Effect of elevated CO_2 concentration on growth, development and postharvest characteristics of sweetcorn (*Zea mays* L. var. *saccharata*)

Dlulisa, Balungile Precious

BSc (Agricultural Science); BSc Honours in Agriculture Science (Plant Breeding)

A dissertation submitted in partial fulfilment of the academic requirements for the degree of
Master of Science in Agriculture (Horticultural Science)

Discipline of Horticultural Science

School of Agricultural, Earth and Environmental Sciences

College of Agriculture, Engineering and Science

University of KwaZulu-Natal

Pietermaritzburg

South Africa

February 2022

Supervisor: **Prof. Bertling I.**

Co-supervisor: **Prof. Clulow A. D.**

Declaration

I, **Dlulisa Balungile Precious** (220077462), hereby declare that:

1. The dissertation submitted by me for the **Master of Science** in **Agriculture** (**Horticultural Science)** degree at the University of KwaZulu-Natal, except where stated otherwise, is of my own independent strength.
2. This dissertation has not been formerly submitted to any degree or examination at any institution of higher education or faculty.
3. I freely grant the copyright of the **Dissertation** in favour of the University of KwaZulu-Natal.
4. This dissertation contains no other person's writing, unless clearly acknowledged as being sourced from other researchers.

a. Their words have been re-scripted nonetheless, the information attributed to them being referenced
b. In the instance where their precise words have been utilised, then their articulation has been illustrated in italics and within the quotation marks and referenced.

5. This dissertation contains neither text, tables or graphics taken from the internet, unless acknowledgements have been specified as such, and the source being detailed in the dissertation as well as in the references.

Signed:

…………09 February 2022……

Dlulisa Balungile Precious (Student) Date

As the candidate's supervisors, we fully agree to the submission of the dissertation:

…………. ……..…09 February 2022…..…

Prof. Isa Bertling (Supervisor) Date

……… ………….. ………09 February 2022……...

Prof. Alistair D. Clulow (Co-Supervisor) Date

Dissertation output

Symposium (Oral presentation)

- Dlulisa B. P.; Bertling I. and Clulow A. D. The impact of elevated CO_2 on growth, development, and reproduction on sweetcorn (*Zea mays* L. var. *saccharata*).

  University of KwaZulu-Natal. College of Agriculture, Engineering and Science ONLINE Postgraduate Research & Innovation Symposium (PRIS) 2021 Symposium. 09-10 December 2021.

  ONLINE Combined Congress, Annual congress of SSSSA, SASHS, & SASCP Symposium. 25-27 January 2022.

General Abstract

Sweetcorn (*Zea mays* L. var. *saccharata*) is one of the world's most consumed crop in the world and, available in different forms, as fresh maize on a cob and as canned food (baby maize and kernels). Sweetcorn is a horticultural vegetable of high nutritional and mineral quality with increasing demand. Alterations in atmospheric greenhouse gases (GHGs), specifically in carbon dioxide (CO_2) has resulted in complex interactive effects on plants, particularly in the photosynthetic process. Most C4 crops, such as maize are particularly affected by the 'fall-out' of global warming, due to increasing temperatures hindering CO_2 uptake due to stomatal closure. Information on the impact of elevated CO_2 [eCO_2] on C4 crops compared with C3 crops is scarce. Since CO_2 enrichment has been reported to have a positive effect on total biomass of crops, sweetcorn plants grown in glasshouses, were exposed to eCO_2. The use of structures, such as greenhouse, open field and open top chambers (OTC) have been used in order to maintain a certain eCO_2 concentration and determine the effects of such eCO_2 on certain crops. In the current study, 'Mycelium CO_2 generator bags' were used to increase the CO_2 concentration in one of the two adjacent glasshouses. The aim of the study was to subject selected sweetcorn hybrid cultivars ('Assegai' and 'STAR7719') to eCO_2 and to record vegetative and reproductive parameters in response to eCO_2. Such variables include growth, development and reproduction attributes. In addition, mineral nutrients of the fruit (kernels), leaf pigment concentrations and phytochemical parameters, such as ascorbic acid, and carotenoid concentrations, total kernel protein and total soluble solids (TSS) were analysed with the aim of recording potential alterations due to eCO_2.

Elevated CO_2 significantly affected plant growth (plant height and leaf number per plant), leaf area was not significantly increased by eCO_2. A shortened time to maturity was recorded in sweetcorn grown under eCO_2 as well as an increased total above fresh and dry mass, total fresh and dry root mass, and an increased cob mass compared with sweetcorn maintained at 430 ppm CO_2. Mineral nutrient and leaf pigment concentrations were also significantly lower under eCO_2. Growing sweetcorn under eCO_2 also positively impacted some of the phytochemical parameters, such as kernel total soluble solids (TSS), and ascorbic acid concentrations, while total protein and fruit carotenoid concentrations were significantly lower. Although sweetcorn plants under eCO_2 were only grown under a difference of 70 ppm more CO_2 than the control, significant differences in morphology and in kernel phytochemical composition were observed. This study, therefore, points

towards major alterations in sweetcorn quality parameters, when plants are grown under only slight eCO_2. It will be important to determine which effect an elevation to higher CO_2 levels will have, as certain model predict a rise to 600-800 ppm by the year 2100.

Acknowledgements

This consolidation would have not been fruitful without the guidance from of my supervisors: Prof. Isa Bertling and Prof. Alistair Clulow for their amazing interest, encouragement, and guidance accompanied by constructive optimism. The interest they had shown to my project, excellent supervision, constantly editing my work with constructive criticism, the excel drive they instilled in me, and their willingness to training me to be successful. I am profoundly grateful for the provision of agrometeorology technical tools used to run this project. Thank you for making sure this project was a success. I wish to express my sincere gratitude to Maize Trust (MT) for generously providing funding for my study. This project would have been hard to implement without their support, I honestly appreciate it.

My deepest gratitude is owed to the following people, the backbone for the success of this research project:

- I am grateful to Mr. Vivek Naiken. You deserve a special thanks for the amazing input you had poured out to my study. Your confidence in this project gave it “THAT THING”! You believed in idea so much that you had given it your all and excelled amazingly in setting up the technical instrumentation in the glasshouses, and your support was incredible. Thank you for making sure that all the agrometeorology technical equipment required for my project were provisioned. I cannot thank you enough.
- To Mr. Thokozani Nkosi, a humble yet funny and contained soul. I would like to thank you for your amazing support through-out the duration of my study. You persistently gave me hope, and your willingness to frequently stretch out your hand for an assistance. Your cheerfulness taught me to never take minor setbacks seriously. Izandla zidlula ikhanda. I am grateful also to my fellow students, soon to be Dr. Bonga Lewis Ngcobo and Mbalenhle Nkonyane for familiarising me with UKZN laboratories, Sydney Ngwenya, Takudzwa Mandizvo and Sharon Migeri as well as Mr. A. Hokwana, Mr. N. Zuma and Mr. Mlotshwa for their constant willingness to assist me with my experimental trials in the glasshouses, we as students are quite fortunate to have such dedicated university staff.

