist framework, the author argues that a constructivist epistemology would also serve as a feasible research framework to identify students' difficulties with the interpretation of ERs used in the teaching and learning of biochemistry. In this regard, Treagust *et al.* (2002) suggest in terms of the constructivist paradigm that, "learning in science requires students to take ownership of an idea or concept, reconstruct it, internalise it and be able to communicate it to others." (p. 367). In an extension of this sentiment in terms of the current study, Mayer (2003) and Kosslyn (1985) suggest that individuals learn from ERs via an active process characterised by them making sense of, and integrating the external information themselves. This process is in contrast with otherwise traditional views that see learners internalising the external information passively and directly (e.g. Ward and Wandersee, 2002; Gall *et al.*, 1996; Grayson, 1995). Thus, when interpreting ERs, it can be expected that each individual will construct a unique mental model of the scientific phenomenon that is represented by a particular ER (e.g. Lohse *et al.*, 1991). In order for learners to make sense of the visual information represented by the ER, the information has to be internally processed through learners' "theoretical lenses" (Stylianidou *et al.*, 2002, p. 257). Hence, for learners to construct meaningful concepts from ERs as well as to be able to reason with them, information has to be processed through already existing knowledge (e.g. Ward and Wandersee, 2002) and experiences. The above sentiments form the basis for the science education context of the theoretical framework employed in this thesis.

In summary, a constructivist philosophy has been introduced and discussed in order to provide a theoretical explanation of how students' are thought to *learn* new knowledge, and integrate already existing information. The feasibility of such a theoretical framework for investigating students' interpretation and processing of ERs in biochemistry has been argued to be favourable. Following the above outline of a suitable theoretical framework from which to base the research questions (Chapter 1), in the next section, we show how this theoretical framework, described in section 3.3, informs the methodological framework employed in this thesis.

3.4 Methodological framework

Now that a theoretical foundation, based on the nature of representation of antibody molecules in biochemistry and a constructivist epistemology has been provided for this thesis, the following questions need to be posed. What methodological framework would be compatible with the theoretical framework described in section 3.3? What are the most appropriate methods that can be used as instruments for the collection of data on students' interpretation of ERs of antibody structure and interaction with antigen, and why (i.e. what key research findings support their validity)? Finally, what are the limitations of such methods?

The methodological framework that was employed in this study is discussed in sections 3.4.1, 3.4.2, 3.4.3 and 3.4.4, respectively. Firstly, the general methodological approach that was adopted within the overall methodological framework of this study is outlined. Secondly, the research instruments used to collect data are presented. This includes a description of the selection of the data-gathering instruments based on similar methods used by other workers in the field. The aim of this is to demonstrate the acceptability of the chosen instruments amongst the community of science educators. Thirdly, methods for analysing the data are discussed and finally, the validity and reliability of the methods are scrutinised.

3.4.1 General methodological approach

According to Gall *et al.* (1996), educational researchers who subscribe to the constructivist learning theory (section 3.3.2) are of the opinion that methods that are strictly analytical are

not appropriate for measuring and understanding the unique interpretations that individuals construct. This position, known as postpositivism, is a direct reaction to the positivistic epistemology, which suggests that, "physical and social reality is independent of those who observe it, and that observations of this reality, if unbiased, constitute scientific knowledge" (Gall *et al.*, 1996, p. 18). Accordingly, as informed by the assumptions contained in the theoretical framework discussed in section 3.3, the methodological approach in the current study is based on the notion that human behaviour cannot be studied completely objectively and separately from any social context (e.g. Lincoln and Guba, 1985). This is because individuals' interpretations do not remain constant and human behaviour shows complex interactions (e.g. Gall *et al.*, 1996). Therefore, it was necessary to adopt a qualitative and interpretive approach to address the research questions (Chapter 1) of this thesis, rather than a quantitative approach, which would have been employed by positivistic investigators (e.g. Gall *et al.*, 1996; Rosnow and Rosenthal, 1996). Qualitative methods, such as the ones adopted in this study, are concerned with collecting verbal and observational data from participants and then subjecting the data to analytic induction (e.g. Mouton, 2001), rather than subjecting data to strict statistical treatments for the purpose of making generalisable deductions (e.g. Anderson and Arsenault, 1998; Rosnow and Rosenthal, 1996).

In addition to the above, the qualitative approach adopted in this study was informed by the scientific method (e.g. Anderson and Arsenault, 1998). In this regard, and according to Rosnow and Rosenthal (1996, p. 6), the scientific method, "... is not synonymous with any single, fixed procedure; it is instead a philosophical outlook as much as an evolving collection of tools and techniques. This outlook is primarily characterized by empirical reasoning, which in turn encompasses... quantitative as well as qualitative procedures." In terms of the former sentiment, pursuit of the scientific method in the current study was in no way compromised by the fact that the study was qualitative in nature. Instead, all the qualitative methods that were employed in the current study were *empirical* in nature insofar as they were concerned with the observation and measurement (e.g. Rosnow and Rosenthal, 1996) of particular human behaviours, just as any quantitative study may endeavour to do. In addition, an empirical approach was sustained in the current project by the systematic and purposeful investigation (e.g. McMillan and Schumacher, 1993) of the research questions (Chapter 1). Furthermore, related to the engagement of the scientific method in the current study, the methods and subsequent analysis of the data was characterised by an *evolving dynamic* (Anderson and Arsenault, 1998, p. 38), a situation where new research questions often arose

when the study was already far in progress. To empirically address new research questions, which emerged naturally, the current study involved "...an evolving collection of tools and techniques" (Rosnow and Rosenthal, 1996, p. 6). In this regard, the development of novel methods was sometimes necessary for pursuing new research questions encountered during the study (Chapter 1).

As part of the qualitative design, the methods employed in the current research were naturalistic and were concerned with the human as instrument (e.g. Lincoln and Guba, 1985, p. 39). In this regard, student data was collected through verbal (oral and written) means, pictorial means (students' own diagrams) and by observing particular student behaviours (e.g. student gestures related to ER interpretation and students' generation and modification of their own diagrams during ER interpretation). In addition to being of a qualitative and naturalistic design, the term *interpretive research* is often used to describe studies of this nature (e.g. Mouton, 2001; Gall *et al.*, 1996). These research designs, as the one reported in the current study, place less emphasis on strict experimental and laboratory-type conditions (e.g. Anderson and Arsenault, 1998; Lincoln and Guba, 1985) and are more concerned with understanding the meanings that individuals (or a group of individuals) create in a particular situation (e.g. Gall *et al.*, 1996). As a result, the aim of this approach is to *discover* the data (e.g. Gall *et al.*, 1996) by studying the meanings that individual's construct within a context, and to make holistic observations within that context (e.g. Lincoln and Guba, 1985). As part of this inductive approach, it is possible to reach certain generalisations about a group of individuals within the same study context (e.g. Verma and Mallick, 1999). In other words, if the researcher observes that similar patterns of observation emerge during the study, it may be possible to make generalisable observations about a group of individuals in the same context.

Qualitative designs used in educational research such as this, often employ multi-method approaches (e.g. Anderson and Arsenault, 1998) to address research questions (Chapter 1). In addition, it is well accepted that most methods used have at least some tenets in common with other methods (e.g. Verma and Mallick, 1999). In the present study, all methods had some commonality with each other but varied according to the objectives of the study and the manner in which information was gathered from students (e.g. McMillan and Schumacher, 1993). The methods used to generate data in this study were characterised by rigorous and exhaustive data analysis (see section 3.4.3 below) (e.g. McMillan and Schumacher, 1993). In line with an interpretive and naturalistic approach, emphasis was placed on discovering

generalisations from the data within a specific context and then to explain the possible sources of these phenomena (Bell, 1999; Verma and Mallick, 1999). This approach is in contrast with quantitative approaches that use preconceived determinants to analyse data and then use statistical analysis to generalise the findings to a population (e.g. Gall *et al.*, 1996; Rosnow and Rosenthal, 1996).

In addition to the above approach, some of the methodology used in this work was also *descriptive* in design (e.g. Anderson and Arsenault, 1998; McMillan and Scumacher, 1993) since it sometimes, in part, aimed to supply quantitative (in addition to qualitative) descriptions of observations that were made. A descriptive approach to the work was pursued by *describing* what observations were being made and how the observations were related to one another (e.g. Gall *et al.*, 1996; Rosnow and Rosenthal, 1996). In this instance, our qualitative research design sometimes included a degree of numerical measurement (e.g. Verma and Mallick, 1999) in that the incidence of particular student difficulties with the interpretation of ERs was often calculated. In so doing, in addition to obtaining and describing the verbal, pictorial and observational data that emerged from the data, patterns that emerged from the data were sometimes *described* by the calculation of such incidences.

The above description of the general methodological approach underpinning the present project serves as an introduction to the types of data-gathering methods employed in this work. These instruments are discussed in the next section.

3.4.2 Data collection instruments

Three major data-gathering instruments were used to address the research questions (Chapter 1) namely, written responses, interviews and student generated diagrams. Before outlining the data-gathering instruments that were employed, the nature of such methods used to gather data on students' interpretation of ERs in the current study is discussed.

3.4.2.1 Nature of the methods used to collect data on students' interpretation of ERs

Research on learners' interpretation of *linguistic* representations in science education (e.g. text and sentences) is well established. Methods for measuring learners' processing of textual representations include a variety of reliable and standardised test batteries that are easily assessed (e.g. Van Dusen, 1999; Denis, 1989). In the field of ER research however, not many *systematic* research tools are available to researchers for studying learners' interpretation of ERs in science and even fewer methods for analysing the data (e.g. Lewalter, 2003; Henderson, 1999; Lowe, 1993a). Nevertheless, in recent years, together with the development of theories explaining the interpretation of ERs (e.g. Mayer, 2003; Larkin and Simon, 1987), the field of ER research has developed various methods that have proven useful for investigating students' interpretation of scientific ERs (e.g. Lowe, 2003; Blackwell *et al.*, 2001; Anderson and Armen, 1998). On this note, and with respect to the methods employed in this project, four general guiding principles were considered when designing instruments with which to collect data on students' interpretation of scientific ERs.

Firstly, Lowe (1993a) suggests that methods aimed at understanding learners' interpretation of ERs in science should investigate both a *product* component, concerned with the *results* obtained from learners' interpretation of ERs; and a *process* component, concerned with isolating the *cognitive strategies* that learners use when interpreting ERs. By following this guiding principle in the current study, the methods aimed to first diagnose students' conceptual and reasoning difficulties with the interpretation of ERs and then aimed to understand the cognitive processes responsible for the difficulties.

Secondly, even though some researchers (e.g. Lowe, 1994b, 1993a) have stated that studying ERs which contain textual adjuncts makes it difficult to isolate which representational mode (picture or text) is more involved in the construction of mental representations, other researchers (e.g. van Dusen *et al.*, 1999; Mayer *et al.*, 1995; Fry, 1981) have found it extremely difficult to study visual processing *divorced* from verbal processing. This issue is compounded by dual-coding theory (Mayer and Sims, 1994), which postulates that mental model construction is a result of the *integration* of verbal *and* pictorial modes. Nevertheless, some researchers have endeavoured to study ERs as being divorced from text with fruitful

outcomes. For example, Lowe (1993a) argues that his work with weather map ERs (Chapter 2) allows them to be studied in isolation because the ERs do not depend on any textual adjuncts to convey meaning, and are said to "stand alone". However, other ER researchers (e.g. Mayer *et al.*, 1995; Winn and Solomon, 1993; Glenberg and McDaniel, 1992; Holliday *et al.*, 1977) have used combinations of both modes to investigate ER processing. In these combinations, the pictorial component of the ER is present in a much larger proportion than the textual component, something also common to paper-based and computer-based ERs in biochemistry and, common to the ERs studied in the current project. Theoretical implications of dual-coding theory (section 2.4) served as a further guiding principle that was followed by the current study when selecting appropriate methods for addressing the research questions (Chapter 1).

Thirdly, much of the research data on the mental representation of ERs has been gathered verbally or through written responses (e.g. Levie and Lentz, 1982). In this regard, Lowe (1993a) says that it is unsuitable to collect such data *solely* in textual or verbal format, because students can create distortions when expressing their mental interpretations through verbal outputs alone. Instead, Lowe (e.g. 2003, 1993a) has called for further means with which to collect data. These methods should also include techniques such as getting students to physically manipulate ER information and to generate their own ERs. Consequently, in the present thesis this guiding principle was responded to by employing other methods of data output when gathering information on students' interpretation of ERs, including "think-aloud" tasks, student-generated diagrams (SGDs) and observing other tacit behaviours.

Fourthly, ER research literature suggests that researchers should ensure that the validity and reliability of the data-gathering methods is of the highest degree possible. For instance, Lowe (1993a) says that when designing data-gathering instruments, one should ensure that the data obtained actually embodies students' mental representations and isn't just an artefact of the methodology. He thus highlights the significance of data analysis in such studies and emphasises that, because mental representations cannot be observed *directly*, researchers have to be careful when formulating their findings. This opinion has been supported by Sanders (pers. comm.) who has pointed out that ER researchers should take care to ensure that their instruments are measuring what they are designed to measure and that the data obtained corresponds to what is being searched for. This guiding principle corresponding to issues

surrounding validity and reliability of the methods employed in the current study is addressed in section 3.4.4.

As stated previously in section 3.4.1, due to the qualitative and interpretive approach adopted by this study, methods often displayed a degree of overlap and were thus, *mixed* in design (e.g. Kozma, 2003; Verma and Mallick, 1999). Therefore, the three methods that were employed in the project were frequently used in conjunction with one-another with more emphasis being placed on one approach than another, depending on the particular study in question. Consequently, in line with the *evolving dynamic* (e.g. Anderson and Arsenault, 1998, p. 38) adopted by this work, the methods employed depended on the results obtained and on any additional research objectives that emerged during the study (e.g. McMillan and Schumacher, 1993). In the next section, in addition to describing the three data-gathering instruments and acknowledging the afore-mentioned guidelines employed in the current study, descriptions of similar methods used by other workers in the field are also given attention. By doing so, the author will motivate for the selection and acceptability of the instruments used in the project to the community of science educators.

3.4.2.2 Written instruments

Gathering written verbal outputs is one way in which the mental representation and processing of ERs can be investigated (e.g. Lowe, 1993a). Studies in the field often report the use of written instruments (or probes) that are "open-ended" or "free response" in nature (e.g. Noh and Scharmann, 1997). These free response instruments allow the learner to write "what comes to mind" without being forced into a particular way of thinking (e.g. Grayson *et al.*, 2001). For example, Stylianidou *et al.* (2002) have used this approach to investigate students' interpretation of energy ERs and included the following free-response items: "What do you notice first about this picture?" and, "What do you have to do or think about to make sense of this picture?" Similarly, Schollum (1983) has used questions such as, "When you see a diagram like this what does it mean to you?" to probe students ideas on scientific ERs of food chains, matter and gravity. Written free response techniques such as these were also utilised in the current study.

Even though the use of students' drawings as a way to gather data will be discussed shortly in section 3.4.2.4, this method of data collection is often attached to the free response probing technique described above. For example, Peña and Quílez (2001) investigated students' interpretation of ERs with the following free-response instruments: "Make a diagram of the location of the Sun-Earth-Moon indicating their relative movements in such a way that the phases of the moon are clearly laid out" (p. 1127) and, "You...call a friend who is in a spaceship. You tell him to pop his head out of the window so that he can see the beautiful full moon at the same time as you. But he answers that what he sees is a beautiful moon in its final quarter. Do you think that is possible? Justify you answer with the help of drawings" (p. 1127). The use of students drawings attached to free response probing was used in the current investigation.

In naturalistic research designs (e.g. Gall *et al.*, 1996; Lincoln and Guba, 1985) such as the one reported here, after obtaining free-response data, researches often progress to written instruments that focus more specifically on learners' interpretations that emerged during free response. The design of such questions is informed by the data obtained from free-response probes, where specific patterns that emerge are probed further, in a more purposeful manner (e.g. Grayson *et al.*, 2001). For example, du Plessis *et al.* (2003), Hull (2002) and Treagust (1988) have all used such focused written probes to obtain information and a similar process for obtaining students' responses was utilised in this project.

3.4.2.3 Clinical interviews

According to Posner and Gertzog (1982), clinical interviews have the general objective of gathering information about the nature and extent of a person's cognitive structure and knowledge about a certain idea. A further aim of a clinical interview is to identify how an individual's conceptions are related to one-another (e.g. White and Gunstone, 1992). The *clinical* element was born out of Piaget's approach (e.g. Bukatko and Daehler, 1992) where, while the learner speaks *freely*, the interviewer probes further where s/he thinks deeper information, relating to the concept of interest, resides. Through further probing, the clinical method is aimed at delving into an individual's cognitive structure to get even deeper information to emerge (e.g. Posner and Gertzog, 1982). Although clinical interviews mainly gather verbal responses, they may also include diagram-generating tasks (e.g. Beilfuss *et al.*,

2004). Usually, verbal outputs obtained during interviews are audiotaped and transcribed verbatim (e.g. Ametller and Pintó, 2002; Simonneaux, 2000). Modern times have also seen clinical interviews being videotaped (e.g. Pavlinic *et al.*, 2001; Sumfleth and Telgenbüscher, 2001). In this case, researchers can make use of other observational methods such as the analysis of tacit gestures like "pointing" and "indicating" and the observing of further diagram-related behaviours such as learners' modification or adjustment of their drawings (e.g. Sumfleth and Telgenbüscher, 2001; Kindfield, 1993/1994; Lowe, 1993a). Examples of data sources analysed during clinical interviews include electronic transcripts, videotapes and observation sheets (e.g. Pavlinic *et al.*, 2001). All of the above clinical interviewing techniques were used in the present project.

The following are selected examples of other studies in which data on learners' interpretation of scientific ERs was collected, thus providing a strong motivation that such methods are applicable to the current project. Novick and Nussbaum (1978) used Piagetian type interviews to obtain data on students' understanding of ERs of matter. Information was obtained through structured questions as well as students' drawings generated during the interviews. The authors suggested that a probing interview procedure allowed for a deep and thorough investigation of students' conceptual knowledge. In another example, Ametller and Pintó (2002) used clinical interviews to identify students' difficulties with ERs representing energy. Their interview protocol consisted of general and specific questions, depending on the nature of the responses. The start of each of their interviews contained the same question, "If you found this image in a textbook, how would you interpret it, what does it suggest to you?". Furthermore, Sumfleth and Telgenbüscher (2001) conducted semi-structured interviews to evaluate students' interpretation of ERs in chemistry. The process consisted of four parts, a prediction-observation-explanation (POE) task, a recall task, a problem-solving task and a reflection task. As a variation of the clinical method, Pavlinic *et al.* (2001) observed students while they interpreted ERs of 2-D and 3-D chemical structures. Observations were recorded on an observation sheet and students were later interviewed while viewing a videotape of their performance.

Often, encapsulated within the clinical interview method, is the use of *think-aloud* tasks (e.g. Posner and Gertzog, 1982). Bowen (1994) has referred to these methods as instruments for obtaining information as to, "what is going on in the mind" (p. 185). Since clinical interview approaches sometimes consist only of think-aloud tasks, the two terms, *think-aloud methods*

and *clinical interview methods* are often used interchangeably. In terms of ER research, think-aloud techniques have proven to be powerful instruments for gaining insight into learners' utilisation of their mental models during interpretation, problem solving or reasoning with ERs as shown by studies conducted by Kozma (2003), Lewalter (2003) and Kindfield (1993/1994). Use of think-aloud methods was adopted by the current project when interviewing students during the interpretation of ERs.

3.4.2.4 Student-generated ERs

These days, researchers place greater emphasis on the collection of *non-verbal* data in order to obtain more precise inferences about ER processing (e.g. Gobert and Clement, 1999; Lowe, 1993a). Scientists such as Pauling and Einstein claimed to use only mental pictures, rather than words, when thinking and generating new ideas (e.g. Glynn, 1997; Lowe, 1987; Larkin and Simon, 1987). This thought process often resulted in them *drawing* their mental images as a way to express their thinking. For these scientists, the process of visual thinking through the construction of ERs was a powerful cognitive tool (e.g. Lowe, 1988a). Modern researchers have learnt from such strategies in that a useful technique for investigating how learners' process ERs in science is to get them to construct their own ERs. In the opinion of major workers in the field, this enables them to trace and probe students' mental models of scientific ERs (e.g. Beilfuss *et al.*, 2004; Gobert and Clement, 1999). As noted by Glynn (1997), when students draw diagrams of their mental representations, they are essentially sketching their mental models of a particular concept. Hence, the "drawing" of mental models can be seen as a diagnostic tool that can help researchers isolate conceptual and reasoning difficulties and alternative models that students may possess (e.g. Glynn, 1997; Kindfield, 1993/1994). This approach was considered appropriate for the present study.

Examples of such data collecting strategies that constitute a strong motivation for employing this approach in the present project are as follows. Gobert and Clement (1999) investigated students' diagrammatic outputs of concepts surrounding plate tectonics. Through analysis of student-generated diagrams, the researchers were able to trace students' construction of mental models and their conceptual understanding. An example of one of their probes was, "Thinking back to what you just read, draw a picture of the different layers of the earth. Include and label all the information about these layers that you can" (p. 42). In another study

by Galili *et al.* (1993), students' understanding of image formation from light rays was obtained by asking students to draw a light diagram after observing the formation of the images. Analysis of the data allowed researchers to distil learners' core concepts. Furthermore, Dwyer (1973) has made use of a "drawing test" to investigate students interpretation of ERs of the human heart. Student drawings were evaluated by assessing the placement of students' textual labels on their drawings. Lastly, Berg and Phillips (1994) investigated students' interpretation of line graphs by allowing them to construct their own graphs while students provided accompanying verbal explanations.

In similar studies, Reiss *et al.* (2002) and Reiss and Tunnicliffe (2001) have investigated students' understanding of their internal bodily structures by getting them to generate drawings of what they think is "inside them". The results suggested that through this technique, much valuable insight could be gathered about students' understanding of these concepts. Similarly, Ramadas and Nair (1996) investigated students understanding of the human digestive system with an open-ended drawing instrument. This was followed by structured but flexible interview sessions where, although a set of questions was previously designed, the probes were adjusted and student responses followed up on where necessary. In general, Reiss *et al.* (2002) have suggested that approaches such as gathering data through drawings are rarely used in science education research. Obtaining student diagrams was a major feature of the methods used to obtain students' interpretation of ERs in the present study.

3.4.3 Data Analysis

The data collected from the above three data gathering instruments (section 3.4.2) used in the current study were subjected to analytic induction (e.g. Mouton, 2001; Gall *et al.*, 1996). This approach to data analysis is concerned with "inducing" (Gall *et al.*, 1996, p. 25) common themes from the data as a process of discovery rather than subjecting previously enforced themes to the data before any analysis (e.g. Bell, 1999). Inductive analysis of the data constitutes a research process where patterns are uncovered and "made explicit" from "embedded" information that resides in the data (Lincon and Guba, 1985, p. 203).

During inductive analysis of the data in the current project, patterns of meaning and evidence were allowed to emerge from the data themselves (e.g. Anderson and Aresenault, 1998; Lincoln and Guba, 1985) without being previously enforced (Mcmillan and Scumacher, 1993). In addition, interpretations were drawn and described once all information was gathered (e.g. Verma and Mallick, 1999). Such an inductive approach is also often viewed as a descriptive synthesis of the data rather than a process of data reduction (McMillan and Scumacher, 1993, p. 480). In this regard, the researcher in the current project was concerned with providing a natural and detailed description of the patterns that emerged from the data (Gall *et al.*, 1996). Furthermore, the method of data analysis employed in the current project was viewed as being *grounded* in theory (e.g. Gall *et al.*, 1996; McMillan and Schumacher, 1993; Lincoln and Guba, 1985). This was because descriptions and explanations of phenomena came from the data themselves rather than with a view to an already pre-existing theory. This approach to data analysis is in contrast with other solely deductive forms of analyses often associated with positivistic designs (e.g. Verma and Mallick, 1999).

During an analysis of data corresponding to students' interpretation of ERs, categories of student difficulties emerged from the data themselves, rather than being pre-determined (e.g. Anderson and McKenzie, 2002; Anderson *et al.*, 1999; Boo, 1998; Bowen, 1994; Kuiper, 1994). As the process of sorting students' responses to questions (probes) proceeded, the nature of the categories, and hence the underlying difficulties, becomes clearer and sub-categories could emerge (Lincoln and Guba, 1985). The four-level methodological framework (Fig. 3.2) of Grayson *et al.* (2001) was used to classify the difficulties according to how much information and understanding the author had about the nature of each difficulty.

Difficulties that are well established by research across varying contexts (e.g. different courses, student groups and institutions) and for which there is a stable description are classified at Level-4 or established (Fig. 3.2), while those that are known to researchers but have not been extensively explored are classified at Level-3 or partially established (Fig. 3.2). Level-2 difficulties are those that are suspected based on teaching or learning experience or on very limited research (Fig. 3.2). Difficulties that emerge unexpectedly from analysis of the data are classified at Level-1 (Fig. 3.2).

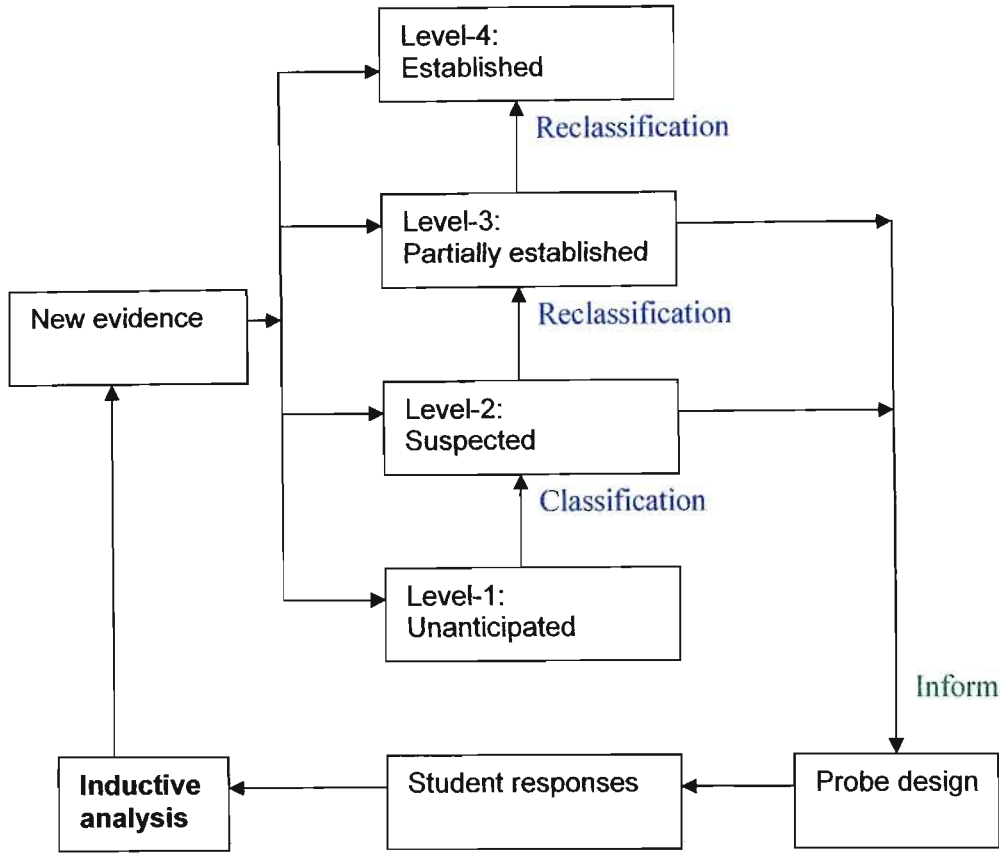


Figure 3.2 Four-level framework used to classify students' difficulties with the ERs (adapted from Grayson *et al.*, 2001, p. 615)

Application of the four-level framework (Fig. 3.2) to identify and classify student difficulties was as follows. If during free response, an unexpected difficulty emerges from the data, it is classified at Level-1. This difficulty is now suspected and therefore automatically reclassified as suspected at Level-2. If the difficulty re-emerges upon further probing, it is re-classified at Level-3 as partially established. If the same difficulty is further probed for in a different context, and emerges again, then it attains the status of being well established at Level-4 (Grayson *et al.*, 2001). Since the author found no documented research on student difficulties with the interpretation of ERs of antibody structure and interaction with antigen, the written probes and interview questions were designed to initially investigate various Level-2 suspected difficulties as well as any Level-1 difficulties that may have emerged from the free-response data (Chapter 4). In each case, the incidence of the difficulty was calculated and recorded.

After obtaining free-response data, questions that focus more specifically on learners' interpretations that emerged during free response are designed (Fig. 3.2). The design of such questions is informed by the data obtained from the free-response probes, where specific patterns that emerge are probed further (e.g. Grayson *et al.*, 2001). In this regard, a difficulty might first emerge at a low level of incidence during analysis of the free response data because not all students might reveal the difficulty. Thereafter, as the difficulty is reclassified at higher levels on the framework (Fig. 3.2), by utilising probes that focus more specifically on the difficulty, the incidence of that same response pattern would increase.

3.4.4 Validity and Reliability of the data collection instruments

Researching students' interpretation of ERs is a challenging undertaking. This is because, as pointed out in section 3.4.2.1, not many systematic tools exist for "tapping" into the human mind *directly*. There is no choice but to rely on *indirect* methods for gathering information such as interviews, written responses, observation and students drawing of ERs. Such methods were chosen based on the theoretical framework outlined in section 3.3 and were therefore, used to address the research questions in the present study. Each of these methods as a tool for generating appropriate data has both advantages and limitations. Advantages of the methods chosen for the current study were discussed in section 3.4.2. The limitations of the specific methods chosen for the present project in conjunction with issues surrounding validity and reliability of the instruments and data analysis are addressed below in sections 3.4.4.1 - 3.4.4.5.

3.4.4.1 Nature of validity and reliability in the current project

Validity is defined as the degree to which an instrument measures what it is designed to measure and *reliability* is defined as the degree to which the same responses would be yielded if the same instrument was used with the same sample of participants on a different occasion (e.g. Verma and Mallick, 1999; Gall *et al.*, 1996; Rosnow and Rosenthal, 1996; Bukatko and Daehler, 1992; White and Gunstone, 1992). Since the current project was concerned with interpreting and describing students' constructions in a certain context (e.g. Lincoln and Guba, 1985), rather than statistically generalising the findings to an entire population divorced of

context (e.g. Gall *et al.*, 1996), striving for validity became more important than striving for reliability (e.g. White and Gunstone, 1992). In this regard, Phelps (1994) has suggested that no naturalistic study, such as the one reported in the current project, will be able to be replicated entirely. This is mainly because such research deals with human subjects whose knowledge could change from one test to the next and even be influenced by the test questions themselves. Thus, in lieu of the postpositivistic approach adopted in the current project, the researcher was satisfied with the degree of *face validity* (e.g, Gall *et al.*, 1996) shown by the experimental design. In this regard, the author felt that a large degree of face validity was maintained by selecting an appropriate theoretical (section 3.3) and methodological (section 3.3) framework to address the research questions (Chapter 1). This is not to say that maintaining reliability of the instruments was of no importance; the current study aimed to achieve this whenever possible.

3.4.4.2 Using the four-level framework to pursue validity and reliability of the data

One way of pursuing validity and reliability of the data in the current study was to use the four-level methodological framework of Grayson *et al.* (2001) to classify student responses according to how much information and understanding the author had about the nature of each difficulty (section 3.4.3). In this case, reliability of the emerging responses was extended each time a difficulty was classified at a higher level on the framework (Fig. 3.2). This was because "movement" of a difficulty up the levels required repeated investigations of the same difficulty using the same or, a highly similar (in terms of prior knowledge), population of students. The degree of validity of a certain probe could be measured by comparing students' responses generated from the same probe. For example, if a probe was found to deliver *both* scientifically sound as well scientifically unsound responses (e.g. White and Gunstone, 1992), the researcher could be sure that the probe was soundly answered by a proportion of the participants, which in turn, demonstrated the presence of a *valid* probe. In contrast, we rejected probes as being invalid if the majority of students delivered poor answers. With respect to pursuing *inter-rater reliability*, the degree to which two or more researchers agree on the meaning of a question or response (e.g. Gall *et al.*, 1996; McMillan and Schumacher, 1993; Bukatko and Daehler, 1992), both the author and supervisor perused, and then agreed, on the written and interview probe sets before administration to students.

Furthermore, non-leading probes were designed to remove any emotive or leading language or potential ambiguity (e.g. Bell, 1999). As a further means of enhancing inter-rater reliability, the author and supervisor both perused the student data and resulting classifications.

3.4.4.3 Validity and reliability of the written instruments

Even though the use of written probes is a useful way for collecting data on students' interpretation of ERs, Lowe (1993a) suggests that, when interpreting ERs, subjects create "distortions" when expressing their visual interpretations through written representations. In this regard, learners may *adjust* their mental information of an ER when expressing their experiences in a verbal form (e.g. Lowe, 1993a). In other words, a student's written description is just a verbal representation of their interpretation and not an exact one-to-one replica of the mental model they may be wishing to describe. To counter this potential problem, the author had to be certain of the *consequential validity* (Gall *et al.*, 1996) of the probes. In doing so, the author had to make sure that sound research inferences could be drawn from the student data that the probes delivered. Such consequential validity was strengthened in the current study when the author noticed that regular patterns often emerged from the data. This observation contributed to *internal validity* (e.g. Anderson and Arsenault, 1998) since the researcher could be confident that the data accurately reflected the student context employed in the project. In addition, a high *external validity* (Anderson and Arsenault, 1998; Gall *et al.*, 1996) of the data was demonstrated when the author found that a particular student difficulty showed a high incidence in the student population under study.

In addition to the above, responses obtained from written instruments may be biased if learners lack the required linguistic skills, or if some students participating in a study are not as forthcoming as others might be in their written responses (e.g. Reiss and Tunnicliffe, 2001). A further extraneous factor may be the fact that not all participants possess English as their mother tongue, which could have distorted the data. In this regard, attempts were made to keep the English as clear and as simple as possible when designing probes and to take cognisance of the fact that some of the student responses might show a linguistic rather than a scientific problem. We felt that this potential problem was also well covered by the above validity checks.

3.4.4.4 Validity and reliability of the clinical interviews

Although researchers have recognised that the clinical interview method offers definite advantages for obtaining data in ER research, there has been a fair amount of critique levelled at it, particularly with respect to the reliability of such techniques. Common problems that can affect the reliability of data collection during interviews include the subjects feeling uneasy and anxious in the interview environment; guessing during tasks; and the manifesting of "artificial" metacognitive behaviours (e.g. Anderson and Arsenault, 1998; Rubin and Rubin, 1995; Bowen, 1994). Other factors affecting the reliability of interview data may include students lacking confidence in their responses; the motivation levels of the students to deliver clear and detailed responses; the mood of the students when the interviews were conducted; and students' concentration spans. Thus special care was taken in the present study to minimise the above problems during interviews by ensuring that the subject was a relaxed, interested and motivated participant.

As pointed out in section 3.4.2.1, when performing ER research, it is unsuitable to collect data *solely* in a textual or verbal format. Therefore, think-aloud tasks are often employed in interview protocols as further "forms of output" with which to collect data (e.g. Lowe, 1993a). These forms of output include student-generated diagrams (section 3.4.2.4) and other observable behaviours. Even though it is important to obtain these data sources, some participants may lack the appropriate visual communication skills necessary for expressing their interpretations (e.g. Reiss and Tunnicliffe, 2001). Related to the former, students' may not expose the necessary *tacit knowledge* (the understanding manifested in gestures such as "nodding" "pointing" and "indicating") during an interview (e.g. Gall *et al.*, 1996), which may dilute both the verbal and drawing data obtained during the interview. Participants who are shy and timid may lack confidence in exposing their tacit knowledge, which may make the data less useful. As a way of countering this potential problem in the present study, large efforts were made to relax the subject in the interview environment and encourage the student to respond freely where possible, whatever the nature of the responses might have been.

In addition, two other factors may potentially distort interview data. Firstly, the "Hawthorne effect" is a phenomenon in which, when participants know they are part of a research study, they change their behaviour to suite what they think the researcher wants to see or hear (e.g.

Rosnow and Rosenthal, 1996) leading to a bias termed *subject reactivity* (e.g. Bukatko and Daehler, 1992). Secondly, a researcher's own involvement in an interview might also potentially affect the way the student answers questions (Coon, 2001). Sometimes, an interviewer might perceive the subject in a favourable way based solely on appearance for example. As a result, the interviewer may make erroneous inferences based on this initial impression alone. Doing so will create a distortion in the data since these traits will seem to outweigh others that haven't been exposed, a situation known as the "Halo effect" (Rosnow and Rosenthal, 1996) or *observer bias* (e.g. Bukatko and Daehler, 1992). In the present study, care was taken to avoid these potential problems.

Additionally, Lewalter (2003) states that even though interviewing methods have been found to be very effective, sometimes one learning or interpretation pattern may be more overt than another pattern. In the current study, the author aimed for a high degree of *content validity*, which involved designing interview probes that represented the kind of scientific content that they were meant to represent (e.g. Gall *et al.*, 1996; Rosnow and Rosenthal, 1996). As the reader shall observe in subsequent sections, interview probes were sometimes piloted to measure whether they delivered the data that they aimed to deliver. This process contributed towards maintaining the *construct validity* (e.g. Mouton, 2001; Gall *et al.*, 1996) of the probes.

Lastly, another problem with interviews is that the data obtained from the clinical method is in a form that is not suitable for immediate analysis. For example, a one-hour interview generates about twelve to fifteen pages of transcript text as well as one hour of corresponding video footage and it is often necessary to analyse the raw data more than one once (e.g. Bowen, 1994). Consequently, familiarity and experience with these types of qualitative analyses is required. Experience with such analyses improves *observer reliability*. In this regard, the author made sure that he was proficient in these techniques, before collecting any data.

3.4.4.5 Using triangulation to improve validity and reliability of the instruments

In a commentary, relating to issues of validity and reliability in science education research, Sanders (1998) has called for ER researchers to "open their minds" (p. 1) during data analysis to prevent any hasty conclusions being drawn and to consider as many factors as possible that could distort the data (e.g. Sanders and Mokuku, 1994). As a reaction to this sentiment, one general way to extend the validity and reliability of the research instruments and subsequent analyses in the current project was to employ a *range* of multifaceted methods (section 3.4.2) to address the research questions (e.g. Cohen and Manion, 1994; McMillan and Schumacher, 1993). Thus in an attempt to eliminate bias, maintain balance, verify and validate results, and find regular patterns in the data, this study relied heavily on the concept of triangulation (e.g. Gall *et al.*, 1996; Rosnow and Rosenthal, 1996). As discussed above, since all the methods utilised in this study were limited to some extent (e.g. Gall *et al.*, 1996), a multi-method approach to collect data rather than only a single method was used in order to "zero in" (Rosnow and Rosenthal, 1996, p. 74) and cross-validate the meanings embedded in the data. In the present study, triangulation (e.g. Bell, 1999; Verma and Mallick, 1999; Anderson and Arsenault, 1998; Gall *et al.*, 1996; Cohen and Manion, 1994; McMillan and Scumacher, 1993; Lincoln and Guba, 1985) was pursued by obtaining data from two or more data sources and through different data-generating mechanisms including written probes, interview probes, student-generated diagrams and other observation methods. In addition, data was collected from multiple samples of participants and during at least three different time frames (e.g. Verma and Mallick, 1999; Anderson and Arsenault, 1998; Cohen and Manion, 1994; McMillan and Schumacher, 1993).

3.5 Summary

The methods presented in this thesis were based on a postpositivistic epistemology that followed the tenets laid out by the learning theory of constructivism. Based on this theoretical foundation, a suitable methodological framework was described to include the use of written instruments, clinical interviews, student-generated diagrams and other observational methods to gather data on students' interpretation of ERs of antibody structure and interaction with antigen. In presenting the methods employed in this project, care has been taken to provide

examples of other workers who have also employed similar data-generating strategies to argue for their applicability as research instruments in the current thesis. The discussion has also offered pertinent viewpoints relating to the validity and reliability of the methods used in the current project.

With a theoretical and methodological platform in place, findings obtained from students' interpretation of three ERs of antibody structure are explored in the next chapter.

4 STUDENT DIFFICULTIES WITH ERS OF IMMUNOGLOBULIN G (IgG) AND ITS INTERACTION WITH ANTIGEN

4.1 Introduction

In Chapter 1, it was pointed out that the interpretation of scientific ERs is a cognitively demanding task (Lowe, 1996), which can induce misconceptions and incorrect ways of reasoning. As was shown in the review of ER research in Chapter 2, extensive literature exists on students' difficulties with the interpretation of ERs. However, as argued in Chapter 1, very few research reports have been published on the effectiveness of ERs in the learning and teaching of biochemistry. This is rather surprising given the variety of visual means available for representing a single biochemical phenomenon. For instance, as presented in Chapter 3, the propositional (scientific) knowledge for concepts of IgG antibody structure and interaction with Ag can be visually represented in multiple ways. In lieu of this, it was argued that the diverse pictorial representation of these concepts might pose potential difficulties for students. Thus the aim of this aspect of the study was to investigate this possibility by studying students' interpretation of three typical textbook ERs depicting Ab structure and interaction with antigen since, to the author's knowledge, no such investigation has ever been carried out.

In this chapter research questions 1 and 2 (see Chapter 1) are addressed namely, what types of difficulties do students have with ERs used in the teaching and learning of biochemistry; what are the sources of such difficulties; and therefore, what are the factors affecting students' ability to interpret ERs? The following approach was employed to address these questions. Firstly, based on the author's teaching and learning experience, three ERs, representing the structure of immunoglobulin G (IgG) and its interaction with antigen were screened for potential student difficulties. Following this, both free response and specific probes (Chapter 3) were designed to generate data on students' interpretation of the three ERs. As part of the data analysis, the four-level research framework (Grayson *et al.*, 2001) (section 3.4.3) was used to identify and then classify students' conceptual and reasoning difficulties with the ERs.

Possible sources of the difficulties were also considered. Based on the results, the chapter discusses the potential factors affecting students' ability to interpret ERs in biochemistry.

4.2 Methods

4.2.1 Study groups and ERs under study

The study involved a total of 130 second-year biochemistry students who had studied a module on immunology in the year 2000 as well as 21 third-year students who had studied the same course the previous year (1999). Students in both years were enrolled in undergraduate biochemistry courses at the University of KwaZulu-Natal, South Africa and the data was collected in May 2000. All of these students responded to written probes. In addition, of the 130 second-year students, 10 students participated in clinical interviews. Table 4.1 outlines the dates on which data was collected together with the grouping of the different student samples and the corresponding types of data collected from each group.

Table 4.1 Student populations, data collection dates and the corresponding types of data collected from each group

Student groups	Data collection date	Year of undergraduate study	Responded to written probes	Free-response type probes	Focused type probes	Participated in clinical interviews	ER under study (Fig. 4.1)
70	9 May	2nd	Yes	Yes			A
21	9 May	3rd	Yes	Yes			A
45	10 May	2nd	Yes	Yes			C
69	16 May	2nd	Yes	Yes			D
23	23 May	2nd	Yes		Yes		C
13	25 May	2nd	Yes		Yes		A
10	18-24 May	2nd				Yes	A, B and C

For the convenience of the reader, a flip-out page of all four ERs used in this study is supplied on p. 76. Two of the textbook ERs (Fig. 4.1A and C) used in the study were presented to students as part of coursework notes with accompanying text and additional oral explanation. Fig 4.1D was obtained from one of the recommended textbooks (Stryer, 1995) for the second-year biochemistry course. Fig 4.1B was an adapted version of Fig 4.1A and was used as an additional ER during interviews (Table 4.1). Fig 4.1 A represents the tertiary structure of IgG with its variable (V) and constant (C) domains shown in light red and grey, respectively.

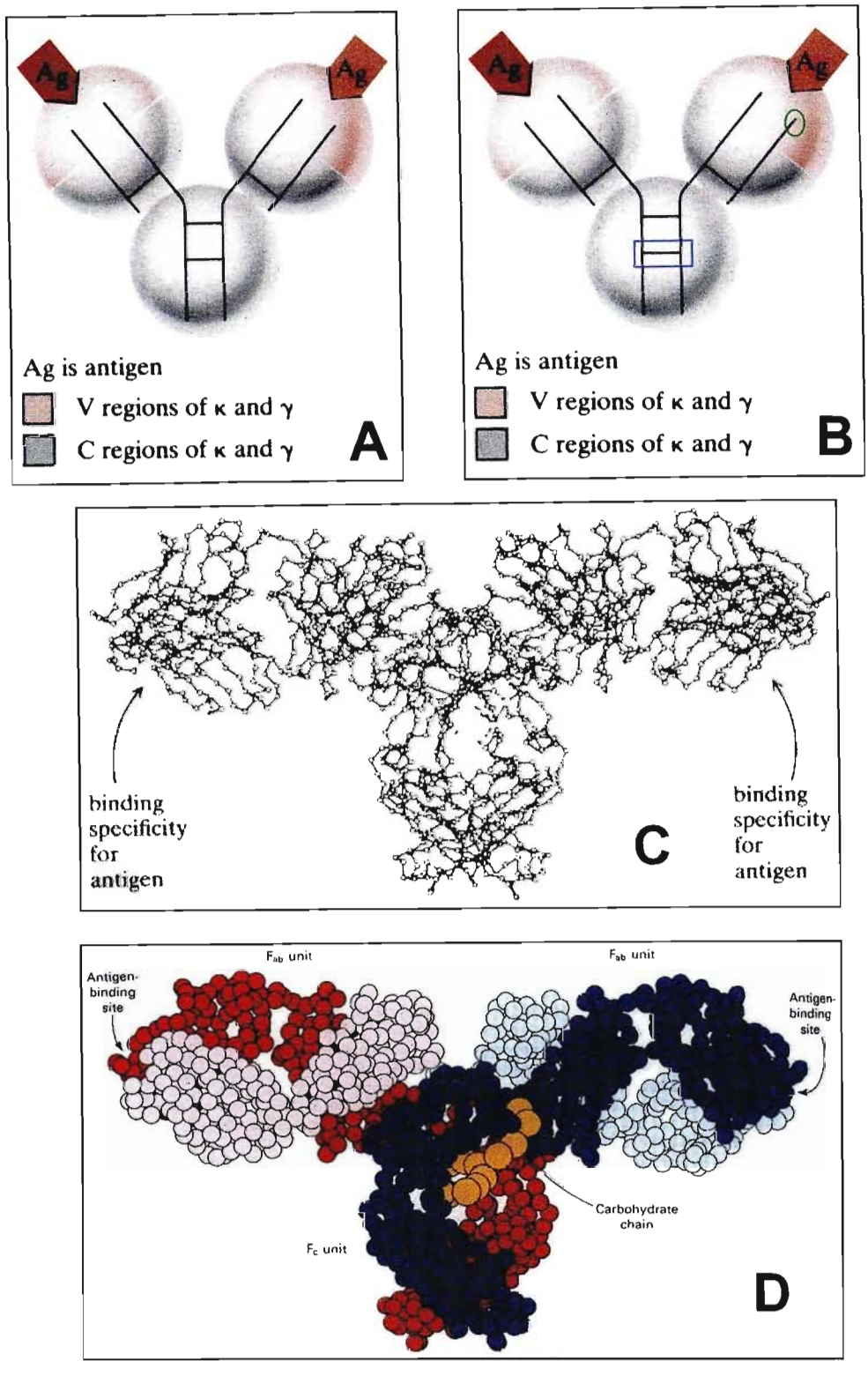


Figure 4.1 Four ERs each showing the three-dimensional structure of an IgG antibody molecule. (**A**) and (**B**): Tertiary structure showing V and C regions (Bohinski, 1987), (B) is an adapted version of (A) with a blue box drawn around one disulfide linkage and a green oval around an N-terminus of a light chain; (**C**): Tertiary structure in chain form (Bohinski, 1987); (**D**): Three-dimensional structure showing one of the H chains in dark red and the other in dark blue. One of the L chains is shown in light red, the other in light blue. A carbohydrate unit attached to a CH_2 domain is shown in yellow (Stryer, 1995).

The solid black lines in Fig. 4.1A represent the two identical heavy (γ) and two identical light (κ) polypeptide chains, connected by interchain disulfide bonds. These lines also depict the characteristically presented 'Y' shape of the IgG molecule (see Chapter 3) in a $\kappa_2\gamma_2$ structural designation. The bivalency of the IgG molecule is represented in Fig 4.1A by two antigen molecules (shown in dark red) attached to the variable regions of the antigen-binding domains of the antibody. Fig 4.1B was adapted from Fig. 4.1A to display a blue box that enclosed a disulfide linkage between the two heavy chains in the C-region of the Ab and a light green oval to enclose the approximate region of an N-terminus on one of the light chains.

Fig 4.1C is a 2-D representation of the three-dimensional structure of an IgG antibody molecule. Small circles represent the position of α-carbon atoms (amino acid centres but not whole amino acid residues), and lines between the atoms constitute the formation of the α-carbon backbone (not covalent bonds of any sort). The ER also shows the bivalency of IgG by indicating the "binding specificity for antigen" by means of two curved arrows. The Fab arms of the antibody are aligned horizontally representing the molecule in an overall T-shaped configuration (Silverton *et al.*, 1977). In this view, a vertical two-fold axis of symmetry bisects the molecule through the Fc portion of the structure (Silverton *et al.*, 1977). Fig 4.1D is a 2-D representation of the three-dimensional structure of an immunoglobulin G molecule. One of the light chains is shown in light red, the other in light blue. One of the heavy chains is shown in dark red, the other in dark blue. Each of the coloured circles in the chains represents an amino acid residue. A carbohydrate is shown by means of the yellow circles, attached to the C_H2 domain. The ER also uses textual labels to indicate the positions of both Fab units and the Fc unit. Arrows and text, that indicate the approximate binding locations on the antibody, also indicate regions for antigen binding. Each of the four ERs in Fig. 4.1 will be referred to as "ER A", "ER B", "ER C" and "ER D", respectively.

4.2.2 Screening the ERs for potential student difficulties

Prior to obtaining data on students' interpretation of the three ERs (Fig 4.1), the ERs were screened by the author for any potential difficulties that students may have when interpreting the graphical features of the ERs. The purpose of doing this was to obtain an appreciation of what potential difficulties to suspect (see Level-2: Fig. 3.2, section 3.4.3), which would in turn, inform the design of probes (section 4.2.3 below) to check if our suspicions were correct.

Such knowledge would also help inform the author's interpretation of the difficulties, should they emerge. The ERs (Fig. 4.1) were screened for potential difficulties by subjecting them to an informal visual analysis process (e.g. Kosslyn, 1989; Fleming, 1967) where the graphical markings were scrutinised (e.g. Bos and Tarnai, 1999a, b; Tamir, 1985), based on the analysers' knowledge, learning and teaching experience. The visual analysis was similar to other analyses conducted by Bell (2001), van Leeuwen and Jewitt (2001) and Lohse *et al.* (1991), where the aim was to investigate the knowledge communicated by the ERs as well the graphical markings contained within the ERs. The suspected student difficulties that were generated for the three ERs (ER A, C and D; Fig. 4.1) are presented in Figs 4.2, 4.3 and 4.4, respectively.

1.	IgG is made up of three spherical structures.
2.	IgG molecules do not exist as 3-D structures.
3.	There are structural 'spheres' surrounding the heavy and light chains.
4.	A 'V' region consists of half of a structural 'sphere'.
5.	A 'C' region consists of a whole and/or half a structural 'sphere'.
6.	The IgG molecule is supported, protected or encapsulated by structural 'spheres'.
7.	Three 'structural' spheres constitute the IgG structure.
8.	The Y-shaped molecule serves as a skeleton-like structure holding the three structural 'spheres' together.
9.	Ag molecules bind to the spheres by means of an elongated 'head' that protrudes into a structural 'sphere'.
10.	Ag molecules have arrow-like and pointed shapes.
11.	IgG molecules do not contain carbohydrate residues.
12.	IgG molecules do not consist of different domains.
13.	IgG molecules exist as 2-D Y-shaped structures.
14.	IgG molecules do not contain intra-chain disulfide bonds.