I am profoundly indebted to my father, mother, sisters, and my family and friends for their constant support and prayers to uplift my spirit. Finally, my sincere highest appreciation is owed to the Almighty Creator (Amen-Ra) onguMvelingqangi, Amadlozi, and my maternal and paternal Ancestors, Izandla zidlula ikhanda for your utmost presence in my life!

Dedication

I dedicate this thesis project to my dearest father. You have supported me through it all by making sure that I succeed in academics and in life. Thank you, father for being my dearest daddy, thank you for your love and constant encouragement. With this work, I strongly trust that I have made you profoundly proud.

Abbreviations / Acronyms

aCO_2 ----------------------------- Ambient carbon dioxide

Al ----------------------------- Aluminium

ANOVA --------------------------- Analysis of variance

ATP ---------------------------- Adenosine triphosphate

AT ----------------------------- Air temperature

BSA ----------------------------- Bovine serum albumin

Bt ----------------------------- *Bacillus thuringiensis*

C ----------------------------- Carbon

Ca ----------------------------- Calcium

$CaMgNO_3$ -------------------- Calcium magnesium nitrate

CH_4 ----------------------------- Methane

Cu ----------------------------- Copper

DTE ----------------------------- Days to emergence

DM ----------------------------- Dry mass

eCO_2 ----------------------------- Elevated carbon dioxide

Fe ----------------------------- Iron

FM ----------------------------- Fresh mass

GHGs ----------------------------- Greenhouse gases

H_2O ----------------------------- Water

HPLC ----------------------------- High Performance Liquid Chromatography

K ----------------------------- Potassium

LSD ----------------------------- Least significant difference

Mg ----------------------------- Magnesium

Mn ----------------------------- Manganese

N ----------------------------- Nitrogen

Na ----------------------------- Sodium

N_2O ----------------------------- Nitrous oxide

NADPH ---------------------------- Nicotinamide adenine dinucleotide phosphate

NH_3 ----------------------------- Ammonia

ns ----------------------------- Non-significant

OTC ----------------------------- Open Top Chambers

P ----------------------------- Phosphorus

PAR ----------------------------- Photosynthetically Active Radiation

PEP ----------------------------- Phosphoenolpyruvate

PEPCase ----------------------------- PEP carboxylase

PNUE ----------------------------- Photosynthetic nitrogen use efficiency

pH ----------------------------- Potential of hydrogen

PVC ----------------------------- Polyvinyl chloride

RH ----------------------------- Relative humidity

RuBP ----------------------------- Ribulose-1,5-biphospate

s**; * ----------------------------- Significant

S ----------------------------- Sulphur

SA ----------------------------- South Africa

SED ----------------------------- Standard error difference

SSA ----------------------------- Sub-Saharan Africa

SIS ----------------------------- Small-holder irrigation systems

SRM ----------------------------- Solar radiation monitoring

TNCs ----------------------------- Total non-structural carbohydrates

Tris ----------------------------- Trizma

TSS ---------------------------- Total Soluble Solids

VDA ---------------------------- Vitamin A Deficiency

USA ---------------------------- United States of America

WUE ---------------------------- Water Use Efficiency

Zn ---------------------------- Zinc

Units of measurements

^{o}Brix ---------------------------- Degrees Brix

^{o}C ---------------------------- Degrees Celsius

g ---------------------------- Gram(s)

h ---------------------------- Hour(s)

Kj ---------------------------- Kilojoule(s)

Km ---------------------------- Michaelis constant

L ---------------------------- Litre

mg ---------------------------- Milligram(s)

min ---------------------------- Minute(s)

mL ---------------------------- Millilitre(s)

ppm ---------------------------- Parts per million

s ---------------------------- Second(s)

µbar ---------------------------- Micro-bar(s) = 16^{-6} bars

µg ---------------------------- Micro-gram(s) = 10^{-6} gram

µL ---------------------------- Microlitre(s) = 10^{-6} litre

µmol ---------------------------- Micro-molar

W/m^2 ---------------------------- Watts per meter square

Table of Contents

Contents

List of figures

List of tables

Appendix

General Introduction

1. Background to the dissertation

1.1 Sweetcorn production and worldwide importance

In Sub-Saharan Africa and Latin America, white maize is the most preferred form of maize, *Zea mays*, it is used as a staple food as well as consumed as fresh maize or as maize meal by more than 1.2 billion people. It makes up 30-50% of small household income expenses, particularly in Eastern and southern Africa (Usman *et al.*, 2015). Maize is an important crop produced on many commercial farms, particularly in Free State, but also in small-holder irrigation systems (SIS) in South Africa, specifically in the Eastern Cape Province. Small-holder maize production is, however, not limited to the Eastern Cape Province, but it is also produced KwaZulu-Nata (KZN) in Mjindi, Ndumo, and in the Tugela Ferry Irrigation System (Fanadzo *et al.*, 2010). Small-holder production of maize is also currently recognized in Camperdown and Vryheid in KZN (Fanadzo *et al.*, 2010).

Escalated global temperatures and elevated carbon dioxide (eCO_2) are said to have intense interactive effects on the process of photosynthesis; further, the negative effect of heat stress on plants affects photosynthesis, as, particularly in C4 plants, such as maize, high temperature will result in stomata remaining closed for longer, not allowing optimal CO_2 uptake (Schulze *et al.*, 1975; Berry and Björkman, 1980; Heckathorn *et al.*, 1998, 2002). It is reported that there is minimal research on the impact of eCO_2 on C4 plants compared with research on C3 plants (Bowes, 1996; Drake *et al.*, 1997). Therefore, subjecting sweetcorn to eCO_2 conditions is highly significant to understand the effects on physical and physico-chemical attributes as well the as chemical composition attributed to eCO_2. In addition, eCO_2 may impact perishability of the commodity, because in sweetcorn, the harvested product is still immature, resulting in dehydration after harvest, aggravating loss in fruit mass, flavour, colour, odour, as well as texture (Antoniali and Santos, 2012).

Sweetcorn also referred to as sugar maize, is an important horticultural vegetable widely consumed in fresh, frozen, and processed form (Siddiq and Pascali, 2018). Although sweetcorn is a versatile vegetable, it is necessary that the crop's environmental and cultural requirements are met for producing viable yield (Bender *et al.*, 2013). To produce such yield, sweetcorn requires a considerable amount of nitrogen fertiliser (Teasdale *et al.*, 2008). Compared with white maize, the most-common form of *Zea mays* in South Africa, sweetcorn has certain benefits, such as a shorter growing season and an enhanced cropping index, which increases the producer's costs as well as returns (Fahrurrozi *et al.*, 2016). In Nigeria it has been reported that sweetcorn (*Zea mays* L. var. *saccharata*) is regarded as a fruit vegetable with potential to substantially contribute to food security (Olaoye *et al.*, 2009).

Sweetcorn belongs to the Poaceae family and is a monoecious, annual crop, carrying male (tassels) and female (spadix) inflorescences. Sweetcorn has become the most significant cereal crop in Africa, accounting for 40% of the cereal production in Sub-Saharan Africa (SSA) (FAOSTAT, 2016), and is consumed by a vast number of people in West and Central Africa (Badu-Apraku and Akinwale, 2011), as well as being staple food in many parts of the world. Severe weather conditions, due to climate change, can led to inter-seasonal alterations in maize yield (Lin *et al.,* 2017). In the United States of America (USA), sweetcorn is considered popular food and its acceptability is increasingly scattering around the world (Revilla *et al.*, 2021).

Recently, various studies on sweetcorn uncovered its importance as a crop in current-day Mexico probably about 7000 years ago (Shukla *et al.*, 2013). In terms of maize production, the USA is considered the largest producer globally, followed by Brazil, while China is ranked third (Ragasa *et al.*, 2013). African countries with the highest maize production, are largely situated on the Eastern and southern part of the continent. The production of maize dominates 75% of the cereals produced, with similar outputs from countries such as Kenya (79%), Malawi (89%), Zambia (78%), and Zimbabwe (75%) producing maize. In western part of the continent, Nigeria and Ghana are the largest maize producers, nevertheless, a small percentage (55% and 75%) of the maize produced there is intended for human consumption, as fresh maize and or maize meal.

The majority of maize produced is destined for food per capita consumption (Smale and Jane, 2003). Maize has a high nutritional quality for humans (Barros and Calado, 2014). Nutritionally, dry, white maize contains about 10% protein, 72% starch, and about 4% lipids; and accounts for 1527 KJ/100g of energy density compared with rice and wheat, nevertheless, it is rather low in protein compared with wheat (*Triticum aestivum*) (Dale and Niernberger, 1982; Nuss and Tanumihardjo, 2010). Sweetcorn, however, has a high provitamin A content (Yang *et al.*, 2014). Sweetcorn also serves as an important source of vitamin B, protein, iron, and other minerals (Badu-Apraku and Akinwale, 2011).

In Brazil, maize cultivation for the production of fresh maize is a vital agricultural enterprise, specifically in the northeast region State of Rio Grande do Norte because, it is favoured by consumers over dry grain maize. This makes it a feasible alternative for subsistence maize farmers (Albuquerque *et al.*, 2008; Fritsche-Neto and Silva, 2011). Such fresh maize is consumed in several forms, in Brazil (Fritsche-Neto and Silva, 2011) and in South Africa (Qwabe, 2011) as vegetable, as a side dish, or even canned. Although the consumer price of fresh maize is of higher than that of mature, dry maize (Barros and Calado, 2014), sweetcorn spoils easily, due to its high-water content between (70 and 80%) leading to losses in mass and, thus, the producer needs to quickly supply the commodity to the market (Antoniali and Santos, 2012).

Even though KwaZulu-Natal is not a major maize producing province in South Africa, it has suitable climate to grow and produce sweetcorn in the summer months, when temperature seldom decline below 10^{o}C and annual rainfall rarely below 350 mm (Smith, 2006). In South Africa, commercial maize producers contribute equally to staple and industrial markets (Sihlobo, 2014). Meanwhile, small-holder farmers are responsible for producing sweetcorn primarily for food security and income (Weatherspoon and Reardon, 2003).

2. Rationale for study focus

Reports show that the Earth's surface temperature has increased over last three consecutive decades (IPCC, 2014), triggering higher average temperature compared with the past decades. The phrase "climate change" as defined by the IPCC, (2007) is "a variation in the state of the climate that can be characterized by adjustments in the mean and/or the variability of its properties, and that continue for a prolonged time, usually decades or even longer". According to the United Nations Framework Convention on Climate Change (UNFCCC), climate change is an "alteration of climate that is linked directly or indirectly to human activity that changes the arrangement of the global atmosphere, and that is in addition to natural climate variability observed over comparable time decades" (IPCC, 2007).

Global climate change is characterized by an elevation in global temperature, altering, amongst others, soil moisture content and precipitation patterns. Methane (CH_4) and nitrous oxide (N_2O) emissions from crop cultivation also pose a threat to the ozone layer; therefore, lessening these emissions from agricultural activities is crucial (Forster *et al.*, 2007). This change in gas alteration in the atmosphere is predicted to cause a decline in yield of all food crops and will carry on for the next 50 years (Thomson *et al.*, 2005). On the other hand, climate change is likely to have positive spin-offs for crop production as it is projected to stimulate the productivity of certain crops (Thomson *et al.*, 2005) by means of carbon sequestration into the soil. Such carbon sequestration entails the transfer of atmospheric CO_2 into prolonged global pools, such as marine, pedologic, biotic or geological layers (Lal, 2008).

Numerous investigations have examined the impacts of climate change on maize production and productivity, enforcing the need for adaptation strategies to be encouraged to counter the harmful impacts of climate change (Mulungu and Tembo, 2018; Amadu *et al.,* 2020). Maize yields have been significantly reduced in numerous countries in Sub-Saharan Africa (SSA) by climate change (IPCC, 2014). Consequently, even compared with the top five countries producing maize globally, yields continue to stagnate in SSA at two tons per hectare and 1.5 tons per hectare in Western and the southern parts of Africa (Cairns *et al.,* 2013). Enhancing such low yields could be achieved by the addition of carbon to soil, potentially presenting a means to enhance maize growth and yield.

It is, consequently, important to investigate how sweetcorn cultivars would perform when subjected to elevated CO_2 (eCO_2) concentration, especially under South African conditions, and to investigate whether eCO_2 would reduce sweetcorn as well as determining the effect on of nutritional components of sweetcorn cultivars under such conditions.