Figure 4.2 Statements describing suspected difficulties with Fig. 4.1A generated by the researcher from the visual analysis

1.	IgG only exists in a T-shaped conformation.
2.	The antigen-binding sites are not identical in structure.
3.	Ag binds to the underside of the Fab arms.
4.	The binding area for antigen is flat and planar.
5.	Ag does not have to be specific to bind to Ab.
6.	The chemical components that make up IgG are all the same size, shape and type.
7.	IgG has no secondary structure.
8.	The lines indicating the α-carbon skeleton are physically part of the IgG structure.
9.	IgG has a skeletal, mesh-like structure with many "gaps" and "holes".
10.	The lines between α-carbons are covalent bonds.
11.	IgG has no intra-chain disulfide bonds.
12.	IgG has no inter-chain disulfide bonds.
13.	IgG contains no carbohydrate residues.
14.	IgG does not consist of light and heavy chains.
15.	An antibody protein has no N- and C- termini.
16.	An antibody protein has more than two N- termini and two C-termini.
17.	IgG consists of six domains instead of twelve.

Figure 4.3 Statements describing suspected difficulties with Fig. 4.1C generated by the researcher from the visual analysis

1.	The Amino acid residues in IgG are all the same size and shape.
2.	The amino acids in IgG are spherical in shape.
3.	Every constituent atom that makes up the IgG molecule is represented.
4.	Amino acids are red, light red, blue and/or light blue in colour.
5.	IgG is constructed from molecular units in the shape of 'spheres'.
6.	The units making up the carbohydrate chain are bigger than the units making up the IgG molecule.
7.	The carbohydrate chain is yellow in nature.
8.	IgG doesn't consist of different structural domains.
9.	The heavy chain is intimately 'fused' to the light chain.
10.	IgG has no intra-chain disulfide bonds.
11.	IgG has no inter-chain disulfide bonds.

Figure 4.4 Statements describing suspected difficulties with Fig. 4.1D generated by the researcher from the visual analysis

In conclusion, even though the principles of inductive analysis (Chapter 3) were strictly adhered to when subsequently analysing students' responses (see section 4.2.4 below), the lists of suspected difficulties (Figs 4.2 - 4.4) nevertheless served as possible hypotheses of what response patterns may have emerged from the data, without biasing the author's approach to the analysis at all. In the next section, we show how the derived suspected difficulties were used to inform probe design.

4.2.3 Probing students' interpretation of the ERs

Student understanding of the ERs (Fig 4.1) was investigated at the end of the module by means of written tests and interview questions. For each ER, both the ER and its caption were supplied to students during all questioning processes but only one ER was supplied at a time. Captions supplied were as provided in Fig 4.1 except for the following modification. For ER D, the statement, "each amino acid residue is represented by a small sphere" (Stryer, 1995, p. 376) was removed as we wished to gauge students' own interpretations in this regard. Written questions were given to groups of students as described in Table 4.1. The second-year students answered both the free-response and the more focused questions, whereas the third year students answered only the free-response types. Three different sets of written questions were administered to both groups of students. The written questions (Figs 4.5 - 4.7) were given to students either at the commencement of lectures, laboratory sessions or tutorials. Students were allowed a more than adequate amount of time (approximately 5 - 10 minutes per question) to answer the questions to ensure that time pressure did not affect the nature of the answers.

More detailed information on the nature of students' interpretations was obtained by means of clinical interviews with ten volunteers from the second-year student sample (Table 4.1). During interviews, participants were asked about their understanding and interpretation of ERs A, B and C (interviews with ER D were not conducted). The general interview methods for gathering information about student understanding corresponded to those outlined in Chapter 3 (section 3.4.2.3). The same two free-response questions given in the written probes (see Fig. 4.5 below) also served as the initial free-response questions in the interviews. If no significant patterns of reasoning or conceptual understanding emerged at the time, the interviewer asked structured questions that were similar to those used in the more focused written questions (Figs 4.6 and 4.7). The interviews lasted about one hour each and were audiotaped and transcribed. Transcripts were analysed qualitatively in order to identify conceptual and reasoning difficulties (e.g. Kindfield, 1993/1994). In particular, the interview data was used to elaborate several difficulties that had emerged from the written data, as well as to expose unanticipated (Level-1) difficulties (section 3.4.3).

Initially, only free-response type questions (also termed "probes" as we use the questions to probe for student understanding and difficulties) were used to collect data during the written tests and interviews. This ensured that students were free to respond with what came to mind and reveal their understanding of the ER, without being led into giving a particular answer. Examples of this type of probe used for all three ERs (Fig 4.1) are shown below in Fig 4.5.

1.	Describe everything you think this diagram represents or shows.
2.	Is there anything in the diagram that you don't understand or find confusing? If so specify.

Figure 4.5 Examples of two free-response probes used to collect data during the written tests and interviews

As more insight was gained into the nature of each difficulty, the probes became increasingly more focused, and more specific for each difficulty that emerged. This was not the only method used by the researcher to focus the probes. As pointed out in section 4.2.2, probes were also focused through the author's lists of suspected difficulties (Fig. 4.2 and 4.3). For instance, the author's suspicion that students' may have thought the large "spheres" represented in ER A (Fig. 4.1) to be structures separate from the antibody itself, prompted the design of probes 4 and 5 (Fig. 4.6). In another example, based on the author's suspicion that students' may misinterpret the alpha-carbons of the amino acids shown in ER C, probe 9 (Fig.

4.7) was specially designed. The set of focused probes designed for ERs A and C and answered by the second-year students (Table 4.1) are provided in Figs 4.6 and 4.7.

3.	With the aid of separate sketches, explain which part of the diagram represents: i) The antibody; ii) The antigen
4.	What do the various black lines on the diagram represent?
5.	What do the coloured areas represent?
6.	How do the coloured areas relate to the black lines on the diagram?
7.	Use the diagram to explain what happens to the antigen (i.e. what does it do?) after it has bound to the antibody.

Figure 4.6 Examples of focused probes designed for ER A to collect data during the written tests and interviews

8.	What level of structure (e.g. primary, secondary, tertiary or quaternary) does this diagram represent? Explain your answer.
9.	What do the circles and lines represent?
10.	Where and how does antigen bind to the antibody?
11.	Compare the structure (primary, secondary, tertiary or quaternary) of the two antigen binding sites.
12.	What does the diagram tell you about the specificity of the antigen?

Figure 4.7 Examples of focused probes designed for ER C to collect data during the written tests and interviews

In summary, in addition to focusing probes by virtue of the patterns that emerged during free response data itself, the list of suspected difficulties (Figs 4.2 and 4.3) also informed the design of probes to investigate whether there were any other serious difficulties that had not been exposed, since free-response probes alone (by their very nature), would not on their own necessarily reveal all possible difficulties.

4.2.4 Analysis and classification of student responses

Student answers were analysed by inductive analysis (see section 3.4.3). The four-level methodological framework of Grayson *et al.* (2001) was used to classify the difficulties at Level 1, 2, 3 or 4, according to how much information and understanding the author had about the nature of each difficulty (see section 3.4.3). In each case, the incidence of the difficulty was calculated and recorded.

4.3 Results and discussion

Investigations into students' interpretation of the three ERs (Fig 4.1), revealed three general categories of difficulties, which we termed *process-type* difficulties, *structural-type* difficulties, and *DNA-related* difficulties, as well as seventeen sub-categories of difficulties. Each sub-category was classified separately on the research framework of Grayson *et al.* (2001). To facilitate discussing the student quotations presented in support of each category and sub-category, the quotations have been numbered where relevant.

For the convenience of the reader, a flip-out table of all difficulties reported in this chapter is supplied on p. 84.

4.3.1 Process-type difficulties

Students demonstrating the general process-type difficulty (**P**) thought that the three IgG antibody ERs (Fig 4.1) represented various complex processes, rather than a simple non-covalent binding interaction between antibody and antigen molecules. Within this general category, six sub-categories of difficulties were discovered belonging to the parent type, which were exposed when students interpreted one or more of the three ERs, A, C, or D (Fig 4.1). Between 7% and 70% of students showed the general **P** category of difficulty depending on the particular ER and probe employed. Students within this general category of incidence showed one or more of the sub-categories of difficulty. Table 4.2 presents a summary of the descriptions of these sub-categories, the ER from which they were generated, as well as their classification on the Grayson *et al.* (2001) research framework.

4.3.1.1 P_1 Sub-category: Antigens "attack" antibodies

In the first sub-category (Table 4.2), labelled P_1, some students interpreted the three ERs (Fig 4.1) as showing an antigen in the process of attacking the antibody, analogous to the way a foreign agent is said to "attack" or "invade" a host. Examples of student quotes that exposed this difficulty, either in interviews or in written responses to ER A, C or D (Fig 4.1), are illustrated below:

1. "The diagram is trying to represent regions...regions where an antigen may attack." [response to probe 1; ER A]

2. I: All right, now...let's talk about the antigen. What is the antigen doing here [no pointing]?
   S: I think it [Ag] is trying to attack some of the...it's trying to attack the immunity of...the cell...trying to cause some disease or something. [Interview extract; ER A]

3. "Bonds between molecules, its structural configuration i.e. 3D. Sites for other molecules to attack and the effects the binding molecule will have on the structure." [Response to probe 1; ER C]

4. "...antigen (which we can call an inhibitor) attacked and binded in the available site..." [Response to probe 1; ER D]

Quotes 1-4 clearly demonstrate students' interpretation of the Ag as an agent that on its own attacks the antibody, which in turn has the capacity to actively "fight" the "disease". The students who showed the P_1 difficulty may have incorrectly linked the everyday meaning of the body being prone to an "attack" to their interpretation of the single biomolecular event of primary interaction between antibody and antigen. Interestingly, all three ERs (A, C and D) exposed the P_1 difficulty, despite the fact that ERs C and D did not explicitly represent the presence of antigen structure(s) graphically, but merely inferred the location of antigen binding with the aid of arrow symbols.

The use of terms such as "attack" and "fight" to describe antibody-antigen binding is in line with the work of Simonneaux (2000) who has shown that students often view the immune system in terms of a "warrior metaphor". Simonneaux (2000) suggests that this image of the immune system has its roots in social representations of health as well as the associated *military* terminology used to describe it. Terms such as 'invasion', 'defence', 'fighting' and 'antibody' form part of the vocabulary that is used to describe and understand the immune system. The P_1 difficulty initially emerged unexpectedly from free-response data and is classified as a sub-category of **P** since students ascribed a process other than non-covalent Ab-Ag binding to the ERs. Since it was re-exposed during interviews, it was classified as partially established (Level-3) on the research framework (Table 4.2) meaning that studies are still required in multiple contexts in order to fully establish the difficulty at Level-4.

Table 4.2 Descriptions of sub-categories making up the process-type (**P**) difficulty category for interpretation of the three ERs, ranging in student incidence from 7% to 70%

Difficulty category Code	General category and sub-category description of difficulty	Shown for ER (Fig 4.1):	Level on framework
P	IgG antibody ERs represent various complex processes, rather than a simple non-covalent binding interaction between antibody and antigen molecules.	All ERs	N/a
P_1	An antigen attacks the antibody, analogous to the way a foreign agent might "attack" or "invade" a host.	A, C and D	3
P_2	The antigen enters, protrudes into or penetrates into the antibody structure.	A, C and D	3
P_3	The antibody itself is capable of performing the major immune function of eliminating the antigen.	A,C and D	3
P_4	Antibodies can fuse or split into different structures.	A	3
P_5	Heavy and light polypeptide chains are able to emanate from within the structure of the antibody.	A	1
P_6	Heavy and light chains are information-carrying devices that can communicate between different parts of the immunoglobulin.	A	1

Table 4.3 Descriptions of sub-categories making up the structural-type (**S**) difficulty category for interpretation of the three ERs, ranging in student incidence from 3% to 70%

Difficulty category Code	General category and sub-category description of difficulty	Shown for ER (Fig 4.1):	Level on framework
S	Misinterpreted various ER graphical markings representative of antibody structural components.	All ERs	N/a
S_1	Misinterpreted the arrow symbolism used to represent the antigen (and/or its site of binding to the antibody).	A, C and D	3
S_2	Misinterpreted the symbolism representing the disulfide linkage joining light and heavy chains.	A and B	3
S_3	Misinterpreted the symbolism depicting end termini on polypeptides.	A and B	2
S_4	Misinterpreted the 'spheres', depicting variable and constant regions, as other structural entities.	A	3
S_5	Misinterpreted the "Y-shaped", black line as a support structure.	A	3
S_6	Misinterpretation of the level of protein structure represented.	C	3
S_7	Interpreted ER depicting antibody structure as representing a T-cell.	A and C	1
S_8	Misinterpreted the symbolism used to represent amino acids.	C and D	3
S_9	Interpreted antigen-binding sites as not being identical in structure or composition.	C and D	3

Table 4.4 Descriptions of sub-categories making up the DNA-related (**D**) difficulty category for interpretation of the three ERs, ranging in student incidence from 4% to 19%

Difficulty category Code	General category and sub-category description of difficulty	Shown for ER (Fig 4.1):	Level on framework
D	Incorrectly interpreted the ERs as representing a form of DNA structure and/or DNA processing.	All ERs	N/a
D_1	Misinterpretation of antibody structure as representing DNA structure or function.	A, C and D	3
D_2	Inappropriately combining immunology concepts with DNA concepts.	A and D	3

4.3.1.2 P_2 Sub-Category: Antigens can enter antibody structures

In a possible extension of the erroneous thinking shown by the P_1 sub-category, and classified as the P_2 process-type difficulty (Table 4.2), some students' interpreted ERs A, C and D (Fig. 4.1) as representing antigen in the process of either entering, protruding into, or penetrating into the antibody structure itself. The following students quotations, obtained from their interpretation of the three ERs (Fig.4.1), illustrate this difficulty:

1. "Antigen entering the κ and γ. Shows the pathway on which the antigen goes through. The V region first, then through the C region." [Response to probe 1; ER A]

2. I: We've established now that the antigen binds here [points to Ag binding site].
S: Yeah...I think it [antigen] goes straight and breaks those two strands [S-S bonds]...
[Interview extract; ER A]

3. "  represent[s] the direction of the molecule of Ag and the way the[y] attack the C region of κ and γ." [Response to probe 1; ER A]

4. "The path of the antigen." [Response to probe 4; ER A]

5. I: Over here in this region [points to Ag-Ab binding region] how does it [the antigen] come to be there?
S: [long pause]..ah...maybe it broke the membrane of...of the antibody... And came into the antibody [points to Ag-Ab interaction site] [Interview extract; ERA]

6. S: Here [points to binding site] and here [points to other binding site]. You have this piece of antibody and the antigen will come and bind here [points to binding site] and sticks there or goes inside... It's sort of like it [Ag] recognises some chemicals...that are in this bond region [points to b. site] and it [Ag] goes in... if it [Ag] has to get inside it would get inside the antibody... [Interview extract; ER C].

7. "The antigen[s] get/enter into the red blood cells on both sides shown in the diagram. They then cross over to different parts of the blood cells spreading all over. Where the sort of ladder is, i.e. in that cell, it is where all the antigens get together and sort of like summarise everything (diseases) found in the blood. Then they can report or give sign to the responsible organelles to attack those diseases found." [Response to probe 1; ER A]

8. "The diagram represent[s] different substrates with binding sites where the enzyme was supposed to bind. Instead, antigen (which we can call an inhibitor) attacked and binded in the available site and the antigen is spreading." [Response to probe 1, ER D]

As shown by quotes 1-4 some students, when analysing ER A, interpreted the antigen as being able to follow a specific path (quotes 1 and 4) when entering the Ab in the direction of a "channel" (quote 3) between the light and heavy polypeptide chains and then attacking the disulphides (quote 2) at the end of the channel. The latter difficulty might be due to the relative spatial organisation of the arrow-like structure pointing to, and being of the same width as the "channel" between light and heavy chains. Furthermore, it is also interesting that

two students, besides manifesting the P_2 difficulty, interpreted the shaded spherical markings in ER A as representing structural entities such as cells (quote 7) while another student interpreted the spherical marking as a membrane (quote 5) which could be penetrated by the antigen. These misinterpretations of the graphical markings as separate structural entities will be presented as a separate major sub-category of difficulty in section 4.3.2. The P_2 difficulty was exposed across all three ERs (quotes 6 and 8). Of particular interest in quote 8 was that the student thought the antigen, having entered the antibody structure, was in some way "spreading", perhaps in an analogous way to how disease is perceived to multiply.

The P_2 sub-category of difficulty was initially suspected on the basis of the visual analysis of the ERs performed by the author (statement 9, Fig. 4.2). After being exposed during free response, interviews and focused probing across all three ERs, the P_2 difficulty was reclassified from Level-2 to Level-3 on the Grayson *et al.* (2001) research framework. The P_2 difficulty is classified as a sub-category of the **P**-type category as students were unable to interpret the three ERs as only showing the idea of non-covalent Ab-Ag interaction.

4.3.1.3 P_3 Sub-Category: Antibodies eliminate antigens

Students who showed the P_3 difficulty (Table 4.2) interpreted the antibody represented in the three ERs (Fig 4.1) as the entity capable of performing the immune function of destroying or "breaking down" the antigen, in order to "act against" and eliminate it. For this reason, the P_3 difficulty, also implying a complex process, was classified as a sub-category of the parent **P**-type category and is shown by the following examples of student quotations obtained from interpretation of all three ERs (Fig. 4.1):

1. "It [ER A] is meant to show how the antigen Ag attack[s] the cell and how the antibody fights the antigen and get[s] rid of it. Region[s] V and C show the different parts of the antibody which are meant to destroy the antigen. The composition of chemicals released in region V are different to the one[s] in region C. [response to probe 1; ER A]

2. "After binding to the antibody, the antigen is destroyed by the antibody. The antigen cannot do anything as it is tight[ly] bound to the antibody, which will then destroy the antigen." [Response to probe 7; ER A]

3. S: ...should the antigen be detected, these black lines will be arranged in a certain order... making themselves ready to attach to certain sites [on Ag]... these black lines over here should interact with the [Ag] molecule and break it down. [Interview extract; ER A]

4. S: ...Ok, they're [Ag] just going to go inside [the Ab]...that's how they going to get digested, inside there, obviously inside...the antibody, they're going to get digested. They go inside the

antibody to get digested... they [Ags] will be engulfed...it [Ab] forms like a thing around it [Ag] and sucks it in... [Interview extract; ER A]

5. S: ...the antibodies would change their [Ag] structures, and they [Ags] won't be able to function anymore.
I: Change of structure?
S: Destroys them [Ag].
I: Where would that occur?
S: As they're attached. [interview extract; ER C]

6. I: Ok, once it is joined [Ag] what does it do?
S: ...then I would think that the antibody surrounds it [Ag] and kills it. [interview extract; ER C]

7. "The representation of an immune response by an antibody which is an IgG molecule. Molecule capable of binding with antigen at binding site so as to act against an antigen." [response to probe 1; ER D].

8. I: Now, tell me more about the colours represented here [V and C regions].
S: Oh, ok, well grey is more kind of like an inact[ive] ... in an infection or something it is like red... they kind of look active because they're red, you know...like if you switch on like an oven or stove or something, when it is inactive, it's black...and when it's hot it's kind of red...I would think that's why I think they're the active parts because they're red... [interview extract; ER A]

The quotes above confirm some students' misinterpretation, across all three ERs, that the Ag could be "acted against" (e.g. quote 7), destroyed or eliminated by the antibody in some manner. This erroneous interpretation is also clear in quotes 1 and 2 in which students thought that antibody is capable of "destroying" antigen. In addition, quote 3 suggests that the antigen could get broken down by the black lines representing the polypeptide chain of the antibody, while quote 5 suggests that antibody induces a change in Ag structure and quotes 4 and 6 both suggest that Ab engulfs the antigen. In addition to confirming the P_3 difficulty, quote 8 specifies a link to the red-shaded colouring used to depict the variable regions of the antibody molecule. In this regard, and with respect to ER A, a possible source of the P_3 difficulty could be that, in everyday visual displays, the colour red is often associated with ideas of "danger", "infection", "activity" and "heat". Students showing the difficulty may have simply associated their everyday understanding of the red colouring to the context of primary interaction between Ab and Ag. As a result, ER A was interpreted as depicting an antibody "warrior" of sorts (e.g. Simonneaux, 2000). Students who showed the P_3 difficulty were probably unable to distinguish between the concept of antibody-antigen binding as a primary reaction and other cellular immune response reactions responsible for digestion and elimination of foreign bodies.

The P_3 sub-category initially emerged during free-response but was also exposed during more focused written responses and interviews, which allowed it to be classified as

partially established at Level-3 on the framework (Table 4.2). Thus although we are confident about the nature of the difficulty, further research is required to establish (Level-4) its occurrence in multiple contexts.

4.3.1.4 P_4 Sub-category: Antibodies can split or fuse into different structures

Some students thought that ER A (Fig. 4.1) represented a structural entity in the process of "splitting" into, or forming more than one structural entity. The following quote supports this interpretation, which was coded the P_4 difficulty (Table 4.2):

1. "Cell (C), cell division takes place, two cells (V) are formed. Cell (C) old mature structure attaches 2 cells with black lines or bonds. Young immature cells (V) are attacked by Ag." [response to probe 1; ER A]

Quote 1 shows that the student interpreted ER A as representing the process of cell division. Other student responses suggested related processes, where it was thought that ER A either represented two antigens, antibodies or other structural units 'coming together' or 'combining' resulting in some type of cellular or structural fusion. This further aspect of the P_4 sub-category (Table 4.2) was supported by the following quotes obtained from the interpretation of ER A:

2. "... The arrow points to the area which the Ag antigen connects to the V region. The 2 antibodies join +[and] become one, which is represented by the black lines." [response to probe 1; ER A]

3. "It is a combination of two antigens having two colours resulting in an antigen with one colour. C regions of κ and γ also come together." [response to probe 1; ER A]

4. "Ag becomes part of the Ab." [response to probe 7; ER A]

5. S: ...these are antigens [points to top two spheres], and this [lower sphere] is...an antibody and this [top sphere(s)] is trying to look like it [lower sphere] so that they can react...they [Ab and Ag] can form one big molecule...it [top lt sphere] was red in colour, and then when it joined to this one [lower sphere]...then it [Student's Ags: top two 'spheres'] changed and tried to look like this [lower sphere] so that it could fit.
[...]
I: How would they [Ab and Ag] react [S stated this earlier]?
S: ...they [Ab and Ag] had sequences of amino acids that could pair with the sequences of the antigen [top sphere]... these [Student's Ags: top spheres] will change into antibodies... It is an antibody formation.
[interview extract; ER A]

It is evident from the above data that these students interpreted the spherical shapes on ER A (Fig. 4.1) as portraying separate structural components that were somehow in the process of fusing into one structural component. This thinking is depicted in Quote 2 where the student suggests that the antigen and two antibodies (each "sphere" was interpreted as one antibody) are in the process of "becoming one". In addition, Quotes 3, 4 and 5 suggest that there is a similar fusion process occurring, where antigens are fusing (quote 3) or antigens are becoming "part" of the Ab structure (quotes 4 and 5). Regarding quote 5, the student thought that the antigens composed the entire top two 'spheres' and thought that they were trying to "look like" the antibody, which was interpreted as the lower grey 'sphere'. In addition, the same student thought that the antigens (top two spheres) were in the process of forming a single structure. This was further supported in that, even though the student correctly identified the black "lines" to represent amino acids, the student maintained that amino acids "on" the antibody could "pair" with the sequences of the antigen allowing for a fusion process. In this regard, the student interpreted the antigens as slowly altering their colour from red to grey once they had "reacted" with the antibody. The source of this misinterpretation could very well be due to the manner in which the student perceived the colouring on ER A (Fig 4.1). As discussed in section 4.3.1.3, perhaps the red colour was viewed by the student as some type of disease-causing or attacking agent that could be transformed by a grey-coloured 'neutralising' entity.

The P_4 difficulty first emerged unexpectedly during free response probing and was then re-classified at Level-3 after interviews and more focused written probing. It exists as a sub-category of the **P**-type category since some students ascribed biological processes to ER A when only a non-covalent binding interaction between Ab and Ag was represented.

4.3.1.5 P_5 Sub-category: Polypeptide chains can emanate from antibody structure

In the P_5 sub-category of the process-type difficulties, some students interpreted the heavy and light polypeptide chains in ER A (Fig. 4.1) as being able to grow or originate from within the structure of the antibody itself (Table 4.2). Therefore, this sub-category of misinterpretation is related to the overall **P**-type category and is demonstrated by the following two interview quotes:

1. S: These strings [polypeptide chains]...I would say they originally came from this big black molecule [C-region and lower sphere] ...they [H/L chains] come apart [indicates at hinge region], they bind into the antigens...
[...]
S: They come from here [indicates the lower sphere] ... they [H/L chains] growing from here [points to lower sphere]... [interview extract; ER A]

2. I: ...you're saying the black lines are part of the antibody [S stated this earlier]?
S: Ja [yes], I think they're [H/L chains] originating from here [lower 'sphere']
[...]
I: ...if the antibody was by itself here, if the antigens weren't here on this picture, how would it [ER A] look?
S: These black lines [heavy/light chains] wouldn't be out here [points], it [they] will [would] be compacted inside so there's just one sphere [lower sphere]... and then... it [H/L chains] will come into contact with the antigens, and then sense the contact, and then these lines will protrude in... [interview extract; ER A]

Regarding quotes 1 and 2, both students thought that the polypeptide heavy and light chains would be able to "grow" out, or emerge from, the antibody molecule represented as the lower "sphere". In the first case (quote 1), the heavy and light chains were interpreted as being able to "go into" the antigens when binding to them. Similarly, the second student (quote 2) thought that the heavy and light chains were able to "protrude" into the antigen structure, upon "sensing the contact" of the antigens. Both students thought that the antigens were represented in ER A by the entire top two spheres. This was probably because they interpreted the arrow-shaped graphical feature, used to depict antigen, simply as a diagram label. This notion of interpreting the arrow-shaped antigen molecules as diagram labels will be dealt with in more detail in section 4.3.2.

The P_5 difficulty emerged only during interviews. Therefore, the difficulty was classified as unanticipated at Level-1. Further investigations are required to clarify the nature of the difficulty more fully and classify it at a higher level on the framework (Grayson *et al.*, 2001).

4.3.1.6 P_6 Sub-category: Polypeptide chains are information carriers

In the P_6 process-type difficulty, the black heavy and light chains, depicted in ER A (Fig. 4.1), were interpreted as being information-carrying devices that could somehow communicate between different parts of an immunoglobulin structure (Table 4.2). Since this process-type difficulty was only exposed by interviews, it was classified as unanticipated at Level-1. The following quotation illustrates this process-type difficulty:

I: ...what would these be [points to black heavy and light chains]...these black lines here?
[...]
S: A pathway...to transport the information...
[...]
S: ...this bridge...[points to first S-S in C-region]...bridges the information from this ball [top lt sphere]...and this ball [top rt sphere]. Yeah. And the second one [bot. sphere], I think it takes overall information from the two balls [top two spheres].
[...]
I: These lines over here... [points to H/L chains]...Tell me a bit more about those lines.
S: ...I think here it [black lines] carries two different [types of] information than here [black lines]...the pink and the black...the information is totally different ...I think the pink one [red areas on top two spheres] represent[s] the information before it was translated so that this ball [lower sphere] can understand the information from these two balls [top two spheres]... [interview extract; ER A]

It is evident from the above interview extract, that the student interpreted the black heavy and light chains as information-carrying devices. Furthermore, the student thought that certain information, provided by the red areas in the top two spheres, was being translated by the grey "sphere" at the bottom of ER A and that the heavy and light chains made this communication possible. Therefore, this sub-category of difficulty is related to the parent **P**-type because the student interpreted ER A as showing a process other than simple Ab-Ag interaction. A possible source of this difficulty could be the fact that even though proteins can "communicate" through conformational changes or "signal" each other during protein synthesis (e.g. Campbell and Smith, 2000), students may have confused these ideas during their interpretation of ER A.

4.3.1.7 Sound interpretations of the ERs relative to the Process-type difficulty category

In contrast to the process-type (**P**) difficulties (Table 4.2) shown by students with the three ERs (Fig 4.1), some students showed evidence of scientifically sound interpretations of the same three ERs (Fig 4.1). The following are examples of such responses:

1. "The whole complex agglutinates and allow[s] the body to recognise it and remove it." [response to probe 7; ER A]

2. I: ...and...this colour? [pink-brown V region]
   [...]
   S: ...the variable region will be able to form a kind of...stereospecific structure... which would have particular sites which would bind to particular sites on a particular structure of antigen. So, different antigens would have different potential binding sites on them... you have a structure [on Ab] which is stereospecific to one antigen.
   [...]
   I: ...Why is it actually there...the antigen?

S: ...it might be a chemical on a foreign bacteria or it could be a virus, it's a foreign particle that has entered the system...the next step would be for white blood cells...the lymphocytes...they'd come bind to this region [bot. of C-region]...they got a protein in their cell membrane which recognises to bind to that and then consume...put it into a lysosome...lyse the whole IgG with the antigen on it.
[interview extract; ER A]

3. I: What does this diagram [ER C] tell you about the specificity of the antigen?
S: ...well it [Ag] will be... stereospecific to both of the sites on an IgG...
[...]
S: ...the tertiary positioning of...acidic and basic residues, would then form a kind of stereospecificty...the positioning in 3-D space of the potential hydrogen bonding sites... there might be some hydrophobic interaction to an extent, and you might get a non-polar region... [interview extract; ER C]

4. "Show[s] the binding sites for an antigen. The 3-D configuration of an antibody. Antigens bind only to two specific sites on an antibody – these two sites are found on either end of the molecule. [response to probe 1; ER C]

5. "Diagram [ER D] shows 3D structure of a molecule (IgG). Shows 2 antigen binding sites at the extremes of the molecule... since antigens bind to it. H chains (long arm) are shown in dark red and dark blue & L chains (short) are shown in lighter colours..." [response to probe 1; ER D]

It is evident from the above quotes, that some students provided scientifically acceptable interpretations of the three ERs relative to the process-type difficulties (Table 4.2). For instance, during interpretation of ER A, quote 1 correctly suggests that agglutination is one process whereby antigen can be removed from a biological system, while quote 2 suggests that specialised cells are responsible for digesting and eliminating Ag-Ab complexes. In addition, in quote 3, the student provides a detailed explanation for the process of specific interaction between Ab and Ag. This is also supported by quote 4. A sound interpretation of ER D is demonstrated by quote 5, in which the student correctly explains the process of primary interaction between Ab and Ag. Thus the above quotes suggest that the three ERs (Fig. 4.1) could in fact be useful to some students and correctly interpreted even though other students found problems with them. The sound interpretations of the three ERs by those students also served to confirm the validity of the probes designed to generate data (section 4.2.3).

4.3.1.8 Conclusion and possible sources of the Process-type difficulty

In regard to the Process-type (**P**) category of difficulty, six sub-categories of difficulty emerged from the data, with the P_5 and P_6 sub-categories being classified at Level-1 as unanticipated on the Grayson *et al.* (2001) framework, while the P_1 through to P_4 sub-

category were classified at Level-3 as partially established. Thus in all cases further research is required to fully establish whether the difficulties will be found in other contexts such as other institutions and classes, both locally and internationally. Possible sources of the Process-type difficulty are as follows.

With regard to difficulties P_1, P_2 and P_3 (Table 4.2), where students thought that antigens "attacked" antibodies, that antigens could enter Ab structures or that antibodies are themselves responsible for destroying antigens, respectively, it is suggested that students' erroneous conceptual knowledge (or the incorrect application of it), during interpretation of the three ERs, may have contributed to these misinterpretations. In support of this observation, students often interchanged the words "binding site" and "active site" when interpreting the three ERs (Fig. 4.1) during focused probing. This was illustrated by the following quotes:

> "Active site, the antigen blocks the antibody, it's like a key-lock analogy." [response to probe 10; ER A]
>
> "It binds to the active site by lock and key model and induced fit model." [response to probe 10; ER A]
>
> "It binds by forming bonds with the molecules in the active site(s)." [response to probe 10; ER A]

The above inappropriate use of such terminology and concepts by students may have been a major source of the process-type difficulty across the three ERs. For instance, the word "active", with reference to enzyme-substrate binding, rightly suggests the possibility of chemical action or catalysis taking place at the binding interface. However, in Ab-Ag binding, *no* "active" chemistry occurs. Instead, this primary process serves as a precursor to the more "digestive" and "killing" types of cellular immune responses. Similarly, with reference to difficulties P_5 and P_6, students may have been using unsound conceptual knowledge to interpret the ERs, which resulted in ideas such as "growth" of, and "communication" between, polypeptide chains coming to the fore. Alternatively, it could be plausible that a source for the latter difficulties was students' lack of the scientific knowledge necessary for interpreting the ERs.

In addition to the possible sources of the P-type difficulty provided above, upon analysis of the data across the different student samples *relative to each ER*, it was found that ER A showed the highest incidence for the process difficulty category at a value of 70%. This was followed by ER C and ER D, in which each showed an incidence of 50% and 7%,

respectively. Since more or less the same content knowledge was required to interpret all three ERs, it is likely that these differences in incidences were primarily due to the relative nature of the ERs and how much difficulty students had interpreting their graphical features. Thus ER A seemed to have the most negative influence on students' interpretations, possibly because it makes use of less conventional features (e.g. large spherical markings) than ERs C and D, to represent the protein. By contrast, ER D makes use of a more conventional space-filling representation with which students would be more familiar from their studies of protein structure. The wire frame-like/α-carbon backbone representation of ER C would be less familiar to students, hence the intermediate incidence. Thus this is an example of how the nature of the ER can strongly influence student interpretation and therefore learning. Following on from this argument a possible source of the P_4 difficultly, where students thought antibodies could "split" apart or, "fuse" together, may be the rather unconventional and confusing spherical and "ball-like" graphical means used to depict the V and C regions of heavy and light chains.

A possible source of the P_2 difficulty for ER A might have been the fact that, the antigen is both pointing at the space between the light and heavy chains and is of the same width as the space, suggesting a possible pathway of entry. Thus it is possible that the arrow shape of the antigen and channel-like artistic features of the ER led students to incorrectly consider the processes of phagocytosis and endocytosis when attempting to interpret the ER. Interestingly, the same invalid reasoning was also shown with students' interpretation of ERs C and D by also thinking that Ag could somehow enter the Ab structure. The latter may have been due to the graphical nature of the arrows used to indicate possible areas for antigen-antibody interaction. Instead of interpreting the arrows as depicting possible antigen-binding sites, the arrows may have been interpreted as pointing to a point of entry for the antigen molecule.

A source for the P_1 and P_3 difficulties, where students thought the antibody was under "attack" and/or could itself eliminate antigen, may have been related to the use of the red-like colour to represent variable regions of the antibody on ER A. As shown in the data, students associated this red colour with everyday connotations of "activity" or "danger" and suggested that a chemical or digestive process between antibody and antigen was occurring. The result was that students placed too much importance on the nature of the colours (e.g. Reid and Miller, 1980; Holliday, 1975a) used to represent the various features of the ERs, especially when they related them to everyday language.

Another possible source of the process-type difficulties (Table 4.2) could have been that some students were focussing on the surface-level features of the ERs when extracting meaning from them, a type of reasoning that has been shown to be common amongst novices (e.g. Lowe, 1993a, section 2.6.3). In this thesis, "surface-level reasoning" shall be used to describe the cognitive process employed by students when they focus on surface features of an ER to interpret it, while "deep-level reasoning" shall be used to describe the process in which students focus on the deeper structure of the ER to extract meaning (e.g. Lowe, 2003, 1996; Kozma and Russell, 1997).

In the case of the P_4 difficulty (Table 4.2), students may have inappropriately transferred (Salomon and Perkins, 1989) what they had previously learnt about biology ERs and concepts of cell division, mitosis or binary fission to what was being graphically presented on ER A. Such inappropriate transfer of information may well be a consequence of surface-level reasoning, especially when students rely heavily on the visuospatial information displayed on the ER to make sense of it (Cheng *et al.*, 2001; Olivier, 2001; Lowe, 1996). Furthermore, as was displayed by difficulty P_2 (Table 4.2), students probably interpreted the ERs literally instead of recognising the stylised nature of the ERs (e.g. Lowe, 1989) when suggesting that the arrow-shaped antigen in ER A (or the arrows used to show antigen-binding sites in ERs C and D) could penetrate the antibody (another example of surface-level reasoning). In fact, as we shall see later (Fig 4.8), some students, during focused probing, drew the elongated Ag to actually resemble the shape of an arrow form (see section 4.3.2).

With regard to the P-type difficulties in general, many students were probably over generalising when deciphering the ER (e.g. Hill, 1990). This was especially the case for difficulties P_1 and P_3 where students thought the three ERs (Fig 4.1) represented cellular immune response reactions rather than the primary interaction between antibody and antigen. A source of this erroneous reasoning may be the vocabulary used to describe immunological processes, as processes of "attack", or "killing" (e.g. Simonneaux, 2000). When interpreting the ERs, students may have linked such terminology to their interpretations.

Based on the above analysis of student data corresponding to the **P**-type category of difficulty, it is suggested that the potential sources of students' difficulties, across the sub-categories, were related to either, students' lack of scientifically sound concepts needed to interpret the ERs, students' use of inappropriate processing mechanisms to decipher the ERs or problems

with the nature of the ERs themselves. In the latter instance, the nature of the graphical features on ER A seemed to enhance the P-type category of difficulty amongst students the most.

4.3.2 Structural-type difficulties

Students who showed the structural-type difficulties (**S**) when interpreting the three ERs (Fig 4.1) incorrectly interpreted the way in which various structural features of IgG are visually represented on the ERs. These included the way in which disulfide bonds, variable and constant amino-acid regions, light and heavy chains, end-termini, amino-acid residues (α-carbon centres), antigens, antigen binding sites, level of protein structure and binding site structure were represented on the antibody ERs. Incidences for this general difficulty category ranged from 3% to 70% across the students groups and across all three ERs. Students represented by this range of incidence showed one or more of the sub-categories of difficulty. Table 4.3 presents a summary of the nine sub-categories of the structural-type difficulty (**S**) belonging to the parent type that were exposed through student interpretation of the three ERs (Fig 4.1).

4.3.2.1 S_1 Sub-category: Misinterpretation of arrow symbolism

The S_1 difficulty was exposed through student interpretation of all three ERs (Fig.4.1). These students misinterpreted the arrow symbolism used in the ERs to represent the antigen (ER A) and its site of binding (ER C and D) to the antibody (Table 4.3). Examples of quotes showing the S_1 sub-category of difficulty are as follows:

1. S: I don't know where the antigens...which one's [are] the antigens or the antibody...I thought these are antigens [points to top two 'spheres']... [interview extract; ER A]

2. I: What is the antigen?
   S: According to this, the brown [spherical] part.
   [...]
   I: Say the antigen separates [student stated this earlier], how would this [diagram] look?
   S: ...You won't have this coloured region [points to red 'spherical' region], but you'd still have this line structure. [interview extract; ER A]

3. S: ...[the diagram] shows some structure of the antibody, these lines [L/H] ...and these are antigens...the balls, the big balls here [top two spheres]. [interview extract; ER A]

4. "Circles represent antigens. Lines represent bonds joining the antigens together." [response to probe 9; ER C]

5. "3-D structure of an antigen in a chain form.
 represents bonds between the antigens – since there are about 5 antigens illustrated. Between small molecules that make up the antigen, there are bonds as well." [response to probe 1; ER C]

6. I: How many binding sites are there?
S: There're lots of them...[points to multiple 'clefts' on perimeter of antibody structure on ER C]...fifty to one-hundred [binding sites].
I: So, how many antigens could be bound here?
S: Fifty to one hundred antigens. [interview extract; ER C]

7. "the diagram shows how primary structure of the antigen is converted to tertiary structure and it shows where and how the antigens bind to each other to form a chain. [response to probe 1; ER C]

8. "How an antibody attacks an antigen i.e. how they bind, location of different bonds within the complex of antigen and antibody." [response to probe 1; ER D]

9. "...The antigens seem to bind at the darker areas of the molecule." [response to probe 1; ER D]

10. "...The diagram also illustrates the Antigen binding sites that occur on the H-chains." [response to probe 1; ER D]

In quotes 1 - 3 above, it is evident that these students interpreted the antigens represented in ER A, as the top two spheres instead of the arrow-like elongated shape. As was mentioned in section 4.3.1, this difficulty could have been due to students interpreting "antigen" as a diagram label and, therefore, thinking that the arrow-like antigen shape was "pointing" to the antigen structure. Similarly, for ER C (e.g. quotes 4 -7), these students may have interpreted the arrow showing "binding specificity for antigen" as indicating either the actual antigen structure, many antigens, or separate components of an antigen, rather than merely an *antigen binding area* on the *antibody* structure. Students' interpretation of ER D (quote 8) also revealed a similar difficulty where it was suggested that the ER was representing an antigen-antibody complex, rather than the structure of the antibody. Finally, quotes 9 and 10 show that students probably misinterpreted the arrow form presented on ER D in thinking that antigens could *only* bind to the "darker" red and blue areas. At a superficial level, one can understand this difficulty because, due to the 2-D nature of ER D, the arrows do seem to be pointing only to the *dark* red and blue chains, instead of to the antigen-binding cleft.

The data showed that the S_1 difficulty initially emerged unexpectedly during free response probing. However, since it also emerged during more focused written and oral probes, it was classified at Level-3 as partially established and therefore, requires further research in other

contexts to become fully established (Level-4). The S_1 difficulty can be considered a sub-type of the **S** category since students misinterpreted the symbolism used to portray antigen binding sites on the antibody structure.

4.3.2.2 S_2 Sub-category: Misinterpretation of symbolism depicting disulfide bonds

Students who showed the S_2 sub-category of structural-type difficulties (Table 4.3) did not recognise the black-lined "ladder-like" features between heavy and light chains, as being representative of disulfide linkages in ERs A and B (Fig 4.1). Therefore, this difficulty was classified as a sub-type of the overall **S** category. The following examples of quotes displayed the S_2 difficulty:

1. "Heavy and light chains and [with] H-bonds between them." [response to probe 4; ER A]
2. "Represents the protein structure – Tertiary with beta pleated sheets joined by hydrogen bonds. Hydrogen bonds for the stability of the molecules." [response to probe 1; ER A].
3. S: ... they're [light/heavy chains] connected by hydrogen bonding.
   I: All right, could you show me where the hydrogen bonding is?
   S: Here [points to lower S-S bond]. [interview extract; ER A]
4. I: What is enclosed by this blue rectangle here [points to area]?
   S: I think they should be the same [types of molecules]...I think it [S-S bond] is made up of the same ... units which make up these [heavy/light chains]. [interview extract; ER B]
5. I: ...what can you tell me about what is enclosed in this blue rectangle here [points to blue rectangle]?
   S: ...this is the bridge...and it [the 'bridge'] has vital information about the affected areas. [interview extract; ER B]

Quotes 1-5 show that students interpreted the inter chain disulfide linkages between the heavy and light chains of the antibody as other structural components. For instance, three students (quotes 1-3) interpreted them as representing hydrogen bonding. Even though the student in quote 2 showed deep insight when stating that ER A inferred beta-pleated sheet conformation, s/he still thought that the ladder symbolism depicted hydrogen bonds between the H and L chains. The student who generated quote 4 for ER B thought that the "ladder" symbolism depicted the "same type" of molecules as those used to depict the H and L chains. This was probably due to the use of the same graphical means (black lines) to depict both structural elements. Lastly, the student who produced quote 5 thought that the black line in ER B represented a communication "bridge" between heavy and light

chains (see section 4.3.1.6 and Table 4.2), an example of inappropriate transfer (e.g. Brna *et al.*, 2001; Mayer and Sims, 1994) of knowledge to another domain.

The S_2 difficulty was unanticipated and, therefore, classified at Level-1, but its re-emergence during more focused probing and interviews allowed it to be re-classified at Level-3, as partially established.

4.3.2.3 S_3 Sub-category: Misinterpretation of symbolism depicting polypeptide termini

Students who showed the S_3 difficulty (Table 4.3), could not identify the N and C end termini of the heavy and light chains represented in ER A (Fig 4.1). This sub-category of the parent **S** category of difficulty was revealed in interviews in which students were specifically questioned about the feature enclosed by the green circle in ER B (Fig 4.1). The following interview extracts show this difficulty:

1. S: ...it is the start of the pathway...which transports information. [interview extract; ER B]
2. S: ...well that has come at the end of the strand...at the end of the strand is the phosphate group... phosphate and the sugar. [interview extract; ER B]
3. S: ...it's the site where...the elongation of this strand here [points], is supposed to continue. [interview extract; ER B]

It is clear from the above extracts that the students failed to interpret the graphical marking enclosed within the green circle as depicting an N-terminus of a polypeptide chain comprising the antibody structure. Instead, one student (quote 1) interpreted the graphical marking as the "start" of an information transport pathway. This same student's notion of the black lines representing an information pathway was previously discussed in relation to the P_6 difficulty (section 4.3.1.6). Other students (e.g. quotes 2 and 3) associated ideas of DNA structure and processing to their interpretation of the marking enclosed by the green circle on ER B. Students' invalid association of DNA to the ER will be given greater attention later in section 4.3.3.

The S_3 difficulty emerged during interviews only and was, therefore, classified at Level-2 as suspected and requires further investigations into its nature.

4.3.2.4 S_4 Sub-category: Misinterpretation of the "spheres" as representing other structural entities

Students who showed the S_4 sub-category incorrectly thought that the coloured ball-like 'spheres' in ER A, depicting variable and constant regions of heavy and light chains, were not part of the actual IgG, but represented other structural entities (Table 4.3). Examples of quotes that illustrated this sub-category of difficulty that are related to the overall S-type category are provided below.

1. "The coloured areas represent different areas...of red blood cells" [response to probe 5; ER A]
2. "Show[s] how 3 atoms are bonded to form a molecule. The antigen binds to the V region of the molecule. It shows that all 3 atoms are bonded by the C region..." [response to probe 1; ER A]
3. "What are those circles (3) seen behind the IgG antibody?" [response to probe 2; ER A]
4. I: What are those "balls"?
   S: Antibodies. [interview extract; ER A]
5. I: So these are three different entities [S had stated this earlier]?
   S: Ja [yes], like one antigen, one antigen and one antibody. [points to top spheres as Ag and bot. sphere as Ab]... It [ER A] also... shows that the antibody can work on more than one [Ag] at a time, so these two antigens [top 'spheres'] would be of the same type. [interview extract; ER A]

From the above data (quotes 1-5) it is clear that all five students interpreted the spherical components of ER A as representing other structural entities, rather than the V and C domains of one Ab molecule. Quote 1 suggests that the spheres represent red blood cells, quote 2 atoms, while in quote 3 the student is unsure of what the spheres represent but suggests another entity other than antibody. On the other hand, quotes 4 and 5 demonstrate that two students interpreted each of the spheres to be indicative of entire antibody structures. Finally, in quote 5, the student suggests that the bottom sphere in ER A is the antibody, while the two top spheres are antigen structures (see section 4.3.2.1).

The S_4 difficulty was initially suspected based purely on the visual analysis (Fig. 4.2, statements 4 and 5) performed by the researcher on ER A (Fig 4.1). Since focused probing and interviews subsequently confirmed the existence of the difficulty, it was classified at Level-3 as partially established on the Grayson *et al.* (2001) research framework.

4.3.2.5 S_5 Sub-category: Misinterpretation of the Y-shaped black lines

The S_5 difficulty was displayed by those students who misinterpreted the "Y-shaped", black lines in ER A as a type of backbone or support structure holding structural entities together (also see S_4 difficulty), rather than depicting the antibody's light and heavy polypeptide chains (Table 4.3). Examples of erroneous interpretations illustrating this difficulty are as follows:

1. "The coloured (grey) region represents different amino acid residues attached to the backbone (black line) of the antibody." [response to probe 6; ER A]
2. "Black lines [are] some form of bond or attachment holding the 3 cells together- blood cells, biconcave type shape. Differentiates between V and C regions" [response to probe ; ER A]
3. S: ...it [heavy and light chains] keeps these structures [3 'spheres'] together as you can see. [...]
   S: It [H/L chains] should keep these structures, the other molecules [spheres] together. [interview extract; ER A]
4. "The diagram is trying to represent... 2d [2-D] cross linking present in antibodies" [response to probe 1; ER A]

Quotes 2 and 3 above suggest that the black lines were a "bond" of sorts that allowed for three separate structural units to be attached to each other, while the student in quote 1 interpreted the black lines as being responsible for the attachment of amino acid residues to the Ab structure. Similarly, quote 4 shows the interpretation of antibodies being "cross linked" to one-another.

Since the S_5 difficulty was suspected based on the screening process done by the author described in section 4.2.2 (statement 8, Fig. 4.2), it was initially classified at Level-2. Subsequent exposure of the S_5 difficulty during focused written probes and interviews allowed it to be classified at Level-3 as partially established. Since students who showed the S_5 difficulty misinterpreted the graphical features depicting polypeptide chains in ER A, it was classified as belonging to the overall S-type category.

4.3.2.6 S_6 Sub-category: Misinterpretation and limited understanding of the level of protein structure represented

In the S_6 difficulty that was classified as belonging to the overall S category of difficulty (Table 4.3), some students incorrectly identified the level of protein structure depicted in ER C (Fig. 4.1) as being primary or secondary rather than tertiary. In addition, other students correctly stated that a tertiary structure was being represented, but displayed erroneous reasoning and limited conceptual understanding of this type of structure. The following written student quotes and interview extract constitutes evidence for these difficulties:

1. "Primary, because normally the tertiary and quaternary [structures] are more clear and in the case of this structure you can't see clearly." [response to probe 8; ER C]
2. "Secondary because pieces are forming a helix of double strands. They are not single strands. This is not a large complex." [response to probe 8; ER C]
3. "Tertiary. Shows all the disulfide bonds between the protein chains of the antibody." [response to probe 8; ER C]
4. "Tertiary structure, the structure consists of a folded chain (folded into a particular shape) but is only a single chain." [response to probe 8; ER C]
5. I: ... what level of structure, primary, secondary, tertiary, quaternary, does this diagram represent?
   S: [long pause] Tertiary... because there are only three parts...There are only three parts...one, two, three [points to lt variable region, constant region and rt variable region as three different parts]
   [interview extract; ER C]

The above quotes suggest that these students did not have a clear understanding of the different levels of protein structure, and in this case, how they pertain to an antibody molecule. Quotes 1 and 2 incorrectly identify the structure in ER C as depicting a primary and secondary level of structure, respectively. In contrast, quotes 3-5 correctly suggest that the antibody is being represented at the tertiary level of structure but contain unsound explanations. In quote 3 for instance, the student suggests that tertiary structure can be identified by always having disulfide bonds present while the student in quote 4 indicates that the structure is tertiary because it comprises only a single folded chain. Finally, the student in quote 5 has attached a "three part" structure to the idea of tertiary structure. Interestingly, even though the caption supplied to students with ER C (Fig. 4.1) clearly states a "tertiary" level of structure (see section 4.2.1), some students still identified the structure as representing primary and/or secondary structure (e.g. quotes 1 and 2), suggesting a diagram reading problem.

Related to the above difficulty with levels of protein structure, a study by Mbewe (2000) has shown that definitions of primary, secondary and tertiary structure of a protein are consistent across textbooks whereas the same does not always hold for *quaternary* structure. In this regard, it is generally agreed amongst the community of biochemists that a protein can exhibit quaternary structure if it consists of two or more polypeptide chains or sub-units that can be arranged in space as one ensemble. Nevertheless, when it comes to defining the *exact nature* of interaction between the polypeptide chains (i.e. covalent versus non-covalent interaction), the definition for quaternary structure has been shown to sometimes differ amongst textbook authors and biochemists (see Mbewe, 2000). Although the majority of biochemists are probably in solid agreement that a quaternary structure exists when at least two polypeptide chains are associated by covalent *or* non-covalent forces (e.g. Garrett and Grisham, 1995) other texts (e.g. Ritter, 1996; Bohinski, 1987) define quaternary structure of a protein as the arrangement of polypeptide chains where the forces between chains are of a non-covalent nature *only* (Mbewe, 2000). Interestingly, the International Union of Biochemistry and Molecular Biology make no direct reference to what type of inter-subunit interactions (i.e. covalent or non-covalent) *have to be* involved for a structure to exist at the quaternary level. Bearing this in mind, an antibody is a protein structure that has four polypeptide chains associated by *covalent* disulfide linkages. According to Garret and Grisham (1995) (and probably the majority of biochemists), IgG would constitute a quaternary arrangement, but other texts (e.g. Bohinski, 1987) define it as a tertiary structure. Strictly speaking, from the above analysis, it seems that for the IgG protein molecule anyway (e.g. Mbewe, 2000), a clear definition for its apparent *quaternary* or *tertiary* structure seems to be a point that can be debated. Similar silent debates have been documented for the structural level of classification of both insulin and chymotrypsin proteins among the biochemical community (e.g. Mbewe, 2000). Incidentally, in the original paper wherein the actual X-Ray crystallographic antibody structure represented in ER C (Fig. 4.1) was solved, the structure was stated by the authors as exhibiting a *quaternary* (not tertiary) structure (Silverton *et al.*, 1977). By contrast, the exact same structure is stated in Bohinski (1987), as having a *tertiary* structure (see ER C) and those biochemists that insist that chymotrypsin exhibits no quaternary structure (since the subunits interact though covalent links) would agree with this conjecture. In support of the above sentiments, and related to the S_6 sub-category, data from the present study also showed this interesting irregularity in definition, in that some students spoke about tertiary and others about a quaternary level of structure for the antibody structure depicted in ER C. The following student quotes indicate this divergence:

1. "Tertiary structure illustrating the chains and bonding of polypeptide chains." [response to probe 8; ER C]

2. "Tertiary – more than 1 [one] structure." [response to probe 8; ER C]

3. "Tertiary structure. It has a complex structure, with many folds – make up of a protein bonded together with hydrogen bonding." [response to probe 8; ER C]

4. "Quaternary structure because the structure is a giant molecule of a protein." [response to probe 8; ER C]

5. I: ...why would this [points to ER C] be quaternary [student stated this earlier]?
S: Because there is more than one peptide chain involved. [interview extract; ER C]

6. S: ...the quaternary structure is when... you have more than one amino acid sequence...binded to one another separately, via non-peptide bonds. The quaternary structure is the way in which the subunits... bond together to form a complete protein.
I: In terms of this structure [ER C]... what makes it quaternary [S stated this earlier]?
S: Ok, the subunits are the four chains because they're each a single peptide...the quaternary structure itself is maintained by the disulfide bonds... [interview extract; ER C]

From the above data, three students (quotes 1 -3) suggested that the antibody structure was being depicted at a tertiary level because more than one "chain" or separate "structure" was binding to another. In addition, the student in quote 3 supported the notion of a tertiary level of structure by pointing out that hydrogen bonding is responsible for the association of chains with one another. In contrast, the two students depicted in quotes 4-5 both thought that ER C was representing a quaternary level of structure. These students (quotes 4 and 5) supported their quaternary designation by suggesting that a "giant" molecule was being represented (quote 4) and that there was more than one chain involved in the structure (quote 5). The student in quote 6 supports his/her quaternary designation by suggesting that the polypeptide subunits are held together by covalent disulfide bonds. Thus clearly there is an urgent need to get biochemists worldwide to reach an all-encompassing consensus on a definition for quaternary structure (e.g. Mbewe, 2000). These findings, then, illustrate how science education research into student understanding can expose the need to clarify fundamental biochemical knowledge.

Since the S_6 difficulty was initially suspected based on a probe designed to expose its presence (probe 8, Fig. 4.7), its emergence during focused probing and interviews allowed it to be classified as partially established at Level-3 on the framework.

4.3.2.7 S_7 Sub-category: Misinterpreted ER of antibody structure as representing a T-cell

In a further structural-type difficulty identified as part of the parent S category, classified as the S_7 difficulty, two students during free-response probing thought that ERs A and C (Fig 4.1) were in some manner representative of a T-cell of the immune system (Table 4.3). This was shown by the following quotations:

1. "This [diagram] shows how or where antigen binds to the T-cells or MHC class 1 and that the substrate has carbohydrates." [response to probe 1; ER A]

2. "i. The binding of antigen to the T-cell
ii. Shows that the antigen has a specific shape for the binding of other molecules." [response to probe 1; ER C]

The student in quote 1 above may have inappropriately transferred his/her knowledge of T-cells to that of immunoglobulin structure. Since the plural was used when mentioning T-cells, the student may have thought that each spherical component depicted on ER A was representative of a "T-cell". Furthermore, the student may have been associating the letter "C" on ER A (Fig 4.1) with a "carbohydrate" region instead of a *constant* region. The second student (quote 2) may have superficially associated the "T-shaped" appearance of the antibody molecule on ER C with a "T-cell" instead of with an antibody molecule, but without further data, this remains speculation.

Since this difficulty emerged unexpectedly from the data, it was classified as unanticipated at Level-1 on the research framework. It thus requires substantial research in order to further clarify its nature.

4.3.2.8 S_8 Sub-category: Misinterpretation of symbolism depicting amino acids

In a further sub-category of the structural-type difficulties, coded S_8, it was found that some students incorrectly interpreted the graphical markings used to depict amino acid centres and residues on ERs C and D, respectively (Table 4.3). For ER C (Fig. 4.1), students misinterpreted the black 'circles' and 'lines' used to represent the α-carbon skeleton that

constitutes the polypeptide chains of the antibody molecule. For ER D, some students had trouble identifying the graphical marks (red and blue spheres) used to show the amino acid residues. The following examples of quotes illustrated this S_8 difficulty:

1. "  are Hydrogen bonds." [response to probe 1; ER C]
2. S: Some of the circles are proteins and some of them of sugars ...half protein and half sugar. [interview extract; ER C]
3. "  Oxygen
 bonds." [response to probe 9; ER C]
4. "Circles represent the active sites where the antigen binds, and lines represent different chains that make up the tertiary structure of amino acid[s]." [response to probe 9; ER C]
5. "Antigens are the dominating structures in this molecule, this is because they need to spread around to perform well i.e. tell the antibodies if there is any foreign diseases." [response to probe 1; ER D]
6. " ...I would expect the dark red H chain to be carrying oxygen and the dark blue chain to not be carrying O_2..." [response to probe 1; ER D]
7. "An IgG molecule, made up of H and L chains each formed by atoms." [response to probe 1; ER D]
8. "... Different colour coding for different sub-units are used to show location of certain atoms." [response to probe 1; ER D]

Quote 1 suggests that some students thought that the line joining two 'circles' in ER C represented hydrogen bonding. Furthermore, other students (quotes 2, 3 and 4) thought that the 'circles' on ER C, making up the α-carbon backbone, were representative of "oxygens" (quote 3), sugars (quote 2), or active sites where antigen could bind (quote 4). With regard to quote 2, even though IgG is often referred to as a glycoprotein (Chapter 3), there are only a few carbohydrate hexose units situated between the two C_H2 domains (Silverton *et al.*, 1977; Davies and Padlan, 1990), constituting only about 3% of the entire IgG molecule's composition (e.g. Roitt, 1997). Besides this fact, the sugar units were actually left out of ER C by Bohinski (1987, p. 161). Thus this student (quote 2) may not have been aware of the proportion of carbohydrate residues present on an antibody molecule. With respect to ER D, one student (quote 5) thought that the coloured 'spheres' depicted antigens, while another student (quote 6) thought the red spheres in the H chain were "carrying" oxygen. Furthermore, two students (quotes 7 and 8) identified the coloured 'spheres' on ER D as atoms. In this regard, space-filling ERs that depict protein structure (e.g. ER D) sometimes do represent the van der Waal radii of *all* the individual atoms (e.g. Lehninger *et al.*, 1993, p. 61; Garrett and Grisham, 1995, p. 58) making up the protein. Alternatively, the same types of

ERs sometimes just depict the alpha-carbon coordinates (e.g. Silverton *et al.*, 1977; Ritter, 1996) or just individual amino acid residues (e.g. Stryer, 1995, p. 376) constituting the structure. Moreover, sometimes these ERs exclude some atoms and show all others, like for example, showing all the atoms constituting a polypeptide but excluding the R-chains (side chains) of constituent amino acids (e.g. Ritter, 1996, p. 122). These various modes of representation of protein structure can cause misdirection when students try to visualise the order of magnitude represented by the graphical markings contained in ERs of abstract phenomena.

A focused probe (probe 9, Fig 4.7) was designed to further explore students' interpretation of ER C after the S_8 difficulty had emerged during free-response questioning. Since the S_8 difficulty was initially suspected based on the visual analysis performed by the researcher on ERs C (statement 6, Fig. 4.3) and D (statement 3, Fig. 4.4), it was classified at Level-3 on the framework of Grayson *et al.* (2001). The S_8 difficulty was considered as belonging to the general **S** category because those students who showed it misinterpreted the symbolism used to designate amino acid components of the antibody structures in ER C and D.

4.3.2.9 S_9 Sub-category: Binding sites on IgG are not identical

In the final sub-category of the structural-type difficulties (S_9), students thought that the antigen binding sites depicted on ER C and D were not identical in structure and that a particular IgG could bind two structurally different antigens (Table 4.3). Therefore, the S_9 difficulty was considered as being related to the parent **S** category of difficulty and was illustrated by the following examples of student quotations:

1. "...The binding sites are shown not to be the same in configuration therefore different shaped substrates [antigens] will bind to different binding sites..." [response to probe 1; ER C]

2. I: How does the structure of this antigen-binding site here compare to the structure of this antigen-binding site [points].
   S: This one [lt] is different from that one [rt], so it means a different antigen will bind to this one...and a different antigen will bind to this one ...so it means this site has different sequences of amino acids compared to that one, so it will have a different structure. [interview extract; ER C]

3. "To show that the two different sites of IgG are not of the same type." [response to probe 1; ER D]

Some students (quotes 1-3) upon interpretation of ER C and D thought that the antigen-binding sites are structurally different. Included in these misinterpretations were the notions that both binding sites were of different configurations (quote 1), and of different amino acid sequence or primary structure (quote 2). Thus these students thought that two completely different antigens could bind to the binding sites of the same antibody.

The S_9 difficulty was initially suspected from the visual analysis that was conducted on ER C (statement 2, Fig. 4.3). A focused probe was designed to investigate the author's suspicion (probe 11, Fig. 4.7) and subsequent exposure of the difficulty allowed it to be classified at Level-3 as partially established.

4.3.2.10 Sound interpretations of the ERs relative to the Structural-type category

In contrast to the structural-type (**S**) difficulties (Table 4.3), several students produced scientifically acceptable interpretations of the three ERs (Fig 4.1) when given the same probes. For example, in response to probe 3 (Fig. 4.6), where students were asked to draw which component of ER A represents antigen and which part represents antibody, one student generated the scientifically acceptable diagrams presented in Fig. 4.8 below.

It is evident from the SGD (Fig. 4.8 below) that some students could soundly depict those graphical markings that constitute antigen and antibody structure in ER A. In addition, consider the following quotes obtained from students' interpretation of ERs C and D:

1. "[ER C] Show[s] the binding sites for an antigen. The 3-D configuration of an antibody. Antigens bind only to two specific sites on an antibody – these two sites are found on either end of the molecule. [response to probe 1; ER C]

2. "It [ER D] shows the antibody structure, with its antigen binding sites and different chains making up the antibody." [response to probe 1; ER D]

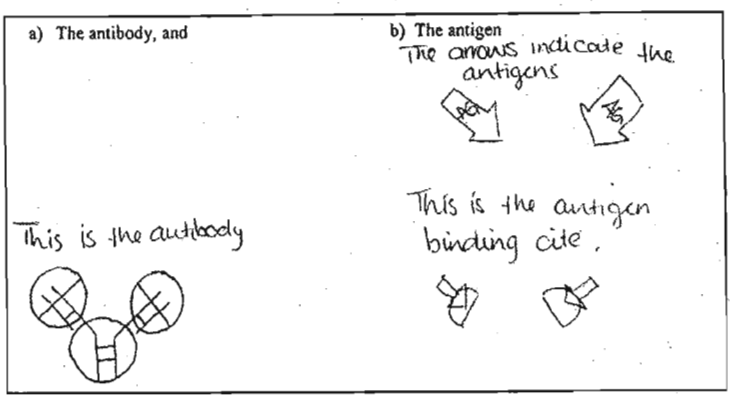


Figure 4.8 SGD obtained in response to probe 3 (Fig 4.6) correctly depicting the "black lines" and "spherical elements" as constituting antibody structure as well as soundly representing antigen components in ER A.

The responses above (Fig. 4.8, quotes 1 and 2) are in contrast with those students who were unable to resolve the role of the arrow symbolism in the ERs (S_1 difficulty) (Table 4.3). Even though Fig. 4.8 represents a sound graphical representation of the structural components depicted in ER A, the reader will notice that the same student has portrayed Ag structure as an arrow form. The prevalence of this notation as a possible source for the S_1 difficulty will be discussed in section 4.3.2.11 below.

The following correct responses corresponded to the S_2 difficulty, for ERs A and B:

1. "They represent polypeptide chains with disulfide bonds in between..." [response to probe 4; ER A]

2. I:...Tell me about the structure enclosed by the blue rectangle there [points].
[...]
S: Yeah, I believe they're disulfide bonds between cysteine residues...like a covalent bond between two sulphur groups on two cysteine residues. [interview extract; ER B]

It is clear from both quotes (1 and 2) that some students correctly interpreted the "black lines" between polypeptide chains as S-S bonds.

The following students (quotes 1 and 2) demonstrated a sound interpretation of the N-terminus represented within the enclosed green circle on ER B and were in contrast with students who showed the S_3 difficulty (Table 4.3):

1. I: Now, the structure within the green circle...
S:...well that would be the terminus of the light chain...part of the antigen binding site. [interview extract; ER B]

2. "It is a drawing of immunoglobulin. It indicates the V & C regions. It shows us where Ag the antigen will bind. The V-regions represent the N-terminus, C-regions the C-terminus." [response to probe 1; ER A]

Even though some students showed the S_4 and S_5 difficulties, by respectively considering the spherical shapes on ER A to be separate structural entities and/or the black "Y" shape to be a support structure of sorts (Table 4.3), some students were able to supply correct interpretations of these graphical markings. For instance, consider the following student quotes:

1. "The coloured areas are different areas of the black lines, it is a 3-D overview of what the black lines are made up of." [response to probe 6; ER A]
2. "The pink area → variable region of antibody, differs in every antibody (specific for an antigen) The grey area → constant region of the antibody which is the same for all antibodies..." [response to probe 5; ER A]
3. S: ...it [H/L chains] is the main protein backbone, the amino acid backbone...[points to H/L chains]. [interview extract; ER A]
4. "The black lines represent proteins – polypeptide chains of a protein." [response to probe 4; ER A]

From the interpretations provided above, it is evident that quotes 1 and 2 demonstrate a sound appreciation of the graphical nature of the coloured spherical areas that represent variable and constant regions of the polypeptide chains in ER A. In addition, quotes 3 and 4 affirm that the "black lines" are representative of the amino acid backbone constituting the Ab structure rather than a support "backbone" holding the spherical components together.

In comparison with students who manifested the S_8 difficulty (Table 4.3) by misinterpreting the small black 'circles' and 'lines' on ER C and/or the coloured spheres on ER D, examples of sound interpretations of these graphical markings across both ERs were as follows:

1. "...The rings are there to represent [the] carbon backbone..." [response to probe 1; ER C]
2. S: ...each circle is representative of ...the main amino acid kind of group...from amino acid to amino acid.
   [interview extract; ER C]
3. "...The small circles [spheres] that make up the chains represent amino acids which form the protein... Different colours to differentiate stereo arrangements of different chains." [response to probe 1; ER D]
4. "This diagram shows an immunoglobulin molecule. The round balls represent individual building blocks that make up the molecule (a protein therefore amino acids)... It shows the difference between H&L chains, and their relative positions." [response to probe 1; ER D]

The students who showed their sound interpretations in quotes 1- 4 above all suggested that the circle markings on both ERs C and D were representative of amino acid centres or residues rather than any other type of structural components.

Lastly, in contrast to students who thought the two antigen-bindings sites on an antibody were not identical (S_9 difficulty), an example of a quote showing a sound interpretation of the binding sites as being structurally identical is given below:

> "It [the antigen] is specific because there are two sites that the antigen can bind to; and the sites have identical but mirror-image constitutions." [response to probe 12; ER C]

In summary, the sound quotes discussed above suggest that, in contrast to the structural-type difficulties induced by the three ERs (Fig. 4.1), the ERs were nevertheless useful to various other students who yielded scientifically acceptable interpretations. In addition, evidence of sound scientific interpretations of the three ERs confirms the validity of the probes designed to generate data of relevance to the **S** category. Furthermore, the reliability of the probes as data-generating tools was supported by the fact that firstly, structural-type difficulties emerged on more than one occasion from more than one test (see section 3.4.4.2) and secondly, that structural-type difficulties emerged across all three ERs (Fig. 4.1).

4.3.2.11 Conclusion and possible sources of the Structural-type difficulty

The occurrence of the structural-type (**S**) difficulty across all student groups showed that there was a general difficulty in the student populations (Table 4.1) with interpreting how the structural features of IgG were externally represented and visually depicted in all three ERs (Fig. 4.1). With respect to the general S-type category, nine sub-categories of difficulty were identified in students' responses (Table 4.3). The S_7 sub-category emerged unexpectedly from the data and was classified at Level 1 on the Grayson *et al.* (2001) framework, while the S_3 sub-category was classified at Level-2 as suspected. All seven of the remaining sub-categories were classified at Level-3 as partially established. Thus we feel confident about the nature of these difficulties but further research is required to establish their occurrence across multiple contexts.

Upon analysis of the data generated across the student groups (Table 4.1) relative to each of the three ERs, it became evident that ER C showed the highest incidence for the structural-type category of difficulty with a prevalence of 70%. This was in contrast to ER A, which also showed a moderately high incidence of 50% followed by ER D, which manifested an incidence of 19% relative to the S-type category. Thus since the conceptual knowledge required to interpret these ERs was more or less the same, these results suggest that the nature of ER C caused students the most problems and ER D the least, when interpreting the symbolism representing structural features of IgG. It should be noted however, that for ER D, only free response probes were used to gather data. It is possible therefore, that the degree of influence reflected by the incidence shown by ER D, could increase should more focused probes be used in other studies.

Students who showed the S_1 difficulty for ER A, C and D (Fig. 4.1) struggled to resolve the function of the arrow symbolism used to graphically represent the Ag and its binding *location* on the Ab structure. As a consequence of the nature of these ER features, it is possible that students could not discriminate between those graphical markings that showed antibody components and those that showed possible interaction between Ag and Ab. In addition, the graphical depiction of the black "lines" on ER A could have been a source of confusion to those students who thought that the shorter ones represented structural components other than disulphide bonds (S_2 difficulty) and that the longer ones represented a type of support structure (rather than amino acid chains) holding "spheres" together (S_5 difficulty). By taking ER A at face value the rigid, frame-like appearance of the black lines does seem to imply a mechanical capability that is "supporting" the ball-like spheres and, *both* the disulphide bond and the polypeptide chain are represented as straight black lines. Related to the former, a source of the S_4 difficulty, wherein the shaded spheres depicting variable and constant regions of a 3-D structure on ER A were interpreted as separate structural entities, could lie in the artistic means chosen to depict V and C regions of the Ab. They look like separate, ball-like structures, leading some students to believe that they were not part of the antibody structure.

Students who showed the S_8 difficulty misinterpreted the graphical marks used to represent amino acids on ERs C and D. A source of this problem could lie in the fact that computer drawn chemical models present in textbooks vary quite widely, in terms of what the graphical units represent. Sometimes, the components making up the structure are representative of atoms, alpha-carbon centres (e.g. ER C), at other times as separate domains (e.g. ER A) or, as

complete amino acid residues (e.g. ER D). In addition, with respect to the S_9 difficulty, which showed that some students considered IgG's two antigen-binding sites represented in ERs C and D to be different in structure and in amino-acid composition, a possible source of this misinterpretation could be as follows. At a superficial level, based on the nature of the artistic embellishments, it does appear that the antigen-binding areas on the structures shown in ERs C and D are not identical. Upon perusal of ERs C and D, the fact that an IgG molecule has two structurally identical antigen-binding sites, may not have been immediately obvious to the respondents.

A possible overriding source for the existence of the S_1, S_2, S_4, S_5, S_8 and S_9 structural-type difficulties could be authors' and ER designers' confusing use of multiple 'conventions' to represent the same structural features in biochemistry. Many of the 'conventions' that are used appear not to be conventions at all but idiosyncratic representations (e.g. Lowe, 1987). For instance, while the disulfide bond is represented as a short straight black line in ER A, it is often represented in other ERs as "-S-S-" or as a yellow coloured bar, presumably to denote the presence of sulphur (this in itself could cause a misconception since not all chemical compounds containing sulphur are yellow in appearance). In other cases (e.g. ERs C and D), the presence of sulphur is not represented at all. Of course, what is represented in the ER is a function of what the author wishes to represent and on the pedagogical goal of the ER. However, in the light of the findings of this study, it is fair to assume that idiosyncratic graphical features do make it more difficult for students to decipher the necessary visual information.

Students reasoning processes could have also contributed to misinterpretations. For instance, when responding to probe 3 (Fig. 4.6) for ER A, students often represented the antigen as an *arrow* form (e.g. Fig. 4.8). The use of an arrow form to reason about the structural relevance of antigen may have itself been a source not only of the S_1 difficulty but also of the earlier discussed P_2 difficulty (section 4.3.1.2), where Ag was thought to enter the Ab structure. By inappropriately alluding to an arrow form to depict antigen structure, students may have incorrectly inferred a *direction* of entry into the antibody structure when processing the ERs. In addition, the S_1 data suggests that students relied heavily on perceptual organisation (e.g. Olivier, 2001) when interpreting the ERs. That is to say, students often relied on salient features to process the ERs and, as a result, neglected the deeper implications of the markings (e.g. Lowe, 2003, 1989). This was especially the case when students deciphered the Ag

structure on ER A, and the arrows on ERs C and D, as diagram labels that were "pointing" to Ag components. Du Plessis *et al.* (2003) and Schollum (1983) have shown similar student difficulties with arrow symbolism in other scientific contexts. In relation to the former, students who associated ideas of the T-cell when processing the antibody structure on ERs A and C (S_7 difficulty), may well have over relied on certain graphical markings when extracting meaning from the ERs (e.g. Lowe, 1989). In this regard, students who exposed this misinterpretation may have been performing surface-level reasoning when interpreting the ERs (Lowe, 1993a; Chi *et al.*, 1981) and may have simply associated the "T" shape of the antibody depicted in ERs C and D to a "T-cell" found in the human immune system.

Other than the graphical nature of the ERs, and the reasoning processes used to decipher them, difficulties are worsened if students do not possess the conceptual knowledge of what different visual conventions mean, do not correctly apply this knowledge or, are not aware that multiple possible conventions are available for depicting the same structural component. In this regard, a source for students' misinterpretation of the 'ends' of the 'black lines' as structures other than end-termini of polypeptide chains (S_3 difficulty) may have been caused by either, a lack of the necessary conceptual knowledge; linking of erroneous conceptual knowledge concerning "growth" or "information carrying systems" to the graphical symbols; or, the failure to bring the appropriate conceptual knowledge to the ER (e.g. Roth, 2002; Cheng *et al.*, 2001; Winn, 1993). In addition, as discussed in section 4.3.2.6, students' misinterpretation of level of protein structure (S_6 difficulty) could have originated from the conflicting propositional knowledge used by biochemists to describe level of structure. This might especially have been enhanced when certain debate surrounds definitions that pertain to the *quaternary* level of protein structure (e.g. Mbewe, 2000). Like the experts, it was clear that students also had differing opinions as to what they understood the level of structure of an antibody protein to be.

Overall, the above discussion suggests that sources of the S-type difficulty may emanate from the reasoning mechanisms used by students to decipher the ERs as well as from the nature of the conceptual knowledge that students used to interpret the ERs. A further potential source of the S-type difficulties could have been the multiple 'conventions' available for depicting the nature of the structural components in the ERs. In this regard, even though the nature of ER D may have been a contributing source for the latter, the nature of the graphical markings

contained in ER C, followed by ER A, seemed to have the most pronounced influence on students' misinterpretations pertaining to the S-type category of difficulty.

4.3.3 DNA-related difficulties

In the DNA-related difficulties (**D**), some students interpreted the three ERs (Fig 4.1) as representing a form of DNA structure and/or DNA processing. The prevalence of the general **D** category of difficulty, across the student groups and across all three ERs (Fig. 4.1) ranged from 4% to 19%. Respondents who showed one or more of the sub-categories of the DNA-related difficulty belonging to the parent **D**-type are included in the incidence range. Two sub-categories of this category as well as their classification on the Grayson *et al.* (2001) framework are presented on Table 4.4.

4.3.3.1 D_1 sub-category: Misinterpreting antibody structure as representing DNA structure or function

In the first sub-category coded D_1, some students misinterpreted certain graphical markings in the three ERs (Fig. 4.1) as being elements of DNA structure or DNA-related mechanisms (Table 4.4). Therefore, the D_1 difficulty was identified as belonging to the overall **D** category. Student quotes that illustrate the D_1 sub-category of difficulty, are shown below:

1. "This is meant to represent a DNA molecule, leading strands and a lagging strand of DNA..." [response to probe 1; ER A]

2. I: ...Can you tell me about what is enclosed by this blue rectangle?
   S: ...Ok, it will be two bases ...one purine and one pyrimidine... Ja [yes]...they're joined by hydrogen bonds. [interview extract; ER B]

3. "circles – DNA
   lines – protein structure". [response to probe 9; ER C]

4. "This represents the structure of a DNA molecule." [response to probe 1; ER D]

5. "...within each molecule there['re] bases." [response to probe 1; ER D]

6. I: ...Is there anything that you find confusing here [on ER A]?
   S: ...Ja [yes]. These black strands, and then if it is replicating, then why...why it [light chain] is on the other side of the strand [heavy chain]. [interview extract; ER A]

7. "Why the RNA template is on the outside of the DNA if the nitrogen base pairs of the DNA are dislodging from inside." [response to probe 2; ER A]

From the quotes above, it is evident that some students interpreted the antibody structures depicted in the three ERs (Fig. 4.1) as representing constituents of DNA structure or DNA-related processing mechanisms. This is clear in quotes 1 and 2 where both these students elude to ideas of "leading" and "lagging" strands as well as purine and pyrimidine base pairs when interpreting ERs A and B (Fig. 4.1). In addition, upon interpretation of ER C, a further student (quote 3) associated ideas of DNA structure to the markings showing α-carbon centres. Similarly, some students interpreted ER D as representing components of DNA structure (e.g. quotes 4 and 5).

Interestingly, two students (quotes 6 and 7) voiced concern as to why the shorter "DNA" strand was on the "outside" or on the "side" of the longer one. In textbook ERs that represent DNA structure and processing (e.g. Hames and Hooper, 1997, p. 137; Stryer, 1995, p. 804) leading and lagging strands are rightly shown as being *within* the replication fork formed by the parent strands. However on ER A, and in the context of IgG structure of course, the light chains (short black lines) are shown to be on the 'outside' of the heavy chains (long black lines) (e.g. Ritter, 1996, p. 154). Thus when an antibody's light chains are shown on the *inside* of the heavy chains (Garrett and Grisham, 1995, p. 924) this could potentially induce the D_1 difficulty, especially if the ER is already interpreted as a DNA-related component. Of course, it has been shown experimentally that the Fab arms are able to rotate (Brekke *et al.*, 1995) so, therefore, either representation of the light and heavy chains' location is scientifically sound. In addition, students who showed the D_1 difficulty for ER D (quotes 4 and 5) may have superficially associated the "coiled" nature of the heavy and light chains to a DNA helical structure.

The D_1 difficulty was exposed across all three ERs (Fig 4.1) and initially emerged unexpectedly from second-year written responses. Following further investigations with interviews, the difficulty was reclassified from Level-2 to Level-3, or partially established.

4.3.3.2 D_2 sub-category: Combining distinctly different concepts inappropriately

The second sub-category of the DNA-type difficulty, coded D_2, represents a situation where students inappropriately combined distinctly separate concepts from two different domains

when interpreting ERs A and D. In these cases, it was found that students erroneously combined concepts reserved for immunology with those of DNA structure or processing (Table 4.4). Consider the following student quotes, which showed the D_2 difficulty upon interpretation of ERs A and D:

1. "-DNA molecule replication
-Where the Ag bind[s] to the DNA molecule." [response to probe 1; ER A]

2. "Structure of DNA as it unfolds due to RNA interpretation of the DNA template. Ag is [a] protein molecule that is required according to the nitrogen base pairing of both the DNA and RNA. The whole process occurs in macrophages which are represented/shown by circles." [response to probe 1; ER A]

3. I: ...if I were to ask specifically about this line [light chain], what would you say?
S: It looks like a new replicating strand of DNA ...possibly replicating the same information which is on this C region [points], and then building it onto the Ag molecules so you're going to get identical molecules with the same DNA conformation...it is nucleotide synthesis, communication...[interview extract; ER A]

4. "This diagram is meant to show how DNA molecule IgG fights the antigen. It has an antigen binding site where antigen binds and will be killed after it is locked by this molecule" [response to probe 1; ER D]

5. "This diagram shows the DNA double helix molecule. How the long and short chains interact with each other and how and where the antigen binds." [response to probe 1; ER D]

It is clear from the above examples (quotes 1- 5) that students often inappropriately combined distinctly different concepts with those reserved for DNA structure and function (Table 4.4). For instance, in quotes 1 and 2 the students suggested that antigen structures were somehow involved in binding with DNA structures. In addition, the student in quote 2 suggests that the described process occurs in macrophage cells. Similarly, quote 3 suggests that DNA processing occurred for the purpose of "building information" onto an antigen molecule while quote 4 suggests that DNA is responsible for "fighting" the antigen. Lastly, the student depicted in quote 5 suggests that antigen binds to DNA components.

The data above provides evidence for the inappropriate fusing of immunology knowledge with that of DNA-related knowledge. As Grayson (2004) has shown in the context of students' understanding of electric circuits in physics, it is possible that the above students were unable to "disentangle" at least two distinctively different concepts from one-another when interpreting ERs A and D. Although in a biochemistry content area, ideas of immunology do intersect with those of DNA in some cases, for example, when the synthesis of IgG molecules through gene segments is considered (e.g. Hames and Hooper, 2000;

Kedzierski, 1992), it is clear that students who showed the D_2 difficulty merged these ideas inappropriately.

The D_2 difficulty initially emerged unexpectedly from the data. Its re-exposure during interviews allowed it to be classified from suspected to partially established at Level-3. The D_2 sub-category of difficulty was considered related to the parent **D** category because those students who exposed it inappropriately incorporated DNA-related knowledge into their interpretations of ERs A and D.

4.3.3.3 Sound interpretations of the ERs relative to the DNA-related category

In contrast with the DNA-related (**D**) difficulties (Table 4.4), that were exposed when students interpreted the three ERs (Fig 4.1), some examples of scientifically sound interpretations of the three ERs were as follows:

1. "...The lines represent the chemical structure of IgG. The V and C regions of κ & γ are also shown by the shading of the circles. [response to probe 1; ER A]
2. "This diagram is meant to show the specific binding sites of antibodies to antigens. It's supposed also to [to also] show the supercoiling of the tertiary structures of proteins." [response to probe 1; ER C]
3. "This [diagram] is meant to represent the 3-dimensional structure of the amino acid backbone of an IgG immunoglobulin, showing the "forking" of the molecule into two chains, each with their own antigen binding site..." [response to probe 1; ER C]
4. "Shows tertiary structure of IgG molecule. Shows how the chains coil around to give an overall structure. Also shows how chains interact together with the other chains i.e. which chain is closer to which." [response to probe 1; ER D]

Quote 1 correctly states that the "lines" representing polypeptide chains in ER A are composed of variable and constant regions. Quotes 2 and 3 also correctly suggest how the "supercoiling" and "forking" of the Ab represented in ER C is related to tertiary or 3-D protein structure, while quote 4 soundly suggests how the arrangement of the heavy and light chains are related to overall antibody structure in ER D. Further evidence for sound interpretations of the three ERs have already been provided in sections 4.3.1.7 and 4.3.2.10.

Another group of students did not expose the DNA-related difficulty but sometimes did mention that ER A reminded them of, or looked similar to, a DNA structure or process. In

doing so, these students provided the author with information on what may have contributed to the D-type difficulty in the other students. Consider the following scientifically sound quotations:

5. I: ... tell me about the hinge region [S referred to "hinge region" earlier].
S: ...The hinge region is actually where you have to cleave the antibody to get the Fab fragments...the hinge region actually shows where the molecule diverts, goes apart, just like a replication fork. It's like a replication fork but this time you are talking about antibodies not about DNA. [interview extract; ER A]

6. S: ...it's [ER A] like in DNA replication...
I: ... how does this [indicates diagram] relate to DNA?
S: I was just giving you an example. [interview extract; ER A]

Unlike the students who displayed the DNA-related difficulty, the students in quotes 5 and 6 were able to appropriately transfer their knowledge from one domain to another (Salomon and Perkins, 1989) and translate (e.g. Ainsworth *et al.*, 1998) between one representation (antibody structure) and another (DNA structure). For these students, the visual appearance of the Y-shaped antibody allowed them to draw a graphical analogy with DNA replication, even though they soundly suggested only a visual similarity. Thus the data above served to inform the author on what exactly may have induced the DNA-related difficulty.

4.3.3.4 Conclusion and possible sources of the DNA-related difficulty

Data corresponding to the DNA-related (**D**) category of difficulty suggested that some students incorrectly interpreted the three ERs as representing a form of DNA structure and/or processing. Within the D-type category of difficulty, two sub-categories of difficulty emerged from the data (Table 4.4). Both the D_1 and D_2 sub-categories were classified at Level-3 on the Grayson *et al.* (2001) framework as partially established. A discussion of the potential sources of the DNA-type difficulties, across both sub-categories, is presented below.

When data across the student groups (Table 4.1) was analysed with respect to each of the three ERs in conjunction with the D-type responses, it was found that ER A, D and C showed incidences of 40%, 10% and 4%, respectively. Thus ER A contributed the most and ER C the least to the D-type category of difficulties.

The nature of the graphical markings constituting the black lines, "spheres" and arrow-like antigens on ER A seemed to be a major source of confusion. It is evident that the black lines

representing the "Y-shaped" heavy and light chains in ER A look similar in appearance to an actual DNA replication fork that shows "lagging" and "leading" strands. In addition, the "supercoiled" arrangement of the heavy and light chains in ER D looks similar to ERs that represent actual DNA components. In the latter, DNA structure is often depicted in a helical nature, with molecular chains twisted around each other in a double helix (e.g. Hames and Hooper, 2000, p. 150). Furthermore, when considered at face value, the "forking" of the polypeptide chains in ER C and the "supercoiling" shown at the base of the Fc region of the Ab molecule, do show visual characteristics similar to ERs that represent DNA structure. The above graphical features of the three ERs may have been one of the sources contributing to the DNA-related category of difficulties.

In addition to the nature of the artistic embellishments on the ERs (Fig. 4.1), students processing mechanisms responsible for interpreting these graphical features may also be a possible source of the DNA-related difficulties. In this regard, for ER A, students' superficial processing (e.g. Lowe, 1994a; Egan and Schwartz, 1979) of the graphical markings described above may have been a source of the D_1 difficulty. In addition, students' inappropriate connections to other concepts in biochemistry when reading ERs A and D probably induced the D_2 difficulty. Evidence for such inappropriate connections during students processing of ERs A and D were found in those quotes in which students thought that antigens were able to interact with DNA. Furthermore, another source of the D_2 difficulty could be that some students were erroneously combining or fusing one distinct concept (e.g. Ab-Ag interaction) with another distinct concept (e.g. DNA structure) when processing the ERs. In this regard, these particular students were probably unable to disentangle distinctly different concepts from one another (e.g. Grayson, 2004) when deciphering the ERs.

In addition to the role of the graphical nature of the ERs and students' processing mechanisms towards contributing to the DNA-related difficulty, students' conceptual understanding may have also been a source of the problem. Prior to this investigation, second-year students (Table 4.1) had just completed a module on nucleic acids in which they had been exposed to ERs of DNA replication and synthesis (e.g. Stryer, 1995). It is possible that the students who showed the DNA-related difficulty were inappropriately transferring their newly constructed conceptual knowledge (Salomon and Perkins, 1989) of DNA elongation or processing to the context of IgG structure. For both the former, this inappropriate transfer of knowledge may be a consequence of a surface-level processing of the ERs (e.g. Cheng *et al.*, 2001).

4.4 Summary and Conclusions

The research findings of the study reported in this chapter identified three general categories of difficulty with students' interpretation of three textbook ERs (Fig. 4.1) depicting antibody structure and interaction with antigen. The three general categories that emerged in the study were the process-type (**P**), structural-type (**S**) and DNA-related (**D**) difficulties. As part of the general categories of difficulty, seventeen sub-categories of difficulty emerged from the data. Each sub-category of difficulty was individually classified on the Grayson *et al.* (2001) research framework, according to how much was known about the nature of each difficulty. Classifications on the research framework ranged from Level-1 through to Level-3 (see Tables 4.2 - 4.4). Further research of the difficulties in a different context (i.e. course, institution and/or student sample) will enable the difficulties to be classified at the highest level on the framework, as established at Level-4.

When incidences of the three categories of difficulty were calculated, relative to *each ER* used in the study, it was shown that different ERs played a greater role in causing a particular difficulty particularly as the conceptual knowledge required to interpret all the ERs was highly similar. For instance, ER A induced the highest incidence for the **P** category at 70% followed by ER C at 50 % and ER D at 7%. By contrast, ER C and ER A caused the highest incidences for the **S** category difficulty with values of 70% and 50%, respectively, while ER D showed an incidence of 19%. Lastly, ER A caused most students to reveal the **D** category difficulty at 40% incidence followed by ER D (10%) and ER C (4%). Thus these incidence values provide an indication of the degree in which the nature of the graphical markings represented within each ER contributed towards a particular category of difficulty. It is clear from the above values that the visual markings in ER A and ER C caused the most problems for students, with ER A having the most negative influence out of the three, across all three categories. Even though ER D showed relatively low incidences in comparison, students interpreted ER D through free-response probing alone and, therefore, the values provided above may not be a complete reflection of the contribution of ER D towards student difficulties. This is because with free response probing not all students will necessarily reveal a difficulty that they might have. Thus for free response probes incidences would be low values. Lastly, the "order of presentation" of the ERs to students during data collection (see Table 4.1) might have contributed to the relative incidences of the difficulties revealed for the

three ERs. For example, ER A given first to students (see Table 4.1), may have influenced their subsequent interpretations of the other ERs used in the study and so on. The author is uncertain to what extent this was a factor as it was not investigated in the present study. This could be a topic of future research in which the actual source of the difficulties could be further clarified.

In consideration of the above incidences relative to each ER, analysis of the data suggested that the *nature of the ER* and its graphical markings played a major role in students' ability to successfully interpret them. For ER A, the arrow-like depiction of antigen as both pointing at the space between the light and heavy chains and being of the same width as the space; the "ball-like" graphical means used to depict V and C regions of heavy and light chains; the use of red-like colouring to represent variable regions of the Ab; and, the black "lines" used to denote polypeptide chains as well as disulfide bonds, all contributed to categories of difficulty. For ER C and D, the graphical nature of the arrows used to indicate possible areas for antigen-antibody interaction often caused induced difficulties when students interpreted them as indicating a point of entry for the antigen molecule. Furthermore, the graphical marks used to represent amino acids on ERs C and D were often misinterpreted, while the "supercoiled" arrangement representing the heavy and light chains in ERs C and D also misled some students. Moreover, at a superficial level, it does appear that the antigen-binding areas on the Ab structures shown in ERs C and D are not structurally identical. Lastly, across ERs A, C and D, students often struggled to resolve the function of the arrow symbolism used to graphically represent the Ag and its binding *location* on the Ab structure. Consequently, students struggled to discriminate between those graphical markings that showed antibody components and those that showed possible sites for interaction between Ag and Ab.

In addition to the nature of the ER and its graphical markings being a major source of student difficulties, the data showed that students' *reasoning processes* also had a large effect on their ability to successfully interpret the ERs. In this regard, it was found that students often focussed on surface-level features of the ERs when extracting meaning from them (e.g. Lowe, 2003, 1996; Kozma and Russell, 1997). This surface-level reasoning (Chi *et al.*, 1981) was characterised by students relying heavily on the visuospatial information displayed on the ER to decipher it (Cheng *et al.*, 2001; Olivier, 2001; Lowe, 1996, 1993a). As a result, students often relied on salient features to process the ERs and neglected the deeper implications of the markings (e.g. Lowe, 2003, 1989; Olivier, 2001). In addition, students often inappropriately

transferred (e.g. Brna *et al.*, 2001; Mayer and Sims, 1994; Salomon and Perkins, 1989) their knowledge from one context to another and therefore, struggled to translate (e.g. Ainsworth *et al.*, 1998) between one representation and another. Furthermore, students were found to interpret the ERs literally instead of recognising the stylised nature of the ERs (e.g. Lowe, 1989) with the result of many students over generalising when deciphering them (e.g. Hill, 1990). The latter reasoning process was exaggerated when students' processed the graphical markings in a superficial manner (e.g. Lowe, 1994a; Egan and Schwartz, 1979). Lastly, some students erroneously combined or fused one distinct concept with another distinct concept when processing the ERs. These particular students were probably unable to disentangle distinctly different concepts from one another (e.g. Grayson, 2004) when deciphering the ERs.

Besides the nature of the ER and students' reasoning processes being major sources of student difficulties, analysis of the data revealed that the nature of students' *conceptual knowledge* also influenced their ability to successfully interpret the ERs. For example, students' erroneous conceptual knowledge (or the incorrect application of it), during interpretation of the three ERs, may have contributed to misinterpretations such as antigens being able to enter antibody structures and antibodies themselves being responsible for destroying antigens. In addition, the inappropriate use of specific scientific terminology such as "binding site" and "active site" may have also been a major source of the difficulties. Furthermore, students' lack of the scientific knowledge necessary for interpreting the ERs (e.g. the knowledge of what certain symbolism meant) or bringing inappropriate conceptual knowledge to the ER (e.g. Roth, 2002; Cheng *et al.*, 2001; Winn, 1993) such as ideas of "growth", "information carriers" and "DNA elongation and processing" were also sources of the difficulties. Lastly, the sometimes-conflicting propositional knowledge used by biochemists also had a negative influence on students' interpretations. An example of this problem was shown by the conflicting scientific definitions provided by both students and experts for quaternary protein structure.

With respect to the evidence provided above, we believe that the data indicates at least three factors that play a major role in students' ability to interpret ERs in biochemistry. These factors are students' ability to *reason* with the ER or with their own conceptual knowledge, students' understanding (or lack thereof) of the *concepts* of relevance to the ER, and the *mode* in which the desired phenomenon is represented in the ER. These three factors often appear

to be interdependent, making it difficult to establish which factor is playing the major role. With respect to the findings reported in the current chapter, there was no definite way in which to observe to what degree each of the factors affected ER interpretation. It was uncertain which of either; students' conceptual knowledge of relevance to the ER (e.g. Ametller and Pintó, 2002; Cheng *et al.*, 2001), the role of the visual markings themselves (e.g. Lowe, 1993a) or, the role of students' employed reasoning processes (e.g. Cox and Brna, 1995) played the most pronounced role. A further complication was that the data also confirmed the presence of all three factors across all the categories of difficulty, but in varying degrees.

In view of the above discussion, we considered it useful to try to resolve each factor independently in order to develop a clearer idea of where the difficulties lie, so that we could further investigate their sources and be in a position to suggest possible remediation. To achieve this, we realised that a suitable instrument was required to gather data pertinent to each of the factors so that their existence and influence upon one another could be further confirmed. The following chapter deals with the design of such a research instrument.

5 A THREE-PHASE SINGLE INTERVIEW TECHNIQUE (3P- SIT) FOR GENERATING EMPIRICAL DATA ON THE FACTORS AFFECTING STUDENTS' INTERPRETATION OF ERS

5.1 Introduction

In Chapter 4, we presented the identification and classification of several students' difficulties with the interpretation of three textbook ERs used in the teaching and learning of biochemistry. This in turn, led to the identification of a range of possible sources of such difficulties and, therefore, the proposal of three factors affecting students' ability to interpret ERs in biochemistry. The possible factors are, students' ability to *reason* with the ER and with their own conceptual understanding (coded **R**), students' understanding (or lack thereof) of the *concepts* of relevance to the ER (coded **C**), and the *mode* in which the desired phenomenon is represented in the ER (coded **M**). To gain greater insight into the nature of the **C**, **R** and **M** factors, and to confirm their validity, we required a specific, if necessary customised, instrument that would yield the necessary empirical data. In this regard, since the clinical interviews had proved to be a powerful research tool (see section 3.4.2.3) for identifying the difficulties reported in Chapter 4 we decided to design an interview technique that would serve our specific purpose.

The aim of this aspect of the study was, therefore, to address the third research question (Chapter 1) namely, how might we obtain empirical data to further investigate the nature of the factors affecting students' ability to interpret ERs? Towards achieving this aim, we developed, and then piloted, a clinical interviewing technique with which to generate data on each of the factors. The results of this developmental and design process are presented in this chapter together with results from the pilot study employed to test the instrument.

5.2 Basic design, structure and rationale of the clinical interview instrument

The overall purpose of the design of the current interview instrument was to provide a window into an individual's knowledge and reasoning processes (e.g. Beilfuss *et al.*, 2004; White and Gunstone, 1992) of relevance to concepts that represent antibody structure and primary binding to antigen. In general, by adopting a Piagetian approach towards gathering student responses, the interview instrument was designed to be *clinical* in nature (e.g. Bukatko and Daehler, 1992), where interview questions are modified in response to a subject's outputs as part of extracting deeper response patterns. The rationale behind this design was that we required the instrument to be an information-gathering device that could serve to expose both the *nature* and *extent* of an individual's conceptual understanding (e.g. Posner and Gertzog, 1982) and reasoning processes (e.g. Kozma, 2003) as well as data on the effect of the mode of representation on such understanding and reasoning.

In pursuing the above rationale, on the one hand, the author was interested in allowing the interviewee to 'speak their minds' while on the other hand, the author was interested in collecting specific information. In this regard, interview methods that are used in science education research adopt a wide array of interview techniques (see Chapter 3, section 3.4.2.3) in an effort to gauge students' conceptual knowledge (e.g. Novick and Nussbaum, 1978), concept construction (e.g. Posner and Gertzog, 1982) and ways of reasoning (e.g. White and Gunstone, 1992). Often, the interview consists of an informal, one-on-one, neutral, and two-way interaction between the student and the researcher (e.g. Simonneaux, 2000) where a flexible and *semi-structured* interview approach is often employed (e.g. Sumfleth and Telgenbüscher, 2001). By adopting a similar semi-structured approach in the current work, the clinical instrument was designed to be flexible in nature such that interview probes could be modified according to student response patterns that emerged during an interview session (e.g. Rubin and Rubin 1995; Posner and Gertzog 1982). The rationale behind this approach was that the initial emphasis should be on gathering free-response data before delving into patterns of interest (e.g. Ametller and Pintó, 2002) should they emerge. By employing the general intentions offered by semi-structured interview protocols in the current instrument, questions were designed that could be modified or adjusted based on the response patterns

that emerged (e.g. Cohen *et al.*, 2000; Rubin and Rubin, 1995) while the interviewer remained neutral at all times.

The design of the instrument reported in this chapter was divided into three interview "phases" the structure of which is shown in Figure 5.1. Upon execution of the instrument, there is a progression through each of the phases from, Phase 1 to Phase 2 and then to Phase 3 (Fig. 5.1). All three phases that comprise the instrument can be executed in a single interview sitting that lasts for approximately one to one-and-a-half hours. Based on this design, the author has termed the current instrument the *Three-Phase Single Interview Technique* (3P-SIT). Phase 1 of 3P-SIT (Fig. 5.1) has the primary objective of exposing a student's conceptual understanding about a scientific idea, for example, antibody structure and its interaction with antigen. Phase 1 is conducted *prior* to the student being exposed to any ER of interest (Fig. 5.1) and is concerned with extracting as much as possible of the conceptual knowledge that a student holds about a particular scientific construct, before the student interprets an ER representing the same scientific ideas. Phase 2 of 3P-SIT (Fig. 5.1) has the objective of probing a student's reasoning processes *during* the interpretation of an ER (e.g. Fig. 5.2 E, F or G) as well as any changes in their conceptual knowledge following interpretation of an ER. Lastly, Phase 3 of 3P-SIT (Fig. 5.1) requires students to evaluate and critique the ER in question (e.g. Fig.5.2 E, F or G). Thus Phase 3 allows the researcher to generate information about the role, effect and nature of the ER *in isolation.* Such information is also supplemented by evaluation of the ER by experts.

Overall, when conducting a 3P-SIT interview, the researcher moves through the three interview phases with an emphasis on first generating uninhibited and natural responses from students and then on delving deeper into those areas where the researcher believes interesting patterns reside. Where relevant, the interviewer probes further into certain conceptual difficulties or particular patterns of reasoning. At all times, the interviewer remains neutral about correct and incorrect responses and ensures that the student is not led into giving a particular response. The rationale behind the structure of 3P-SIT is that it is a flexible and systematic instrument. The instrument consists of probes that allow responses to emerge naturally and impartially.

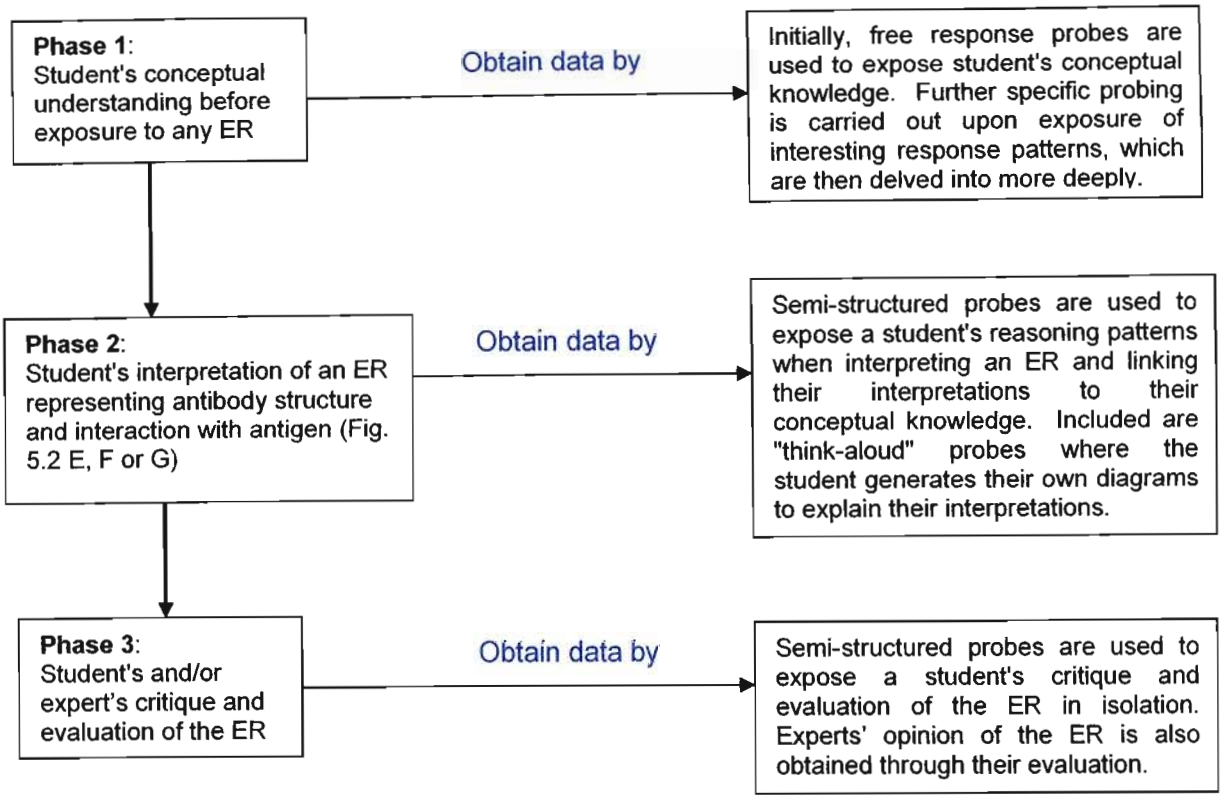


Figure 5.1 Overview of the structure and protocol of 3P-SIT

In the next section (5.3), we describe the participants and ERs, and in section 5.4 the pilot study, used to test 3P-SIT for its usefulness in generating empirical data that allows researchers to further investigate the **C**, **R** and **M** factors. In doing so, we provide examples of probes customised for each of the interview phases and the rationale behind their design. We also present selected student responses and show how the data can be analysed to expose information corresponding to each of the factors.