Maize especially coloured cultivars, has nutraceutical properties, providing certain health benefits; therefore, consumption of such food can assist in the prevention and treatment of disease (DeFelice, 1994). Although white and yellow maize are currently the most-commonly cultivated forms, the cobs can also bear blue and red maize kernels (Sarna-Saldivar, 2016; ŽIlić, *et al.,* 2012). Such colorful kernels are either consumed fresh, immature fruit, being boiled, roasted, steamed, or as a dry, milled product in staple foods, such as ‘pap’ and ‘uphutu’ in South Africa, but dry maize kernels also are manufactured into or part of porridges, snacks or beverages (Ranum *et al.,* 2014).

3. Research aim

The aim of this study is, hence, to determine growth and development as well as quality and yield parameters of sweetcorn cultivars in controlled environments, elevating ambient CO_2 concentrations using ‘Nature Mycelium CO_2 bags’ and compare these CO_2 to ambient CO_2 conditions.

3.1. Research objectives:

To determine, if there is a possible beneficial effect of eCO_2 on size and external appearance of sweetcorn plants and to analyse the effect of such eCO_2 on the nutritional quality of sweetcorn.

- To determine the effect of eCO_2 on anatomical parameters such as leaf number and size, plant height, days to tasseling, anthesis and silking, fresh cob mass and length, root fresh and dry mass and total biomass (experimental chapter 1).

- To determine the effect of eCO_2 on leaf and fruit phytochemical and mineral nutrient concentration (experimental chapter 2) including:

- Leaf chlorophylls and carotenoid concentration as well as
- Iron, zinc, calcium, and magnesium concentrations of the kernel

- To determine phytochemical components in (experimental chapter 3) such as:

- Antioxidant (ascorbic acid) concentrations in all of the kernel
- Fruit carotenoids and chlorophyll concentrations in all of the kernel
- Total protein concentration in all of the kernel
- Total soluble solids (TSS) concentrations in all of the kernel

4. Methodology

The experiment was carried out at the University of KwaZulu-Natal, Pietermaritzburg, (SA), in a growth room (4 x 3,5m^2) using sweetcorn plant grown from hybrid seeds of cultivar; 'Assegai' and 'STAR 7719' obtained from Starke Ayres Pietermaritzburg, (SA). Each sweetcorn cultivar had one treatment with nine replications in one glasshouse with enhanced CO_2 concentrations due to Because Nature Mycelium CO_2 powered generator bags (Windell Hydroponics.co.za, Cape Town, SA), and nine controls for each cultivar in another glasshouse with ambient CO_2. Plants were cultivated in 30 cm diameter plastic pots filled with locally obtained heavy clay-loam soil. Furthermore, the CO_2 concentration in the two glasshouses was monitored throughout the experiments with agrometeorological instrumentation. Recommended doses of macro-elements (N, P and K) were manually applied, as well as minor and micro-nutrients via the irrigation system. Pest and disease management practices were carried out as required.

Outline of dissertation

Specific objectives in the above-mentioned aim were accomplished and addressed mostly in the listed chapters which are independent of each other though constituting one entity. The chapters of the dissertation are as follows:

1. Introduction to dissertation.
2. Chapter 1: Literature review: Enhancement of the morphology and nutritional quality of vegetable crops through eCO_2 concentration
3. Chapter 2: Diurnal variations in CO_2 concentration and soil moisture in pot-grown sweetcorn (*Zea mays* L. *var. saccharata*)
4. Chapter 3: Effects of artificially elevated CO_2 conditions on the morphology of sweetcorn (*Zea mays* L. var. *saccharata*)
5. Chapter 4: Does elevate CO_2 affect the nutritional quality and leaf pigment parameters of sweetcorn (*Zea mays* L. var. *saccharata*)? Mineral composition of fruit and leaf carotenoid and chlorophylls
6. Chapter 5: The effects of CO_2 on biochemical quality of sweetcorn (*Zea mays* L. var. *saccharata*): Phytochemical parameters
7. Chapter 6: Discussion, conclusion, and recommendations

Literature cited

Albuquerque C. J. B.; Von-Pinho R. G. and Silva R. (2008). Performance of experimental and commercial maize hybrids for green maize production. Ciênc. Agrotechnology. Vol. 32(2): 69-76.

Amadu F. O.; Miller D. C. and McNamara P. E. (2020). Agroforestry as a pathway to agricultural yield impacts in climate-smart agriculture investments: Evidence from southern Malawi. Ecological Economics. Vol. 167: 106443.

Antoniali S.; Santos N. C. B. and Nachiluk K. (2012). Organic green maize: Production and post-harvest. Research & Technology. Vol. 9(2): 1-6. (in Portuguese).

Badu-Apraku B. and Akinwale R. (2011). Identification of early-maturing maize inbred lines based on multiple traits under drought and low N environments for hybrid development and population improvement. Canadian Journal of Plant Science. Vol.: 91: 931–942.

Besada H. and Sewankambo N. (2009). Climate Change in Africa: Adaptation, Mitigation and Governance Challenges. The Centre for International Governance Innovation.

Barros J. F. C. and Calado J. G. A. (2014). Culture of maize. Soil Technology and Cultures, Agricultural Basics and General Agriculture Fundamentals. Évora: University of Évora; (in Portuguese).

Berry J. A. and Björkman, O. (1980). Photosynthetic response and adaptation to temperature in higher plants. Annual Reviews in Plant Physiology Vol. 31: 491–543.

Bender B. R.; Haegele J. W.; Ruffo M. L. and Below F. E. (2013). Nutrient uptakes partitioning and remobilization in modern, transgenic insect protected maize hybrids. Agronomy Journal, Vol. 105: 161-170.

Bowes G. (1996). Photosynthetic responses to changing atmospheric carbon dioxide concentration. In: Baker N (ed) Photosynthesis and the environment. Kluwer, New York, (pp 387).

Brown R. A. and Rosenberg N. J. (1999). Climate change impacts on the potential productivity of maize and winter wheat in their primary United States growing regions. Climate Change. Vol. 41: 73–107.

Brown W. L.; Bressani R.; Glover D. V.; Hallauer A. R.; Johnson V. A. and Qualset C. O., (1988). Quality-protein maize: report of an ad hoc panel of the advisory committee on technology innovation, Board on Science and Technology for International Development, National Research Council, in cooperation with the Board on Agriculture, National Research Council. Washington, D.C.: National Academy Press.

Cairns J. E.; Hellin J.; Sonder K.; Araus J. L; MacRobert J. F.; Thierfelder C. and Prasanna B. M. (2013). Adapting maize production to climate change in sub-Saharan Africa. Food Security. Vol. 5(3): 345-360.

DeFelice S. L. (2017). What is a True Nutraceutical? And – What is the Nature and size of the US Nutraceutical Market? 1994, http://www.fimdelice.org./ pp 2462.htm/ (Accessed December 2017).