5.3 Participants and ERs used to test the instrument

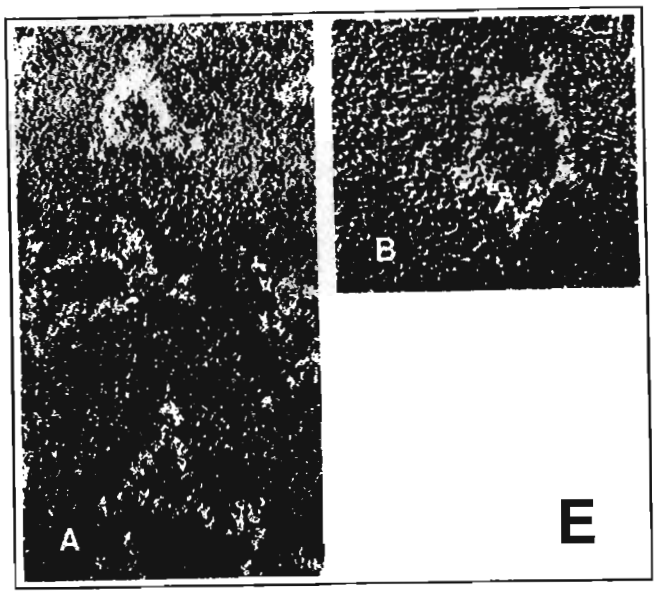
The 3P-SIT instrument was developed and tested from 2000 to 2001 using data obtained from six students at the University of KwaZulu-Natal, South Africa during November 2000. The six student participants had varying biochemistry content knowledge. All six participants had completed a full second-year level biochemistry course that included introductory immunology, as well as at least one biochemistry module at the third year level. The 3P-SIT

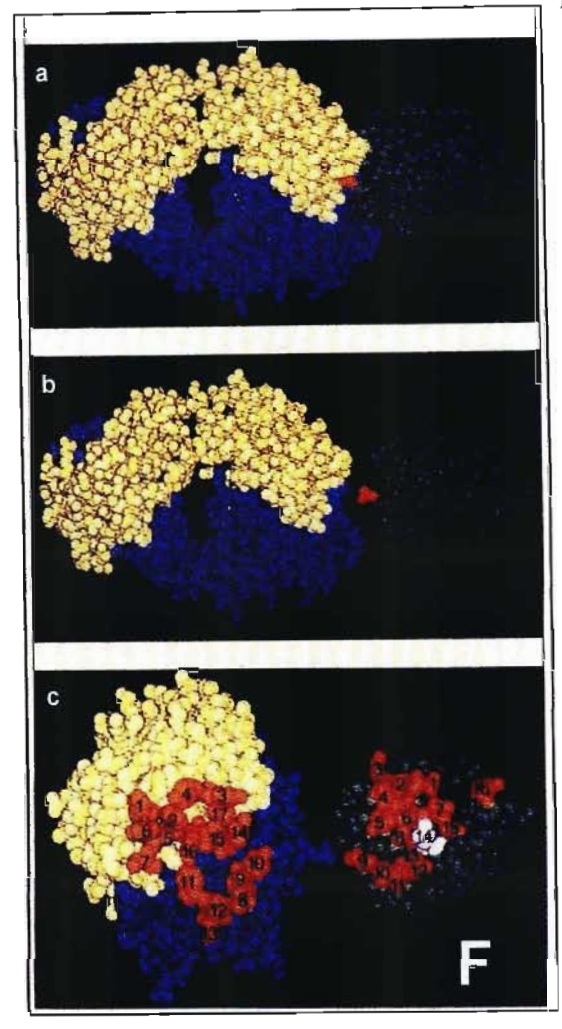
instrument was tested by obtaining students' responses to one of three different ERs (Fig. 5.2) during the interview phases.

For the convenience of the reader, a flip-out page of all three ERs (Fig. 5.2) used in this study is supplied on p. 130.

Two interviews, each with a different participant, were conducted for each of the three ERs (Fig. 5.2), giving a total of six interviews. None of the six participants was interviewed more than once. Two of the three ERs (Fig. 5.2 E and F), used to pilot the interviewing instrument, were obtained from the immunology textbook (Roitt, 1997) prescribed for the course, while a colleague (Jackson, pers. comm.) provided the remaining ER (Fig. 5.2 G). With regard to Fig. 5.2 G, students were familiar with these types of ELISA representations in that the immunology course required them to generate similar ERs during practical work.

The three ERs shown in Fig. 5.2 (E - G) are multiple representations of antibody-antigen interaction that fall on a real to abstract continuum (e.g. Kress and van Leeuwen, 1996; Wheeler and Hill, 1990; Alesandrini, 1984; Fry, 1981; Dwyer, 1967). The electron micrograph (Fig. 5.2 E) can be considered a "real" depiction of antibody and antigen interaction, the space-filling model (Fig. 5.2 F) a "semipictorial" (stylised) representation of antibody-antigen interaction and the graphical plot (Fig. 5.2 G) an "abstract" portrayal of antibody-antigen interaction. The electron micrograph (Fig. 5.2 E) shows trimer and pentamer complexes formed when Y-shaped IgG antibodies bind to the divalent hapten dinitrophenyl (DNP) (Roitt, 1997; Valentine and Green, 1967). Fig. 5.2 F represents a three-dimensional, space-filling display of the binding of an antigen (lysozyme protein) to a Fab fragment of an IgG antibody molecule (Roitt, 1997; Amit *et al.*, 1986). Lastly, Fig. 5.2 G is a Cartesian graph of the quantitative results obtained from an enzyme-linked immunosorbent assay (ELISA) (Jackson, pers. comm.) of the binding interaction between antibody and antigen molecules. Each coloured curve represents results obtained at different weeks of an immunisation schedule. Absorbance at 405 nm is plotted against the negative logarithm of antibody concentration. The presentation of ER G to students also included insertion of a block letter 'P' on the blue curve at an approximate coordinate of 0.33 on the y-axis and 1.75 on the x-axis. A further block letter 'Q' was inserted just after the peak on the blue curve.





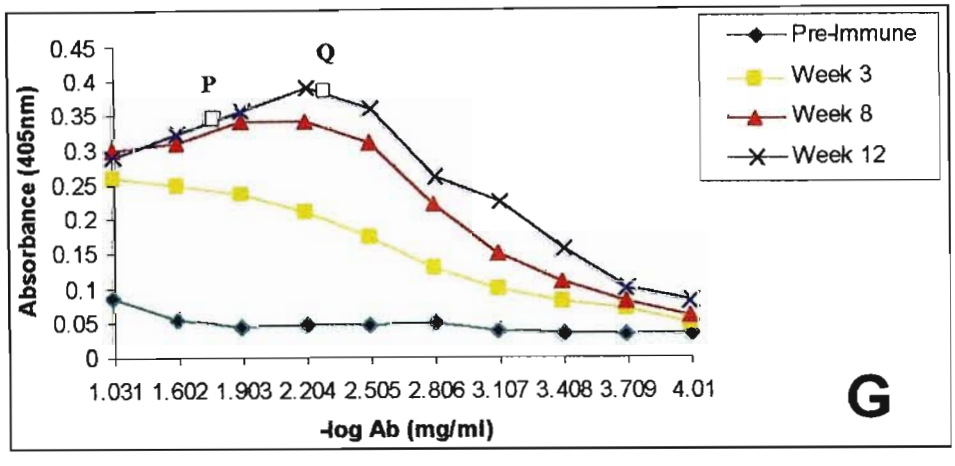


Figure 5.2 Three multiple ERs of antibody-antigen interaction.
(**E**): Electron micrograph (x 1 000 000) of complexes formed on mixing divalent hapten with anti-hapten antibodies. The hapten links together the Y-shaped antibody molecules to form trimers (A), and pentamers (B) (Roitt, 1997); (**F**): Space-filling model showing Fab antilysozyme and lysozyme molecules fitting snugly together. Antibody heavy chain, blue; light chain, yellow; lysozyme, green with its glutamine 121 in red. Fab and lysozyme models are also shown pulled apart in the second frame (Roitt, 1997); (**G**): Antibody response curves obtained from an ELISA showing the relationship between absorbance (405nm) and antibody concentration (mg/ml). Three booster shots were administered and the antibodies collected at the weeks indicated in the text box (Jackson, pers. comm.)

Each of the ERs in Fig. 5.2 will be referred to as "ER E", "ER F" and "ER G", respectively. For each ER, both the ER and its caption were supplied to students during all interviews but only one ER was supplied at a time. Captions supplied were as provided in Fig 5.2 except for the following modification. For ER F, the statement, "In the third frame, both molecules have been rotated 90^0 about a vertical axis and contact residues are shown in red and Gln 121 in light purple" (Roitt, 1997, p. 376), was removed as we wished to gauge students' own interpretations in this regard.

5.4 Probe design and analysis of students' responses

In this section, we provide examples of probes used within each of the 3P-SIT phases and the rationale behind their design. We also present selected student responses, corresponding to the probes and the analysis thereof to demonstrate how 3P-SIT can be used to generate data corresponding to each of the three factors. Since the probes, the responses and their analysis are presented together in sections 5.4.1 - 5.4.3, it is appropriate to first consider the following general approach to the analysis of students responses obtained during 3P-SIT.

During testing of the designed instrument, all interviews were both audiotaped and videotaped (e.g. Hull *et al.*, 2003; Pavlinic *et al.*, 2001; Sumfleth and Telgenbüscher, 2001). The data collected during the interview sessions consisted of video segments, audio-transcripts, student-generated diagrams (SGDs) and researcher-generated field note items. Data was analysed by means of a qualitative, iterative and inductive method (Chapter 3) in which categories of responses emerged from the data themselves, rather than being pre-determined (e.g. Anderson and McKenzie, 2002; Grayson *et al.*, 2001), and in which patterns were uncovered and "made explicit from embedded information" (Lincoln and Guba, 1985, p. 203). With reference to this approach, analysis of the data could best be described as a "descriptive synthesis" rather than a process of data reduction (McMillan and Schumacher, 1993, p. 480).

The following general seven-step process, not necessarily in a linear manner, was used to analyse the data. Firstly, the interviewer made paper-based field notes consisting of any relevant issues that were observed while the interview was in progress. Secondly, each audiotape was transcribed and the relevant data electronically assigned to Phase 1, 2 and 3

categories of 3P-SIT. Thirdly, the researcher used inductive analysis (Chapter 3) of the transcripts, to formulate common patterns of student responses into categories. During this process, in addition to the field notes, the researcher made further notes on the printed transcripts. Fourthly, the researcher analysed the diagrams that were generated by the respondents. Analysis of these SGDs helped facilitate the diagnosis of students' reasoning processes and the extent of their conceptual understanding (e.g. Glynn, 1997; Kindfield, 1993/1994). This approach is supported by other research in which students' *drawing* of their mental images has proved to be a powerful way of measuring thought processes (e.g. Beilfuss *et al.*, 2004; Reiss and Tunnicliffe, 2001; Lowe, 2000, 1988a, 1987; Gobert and Clement, 1999; Novick and Nussbaum, 1978). Fifthly, the researcher used the video footage to supplement the electronic transcripts with additional, relevant information pertaining to students' interpretation of the ERs (e.g. pointing on the ER). This allowed the researcher to gain more information about students' mental processing of the ERs. The ER-related observable behaviours, that were inserted into the transcripts, were those such as students' specific sequence of diagram construction; student's modification, annotation or rejection of their diagrams; their gestures such as 'pointing' and 'indicating' on the diagram and various other observable behaviours (e.g. Kindfield, 1993/1994; Lowe, 1993).

Sixthly, the interrelationships between the data across the 3P-SIT phases were investigated in an attempt to measure how correctly the ER was interpreted and, whether sound or unsound learning had occurred after exposure to the ER. In this regard, data corresponding to Phase 2 (reasoning with the ER) were compared with the response patterns from Phase 1 (conceptual knowledge before exposure to an ER), and similarly for Phase 3 (critique and evaluation of the ER). The success of the *interpretation* of the ER was measured by comparing the student's conceptual knowledge *after* exposure to the ER (Phase 2) to the conceptual (propositional) knowledge represented by the ER. In addition, evidence of any *learning* from the ER, was measured by comparing the student's conceptual knowledge *after* exposure to the ER (Phase 2) to the student's prior knowledge, obtained during Phase 1. Through the latter, it could also be determined whether the construction of a new conception, an alternative conception or a modification of an existing conception had taken place. In addition, through this comparative analysis, we could monitor how existing conceptions *modulated* reasoning with a particular ER (e.g. Lowe, 1996; Winn, 1993), especially when the ER was novel to a student. By comparing data generated from Phase 3 with that of Phase 2, insight could be

gained into how the actual visual-spatial markings on the ER influenced and modulated students' reasoning processes.

Seventhly, similar categories and patterns of difficulties obtained from transcripts and SGDs across the three ERs (Fig. 5.2) were pooled and analysed in order to identify categories that were common to particular students regardless of the nature of the ER. For example, we investigated evidence of particular reasoning (e.g. analogical reasoning) and conceptual patterns (e.g. misconceptions about antibody binding sites) among all students, regardless of the ER in question.

5.4.1 Phase 1: Generating and analysing data corresponding to students' conceptual knowledge (C)

Phase 1 of 3P-SIT (Fig. 5.1), concerned with exposing students' conceptual understanding about a scientific idea *prior* to being exposed to any ER, requires approximately 20 - 30 minutes of interviewer-student engagement. The rationale of Phase 1 is that at first, initial probing is of a free-response nature, followed by specific questions that are posed to the student as deeper patterns of interest emerge. The following free-response type probe was used at the start of Phase 1 in all six interviews to probe students' conceptual understanding prior to being exposed to any ER.

> I: Today I would like us to talk about antibody molecules...[long pause] ... take your time and start thinking about these types of molecules. Take as much time as you want, don't rush, just relax and think about them for a while [long pause]. Try to imagine it; an immunoglobulin molecule...think about everything you know about these types of molecules [long pause]... slowly, let your thoughts flow... [silence]. When you feel like telling me something about these molecules, go ahead...speak slowly and clearly, there is no rush... [after a while]...Ok, what are you thinking about now...tell me slowly and clearly, take your time.

From the above probe, it is clear that the interviewer waits for responses to emerge naturally. Following this the interviewer will delve deeper into the student's conceptual understanding until satisfied or until a certain response is saturated. Subsequent probes do not follow any pre-determined sequence and are solely dependent on the nature of the responses elicited by the student (e.g. Ametller and Pintó 2002; Rubin and Rubin 1995; Posner and Gertzog 1982), an approach that is in agreement with the rationale and objective of Phase 1 (Fig 5.1). Interestingly, in addition to the verbal outputs generated by the probes utilised in Phase 1, it was found that in *all* cases, participants spontaneously requested to draw their own diagrams

to form part of their responses. This activity was encouraged whenever such a request was made.

In view of the above rationale and design of Phase 1, consider the following example of an interview extract obtained from a participant during Phase 1 of the interview process:

> I: I'm interested in antigen recognition, in terms of the structure of the antibody and the antigen...can you explain it [recognition] in a bit more detail?
> S: That [Ag recognition] will depend on the...when the antigens elevate [stimulate] the B-lymphocytes right... it [Ag] activates the B-lymphocyte ...the antibodies that are being produced are complementary or have sites of recognition for that specific antigen.
> [...]
> S: The fact is...let me use a square shape right [indicates with hand gesture], you are going to get antibodies that have the site of binding in the square shape...Specific antibodies bind specific antigens.
> [...]
> S: Each immunoglobulin right, is specific for an antigen... there are variants of them [Abs], depending on the classes, sub classes controlled by chains, heavy chains and light chains...
> [...]
> S: Let's go back to the basics...that antigen when it activates the B lymphocytes right... it [Ag] has secondary structure, it can bind to the antibody right...the B-lymphocyte is flexible, it synthesises antibodies with that particular binding region that can complement the antigen...

The extract above indicates the type of data that the interviewer would obtain during Phase 1 and analyse, to measure a student's conceptual knowledge *prior* to exposure to any ER. For instance, included in the above student's responses are concepts of relevance to the production of antibodies from B-cells following specific and complementary interaction with antigen, as well as conceptual understanding relating to some structural elements of antibody molecules.

Thus Phase 1 of 3P-SIT can be used to generate and analyse data corresponding to one of the factors affecting students interpretation of ERs: students' understanding (or lack thereof) of the *concepts* (**C**) surrounding antibody structure and interaction with antigen. The data collected in Phase 1 also represents a measure of the conceptual understanding that a student would bring (e.g. Cheng *et al.*, 2001) to an ER during Phase 2 when required to respond to questions about the ER. Designing and using probes to measure students' engagement of this conceptual knowledge and their processing of the ER markings during ER interpretation is discussed in the next section.

5.4.2 Phase 2: Generating and analysing data corresponding to students' reasoning processes (R)

Following Phase 1 of 3P-SIT, the student is then exposed to an ER of interest (Fig. 5.2 E, F or G), which marks the beginning of Phase 2 (Fig. 5.1). Phase 2 (Fig. 5.1), which requires about half an hour to forty minutes of engagement, has the primary objective of probing a student's reasoning processes and any changes in their conceptual knowledge, *during* the interpretation of a scientific ER. The researcher uses semi-structured questions to first probe for surface-level reasoning and then more demanding questions to probe for evidence of deep-level reasoning. In doing so, the researcher aims to establish the way in which subjects link their interpretations of an ER to their conceptual knowledge (obtained from Phase 1) and how they go about reasoning with the ER, and the markings contained within them, to acquire meaning. In other words, the probes designed for Phase 2 aim to induce the student into making sense of the graphical markings and visual-spatial features on the ER such as conventions, visual icons, spatial arrangements, topography and the representation of abstraction, while also inducing the student to associate their interpretations of the ER with their already existing conceptual knowledge.

The rationale behind the designed interview protocol for Phase 2 was one in which the probes were purposely arranged to progress from a "surface-type" to a "deeper-type" of questioning. This allowed the interviewer to observe the slow building process of ER interpretation by the student. In this regard, as the interviewer progressed through the probes, the student was required to steadily increase their level of engagement with the ER, as the probes became more cognitively demanding. In addition, the author felt that this approach allowed for both a useful and valid means for tracing any changes in students' ER-reasoning processes as the interview phase developed. As in the research reported in Chapter 4 (section 4.2.2), probe design for Phase 2 was informed by the authors visual analysis of ER E, F and G (Fig. 5.2) for any potential and therefore suspected interpretation difficulties that students may have shown.

When commencing with Phase 2 of 3P-SIT, the interviewer first gave students approximately 2 - 3 minutes with which to familiarise themselves with the ER before continuing with the semi-structured probing. As part of this, the interviewer pointed out the figure caption to the participant (Fig. 5 2) and read it out aloud. The following are semi-structured probes

designed for each of the three ERs (Fig. 5 2 E, F and G). Note how they fit the fundamental rationale of Phase 2 as progressing from a "surface" to "deep" level of questioning involving more cognitively demanding tasks.

Probes designed for ER E (Fig. 5.2) and posed to students in Phase 2 were as follows:

1. Describe the shapes and shades in this picture in as much detail as you can.
2. Can you identify a single antibody in the picture? Explain your thinking.
3. Use this picture to describe the antigen-binding sites. Explain where they are positioned.
4. Describe the shape of the antigen-binding sites. Tell me more about them. How many [antigen-binding sites] are shown on the ER? What do you think is responsible for maintaining this [structural] arrangement in the picture [point to trimer]?
5. Why is that angle in the pentamer [point to an angle within pentamer] greater than that angle in the trimer [point to an angle within trimer]?
6. Draw a diagram to represent what this part of the picture shows you [point to trimer]. Clearly explain what you are drawing.
7. How do you think this arrangement in the picture [point to trimer] could arise? You can use a diagram to aid your explanation.
8. If you had to explain this picture to a fellow student by drawing your own diagrams, how would you do it? Clearly explain what you draw.
9. Tell me about the interaction between the antibody and antigen if a different hapten or antigen were used in the situation described by the ER. You can use diagrams to aid your answer.
10. Sometimes antibody molecules are represented by a 'T' shape, and sometimes by a 'Y' shape in textbooks and other pictures. Why do you think some diagrams show a 'T' shape while some show a 'Y' shape? Sketch diagrams if they will aid your explanation.
11. Imagine you could draw a vertical line down the centre of an antibody molecule and then fold the antibody along this line. Would both sides of the antibody molecule be mirror images of each other? Explain your answer.
12. Why do you think a biochemist would want to look at and analyse this picture?

Phase 2 semi-structured probes designed for ER F (Fig. 5.2) are shown below:

1. Explain what each coloured 'sphere' on the diagram represents [point].
2. How do you think the light blue 'spheres' are "associated" to each other [point]?
3. Explain why one group of 'spheres' is coloured yellow and the other is coloured blue [indicate]. How are the two groups of 'spheres' related to each other?
4. What do you think the numbers on the red 'spheres' represent [point]?
5. What does 'plate (c)' on the ER represent [indicate]?
6. If it were possible to look at this ER from the opposite side, say, if you were looking behind the structure on the ER from the other side [indicate using hand gestures]. Would you still be able to see the red, numbered 'spheres' [point to frame c]?

7. Tell me about the biochemical situation that is represented by this ER.

8. In terms of antibody structure, what is being represented on this ER? Use diagrams to explain your answer. Clearly explain what you are drawing.

9. How would you draw 'frames' 'a', 'b' and 'c' [point] if you were asked to explain this ER to a fellow student? Take your time and sketch the diagrams you would use to explain the ER. Clearly explain what you are drawing.

10. Explain what would happen if different 'spheres' in the same situation replaced the red 'spheres' on the ER [indicate]. If you like, use diagrams to explain your answer.

11. What is the purpose of antibody-antigen binding *in vivo*? By considering the biochemical situation described by the ER, what do you think would happen next?

12. When you look at this ER, and the diagrams that you have drawn, do you think of any other biochemical processes?

13. Why would a biochemist want to look at and analyse this type of representation?

Finally, Phase 2 probes designed to investigate students' interpretation of ER G (Fig. 5.2) were as follows:

1. What graphical relationship is this ER showing?

2. Explain what the four coloured curves mean [point].

3. Why do you think the logarithmic function [point] is used to plot these curves?

4. What is being plotted on the x-axis of the ER [point]? Comment on the antibody concentration as one moves from left to right on the x-axis [indicate].

5. Explain the general, negative slope of the coloured curves [indicate] as the values on the x-axis increase. Explain this relationship.

6. In biochemical terms, what do you think these curves are describing?

7. In biochemical terms, what is responsible for the absorbance values at 405 nm [point to y-axis]?

8. While you are interpreting this ER, what pictures are going through your mind? Try and draw what you are thinking about so that you can explain the images in your mind. In your diagram that you have drawn, what antibody is the antibody that is represented on the ER [x- axis]?

9. Draw a diagram to explain how the biochemical components [the arrangement of antibody and antigen] related to this ER, would look like at point Q on the ER [point]. Clearly explain what you are drawing.

10. Why do you think the curves for week 8 and week 12 first increase and then decrease [indicate]?

11. How would the curves look, if we:
    i) Changed the negative sign in front of the 'log' to a positive sign?
    ii) Still used the negative 'log' function to plot the graph, but plotted μg/ml instead of mg/ml?

12. Normally, the absorbance readings for these curves would be around 0.8. Why do you think that they are lower in the situation shown by the ER?

13. Consider that the experiment that generated the data to plot these curves had ended and the researchers had stopped collecting samples. Draw a rough graph to show how the ER would look if you were to plot values for serum samples collected for week 100.

14. Why would a biochemist be interested in using or plotting an ER such as this?

The above Phase 2 probes, designed for each ER (Fig. 5.2), were administered by the interviewer in a flexible manner in that they were not necessarily posed verbatim to the participants. The precise content of the probes depended on the unique style in which the Phase 2 component of each interview progressed. In addition, due to the naturalistic approach offered by 3P-SIT, it was not always necessary to pose an *entire* set of Phase 2 probes to a student during their interpretation of an ER. Instead, the decision to exclude or include particular probes depended very much on the nature of the responses that were being elicited during a particular interview session. In this regard, the general emphasis in Phase 2 like in Phase 1 was to, once an interesting response pattern had been observed, probe deeper where viable. Furthermore, each set of probes served as a structural framework in that there was always a variety of other probes that could be administered by the researcher if it was found that no patterns of interest emerged, or if the student was not forthcoming in delivering responses. It should be noted, though, that introducing some degree of standardisation into Phase 2 through the use of a pool of probes for each ER, did in no way allow the student to be led. On the contrary, this added structure and logic to Phase 2 allowing interesting patterns to be probed for further, without forcing the student into a specific response. This systematic approach served to instil a degree of reliability into the interview instrument.

As stated above, it is clear that for each ER, the probes were pitched as progressing from a surface to a deeper-level of necessary student engagement. For example, compare probes 1, 3 and 7 for ER G above, where a steady increase in the complexity of the questions can be observed. In terms of the above semi-structured probes for Phase 2, we based our rationale for their design on the following. A surface-level of engagement can be best described as a process of *extracting information* (Kindfield, 1993/1994) from ER features that are salient or stand out (e.g. Lowe 2004, 2003). For example, consider probes 1 - 3 above for each of the ERs, where to respond to the probes successfully the student is required to extract visual information from the particular ER. In contrast, a deeper level of engagement can be described as a process of *extracting meaning* (Kindfield, 1993/1994) from ER features that are not salient. During this process, students have to use the ER *and* engage their own

conceptual knowledge to successfully reason with the ER. For instance, consider probes 8 - 10 above for each of the ERs, where, in order to respond to the probe fruitfully, the student is required to link his/her interpretations of the ER to their already existing knowledge, which is a much more demanding reasoning process than that required for previous probes.

As evident in the Phase 2 probes above, deeper-level probing also consisted of more specific "talk-aloud" or "think-aloud" tasks (e.g. Kozma 2003; Lewalter 2003; Peña and Quílez 2001; Bowen 1994; Lowe 1993; Posner and Gertzog, 1982) in which students were sometimes required to generate their own diagrams (e.g. Glynn, 1997) when interpreting an ER. These types of probes aimed to attain information pertaining to how a student reasoned with an ER or made *use* of it to "solve a problem" (e.g. Cox and Brna, 1995; Mousavi *et al.*, 1995; Hegarty, 1992). Therefore, data generated from these tasks was often both in a verbal *and* diagrammatic form (Chapter 3) enabling the researcher to track a student's ER-related cognitive processes while they expressed their reasoning processes. In addition, as part of these probes, the interviewer also noted students' tacit behaviours (e.g. Gall *et al.*, 1996) such as pointing, indicating to, constructing, annotating and modifying of their generated diagrams (e.g. Kindfield, 1993/1994). When obtaining responses from students during Phase 2, students were prompted to succinctly explain the diagrams that they generated so that the author was in a better position from which to determine the nature of a student's mental models (e.g. Gobert and Clement, 1999; Lowe, 1993a). In lieu of the former, Beilfuss *et al.*, (2004), Lowe (2003), Olivier (2001), Cheng *et al.* (2001), Kindfield (1993/1994), Koedinger and Anderson (1990) as well as Larkin and Simon (1987) have all used similar approaches to obtain data on students interpretation of scientific ERs. As mentioned in Chapter 3 (section 3.4.4.5), obtaining more than a single datum from each response served to triangulate the methods used in this thesis to obtain data corresponding to students' interpretation of ERs.

With respect to the above rationale and design of probes for Phase 2, consider the following interview extract and accompanying student-generated diagram (SGD) (Fig. 5.3) obtained during a student's interpretation of ER E during Phase 2:

I: Tell me about the different shapes that you see [on ER E].
[...]
S: I'd think A [points to region "A" on ER E] is a realistic picture... In this one [trimer arrangement near region A]... the antibody is Y-shaped and the antigen gets in over there [points within "V-cleft" of top Y-shape of trimer near area A], it makes sense...
[...]

S: ...something which was a triangle can get into the Y-shape there [points within trimer arrangement]... I can imagine how [the triangle shape] gets in. Something that is pointed at the end can get into it [Ab].
[...]
S: ...that part [points to "V-cleft" of top Y-shape of trimer near area A] is being opened up in such a way that that antigen that is recognised can get in.
I: ...what opens up?
S: That V-shape [points to top Ab in trimer]...[beg. to gen. Fig. 5.3]
[...]
S: The antibody recognises the antigen while around the blood system, right. It [Ab] will move towards it [Ag], it [Ab] is complementary, it [Ag] binds specifically at the binding region...So for A, the binding region would be here, ok, I will just put it in black [Fig. 5.3]...that is the binding region [marked with black lines on Fig. 5.3].

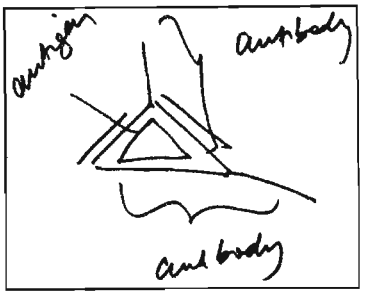


Figure 5.3 SGD obtained during interpretation of ER E in Phase 2 of 3P-SIT. Textual labels in SGD read "antigen" and "antibody"

From the interview extract and SGD (Fig. 5.3) above, it is evident that the student has exposed data relating to their processing and interpretation of the graphical markings on ER E. In this case, analysis of the data indicates that the student interprets the triangle-like markings on ER E to represent two antibodies joined together (see Fig. 5.3). In addition, the same student interprets the dark area within the trimer on ER E to be representative of a triangular-shaped antigen (see Fig 5.3). Other than revealing information on those reasoning processes incorporating the processing of ER E itself, the same student has also revealed data that corresponds to how the student engages certain concepts (obtained from Phase 1) to reason about the ER. In this instance, the student may have possessed an alternative conception that an antibody has only *one* "complementary" site for antigen binding, and this concept may have influenced the reasoning process expressed in Phase 2 for this ER and student.

In addition to the above, consider a further example of an interview extract and SGD (Fig. 5.4) obtained from Phase 2 during a student's interpretation of ER F:

I: What biochemical situation do these pictures [ER F] represent?

[...]
S: ...it is like a chemical process...where...it [Ab] engulfs it [Ag] and takes it [Ag] inside and it [Ab] breaks it [Ag] up into little pieces...
I: ...how does it [Ab] take it [Ag] inside?
S: ...The red [Gln on Ag] has some sort of thing, which it [Ab] would recognise and see it [Ag] as foreign...it [Ab] pulls it [Ag] towards it [Ab] ...the antibody would pull this foreign antigen.
I: How?
S: ...by recognising that it is foreign because these bonds here [points to red Gln on "frame b" of ER F] are complementary bonds... It is like a chemical process where it [Ab] would break it [Ag] down... like digest the dangerous or the harmful things and make it [Ag] less harmful. And then it [Ab] lets it [Ag] go again...then it [Ag] is not so potent when it [Ag] comes out ["gets released"].

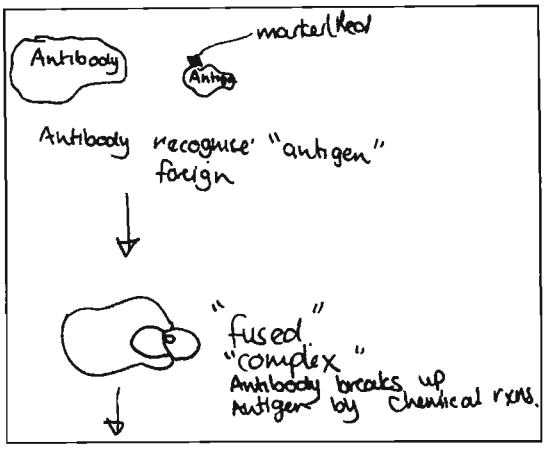


Figure 5.4 SGD obtained during interpretation of ER F in Phase 2 of 3P-SIT

It is evident from the interview extract and SGD (Fig. 5.4) above that the Phase 2 probes generate data corresponding to student's interpretation of the graphical features shown on a particular ER. In this case, perusal of the datum expressed above provides an indication of the reasoning processes surrounding the student's deciphering of the red "spheres" on ER F. Here, the student may have interpreted the red coloured spheres on frames "a" and "b" on ER F representing Gln 121 as having undergone a digestive process, resulting in the two groups of red spheres depicted on frame "c". In addition, inspection of the above datum also indicates that the student's processing of the graphical markings may have been influenced by the conceptual understanding (revealed in Phase 1), which the student brought to the ER. In this instance, the student may have engaged the misconception that the Ab is able to perform cellular immune reactions such as eliminating Ag when reasoning with ER F (see Fig. 5.4).

Lastly, consider the following interview extract obtained from a student's interpretation of the ELISA representation (ER G) during Phase 2 of 3P-SIT:

I: Comment on the concentration of antibodies when you move from the left to right on the x-axis [indicates].
S: It [Ab concentration] appears to be increasing...that is right, they [Ab conc.] are increasing...I also think that is because there are more antigens around, so there would be more antibodies being produced, so that is why the concentration would increase, but it would also explain why absorbance is decreasing. It is because even though there are more antibodies around, there are more antibody-antigen complexes being formed.
[...]
I: What is responsible for these numbers [indicates on y-axis]? Where do they come from?
S: Ok...it is something to do with light scattering...measuring the amount of...light scattering and reading off that...how much the antibody-antigen complexes can absorb the light...I would say the more they [Ab and Ag] form a complex the more they can absorb light, the absorbance readings would decrease the more complexes are formed [S stated earlier that Ab conc. Decreased from lt to rt on x-axis].
[...]
S: The way I picture it is absorbance would be a function of what is going on here [indicates absorbance values on ER G], scattering light...since antigen binds to the receptor sites, it doesn't leave the receptor site open to scattering light.

Analysis of the datum above suggests that, when processing the graphical markings on ER G, the student erroneously associated the numerical increase of the values on the x-axis to an increase in Ab concentration. Therefore, the data gives the author insight into how the student reasoned about the markings contained in the ER. In addition, examination of the datum above also provides information about how the student used his/her conceptual knowledge (determined during Phase 1) to interpret the ER. In this regard, the student's alternative conception that the "more Ab-Ag complexes are formed, the less the absorbance", clearly influenced the student's interpretation of ER G.

In the above examples of data generated from Phase 2 of the instrument for each of the ERs (Fig. 5.2), we have provided evidence of how the data can be scrutinised to demonstrate students' reasoning processes when interpreting the ERs. In this regard, data can be gathered and analysed that corresponds firstly, to students' processing of the graphical markings when reasoning with the ER. These reasoning processes are particularly evident in the above extracts by students exposing language that mimics their active engagement with the visual features of the ERs. Secondly, information could also be gathered on students' use of their conceptual knowledge when reasoning with the ER. These reasoning processes are evident in the above extracts by students' use of language that represents the active engagement of certain concepts when interpreting the ER. Overall, the data generated during Phase 2 corresponds to another of the factors affecting students' interpretation of ERs namely, students' ability to *reason* with the ER and with their own conceptual knowledge (**R**).

5.4.3 Phase 3: Generating and analysing data corresponding to the mode of representation (M)

Phase 3 of 3P-SIT (Fig. 5.1) lasts for about 15 - 20 minutes and requires students to evaluate and critique the ER in question (Fig. 5.2 E, F or G) in response to semi-structured probes. This in turn, helps the researcher generate data about the role and effect of the graphical markings and features of the ER such as conventions, icons, colour, artistic devices, labels and captions on students' reasoning processes. In other words, the rationale behind this approach is that data revealed in Phase 3 helps the researcher measure the nature or influence of the ER *in isolation*, i.e. the role and effect of the *representation mode* on students reasoning processes. This student data is also compared to that from experts' evaluation of the same ER conducted independently of the student interview (see later). Five typical semi-structured probes used in Phase 3 for all 6 interviews and across all three ERs were as follows:

1. Is there anything on the ER in particular that you don't understand or find confusing?
2. What do you think this ER is not showing? Explain your answer.
3. Consider yourself a diagram designer or textbook author. If you could change this ER in any way, what would you do to improve it, if anything?
4. Do you think this is a good and clear representation? Give reasons for your answer.
5. Comment on these types of representations in general, and your feelings on interpreting them.

A further characteristic of the rationale behind the design of the Phase 3 probes was that they required the student to think critically about the ER at hand and to apply a "rating" of its usefulness. In doing so, the probes aimed to induce metacognitive and reflective behaviours (e.g. Ward and Wandersee, 2002; Case *et al.*, 2001) in that students were required to 'take a step back' and consider the ER as an 'outsider' in an effort to evaluate the ER objectively. All the probes utilised in Phase 3 were similar across all three ERs and presented to all of the participants. For each probe, the interviewer pursued the patterns of interest applicable to a particular ER by delving deeper into a students' particular emerging responses while refraining from leading or biasing a student into a particular response (e.g. Ametller and Pintó 2002). Once Phase 3 of the 3P-SIT protocol (Fig. 5.1) had been completed, the interview session was closed.

With regard to the above rationale and design of the Phase 3 probes, consider the following interview extract obtained during a student's evaluation of ER E during Phase 3:

> I: Is there anything that you don't really understand, or that you find confusing in these pictures [ER E]?
> S: They're unclear...they are real pictures right...if they were drawn, they would make a lot of sense... Yeah, because you have to have done some work to remember what the antigen looks like, what the antibody might look like, what happens on binding...if you don't know that, you won't understand the picture.

It is evident from the extract above that the participant identified what graphical marking(s) or features positively or negatively influenced reasoning with the ER. In this case, scrutiny of the above datum shows that the "realistic" nature of ER E was the ER feature that played a key role during the student's interpretation of the ER. The student suggests that the realistic graphical nature of the depicted antibodies and antigens makes the ER challenging to interpret. Therefore, by obtaining data corresponding to this graphical feature of the ER, the researcher is able to identify the graphical markings that are playing a significant role during students' interpretation of the ERs.

In addition to the example above, consider the following interview extract generated during a student's evaluation of ER F in Phase 3:

> I: ...is there anything that you find confusing about this diagram [ER F]?
> [...]
> S: ...I think that [points to white Gln in frame c] is confusing...this here [white Gln on "frame c"] is white, I don't know if that is just the way it is supposed to be... why is this [points to white Gln] different to everything else?
> [...]
> S: I think it is quite interesting that the red [Gln] has actually fitted in and filled the gaps between the heavy and the light chains... It looks like it is almost a complex since the red has joined the blue and yellow.
> [...]
> I: ...do you think that it [ER F] is a clear diagram?
> S: ...I think the [heavy and light chain] regions are quite distinct...I suppose that is because of the colours that are used, so you can see them quite nicely from each other.

In the extract above, the student identifies which graphical feature(s) may have been responsible for a poor (or successful) interpretation of ER F. In this instance, analysis of the datum shows that the student identifies the glutamine amino acid, coloured in white on "frame c" of ER F as one graphical feature that had an influence on his/her interpretation of the ER. The student also delivers further information on what ER features may have also had a positive effect on interpretation of the ER. In this regard, investigation of the datum suggests that the colouring and spatial devices used to depict both the Fab-Ag complex and the light

and heavy chains of the Fab structure were ER markings that had a favourable effect on the student's interpretations.

Lastly, consider the following student's verbal output obtained during interpretation of ER G in Phase 3:

> I: Is there anything that seems confusing to you [on ER G]?
> S: Well, right at the beginning, this whole negative 'log' [points] kind of threw me off...
> [...]
> S: The graph itself is quite straight forward... except these figures [values on x-ax.]...for instance I didn't realise they were increasing, it didn't really stand out to me...

Analysis of the response obtained from the student above suggests that the graphical (textual) features corresponding to the "-log" function on ER G may have contributed to unsuccessful reasoning with the ER. In addition, perusal of the above response indicates that the numerical values on the x-axis may have also been examples of graphical features that could have had a negative effect on ER interpretation.

Based on the above three examples, analysis of the data generated during Phase 3 of 3P-SIT provides a window into the role of the graphical markings such as the spatial arrangement of the ER elements, ER conventions, visual icons, artistic devices, colour, topography, level of abstraction, symbols, labels, captions and other ER embellishments on students interpretation of the ER. Thus generating and analysing data from the Phase 3 probes helps the researcher identify how the external nature of the ER itself influences ER interpretation. Therefore, the data generated during Phase 3 corresponds to the last of the factors affecting students' interpretation of ERs observed in Chapter 4, namely, the *mode* in which the desired scientific phenomenon is represented in the ER (**M**).

5.5 Implications of 3P-SIT as a data-gathering instrument

Analysis of the data generated from the above pilot study, used to test the instrument has revealed the following implications for 3P-SIT as a research tool. Firstly, 3P-SIT can successfully generate data corresponding to three factors (**C**, **R** and **M**) affecting students' ability to interpret ERs identified in Chapter 4. In doing so, greater insight into the nature and validity of the factors can be obtained. Secondly, analysis of the data generated during Phase 1 shows how 3P-SIT can be used to measure the nature and extent of the conceptual

knowledge (**C**) that a student will bring to an ER. Thirdly, analysis of the data from Phase 2 shows how the researcher can obtain information about students' reasoning processes (**R**) corresponding firstly, to the interpretation of the graphical markings in the ER and secondly, to students' engagement of their conceptual knowledge during reasoning. Fourthly, analysis of the data from Phase 3 demonstrates how the researcher can obtain information on the role and effect of the mode of representation (**M**) on students' reasoning processes. Fifthly, by comparing data *across* the 3P-SIT phases, we can measure a student's overall ability to successfully interpret and learn from a particular ER. Sixthly, although Phase 1 delivers information pertaining to a student's conceptual understanding, it is expected that other components of students' conceptual knowledge will also be revealed in Phases 2 and 3 that may have not have been necessarily shown during Phase 1. Seventhly, even though Phase 2 has the primary objective of probing and generating data on student's reasoning processes *during* the interpretation of a scientific ER, it is to be expected that other data pertinent to students' reasoning processes will also exposed in Phases 1 and 3. This is by virtue of the fact that students' are also employing other cognitive processes in Phases 1 and 3 when generating *any* responses whatsoever. Eighthly, using 3P-SIT with another sample of students would serve to further inform its development as a research instrument.

In the next Chapter, 3P-SIT is implemented with a different sample of students to generate empirical data for developing a model of factors determining students' ability to interpret ERs. Expression of such a model could help to not only further confirm the nature and validity of the factors, but may assist in measuring the nature of influence of the factors upon one another. Overall, such a model, in addition to validating the factors, will also serve to frame researchers' thinking on the nature of the factors determining students' ability to interpret ERs in science.

6 A MODEL OF FACTORS DETERMINING STUDENTS' ABILITY TO INTERPRET EXTERNAL REPRESENTATIONS

6.1 Introduction

Much inquiry in science education and educational psychology has centred on the role and effectiveness of external representations (ERs) in the learning and teaching of science (see Chapter 2). Seminal papers in this area include those by Lowe (2004, 2003, 1999), Mayer (2003, 1999), Ametller and Pintó (2002), Roth (2002), Treagust *et al.* (2002), Peña and Quílez (2001), Kozma and Russell (1997), Scaife and Rogers (1996), Cox and Brna (1995), Stenning and Oberlander (1995), Wandersee (1994), Kindfield (1993/1994), Winn (1993, 1991), Lord (1987a, b), Holliday *et al.* (1977) and Dwyer (1972, 1967). These and other studies have focused on various ERs including, inter alia, static pictures, diagrams, graphs, photographs, micrographs, maps, flowcharts and computer-based dynamic visuals. Although ERs are usually assumed by science lecturers to be excellent learning tools for constructing knowledge, various research reports (e.g. Stylianidou *et al.*, 2002; Cheng *et al.*, 2001) have suggested that they do not always improve understanding and may in fact cause difficulties. This problem is largely due to naïve assumptions by lecturers that what works for experts will also be good for novices (e.g. Lowe and Schnotz, 2003; Scaife and Rogers, 1996).

The aim of this aspect of the study was to address research questions 4 and 5 (see Chapter 1) namely, can the factors identified in Chapter 4 and, further investigated in Chapter 5, be incorporated into an appropriate model and how might we obtain empirical data to confirm the validity of the model? To address these questions, the modelling process of Justi and Gilbert (2002) was used to develop the model and the 3P-SIT instrument (Chapter 5) was used to generate empirical data for the development of operational definitions of each factor, and for the validation of the model.

6.2 Methods

6.2.1 Participants and descriptions of the external representations

The study was conducted from 2001 to 2002 with nine biochemistry students at the University of KwaZulu-Natal, South Africa, who had all completed a third-year level module on immunology. Each student was interviewed three times between July and November 2001 – an interview for each of three different ERs (Fig. 5.2) giving a total of 27 interviews. The same three ERs used for the development of the 3P-SIT instrument reported in Chapter 5, were used for the investigation described in this chapter.

For convenience, we ask the reader to consult the flip-out copy of Fig. 5.2 on p. 130.

6.2.2 Development of the model

The modelling framework of Justi and Gilbert (2002) was used to develop and test a model of the factors, identified in Chapter 4, that influence student interpretation, processing and understanding of ERs used in the teaching and learning of science. Although the modelling method proposed by Justi and Gilbert (2002) is concerned with those models associated with scientific knowledge, such as historical and current scientific models or those associated with modelling curricular models, the author found their process to also be applicable to the process used to develop the present model. In this case, the modelling framework was used in a more abstract manner; that of expressing the factors that influence student interpretation, processing and understanding of ERs. In this regard, the current author believes that a thinking process highly similar to the one framed by Justi and Gilbert (2002) (Fig. 6.1) enabled the current model to be developed. Adopting this approach in itself suggests that all thinking related to modelling any phenomenon of knowledge must follow some type of logical pattern. Given this opinion, we believe that the modelling framework set out by Justi and Gilbert (2002) (Fig. 6.1) enabled us to follow such a logical pattern and we therefore, considered it a suitable and rigorous framework for guiding the development of our own model.

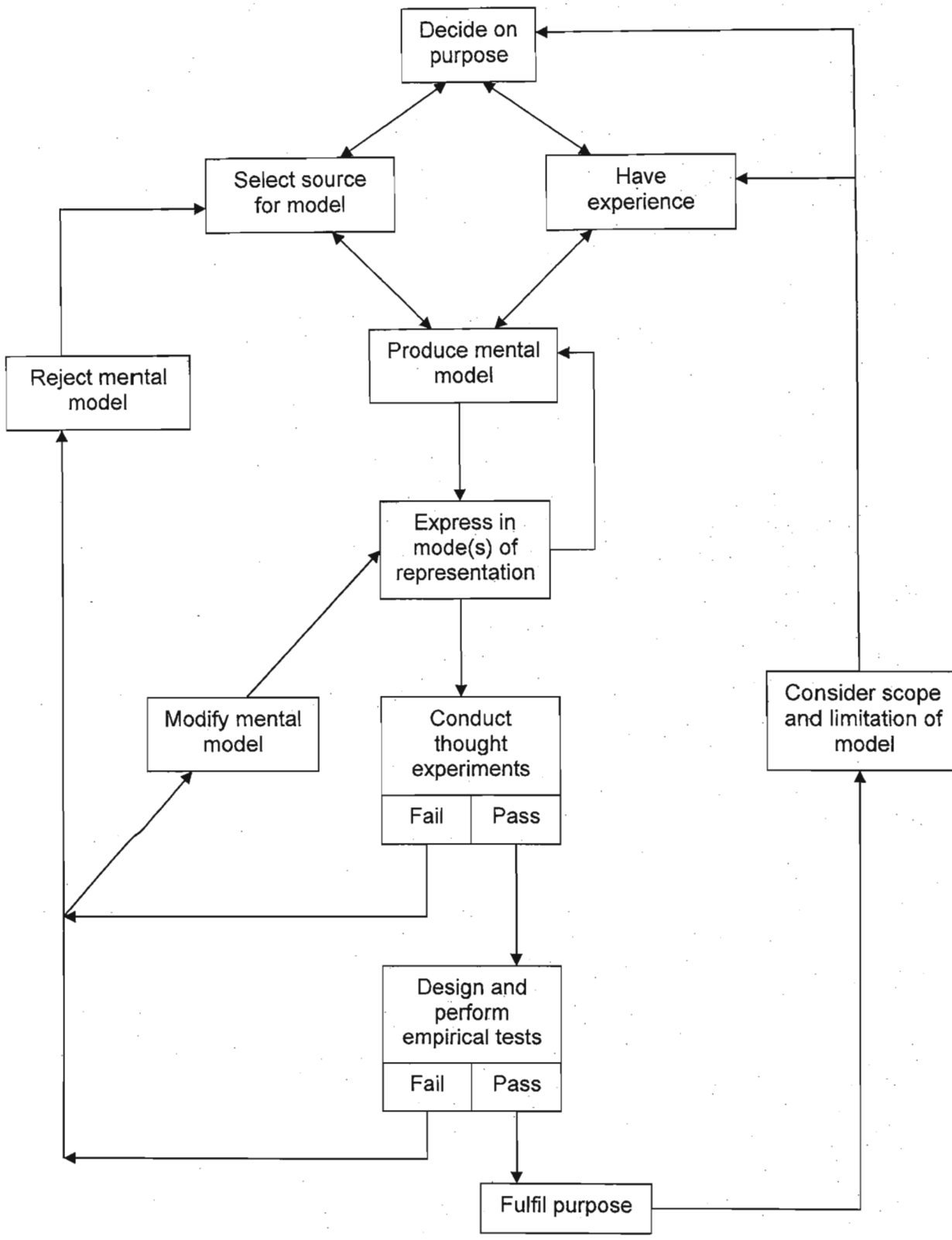


Figure 6.1 The modelling framework used to develop and express the model of factors (Adapted from Justi and Gilbert, 2002, p. 371)

The modelling process involved a five-stage cyclical process (Fig. 6.1). Firstly, the *purpose* of the model was decided upon based on the factors identified in Chapter 4, the author's prior knowledge and experience of student difficulties with ERs, and a thorough analysis of the literature (Chapter 2) on learning and teaching with ERs in science. Secondly, a *mental model* was constructed and thirdly, the mental model was externalised as an *expression model.* Fourthly, conduction of various *thought experiments* as well as extensive discussion of the expression model with the supervisor helped decide on the validity of the model and whether to modify it. Stages 2 – 4 were repeated several times so as to optimise the expression model. Fifthly, empirical tests (see 6.2.3. below) were designed and performed in order to decide whether to modify, reject or accept the model as a *consensus model.* Of pivotal importance during the fifth stage was to establish whether the resulting *consensus model* satisfied its purpose and, to consider what the actual applications and limitations of the model would be.

6.2.3 Empirical testing of the model (stage 5)

Empirical testing of the model was performed, using 3P-SIT (Chapter 5), in order to investigate the nature of the interaction between the factors of the model, to formulate clear operational definitions for its component factors and to validate the consensus model. A description of each phase of the 3P-SIT interview method including samples of the probes employed and examples of student data and their analysis were described in Chapter 5.

6.2.4 Analysis of the interview data

All interviews were both audiotaped and videotaped (e.g. Hull *et al.*, 2003; Pavlinic *et al.*, 2001; Sumfleth and Telgenbüscher, 2001). The data collected during the interview sessions consisted of 27 video segments, 27 audio-transcripts, 134 student-generated diagrams (SGDs) and 27 researcher-generated field note items. Details pertaining to analysis of the data generated from the 3P-SIT instrument are discussed in section 5.4 (Chapter 5).

6.3 Results and Discussion

6.3.1 Development of the model

The modelling process (Fig. 6.1) of Justi and Gilbert (2002) enabled us to successfully design the model presented in Fig. 6.2 below. Regarding the purpose of the model, it was decided that it should serve as a tool with which to frame (guide) our thinking on the factors that affect a student's ability to interpret a scientific ER. Through the use the factors identified in Chapter 4, the model was first conceptualised as a mental model and then as an expression model. Initially, the model was expressed as a *triarchic* model, defined by three apexes of a triangle. The three apexes represented the three factors proposed in Chapter 4, namely, students' ability to *reason* with the ER, students' understanding of the *concepts* of relevance to the ER, and the nature of the *mode* in which the desired phenomenon was represented through the ER. Following lengthy debate with the supervisor and thought experiments, the triarchic model was modified to include the *interaction* or *relationship* between each of the three factors. This decision was motivated by the realisation that the factors were strongly interdependent in that, for example, reasoning could not occur without something to reason with - in this case a student's conceptual knowledge and the ER mode. Thus this modified model, composed of seven factors (**C**, **R**, **M**, **R-C**, **R-M**, **C-M** and **C-R-M**) as shown in Fig. 6.2, was better represented in the form of Venn logic in that the factors would overlap. Empirical testing of the model using 3P-SIT (see section 6.3.2 and 6.3.3 below) allowed operational definitions for each factor to be established and further confirmed the importance of the seven factors affecting ER interpretation. This reinforced our opinion that the Venn diagram (Fig. 6.2) was the most useful representation for communicating the purpose and nature of the model. Thus, based on these results, we decided to accept the model as a consensus model.