Drake B. G.; Gonzalez-Meier M. A. and Long S. P. (1997). More efficient plants: a consequence of rising atmospheric CO_2? Annual Reviews of Plant Physiology. Plant Molecular Biology, (pp 48: 609).

Dale E. and Niernberger F.F. (1982). Economic models of dry maize milling: part I-model design and system specifications. Association of Operative Millers Technology Bull., (pp 3994–3402).

FAOSTAT (2016). Food Balance Sheet, Available: http://faostat.fao.org/site/345/default.aspx.

Fahrurrozi Z. M.; Dwatmadji N. S.; Sigit S. and Muhammad C. (2016). Growth, and yield responses of three sweetcorn (*Zea mays* L. var. *saccharata*) varieties to local-based liquid organic fertilizer. International Journal on Advanced Science Engineering Information Technology. Vol: 6 ISSN: 2088-5334.

Fanadzo M..; Chiduza C. and Mnkeni P. N. S. (2010). Comparative performance of direct seeding and transplanting green maize under farmer management in small scale irrigation: A case study of Zanyokwe, Eastern Cape, South Africa. African Journal of Agricultural Research. (pp. 524-531) available online.

Forster P.; Ramaswamy V.; Artaxo P.; Berntsen T.; Betts R. and Fahey D. (2007). Changes in atmospheric constituents and in radiative forcing. In: Climate change 2007: The Physical Science Basis. Contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change, Cambridge University Press, and (pp129–234).

Fritsche-Neto R., Silva P. S. L. (2011). Selection index for maize cultivars with dual aptitude: Maize and green maize. Bragantia. Vol. 70(4): 781-787.

Heckathorn S. A.; Downs C. A.; Sharkey T. D. and Coleman, J. S. (1998). The small, methionine rich chloroplast heat-shock protein protects photosystem II electron transport during heat stress. Plant Physiology. Vol. 116: 439–444.

Heckathorn S. A.; Ryan S. L.; Baylis J. A.; Wang J. A.; Hamilton E. W. and Cundiff L. (2002). In vivo evidence from an Agrostis stolonifera selection genotype that chloroplast small heat-shock proteins can protect photosystem II during heat stress. Functional Plant Biology. Vol. 29: 933–944.

Inglett G. E. (1970). Kernel structure, composition, and quality. In: Inglett G. E., editor. Maize: culture, processing, products. Westport, Connecticut: Avi Publishing Company, Inc., (pp 123–37).

IPCC. Climate Change (2007). Synthesis Report. Fourth Assessment Report. Valencia: Intergovernmental Panel on Climate Change, IPCC; 2007.

IPCC. Climate Change (2014). Synthesis Report. Fifth Assessment Report. Geneva: Intergovernmental Panel on Climate Change, IPCC; 2014.

Kimball Bruce A. (1993). Ecology of Crops in Changing CO_2 Concentration. Journal of Agricultural Meteorology. Vol. 48 (5): 559 – 566.

Lal R. (2008). Carbon sequestration. Philosophical Transactions of the Royal Society B. Vol. 363: 815-830.

Lara M. V. and Andreo C. S. (2011). C4 Plants Adaptation to High Levels of CO_2 and to Drought Environments. Abiotic Stress in Plants – Mechanisms and Adaptations, Prof. Arun Shanker (Ed.), ISBN: 978-953-307-394-1, In Tech, Available from http://www.intechopen.com/books/abiotic-stress-in-plantsmechanisms-and-adaptations/c4-plants-adaptation-to-high-levels-of-co2-and-to-drought-environments.

Lin E.; Xiong W.; Ju H.; Xu Y.; Li Y.; Bai L. and Xie L. (2005). Climate change impacts on crop yield and quality with CO_2 fertilization in China. Philosophical Transaction B. The Royal Society Publishing London Book of Biology Science. Vol. 360 (1463): 2149–2154.

Lin Y.; Feng Z.; Wu W.; Yang Y.; Zhou Y. and Xu C. (2017). Potential Impacts of Climate Change and Adaptation on Maize in Northeast China. Agronomy Journal. Vol. 109: 1476-1490.

Mertz E. T. (1970). Nutritive value of maize and its products. In: Inglett GE, editor. Maize: culture, processing, products. Westport, Connecticut: Avi Publishing Company, Inc., (pp 350–9).

Mulenga B. P.; Wineman A. and Sitko N. J. (2017). Climate trends and farmers' perceptions of climate change in Zambia. Environmental Management. Vol. 59: 291-306.

Müller C.; Cramer W.; Hare W. L. and Lotze-Campen H. (2011). Climate change risks for African agriculture. Proceedings of the National Academy of Sciences. Vol. 108: 4313-4315.

Mulungu K. and Tembo G. (2018). Effects of weather variability on crop abandonment. Sustainability. Vol. 7: 2858-2870.

Nuss E.T. and Tanumihardjo S. A. (2010). Maize: A paramount staple crop in the context of global nutrition. Comprehensive Reviews in Food Science Food Safety. Vol. 9: 417-436.

OECD (2006), "Section 3 - Maize (*Zea mays* subsp. *mays*)", in Safety Assessment of Transgenic Organisms, Volume 1: OECD Consensus Documents, OECD Publishing, Paris.

Olaoye G.; Bello O. B.; Ajani A. K. and Ademuwagun T. K. (2009). Breeding for improved organoleptic and nutritionally acceptable green maize varieties by crossing sweetcorn (*Zea* *mays* L. var. *saccharata*): changes in quantitative and qualitative characteristics in F1 hybrids and F2 population. Journal of Plant Breeding and Crop Science. Vol. 1: 298-305.

Parry M. L.; Rosenzweig C.; Iglesias A.; Livermore M. and Fischer G. (2004) Effects of climate change on global food production under SRES emissions and socio-economic scenarios. Global Environmental Change. Vol. 14: 53–67.

Phillips D. L.; Lee J. J. and Dodson R. F. (1996). Sensitivity of the US maize belt to climate change and elevated CO_2. 1. Maize and soybean yields. Agriculture Systems Vol. 52: 481–502.

Ragasa C. and Dankyi A. Acheampong P. Wiredu A. N., Chapo-to A. Asamoah M., *et al.* Patterns of Adoption of Improved Maize Technologies in Ghana. International Food Policy Research Institute, Working paper 36; 2013.

Ranum P.; Pena-Rosas J. P. and Garcia-Casal, M. N. 2014. Global maize production, utilisation, and consumption. Annals of the New York Academy of Science, (pp 105-112).

Revilla P.; Anibas C. M. and Tracy W. F. Sweet maize research around the world 2015–2020. Agronomy 2021, 11, 534.