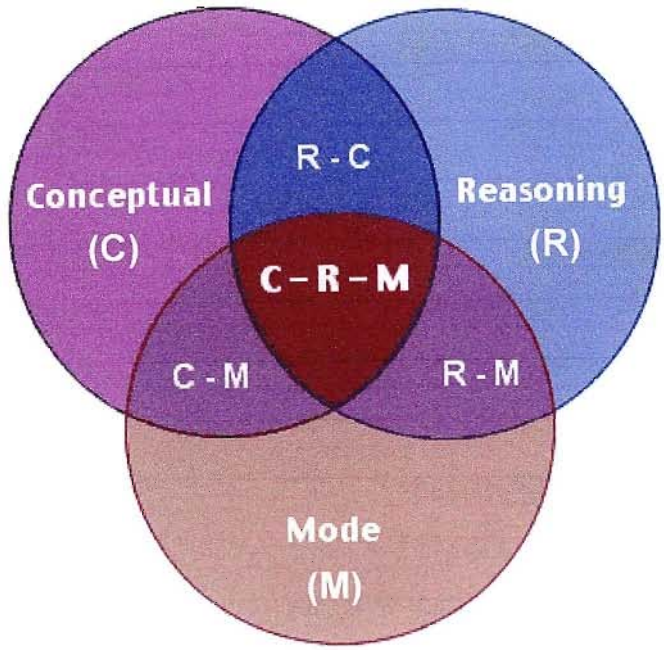


Figure 6.2 Venn diagram representing a model of seven factors that determine students' ability to interpret ERs. The model expresses three factors and four interactive factors affecting students' ability to interpret an ER

6.3.2 Operational definitions for factors of the expressed model

Empirical testing of the proposed model enabled specific operational definitions for each factor of the expressed model to be formulated. These are outlined below and presented in further detail in Table 6.1. To assist the reader to assimilate the interpretation of the empirical data presented in section 6.3.3 the operational definitions derived from the data are presented first. For the convenience of the reader, a flip-out page of the operational definitions is supplied on p. 154.

We defined the conceptual factor (**C**) (Table 6.1) of the model as the existing conceptual understanding and prior knowledge that a student holds *before* exposure to any ER. It embodies the collection of a student's preconceptions, conceptions, conceptual schemata, conceptual frameworks, semantic networks, mental models and alternative conceptions of relevance to the ER. Alternative conceptions, also termed conceptual *difficulties*, can be described as those conceptions that are inconsistent with accepted propositional scientific knowledge or worldviews (e.g. Osborne and Wittrock, 1983). They are *specific* to a certain

scientific context (Grayson *et al.*, 2001), such as to the idea of antibody-antigen structure and binding in biochemistry.

Since reasoning is a *process*, one has to have something to reason with and therefore, reasoning processes cannot be defined in isolation. In terms of the model, we defined the reasoning factor (**R**) (Table 6.1) as representing those *cognitive processes* and *reasoning skills* that a student employs when reasoning with the ER and with his or her own conceptual knowledge that is of relevance to the ER. More specifically, factor **R** represents a student's total reasoning ability, i.e. the skills needed to decode and perceive visual markings on an ER (e.g. Ward and Wandersee, 2002; Coon, 2001; Lowe, 2000; Dwyer, 1969), to access and retrieve conceptual knowledge from long term into working memory (e.g. Baddeley, 1992; Kosslyn, 1989; Jonassen and Hawk, 1984) in order to perform ER-related reasoning or problem solving; and, to assimilate information that is first perceived from an ER and then incorporated into already existing knowledge (e.g. Bukatko and Daehler, 1992). In agreement with a constructivist paradigm, cognitive mechanisms associated with **R** can never be passive (e.g. von Glasersveld, 2003) in that reasoning is considered an active process (e.g. Treagust *et al.*, 2002; Bruner, 1960), characterised by students' *constant* selection, organisation, integration and encoding of information (e.g. Mayer, 2003, 1999). Unlike a conceptual difficulty, which is context-dependent, a *reasoning difficulty* is *independent* of context (e.g. Grayson *et al.*, 2001) and can be observed in multiple scientific content areas.

For instance, localised reasoning is an example of an ER-related reasoning difficulty identified in the contexts of electricity (Cohen *et al.*, 1983), metabolism (Anderson *et al.*, 1999) and biological food webs (Griffiths and Grant, 1985). With respect to the model, a reasoning difficulty can span across several ERs within a specific context (e.g. across antibody-antigen binding), across several ERs from different contexts (e.g. across antibody-antigen binding *and* the particulate nature of matter), across ERs in an even larger context (e.g. across biochemistry or physics), or across science as a whole

The representation *mode* factor (**M**) (Table 6.1) of the model encapsulates the nature of the ER. By the nature of the ER, we mean the characteristics of the ER such as the graphical and diagrammatic features, the spatial arrangement of the ER elements, ER conventions, visual icons, visual cues, artistic devices, colour, topography, level of abstraction, symbols, labels,

Table 6.1 Operational definitions of the factors and interactive factors of the model affecting students' ability to interpret an ER

Factor	Operational Definition	Method of Evaluation
C	Represents a student's **PRIOR KNOWLEDGE** of all the concepts that are represented by the ER (C-M), before exposure to the ER. Such knowledge includes: all the student's preconceptions, conceptions, conceptual schemata, conceptual frameworks, semantic networks, mental models and alternative conceptions of relevance to the ER.	Phase 1 of 3P-SIT
R	Represents a student's **TOTAL REASONING ABILITY (SKILLS)** he/she has available for interpreting the ER. It includes the student's ability to reason with both the ER (R-M) and his/her conceptual knowledge (R-C) of relevance to the ER (C-M). It represents both sound reasoning and any reasoning difficulties including: surface-level reasoning; inappropriate analogical reasoning; transfer; translation between ERs; and, superimposing of one concept upon another.	Evaluated by combining data obtained for R-C and R-M interactive factors.
M	Represents the **NATURE OF THE ER** and how well (or poorly) its features represent the concepts, structures or processes it is designed to represent. These include the effective and ineffective use of graphical and diagrammatic features; the clarity of and relationship between representations; and, the spatial arrangement of elements, conventions, visual icons, visual cues, artistic devices, colour, complexity, topography, level of abstraction, symbols, labels and captions.	Evaluated by experts such as scientists, researchers and graphic artists as well as students in isolation from interpretation of the ER during Phase 3 of 3P-SIT
R-C	Represents a student's **ABILITY TO REASON WITH HIS/HER CONCEPTUAL KNOWLEDGE** of relevance to the ER. It includes ability to perform cognitive processes such as: memory-recall including accessing, selection and processing of existing information of relevance to the ER; the assimilation, accommodation and, integration of new knowledge learnt from the ER. It also includes reasoning processes such as analogical reasoning, transfer, superimposing of one concept upon another, inductive and deductive reasoning etc. It includes both sound reasoning and unsound/ inappropriate reasoning difficulties.	Phases 1 and 2 of 3P-SIT
C-M	Represents the nature of the **CONCEPTUAL (PROPOSITIONAL) KNOWLEDGE REPRESENTED BY THE ER** and its symbolism. It includes the extent, complexity and soundness of the knowledge represented by the ER and therefore, how cognitively demanding it might be (a complex ER is more difficult to assimilate).	Obtained from text, captions and expert evaluation of the ER and the knowledge represented by the ER, in terms of extent, complexity and soundness. Obtained in isolation from students' interpretation of the ER.
R-M	Represents the student's **ABILITY TO REASON WITH THE ER** and its graphical features. It includes ability to perform cognitive processes such as decoding; deciphering; recognition; perception; visualisation; and organisation of patterns, shapes and colours; visuo-spatial operations; distinguishing relationships between ER features; organising visual information on the ER; analogical reasoning, symbolic reasoning, as well as surface-level and deep-level reasoning; formation of superficial mental models; transfer; and, translation between ERs. It includes both sound reasoning and unsound/inappropriate reasoning difficulties and students' inability to perform any of the above cognitive processes.	Phase 2 of 3P-SIT
C-R-M	Represents a student's **ABILITY TO SUCCESSFULLY INTERPRET, VISUALISE AND LEARN FROM THE ER.** This includes the student's ability to engage all factors of the model by using reasoning skills (R) to reason with both their conceptual knowledge (C and R-C) of relevance to the ER and with the symbolism and features of the ER itself (R-M) to make sense of the graphical features of the ER (M) and visualise the conceptual knowledge represented by the ER (C-M). This will reveal any improvement in the student's science conceptual knowledge, any conceptual changes that may have occurred, as well as any new alternative conceptions, alternative frameworks or models that may have developed as a result of the student's interaction with the ER.	Measured by how correctly the ER is interpreted and the improvement in understanding and/or development of alternative conceptions that occurs after exposure to the ER. The success of the **interpretation** of the ER, **and of any learning** from the ER, is measured by comparing the student's conceptual knowledge after exposure to the ER (Phase 2) to the conceptual knowledge represented by the ER (i.e. C-M) and to the student's prior knowledge (C), respectively.

captions and so on. Factor **M** can be considered distinct from both **C** and **R**, since it does not depend on any human constituent during the interpretation process and remains constant unless the ER is modified (e.g. during animation).

Thought experiments, and empirical testing, revealed that it was appropriate to include a further four-factors in the model, representing the *interaction* or *relationship* between factors **C**, **R** and **M** (Fig. 6.2). This was because, at any one time, none of the three factors would influence ER interpretation in isolation. For example, the student would have to be reasoning either with the ER or with their conceptual knowledge. Thus, the interactive factors can help us describe the possible scenarios at play when two, or all three, of the factors **C**, **R** and/or **M** influence a student's interpretation of an ER.

The interactive factor that was defined as representing the relationship between the reasoning (**R**) and conceptual (**C**) factor, termed **R-C** (Table 6.1), represents cognitive processes such as when a student selects, retrieves, actively adjusts or adds to their existing knowledge. **R-C** is indicative of a student's ability to reason with their conceptual knowledge of relevance to the ER because, in effect, they are using the collection of their concepts in order to 'think about something' or to 'solve' a problem. Congruently, within **R-C**, cognitive processes such as assimilation and accommodation can also be represented (section 3.3.2). This is so because a student may add to, or adjust, their conceptual structure, especially when concepts are constructed that did not form part of an original schema.

The **R-M** interactive factor (Table 6.1) between the representation mode (**M**) factor and the reasoning (**R**) factor exemplifies a student's ability to decipher, process and reason with an ER and its graphical features. For instance, when reading an ER, a student will employ perceptual cognitive mechanisms such as recognition and organisation of patterns, shapes and colours (e.g. Kosslyn 1989, 1985), visuo-spatial operations (e.g. Lowe, 1993; Lord, 1987a), visualisation (McCormick *et al.*, 1987), distinguishing relationships between ER features (e.g. Shubbar, 1990) and mentally organising the visual information on the ER (e.g. Ward and Wandersee, 2002).

The **C-M** interactive factor (Table 6.1) of the model was defined as representing the nature of the conceptual (propositional) knowledge represented by the ER, including the extent, complexity and soundness of such knowledge. It also includes both the conceptual

knowledge that is communicated through, or represented by, the graphical markings and symbolism used to construct the ER, and the knowledge of the meaning of the symbolism and conventions employed in the ER to communicate the science. For example, the meaning of the blue symbol " **x** " in Fig. 5.2 G i.e. that it is an x, y coordinate.

The **C-R-M** interactive factor represents a student's ability to engage all factors of the model, by utilizing their reasoning skills (**R**) to reason with both their conceptual knowledge of relevance to the ER (**C** and **R-C**) and with the ER itself (**M** and **R-M**) so as to successfully interpret, visualise and learn from the conceptual (propositional) knowledge represented by the ER (**C-M**) (Table 6.1). For example, the process could take the following form. Upon reading the ER, the individual deciphers and decodes the visual information on the ER (**R-M**) and, in so doing, links their interpretation to, and filters their interpretation through, already existing current knowledge (**R-C**) (e.g. Anderson *et al.*, 2000). The outcome of this process could result in the construction of a unique conception consistent with accepted scientific knowledge (**C-M**) or an erroneous conception, inconsistent with a scientific worldview (e.g. Osborne and Wittrock, 1983; von Glasersveld, 1983). Hence, the scenario described above would be based on a combination of all three factors (**C-R-M**), during which all factors comprising the model would, at some time or other, be engaged resulting in the student hopefully interpreting, visualising and learning from the ER.

6.3.3 Using 3P-SIT to empirically validate the model

The following empirical data validated the model and its component factors and informed the development of the above operational definitions for each factor.

6.3.3.1 Validation of the Conceptual Factor (C)

Data from Phase 1 of 3P-SIT allowed us to validate the importance of students' prior knowledge, i.e. the conceptual factor (**C**), as one component of the model affecting students' ability to interpret an ER. To do this, students' prior conceptual understanding of antibody structure and antibody-antigen interaction was obtained before exposure to any ER. For example, the following two student quotations from Phase 1 show a sound scientific knowledge of the nature of Ab-Ag binding:

S: ... ok, you'd have two binding regions that look the same on an antibody molecule ...and ... they'll [binding regions] recognise the same antigen.

S: The structure of an antibody...consists of four chains... two light chains and two heavy chains. On the N-terminal is where the antibody binds to the antigen...one antibody can bind to two antigens... there are two binding sites for binding two antigens.

In addition to the first quote above, the same student drew the following SGD (Fig. 6.3), which supported a sound visualisation and scientific understanding of the bivalent nature of Ab-Ag binding:

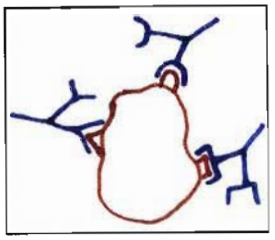


Figure 6.3 Student-generated diagram portraying a sound conceptual knowledge of the binding interaction between Ab and Ag

In contrast to the above, various students showed a range of conceptual difficulties, in support of the **C** factor. For example, three students erroneously thought that an antibody only had *one* possible binding site for an antigen and that this site was the entire 'V' cleft of the Y-shaped antibody, instead of the two variable binding domains. Two typical student quotes that showed this conceptual difficulty, were as follows:

I: Ok, how does that [the antigenic binding site] actually look?
S: ... it is Y-shaped.
I: Ok, Y-shaped...
S: It [Ag] is kind of like an upside down pyramid that tries to fit into that Y-shape.

S: This is the antigen [inserts and labels Ag on Fig. 6.4(b)] ...ja [yes], the antigen. And the antibody would be like that [inserts top rt Ab]... it [Ab] forms a complex when it binds on. That would be like one antibody to one antigen... the normal thing that happens is one antigen to one antibody... that's the specificity.
I: Where is the actual binding, on your diagram?
S: Well, it should be complementary, there must be a site here, a binding site on the antigen that it [Ab] binds to... for example, this head area here [points within V-cleft of top rt Ab]... that area has the sequence that binds on to the antigen [inserts triangular epitope on top rt of Ag on Fig. 6.4(b)], and that is how it [Ab] binds onto it [Ag].

The above quotations illustrate that the first student had the idea of an "upside down pyramid", which tries to fit into the V-cleft, while the second student associated the specificity of antibody-antigen binding to the fact that only a single antibody can bind to a single antigen.

These findings are affirmed by the following diagrams (Fig. 6.4), generated by the same two students, which both depict a single antigen-binding site on the antibody with the 'V' cleft of the Y-shaped antibody accommodating the antigen.

(a) (b)

Figure 6.4 Two examples, (a) and (b) of student-generated diagrams obtained during students' verbal explanations of antibody-antigen binding

It is clear from the above examples that the idea of *specificity* between antibody and antigen was very pronounced. For example, accompanying students' explanations of antibody-antigen binding were statements such as, 'a key unlocking a specific lock', 'complementary shapes', 'two-piece puzzle', 'specific fit', 'fit into a pocket', 'compatibility', and 'join perfectly'.

Two other students showed an interesting variation of the above Ab-Ag binding conceptual difficulty. As illustrated in the following quotation, and accompanying SGD (Fig. 6.5) from one of the students, even though they accurately represented *both* antigen binding sites (see two black circles), they nevertheless still believe that the antigen binds into the V-cleft of the antibody.

I: ...where are the actual binding sites on the antibody molecule?
S: Ok... [S beg. to gen. Fig 6.5]... these are your binding sites here [inserts black circular shaped sites on Ab]...the components within these two domains are responsible for recognising antigen.
I: ... show me where the antigen would be when there is an antigen-antibody complex.
S: Ok... these are the binding sites [points to black circular shapes]... the antigen will basically fit in between here... between these domains [draws green antigen fitting into V-shape of Ab]. So, that would be your antigen. Probably some kind of interaction occurs there [points to circular shapes].
I: And what are these regions over here [points just below circular binding domains on V cleft of Fig 6.5]?
S: I would say they are also part of the binding domains, because this is where the antigen binds to [indicates entire V-cleft], so one would have to assume that this whole kind of region here [indicates by inserting red bracket on side of Ab] will also be part of the binding site.

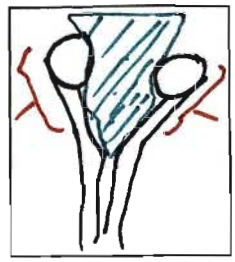


Figure 6.5 Student-generated diagram accurately depicting both antigen-binding sites on the antibody, but erroneously portraying antigen as binding into the V-cleft of the antibody

Thus clearly, some students possess conceptual difficulties with the structural mechanisms of antibody-antigen binding in that they believe that both the two binding sites and the "V-cleft" are simultaneously responsible for recognising a single antigen. This constitutes an example of how some students can hold two different mental models of the same concept (e.g Ainsworth *et al.*, 1998), in this case one sound and one unsound.

Possible origins of the binding misconception include the following. The first possible source could be students' understanding of the 'lock-and-key' analogy, used by instructors and textbooks to describe specific binding interactions between biomolecules (e.g. enzyme-substrate binding). The analogy emphasises that for a *fit* between biomolecules to occur, both participating elements must have a complementary and specific shape (e.g. Stryer, 1995; Mathews and van Holde, 1990). The following two student quotes illustrate expression of the analogy:

> S: The antibody binds to the antigen by a lock and key method... which means that it [Ab] has like a specific shape, and that is how it will bind. [...] if the antigen wasn't a specific shape it wouldn't bind to the receptor site on the antibody.

> S: ...It's a very specific interaction between antigen and antibody. The antibody has to be specific to the epitope found on the antigen, which is with regard to the lock-and-key mechanism, which I keep reverting to. It has to fit properly otherwise it won't bind. So, it actually has to be compatible...

As demonstrated by the quotes above, the lock-and-key metaphor was very ingrained in the students' conceptual understanding of antibody-antigen binding. In fact, it was largely shared by all the participants in the study. Although, at its inception, Fischer (1894) used the lock-and-key metaphor to exclusively describe enzyme-substrate interaction. The metaphor can clearly also be applied to antibody-antigen binding since the structural basis of binding is synonymous in both cases (e.g. Roitt, 1997; Amit *et al.*, 1986). However, for enzyme-substrate reactions, typical lock-and-key ERs usually show the simple situation of a *single*

enzyme binding to a *single* substrate (e.g. Ritter, 1996). When applying the lock-and-key analogy to the context of Ab-Ag binding it is possible, therefore, that students may have thought that a *single* immunoglobulin G molecule can only bind to a *single* antigen molecule. Thus, one possible source of the binding difficulty is that students did not envisage *two* lock-and-key scenarios: one occurring at each binding site. Also, some students' may have considered the analogy *itself* to be a complete and realistic depiction of actual antigen-antibody interaction *in vivo* (e.g. Orgill and Bodner, 2004). As a result, students may have interpreted the analogy literally, instead of taking the analogy to be only a *representation* of reality (e.g. Hill, 1990), resulting in the alternative conception of Ag binding into the "V-cleft" of an antibody.

A second possible source of the binding misconception is that, in addition to enzyme-substrate reactions, students may have been associating other 'single' biochemical interactions with their ideas of antibody-antigen binding. These could include a single ligand binding into a singular receptor site, as is the case when a peptide binds within the single cleft of an MHC molecule (e.g. Roitt, 1997). Additionally, antibody 'advert' molecules present on B-cell membranes are often depicted, for simplicities sake, as having only a *single* binding cleft (Gupthar, pers. comm.) to highlight the fact that it is specificity between Ab and Ag molecules which render B-cells active. Thus students' may have constructed mental models and concepts of Ab-Ag binding from ERs that are oversimplified. As a result, some students expressed limited and literal mental models of Ab-Ag binding, failing to consider the strengths and weaknesses of these ERs and their own conceptual understanding.

A third possible source of the binding misconception could be students' lack of any other explanative models to describe binding, other than the lock-and-key analogy. For instance, Koshland's (1963) notion of an induced-fit between Ab and Ag was found to be almost completely absent from students' conceptual knowledge, with only a single student exposing the idea. In this case, students seemed to only expose conceptual knowledge relating to the "physical fit" between Ab and Ag and possessed little conceptual understanding of other stereospecific considerations such as the role of amino acid side chains or intermolecular forces such as hydrophobicity and electrostatic interaction during binding.

In summary, the **C** factor as defined in Table 6.1 is clearly a key and indispensable component of the model in that the nature of a student's prior knowledge, whether sound or

erroneous, will seriously affect their ability to interpret an ER representing such knowledge (e.g. Lowe, 1993; Winn, 1993). The above analysis has also shown how the conceptual difficulty data, corresponding to the **C** factor, can be used to isolate the possible source(s) of a certain conceptual difficulty. In chapter 7 we will show how knowledge of the nature of a difficulty and its source can inform the design of possible remediation strategies (e.g. Pintó and Ametller, 2002).

6.3.3.2 Validation of the Reasoning Factor (R)

As described in section 6.3.2 (Table 6.1), the **R** factor of the expressed model (Fig. 6.2) represents those *cognitive processes* whereby students reason with both the ER and their own conceptual knowledge in order to interpret a scientific ER. The present study identified at least five different reasoning mechanisms associated with students' interpretation of ERs. Firstly, some students employed *surface-level reasoning* (Chi *et al.*, 1981) when processing the graphical markings on the ERs. These students interpreted the ER markings literally and at face value, without considering the deeper meaning of the markings (e.g. Ametller and Pintó, 2002; Cheng *et al.*, 2001; Lowe, 1993). As Olivier (2001) has pointed out, students who employ surface-level reasoning rely heavily on perceptual processes when interpreting ERs, rather than deeper knowledge structures. Secondly, our data suggested that some students performed *inappropriate analogical reasoning* when interpreting the ERs (e.g. Sumfleth and Telgenbüscher, 2001). As introduced during the validation of the **C** factor above, this was found to be the case especially when students battled to use the lock-and-key analogy as a tool with which to explain the nature and specificity of antibody-antigen binding. Thirdly, some students were found to engage in *inappropriate transfer* (Salomon and Perkins, 1989) when interpreting the ERs. Here, the students inappropriately transferred a particular biochemical concept (e.g. destruction of invading pathogens) from the context of cellular immune responses to the context of primary antibody-antigen binding. Fourthly, and related to the former, some students found it difficult to *translate* between different ERs, which all represent the same concept or phenomenon. In particular, these students could not map between one ER and another, probably because students treated each ER as a unique situation, instead of viewing all the ERs as being multiple representations of the same scientific concept (e.g. Gobert and Clement, 1999; Ainsworth *et al.*, 1998). Fifthly, we also discovered what we have termed the apparent *superimposing* of one concept upon another. Here, some students

tended to fuse two or more distinctively different concepts together into a single explanative model, leading to the moulding of scientifically inaccurate conceptions (see for example section 4.3.3.2 in which DNA and Ab concepts were fused). The superimposing of concepts could be related to a recent finding by Grayson (2004), who has referred to a similar phenomenon in the context of electric circuit ERs. In this case, it was found that some students struggled to *disentangle* the distinctively different concepts of current and energy from one another.

Now that the above five reasoning processes, corresponding to the **R** factor of the model, have been introduced, data is presented that supports the influence and importance of these processes as components of the **R-M** and **R-C** factors of the model. In this regard, these isolated reasoning mechanisms acquire meaning only when they are observed as cognitive processes in *action.* In other words, reasoning processes can only be observed if there is something to reason with, in this case with the ER (**R-M**) and with students' own conceptual knowledge (**R-C**). Hence, the reason for including the **R-C** and **R-M** factors as components of the model is that each can be considered a subset of the overall reasoning factor (**R**), which has an indispensable effect on a student's ability to interpret an ER. Therefore, empirical data pertaining to the **R** factor is composed of that empirical data corresponding to both the **R-M** (see section 6.3.3.4) and **R-C** (see section 6.3.3.5) factors below.

6.3.3.3 Validation of the Representation Mode Factor (M)

During Phase 3 (see section 5.4.3) of the 3P-SIT interview process (Chapter 5), we were concerned with collecting data that supported tenets of the **M** factor (Table 6.1) of the model. As outlined in the operational definition (Table 6.1), the **M** factor is concerned with that information that corresponds to the nature of the ER in isolation and how well (or poorly) the graphical markings that constitute the ER represent what it is designed to represent (Table 6.1). By validating the **M** factor, we attempted to identify those external characteristics of the display that may cause student difficulties. In other words, data corresponding to the **M** factor centres around the effective or ineffective use and clarity of the graphical and diagrammatic features, namely, the spatial arrangement of the ER elements, ER conventions, visual icons, visual cues, artistic devices, colour, topography, level of abstraction, symbols, labels, captions

and other ER features. Thus the objective is to better understand what external features of the ER may be giving students problems, or initiating particular reasoning patterns.

As pointed out in Table 6.1, information pertaining to factor **M** can be obtained from experts including scientists, researchers and graphic artists as well as students' evaluation of the ER. In addition, similarly to the study reported in Chapter 4 (section 4.2.2), information corresponding to the graphical features of an ER (**M**) was also obtained from the author's informal visual analysis of the ERs in which they were screened to identify those ER markings that could potentially induce erroneous interpretations. For example, upon an informal visual analysis of ER E, prior to exposing it to the participants, the author suspected that the visual clarity of the "realistic" graphical depiction of structural features representing antibody structure and binding to antigen might have been a possible source of confusion for students, which is shown by the following opinion:

> "Due to the realistic nature of the electron micrograph obtained at such a high magnification, students may think that antibodies can "join" to form trimer arrangements through bonds that are not non-covalent. In light of this, it may be difficult for students to identify the approximate location of a single antibody structure and its antigen-binding sites within the trimeric and pentameric shapes."

The same graphical feature on the electron micrograph (ER E) was considered by an expert immunologist (Coetzer, pers. comm.) to be a potential problem for students. Consider the following expert opinion, which supports this:

> "Students would possibly have some difficulty in interpreting the electron micrograph without an explanation for the way this negative stain was obtained and that the spiky bits sticking out are the Fc fragments..."

The expert and author opinion above is supported by the following two student extracts pertaining to the clarity of the same graphical features on the electron micrograph (ER E) and constitute further evidence for the nature of the ER (**M**):

> I: Is there anything that you don't understand or find confusing on this representation [ER E]?
> S: ...The only thing is like...where the bonds form between the different antibodies.

> S: What I cannot see is the hapten, yeah. From the information [points to caption of ER E] I can have the assumption that the haptens should be on the N-terminals of these antibodies... yeah. I also can't see if these antibodies have two chains... but I know that, in reality, they have two light chains and heavy chains... yeah.

The above students' extracts demonstrate how the graphical features representing the nature of the visual clarity of the trimer and pentamer Ab-Ag complexes influenced their reasoning. In the first case, the student thought that the Y-shaped antibodies were somehow joined together,

rather than being bonded to haptens present in between the antibodies. Due to the clarity of the visual information on the micrograph (ER E), it is impossible to see the hapten (antigen) molecules and, from a purely visual perspective, the antibodies do look like they are 'joined' without hapten. Since haptens are small molecules with low molecular weights, the magnification used to generate the micrograph was not enough to expose their presence as distinct visual features. The second student above realised that the haptens could not be viewed on the micrograph directly and, therefore, reinforced the fact that the lack of clarity of this ER feature (M) might affect students' interpretation of ER E.

During an informal visual analysis of ER F, the author anticipated that the use of the red colouring (see Table 6.1) on the ER might create a problem for students. The author's opinion in this regard was as follows:

> "Use of the same red colour to show the Gln 121 residue (in frames 'a' and 'b') as well as the amino acids involved during Ab-Ag contact (in frame 'c') may cause some students to think that some type of biochemical event has occurred resulting in "more spheres" in the last frame...".

During construction of the opinion above, the author observed that the same red colouring is used to show both the glutamine residue involved in the antibody-antigen binding (frame 'b') and the contact residues between antibody and antigen (frame 'c'). In support of this concern, this colouring feature of the red 'spheres' on ER F led one student to make the following comments:

> I: Is there anything that you find particularly confusing on the diagram [ER F]?
> S: The glutamine... and how it sort of multiplies. There is no sort of step on how to... how they got to so many, or why there are so many [glutamine residues]. Why is it [Ab and Ag] attached first, and then just pulled apart... You know normally, like if you get a negative and a negative, that is how come it will like pull apart, but then it wouldn't make sense if it was attached in the first place. I don't understand how they get from there [points to frame a] to part 'b' and why there are so many glutamine molecules there [points to red spheres on Fab in frame c and then to red spheres in Ag in frame c].... I'm just looking at this diagram and I don't understand the steps and how to get to the next one [step].

It is evident from the above extract that the student thought that 'multiplication' of the single glutamine residue had occurred. It is very possible that this reasoning could have been as a result of the same (red) colouring technique used to show two very different ideas, one idea being the location of the glutamine, and the other being the idea of contact areas between antibody and antigen. In addition, by labelling the frames in ER F as 'a', 'b' and 'c', students may have attached some idea of sequence to the ER and interpreted the ERs as a set of three consecutive events rather than different representations of the same phenomenon. Thus the

inappropriate use of colour on the ER (**M**) (Table 6.1) may have affected students' interpretation of ER F.

Lastly, during the author's visual analysis the ELISA curves (ER G), the author suspected that the graphical features representing the "-log" expression might induce erroneous student interpretations of the ER. The author's thinking is demonstrated by the following quote:

> "Students may think that antibody concentration increases as one reads the graph from 'left-to-right'. Concurrently, this may cause problems since the coloured curves for each week are showing a negative slope as one reads from left-to-right."

In support of the author's observation, an expert (Coetzer, pers. comm.) also identified the "-log" graphical feature in ER G as a potential source of confusion. This was shown by the following opinion:

> "If students are not very familiar with this format of expressing ELISA results, they may be confused by the appearance of the – log (antibody concentration) plot, i.e. that the "big numbers" represent low antibody concentration. Expressing antibody concentration in µg/ml gets around this potential problem..."

The expert's evaluation above reinforces the author's notion that the "- log" graphical feature (**M**) of ER G may pose potential processing difficulties for students. In addition to the above evidence, consider a student's quotation that also emphasises the use of the "-log" expression in the ELISA (ER G):

> S: ...according to the graph... at a high concentration [of Ab] we have less absorbance, which is really confusing me because, the concentration increases with the absorbance. But, I think the thing that makes the graph look like this is this 'log'... It is a bit confusing, really, because now, the absorbance decreases but the concentration still increases [points to x-ax.]...

It is evident from the quotation above that the student identifies the "- log" expression as the graphical feature (**M**) that causes certain confusion. In real terms, since negative values were obtained when the logarithm of Ab concentrations (mg/ml) were calculated, the experimenter (Jackson, pers. comm.) who constructed ER G had to assign a negative value to the calculated values to place the curves in the positive Cartesian quadrant. Students who identified the "-log" as one graphical symbol (**M**) that caused confusion probably drew inferences from the numerical increase on the x-axis rather than considering the deeper arithmetic meaning of the "increase". It is clear that the author's, expert's and student's opinions above all show evidence that the graphical symbolism (**M**) used to portray information can greatly influence the manner in which students interpret ERs.

Overall, by analysing data generated from Phase 3 of 3P-SIT, an appreciation of the potential effect of different diagrammatic, pictorial and graphical markings on students ER-processing could be harnessed. In turn, such data helps us not only to locate and identify specific ER features, which may induce difficulties, but may also help formulate criteria and guidelines for the optimal design and presentation of biochemistry ERs (see Tables 7.4 and 7.5, Chapter 7). The data above has validated the importance of the **M** factor of the model as a key and essential factor contributing to students' ability to interpret ERs in science.

6.3.3.4 Validation of the Reasoning-Mode (R-M) Factor

Following Phase 1 of the interview process, an ER (Fig. 5.2) is presented to the participants for interpretation, which marks the beginning of Phase 2 of 3P-SIT and the collection of data on student reasoning (see section 5.4.2). As discussed, the **R-M** factor is representative of the reasoning processes that operate when a viewer specifically tries to read and make sense of the graphical markings in an ER. An example of empirical data that supported unsound reasoning with the ER, and therefore the **R-M** factor, was shown by two students who interpreted the glutamine residue depicted in red in ER F, as having undergone some type of active digestion process. This thinking was displayed by the following interview extract:

I: What does this plate over here represent [points to frame c in ER F]?
[...]
S: ...interaction [between the antibody and the lyso.] caused the glutamine to break down and join with the antibody [points on frame c]. The antibody is actually working on the glutamine [circular pointing on frame c]... the antibody is probably responding to the lysozyme... the antibody is breaking down the molecule [lyso.].
I: How?
[...]
S: The antibody has receptors that go into this molecule [points to lyso. on frame c] and then works on it [Ag] and breaks it [Ag] down... yeah, and that is how you get this glutamine [points to red spheres on frame c].

The students who showed this unsound interpretation thought that the single red glutamine molecule represented on frames 'a' and 'b' (ER F) had in some manner been degraded, so as to produce the scenario that appears on frame 'c' of ER F. It is evident from the above quotation that the student was interpreting the red 'spheres' on the ER superficially and that an over reliance on the graphical markings had resulted in surface-level processing (Chi *et al.*, 1981), rather than a deeper appreciation of what the markings actually meant (e.g. Cheng *et al.*, 2001; Olivier, 2001). That is, instead of interpreting the red spheres in frame 'c' as contact amino acid residues between Ab and Ag during binding, the student erroneously attributed a

digestive process to the 'increase' in the number of red spheres in frame 'c'. Thus the student inappropriately decoded the symbolism (Table 6.1) used to represent the amino acids involved in binding. Furthermore, since the student viewed the antibody as the entity responsible for the destruction of the Ag, the student transferred her conceptual knowledge inappropriately (Table 6.1) by interpreting a primary interaction as a cellular immune response.

In contrast to the above reasoning difficulty, the following two student quotes provided evidence for sound reasoning with the ER (Fig 5.2 F) in support of the **R-M** factor of the model:

> S: ... this is the antigen [points to lyso in frame c]... the lysozyme... it shows how it fits onto that molecule [points to Fab in frame c]. So, this is the paratope [points correctly] and that is the epitope [points correctly]. And, this [points to red spheres on lyso and Fab in frame c] shows the position of the molecules that facilitate that association.

> S: ...[frame] 'c' shows what is involved in the binding... it shows the actual atoms involved in the binding... by highlighting the specific atoms and numbering them...

As further evidence for the tenets of the **R-M** factor (Table 6.1), eight of the nine respondents struggled to accurately visualise the biochemical structures portrayed in ER F. Whereas the space-filling display (ER F) only represents a single 'arm' or Fab fragment of IgG, these students visualised it as the *complete* Y-shaped antibody. For instance, consider the following SGD (Fig. 6.6) obtained from one of these students:

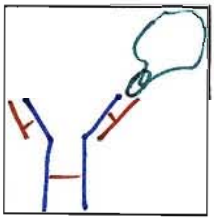


Figure 6.6 An SGD portraying the misinterpretation of the Fab arm as an upright and complete Y-shaped antibody

The student's verbal explanation, which corresponded to Fig. 6.6 above, was as follows:

> I: In terms of structure, what is being shown on this representation [ER F]?
> S: ...you can see the antibody structure... one can see that it consists of the two chains [H and L]... it is actually two heavy chains [points to bot. two 'groups' of blue spheres making up the H-chain simultaneously] and two light chains [points to top two 'groups' of yellow spheres making up the L-chain simultaneously].
> [...]
> I: ...Could you relate the markings that you've drawn on paper [Fig. 6.6] to what is visually represented on the actual diagram [ER F]?

S: Ok... that's [points to Ag in Fig. 6.6] your antigen there [points to green Ag in frame a]. This would be your epitope [points to small green oval on top Ag of Fig. 6.6], your actual region of binding, which is glutamine, so that is your red part [points on ER F], that little blob sort of part sticking out [red Gln in frame a]. Then, these [points to each 'group' of blue spheres on Fab in frame a] are your two heavy chains [points to each lower part of H chains on Fig. 6.6]... these are your two light chains [points to each 'group' of yellow spheres on Fab in frame 'a' and then to each red light chain on Fig. 6.6].
I: If I were to bind an antigen over there [points to lt binding site on Fig. 6.6], how would that look [in ER F]?
S: What I'm thinking is that it [Ag] would actually come in from this side [points to lt of Fab on frame a], so it would actually be more or less a mirror image of this molecule [points to green Ag on frame a], but on that side [points to lt of Fab on frame a].

Based on the above extract, the SGD in Fig. 6.6, as well as an analysis of the student's observable behaviours, such as pointing and indicating to different components of the ER, it was clear that the Fab arm represented by the space-filling ER was interpreted as an entire Y-shaped antibody. This was further supported when the students described how the space-filling ER would appear if another antigen had bound to the other antigen binding-site, depicted in the SGD (Fig. 6.6). In this case, the student indicated that the other erroneous 'binding-site' on the space-filling ER would be where another antigen could bind. In this case the student has attributed the general shape and topography of the grouped cluster of spheres in the ER to the visualisation of a complete and upright Y-shaped antibody. One possible source for this reasoning is that the student could not distinguish between, and organise, the visual information on the ER appropriately (Table 6.1 and e.g. Kozma and Russell, 1997; Bennett and Flach, 1992). As a result, the student erroneously translated (Table 6.1) between the ER portrayed in Fig. 5.2 F and her mental models of other more common textbook ERs (like ER A, Fig. 4.1) that portray antibodies as upright and complete Y-structures (e.g. Brna *et al.*, 2001; Gobert and Clement, 1999; Ainsworth *et al.*, 1998). Note, in addition her SGD (Fig. 6.6) showed no attempt to reproduce a space-filling ER as in ER F. Instead, she switched to a different representation, probably one that was her own mental model. In summary, identification of this difficulty provides concrete evidence that sound reasoning with the ER is crucial for sound interpretation of the ER and thus the **R-M** factor is an important component of the proposed model.

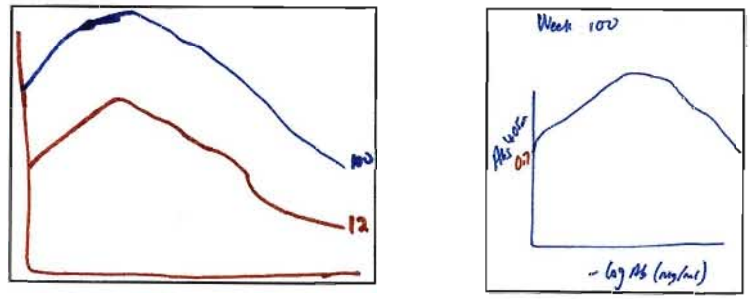
Even though eight students demonstrated the above reasoning difficulty, one student showed evidence of sound processing of ER F with respect to the structural components represented by the ER. This is clear from the following interview extract:

S: ...Basically, on this structure [ER F], you'll be representing one arm of your molecule. You have two of these [arms] on your entire antibody molecule... it [ER F] is just showing one arm.

Lastly, regarding data that supports the tenets of the **R-M** factor, students were asked how the ELISA graph (ER G) would appear if absorbance results for week one hundred were plotted on the same curve. Realistically, at week one hundred, the experimental serum obtained from the laboratory animal would show an antibody concentration very close to pre-immune levels (green curve on ER G), due to the lack of experimental antigen, which is needed to stimulate antibody production. An example of a response demonstrating sound reasoning with the graph (i.e. factor **R-M**) in terms of this scenario, was as follows:

I: Consider that we stopped the experiment...at week one hundred, we took another sample, and we did a plot, how would that look here?
[...]
S: It will be something like the pre-immune... because... there won't be antigens in your system to make you produce antibodies... or increase your antibody production.

However, in contrast to the above, two students thought that the absorbance value for week 100 would be higher than for week twelve. This reasoning was demonstrated by the following SGDs (Fig. 6.7):



(a) (b)

Figure 6.7 Two SGDs (a and b) demonstrating students interpretation of ER G when predicting an absorbance curve for collection of serum samples at week one hundred, after the booster schedule had ceased

Corresponding audiotaped quotations from the same two students was as follows:

I: Say they [the researchers] had finished taking readings and had finished the experiment. Then, they plotted for say, week 100, how would the graphs look then?
S: It would probably be a higher value than twelve, with bigger absorbance values...[student proceeds to draw Fig. 6.7(a)]... here we have week twelve. When I'm looking at this graph [ER G], I would think that week 100 would be somewhere up there, with a similar effect... with it [curve] going higher then coming down.

I: Consider that the experiment had finished...but at a week one hundred, the researchers decided to take a sample and plot it on this curve. How would that curve look?
S: For week hundred... it is going to increase like [traces a curve above the wk 12 curve on ER G with finger], more than these ones [the actual EL curves underneath the trace on ER G]... at point six maybe [points above 0.45 absorbance reading on y-ax.]... [S beg. to gen. Fig. 6.7(b)]. Ok, that is absorbance at 405... it is going to look like this... this peak [higher curve on Fig. 6.7(b)] will be more than that one [wk 12 curve in ER G] over there... the [absorbance] value would be like... point seven.

A possible source of the above difficulty, corresponding to the **R-M** factor (Table 6.1), is that the students may have placed greater emphasis on the visual relationships on the graph (ER G) rather than engaging their knowledge of ELISA concepts to consider the biochemical implications of ceasing booster injections. For instance, these students used the visual trend of a 'higher' graph corresponding to a higher week number to solve the task. Thus they were reasoning in a linear manner with the graphical data rather than thinking deeply about the related biochemistry. In other words, **R-M** was playing a dominant role while **R-C** (see below), was being neglected by the student during the interpretation. As a result, the students showed surface-level reasoning (Table 6.1) when interpreting the ELISA curves and relied heavily on the graphical markings to do so (e.g. Lowe, 1993; Egan and Schwartz, 1979).

In summary, the importance of the **R-M** factor as a component of the model (Fig. 6.2) was illustrated by showing how both sound and unsound cognitive processing of an ER affects the manner in which a student interprets an ER. The synthesis provided above also demonstrates how data corresponding to the **R-M** factor can be analysed to identify the possible sources of students' difficulties with ER interpretation.

6.3.3.5 Validation of the Reasoning-Conceptual (R-C) Factor

When interpreting scientific ERs, students should not only be deciphering and processing graphical features of the ER (**R-M**), but also integrating this information into, and filtering this information through, their already existing conceptual knowledge (e.g. Ward and Wandersee, 2002; von Glasersfeld, 1989). In other words, interpreting an ER also requires a student to engage their conceptual understanding of the scientific phenomenon that is represented by an ER and to use a wide range of cognitive processes to achieve this. The ability of a student to reason with their conceptual knowledge of relevance to the ER is represented by the **R-C** factor of the model (Table 6.1).

Since Phase 1 of 3P-SIT (Chapter 5) allowed us to first establish the nature and extent of a student's prior knowledge of relevance to the ER (Factor **C**), in Phase 2 we were able to establish the extent to which the student engaged this conceptual knowledge when subsequently interpreting an ER. For example, during Phase 2 when interpreting ER E, one student was shown to rely heavily on his/her unsound conceptual understanding (measured in Phase 1) to interpret the ER during Phase 2. This is demonstrated by the following extract obtained during Phase 1 followed by a SGD (Fig. 6.8) and quotation from Phase 2 of the same interview:

> S: ... antibodies... they form complexes with the antigen in order to destroy it or engulf it.
> [...]
> S: ...they [Ab and Ag] will form like a lock and key mechanism and join. Ja [yes], if they're [Ab and Ag] exactly the right sequence on each of them they will join perfectly together.
> S: ...the antibody has certain compounds in it... that infiltrate the antigen, when it [Ab] engulfs it or whatever and binds to it [Ag]... and sort of breaks down the different components in the antigen... the antibody could contain for example an enzyme with it and this enzyme could contain digestive stuff in it... it would sort of engulf... well it would bind onto it [Ag] and then release these things when it binds to the antigen.
> [...]
> S: And then this antibody...this little antibody infiltrates the antigen and releases little granules that contain the digestive enzyme and then these things degrade the whole antigen into smaller things...

Now, consider the following diagram (Fig. 6.8) and corresponding verbal commentary from the same student during interpretation of the electron micrograph (ER E), during Phase 2 of the interview:

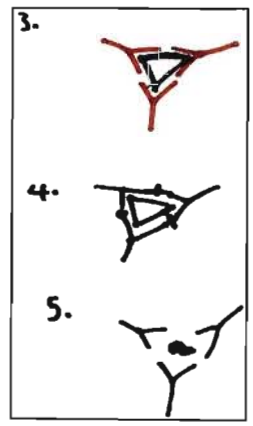


Figure 6.8 SGD from Phase 2 of 3P-SIT showing a student's dependence on certain conceptual knowledge when interpreting ER E

> S: ... step four [labelled '4' on Fig. 6.8], they [Abs] form a trimer. The different antibodies bind to three sites ['V-clefts']...and then they [Abs] join... to form a trimer.
> I: What would happen after [step] four [on Fig. 6.8]?
> S: The antibody has done its function of removing this hapten molecule....the hapten would be gone, taken out of the blood. Yeah, it [hapten] gets broken down and destroyed. Then, the antibody... once it has destroyed this hapten molecule... you just left with antibodies.

I: Ok. Between [step] four and five ['4' and '5' on Fig. 6.8] what is going on?
S: Ok, four...once antibody has bound onto the hapten molecule, um... they [Abs] start their action... whatever they [Abs] have inside in them ... the granules... they move in and then they [granules from Ab] start destroying the hapten molecule...The antibody molecule removes this molecule [hapten] from the blood.

Based on the SGD (Fig. 6.8) and interview extract obtained from the student above, three reasoning processes are of relevance to the **R-C** factor (Table 6.1) of the model. Firstly, the student is clearly demonstrating inappropriate analogical reasoning (Table 6.1) by using the ingrained lock-and-key analogy from his/her conceptual knowledge to facilitate reasoning with ER E. As displayed by Fig. 6.8, the student has applied the lock-and-key analogy by inserting the hapten (antigen) molecule into the centre of the trimer. Unfortunately, the student is not utilising the analogy in the appropriate manner and is thus displaying erroneous analogical reasoning (e.g. Sumfleth and Telgenbüscher, 2001) when interpreting the ER (Table 6.1). This finding has been supported by Orgill and Bodner (2004) who reported that biochemistry students' often lack clear ideas as to the purpose of analogies and how to use them as learning or reasoning tools. Secondly, the student is inappropriately transferring concepts (e.g. Salomon and Perkins, 1989) reserved for cellular immune function to the domain of primary interaction. It is the cellular immune response that is responsible for 'killing' and 'digesting' the antigen (e.g. Simonneaux, 2000) and not the primary response, as suggested by the student. Thirdly, the student is selecting at least two misconceptions from his/her prior knowledge (see Phase 1 quote above) to interpret the ER. Specifically, selection of the misconception that the antibody is the agent that destroys the antigen, as well as the misconception that the Ag binds into the V-cleft of the Ab, had a very pronounced effect on the way the student reasoned with their conceptual knowledge to make sense of ER E. Thus in this case the student relies heavily on the selection of these misconceptions (**R-C**) to interpret the ER.

It is evident from the above data, that the student's interpretation of the ER is significantly affected by reasoning processes represented by the **R-C** factor of the model, in particular with respect to erroneous analogical reasoning, inappropriate transfer of knowledge and selection of scientifically unsound concepts (Table 6.1). In comparison with the data above, consider the following interview extract and SGD (Fig. 6.9) from a student who showed sound reasoning with his/her conceptual knowledge represented by factor **R-C**, when interpreting ER E during Phase 2 of the interview:

S: ...the divalent hapten is going to attract an antibody from each side [indicates on ER E], so that is why it holds those two together [points]... then it holds those two [points] together... and it holds those two [points]. It actually agglutinates and forms a clump.
[...]
S: ...I've drawn the hapten as a sphere, so I've actually drawn the fragment antigen binding-site as um... a little curve to fit the sphere [points to rt b-site on Fig. 6.9]...
I: Ok, so where would a lock-and-key interaction happen here [Fig. 6.9]?
S: Um, well on both sides of the hapten. Because, if you see here [indicates Fig. 6.9], it would happen on this side and on this side [indicates with bot. hapten on Fig. 6.9]. So, there'd be like two lock-and-key interactions on both sides.

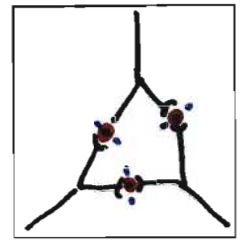


Figure 6.9 SGD showing sound reasoning with the lock-and-key analogy represented by the **R-C** factor of the model

Clearly, this student was able to select and engage sound scientific conceptual knowledge, as well as successfully apply his/her knowledge of the lock-and-key analogy to interpret the ER (Fig 5.2 E). In so doing, the student correctly suggested that linking between antibodies and the divalent antigen allows agglutination to occur rather than, as in the case of the previous student, the antibody itself being responsible for elimination of the antigen. Hence, the data above provides evidence for sound conceptual reasoning processes, represented by factor **R-C**, in that the student is able to engage his/her sound conceptual knowledge when interpreting ER E.

A further intriguing situation, in support of the tenets of the **R-C** factor, was one where students were found to fuse two distinctly different concepts together when attempting to interpret ER E. For example, one student struggled to explain the difference between the lock-and-key analogy as an *analogy* and the actual binding *mechanism* between antigen and antibody. The following SGD in Fig. 6.10 and the corresponding verbal commentary are evidence for this difficulty.

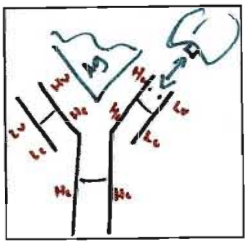


Figure 6.10 SGD showing the integration of two distinctly different ideas into one model

I: Do both sites [of the antibody] have to bind to antigen...?
S: I would say that both would have to bind, because I can't see one binding... I suppose if you have just one binding, it is not going to be a very strong interaction... I don't see how one can just bind... [starts to gen. Fig. 6.10]
[...]
I: Can you represent antigen on your diagram?
S: ...Well, normally, I talk of my lock-and-key thing, which would be here [traces 'V' within V-cleft of Ab on Fig. 6.10]... but, it would have to interact with this whole thing here [points to top lt actual binding site of Ab in Fig. 6.10], see what I'm saying? How can I represent this...well, this is my normal theory that I go back on [inserts V-shaped Ag into V-cleft of Ab on Fig. 6.10].... that is your antigen [inserts label]. So, that is my normal thing of lock-and-key... that it [Ag] has to fit. But, it [Ag] has to interact with this site and this site [inserts dots on top rt binding region of Ab on Fig. 6.10]. So, I'm supposing it's [Ag] sequence specific... so, those amino acids [on Ab] and those amino acids [on Ag] are going to interact. So, if you have an antigen there [inserts Ag at top rt of Ab on Fig. 6.10]... then you going to have an epitope on the antigen which interacts specifically here [inserts arrow].

The extract and SGD (Fig. 6.10) above provided further evidence that this student held two distinct conceptions of the same phenomenon simultaneously; one correct conception of Ab-Ag binding at the variable region of the Ab and one erroneous conception that the Ag binds into the V-cleft of the Ab. It is suggested that the student was *superimposing* both ideas and expressing them as one model, i.e. combining the lock-and-key analogy with the need for specificity between antibody and antigen. Interestingly, during interviews, the same student had stated that she needed two 'theories' to explain antibody-antigen binding. As part of her first 'theory', which can be viewed as one model for representing Ab-Ag binding, she related antibody-antigen binding to a lock-and-key situation, and as part of her second 'theory' or model, she related antibody-antigen binding to sequence specificity between amino acids. It is probable that either, the student's construction of two models to explain antibody-antigen binding was a way to alleviate the obvious conflict that had arisen when this student engaged her conceptual understanding (**R-C**) during reasoning, or, she already possessed both these ingrained models as part of her conceptual knowledge. In an attempt to clarify this issue, the same student was probed in a subsequent interview about the conflict that led to her expressing her 'two theories'. The following is the interview extract that portrayed her explanation:

I: Remember...we were speaking, and you said... your two theories...
S: Ok. Well, my two theories. Well, the one is about the lock-and-key theory... that an antigen and antibody are specific to each other as in a lock and key. But, the other theory was that an antigen binds to the two fragment antigen binding sites. And, it is only in that region that you get that interaction with the variable regions [of the antibody]. Now, why I said it was a bit conflicting because... if you look at it in a diagram, the lock-and-key thing suggests that it goes into the cleft of the antibody whereas ... if you look at the actual structures you can see that it [antigen] actually only interacts with the specific edges of the fragment antigen binding-site as such. If I had to represent it, I won't be able to do it... to represent both ['theories'] as one... as in both theories... as in binding to the fragment antigen binding-site and then also binding to the cleft.