Schulze E. D.; Lange O. L.; Kappen L.; Evenari M. and Buschbom U. (1975). The role of air humidity and leaf temperature in controlling stomatal resistance of Prunus armeniaca L. under desert conditions. II. The significance of leaf water status and internal carbon dioxide concentration. Oecologia. Vol. 17: 159-70.

Serna-Saldivar S. O. Cereal Grains: Properties, Processing, and Nutritional Attributes; CRC Press (Taylor & Francis Group): Boca Raton, FL, 2016.

Sihlobo W. (2014). South African maize market structure and East African export opportunities under the spotlight. SA Graan/Grain August 2014, (pp 88–89). Available at http://www.grainsa.co.za. [accessed 19 June 2016].

Siddiq M. and Pascali M. A. (2018). Peas, sweet maize, and green beans. Handbook of vegetables and vegetable processing 2nd edition, (pp 761–783).

Smale M. and Thom J. Maize in Eastern and Southern Africa: “Seeds” of Success in Retrospect, (2003). EPTD discussion paper no. 97.

Smith B. (2006). The farming handbook. Pietermaritzburg: University of KwaZulu-Natal Press.

Shiferaw B.; Prasanna B.; Hellin J. and Banziger M. (2011). Crops that feed the world 6. Past successes and future challenges to the role played by maize in global food security. Food Security. Vol. 3: 307–27.

Shukla G. N.; Kumar A.; Jha A.; Singh N.; Sharma P. and Singh J. *et al.*, FICCI PwC knowledge Report: Maize Vision 2022; 2013.

Tesfaye K.; Gbegbelegbe S.; Cairns J. E.; Shiferaw B.; Prasanna B. M. and Sonder K., *et al.* (2015). Maize systems under climate change in sub-Saharan Africa: Potential impacts on production and food security. International Journal of Climate Change Strategies and Management, (pp 247-271).

Teasdale J. R.; Abdul-Baki A. A. and Park Y. B., (2008). Agronomy for Sustainable Development. Vol. 28; 559-565.

Thomson A. M.; Brown R. A.; Rosenberg N. J.; Izaurralde R. C. and Benson V. (2005). Climate change impacts for the conterminous USA: an integrated assessment. Part 3. Dryland production of grain and forage crops. Climate Change. Vol. 69: 43–65.

[USDA]. United States Department of Agriculture. (2009). National nutrient database for standard reference. Available from: http://www.nal.usda.gov/fnic/foodcomp/search/. Accessed September and October 2009.

Usman J.; Zalkuwi J.; Bakari U. M. and Hamman M. (2015). Economics of white maize production in future local government area of Adamawa State, Nigeria, International Journal of Scientific Research and Management. Vol. 3(2): 2159-2166.

van Averbeke W.; Ralivhesa K.; Mbuli S.; Khosa T. B. and Manyelo K. W. (August 2013). Growing Green Maize on Canal Schemes in Vhembe: Production Guidelines. WRC Report No. TT 567/13.

Weatherspoon D. and Reardon T. (2003). The rise of supermarkets in Africa: implications for agri-food systems and the rural poor. Development Policy Review. Vol. 21: 333–356.

Windell Hydroponics.co.za, www.hydroponics.co.za.

Young K. J. and Long S. P. (2000). Crop ecosystem responses to climatic change: maize and sorghum. In: Reddy KR & Hodges HF (Eds.), Climate change and global crop productivity, CABI International, Oxon, United Kingdom, (pp. 107-131).

ŽIlić S.; Serpen A.; AkıllıoğLu G.; GöKmen, V. and VančEtović J. (2012). Phenolic Compounds, Carotenoids, Anthocyanins, and Antioxidant Capacity of Coloured Maize (*Zea mays* L.) Kernels. Journal of Agriculture Food Chemistry. Vol. 60(5): 1224–1231.

Chapter 1

Enhancement of the morphology and nutritional quality of vegetable crops through eCO_2 concentration

1.1 Introduction

An elevated CO_2 concentration is likely to enhance leaf and fruit size (Thompson *et al.*, 2005) and, therefore, is likely to alter the appearance of any horticultural crop. Sweetcorn is used in this study as one of the world's favourite vegetable, hence, this chapter discusses studies that have been undertaken on dry grain maize grown under eCO_2. Unfortunately, little literature is available on sweetcorn, since it is a fresh, horticultural crop, therefore, the present study is meant to fill this gap. This study proposes to practically grow sweetcorn cultivars in glasshouses under elevated CO_2 (eCO_2) and ambient CO_2 (aCO_2) concentrations, and to evaluate the response of sweetcorn to eCO_2 concentrations.

Both, C3 and C4 crop species are directly affected by the currently escalating levels of atmospheric CO_2, a condition that ultimately alters morphological features of leaves and their chemistry, thereby also affecting the plant's carbon to nitrogen balance. This phenomenon has been described in C3 agronomical crops, such as tomato (Islam *et al.*, 1994; Behboudian and Tod, 1995; Li *et al.*, 2007) and potato (De Temmerman *et al.*, 2002; Högy and Fangmeier, 2009). The accumulation of CO_2 in the atmosphere is said to have already reached about 550 ppm in certain places of the planet from the 330ppm measured in the middle of the 20th century and is expected to exceed 700 ppm towards the end of the 21st century (IPPC, 2007), with 650~1200 ppm CO_2 by 2100, which could possibly contribute to a global temperature rise of 2 to 4ºC by that time (Lin *et al.*, 2005). Such environmental alterations are likely to be taken advantage of by plants improving their growth and development, and, therefore, plant production and productivity of major horticultural crops. This is likely to have a positive spin-off effect on food security.

Carbon dioxide in C4 plants is captured in minimal quantity during photosynthesis in an outer layer of mesophyll (M) compartment through the activity of phosphoenolpyruvate (PEP) carboxylase (PEPCase), and subsequently concentrates it into an internal compartment, usually a layer of cells situated around the leaf vascular called the bundle sheath (Edwards and Walker, 1983; Hatch, 1987). A rise in the atmospheric CO_2 increases the rate of photosynthesis by reducing photorespiration which is encouraged by rising temperature in C3 plants than in C4 plants (Sage and Monson, 1999); thus, relatively few studies have been conducted to investigate the growth of C4 plants under eCO_2 conditions and the response of C4 plants to

eCO_2 concentrations (Poorter, 1993; Poorter, Roumet and Campbell, 1996; Wand *et al.,* 1999). Carbon sequestration by crops could be viewed as means to reduce greenhouse gas (GHGs), while conserving carbon in the soil. It has been postulated that the impact of climate change will result in a decline of 0.5% in universal food production from year 2020, and a further 2.3% decline is expected by 2050 (Ainsworth and Long, 2005; Calzadilla *et al.,* 2003).