In lieu of the above extract, one of the student's models could well have been conceptualised from interpreting oversimplified diagrams of IgG-antigen binding (e.g. on the surface of B-cells). Her other model may well have stemmed from a scientific and mechanistic definition of antibody-antigen binding, which she had also previously internalised, in that it is the sequences of amino acids between antibody and antigen that render molecules 'specific' to each other. Thus, as Grayson (2004) has found in the context of electrical circuit ERs, this student cannot *disentangle* the two models from one another.

Lastly, as further evidence of inappropriate conceptual reasoning, that corresponds to the **R-C** factor (Table 6.1), some students misinterpreted the 'increase' in absorbance or positive gradient for the week 8 and/or 12 curves in ER G. Before examining the described misinterpretation, for comparative purposes, first consider the following student quote showing a sound interpretation of the increase in absorbance shown in the week 8 and/or 12 curves:

S: Ok, the [absorbance] increase in that region here [traces wk 12 curve starting from y-ax. until halfway between P and Q with finger] could be due to the steric hindrance. You have so many antibodies that they compete for binding and eventually they shove each other off. And because there is so many [Abs] they can't bind strongly because there's too many and... so they get washed out in your wash step. It looks like you have a lower concentration of them [points on the wk 12 curve], but, as you dilute it, you have less of the steric hindrance and once you get proper binding... strongly, then you can detect it with your secondary antibody.

In comparison with the above example of sound reasoning, with the student's conceptual knowledge to interpret ER G, four students attributed the positive gradient in each of the curves to an increase in antibody concentration *per time*, rather than to factors such as steric hindrance between antibody molecules and competition for binding sites, as stated by the student above. For instance, consider the following example of a student quote that represented this difficulty:

I: Compare area 'P' and 'Q' [ER G] in terms of that blue line [points].

S: In area 'P'... in week 12 [points to P on ER G], the antibody concentration is increasing, it is on the rise, that means antibodies are being made in the system. And at 'Q' it is showing that the immune response is declining that means that less antibodies are being made... yeah [points to Q]. At 'P'... this is like a growth phase or log phase of the graph [traces graph wk12 from y-ax. to prior to Q]... it is just showing the steady growth or increase in antibody count in the immune system... it is after the booster injection has been put in, that the immune response increases... after the booster injection is put they're [Abs] reacting to this booster injection [points to wk 8 & 12], therefore they are increasing [traces wk 8 & 12 grad. prior to Q]... there is an immune response, and the amount of antibody... the count is getting higher, that means more antibodies are being made.

It is clear from the above data that the student thought that the increase in absorbance of the week 8 and 12 curves was due to an immune response that had produced an increased number of antibodies. Even though the immune response, following booster shots, *is* represented on ER G, the *three* immune responses are represented by the three curves, not *within* each curve. Thus the students who showed the difficulty were interpreting the graph as if "time", rather than "-log Ab", was plotted on the x-axis.

The four students, who manifested the above difficulty, were probably erroneously transferring and integrating their conceptual knowledge (Table 6.1), obtained from other graphs, into the interpretation of the ELISA graph (ER G). Conceptual knowledge gained from those graphs that plot antibody concentration versus time, probably influenced students' reasoning leading to them erroneously selecting this conceptual knowledge to make sense of the ER. With concentration versus time graphs, a positive gradient would indeed be represented *within* a single curve. In support of this inappropriate reasoning pattern, Scanlon (1998), has shown in a study of students' interpretation of graphs of motion, that sometimes over-generalised rules were used to interpret a graph such that distance-time graphs were treated as velocity-time graphs.

In support of the above findings, other ER research has also shown that the successful interpretation of an ER depends to a large extent on the knowledge that an individual brings to the ER (e.g. Roth, 2002; Cheng *et al.*, 2001; Lowe, 1996). These authors have shown that interpretation of an ER is indeed 'modulated' by this knowledge and the modulation plays a crucial role in determining whether the ER will be successfully interpreted or not. Thus in terms of the expressed model (Fig. 6.2) the **R-C** factor, which represents a student's ability to reason with their conceptual knowledge of relevance to the ER, is an essential component affecting students' ability to interpret scientific ERs.

6.3.3.6 Validation of the Conceptual-Mode (C-M) Factor

We defined the **C-M** factor as representing the nature of the conceptual (propositional) knowledge represented by the ER and its symbolism. It includes the extent, complexity and soundness of the knowledge represented by the ER (Table 6.1). Like for factor **M**, it requires experts to judge or evaluate something in isolation from student interaction with an ER - in this case propositional knowledge. The propositional knowledge is obtained from textbook authors', from surrounding text that describes an ER, from the captions (figure legends) used by the author to describe an ER and, from authors' descriptions of ERs and scientific findings that are presented in journals and other scientific documents. The evaluation of the propositional knowledge is done by experts, which can include the researcher or any other experienced scientist. In the current study, data corresponding to the **C-M** factor was obtained from the primary sources where the ERs were located and described (see Fig. 5.2 caption), namely two scientific papers and the prescribed textbook for the immunology module (section 6.2.1) for ER E and F and, discussions with a colleague for ER G (Jackson, pers. comm.).

The conceptual (propositional) knowledge represented by **C-M** is a significant and indispensable factor that affects a student's interpretation of an ER. This is because the complexity, soundness and extent of knowledge that the ER is designed to represent will have a profound affect on how well the ER is interpreted. For example, a highly complex ER that contains a large and complicated body of knowledge will be difficult to interpret. For the same reason, an ER that represents unsound or conflicting propositional knowledge could also negatively affect a student's interpretation of an ER. For example, the study reported in Chapter 4 (section 4.3.2.6) showed that for ERs of protein molecules in biochemistry, the propositional knowledge used to describe *quaternary* structure in these ERs is often inconsistent with some scholars taking one view and the other another (**C-M**). This inconsistency may affect the manner in which students go on to interpret the ERs (e.g. Mbewe, 2000).

By obtaining a student's conceptual knowledge of relevance to an ER (**C**) in Phase 1 and 2 and then comparing it to the knowledge represented by the ER (**C-M**), we can establish the student's sound/unsound knowledge prior to exposure to the ER and, whether the student

constructed any new sound/unsound knowledge (or adjusted their prior knowledge) when interpreting the ER, i.e. whether learning took place (see section 6.3.4 below).

6.3.3.7 Validation of the Conceptual-Reasoning-Mode Factor (C-R-M)

In the previous sections (6.3.3.1 - 6.3.3.6) of this chapter, the aim was to separately confirm the validity of the three factors **C**, **R** and **M** and the three interactive factors **R-M**, **R-C** and **C-M** (Fig. 6.2) that influence a student's ability to interpret an ER. The aim of the current section of the work was to obtain data that would confirm the validity of the model as an integrated whole as implied by the overlapping nature of the factors (**C-R-M**) in the Venn representation (Fig. 6.2). To test the validity of the model, data was needed that demonstrated that students were required to engage *all* factors of the model in an integrated manner in order to successfully interpret and learn from the ER. That is, we needed to confirm the *indispensable* nature of each component of the expressed model (Fig. 6.2). Our hypothesis was that interpretation of the ER required the learner to use reasoning skills (**R**) to reason with both their conceptual knowledge (**C** and **R-C**) of relevance to the ER and with the symbolism and features of the ER itself (**R-M** and **M**) to make sense of the conceptual (propositional) knowledge represented by the ER (**C-M**). Therefore, the hypothesis that we wished to test was whether students needed to engage all factors of the model in order to successfully interpret the ER.

To validate the **C-R-M** factor, we present two types of data. Firstly, in section 6.3.3.7.1 below, three examples of interview extracts, from different students and different ERs (Fig. 5.2), are used to show engagement of all factors of the model during a highly successful process of ER interpretation. Secondly, in section 6.3.3.7.2 below, data obtained from two students during the interpretation of an ER is provided to not only retest the hypothesis that a student is required to engage all factors of the model during interpretation, but to also show that the relative nature and *degree of influence* or contribution of one or more of the factors greatly affects a student's ability to correctly interpret an ER. Such data could also be used to validate the **C-R-M** factor.

6.3.3.7.1 Validation of the C-R-M factor through engagement of all factors of the model

The **C**, **M** and **C-M** factors of the model (Fig. 6.2) are implicit to the process of ER interpretation. In other words, there has to be an ER (factor **M**) available for an individual to interpret and all individuals bring a degree of conceptual knowledge (factor **C**) to the ER and, all ERs represent some type of propositional knowledge (factor **C-M**). Therefore, as shown in previous sections, **M**, **C** and **C-M** are valid factors of the model. However, as described in section 6.3.3.2, since reasoning processes can only be observed if there is something to reason with, in this case with the ER (**R-M**) and/or with students' own conceptual knowledge (**R-C**), each can be considered a subset of the overall reasoning factor (**R**), which is also a factor that affects ER interpretation. Therefore, when analysing a student quote, it is only possible to explicitly observe factors **R-M** and **R-C** *in action* during the interpretation process. Embedded within data corresponding to **R-C**, would be factors **R** and **C**. Similarly, embedded within factor **R-M** would be factors **R** and **M**. It follows that, from a student quote *alone*, the only direct observation that can be made of the engagement of factors **R**, **M** and **C** is when they are being expressed *as part* of factors **R-M** and **R-C**, respectively. Therefore, by coding a student response as **R-M** the author is validating the engagement of *both* the **R** and **M** factors. Similarly, by coding a response **R-C**, the author is validating the engagement of both the **R** and **C** factors. The validation of factors **R-M** and **R-C** and therefore, validation of the **C-R-M** factor of the model, was done by using a red colour code to identify the engagement of the **R-M** factor and a blue colour code to identify the engagement of the **R-C** factor of the model during ER interpretation. This would enable us to establish whether all factors were engaged during the interpretation process.

In view of the above rationale, the criteria for coding verbal segments of student interview extracts either as corresponding to the **R-M** or **R-C** factors was based on an analysis of the nature of the *language* contained in a student quote. For example, when expressing data corresponding to the **R-M** factor, the student used specific verbs such as "seeing" and "looking"; adjectives such as "distinct", "blob-like", "close" and "twisted"; and nouns such as "triangle", "Y-shape", "part" and "area" to reason (**R**) about the graphical features on the ER (**M**). In contrast, when expressing data corresponding to the **R-C** factor, the student linked specific words or reasoning phrases (**R**) such as "since", "therefore", "because of", "that means", "even though", "I can see it now" and "that is why" to reason with specific concepts

(**C**) like "amino acid sequence", "covalent bonds", "lock-and-key" and "antigen-antibody complex". To illustrate this approach, consider the coding of the following three extracts obtained from different students' interpretation of ER E, F and G, respectively.

Interpretation of Fig ER E:

S: ...in 'A' [ER E]... what I am seeing is a triangular shape... it is very distinct. And, the actual points of the triangle seem to be like a little blob [points to each 'blob-like' Fc region in area A].
[...]
...in this figure legend... it says Y-shaped antibody molecules... right. But, I don't see how all Y-shaped antibody molecules are going to form a triangle [points to trimer in ER E]. No, wait, I can see it! I can see it now! ...Ok, there is one antibody here [points to lower rt Ab in ER E], there is another antibody there [points to top Ab] and there is another antibody there [points to bot. lt Ab]. And, your haptens are actually here, there and there... [points correctly to three binding areas].... This would be your... basically, the tail of your 'Y' [points to Fc 'stalk of bot. rt Ab] that is forming those little blobs there, from what I see. Umm, your haptens are actually...from what it seems to me, they [haptens] seem to be shared between ...like these two antibodies here, there'd be one hapten there [points correctly within trimer]... and again here [points correctly within trimer]... there'd be one hapten between these two antibodies here [points] and one between those two antibodies [points].
[...]
I: How does the hapten stay there [in trimer arrangement] other than it being specific?
S: ...it is because of the sequences, the amino acid sequence on the hapten would be forming bonds with the... the variable region of that antigen binding site.
[...]
I: Ok, so where would a lock-and-key interaction happen here [S mentioned this earlier]?
S: ...well on both sides of the hapten. Because, if you see here, it would happen on this side and on this side [indicates with bot. hapten on trimer in ER E]. So, there'd be like two lock-and-key interactions on both sides.
[...]
S: ... because the antibodies want to bind to the hapten, they're going to have to stretch out more, to bind to it [hapten]... these antibodies have a hinge region... like a door has a hinge... it has flexibility to stretch out more because of that hinge.
I: Ok, consider if we had introduced a different type of hapten into this situation...
S: Then it is not going to be specific to the antibody. So, I don't think that you'd have the shapes forming at all [trimer and pentamer in ER E], because it won't be able to clump it together... it won't be able to from this stable... clumping structure [region A showing trimer] because it is not specific.
[...]
I: Is there anything on the visual display that is confusing?
S: Well, this is an electron micrograph, so you expect background basically... this fuzziness...I looked and I saw that is a big 'Y' [bot. rt Ab in trimer]... afterwards I saw that it has to form a triangle.

It is evident from the data presented above that, in order to successfully interpret the scientific knowledge (**C-M**) contained in ER E, the student has to engage all factors of the model. For instance, in order to successfully interpret the "triangular-shape" of the trimer (**R-M** and **M**), the student has to engage her sound conceptual knowledge (**R-C** and **C**) surrounding the nature of the lock-and-key interaction between Ab and Ag. Here, the student correctly suggests that, "there'd be like two lock-and-key interactions on both sides" of the Ab molecule. Further evidence for the above student's engagement of her conceptual knowledge

(**R-C** and **C**) upon interpretation of ER E is provided by her analogical reasoning used to suggest that, "... these antibodies have a hinge region... like a door has a hinge... it has flexibility to stretch out more because of that hinge." By contrast, the student is reasoning with the ER (**R-M**) when she makes statements such as, "...in [region]'A' [ER E]... what I am seeing is a triangular shape... it is very distinct... the actual points of the triangle seem to be like a little blob."

Interpretation of ER F:

I: ...How are the yellow 'spheres' associated to each other [ER F]?
S: ... by bonds... They would be covalent bonds.
I: Why is this group coloured yellow [points on frame a] and that coloured blue [points on frame a]?
S: Because that is specifically the light chain [points on frame a] and that's the heavy chain [points on frame a] of the fragment antigen binding region.
[...]
S: ...they [H and L chains] are close to each other, because they form the antibody molecule, so they would be bound... connected by disulfide bridges to pull them close together so that you have the variable regions close enough to form your paratope.
[...]
I: What is 'c' actually showing you... I mean the third frame [points]...
S: ...This is the antigen [points to lyso. in frame c]... the lysozyme... it shows how it fits onto that molecule [points to Fab in frame c]. So, this is the paratope [points] and that is the epitope [points]. And, this [frame c] shows the position of the molecules that facilitate that association... it seems to be in a different orientation... it's twisted [hand gesture]... at [frame] 'b' you're looking it from a length-wise angle [hand gesture] and then in [frame] 'c' you're looking at it from the top, and cutting it open... yeah, looking at the surface... basically you're looking at this section here [points to Ab-Ag interaction in frame b] in cross-section, rather than from a longitudinal section...
[...]
S: ...Basically, on this structure [ER F], you'll be representing one arm of your molecule. You have two of these [arms] on your antibody molecule, on your entire antibody molecule... you have two heavy chains and two light chains... Yeah...it [ER F] is just showing one arm...Yeah, because it is a single arm, I'm assuming you'd have papain cleavage...
I: Ok. Let's look at these red spheres here [points in frame b]. How would the situation alter, if it would alter, if we replaced the glutamine 121, with a different amino acid?
S: You wouldn't get recognition, antibody recognition, because it recognises the glutamine 121 specifically, and if it is not there, it won't recognise the next molecule. But, a different antibody might recognise the replaced [amino acids]...

In terms of the above student's interpretation of the propositional knowledge (**C-M**) represented by ER F, in order to successfully visualise the ER (**R-M**) as showing only one Fab arm of an antibody (**M**), the student is required to engage his/her conceptual knowledge (**R-C** and **C**) represented by the ER and also reason with the ER (**R-M**). For example, the student correctly reasons with the ER (**R-M**) by saying, "...Basically, on this structure [ER F], you'll be representing one arm of your molecule." And then consults with his/her conceptual knowledge (**R-C**) by stating that, "You have two of these [arms] on your antibody

molecule, on your entire antibody molecule… you have two heavy chains and two light chains…".

Interpretation of ER G:

I: Ok. Tell me about the four coloured lines in a bit more detail [ER G].
S: Well, the green one is pre-immune, that's before you've immunised the animal with a particular antigen. And, then they yellow one is at three weeks, the red one at eight weeks and the blue one is at twelve weeks.
I: Why do they use the negative 'log' here [on the x-ax.]?
S: …it's easier to represent those concentrations, because they get very small… which makes the graph easier I guess.
[…]
I: What happens to antibody concentration as we move… from left to right [indicates on x-ax.]…
S: It is decreasing…the negative 'log' increases… that means that the concentration is decreasing and you can see with your absorbance [indicates y-ax.]… the absorbance is greater and you measuring the absorbance of your antibodies. It [Ab conc.] is greater over here [points to lt of x-ax.] than down there [indicates toward rt of x-ax.].
[…]
I: …What is responsible for the absorbance?
S: It is the substrate… that is converted to product. You would add a detector antibody to your plate and that will bind to your antigen-antibody complex. Then, you add substrate and that will be converted to product if your detection antibody bound to that antigen-antibody complex. And, that will only happen if you have got your antibody that you're looking for.
[…]
I: Could you compare [points] 'P' and 'Q' [ER G].
S: 'P' seems to have a lower absorption than 'Q', even though the concentrations of the antibody at 'P' is greater than that at 'Q'… and that is just basically because there is too much antibody present to bind to all the antigen, in the well… there is a number of things like steric hindrance… that prevented those antibodies from binding as well. So, in the next washing step you'll wash off some antibodies, that is why it looks like there is less [Ab concentration].

In order to successfully interpret the scientific knowledge (**C-M**) depicted in ER G, the student in the above quotation engages sound conceptual knowledge (**R-C** and **C**) to reason with the graphical features (**R-M** and **M**) of the ER. In this case, the integration of all factors of the model allows the student to suggest that, "P seems to have a lower absorption than Q (engagement of **R-M**), even though the concentrations of the antibody at P is greater than that at Q… and that is just basically because there is too much antibody present to bind to all the antigen, in the well…(engagement of **R-C**)".

Thus it is evident from the above three student extracts that, at some time or other, a student is required to engage and integrate all factors of the model in order to successfully interpret an ER. By coding the engagement of factors **R-M** and **R-C** within student quotes, the data above demonstrates the indispensable nature of each factor of the model for sound interpretation of an ER and as a result, serves as the first validation of the **C-R-M** factor. In

addition, it is noteworthy that the two cognitive processes corresponding to **R-M** and **R-C** are not engaged in any specific sequence (i.e. **R-M** first then **R-C** or vice-versa). Instead, students continually switch back and forth between reasoning with the ER (**R-M**) and with their conceptual knowledge (**R-C**) during the process of interpretation.

6.3.3.7.2 Validation of the C-R-M factor through the relative degree and nature of influence of one or more of the factors of the model

In addition to the data presented in section 6.3.3.7.1 above, data obtained from two students during interpretation of an ER further validated the **C-R-M** factor. Not only does the data presented in this section support the need for a student to engage all the factors of the model to interpret an ER, but also illustrates how the *degree* and *nature of influence* or contribution of one or more of the factors plays a major role in determining a student's overall ability to correctly interpret an ER (**C-R-M**). For example, poor interpretation of an ER might result from either; failure of the student to adequately engage conceptual knowledge (low degree of contribution from **R-C**) or, conceptual knowledge fraught with misconceptions, might be adequately engaged (**R-C**).

Factor **M** makes a constant contribution to interpretation because the ER and its graphical features do not change during interpretation. In other words, the ER is not altered during interpretation. This is of course only true for static ERs and not for animations, which is why the latter are more complex and cognitively demanding for students (see Chapter 2). Factor **C-M** also does not change during ER interpretation, but might change during the course of time as part of the progress of science wherein there is an adjustment or modification of the propositional knowledge represented by the ER. Factor **C** might change in a limited way depending on whether student knowledge is unaffected by interpretation of the ER or whether learning takes place or alternative conceptions develop. Thus in the case of factors **M**, **C-M** and **C**, their contributions for all intents and purposes remain constant during the process of interpretation although the quality of the ER, the soundness of the propositional knowledge and the student's prior knowledge, respectively, will still affect overall interpretation. On the other hand, as already demonstrated in section 6.3.3.7.1, the relative contribution of factors **R-M** and **R-C** during ER interpretation can fluctuate dramatically during interpretation depending on whether the student is consulting with the ER (**R-M**) or their conceptual knowledge (**R-C**). Therefore, the researcher can analyse the colour-coded quotes

corresponding to factors **R-M** and **R-C** to show how each factor makes a variable contribution during interpretation of an ER. As for section 6.3.3.7.1, the same colour codes are used to illustrate engagement of the **R-M** and **R-C** factors of the model.

The first example, coded **Q1**, shows how a student's prior conceptual knowledge (**C**), even if it is excellent, may still lead to the unsuccessful interpretation of an ER. In this case one student's prior conceptual understanding (**C**) about general antibody structure and primary interaction with antigen binding, before exposure to any ER, was shown to be rich and extensive. Additionally, the student's reasoning with these concepts (**R-C**) was shown to be consistently excellent. For instance, consider the following extract obtained from the student, during Phase 1 of 3P-SIT, before exposure to any ER:

> S: ...both antigen and antibody are proteins... antibody structure varies according to the type of antibody... they vary in sub-classes and classes with the respective chains that make them up... the interaction with the antigen... is through the variable regions on the heavy and light chains of the antibody. They react with the epitopes of the antigen. The antibody has to be specific to the epitope found on the antigen ...It has to fit properly, otherwise it won't bind. So, it actually has to be compatible ...the interaction is actually on the antibody with the variable regions, rather than the constant regions, because those constant regions are found on most antibodies... that is why they're called 'constant'... whereas the variable regions change... are variable because they're specific to an antigen's epitope.

After being exposed to ER F however, it was found that the student did not reason with the ER appropriately and thought that the ER was showing a complete Y-shaped antibody instead of a single Fab arm. This reasoning was demonstrated by the following quote:

> I: In terms of structure, what is being shown on this representation [ER F]?
> S: ...you can see the antibody structure... one can see that is consists of the two chains [H and L]... it is actually two heavy chains [points to bot. two 'groups' of blue spheres simultaneously] and two light chains [points to top two 'groups' of yellow spheres simultaneously].

It is evident from the quote above that when reasoning with the ER (**R-M**) the student erroneously thought that ER F (**M** and **C-M**) represented a complete antibody. Even though this data validates the **C-R-M** factor of the model by showing that ER interpretation requires all factors of the model to be engaged, the student did not reason soundly with the ER and thus the **R-M** factor was adversely influencing interpretation. In this case, as is evident in the quote above, the nature of the ER (factor **M**), i.e. the spatial arrangement of the graphical markings, influenced the student to incorrectly reason (**R-M**) that ER F represented an entire intact antibody. This is despite the fact that the student's prior conceptual understanding (**C**)

was shown to be outstanding. Therefore, factors **M** and **R-M** had a large degree of influence on the student's ability to successfully interpret the ER (**C-R-M**).

The second example, coded **Q2**, shows how a student's poor prior conceptual knowledge (**C**) may lead to a poor interpretation of an ER. In this case, the following student's prior conceptual understanding pertaining to antibody structure and interaction with antigen (**C**), before exposure to any ER, was found to be not as extensive and sound as the first student's (**Q1**) knowledge above. In addition, the student's prior knowledge showed a strong reliance on the application of the lock-and-key analogy to describe Ab-Ag binding when reasoning about these concepts (**R-C**). For instance, consider the following quote, obtained from the student during Phase 1 before exposure to any ER:

> I: What is it about antibody structure that allows it to form a lock and key with the antigen [S stated this earlier]?
> S: Well, it is the light chains of the antibody, which has got the 'V' part. Ok, you get the heavy chain which is the 'stalk' and then you get the 'V' on top of the 'stalk'... and, the light chains are the 'V' part... that region ('V') is the area that they [Ag] bind to.
> I: At what area specifically, do they [Ag] bind to?
> S: ...specifically to the variable site... in order for specificity to come into it... yeah, that region there [gesture]... the 'V' part, the whole 'V' part...that is the main area that they bind to.

In addition to expressing the lock-and-key analogy strongly, it is evident in the quote above that the student showed a misconception (**C**) by stating that the entire "V" part of the antibody is representative of the antigen binding site, instead of two separate binding domains. Upon exposure to ER E, the same student carried this misconception over by misinterpreting the trimer arrangement depicted by the micrograph as representing a single antigen (hapten) inside the trimer, even though this was not succinctly conveyed by the ER (**M**). The following SGD and accompanying verbal output generated by the student demonstrates this misinterpretation:

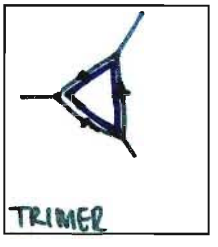


Figure 6.11 Student-generated diagram obtained from the interpretation of the trimer arrangement on ER E

S: ...I can see the triangle there [points on ER E] and the Y-shaped antibodies, you can actually see them...forming a trimer. And, they're very light, that area where the antibody is, is a very light area ... in the middle of the trimer it is dark ...that is where the hapten is, where the antibody is binding onto it...
[...]
S: ...[gen. Fig. 6.11]...this [Ab] binds with a complementary fit to that [V edge of hapten]. All these [3 V edges of hapten] have to somehow fit into these antibody binding sites, the 'V' shape in order to be... like a lock and key mechanism, it [Ab] has to fit into this thing [hapten], so the shape has to be similar. Yeah, and the antibody just binds onto that [V edge of hapten], that shaped area...

Although the above data validates the **C-R-M** factor of the model by showing that the student is required to engage all the factors of the model to interpret ER E, it is clear that the student's interpretation of the ER was based on an over reliance on reasoning (**R-C**) with very ingrained ideas (**C**) such as the lock-and-key analogy. This resulted in the student erroneously suggesting that a single hapten (antigen) could bind within all three "V-clefts" of the three antibodies constituting the trimer on ER E (Fig. 6.11). In this case, the student's reasoning processes corresponding to factor **R-C** and his/her conceptual knowledge (**C**) were most limiting and therefore, these factors had a major influence on the student's ability to interpret the ER (**C-R-M**).

In summary, a student's overall ability to interpret, visualise and learn from an ER depends on both the engagement of all the factors represented by the model (**C-R-M**) (Fig. 6.2) and the nature of the contribution of each factor in terms of whether, for example, the student uses scientifically sound or unsound conceptual knowledge and reasoning, whether the ER represents sound or unsound propositional knowledge and/or, whether the ER is graphically misleading or appropriate. Thus each of the six factors (Fig. 6.2), corresponding to **C-R-M,** represent key and indispensable components of the model. In addition, since some of the data representative of a particular factor contained evidence for one or more of the influence of other factors at the same time, it is often impossible to totally resolve the influence of only one factor alone. This supports the use of Venn logic for conceptualising the nature of the model in that it is still possible to show the influence of one particular factor at one time, even though there may be evidence for the simultaneous influence of another factor. This in itself validates and provides good evidence for the integrated nature (**C-R-M**) of the model.

6.3.4 Uses and applications of the expressed model

Sections 6.3.1 to 6.3.3 discussed the development and validation of the expressed model. Now that empirical data, confirming the operational definitions (Table 6.1) of the model have been provided, the practical application of the model (as per research question 6) needs to be considered. The following seven uses of the expressed model (Fig. 6.2) were identified:

1. The model can be used to establish whether a student's overall *interpretation* of an ER was successful or not as per factor **C-R-M**. This can be done, by comparing the student's "post" knowledge after exposure to an ER (Phase 2) with the conceptual knowledge represented by the ER (**C-M**). For example, with respect to **Q1** and **Q2** (section 6.3.3.7.2), when data from Phase 2, after interpretation of the ER corresponding to factor **C**, was contrasted with the propositional knowledge corresponding to **C-M**, it was evident that both students had misinterpreted ER E and F, respectively.

2. In relation to (1.), the model can also be used to determine which of the six factors (Table 6.1) *positively or negatively* influence a student's interpretation of a particular ER the most and, which the least. As demonstrated by the **Q1** and **Q2** data in section 6.3.3.7.2, the nature and *relative contribution* of a particular factor can be measured for a particular student interpreting a specific ER at a particular time. For example, for **Q1**, it was suggested that factor **R-M** had the most negative influence while for **Q2**, factors **C** and **R-C** had the most negative influence on the student's ability to successfully interpret the ER. The relative contribution of each factor towards ER interpretation will be different for different students, ERs and scientific contexts. For instance, an individual may bring insufficient or poor conceptual knowledge to an ER (**C**). As a result, the student may depend largely on the interaction represented by **R-M** to make sense of the ER, in an attempt to reach some type of understanding. Conversely, a student with a rich and scientifically sound conceptual understanding of a scientific phenomenon, may rely less heavily on **R-M**, and depend more on reasoning with already existing concepts (**R-C**) to try and understand the ER. Alternatively, since some ERs of a scientific phenomenon are not always meaningful or scientifically accurate representations of the idea they convey (**C-M**), it is possible that a student may have an excellent conceptual understanding (**C**) and reasoning skills (**R-M** and **R-C**), but might still interpret the ER in an unsuccessful manner. The results presented in this

chapter show that this degree of 'weightedness' during interpretation of an ER is measurable, albeit in qualitative terms. The ultimate aim, of course, is to ensure that students successfully engage all factors in order to optimise interpretation of the ER (**C-R-M**) (section 6.3.3.7).

3. The model can also be used to establish whether sound or unsound *learning* has occurred as a result of a student's interpretation of an ER. To establish whether learning has occurred from the ER, the student's "post" knowledge (**C**) obtained after exposure to and interpretation of an ER (after completion of Phase 2 of an interview) is compared with data corresponding to their prior knowledge (**C**) obtained during Phase 1 of 3P-SIT. Through this comparison, the researcher can establish whether the student has altered, or added to, their conceptual knowledge (**C**) after being exposed to an ER, to establish whether learning has occurred. In this regard, it is possible for a student to interpret an ER perfectly (see 1. above) but not learn anything new. In addition, it is also possible for the researcher to measure whether a student improved their knowledge and understanding of the concepts represented by the ER or developed any new alternative conceptions that were not diagnosed in Phase 1. Furthermore, it is also possible to measure any conceptual change that a student may have undergone by comparing misconceptions identified in Phase 1 with any sound knowledge that may have been constructed after exposure to an ER.

4. With reference to points 1, 2 and 3 above, the expressed model therefore serves as a general *diagnostic* framework that can guide practitioners' and researchers' discussion, thinking, identification and data analysis relating to the nature of a student's difficulty with an ER. That is, whether the student has a conceptual (**C**) or reasoning (**R-M** or **R-C**) difficulty or, whether the difficulty lies with the nature of the graphical features of the ER (**M**). With respect to a conceptual difficulty, the model can assist us to determine the degree in which a student's conceptual understanding is lacking or erroneous, as well as to determine the nature of any alternative knowledge. With respect to a reasoning difficulty, the model can assist us to define the nature of the reasoning difficulty. For instance, the model can help define what particular cognitive process may be the cause of such a reasoning difficulty (Table 6.1). With respect to a difficulty resulting from the nature of the ER, the model can assist us to determine what particular ER graphical markings or symbolism are responsible for inducing either inappropriate reasoning or alternative conceptions. This guiding role of the model has proved invaluable in facilitating discussion in the Science Education Research Group (SERG) at the University of KwaZulu-Natal, South Africa.

5. The model enables the prediction of the *potential source(s)* of difficulties with ER interpretation. This is because the interactive factors **R-M**, **R-C** and **C-M** frame our thinking about a student difficulty as to the combination of which two factors (**C**, **R** or **M**) play the most influential role during ER interpretation and therefore, what the source of the problem might be. For instance, data might reveal that one source of a particular problem was a student's surface level interpretation of the ER, which would correspond to **R-M**. Another example of a source of difficulty could be a student's inappropriate transfer of their conceptual knowledge from one domain to another when interpreting an ER, which would correspond to **R-C**. Lastly, an example of a source of difficulty corresponding to **C-M** could be misleading symbolism and graphical features used to represent the scientific propositional knowledge.

6. Since the model informs potential sources of a student's difficulty with the interpretation of an ER (5.), we can use this knowledge together with that of the nature of the difficulty to design and develop *approaches* to teaching and learning including intervention strategies for improving the student's interpretation of and learning from ERs. For example, with regard to **Q1** (section 6.3.3.7.2), possible interventions for improving this student's interpretation of ER F could include the following. The design and presentation of ER F as a means for portraying conceptual understanding (**C-M**), could be scrutinised, and student understanding of the nature of the graphical symbolism used in these ERs (**M**) and how best to decode them (**R-M**), could be facilitated. Regarding example **Q2** (section 6.3.3.7.2), intervention could include explicitly facilitating student learning and understanding of the sound propositional knowledge pertaining to Ab-Ag binding, necessary for interpreting an ER such as ER E. In terms of proposing intervention strategies in general, if all factors were found to be successfully engaged except the **M** and/or **R-M** factors, intervention strategies could include reconsidering the design and nature of the graphical presentation/representation of the ER, providing the student with insight into the nature of the graphical symbolism used in the ER, as well as "teaching" the student how best to decode the symbolism. Alternatively, if all factors were found to be successfully engaged except the **C** and/or **R-C** factors, one intervention strategy could include supplying the student with the sound propositional scientific knowledge necessary for interpreting the ER. Further details pertaining to remediation strategies with respect to teaching and learning with ERs are dealt with in Chapter 7.

7. The model has a generic application to *all types* of ERs including not only static representations but also dynamic, animated and multimedia representations (see sections 2.6.6 and 2.6.7). Such applications of the model will be the target of future research.

6.4 Summary and Conclusions

The Justi and Gilbert (2002) modelling process was used to develop and express a model of factors determining a student's ability to interpret a scientific ER. Empirical data corresponding to each of the seven factors, constituting the expressed model were gathered with a specially designed clinical interviewing method, termed 3P-SIT (Chapter 5). Data generated from 3P-SIT was analysed by a qualitative and iterative method to illustrate the importance and validity of each factor comprising the expressed model (Fig. 6.2). In so doing, each factor constituting the model was validated and defined as making an indispensable contribution to a student's ability to interpret an ER. As a result, the researcher could generate specific operational definitions (Table 6.1) that represent the meaning and nature of each factor of the model.

In order for the model to be representative of a student's ability to successfully interpret, visualise and learn from the ER, as implied by the Venn logic used to depict it, empirical results were required with which to validate the **C-R-M** factor of the model. This was carried out by first showing that engagement of all six factors of the model was essential for ER interpretation and secondly, that the nature and relative degree of influence of one or more of the factors of the model plays a major role in the success of any interpretation. Lastly, the chapter has also demonstrated how the model can be used and applied in a wide range of educational and/or research settings. In particular, the model can be used as a framework with which to establish firstly, whether ER interpretation was successful or not and secondly, whether learning from the ER was sound or unsound and thirdly, to identify which factor(s) of the model play the most influential role during interpretation.

7 GENERAL DISCUSSION AND IMPLICATIONS

The three studies reported in Chapters 4-6 respectively, addressed the following five research questions, which were posed in Chapter 1:

1. What types of difficulties do students have with ERs used in the teaching and learning of biochemistry?
2. What are the sources of such difficulties and, therefore, what are the factors affecting students' ability to interpret ERs?
3. How might we obtain empirical data to further investigate the nature of the factors affecting students' ability to interpret ERs?
4. Can the factors be incorporated into an appropriate model?
5. How might we obtain empirical data to confirm the validity of the model?
6. What practical applications will the model have and will it be generalisable to all ERs in biochemistry and science?
7. What guidelines can be suggested for teaching and learning with ERs?
8. What guidelines can be suggested for improving ER design?

In response to question 1 above, the work reported in Chapter 4 successfully identified three general categories and seventeen sub-categories (Tables 4.2, 4.3 and 4.4), of student difficulties with the interpretation of three textbook ERs (Fig. 4.1 A, B, C and D), depicting antibody structure and interaction with antigen. The three general categories included the process-type (**P**), structural-type (**S**) and DNA-related (**D**) difficulties. Thirteen of the seventeen sub-categories of difficulties were classified on the Grayson *et al.* (2001) research framework (Fig. 3.2) at Level-3 (partially established), one was classified at Level-2 (suspected) and three were classified at Level-1 as unanticipated. Thus although we feel confident about the nature of the identified difficulties, further research is required, in multiple contexts with a broader range of ERs in order to fully establish the nature of the difficulties at Level-4. For the process-type difficulty, incidences ranged from 7 to 70%, for

the structural-type difficulty from 3 to 70% and, for the DNA-related difficulty from 4 to 19% across the student populations and across all three ERs. This wide range of incidences was mainly due to differences between ERs, between second and third-year samples, and between the nature of the probes administered to the participants. Free response probes, for instance, give a minimum incidence because not all students revealed their difficulties, whereas more specific probes give higher incidences because they focused more specifically on a difficulty.

Clarification of the nature of the student difficulties enabled us to start addressing question 2 by suggesting possible sources of the difficulties. In so doing, three major categories of difficulty sources were identified. These included: the *nature of the ER* and its graphical features, students' *reasoning processes* and, the nature and extent of students' *conceptual knowledge*. This in turn informed the identification of at least three factors that could play a major role in students' ability to interpret ERs in biochemistry. The three factors are: students' ability to *reason* with the ER and with their own conceptual knowledge, students' understanding (or lack thereof) of the *concepts* of relevance to the ER, and the *mode* in which the desired phenomenon is represented in the ER. During analysis of the data supporting these factors (see Chapter 4), the author observed that it was difficult to pinpoint the overt effect of only one factor alone on students' interpretations. That is, there was a measure of interdependence of the factors on each other across all categories of difficulty and across all three ERs. For example, it was found that reasoning ability was often dependent on the nature of the ER that was being "reasoned with". Coupled to this was the fact that it was difficult to establish to what degree each of the factors (or combinations of them), positively or negatively, influenced ER interpretation. Stated differently, it was uncertain which of either; students' conceptual knowledge of relevance to the ER (e.g. Ametller and Pintó, 2002; Cheng *et al.*, 2001); the role of the visual markings themselves (e.g. Lowe, 1993a); or, the role of students' employed reasoning processes (e.g. Cox and Brna, 1995) played the most influential role during the interpretation of a certain ER.

In order to try to resolve and further investigate the nature of each of the above factors, as well as their above-mentioned interdependence, a research instrument was needed to generate empirical data pertinent to each factor. Thus in response to research question 3, the study reported in Chapter 5 was concerned with the design and testing of a *three-phase single interview technique* (3P-SIT) that could be used to obtain empirical data corresponding to each of the three factors so that they could be confirmed as factors that affect students' ability

to interpret ERs. At this stage, the factors were coded **C** for the conceptual factor, **R** for the reasoning factor and **M** for the representation mode factor. In addition to confirming the existence of the three factors, data obtained from the pilot study generated further evidence of the relative *influence* of one factor upon another although, at this stage, the author was unsure of the extent and nature of this influence. The findings from this study led to a decision to implement 3P-SIT in the more in depth study reported in chapter 6 in which the instrument would be used to further investigate the nature of each factor and the nature in which the factors influence one-another upon students' interpretation of an ER.

In Chapter 6 research question 4 was addressed by employing the Justi and Gilbert (2002) modelling process to develop a model of the factors determining a student's ability to interpret a scientific ER. This led to the identification of seven factors (i.e. a further four) influencing students' ability to interpret three ERs (Fig. 5.2 E, F and G) of antibody-antigen interaction. The seven factors that comprise the model are the conceptual (**C**), reasoning (**R**), reasoning-mode (**R-M**), reasoning-conceptual (**R-C**), representation mode (**M**), conceptual-mode (**C-M**) and conceptual-reasoning-mode (**C-R-M**) factors. In response to research question 5 each factor of the model was validated using 3P-SIT to generate empirical data corresponding to each factor. Validation of the interactive factors confirmed that the seven factors were appropriately represented by Venn logic. However, if the model was to be at all representative of a student's ability to successfully interpret, visualise and learn from the ER, as implied by the Venn logic used to depict it (Fig. 6.2), empirical results were required to validate the conceptual-reasoning-mode (**C-R-M**) factor of the model. In this respect, validation of the **C-R-M** factor was carried out through two avenues. Firstly, validation of the **C-R-M** factor was demonstrated by providing data that showed the *indispensable* nature of all six aforementioned factors of the model by demonstrating that a student is required to engage and integrate all factors of the model in order to successfully interpret an ER (see section 6.3.3.7.1). Secondly, the **C-R-M** factor was validated by providing data that showed the relative degree of influence of one or more of the factors of the model during students interpretation of an ER (see section 6.3.3.7.2). Through the expression of the model and empirical validation of its constituent factors (see sections 6.3.3.1 - 6.3.3.7), we were able to construct and formalise operational definitions for each of the factors comprising the model (see Table 6.1). This in turn, allowed us to address research question 6 by developing at least seven practical applications of the model (section 6.3.4). These include:

a. The model can be used to establish whether a student's overall *interpretation* of an ER is successful or not.
b. The model can be used to determine which of the six factors *positively or negatively* influence a student's interpretation of a particular ER the most and, which the least.
c. The model can be used to establish whether sound or unsound *learning* has occurred as a result of a student's interpretation of an ER.
d. The model serves as a *diagnostic* framework that can guide researchers' and practitioners' discussion and thinking relating to the nature of a student's difficulty with ER interpretation. That is, whether the student has a conceptual (**C**) or reasoning difficulty (**R**) or, whether the difficulty lies with the nature of the graphical features of the ER (**M**).
e. The model enables the prediction of the *potential source(s)* of difficulties with ER interpretation. This is because interactive factors **R-M**, **R-C** and **C-M** frame our thinking about a student difficulty as to the combination of which two factors (**C**, **R** or **M**) play the most influential role during ER interpretation and therefore, what the source of the problem might be.
f. Since the model informs potential sources of a student's difficulty with the interpretation of an ER, we can use this knowledge, together with that of the nature of the difficulty, to design and develop *approaches* to teaching and learning including intervention strategies for improving the student's interpretation of and learning from ERs. This application of the model will be discussed below.
g. Based on the nature of the model and the operational definitions of its constituent factors, the model has a generic application to *all types* of ERs in science including not only static representations but also dynamic, animated and multimedia representations. Such application of the model for teaching and learning with the latter ERs could be the target of future research.

The advantages and limitations of using the model for the purposes described above are as follows. In terms of the advantages, once student difficulties with the interpretation of ERs in biochemistry have been identified by using a rigorous categorisation framework (e.g. Grayson *et al.*, 2001), the expressed model can act as a powerful frame of reference in the identification process and for guiding our thinking on the nature of the identified difficulties. In this regard, the model by virtue of its seven component factors helps inform the process of identification of difficulties (including probe design and data analysis), the clarification of

their nature, the prediction of possible sources of the difficulties, and the design of guidelines for teaching and learning with ERs including the prevention and remediation of difficulties. In other words, the model can be applied to guide our thinking about whether the nature of a difficulty is due to the influence of the conceptual (**C**), reasoning (**R**) or representation mode (**M**) factors, or a combination thereof (factors **R-M**, **R-C** or **C-M**). Hence, an advantage of applying the model in this manner is that the process is not limited to students' interpretation of ERs in biochemistry alone, but can be applied generally to students' interpretation of ERs in any scientific context. This is because, to successfully interpret, or learn from *any* ER in science, a student is required to posses the necessary scientific conceptual knowledge of relevance to that ER and, is required to possess the reasoning skills necessary to reason not only with their conceptual knowledge but to also reason with that ER. Thus a great advantage of the model is that it has potential generic application across all disciplines of science and is very powerful due to its simplicity.

Like any other useful model, a disadvantage (or advantage depending which way you look at it) of the current expressed model is that it provides only a restricted representation of the phenomenon that it aims to depict. The model is only a limited representation of the factors affecting students' interpretation of ERs since there are several other factors that could also influence ER interpretation in science. For instance, ER interpretation may also be affected by the social context from where the data was drawn; psychosocial factors such as cultural and gender dispositions; psychological factors such as students' past experiences, personality traits, value systems, confidence levels, motivation levels (e.g. Wheeler and Hill, 1990) and attitudes towards ER interpretation (e.g. Sumfleth and Telgenbüscher, 2001); and language competence. In this regard however, since the very nature of human-as-instrument studies (see Chapter 3) makes it difficult to control all possible influencing variables, the data obtained in the thesis has nevertheless provided a valid and reliable account of the role of at least seven integral factors in students' interpretation of ERs in science. In addition, by generating only qualitative data to develop and validate the model may have been an incomplete empirical account of its nature. Future work could be concerned with validating the model through quantitative means, which would lend itself to statistical analyses of the data pertaining to the isolated factors.

From the above applications of the model it is clear that the majority of applications would require specialised knowledge, research expertise and further research before teachers and

learners could benefit more directly from them. Towards achieving this goal, it is appropriate at this stage to consider the general pedagogical implications of the model for *improving* the use of ERs in the learning and teaching of biochemistry and, *science in general.* This will include addressing our final two research questions 7 and 8. An outline of the pedagogical implications of the model is represented in Fig. 7.1 below. As described pictorially in Fig. 7.1, the nature of a student difficulty and the source of that difficulty *both* inform the design of strategies to remediate or prevent the difficulty. As part of this process, the author argues therefore, that the model can be used to frame teachers' and researchers' thinking about not only the nature of a difficulty but also the source of the difficulty (Fig. 7.1). This guiding role of the model has proved invaluable in facilitating discussion in the Science Education Research Group (SERG) at the University of KwaZulu-Natal, South Africa, where teachers and science education researchers have used the model to frame their thinking of students' difficulties across a variety of scientific contexts as well as across different types of ERs (Fig. 7.1).

Applying the model to guide teachers' and researchers' thinking about the nature of student difficulties and sources of the difficulties enables the proposal of strategies and guidelines for improving the teaching and learning with ERs and for preventing and remediating difficulties (Fig. 7.1). In response to research questions 7 and 8, both the findings of this thesis and other relevant literature have informed the proposal of such guidelines and are presented in Tables 7.1 - 7.5 below. The guidelines are discussed with respect to each of the six factors constituting the model (Fig. 6.2). In this regard, since the six factors affect students' ability to interpret ERs, it makes good sense that any teaching and learning remediation strategies should be designed to explicitly address each factor. Hence, in the discussion below, strategies relating to each factor of the model are addressed one at a time. Strategies for improving *teaching and learning* with scientific ERs (research question 7) have emanated from considering the role of the **C**, **R**, **R-M** and **R-C** factors of the model. In the commentary below, we have fused teaching strategies with learning strategies. This is because good learning approaches are often good teaching approaches and vice versa, which makes it rather illogical to distinguish between them. Guidelines for improving the *design* of scientific ERs (research question 8) have emanated from considering the role of the **M** and **C-M** factors of the expressed model.

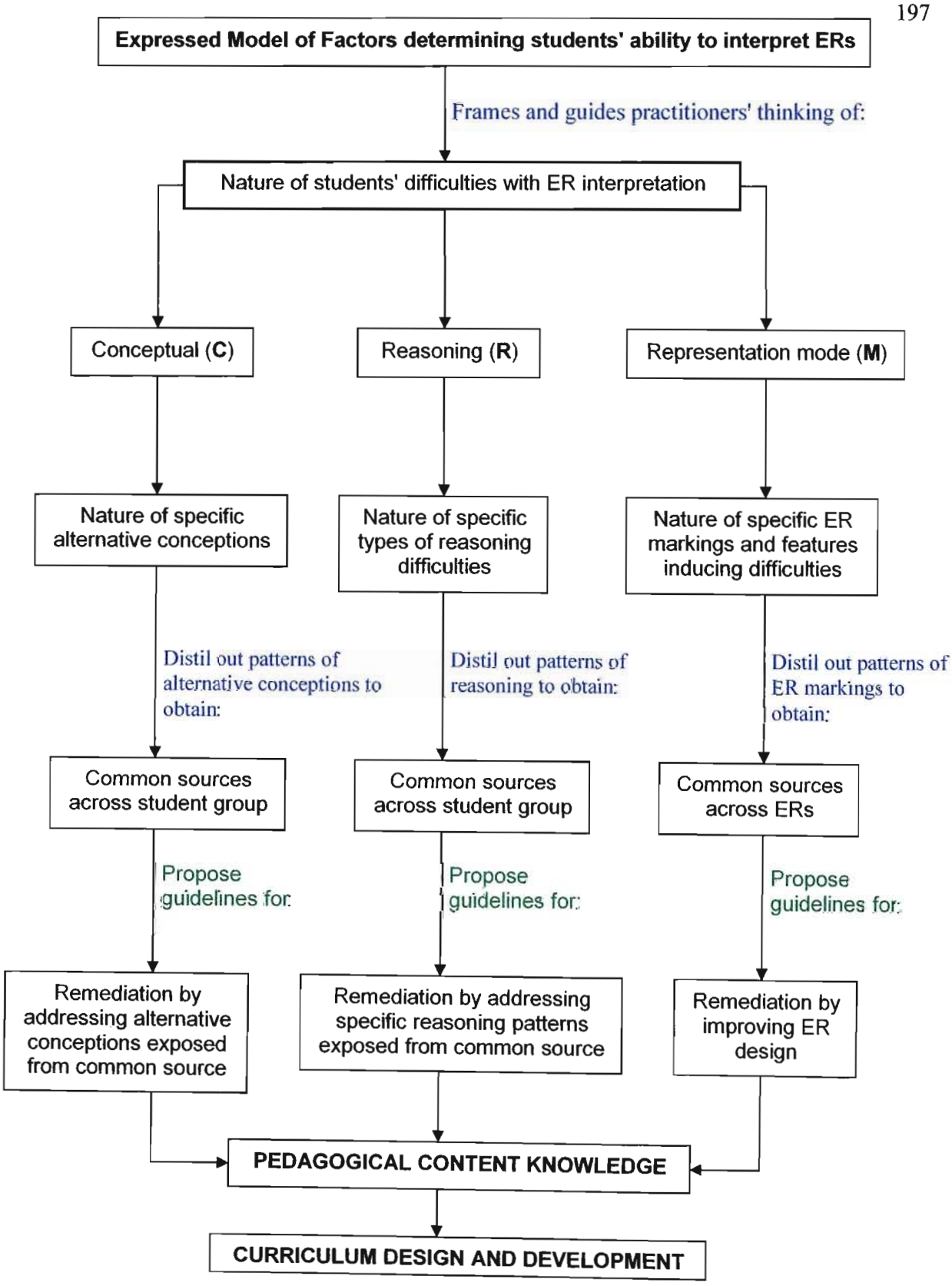


Figure 7.1 Use of the model for improving learning and teaching with ERs in science

During the process of formulating strategies to improve learning and teaching with ERs and ER design, it is important to acknowledge the importance of pedagogical content knowledge (PCK) (Fig. 7.1). PCK is that pedagogical knowledge which *includes* knowledge of how to teach different topics, concepts and phenomena. It includes taking cognisance of the nature of relevant difficulties and their possible sources (e.g. de Jong, 1997; Shulman, 1986) when designing teaching and learning activities and course curricula. In addition, PCK not only includes the knowledge of students' misconceptions but also includes the knowledge that teachers themselves may also possess such as alternative conceptions not consistent with scientific worldviews (e.g. de Jong, 1997). Therefore, each alternative conception or reasoning difficulty that a student possesses might require a different teaching approach depending on the nature of the difficulty typically encountered by students. It is thus the opinion of science education researchers (e.g. Grayson *et al.*, 2001) that acquiring PCK can lead to effective teaching and learning and as a result, effective curriculum design (Fig. 7.1). It is pivotal therefore, that teachers, ER designers and curriculum developers acknowledge the importance of PCK during the teaching and learning with ERs and during the remediation or prevention of ER-related difficulties (Fig. 7.1). In the sections below, ideas of strategies for improving teaching and learning with ERs and ER design, that have emanated from the findings presented in this thesis, and those that the author has deduced based on his own deductive reasoning, are marked with * in the second column of Tables 7.1 - 7.5. In addition, in cases where applicable guidelines have emerged from studies reported in the literature, the respective authors have been cited in the same column in the Tables below. The latter guidelines from the literature arose out of thinking about the results conveyed in this thesis but weren't substantiated in the thesis by research and could therefore, be the target of future research.

*Guidelines drawn from the Conceptual (**C**) Factor.* Various strategies can be used to develop students' conceptual knowledge of relevance to an ER as well as to remediate or prevent *alternative conceptions* or *conceptual difficulties* that affect their ability to interpret and learn from a particular scientific ER, or set of ERs. Effective guidelines may include those listed in Table 7.1 below. Strategies drawn from the conceptual factor (**C**) include making the conceptual knowledge contained within the ER explicit (point 1, Table 7.1). In this regard, in the present study we made it explicit to students what specific part of the immune reaction was being covered when teaching about IgG structure and interaction with antigen, including

Table 7.1 Guidelines for improving teaching and learning with ERs based on the Conceptual (**C**) Factor of the model

General Guideline Category	Specific Guidelines	Ideas obtained from present study (*) and/or literature
1. Making conceptual knowledge explicit.	a) Teachers should make explicit to students *what* specific conceptual knowledge shall (or is) be(ing) covered during instruction.	*
	b) Placing the conceptual knowledge being taught in an overall and meaningful context. .	*
	c) Explaining and clarifying to students what particular conceptual knowledge the ER *is*, and is *not* representing. Teachers should endeavour to expend great effort when explaining the conceptual understanding implied by an ER.	* Lowe (2003, 1989); Stylianidou *et al.* (2002)
	d) Instructors should make the specific learning outcomes of the ER clear to students.	* Henderson (1999)
	e) Instructors should endeavour to use analogies when representing the conceptual knowledge represented by an ER.	Rigney and Lutz (1976)
2. Ensuring knowledge of conventions.	a) Instructors should actively question students about the ER and the conventions and symbolism that the ER utilises to denote conceptual knowledge.	* (section 5.4.2) Henderson (1999); Kosslyn (1985)
	b) Teachers should ensure that students' acquire and/or possess the knowledge of the conventions implied by scientific ERs.	* Lowe (1991)
	c) Explicitly "teaching the ER" to students will promote learners to gain an adequate and representative conceptual structure when required to construct new conceptions. By explicitly teaching pictorial conventions to students, whenever learners are exposed to them, helps students acquire the necessary conceptual knowledge.	* Henderson (1999); Constable *et al.* (1988); Lowe (1987)
3. Using learner-generated graphic organisers and other ERs.	a) Students' production of concept maps and flowcharts help learners' structure, organise and compare *concepts* graphically.	Lumer *et al.* (2003); Britton and Wandersee (1997); Winn (1991); Alesandrini (1984)
	b) By generating their own ERs, students' are stimulated to become more metacognitive thinkers, which allows for a deeper thinking about conceptual understanding that is abstract, the construction of more meaningful knowledge structures and the remediation of conceptual difficulties.	Ward and Wandersee (2002)
	c) Learners should be stimulated to generate their own ERs of a concept before instructors commence with using textbook or computer ERs prescribed to the concept. After observing students' externalisations, the instructor is in a position to understand potential alternative conceptions that a student may bring to an ER or to a course.	Gobert and Clement (1999); Glynn (1997)
	d) Instructors should demonstrate to students how they themselves "draw their own mental models" so students can observe the process. While doing so, teachers should provide detailed explanations of what concepts they are expressing and should ensure that they draw clearly.	Glynn (1997); Maddox and Loughran (1977)
4. Applying the dual-coding approach.	Instructors should adopt the tenets of dual-coding when teaching with ERs. In this way, students have more than one system (graphical and verbal) for integrating knowledge, which allows for a more meaningful conceptual structure.	Mayer (2003); Paivio (1986)

what was pivotal for preventing alternative conceptions. We considered it is also necessary to place the role of IgG structure and function in the context of the overall immune system with, for example, the use of an overview flow diagram. In addition, the findings of this thesis suggest that students should be made aware of the learning outcomes of the ERs that are to be

used in course notes and other educational materials. Following on from this, the results of this thesis suggest, that it is importance to ensure that students have a suitable *knowledge* of the conventions contained within the ERs (point 2, Table 7.1), by explicitly "teaching" the ER to students, especially when engaging with highly abstract ERs. Furthermore, stimulating learners to generate their own ERs (point 3, Table 7.1) can also improve the conceptual knowledge represented by ERs, while adopting a dual-coding approach (point 4, Table 7.1) to teaching (see sections 2.4 and 2.6.7) will provide learners with rich mental models of the scientific phenomenon represented by the ER and as a result, make their conceptual knowledge structures more integrated.

*Guidelines drawn from the Reasoning (**R**) factor.* As previously discussed, the **R** factor is representative of those *cognitive processes* whereby students reason with the ER (**R-M**) and/or, reason with their conceptual knowledge (**R-C**) in order to interpret a scientific ER. Thus, a student's reasoning ability (**R**) for interpreting the ER includes both sound reasoning and any reasoning difficulties. As shown by the findings of this thesis (see sections 6.3.3.3 and 6.3.3.4), these reasoning mechanisms acquire meaning only when they are observed in *action*, either by students reasoning with the ER and/or with their own conceptual knowledge. Therefore, guidelines for improving teaching and learning with ERs with respect to the reasoning (**R**) factor are presented below in Tables 7.2 and 7.3 respectively, as emanating either from the **R-M** or **R-C** factors of the model.

*Guidelines drawn from the Reasoning-Mode (**R-M**) factor.* Various guidelines can be suggested for developing students' reasoning skills or preventing or remediating any reasoning difficulties that occur from when students reason with the ER and its graphical features. Such reasoning difficulties may emanate from students' ability to perform cognitive processes such as decoding; deciphering; recognition; perception; visualisation; and organisation of patterns, shapes and colours; visuo-spatial operations; distinguishing relationships between ER features; organising visual information on the ER; analogical reasoning, symbolic reasoning, as well as surface-level and deep-level reasoning; transfer; and, translation between ERs, or set of ERs. Suggested guidelines for remediating or preventing difficulties related to the **R-M** factor are presented in Table 7.2 below.

Table 7.2 Guidelines for improving teaching and learning with ERs based on the Reasoning-Mode (**R-M**) Factor of the model

General Guideline Category	Specific Guidelines	Ideas obtained from present study (*) and/or literature
1. Empowering students with the skills needed to process ERs.	a) Be aware that little attention has been directed to actually explicitly "training" students to process ERs in science and knowing how to "read" an ER is a skill in itself which must be learned.	* Yair *et al.* (2003); Stylianidou *et al.* (2002); Lowe (1991,1986); Griffiths and Grant (1985); Holliday (1976, 1975a)
	b) Instructors must encourage students to adopt a strategic and purposeful approach to ER processing.	* Moore *et al.* (1993)
	c) Since not all ERs are interpreted in the same manner, instructors must supply students with the necessary domain-specific ER skills. For example, reading a complex electric circuit ER, or reading an ER of an algebraic function requires very different skills than reading an ER that depicts tertiary protein structure or one that portrays a genomic map.	* Roth (2002); Cheng *et al.* (2001); Henderson (1999); Kindfield (1993/1994); Gillespie (1993); Hill (1988)
	d) Students should perform tasks that require visual-spatial cognition. This is one way to gain ER-processing skills because *practice* with these tasks aids mental model construction.	Sanders (2002); Winn *et al.* (1991); Lord (1990, 1987a)
2. Teaching the "visual language" used by ERs.	a) Like oral communication, the *visual language* contained in ERs should be explicitly taught to learners so that they gain the necessary ER-processing skills.	* (section 3.3.1) Pintó and Ametller (2002); Peña and Quílez (2001); Lord (1987a, 1990)
	b) It is imperative that students get to process and learn ER conventions if they are to read ERs effectively. In this regard, students should expose themselves to a range of different pictorial conventions whenever possible.	* (section 4.3.1; 4.3.2; 6.3.3.3; 6.3.3.4) Roth (2002); Lowe (1991, 1986)
	c) Instructors should cue students to think more deeply about the markings contained in ERs when processing them in order to promote further deep-level reasoning in students.	Kozma (2003); Cheng *et al.* (2001); Lowe (1994a)
3. Encouraging students to use and interpret ERs in a meaningful way.	a) For abstract sciences, pictorial skills such as, using an ER as a "tool to think with", using an ER as a means to lessen the load placed on working memory and, using an ER to make important visual features salient during problem solving all contribute to favourable ER processing.	Kindfield (1993/1994, 1992)
	b) Learners should be encouraged to study ERs *before* commencing reading of associated text, and *again*, when a section of reading has been completed.	Moore *et al.* (1993)
	c) Teachers should encourage students to expose themselves to ERs with which they are familiar before having to deal with the processing constraints imposed by novel or more demanding ERs.	Lowe (1991)
	d) Even though it is important that viewers be acquainted with different forms of ER, experience with reasoning with static ERs should be the precursor to processing more dynamic and animated types.	Scaife and Rogers (1995)
4. Exposing students to multiple ERs of the same phenomenon.	a) Expose students to multiple ERs that all represent the same phenomenon. As a result, students should be stimulated to *practice* processing *different* ER conventions that depict identical ideas thereby improving their translation skills between one ER and another.	* Seufert (2003); Treagust *et al.* (2002); Pavlinic *et al.* (2001); Dufresne *et al.* (1997)
5. Predicting potential ER processing	a) Instructors and colleagues should process ERs themselves before exposing them to students, so that they can informally measure whether the ERs may induce erroneous processing.	Winn (1991)

problems.		
	b) Teachers should constantly query the manner in which students utilise ERs in order to gauge possible processing difficulties.	* (section 5.4.2; 5.4.3) Stylianidou *et al.* (2002)
	c) Teachers should imagine themselves as students in order to predict the mental operations a student might employ to process an ER.	Holliday (1975a)
6. Employing Learner-generated ERs.	a) Students' generation of their own diagrams is considered a powerful method for improving ER-processing that enhances scientific *visual literacy*.	Cheng *et al.* (2001); Gillespie (1993); Kindfield (1993/1994); Wheeler and Hill (1990); Lowe (1988a, 1988b, 1987); Alesandrini (1984); Fry (1981)
	b) Planning, constructing and refining their own ERs is one way for students to improve the processing of abstract ERs.	* (section 4.3.2) Glynn (1997); Lowe (1991); Rigney and Lutz (1976)

In consideration of Table 7.2 above, development of strategies for improving teaching and learning with ERs that have been drawn from the **R-M** factor of the model consist of six important general guidelines. Firstly, it is important that students posses the necessary skills to interpret scientific ERs (point 1, Table 7.2). In this regard, the results of the present project suggest that reading abstract ERs is a specialised skill and therefore, students should employ a strategic approach to ER-processing. Secondly, as a way to obtain these skills, teachers must ensure that they "teach" students the necessary visual language used by particular ERs (point 2, Table 7.2) and that students take ownership of their own role in this process. The results reported in this thesis have shown that processing of visual language used in biochemical ERs has a direct bearing on the interpretations (mental models) that students construct. One way of empowering students with these skills is to expose them to a variety of ERs that depict the multiple symbolic markings used by biochemical ERs. Thirdly, students should adopt a goal-orientated approach to ER-processing that ensures their ability to process ER markings in a meaningful manner (point 3, Table 7.2). Fourthly, related to students' exposure to a range of conventions for depicting scientific phenomena, it is vital that learners also consult a wide variety of ERs that depict the same phenomenon (point 4, Table 7.2). Fifthly, as a way to prevent ER processing difficulties, instructors should process ERs themselves before lectures or classes so that they can informally predict whether a certain ER (or set of ERs) may pose potential problems for students (point 5, Table 7.2). As suggested in this thesis, this process may include instructors acknowledging the role of PCK when teaching with ERs. Lastly, in relation to guidelines developed from the **C** factor (Table 7.1), by engaging in the activity of generating their own ERs (point 6, Table 7.2), students also develop their ER-processing

skills. By analysing the SGDs produced as part of the findings in the current project, it seems as though student diagramming is useful for improving ER processing of abstract ERs.

Guidelines drawn from the Reasoning-Conceptual (R-C) factor. According to the model, the **R-C** factor represents students' ability to reason with their conceptual knowledge of relevance to the ER. These reasoning mechanisms include memory-recall such as accessing, selection and processing of existing information of relevance to the ER; and, the assimilation, accommodation and integration of new knowledge learnt from the ER. In addition, reasoning processes represented by **R-C** also include analogical reasoning; transfer; superimposing of one concept upon another; and inductive and deductive reasoning. Like **R-M**, both sound and unsound reasoning is represented by the **R-C** factor. Difficulties that arise out of students' erroneous reasoning with their conceptual knowledge of relevance to the ER could be remediated by the approaches listed in Table 7.3.

Table 7.3 Guidelines for improving teaching and learning with ERs based on the Reasoning-Conceptual (**R-C**) Factor of the model

General Guideline Category	Specific Guidelines	Ideas obtained from present study (*) and/or literature
1. Cuing links and encouraging transfer between knowledge structures.	a) Students should be cued to make links to appropriate conceptual knowledge structures whenever possible.	*
	b) Students should be encouraged to transfer their conceptual knowledge to different contexts, thus making their knowledge more flexible and developing their transfer skills.	* Brna *et al.* (2001); Grayson (1995)
	c) When having to interpret novel ERs, learners should be encouraged to engage their previous positive experiences such as the confidence that might have been gained from interpreting challenging ERs.	*
2. Ascertaining the limitations of only a single ER.	a) It is necessary that educators and students alike, consciously analyse, scrutinise, critically examine and discuss each scientific ER.	Lowe (1999); Ben-Zvi and Genut (1998); Cox (1996); Barlex and Carré (1985)
	b) Students and instructors alike should constantly employ cognitive strategies that help to highlight differences in the understanding implied amongst ERs in order to ascertain the limitations of only a single ER.	* (section 6.3.3.5; 6.3.3.6) Treagust *et al.* (2002); Sumfleth and Telgenbüscher (2001); Hill (1990)
	c) Ascertaining the limitations of an ER should be especially followed in abstract sciences to avoid students thinking that the ER is the "reality", rather than only a *representation* of the reality.	* (sections 6.3.3.5; 6.3.3.6) Roth (2002); Nottis and McFarland (2001); Hill (1988)
3. Fostering a multiple representations approach.	a) Since a single ER lacks the power to show all aspects of an abstract scientific concept, students should aim to interpret multiple ERs simultaneously and link their interpretations to already existing knowledge to obtain different perspectives of a	Piez and Voxman (1997); Dickey (1993)

	concept.	
	b) Interpreting a range of ERs builds powerful and integrated internal representations of a scientific phenomenon that can be utilised when a student engages with further novel ERs that depict the same idea. In turn, "overloading" of the learner's *internal* representations (mental models) is avoided.	* Seufert (2003); Lowe (1993a); Levie and Lentz (1982)
	c) By "switching" from one representational system to another when interpreting multiple ERs, students equip themselves with a variety of cognitive strategies for solving ER-related problems.	Sumfleth and Telgenbüscher, (2001); Bodner and Domin (2000); Cox and Brna (1995); Lohse *et al.* (1991); Lord (1990, 1987a)
	d) When teaching with multiple ERs, research suggests that educators first use analogical ERs to link new concepts to existing knowledge. Following this, educators are encouraged to use abstract ERs to communicate the nature of the concept(s) to learners. Finally, where appropriate, realistic ERs should be used to help learners make newly constructed knowledge clearly distinguishable.	Alesandrini (1984)
4. Developing metacognitive and other mental processing skills.	a) Through the gradual employment of a multiple representations approach to learning from ERs (3. above), students benefit by being able to *reflect* upon their interpretations, which leads to a more powerful and meaningful integration of their conceptual knowledge.	* (section 5.4.3) Dufresne *et al.* (1997)
	b) Learners should be taught skills that help model expert thinking. Acquiring these *cognitive-operational* skills improves ER-related problem solving and induces students to think deeper about the meaning implied by ERs.	Lowe (2003, 1989); Grayson (1995)
5. Generating ERs to integrate new knowledge.	a) Students' integration of, and reasoning with, their knowledge can be improved by drawing their own diagrams of the same phenomenon depicted by the ER.	* (section 5.4.2) Kindfield (1993/1994); Lowe (1987)
	b) The drawing process enhances mental imagery and assists in making scientific concepts concrete as well as to integrate new ideas.	Levie and Lentz (1982); Lowe (1988b, 1987).
	c) Students' construction of an ER is a form of *sense-making* that helps students transfer their conceptual understanding to a particular task.	Brna *et al.* (2001); Bodner and Domin (2000); Glynn (1997)

In terms of proposed guidelines for improving teaching and learning with ERs corresponding to the **R-C** factor of the model, Table 7.3 above presents five general categories of guidelines. Firstly, it is important that when interpreting ERs, students are encouraged to make links to their already existing knowledge, and should be stimulated to make their knowledge more flexible by engaging in the transfer of their knowledge to new contexts (point 1, Table 7.3). In this regard, the studies reported in this thesis suggest that this process of transfer should be developed in students especially in situations where they are required to integrate the conceptual understanding gained from reading abstract ERs. Cuing appropriate transfer of knowledge will also help minimise inappropriate transfer such as in the case of the DNA-related difficulty identified in this thesis (see section 4.3.3). In a more general example, biochemistry students should be stimulated to transfer their knowledge of thermodynamics to the context of chemistry and vice-versa to integrate their understanding. Empowering students in this manner will provide them with the confidence to read challenging ERs as well

as integrate those abstract concepts depicted by these ERs. Secondly, it is pivotal, especially in the abstract sciences, that students and teachers alike, constantly examine the limitations of any single ER (point 2, Table 7.3) that represents a scientific idea in order to internalise the fact that ERs are just limited models of a particular aspect of a phenomenon. Findings in this study have shown that, since biochemistry students often interpret abstract ERs literally as complete depictions of *reality*, educators should get learners to "practice" internalising the idea that these ERs are just representations of a concept that are suitable in some cases but not in others. Thirdly, even though the importance of the use of multiple ERs for learning and teaching were discussed with regard to the **R-M** factor (Table 7.2), a *multiple representations approach* to teaching and learning with ERs (point 3, Table 7.3) is of paramount importance for constructing powerful and meaningful knowledge structures and for integrating new knowledge with already existing knowledge. In terms of the current study, those students who were best equipped to deal with novel ERs seemed to have an integrated knowledge structure far superior to other students, which implies that it is essential that learners engage in a multiple representations approach towards ER interpretation. Fourthly, through reflective and metacognitive approaches (point 4, Table 7.3), students should endeavour to "think about their own thinking" which will empower them to think more deeply about the meanings implied by ERs. This study has shown how this process (used particularly during interviews) induces students to "take a step back" and view the ER in a critical light, a skill that should be learnt and practiced. Lastly, in relation to guidelines emanating from both the **C** and **R-M** factors, students' generation of their own ERs with respect to the **R-C** factor (point 5, Table 7.3) is a powerful tool for mental imagery and the integration of knowledge. This has been shown to be true in the current study through the use of SGDs during interviews, where it is suggested that diagramming helps place students' interpretations in a different perspective, which may help them integrate their knowledge structures in unique and powerful ways.

Discussion of the guidelines outlined in Tables 7.1 - 7.3 above was concerned with strategies for improving *teaching and learning* with scientific ERs (research question 7). These guidelines have emerged from considering the role of the **C**, **R-M** and **R-C** factors of the model. Potential guidelines for improving the *design* of scientific ERs (research question 8) are now considered in terms of the **M** and **C-M** factors of the expressed model. These guidelines are outlined in Tables 7.4 and 7.5 below.

Guidelines drawn from the Representation mode (M) factor. This thesis has demonstrated that not only do internal (cognitive) characteristics of the ER viewer play a role in students interpretation of ERs but also the external characteristics, or mode of representation (**M**) of the ER. As defined previously, factor **M** represents the *nature of the ER* and how well (or poorly) its features represent the concepts, structures or processes it is designed to represent. These include the effective and ineffective use of graphical and diagrammatic features; the clarity of and relationship between representations; and, the spatial arrangement of elements, conventions, visual icons, visual cues, artistic devices, colour, complexity, topography, level of abstraction, symbols, labels and captions. Sources of difficulty associated with the **M** factor may include ER design features such as the artistic embellishments, the particular visual markings, devices and symbols used to represent the elements of the real phenomenon and, the confusing similarity of certain ERs across different contexts. Guidelines for the prevention or remediation of difficulties with respect to the **M** factor may be provided through the avenues outlined in Table 7.4 below:

Table 7.4 Guidelines for improving the design of ERs based on the Mode (**M**) Factor of the model

General Guideline Category	Specific Guidelines	Ideas obtained from present study (*) and/or literature
1. Acknowledging that not every ER is a good learning tool and therefore, that ER design should be scrutinised.	a) It is of pivotal importance for instructors *and* learners to acknowledge that the nature and composition of many ERs do not satisfy their instructional purposes.	* (section 6.3.3.5) Lohse *et al.* (1991); Hurt (1987)
	b) Instructors and ER designers must realise that an ER that seems clear to them may not be clear to a learner and that its features might need to be adjusted to assist the learner.	* Henderson (1999)
	c) Since ER research indicates that students often struggle with ERs that are highly abstract in nature, specific attention needs to be directed to their construction and design. The design characteristics of an ER play a vital role in determining how the ER is processed.	* (sections 6.3.3.5; 6.3.3.6) Stylianidou *et al.* (2002); Winn and Solomon (1993)
	d) Since scientific ERs are of such high instructional importance, ER designers must make every effort to increase the consideration that is given to them as education tools.	* Brna *et al.* (2001); Bernard (1990); Macdonald-Ross (1989)
2. Using ER conventions and graphical features that are well defined.	a) ER designers should use conventions that are well established in a particular scientific discipline because novices show difficulties when the graphical and spatial arrangements within an ER are unfamiliar to them.	* (section 6.3.3.3) Winn and Solomon (1993)
	b) Ideally, if possible, the graphical markings and conventions making up the ER should be conveyed in true proportion to the real world.	Holliday (1990)
	c) ER designers should take care when merging symbolic and real features on the same ER to prevent erroneous	Stylianidou *et al.* (2002)

	interpretations, as is often the case when depicting abstract scientific phenomena. For example, consider a "force" ER, which shows both a *real* object (e.g. a wheelbarrow) as well as *symbolic* markings (e.g. arrows) to depict forces acting on the object.	
	d) If guideline (a.) is not possible, conventions should be universally standardised or specially designed to deliver optimal clarity.	* (section 3.3.1) Lowe (1996); Hoffmann and Laszlo (1991); Guri-Rozenblit (1988)
	e) In biochemistry, a solution to the inclusion of idiosyncratic markings in ERs could be the formulation of specific *ER nomenclature* that could act as a framework for the standardisation of conventions across all ERs.	*
	f) Designers and authors must acknowledge that *universal* ER conventions have developed *together* with the particular scientific concept that they represent, often over many decades. Albeit so, it is common to find many scientific ERs where designers seem to have neglected this fact.	* (section 3.3.1)
	g) Some biochemistry textbook authors present the various conventions, colour codes and symbols used in the textbook, in a specially presented user guide to inform readers on the use of ER markings throughout the textbook. Other authors should endeavour to follow this example.	Roitt (1997); Garrett and Grisham (1995)
3. Considering the use of colour in ER design.	a) Use of colour in an ER is valuable when a particular visual feature needs to be highlighted or discriminated between or when a feature requires a learner's attention.	Holliday (1990); Dwyer (1970)
	b) When colour is plentiful, effectiveness of the ER is often lessened because the impact and contrast between colours is lessened and viewers are induced to process irrelevant ER features.	Winn (1991); Szlichcinski (1979)
	c) The use of the *same* colour to represent two distinctly different features should be avoided whenever possible.	* (section 6.3.3.3)
	d) Where possible, the use of colour should be as realistic as possible, in that it should correspond to the colour of the real world entity that is represented. It is acknowledged that this is extremely difficult to follow when representing scientific phenomena that are highly abstract in nature (e.g. "atom" or "binding-site").	de Lange (1999)
4. Considering the level of detail in an ER.	a) An increase in detail does not automatically mean that learning increases and often, for aesthetic reasons alone, more detail is provided than necessary.	de Lange (1999); Winn (1988); Holliday (1975b)
	b) ER designers should match the level of detail presented in an ER to the nature of the task that is required of a learner. For example, detail should be increased when learning requires the memorisation of specific concepts, but decreased when the learner is required to learn a certain process represented by an ER.	Winn (1988)
	c) As a way to deal with the level of detail in ER design, research suggests that ERs should be designed to present varying levels of abstraction *as well* as detail.	Do and Gross (2001)
	d) ERs are most effective when the amount of visual information present in the ER does not "overload" a learner's ability to interpret it and therefore, an ER should be presented as to first *attract* and then *direct* a learner.	Reid (1990a, b)
	e) With respect to guidelines (a. - d.) above, depicting structure and function in the same ER can cause confusion.	* (section 4.3.1) Crossley *et al.* (1996)
5. Encouraging consultation between ER designers and textbook authors.	a) The usual lack of any systematic interplay between ER designer and textbook author suggests that more attention should be given to suitable design principles for ERs in science. A union between graphic designer and textbook author is therefore desirable so that ER design becomes a significant and formal educational function.	Mayer (2003); Lowe (1997); Bernard (1990); Kosslyn (1989); Hurt (1987); Duchastel and Waller (1979); Duchastel (1978)
	b) The *presentation* of the display should share a high	Lowe (1999); Hardin

	correlation with the *content* of the display and the ER should convey information so efficiently that students and instructors never have to explicitly question its design.	(1993)
6. Designing ERs with cognitive constraints in mind.	a) A well-designed ER is one that is easily encoded by the human visual information-processing system and one that provides information in the clearest way possible.	Kosslyn (1989); Szlichcinski (1979)
	b) A well-designed ER should allow the viewer to successfully group or discriminate between external markings.	Cheng *et al.* (2001); Holliday (1990)
	c) ERs should be designed in such a way as to allow students to "chunk" visual markings appropriately. ER markings should be arranged in a way that novices are able to pay most attention to those features that correspond to the target phenomenon.	Lowe (1996); Egan and Schwartz (1979)
	d) ER designs should complement a learner's level of mental development and the "level of abstraction" used in an ER should match a learner's mental ability.	Gabel and Sherwood (1980); Arnheim (1970)

With respect to the guidelines for improving the design of ERs that have emerged in consideration of the **M** factor of the model, Table 7.4 has outlined six general guidelines. Firstly, it is important that both instructors and students realise that ERs do not always satisfy their learning objectives (point 1, Table 7.4). In this regard, as supported by the findings of the current project, it is necessary for educators (and learners) to scrutinise the nature of design of an ER. In addition, findings of the current thesis imply that biochemistry instructors should not simply assume that biochemical ERs are without design fault and that just because they appear simple to an instructor, that the same will hold for a learner. In this regard, a high educational priority should be given to the role of abstract ERs in the teaching and learning of biochemistry. Secondly, as pointed out in this work and by other authors, it is pivotal that when constructing ERs, designers make use of conventions that are accepted by that particular scientific discipline (point 2, Table 7.4) and refrain from using idiosyncratic markings. In this regard, the current project has pointed out the pitfalls of using symbolism that is unfamiliar to students and emphasised the importance of biochemistry developing their own set of conventions where feasible. For example, the multiple notations available for depicting the S-S bond may confuse learners. However, it is acknowledged that even though this is not always plausible in abstract sciences such as biochemistry, where little standardisation exists and often no formal "conventions" are available from which to draw, attempts must be made to refrain from using idiosyncratic markings in ERs. Having stated this, some ER designers in biochemistry nevertheless, seem to be more concerned with generating idiosyncratic symbols then using those conventions that are considered as "standardised" such as the "ribbon" and "space-filling" notations. Thirdly, even though the use of colour has been shown to be an important ER design variable, its use should be carefully considered by ER designers (point 3, Table 7.4) because an overuse, or poor use of colour, can induce processing difficulties. For

example, in one case described in this thesis, the same red colouring was used to depict contact points between amino acids as well as to depict only one amino acid alone, which caused much confusion among some students. Fourthly, the role of the amount of detail contained within an ER (point 4, Table 7.4) should be considered during the design of scientific ERs since a minor increase or decrease in level can induce processing problems. This was illustrated by the fact that ER A (of relative low detail) caused a high percentage incidence of difficulty than did ER D (of relative high detail). In addition, this thesis has shown that problems arise when ideas of structure are presented together with ideas of function on the same ER (e.g. ERs A, C and D all represent Ab structure as well as possible interaction with Ag). Related to this, in the context of biochemistry, many ERs that depict the mitochondrion show ideas of structure *together* with ideas of function (Crossley *et al.*, 1996). Like all good models, ERs should be distinct representations of a structure, function or process. Fifthly, it is pivotal that there is significant consultation between design and content experts (point 5, Table 7.4) during textbook productions that contain ERs. This process should be seen as a high priority and a formal educational task. Lastly, if ERs are to be well-designed learning tools, it is essential that designers acknowledge the role of cognitive science in the process so that implications for learning can be better assessed (point 6, Table 7.4).

*Guidelines drawn from the Conceptual-Mode (**C-M**) factor.* As defined in this thesis, the nature of the conceptual (propositional) knowledge represented by the ER and its symbolism is represented by the **C-M** factor. This factor includes the extent, complexity and soundness of the knowledge represented by the ER, and therefore, how cognitively demanding it is. The nature of the conceptual knowledge depicted by an ER is often a source of student difficulties. Guidelines for remediating or preventing difficulties that arise from the propositional knowledge reflected by the ER and its markings (**C-M**) are presented in Table 7.5.

Table 7.5 Guidelines for improving the design of ERs based on the Conceptual-Mode (**C-M**) Factor of the model

General Guideline Category	Specific Guidelines	Ideas obtained from present study (*) and/or literature
1. Scrutinising ERs to establish whether they are representing scientifically sound knowledge.	a) Instructors should examine textbook and computer-based ERs to see that they are representing the scientific (propositional) knowledge that they are designed to represent.	* (sections 3.3.1; 4.2.2.; 6.3.3.6)
	b) Instructors should gauge whether the propositional knowledge represented by a particular ER is also shared by	* (sections 3.3.1; 4.2.2.; 4.3.2.6)

	other ERs that represent the same phenomenon, i.e. consistency across ERs representing the same idea.	
2. Defining the role of surrounding text and figure captions for representing the conceptual knowledge depicted by an ER.	a) Authors and instructors must ensure that the text surrounding an ER is an accurate scientific description of the graphical markings represented in the ER.	* (section 6.3.3.6)
	b) ERs that are placed within expository text should be directly applicable to the surrounding text and ERs that show conflict with the semantics in the text should be avoided. It is important that the two mediums support each other so that communication of the science can be enhanced.	de Lange (1999); Hurt (1987); Joseph and Dwyer (1984); Rigney and Lutz (1976)
	c) ERs inserted within expository text, should aim to *explain* rather than simply represent.	de Lange (1999); Hartley (1990)
	d) It is important for ERs to be placed in a pre-empted, logical and systematic manner within scientific prose.	Holliday and Harvey (1976)
	e) Authors and instructors must ensure that the figure caption is a scientifically accurate depiction of the graphical markings contained within an ER.	* (sections 4.3.1; 4.3.2; 6.3.3.6) Guri-Rozenblit (1988)
3. Determining the role of ER conventions and graphical markings for representing scientific knowledge.	a) Instructors should help learners appreciate in what *cases* particular pictorial conventions are used for which particular scientific ideas.	* (sections 4.3.1; 4.3.2; 6.3.3.6) Winn *et al.* (1991)
	b) Instructors should make clear to students the specific *role* that the conventions are playing within an ER with respect to the portrayed science.	* (sections 4.3.1; 4.3.2; 6.3.3.6) Cheng *et al.* (2001)
	c) Learners should be aware of two components related to ER *conventions*: conventions of *style* as well as conventions of *meaning*. Conventions of meaning are what result when a scientific concept is 'transformed' as a certain graphical feature on an ER. Conventions of style are those graphical features that are related to shape, texture and colour.	Lowe (1988)
	d) If learners are to interpret the science conveyed by an ER appropriately, they should be aware of how visual conventions are related to the real world.	Lowe (1991)
	e) Students can empower themselves with understanding the nature of the propositional knowledge conveyed by the ER by realising that multiple ERs and conventions are *subject to change*.	Roth (2002)
4. Realising that there are multiple ways for representing the same scientific knowledge.	a) Teachers must stress that there are many possible means for representing a single scientific concept and no absolute ER exists for a particular scientific concept, especially one that is abstract.	* (sections 3.3.1; 6.3.3.5; 6.3.3.6)
	b) In order to appreciate the nature of the scientific (propositional) knowledge conveyed by the ER, teachers and students should offer suggestions for alternate methods of representation of an ER.	Gillespie (1993)
	c) In order to gain a deeper appreciation of the science conveyed by an ER, instructors should stimulate students to explicitly describe the graphical elements making up an ER, explain the relationships between the components and explain which graphical components are *not* represented in the ER.	* (section 5.4.3) Pintó and Ametller (2002); Peña and Quílez (2001)
5. Stimulating the interplay between scientists and ER designers for the purpose of representing	a) An interaction between content and design experts should aim to represent scientific content in the clearest possible way to aid the viewer and the most applicable graphical features should be chosen for design, which relate directly to the particular scientific context in question.	Pintó and Ametller (2002)

scientifically sound knowledge in ERs.		
	b) Interaction between designer and author should include a prediction of how novices will respond to an ER in order to measure how well the propositional knowledge is represented by the ER. A way to achieve this would be to use students to pilot the ERs before distribution in subsequent textbooks and other educational resources.	Stylianidou *et al.* (2002); Lowe (1993a)

In addition to guidelines for ER design obtained from the **M** factor (Table 7.4), guidelines that have emanated from the **C-M** factor above (Table 7.5) also inform the design of ERs for remediating or preventing student difficulties. With regard to the **C-M** factor, five general strategies have been put forward. Firstly, educators and learners should scrutinise ERs to ascertain whether an ER accurately represents particular scientific knowledge (point 1, Table 7.5). In this regard, an implication of the current study was that instructors should validate the propositional knowledge represented by an ER by checking multiple sources representing the same propositional knowledge to see if it is scientifically correct. For instance, this study has highlighted misgivings in the propositional knowledge depicted in ERs that convey quaternary protein structure (see section 4.3.2.6). Secondly, it is important to appreciate the role of surrounding text and figure captions for representing the propositional knowledge contained within an ER (point 2, Table 7.5). The author and supervisor of the current study have deduced that it is important that the surrounding biochemistry text of an ER succinctly explains, in the clearest way possible, the markings contained in the ER and the relationships between them. Some biochemistry textbook authors do endeavour to describe any graphical markings (e.g. by means of a key in a preface) that may be a source of confusion and do not merely assume that readers will know what science is being represented. In this regard, results from the current project suggest that this practice should become a formal function in biochemistry education. Thirdly, as was also discussed in terms of guidelines emerging from both the **M** and **R-M** factors, the nature of the graphical features and ER conventions used to portray scientific knowledge (point 3, Table 7.5) is an important variable affecting students' potential interpretations. In the context of biochemistry, as stated previously and reinforced by the results of the current work, it is crucial that instructors *define* the role of a "convention" to learners and ensure that learners realise that the use of conventions is necessary for communicating abstract ideas because as yet, we cannot physically see the submicroscopic environment. Fourthly, students and educators should appreciate that many different ERs are available for representing a specific scientific concept (point 4, Table 7.5) and that no single ER is an exhaustive pictorial account of a scientific idea. As demonstrated in the current thesis, each ER is simply a representation of certain propositional knowledge that is not a

model of explanation applicable to all possible cases. This understanding should be stressed both to students and textbook authors. As a way to achieve this understanding in students, results from this study, suggest that instructors should induce students to explicitly describe *what* symbolism is used in an ER, *why* it is used and, *how* it is used. Lastly, as was also discussed with respect to factor **M**, in order to portray a scientifically acceptable representation of any propositional knowledge with an ER, it is essential that scientists and ER designers collaborate during ER design (point 5, Table 7.5).

The guidelines and strategies above (Tables 7.1 - 7.5) for preventing and remediating students difficulties with ER interpretation have been informed by considering each factor of the expressed model (Fig. 7.1). The presented guidelines, in addition to addressing ER-related difficulties, also aim to develop learners' reasoning skills. Furthermore, the proposed strategies aim to optimise the interpretation of the propositional knowledge conveyed by an ER. Moreover, due to the overlapping nature and interrelationships between the factors of the model (Fig. 6.2), some of the above guidelines and strategies (Tables 7.1 - 7.5) are common to more than one factor of the model suggesting that future work could involve the development of fewer strategies in which several sub-categories are incorporated. The proposed guidelines and strategies may be implemented by researchers, authors and educators for improving the use of ERs in the learning and teaching of biochemistry and, science in general. In lieu of this, we realise that many of the guidelines proposed above are clearly far too complex for teachers to implement immediately in practical settings. Nevertheless the identification of the above strategies serves as a solid foundation upon which more "user-friendly" guidelines can be devised and implemented in the future. Thus translation of the guidelines discussed in this work into less complicated and "do-able" strategies for improving the interpretation of ERs in science remains an important focus of this author's future research endeavours.

Implementation of any of the stated guidelines would inform PCK and therefore, the design and development of curricula (Fig 7.1) where there would be a strong emphasis on visual literacy and the use of ERs in scientific contexts. In this regard, as early as 1981, Fry called for curriculum designers to acknowledge the importance of ERs in the discourse of science. Following this, other writers have called for the formal implementation of a *visual literacy* into the curriculum (e.g. Gobert and Clement, 1999; Szabo *et al.*, 1981). Furthermore, other workers who have echoed this view (e.g. Brna *et al.*, 2001; Guri-Rozenblit, 1988) have called for the formal assessment of *visual skills* to be implemented as part of scientific curricula.

According to Lowe (2000; 1988a, 1986), learners' capacities to process scientific ERs should be developed and nurtured from a very young age and should be formally assessed at all levels of science education. It is the opinion of the current author, therefore, that if the pedagogical importance of visual literacy and ER processing is taken seriously, then this will be a vehicle for national and worldwide curriculum development and reform (Fig. 7.1).

In lieu of the importance of ERs in science education, five succinct fundamentals have been identified, which the author believes represent the cornerstones of the abovementioned curriculum development and design. The five research elements are the *meaningful learning* element, the *knowledge* element, the *skill* element, the *design* element and the *expert versus novice* element. Based on an extensive exploration of the literature and on the findings of this thesis, the five fundamentals, which we have termed *research elements,* could also serve as the basis for future research on learning and teaching with ERs in science education.

Meaningful learning element. Current literature motivates that curriculum designers and future researchers should take cognisance of current theories on how people are thought to learn from ERs (e.g. Mayer, 2003, 1997, 1993, 1989). Meaningful learning is an active and generative process (Osborne and Wittrock, 1983) characterised by the construction of understanding (von Glasersfeld, 1989), rather than a passive absorption or recall of rote-learned knowledge (e.g. Ward and Wandersee, 2002). Instructional approaches where students are seen as passive vehicles are not very effective (Grayson, 2004, 1995). However, although ERs play a substantial role in student-teacher communication (e.g. Brna *et al.*, 2001), one big problem is that teachers (and learners) often view ERs as being self-explanatory (Sumfleth and Telgenbüscher, 2001; Lowe, 2000). Instead, both instructors and students should adopt a meaningful and active learning approach that is concerned with constructing useful mental models. As demonstrated by the findings of this thesis, if learners' mental models correlate favourably with the target phenomenon, then in effect, the learner is actively *generating* sound scientific understanding (e.g. Peña and Quílez, 2001; Kindfield, 1993/1994; Lowe, 1993b). To promote meaningful ER-processing, teachers should follow postulates of external and distributed cognition (Scaife and Rogers, 1995; Zhang and Norman, 1994) where learning from ERs is considered as a representational system: as external *and* internal dimensions that exist together (e.g. Brna *et al.*, 2001). In this regard, any science curriculum must allow for the crucial role played by mental models in active learning. Furthermore, the design features making up an ER and/or textbook should match the

processing required for *meaningful* learning where Mayer *et al.* (1995) have shown that even small adjustments to scientific ERs and textbooks can have dramatic and positive effects on learning.

Knowledge element. ER research, including that presented in this thesis, has shown that deficiencies in conceptual knowledge attributed to the ER or, a lack of knowledge of the visual language used to represent scientific content in an ER, can both contribute to learning difficulties (e.g. Lowe, 2003, 1989; Pintó and Ametller, 2002; Table 7.1). A definite obstacle that learners in science often face is that they lack the knowledge of the graphical markings (conventions) used to represent scientific ideas in the ER (e.g. Lowe, 1989). As a result, constructing useful mental models from abstract ERs can be enormously challenging, especially when students do not have knowledge of the graphical conventions as part of their direct experience (e.g. Lowe, 1996; Table 7.1). Ideally, as shown in the studies reported here, effective interpretation of scientific ERs requires an ability to draw inferences from the ER and to link these to current knowledge to construct the appropriate understanding (e.g. Wandersee, 1994; Reinking, 1986; Table 7.3). Additionally, Wheeler and Hill (1990), Winn (1982) and Szlichcinski (1979) have stated that the *manner* in which information is obtained from an ER depends both on the viewer's prior knowledge as well as the viewer's knowledge of the objective, plan or purpose associated to reading the ER. Thus both curriculum designers and future ER researchers should realise the importance of developing sound conceptual and graphical knowledge among students that use ERs to learn with.

Skill element. In addition to the findings of this thesis, other ER-related studies in science education (e.g. Gobert and Clement, 1999) have also shown that students' are often totally unaware of *how* to *read or process* ERs appropriately. On top of this, since different types of ERs convey different types of information, learners' processing mechanisms need to be different for different ERs (Winn, 1982). Often, learners do not posses the cognitive skills necessary for the required ER processing (e.g. Schnotz, 1993a; Kindfield, 1993/1994; Lowe, 1991; Guri-Rozenblit, 1988; Table 7.2). As an explanation for this, Egan and Schwartz (1979) suggest that processing the visual information within an ER requires a large degree of *perceptual* skill. Additionally, reading an ER is also an *acquired* skill because learners have to learn the graphical notations (e.g. conventions) explicitly if understanding is to be fruitful at all (e.g. Petre and Green, 1993). Hence, the information drawn from an ER depends largely on what the viewer has 'learned' to look for (e.g. Petre and Green, 1993; Winn, 1993).

Furthermore, when reading ERs, students' often use cognitive skills that they are not *used* to, which also contributes to difficulties (e.g. Winn, 1987). It has been shown (e.g. Larkin and Simon, 1987) that individuals who possess the necessary ER processing skills gain more value out of the interpretation than those individuals who do not. In general, reading a scientific ER requires very different skills to those required for reading everyday pictures. When reading scientific ERs, especially those that are abstract, it is merely assumed that the information presented will not be taken literally (Lowe, 2000; Table 7.3). However, as demonstrated by the results of this thesis, abstract scientific ERs use a variety of graphical conventions to represent the real world, which makes the unskilled viewer's task even more difficult (e.g. Lowe, 1994b, 1993; Winn *et al.*, 1991; Wheeler and Hill, 1990; Tables 7.2, 7.3). Thus curriculum designers should realise the importance of incorporating ER-skill development into science curricula. As part of this process, researchers should study all facets of this topic to promote such ER-related skills.

Expert versus novice element. Instructors (experts) are often not aware that ERs can lead to learning difficulties for students (novices). Experts already possess the necessary ER-processing skills and knowledge (e.g. Henderson, 1999). For instance, experts *know* what to look for and *where* to look for it in the ER (e.g. Winn, 1993). It is not surprising therefore, that one of the main activities of professional scientists is the construction of their own ERs (e.g. Bowen *et al.*, 1999; Roth *et al.*, 1999; Kozma and Russell, 1997). However, as portrayed by the results of this thesis, inexperienced students are not as competent and interpret ERs very differently to experts, in manners not anticipated (e.g. Lowe, 1993a; Constable *et al.*, 1988). As a solution, Lowe (1989) stresses the pedagogical importance of *visual learning* in science and suggests that textbook authors, professional scientists and science educators must realise that some students find scientific ERs very difficult to perceive. One problem is that experts concentrate more on processing the actual conventions used in the ERs while inexperienced students do not (e.g. Wheeler and Hill, 1990; Lowe, 1989). For example, experts easily relate arrow length to the magnitude of a force, or relate schematic 'circles' to the representation of particles of matter in such ERs, while novices have problems performing such processing. Another problem is that, in the past, only skilled individuals interacting in specialised contexts have been privy to the use of abstract ERs to communicate information (Lowe, 1993a). However, these days there is a huge availability of such ERs in science education, and novices are expected to understand ERs even though they may not possess the required expertise (e.g. Lowe, 1993a). Thus a student's success with ER interpretation

depends largely on the level of expertise that the viewer brings to the ER. In this regard, it is important that the "novice/expert issue" is brought into consideration when designing science curricula and when carrying out ER research.

Design element. Scientific ERs that have not been designed with the goal of communicating intended meaning contribute to learning difficulties (e.g. Pintó and Ametller, 2002; Blackwell, 2001; Duchastel, 1988; Table 7.4). As shown in the present project, although written languages (e.g. German) and symbolic languages (e.g. Algebra) have formal rules for their expression and notation, visual expression in ERs is not bound by any unified system of convention (e.g. Lowe, 1987) particularly in the life sciences (e.g. Table 7.4). The lack of any rule-based method for expressing and presenting ERs is also a source of conceptual and reasoning difficulties (e.g. Henderson, 1999). As shown in the findings of the current work, even ERs such as those of IgG structure and function (Figs 4.1 and 5.2), that appear 'simple' on the surface can still place high processing demands on viewers (Lowe, 1989). Likewise, as there are varying levels of difficulty for reading text, some ERs are more difficult to read than other ERs (Lowe, 1994b, 1991). In these instances, ERs are sometimes restricted in their "representational power" (the potential of the ER to convey the intended meaning to the viewer) because different students' often interpret the very same ER differently (Stenning and Lemon, 2001). Hence, ER processing mechanisms are also largely determined by the nature of ER design (e.g. Table 7.5). Evidently, difficulties are enhanced when there are deficiencies in ER design (e.g. Blackwell, 2001). Associated with the design element, is the sometimes-poor ability of learners to interact with multiple ERs of a scientific phenomenon (e.g. Seufert, 2003). Here, students often concentrate only on a single ER design that is familiar or concrete, rather than consulting a range of ERs that express the same idea (e.g. Table 7.5). Overall, ER designers should strive for favourable correlation between viewers' constructed mental models and their own intended message (Lowe, 1993a). However, even ERs considered of 'good' design may sometimes cause difficulties, resulting in the viewer's understanding being different to that intended by author/designer. Thus it is essential that curriculum materials should consist of well designed ERs and science education research should actively focus on optimising ER design and therefore, teaching and learning with ERs.

In summary, the following specific research outcomes were achieved when addressing research questions 1-8 (Chapter 1):

1. Three categories and seventeen sub-categories of students' difficulties with the interpretation of three ERs of antibody-antigen interaction have been identified.
2. Potential sources of the above students' difficulties were uncovered and informed the proposal of three factors affecting students' ability to interpret ERs namely, students' ability to *reason* with the ER and with their own conceptual knowledge (**R**), students' understanding (or lack thereof) of the *concepts* of relevance to the ER (**C**) and, the *mode* in which the desired phenomenon was represented in the ER (**M**).
3. A three-phase single interview technique (3P-SIT) was designed and tested to further investigate, and generate empirical data on, the above three factors.
4. The 3P-SIT instrument proved to be a novel and reliable instrument for generating empirical data on the three factors and its use led to the identification of four further factors affecting students' ability to interpret ERs, namely the reasoning-conceptual factor (**R-C**), the reasoning-mode factor (**R-M**), the conceptual-mode factor (**C-M**) and the conceptual-reasoning-mode factor (**C-R-M**).
5. Through the modelling process of Justi and Gilbert (2002) and the use of 3P-SIT to generate empirical data, a novel model of seven factors that determine students' ability to interpret ERs in biochemistry has been expressed and operational definitions for each factor have been validated.
6. The model can be applied to qualitatively determine the nature and extent of the influence of a factor during students' interpretation of ERs.
7. The model makes a major contribution to how data on student difficulties with ERs could be analysed and in doing so, the model informs and guides this analytical process.
8. Once a worker has obtained data applicable to the **C**, **R-C** and/or **R-M** factors constituting the model, the model can be used to frame guidelines for improving teaching and learning with an ER in science, including PCK and remediation.
9. Once a worker has obtained data applicable to the **M** and **C-M** factors constituting the model, the model can be used to frame guidelines for improving the design of scientific ERs.
10. Findings from this thesis coupled to other relevant literature have provided a platform, in the form of five research elements, from which to base curriculum design and future research on the use of ERs in science education.

11. The findings described in (**1.**) confirm the results of other studies (Chapter 2), which show that misinterpretation of ERs in science can lead to conceptual and reasoning difficulties.
12. The findings in (**1.**) constitute an important and novel contribution to the little known research area of learning and teaching in biochemistry, since no other study has identified and classified students' difficulties with the interpretation of ERs that show antibody-antigen interaction.
13. This is the first study to consider the sources of students' difficulties (see **2.**) with the interpretation of ERs that show antibody-antigen interaction.
14. The 3P-SIT instrument (see **3.**) shows great potential for use in other scientific contexts by other workers to obtain data that reflects the seven factors affecting students' ability to interpret scientific ERs.
15. The model expressed by this research (see **5.**) can be applied to any scientific context for framing and guiding researchers', educators' and authors' thinking about the nature of students' difficulties with ER interpretation and their prevention and remediation.
16. The model is unique in that it provides a generalisable means for workers to consider their findings and the implications thereof in the context of science education research. The model may serve as a guiding framework with which to base future research on students' interpretation of ERs in science.

In conclusion, the author believes that future ER research in science education will be greatly shaped by the disciplines of cognitive science and cognitive psychology. From a cognitive perspective, the current standing today is that not a lot is known about the higher-order cognition of ERs (Peña and Quílez, 2001; Scaife and Rogers, 1996; Lowe, 1993a; Schnotz, 1993a), even though some promising inroads are currently being made in the context of dynamic and animated ERs (e.g. Chandler, 2004; Hegarty, 2004; Lowe, 2003). Additionally, there is only a limited appreciation of the cognitive mechanisms responsible for the processing of ERs within text (e.g. Glenberg and Langston, 1992). Furthermore, the *way* ERs are processed is poorly understood because a huge diversity of ER forms is available to learners, each with their own instructional goals (Blackwell, 2001). Recently, Blackwell (2001) has advised that theoretical studies, which explore the deficiencies in ER design as well as teachers and students use of ERs, are long overdue. The ultimate aim would be to propose an integrated theory on which *practical* interventions for the use of ERs in science education could be based (Mayer and Anderson, 1992). On this score, recent commentary

suggests that much *more* work is needed to understand how students learn from, translate between, and use ERs during learning (e.g. Ploetzner and Lowe, 2004; Reimann, 2003; Brna *et al.*, 2001). If we are to arrive at anything of use, then it is essential that workers always consider the *cognitive* constraints associated to learning with ERs (e.g. Chandler, 2004; Hegarty, 2004; Seufert, 2003). The findings represented in this thesis have contributed to solving some of the above deficiencies in knowledge.

REFERENCES

Ainsworth, S.E., Bibby, P.A. and Wood, D.J. (1998) Analysing the costs and benefits of multi-representational learning environments. In: M.W. van Someren, P. Reimann, H.P.A. Boshuizen and T. de Jong (Eds.), *Learning with Multiple Representations*, Amsterdam: Elsevier Science, pp. 120-134

Alesandrini, K.L. (1984) Pictures and adult learning. *Instructional Science*, **13**, 63-77

Ametller, J. and Pintó, R. (2002) Students' reading of innovative images of energy at secondary school level. *International Journal of Science Education*, **24**(3), 285-312

Amit, A.G., Mariuzza, R.A., Phillips, S.E.V. and Poljak, R.J. (1986) Three-dimensional structure of an antigen-antibody complex at 2.8 Å resolution. *Science*, **233**, 747-753

Anderson, J.R. (1978) Arguments concerning representations for mental imagery. *Psychological Review*, **85**, 249-277

Anderson, G. and Arsenault, N. (1998) *Fundamentals of Educational Research, 2nd ed.*, London: Falmer Press

Anderson, M. and Armen, C. (1998) Diagrammatic reasoning and color. *Paper presented at the AAAI Fall Symposium on Formalization of Reasoning with Visual and Diagrammatic Representations*, 23-25 October, Orlando

Anderson, T.R and Grayson, D.J. (1994) Improving students' understanding of carbohydrate metabolism in first-year biochemistry at tertiary level. *Research in Science Education*, **24**, 1-10

Anderson, T.R. and McKenzie, J.M. (2002) Using Meta-analysis to Develop a Database of Students' Conceptual and Reasoning Difficulties (CARD). *Proceedings of the Tenth Annual Conference of the Southern African Association for Research in Mathematics, Science and Technology Education* (C. Malcolm and C. Lubisi, Eds.), pp. III-11 - III-17

Anderson, T.R., Crossley, L.G. and Grayson, D.J. (1999) Identifying students' conceptual and reasoning difficulties with biochemistry. In: M. Komorek, H. Behrendt, H. Dahncke, R. Duit, W. Graber and A. Kross (Eds.), *Research in Science Education, Past, Present and Future*, Kiel: IPN, pp. 86-88

Anderson, T.R., Grayson, D.J., Crossley, L.G., and Schönborn, K.J. (2000) Do students really understand metabolism the way we think they do? *Paper presented at the 18th International Union of Biochemistry and Molecular Biology (IUBMB) Congress: Beyond the Genome*, 16-20 July, Birmingham

Arnheim, R. (1970) *Visual Thinking*, London: Faber and Faber

Arons, A. B. (1990) *A Guide to Introductory Physics Teaching*, New York: John Wiley

Ausubel, D. (1968) *Educational Psychology*, New York: Holt, Rinehart & Winston

Baddeley, A. (1992) Working memory. *Science*, **255**, 556-559

Baker, S.R. and Hill, D. (1983) A Proposed Typology for the Visualisation Process. *Journal of Mental Imagery*, 7(2), 67-74

Baker, S.R. and Talley, L.H. (1974) Visualization skills as a component of aptitude for chemistry - a construct validation study. *Journal of Research in Science Teaching*, **11**, 95-97

Barke, H.-D. (1993) Chemical education and spatial ability. *Journal of Chemical Education*, **70**(12), 968-971

Barlex, D. and Carré, C. (1985) *Visual Communication in Science*, London: Cambridge University Press

Beilfuss, M., Dickerson, D., Libarkin, J.C. and Boone, W. (2004) Exploring conceptual understanding of groundwater through college student's interviews and drawings. *Proceedings of the National Association for Research in Science Teaching (NARST)*, April 1-3, Vancouver, Electronic paper number 204743.