This predicament, therefore, requires rapidly expanding the germplasm to include selection of major crops accustomed to increased temperature, CO_2 concentration and drought. Such cultivars should be able to withstand the environmental stresses brought by climate change, while still providing high yields. Developing such cultivars would strengthen the germplasm of sweetcorn, and subsequently alleviate escalating food insecurity. Therefore, a great need to grow agricultural crops is of a particular interest mainly due to the strong fears for the future food security, and the significance of exploring the potential advantage strongly associated with rising CO_2 gas, which is said to stimulate plant growth (FAO, 2016).

South Africa has been identified as a main producer of GHGs, liable for generating 65% of the entire continent's emissions (SA - MDGCR, 2013). Consequently, it is highly likely that crop production is impacted by the escalating eCO_2, and, thus, global warming effects on crop production become prevalent. Irregular, global climate change constantly alters the 'usual' weather patterns and results in abnormal, often catastrophic, weather conditions. This occurrence has been aligned with the industrially driven production systems that contribute to the accumulation in GHGs CO_2, in the atmosphere (IPCC, 2014), subsequently resulting in the accumulation of these compound in the atmosphere.

The United Nations Framework Convention on Climate Change (UNFCCC) was established with the purpose of initiating discussions on how to reduce concentration of greenhouse gases (GHGs) in the atmosphere at a level that would prevent negative, anthropogenic effects on the atmospheric gas composition (United Nation, 1992). An increase in eCO_2 to a certain level encourages photosynthesis CO_2 compensation points for C3 crops which is 13-26 ppm (Yeoh *et al.,* 1980). Ghannoum *et al.* (2000) reported a yield increase in C3 crops, while the impact might not be so prominent in C4 crops. Whereas Bauer and Martha (1981) investigated a CO_2 compensation point for major C3 crops (re-examination I) and found that compensation points for spinach (*Spinacia oleracea* L.) was 35.5 ppm, rice (*Triticum aestivum* L.) was 34.5 ppm, and sunflower (*Helianthus annuus* L.) was 34.3 ppm, respectively.

In C4 plants the enzyme Rubisco is responsible for the first step of fixing CO_2, but it is not operating at optimal gas levels under the current atmospheric conditions (Andrews and

Lorimer, 1987; Tcherkez *et al.*, 2006). Photosynthesis in C4 plant crops, first captures CO_2 at in in minimal concentration in an outer layer mesophyll (M) compartment through of phosphoenolpyruvate (PEP) carboxylase (PEPCase) activity, and consequently concentrate it into an internal partition, typically a layer of cells around the leaf vasculature called the bundle sheath (Edwards and Walker, 1983; Hatch, 1987). Photorespiration involves the loss of fixed carbon in a form of CO_2 in plants in the presence of light. Photorespiration is initiated in plant chloroplasts and during this process, energies in a form of ATP or NADPH are not produced which is considered a wasteful process. This process takes place during higher accumulation of oxygen (O_2). Photorespiration occurs by a biochemical CO_2 pump and depend.00s on a spatial separation of the CO_2 fixation and absorption (Edwards *et al.*, 2004; Lara *et al.*, 2002; Lara and Andreo, 2005).

In C3 species, the enzyme Rubisco, liable for the photosynthesis is responsible for about 30% of the leaf nitrogen content (Lawlor *et al.*, 1989), but in C4 species, Rubisco enzymes is responsible for only 4-21% of leaf nitrogen (Evans and von Caemmerer, 2000; Sage *et al.*, 1987). In C4 plants, photosynthesis occurs as an adaptation of the C3 pathway that suppresses the boundaries of photorespiration, thus, enhancing photosynthetic efficiency and lessening the water loss during hot and dry environments (Edwards and Walker, 1983).

Sequestration of carbon simply involves the transferring of the atmospheric CO_2 into protracted global pools, such as marine, pedologic, biotic or geological layers (Lal, 2008). Sequestration of CO_2 from the atmosphere, requires an integrated approach that meets the biochemical and ecosystem norms (Lal, 2004). Therefore, as a C4 plant, maize has the capacity to assimilate large amounts of $CO_{2,}$ more so than other starchy crops (potato, beans, and wheat) do, (Ghannoum *et al.*, 2011).

According to Ghannoum *et al.* (2011), C4 plants have, compared with C3 plant, a better plant water use efficiency, which is due to both, elevated photosynthetic rates per unit leaf area and a lessened stomatal conductance, allowing C4 plants to have better absorption of $CO_{2,}$ mostly significantly having the ability to separate CO_2 assimilation from the biochemical pathway of producing sugars from the biochemical pathway of producing sugars from CO_2 and H_2O. Elevated CO_2 concentrations are likely to substantially influence maize plant growth and metabolism.

In the 1970's plant scientists tried to analyse the impact of atmospheric CO_2 concentrations on plant development (Fleisher *et al.*, 2011). Several authors (Kimball, 1983; Lonbg *et al.*, 2004) reported that the impact of eCO_2 is associated with an increase in yield of various crops, including several vegetables, such as tomato, sweet potato, and potato. Nevertheless, little information is available on the influence that eCO_2 has on the nutritional quality of vegetables (Gruda, 2005; Moretti *et al.*, 2010). Reports by Yeoh *et al.* (1980) demonstrated that plant growth and development of more than 100 plant species is positively influenced by eCO_2 concentration when he measured the K_c and found that the K_c compensation point for C3 species ranged from 13-26 ppm (389-778 µbar), while C4 species ranged from 28-64 ppm (838-1916 µbar).

Due to the ever-changing climate resulting in altered precipitation patterns and global warming; numerous improvements and techniques need to be applied still to improve green mealies production. Only little information seems available on the effect of climate change in African countries on the effect of eCO_2 on plant production. Numerous papers have reported the reduction of stress under eCO_2, however, the eCO_2 effect on crop plants seems to have been studied less on reviews (AbdElgawad *et al.*, 2016), even though it is of importance for future agricultural crop productions (Berry and Bjorkman, 1980).