Bell, J. (1999) *Doing your research project: a guide for first-time researchers in education and social science, 3rd ed.*, Buckingham: Open University Press

Bell, P. (2001) Content Analysis of Visual Images. In: T. van Leeuwen and C. Jewitt, (Eds.), *Handbook of Visual Analysis*, London: Sage, pp. 11-34

Bennett, K.B. and Flach, J.M. (1992) Graphical displays: implications for divided attention, focused attention, and problem solving. *Human Factors*, **34**(5), 513-533

Ben-Zvi, N. and Genut, S. (1998) Uses and limitations of scientific models: the periodic table as an inductive tool. *International Journal of Science Education*, **20**(3), 351-360

Berg, C.A. and Phillips, D.G. (1994) An investigation of the relationship between logical thinking structures and the ability to construct and interpret line graphs. *Journal of Research in Science Teaching*, **31**(4), 323-344

Bernard, R.M. (1990) Using extended captions to improve learning from instructional illustrations. *British Journal of Educational Technology*, **21**(3), 215-225

Birk, J.P. and Kurtz, M.J. (1999) Effect of experience on retention and elimination of misconceptions about molecular structure and bonding. *Journal of Chemical Education*, **76**(1), 124-128

Blackwell, A.F. (2001) Introduction: thinking with diagrams. *Artificial Intelligence Review*, **15**(1-2), 1-3

Blackwell, A.F., Whitley, K.N., Good, J. and Petre, M. (2001) Cognitive factors in programming with diagrams. *Artificial Intelligence Review*, **15**(1-2), 95-114

Boardman, D.J. (1976) Graphicacy in the curriculum. *Educational Review*, **28**, 118-125

Bodner, G.M. and Domin, D.S. (2000) Mental models: the role of representations in problem solving in chemistry. *University Chemistry Education*, **4**(1), 22-28

Bohinski, R.C. (1987) *Modern Concepts in Biochemistry, 5th ed.*, Boston: Allyn and Bacon, p. 161

Boo, H.K. (1998) Students' Understandings of Chemical Bonds and the Energetics of Chemical Reactions. *Journal of Research in Science Teaching*, **35**(5), 569-581

Borrell, P. and Dixon, D.T. (1984) Electrode potential diagrams and their use in the Hill-Bendall or z-scheme for photosynthesis. *Journal of Chemical Education*, **61**(1), 83-86

Bos, W. and Tarnai, C. (1999a) Content analysis in empirical social research. *International Journal of Educational Research*, **31**, 659-671

Bos, W. and Tarnai, C. (1999b) Editorial: guest editor's introduction. *International Journal of Educational Research*, **31**, 657-658

Bowen, C.W. (1994) Think-aloud methods in chemistry education. *Journal of Chemical Education*, **71**(3), 184-190

Bowen, G.M. Roth, W.-M. and McGinn, M.K. (1999) Interpretation of graphs by university biology students and practicing scientists: toward a social practice view of scientific representation practices. *Journal of Research in Science Teaching*, **36**(9), 1020-1043

Boyes, E. and Stanisstreet, M. (1991) Misconceptions in first-year undergraduate science students about energy sources for living organisms. *Journal of Biological Education*, **25**, 209-213

Braden, R.A. and Hortin, J.A. (1982). Identifying the theoretical foundations of visual literacy. *Journal of Visual/Verbal Languaging*, **2**, 37-42

Brekke, O.H., Michaelsen, T.E. and Sandlie, I. (1995) The structural requirements for complement activation by IgG: does it hinge on the hinge? *Immunology Today*, **16**(2), 85-90

Britton, L.A. and Wandersee, J.H. (1997) Cutting up text to make moveable, magnetic diagrams: a way of teaching and assessing biological processes. *The American Biology Teacher*, **59**(5), 288-291

Brna, P., Cox, R. and Good, J. (2001) Learning to think and communicate with diagrams: 14 questions to consider. *Artificial Intelligence Review*, **15**(1-2), 115-134

Bruner, J. (1960) *The Process of Education*, Cambridge, MA: Harvard University Press.

Bruner, J. (1986) *Actual minds, possible worlds*, Cambridge, MA: Harvard University Press

Bukatko, D. and Daehler, M.W. (1992) *Child development: a topical approach*, Boston: Houghton Mifflin

Cammack, R. (2001) Nomenclature Committee of IUBMB, King's College, *Personal Communication*

Campbell, P.N. and Smith, A.D. (2000) *Biochemistry Illustrated, 4th ed.*, Edinburgh: Churchill Livingstone

Case, J, Gunstone, R. and Lewis, A. (2001) Students' metacognitive development in an innovative second year chemical engineering course. *Research in Science Education,* **31**(3), 313-335

Chandler, P. (2004) The crucial role of cognitive processes in the design of dynamic visualizations. *Learning and Instruction*, **14**, 353-357

Cheng, C.-H., Lowe, R.K. and Scaife, M. (2001) Cognitive Science Approaches To Understanding Diagrammatic Representations. *Artificial intelligence Review*, **15**(1-2), 79-94

Chi, M.T.H., Feltovich, P.J. and Glaser, R. (1981) Categorization and representation of physics problems by experts and novices. *Cognitive Science*, **5**, 121-152

Clark, J.M. and Paivio, A. (1991) Dual coding theory and education. *Educational Psychology Review*, **3**(3), 149-210

Coetzer, T.H.T. (2004) School of Molecular and Cellular Biosciences, University of KwaZulu-Natal, *Personal Communication*

Cohen, R., Eylon, B. and Ganiel, U. (1983) Potential difference and current in simple electric circuits. *American Journal of Physics*, **15**(5), 407-412

Cohen, L. and Manion, L. (1994) *Research methods in education, 4th ed.*, London: Routledge

Cohen, L., Manion, L., Morrison, K. (2000) *Research methods in education, 5th ed.*, London: RoutledgeFalmer

Constable, H., Campbell, B. and Brown, R. (1988) Sectional drawings from science textbooks: an experimental investigation into pupils' understanding. *British Journal of Educational Psychology*, **58**, 89-102

Coon, D. (2001) *Introduction to Psychology: Gateways to mind and behaviour, 9th ed.*, Belmont: Thomson Learning

Cox, R.A. (1996) Is it naïve to expect school science books to be accurate? *School Science Review*, **78**(282), 23-31

Cox, R. and Brna, P. (1995) Supporting the use of external representations in problem solving: the need for flexible learning environments. *Journal of Artificial intelligence in Education*, **6**(2/3), 239-302

Crossley, L.G., Anderson, T.R. and Grayson, D.J. (1996) Redox poses difficulties for undergraduate biochemistry students studying oxidative phosphorylation. *Paper presented at the 14th International Conference on Chemical Education*, 14-19 July, Brisbane

Davies, D.R. and Padlan, E.A. (1990) Antibody-Antigen Complexes. *Annual Review of Biochemistry*, **59**, 439-473

Dean, R.S. and Enemoh, P.A.C. (1983) Pictorial organization in prose learning. *Contemporary Educational Psychology*, **8**, 20-27

de Jong, O. (1997) Protocol analysis as a fruitful tool for problem posing teacher education. *European Journal of Teacher Education*, **20**(2), 191-201

de Lange, R.W. (1999) Typographic and picture variables for web-based instructional material. *Paper presented at the Technology in Interactive Education Conference*, 27-29 January, Durban

Denis, M. (1989) *Image and cognition*, New York: Harvester Wheatsheaf, pp. 155-168

Dickey, E. M. (1993) The golden ratio: a golden opportunity to investigate multiple representations of a problem. *The Mathematics Teacher,* **86**, 554-557

Do, E.Y.-L. and Gross, M.D. (2001) Thinking with diagrams in architectural design. *Artificial intelligence Review*, **15**(1-2), 135-149

Driver, R. (1989) Students' conceptions and the learning of science. *International Journal of Science Education,* **11**, 481-190

Duchastel, P.C. (1978) Illustrating instructional texts. *Educational Technology*, **18**(11), 36-39

Duchastel, P.C. (1988) Display and interaction features of instructional texts and computers. *British Journal of Educational Technology*, **19**(1), 58-65

Duchastel, P. and Waller, R. (1979) Pictorial illustration in instructional texts. *Educational Technology*, **19**, 20-25

Dufresne, R.J., Gerace, W.J. and Leonard, W.J. (1997) Solving physics problems with multiple representations. *The Physics Teacher*, **35**, 270-275

du Plessis, L., Anderson, T.R. and Grayson, D.J. (2003) Student difficulties with the use of arrow symbolism in biological diagrams. In: J. Lewis, A. Magro and L. Simonneaux (Eds.), *Biology Education for the Real World: Student-Teacher-Citizen*, Toulouse: Ecole Nationale de Formation Agronomique, pp. 89-103

Dwyer, F.M. (1967) The relative effectiveness of varied visual illustrations in complementing programed instruction. *The Journal of Experimental Education*, **36**(2), 34-42

Dwyer, F.M. (1968) The effectiveness of visual illustrations used to complement programed instruction. *The Journal of Psychology*, **70**, 157-162

Dwyer, F.M. (1969) Student perception of the instructional value of visual illustration. *Medical and Biological Illustration*, **19**, 42-45

Dwyer, F.M. (1970) Exploratory studies in the effectiveness of visual illustrations. *Audio Visual Communication Review*, **18**(3), 235-249

Dwyer, F.M. (1972) The effect of overt responses in improving visually programed science instruction. *Journal of Research in Science Teaching*, **9**(1), 47-55

Dwyer, F.M. (1975) On visualized instruction effect of students' entering behaviour. *The Journal of Experimental Education*, **43**(3), 78-83

Egan, D.E. and Schwartz, B.J. (1979) Chunking in recall of symbolic drawings. *Memory & Cognition*, **7**(2), 149-158

Fischer, E (1894). Einfluss der configuration auf die wirkung derenzyme. *Ber. Dt. Chem. Ges.* **27**, 2985-2993

Fisher, K.M. (1985) A misconception in biology: amino acids and translation. *Journal of Research in Science Teaching*, **22**(1), 53-62

Fleming, M. (1967) Classification and Analysis of Instructional Illustrations. *AV Communication Review*, **15**(3), 246-258

Fleming, M.L. (1977) The picture in your mind. *AV Communication Review*, **25**(1), 43-62

Flores, F., Tovar, M.E. and Gallegos, L. (2003) Representation of the cell and its processes in high school students: an integrated view. *International Journal of Science Education*, **25**(2), 269-286

Fry, E. (1981) Graphical literacy. *Journal of Reading*, **24**, 383-390

Gabel, D. and Sherwood, R. (1980) The effect of student manipulation of molecular models on chemistry achievement according to Piagetian level. *Journal of Research in Science Teaching*, **17**(1), 75-81

Galili, I., Bendall, S. and Goldberg, F. (1993) The effects of prior knowledge and instruction on understanding image formation. *Journal of Research in Science Teaching*, **30**(3), 271-301

Gall, M.D., Borg, W.R. and Gall, J.P. (1996) *Educational research: an introduction, 6th ed.*, New York: Longman

Garnett, P.J., Garnett, P.J. and Hackling, M.W. (1995) Students' alternative conceptions in chemistry: a review of research implications for teaching and learning. *Studies in Science Education*, **25**, 69-95

Garrett, R.H. and Grisham, C.M. (1995) *Biochemistry*, Fort Worth: Saunders College

Gillespie, C.S. (1993) Reading graphic displays: what teachers should know. *Journal of Reading*, **36**(5), 350-354

Glasgow, J. (2001) School of Computing, Queen's University, *Personal Communication*

Glenberg, A.M. and Langston, W.E. (1992) Comprehension of illustrated text: pictures help to build mental models. *Journal of Memory and Language*, **31**, 129-151

Glenberg, A.M. and McDaniel, M.A. (1992) Mental models, pictures, and text: integration of spatial and verbal information. *Memory & Cognition*, **20**(5), 458-460

Glynn, S. (1997) Drawing Mental Models. *The Science Teacher*, **64**(1), 30-32

Gobert, J.D. and Clement, J.J. (1999) Effects of student-generated diagrams versus student-generated summaries on conceptual understanding of causal and dynamic knowledge in plate tectonics. *Journal of Research in Science Teaching*, **36**(1), 39-53

Goldman, S.R. (2003) Learning in complex domains: when and why do multiple representations help? *Learning and Instruction*, **13**, 239-244

Gordin, D.N., Polman, J.L. and Pea, R.D. (1994) The climate visualizer: sense-making through scientific visualization. *Journal of Science Education and Technology*, **3**(4), 203-226

Grayson, D.J. (1995) Science education research and implications for university science instruction. *South African Journal of Science*, **91**, 168-172

Grayson, D.J. (2004) A teaching strategy for helping students disentangle related physics concepts. *American Journal of Physics*, **72**(8), 1126-1133

Grayson, D.J., Anderson, T.R. and Crossley, L.G. (2001) A four-level framework for identifying and classifying students conceptual and reasoning difficulties. *International Journal of Science Education*, **23**(6), 611-622

Griffiths, A.K. and Grant, B.A.C. (1985) High school students' understanding of food webs: identification of a learning hierarchy and related misconceptions. *Journal of Research in Science Teaching*, **22**(5) 421-436

Gropper, G.L. (1963) Why is a picture worth a thousand words? *AV Communication Review*, **11**(4), 75-95

Gupthar, A.S. (2002) School of Molecular and Cellular Biosciences, University of KwaZulu-Natal, *Personal Communication*

Guri-Rozenblit, S. (1988) The interrelations between diagrammatic representations and verbal explanations in learning from social science texts. *Instructional Science*, **17**, 219-234

Hames, B.D. and Hooper, N.M. (2000) *Instant Notes Biochemsitry, 2nd ed.*, Oxford: BIOS Scientific Publishers

Hardin, P. (1993) A theoretical framework for diagrams and information. : *Visual literacy in the digital age: selected readings from the annual conference of the 25th international visual literacy association*, October 13-17, New York, pp. 20-25

Harris, L. J., Larson, S. B., Hasel, K. W. and McPherson, A. (1997) Refined structure of an intact IgG2a monoclonal antibody. *Biochemistry*, **36**(7), 1581-1597

Harrison, A.G., Grayson, D.J. and Treagust, D.F. (1999) Investigating a grade 11 student's evolving conceptions of heat and temperature. *Journal of Research in Science Teaching*, **36**(1), 55-87

Hartley, J. (1990) Textbook design: current status and future research. *International Journal of Educational Research*, **14**(6), 533-541

Hasweh, M. (1988) Descriptive studies of students' conceptions in science. *Journal of Research in Science Teaching*, **25**, 121-134

Hayes, D.A. and Readence, J.E. (1983) Transfer of learning from illustration-dependent text. *Journal of Educational Research*, **76**(4), 245-248

Hegarty, M. (1992) Mental animation: inferring motion from static displays of mechanical systems. *Journal of Experimental Psychology: Learning, Memory and Cognition*, **18**(5), 1084-1102

Hegarty, M. (2004) Dynamic visualizations and learning: getting to the difficult questions. *Learning and Instruction*, **14**, 343-351

Henderson, G. (1999) Learning with diagrams. *Australian Science Teachers' Journal*, **45**(2), 17-25

Hill, D.M. (1988) Difficulties with diagrams. *Journal of Science and Mathematics Education in South-East Asia*, **11**(2), 32-40

Hoffmann, R. and Laszlo, P. (1991) Representation in chemistry. *Angewandte Chemie: International Edition in English*, **30**(1), 1-16

Holliday, W.G. (1973) Critical analysis of pictorial research related to science education. *Science Education*, **57**(2), 201-214

Holliday, W.G. (1975a) What's in a picture? *The Science Teacher*, **42**(10), 21-22

Holliday, W.G. (1975b) The effects of verbal and adjunct pictorial-verbal information in science instruction. *Journal of Research in Science Teaching*, **12**(1), 77-83

Holliday, W.G. (1976) Teaching verbal chains using flow diagrams and texts. *Audio Visual Communication Review*, **24**(1), 63-78

Holliday, W.G. (1990) Textbook illustrations. *The Science Teacher*, **57**(9), 27-29

Holliday, W.G. and Harvey, D.A. (1976) Adjunct labeled drawings in teaching physics to junior high school students. *Journal of Research in Science Teaching*, **13**(1), 37-43

Holliday, W.G., Brunner, L.L. and Donais, E.L. (1977) Differential cognitive and affective responses to flow diagrams in science. *Journal of Research in Science Teaching*, **14**(2), 129-138

Huddle, P.A. and Pillay, A.E. (1996) An in-depth study of misconceptions in stoichiometry and chemical equilibrium at a South African university. *Journal of Research in Science Teaching*, **33**(1), 65-77

Hull, T.L. (2003) Students' use of diagrams for the visualisation of biochemical processes. *Unpublished MSc. Thesis*, University of Natal, South Africa

Hull, T.L., Anderson, T.R. and Grayson, D.J. (2002) Student difficulties with a diagram of the complement pathways in the immune system. *Proceedings of the Tenth Annual Conference of the Southern African Association for Research in Mathematics, Science and Technology Education*, (C. Malcolm and C. Lubisi, Eds.), pp. III-129-III-134

Hurt, J.A. (1987) Assessing functional effectiveness of pictorial representations used in text. *Educational Communication and Technology Journal*, **35**(2), 85-94

Jackson, J.G. (2000) School of Molecular and Cellular Biosciences, University of KwaZulu-Natal, *Personal Communication*

Janeway, C. (2001) Department of Immunobiology, Yale University School of Medicine, *Personal Communication*

Johnson-Laird, P.N. (1983) *Mental models: towards a cognitive science of language, inference, and consciousness*, Cambridge: Cambridge University Press

Johsua, S. (1984) Students' interpretation of simple electrical diagrams. *European Journal of Science Education*, **6**(3), 271-275

Jones, B.L. and Lynch, P.P. (1987) Childrens' concepts of earth, sun and moon. *International Journal of Science Education*, **9**(1), 43-53

Jonnassen, D.H. and Hawk, P. (1984) Using graphic organizers in instruction. *Information Design Journal*, **4**(1), 58-68

Joseph, J.H. and Dwyer, F.M. (1984) The effects of prior knowledge, presentation mode, and visual realism on student achievement. *Journal of Experimental Education*, **52**(2), 110-121

Justi, R.S. and Gilbert, J.K. (2002) Modelling, teachers' views on the nature of modelling, and implications for the education of modellers. *International Journal of Science Education*, **24**(4), 369-387

Kedzierski, M. (1992) Vaccines & Immunisation. *New Scientist*, **8**, 1-4

Kindfield, A.C.H. (1992) In: Narayanan, N.H. and H. Simon (Cochairs), *Reasoning with Diagrammatic Representations*, Technical Report SS-92-02 from the 1992 Spring Symposium, Menlo Park: AAAI Press, pp. 41-46

Kindfield, A.C.H. (1993/1994) Biology diagrams: tools to think with. *The Journal of the Learning Sciences*, **3**(1), 1-36

Knight, D. (2003) 'Exalting understanding without depressing imagination': depicting chemical process. *HYLE: International Journal for Philosophy of Chemistry*, **9**(2), 171-189

Koedinger, K.R. and Anderson, J.R. (1990) Abstract planning and perceptual chunks: elements of expertise in geometry. *Cognitive Science*, **14**, 511-550

Koshland, D.E. (1963) Correlation of structure and function of enzyme action. *Science*, **142**, 1533-1541

Kosslyn, S.M. (1981) The medium and the message in mental imagery: a theory. *Psychological Review*, **88**(1), 46-66

Kosslyn, S.M. (1985) Graphics and human information processing: a review of five books. *Journal of the American Statistical Association*, **80**(391), 499-512

Kosslyn, S.M. (1987) Seeing and imagining in the cerebral hemispheres: a computational approach. *Psychological Review*, **94**(2), 148-175

Kosslyn, S.M. (1989) Understanding Charts and Graphs. *Applied Cognitive Psychology*, **3**, 185-226

Kozma, R. (2003) The material features of multiple representations and their cognitive and social affordances for science understanding. *Learning and Instruction*, **13**(2), 205-226

Kozma, R.B. and Russell, J. (1997) Multimedia and understanding: expert and novice responses to different representations of chemical phenomena. *Journal of Research in Science Teaching*, **34**(9), 949-968

Kress, G. and Van Leeuwen, T. (1996). Reading images: the grammar of visual design, London: Routledge

Kuiper, J. (1994) Student ideas of science concepts: alternative frameworks? *International Journal of Science Education*, **16**(3), 279-292

Landry, S.J. (2001) Department of Biochemistry, Tulane University School of Medicine, *Personal Communication*

Larkin, J.H. and Simon, H.A. (1987) Why a diagram is (sometimes) worth ten thousand Words. *Cognitive Science*, **11**, 65-99

Laseau, P. (1980) *Graphic problem solving for architects and builders*, New York: Van Nostrand Reinhold

Lazarowitz, R. and Penso, S. (1992) High school students' difficulties in learning biology Concepts. *Journal of Biological Education*, **26**, 215-223

Lehninger, A.L., Nelson, D.L. and Cox, M.M. (1993) *Principles of Biochemistry, 2nd ed.*, New York: Worth Publishers

Levie, W.H. and Lentz, R. (1982) Effects of text illustrations: a review of research. *Educational Communication and Technology Journal*, **30**(4), 195-232

Levin, J.R., Anglin, G.J. and Carney, R.N. (1987) On empirically validating functions of pictures in prose. In: D.M. Willows and H.A. Houghton (Eds.), *The psychology of illustration, Vol. 1., Basic research*, New York: Springer-Verlag, pp. 51-85

Lewalter, D. (2003) Cognitive strategies for learning from static and dynamic visuals. *Learning and Instruction*, **13**(2), 177-189

Lewis, N.A. (1980) Potential energy diagrams. *Journal of Chemical Education*, **57**(7), 478-483

Lincoln, Y.S. and Guba, E.G. (1985) *Naturalistic inquiry*, Newbury Park: Sage, pp. 39, 202-208, 268-273, 340-344

Lohse, G., Walker, N., Biolsi, K. and Rueter, H. (1991) Classifying graphical information. *Behaviour & Information Technology*, **10**(5), 419-436

Lord, T.R. (1987a) Spatial teaching. *The Science Teacher*, **54**(2), 32-34

Lord, T.R. (1987b) A look at spatial abilities in undergraduate women science majors. *Journal of Research in Science Teaching*, **24**(8), 757-767

Lord, T.R. (1990) Enhancing learning in the life sciences through spatial perception. *Innovative Higher Education*, **15**(1), 5-16

Lowe, R. (1986) The scientific diagram: is it worth a thousand words? *The Australian Science Teachers Journal*, **32**(3), 7-13

Lowe, R.K. (1987) Drawing out ideas: a neglected role for scientific diagrams. *Research in Science Education*, **17**, 56-66

Lowe, R. (1988a) Drawing comparisons: school science and professional science. *The Australian Science Teachers Journal*, **33**(4), 32-39

Lowe, R.K. (1988b) Diagram construction and the improvement of science. *Journal of Science and Mathematics Education in South East Asia*, **11**(2), 22-31

Lowe, R.K. (1989) Search strategies and inference in the exploration of scientific diagrams. *Educational Psychology*, **9**(1), 27-44

Lowe, R. (1991) Expository illustrations: a new challenge for reading instruction. *Australian Journal of Reading*, **14**(3), 215-226

Lowe, R.K. (1993a) Diagrammatic information: techniques for exploring its mental representation and processing. *Information Design Journal*, **7**(1), 3-17

Lowe, R.K. (1993b) Constructing a mental representation from an abstract technical diagram. *Learning and Instruction*, **3**, 157-179

Lowe, R.K. (1994a) Diagram prediction and higher order structures in mental representation. *Research in Science Education*, **24**, 208-216

Lowe, R.K. (1994b) Selectivity in diagrams: reading beyond the lines. *Educational Psychology*, **14**(4), 467-491

Lowe, R.K. (1996) Background knowledge and the construction of a situational representation from a diagram. *European Journal of Psychology of Education*, **11**(4), 377-397

Lowe, R. (1997) How much are pictures worth? *Paper presented at the UniServe Workshop*, 15-16 July, Australia

Lowe, R.K. (1999) Extracting information from an animation during complex visual learning. *European Journal of Psychology of Education*, **14**(2), 225-244

Lowe, R. (2000) Visual literacy and learning in science. *ERIC Digests*, ERIC Database # ED463945,
<www.ericfacility.net/ericdigests/ed463945.html>

Lowe, R.K. (2003) Animation and learning: selective processing of information in dynamic graphics. *Learning and Instruction*, **13**(2), 157-176

Lumer, J., Picard, F. and Hesse, M. (2003) "Proteins have the bases uracil, thymine, adenine, guanine, cytosine" concept mapping as a tool to clarify two biochemical concepts. In: J. Lewis, A. Magro and L. Simonneaux (Eds.), *Biology Education for the Real World: Student-Teacher-Citizen*, Toulouse: Ecole Nationale de Formation Agronomique, pp. 199-209

Macdonald-Ross, M. (1989) Towards a graphic ecology. In: H. Mandl, J.R. Levin (Eds.), *Knowledge Acquisition from Text and Pictures*, Amsterdam: Elsevier, pp. 145-154

Maddox, H. and Loughran, R.J. (1977) Illustrating the lecture: prepared diagrams vs. built-up diagrams. *Audio Visual Communication Review*, **25**(1), 87-90

Marek, E. (1986) Understandings and misunderstandings of biology concepts. *The American Biology Teacher*, **48**(1), 37-40

Martin, A.C.R. (2001) Department of Bioinformatics, University of Reading, *Personal Communication*

Mathews, C.K. and van Holde, K.E. (1990) *Biochemistry*. Redwood City: Benjamin/Cummings

Mayer, R.E. (1989a) Models for understanding. *Review of Educational Research*, **59**(1), 43-64

Mayer, R.E. (1989b) Systematic thinking fostered by illustrations in scientific text. *Journal of Educational Psychology*, **81**(2), 240-246

Mayer. R.E. (1993) Commentary: comprehension of graphics in texts: an overview. *Learning and Instruction*, **3**, 239-245

Mayer, R.E. (1997) Multimedia learning: are we asking the right questions? *Educational Psychologist*, **32**(1), 1-19

Mayer, R.E. (1999) Multimedia aids to problem-solving transfer. *International Journal of Educational Research*, **31**, 611-623

Mayer, R.E. (2003) The promise of multimedia learning: using the same instructional design methods across different media. *Learning and Instruction*, **13**(2), 125-139

Mayer, R.E. and Anderson, R.B. (1991) Animations need narrations: an experimental test of a dual-coding hypothesis. *Journal of Educational Psychology*, **83**(4), 484-490

Mayer, R.E. and Anderson, R.B. (1992) The instructive animation: helping students build connections between words and pictures in multimedia learning. *Journal of Educational Psychology*, **84**(4), 444-452

Mayer, R.E. and Gallini, J. (1990). When is an illustration worth ten thousand words? *Journal of Educational Psychology*, **82**, 715-726

Mayer, R.E and Sims, V.K. (1994) For whom is a picture worth a thousand words? Extensions of a dual-coding theory of multimedia learning. *Journal of Educational Psychology*, **86**(3), 389-401

Mayer, R.E., Steinhoff, K., Bower, G. and Mars, R. (1995) A generative theory of textbook design: using annotated illustrations to foster meaningful learning of science text. *Educational Technology Research and Development*, **43**(1), 31-43

Mayer, R.E., Bove, W., Bryman, A., Mars, R. and Tapangco, L. (1996) When less is more: meaningful learning from visual and verbal summaries of science textbook lessons. *Journal of Educational Psychology*, **88**(1), 64-73

Mbewe, B. (2000) Quaternary structure as defined by whom? *Biochemistry and Molecular Biology Education*, **28**(5), 240-241

McCormick, B.H., DeFanti, T.A. and Brown, M.D. (1987) Visualization in scientific computing - a synopsis. *IEEE Computer Application Graphics*, **7**, 61-70

McIntyre, P.J. and Reed, J.A. (1976) The effect of visual devices based on Bruner's modes of representation on teaching concepts of electrostatics to elementary school children. *Science Education*, **60**(1), 87-94

McKee, T. and McKee, J.R. (1996) *Biochemistry: an introduction*, Boston: WCB

McMillan, J.H. and Schumacher, S. (1993) *Research in education: a conceptual introduction, 3rd ed.*, New York: HarperCollins

Menger, F.M., Zana, R., Lindman, B. (1998) Portraying the structure of micelles. *Journal of Chemical Education*, **75**(1), 115

Moore, P.J. and Skinner, M.J. (1985) The effects of illustrations on children's comprehension of abstract and concrete passages. *Journal of Research in Reading*, **8**(1), 45-56

Moore, P.J., Chan, L.K.S. and Au, W.K. (1993) High school students' use of diagrams during reading. *Journal of Research in Reading*, **16**(1), 57-71

Mousavi, S.Y., Low, R. and Sweller, J. (1995) Reducing cognitive load by mixing auditory and visual presentation modes. *Journal of Educational Psychology*, **87**(2), 319-334

Mouton, J. (2001) *How to succeed in your master's and doctoral studies*, Pretoria: Van Schaik Publishers

Nerdel, C., Prechtl, H. and Bayrhuber, H. (2003) Interactive animations and understanding of biological processes: an empirical investigation of the effectiveness of computer-assisted learning environments in biology instruction. In: J. Lewis, A. Magro and L. Simonneaux (Eds.), *Biology Education for the Real World: Student-Teacher-Citizen*, Toulouse: Ecole Nationale de Formation Agronomique, pp. 45-58

Noh, T. and Scharmann, L.C. (1997) Instructional influence of a molecular-level pictorial presentation of matter on students' conceptions and problem-solving ability. *Journal of Research in Science Teaching*, **34**(2), 199-217

Nottis, K.E.K. and McFarland, J. (2001) A comparative analysis of pre-service teacher analogies generated for process and structure concepts. *Electronic Journal of Science Education*, **5**(4),
<http://unr.edu/homepage/crowther/ejse/knottisetal.html>

Novick, S. and Nussbaum, J. (1978). Junior high school pupils' understanding of the particulate nature of matter: an interview study. *Science Education*, **62**(3), 273-281

Nuñez de Castro, I. and Alonso, F.J. (1997) Energy diagrams for enzyme catalysed reactions: a confusing point in the textbooks. *Biochemical Education*, **25**(2), 87-89

Odom, A.L. (1995) Secondary & college biology students' misconceptions about diffusion & osmosis. *The American Biology Teacher*, **57**(7), 409-415

Olivier, P. (2001). Diagrammatic reasoning: an artificial intelligence perspective. *Artificial Intelligence Review*, **15**(1-2), 63-78

Orgill, M. and Bodner, G.M. (2004) Locks and keys: how analogies are used and perceived in biochemistry classes. *Proceedings of the National Association for Research in Science Teaching (NARST)*, April 1-3, Vancouver, Electronic paper number 203366.

Osborne, R.J. and Wittrock, M.C. (1983) Learning science: a generative process. *Science Education*, **67**(4), 489-508

Paivio, A. (1971) *Imagery and verbal processes*, New York: Holt, Rinehart & Winston

Paivio, A. (1986) *Mental representations: a dual coding approach*, Oxford: Oxford University Press

Pavlinic, S., Buckley, P., Davies, J. and Wright, T. (2001) Computing in Stereochemistry: 2D or 3D Representations? In: H. Behrendt *et al.* (Eds.), *Research in Science Education-Past, Present, and Future*, Kiel: IPN, pp. 295-300

Peña, B.M. and Quílez, M.J.G (2001) The importance of images in astronomy education. *International Journal of Science Education*, **23**(11), 1125-1135

Petre, M. and Green, T.R.G. (1993) Learning to read graphics: some evidence that 'seeing' an information display is an acquired skill. *Journal of Visual Languages and Computing*, **4**, 55-70

Pfundt, H. and Duit, R. (1994) *Bibliography: students' alternative frameworks and science education, 4th ed*, Kiel: Institute for Science Education

Phelps, A. J. (1994) Qualitative methodologies in chemical education research. *Journal of Chemical Education*, **71**(3), 191-194

Piaget, J. (1952) *The origins of intelligence in children*, New York: W. W. Norton

Piez, C. N. and Voxman, M.H. (1997) Multiple representations: using different perspectives to form a clearer picture. *The Mathematics Teacher*, **90**, 164-166

Pincus, S. (2001) Department of Microbiology, Montana State University, *Personal Communication*

Pintó, R. and Ametller, J. (2002) Students' difficulties in reading images. Comparing results from four national research groups. *International Journal of Science Education*, **24**(3), 333-341

Ploetzner, R. and Lowe, R. (2004) Dynamic visualisations and learning. *Learning and Instruction*, **14**, 235-240

Posner, G.J. and Gertzog, W.A. (1982) The clinical interview and the measurement of conceptual change. *Science Education*, **66**(2), 195-209

Pribyl, J. R. and Bodner, G. M. (1987) Spatial ability and its role in organic chemistry: a study of four organic courses. *Journal of Research in Science Teaching*, **24**, 229-240

Pyle, E.J. (1999) The effects of visually enhanced instructional environments on students' conceptual growth. *Electronic Journal of Science Education*, **3**(3), <http://unr.edu/homepage/crowther/ejse/pyle.html>

Ramadas, J. and Nair, U. (1996) The system idea as a tool in understanding conceptions about the digestive system. *International Journal of Science Education*, **18**(3), 355-368

Reid, D. (1990a) The role of pictures in learning biology: part 1, perception and observation. *Journal of Biological Education*, **24**(3), 161-172

Reid, D. (1990b) The role of pictures in learning biology: part 2, picture-text processing. *Journal of Biological Education*, **24**(4), 251-258

Reid, D.J. and Beveridge, M. (1986) Effects of text illustration on children's learning of a school science topic. *British Journal of Educational Psychology*, **56**, 294-303

Reid, D.J. and Beveridge, M. (1990) Reading illustrated science texts: a micro-computer based investigation of children's strategies. *British Journal of Educational Psychology*, **60**, 76-87

Reid, D.J. and Miller, G.J.A. (1980) Pupils' perception of biological pictures and its implications for readability studies of biology textbooks. *Journal of Biological Education*, **14**(1), 59-69

Reid, D.J., Beveridge, M. and Wakefield, P. (1986) The effect of ability, colour and form on children's perceptions of biological pictures. *Educational Psychology*, **6**, 9-18

Reid, D.J., Briggs, N. and Beveridge, M. (1983) The effect of picture upon the readability of a school science topic. *British Journal of Educational Psychology*, **53**, 327-335

Reimann, P. (2003) Multimedia learning: beyond modality. *Learning and Instruction*, **13**, 245-252

Reinking, D. (1986) Integrating graphic aids into content area instruction: the graphic information lesson. *Journal of Reading*, **30**, 146-151

Reiss, M.J. and Tunnicliffe, S.D. (2001) Student' understandings of their internal structure as revealed by drwings, In: H. Behrendt *et al.* (Eds.), *Research in Science Education-Past, Present, and Future*, Kiel: IPN, pp. 101-106

Reiss, M. J., Tunnicliffe, S. D., Andersen, A. M., Bartoszeck, A., Carvalho, G. S., Chen, S.-Y., Jarman, R., Jónsson, S., Manokore, V., Marchenko, N., Mulemwa, J., Novikova, T., Otuka, J., Teppa, S. and van Rooy, W. (2002) An international study of young peoples' drawings of what is inside themselves. *Journal of Biological Education*, **36**(2), 58-64

Richardson, D.C. and Richardson, J.S. (2002) Teaching molecular 3-D literacy. *Biochemistry and Molecular Biology Education*, **30**(1), 21-26

Rigney, J.W. and Lutz, K. A. (1976) Effects of graphic analogies of concepts in chemistry on learning and attitude. *Journal of Educational Psychology*, **68**, 305-311

Ritter, P. (1996) *Biochemistry: a foundation*, Pacific Grove: Brooks/Cole, p. 135, 155, 181

Roitt, I.M. (1997) *Roitt's Essential immunology, 9th ed.*, Oxford: Blackwell Science

Roitt, I. and Delves, P. J. (2001). *Roitt's Essential Immunolgy, 10th ed.*, London: Blackwell Science

Rosnow, R.L. and Rosenthal, R. (1996) *Beginning behavioural research: a conceptual primer, 2nd ed.*, New Jersey: Prentice-Hall, pp. 4-5, 101-103, 118-119

Roth, W.-M. (2002) Reading graphs: contributions to an integrative concept of literacy. *Journal of Curriculum Studies*, **34**(1), 1-24

Roth, W.-M., Bowen, G.M. and McGinn, M.K. (1999) Differences in graph-related practices between high school biology textbooks and scientific ecology journals. *Journal of Research in Science Teaching*, **36**(9), 977-1019

Rubin, H.J. and Rubin, I.S. (1995) *Qualitative Interviewing*, London: Sage

Salomon, G. and Perkins, D.N. (1989) Rocky roads to transfer: rethinking mechanisms of a neglected phenomenon. *Educational Psychologist*, **24**(2), 113-142

Sanders, M. (1993) Erroneous ideas about respiration: the teacher factor. *Journal of Research in Science Teaching*, **30**(8), 919-934

Sanders, M. (1995) The nature and remediation of spatial problems associated with interpreting diagrams of biological sections. *Unpublished PhD thesis*, University of Cape Town, South Africa

Sanders, M. (1998) Improving the quality of science education research by promoting the "rival hypothesis". *Paper presented at the Annual Conference of the National Association for Research in Science Teaching*, 19-22 April, San Diego

Sanders, M. (2001) Spatial ability and success in the sciences: a cross-sectional study of biology scholars. *Proceedings of the Ninth Annual Conference of the Southern African Association for Research in Mathematics, Science and Technology Education, Vol. 1*, (I.V. Mutimucuio, Ed.), pp. 211-223

Sanders, M (2001) Science Education Curriculum Development and Evaluation Research Group, University of the Witwatersrand, *Personal Communication*

Sanders, M. (2002) Secondary school biology learners' difficulties in interpreting diagrams of biological sections. *Proceedings of the Tenth Annual Conference of the Southern African Association for Research in Mathematics, Science and Technology Education* (C. Malcolm and C. Lubisi, Eds.), pp. II-85 – II-94

Sanders, M. and Mokuku, T. (1994) How valid is face validity? *Paper presented at the Second Annual Meeting of the Southern African Association for Research and Development in Mathematics and Science Education*, 27-30 January, Durban

Scaife, M. and Rogers, Y. (1996) External cognition: how do graphical representations work? *International Journal of Human-Computer Studies*, **45**, 185-213

Scanlon, E. (1998) How Beginning Students Use Graphs of Motion. In: M.W. van Someren, P. Reimann, H.P.A. Boshuizen and T. de Jong (Eds.), *Learning with Multiple Representations*, Amsterdam: Elsevier Science, pp. 67-86

Schnotz, W. (1993a) Introduction to special issue on comprehension of graphics in texts. *Learning and Instruction*, **3**, 151-155

Schnotz, W. (1993b) Some remarks on the commentary: on the relation of dual coding and mental models in graphics comprehension. *Learning and Instruction*, **3**, 247-249

Schnotz, W. and Lowe, R. (2003) Introduction: external and internal representations in multimedia learning. *Learning and Instruction*, **13**(2), 117-123

Schollum, B. (1983) Arrows in science diagrams: help or hindrance for pupils? *Research in Science Education*, **13**, 45-59

Seddon, G.M. and Shubber, K.E. (1984) The effects of presentation mode and colour in teaching the visualisation of rotation in diagrams of molecular structures. *Research in Science & Technological Education*, **2**(2), 167-176

Seels, B.A. (1994) Visual Literacy: The Definition Problem. In: D.M Moore F.M. Dwyer (Eds.), *Visual literacy: a spectrum of visual learning*, New Jersey: Educational technology publications:, pp. 97-112

Seufert, T. (2003) Supporting coherence formation in learning from multiple representations. *Learning and Instruction*, **13**(2), 227-237

Shimojima, A. (2001) The graphic-linguistic distinction. *Artificial Intelligence Review*, **15**(1-2), 5-27

Shubbar, K.E. (1990) Learning the visualisation of rotations in diagrams of three dimensional structures. *Research in Science & Technological Education*, **8**(2), 145-154

Shulman, L. (1986) Those who understand: knowledge growth in teaching. *Educational Researcher*, **15**(2), 4-14

Silverton, E.W., Navia, M.A., Davies, D.R. (1977) Three-dimensional structure of an intact human immunoglobulin. *Proceedings of the National Academy of Sciences*, **74**(11), 5140-5144

Simonneaux, L. (2000) A study of pupils' conceptions and reasoning in connection with 'microbes', as a contribution to research in biotechnology education. *International Journal of Science Education*, **22**(6), 619-644

Smith, T. (2001) Department of Biological Sciences, Purdue University, *Personal Communication*

Soyibo, K. (1994) Misleading labellings in biology textbook drawings. *Australian Science Teachers Journal*, **40**(2), 10-14

Spoehr, K.T. and Lehmkuhle, S.W. (1982) *Visual information processing*, San Francisco: W.H. Freeman

Stahly, L.L., Krockover, G.H. and Shepardson, D.P. (1999) Third grade students' ideas about the lunar phases. *Journal of Research in Science Teaching*, **36**(2), 159-177

Stenning, K. and Lemon, O. (2001) Aligning logical and psychological perspectives on diagrammatic reasoning. *Artificial intelligence Review*, **15**(1-2), 29-62

Stenning, K. and Oberlander, J. (1995) A cognitive theory of graphical and linguistic reasoning: logic and implementation. *Cognitive Science*, **19**, 97-140

Stewart, A.M. (1981) The use of print in improving learning, In: C.F.A. Bryce (Ed.), *Biochemical education*, London: Croom Helm, pp. 156-168

Stryer, L. (1995) *Biochemistry, 4th ed.*, New York: W.H. Freeman

Stylianidou, F., Ormerod, F. and Ogborn, J. (2002) Analysis of science textbook pictures about energy and pupils' readings of them. *International Journal of Science Education*, **24**(3), 257-283

Sumfleth, E. and Telgenbüscher, L. (2001) Improving the use of instructional illustrations in learning chemistry, In: H. Behrendt *et al.* (Eds.), *Research in Science Education-Past, Present, and Future*, Kiel: IPN, pp. 289-294

Szabo, M., Dwyer, F.M. and DeMelo, H. (1981) Visual testing: visual literacy's second dimension. *Educational Communication and Technology Journal*, **29**(3), 177-187

Szlichcinski, K.P. (1979) Diagrams and illustrations as aids to problem solving. *Instructional Science*, **8**, 253-274

Taconis, R., Ferguson-Hessler, M.G.M. and Boekkamp, H. (2001) Teaching science problem solving: an overview of experimental work. *Journal of Research in Science Teaching*, **38**(4), 442-468

Talbot, C. (2001) Some common biochemical mistakes. *School Science Review*, **82**(301), 91-92

Tamir, P. (1985) Content analysis focusing on inquiry. *Journal of Curriculum Studies*, **17**(1), 87-94

Tierney, R.J., Readence, J.E. and Dishner, E.K. (1990) *Reading strategies and practices: a compendium, 3rd ed.*, Boston: Allyn and Bacon

Treagust, D. F. (1988) Development and use of diagnostic tests to evaluate students' misconceptions in science. *International Journal of Science Education*, **10** (2), 159-169

Treagust, D.F., Chittleborough, G. and Mamiala, T.L. (2002) Students' understanding of the role of scientific models in learning science. *International Journal of Science Education*, **24**(4), 357-368

Treagust, D.F., Duit, R. and Fraser, B.J. (1996) Overview: Research on students' pre-instructional conceptions - the driving force for improving teaching and learning in science and mathematics. In: D.F. Treagust, R. Duit and B.J. Fraser (Eds.), *Improving Teaching and Learning in Science and Mathematics,* New York: Teachers College Press, pp. 1-14

Treptow, R.S. (1980) Le Chatelier's principle: a reexamination and method of graphic illustration. *Journal of Chemical Education*, **57**(6), 417-420

Tuckey, H. and Selvaratnam, M. (1993) Studies involving three-dimensional visualisation skills in chemistry: a review. *Studies in Science Education*, **21**, 99-121

Valentine, R. C. and Green, N. M. (1967) Electron microscopy of an antibody-hapten complex. *Journal of Molecular Biology*, **27**, 615-617

van Dusen, L.M., Spach, J.D., Brown, B. and Hansen, M. (1999) TRIO: a new measure of visual processing ability. *Educational and Psychological Measurement*, **59**(6), 1030-1046

van Leeuwen, T. and Jewitt, C. (Eds.) (2001) *Handbook of Visual Analysis*, London: Sage

van Someren, M.W., Boshuizen, H.P.A., de Jong, T. and Reimann, P. (1998) Introduction. In: M.W. van Someren, P. Reimann, H.P.A. Boshuizen and T. de Jong (Eds.), *Learning with Multiple Representations*, Amsterdam: Elsevier Science, pp. 1-5

Verma, G.K. and Mallick, K. (1999) *Researching Education: Perspectives and Techniques*, London: Falmer Press

von Aufschnaiter, C. and von Aufschnaiter, S. (2003) Theoretical framework and empirical evidence of students' cognitive processes in three dimensions of content, complexity, and time. *Journal of Research in Science Teaching*, **40**(7), 616-648

von Glasersfeld, E. (1983) Learning as a constructive activity. *Paper presented at the 5th Annual Meeting of the North American Group of PME*, Monteréal,
<http://www.umass.edu/srri/vonGlasersfeld/ onlinePapers/pdf/vonGlasersfeld_080.PDF>

von Glasersfeld, E. (1989) Cognition, construction of knowledge, and teaching. *Synthese*, **80**(1), 121-140

von Glasersfeld, E. (2003) An exposition of constructivism-why some like it radical. *Scientific Reasoning Research Institute*, University of Massachusetts, Unpublished: 8 April 2003,
<http://www.oikos.org/vonen.htm>

Waddill, P.J., McDaniel, M.A. and Einstein, G.O. (1988) Illustrations as adjuncts to prose: a text-appropriate processing approach. *Journal of Educational Psychology*, **80**(4), 457-464

Wandersee, J.H. (1994) Making high-tech micrographs meaningful to the biology student. In: P.J. Fensham, R.F. Gunstone and R.T. White (Eds.), *The Content of Science: A Constructivist Approach to its Teaching and Learning*, London: Falmer, pp. 161-176

Ward, R.E. and Wandersee, J.H. (2002) Struggling to understand abstract science topics: a Roundhouse diagram-based study. *International Journal of Science Education*, **24**(6), 575-591

Weidenmann, .B. (1988) When good pictures fail. An information processing approach to the effect of illustrations. In: H. Mandl, H and J.R. Levin, J.R. (Eds.), *Knowledge Acquisition from Text and Pictures*, Amsterdam: Elsevier, pp. 157-171

Wheeler, A.E. and Hill, D. (1990) Diagram-ease: why students misinterpret diagrams. *The Science Teacher*, **57**(5), 59-63

White, R. and Gunstone, R. (1992) *Probing understanding*, London: Falmer Press, pp. 65-81, 82-95

Winn, W. (1980) The effect of block-word diagrams on the structuring of science concepts as a function of general ability. *Journal of Research in Science Teaching*, **17**(3), 201-211

Winn, W. (1982) The role of diagrammatic representation in learning sequences, identification and classification as a function of verbal and spatial ability. *Journal of Research in Science Teaching*, **19**(1), 79-89

Winn, W. (1987) Charts, graphs and diagrams in educational materials. In: D.M. Willows and H.A. Houghton (Eds.), *The psychology of illustration, Vol. 1., Basic research*, New York: Springer-Verlag, pp. 152-198

Winn, W. (1988) Recall of the pattern, sequence, and names of concepts presented in instructional diagrams. *Journal of Research in Science Teaching*, **25**(5), 375-386

Winn, W. (1991) Learning from Maps and Diagrams. *Educational Psychology Review*, **3**(3), 211-247

Winn, W. (1993) An account of how readers search from information in diagrams. *Contemporary Educational Psychology*, **18**, 162-185

Winn, W. and Solomon, C. (1993) The effect of the spatial arrangement of simple diagrams on the interpretation of English and nonsense sentences. *Educational Technology Research & Development*, **41**(1), 29-41

Winn, W., Li, T.-Z. and Schill, D. (1991) Diagrams as aids to problem solving: their role in facilitating search and computation. *Education Training Research and Development*, **39**(1), 17-29

Wittrock, M.C. (1974) Learning as a generative process. *Educational Psychology*, **11**, 87-95

Yair, Y, Schur, Y. and Mintz, R. (2003) A "thinking journey" to the planets using scientific visualisation technologies: implications to astronomy education. *Journal of Science Education and Technology*, **12**(1), 43-49

Zhang, J. and Norman, D.A. (1994) Representations in distributed cognitive tasks. *Cognitive Science*, **18**, 87-122

RESEARCH PAPERS

A. Schönborn, K.J., Anderson, T.R. and Grayson, D.J. (2002) Student Difficulties with the Interpretation of a Textbook Diagram of Immunoglobulin G (IgG). *Biochemistry and Molecular Biology Education*, **30**(2), 93-97

B. Schönborn, K.J., Anderson, T.R. and Grayson, D.J. (2003) Students' Interpretation of External Representations in Biochemistry. In: D. Fisher & T. Marsh (Eds.), *Making Science, Mathematics and Technology Education Accessible to All, Vol. 1*, Perth: Key Centre for School Science and Mathematics, pp. 423-434

C. Schönborn, K.J., Anderson, T.R. and Grayson, D.J. (2003) Some Cognitive Perspectives Related to Students' Interaction with External Representations in Biochemistry. *Proceedings of the 11th Annual Conference of the Southern African Association for Research in Mathematics, Science and Technology Education*, (B. Putsoa, M. Dlamini, B. Dlamini and V.Kelly, Eds.), Swaziland, 11-15 January, Vol. 1, pp. 464-470

D. Schönborn, K.J., Anderson, T.R. and Grayson, D.J. (2002) Developing and Testing a Model of the Factors Affecting Student Interaction with Scientific Diagrams. *Proceedings of the 10th Annual Conference of the Southern African Association for Research in Mathematics, Science and Technology Education*, (C. Malcolm and C. Lubisi, Eds.), Durban, 22-26 January, Section III, pp. 377-383

E. Anderson, T.R. and Schönborn, K.J. (2003) Education Forum: Do Textbook Diagrams Really Enhance Student Learning? *South African Society of Biochemistry and Molecular Biology News - Special Edition*, 7(1), 12-14

F. Schönborn, K.J., Anderson, T.R. and Grayson, D.J. (2001) Biochemistry Students' Difficulties with the Interpretation of Textbook diagrams. *Proceedings of the 9th Annual Conference of the Southern African Association for Research in Mathematics, Science and Technology Education*, (I. Mutimucuio, Ed.), Maputo, 17-20 January, Vol. 2, pp. 53-61

G. Schönborn, K. and Anderson, T. (2004) Education Forum: The Importance of Visual Literacy in the Education of Biochemists. *South African Society of Biochemistry and Molecular Biology News*, **8**(1), 34-39