1.2 Origin and botanical description and domestication of sweetcorn (Zea mays L. var. saccharata)

Sweetcorn (*Zea mays* L. var. *saccharata*) originated primarily from Mesoamerican region, particularly the Mexican plateaus, about 7000 years ago; this area is regarded as the main centres of origin and, therefore, still is the area with the largest variety of maize types (Shukla *et al.*, 2013). Even though maize is thought to have originated from Mexico (Farnham *et al.*, 2003), other evidence points towards its origin in Africa or Asia (Inglett, 1970). Botanically, maize is a species in the Poaceae family, in the genus of *Zea* and it is an annual plant characterised by an extensive, fibrous root system. Biology of maize, (2011) indicates that within the genus of *Zea*, are four species for which *Zea* mays is of commercial significance. Sweetcorn mostly called standard sugary (*su*) maize, is said to have originated from mutation in the *Peruvian* race *Chullpi* (Brown and Darrah, 1985). The sugary gene in sweetcorn hinders or retards the usual conversion of sugar into starch during development of the endosperm, thus resulting in kernels that accumulate a water-soluble polysaccharide termed "phytoglycogen" (Brown and Darrah, 1985).

Subsequently, the dry, sugary kernels become crinkled and glossy in texture (Brown and Darrah, 1985). Sweetness is therefore, enhanced due to the higher water content-soluble polysaccharide accountable for adding texture quality factor adding on to sweetness (Brown and Darrah, 1985). Other *Zea* species are termed teosintes, which a mainly wild grasses that are native to Mexico and Central America. *Zea mays* has 2n = 20 number of chromosomes. The Andropogoneae tribe consists of seven genera which are old and new groups. It is indicated that the old world consists of Coix (2n = 10/20), Chionachne (2n = 20), Sclerachne (2n = 20), Trilobachne (2n= 20), as well as Polytoca (2n = 20); whereas new world group has *Zea* and Tripsacum (Biology of maize, 2011).

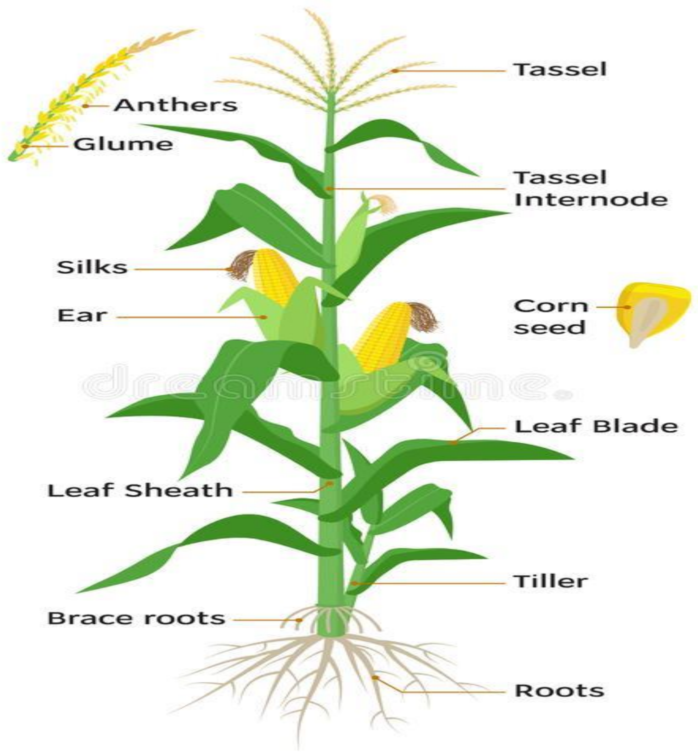


Fig. 1.1 Sweetcorn plant diagram, infographic elements with the parts of maize plant, anthers, tassel, maize ears, cobs, roots, stalks, silk, flowering, seeds fruits Vector encyclopaedic illustration flat design https://www.dreamstime.com/maize-plant-diagram-infographic-elements-parts-corn-plant-anthers-tassel-corn-ears-cobs-roots-stalks-maize-plant-image137290027; Copyright© 2000-2021 Dreamstime.

The plant is termed 'the Indian maize' in the United States (Inglett, 1970; Farnham *et al.*, 2003). Maize is a tall up to 3 m (OECD, 2006) and monoecious annual grass plant that has overlapping leaf sheaths and broad, razor-sharp leaves. The agronomical features of maize involve a pistillate inflorescences encircled in several, large foliaceous bracts (ears), ranging from 7 to 40 cm in length, with spikelets in 8 to 16 rows on a firm axis (cob) found in the leaf axils as well as spikelets in elongated spike-like racemes that are arranged into large spreading terminal panicles (tassels) (OECD, 2006). Maize is one of the oldest crops cultivated by man (FAO, 2006).

1.3 Uses of maize (green and dry grain)

Fresh maize is a versatile crop. It is mainly harvested before physiological maturity, so that the Qfruit (caryopsis – a type of simple dry fruit, the one that is monocarpellate and not opening at maturity stage) is not yet able to germinate and produce a new plant. At this developmental stage kernels can be utilised off the cob for human consumption as a vegetable (Serna-Saldivar *et al.*, 2000), it can be boiled, fried, roasted, ground, as alcohol beverages, gruel, or even fermented especially for baking bread and cakes (Inglett, 1970; Whistler, 1970; Gardner and Inglett, 1971; Alexander, 1987).

Dowswell *et al.* (1996) reported that there are different uses of maize in Africa, resulting in various forms of staple foods from dry maize such as Uphuthu or Insima in South Africa, Ugali in East Africa, Sadza in Zimbabwe, Nsima in Zambia, and Malawi. Maize can be processed into different forms by milling, grinding, fermentation, lime cooking (nixtamalization), boiling and frying. Dry grain maize is used for livestock feed grain, as a silage crop, as Uphuthu and pap for human food, as well as numerous industrial purposes. Due to its vast global distribution, maize is expensive compared to other rice and wheat, because it contains a vast variation of grain types. It has a wide range of biological and industrial properties (Dowswell *et al.*, 1996).

Maize is a staple food for many people globally, while the United States of America about 85% of maize production is destined for livestock feed (Shashidhara, 2008). As a food crop, maize ranks third (Fig. 2.2) in the world as a consumed crop, with an escalation in demand up to 45% from 1997 to 2020 (Young and Long, 2000). Meanwhile, Lucier and Dettmann (2008) reported that green mealies are second ranked and most consumed vegetables in the USA, after canned tomatoes and frozen potatoes. In developing countries, maize is the number one crop utilised for human consumption (FAOSTAT, 2016). A cultivar known commercially as sweetcorn, which is consumed by human and is cooked as green mealies directly on ear or off the ear, a form in which it offered is in frozen and canned foodstuff (Albuquerque *et al.*, 2008; Fritsche-Neto and Silva, 2011). When on cob fruits/kernels are boiled directly in hot water until the kernels are soft and tender, butter is added to glaze the kernels on the cob and seasoned with salt and pepper for taste (Shava *et al.*, 2009).

Sweetcorn is grown mainly for food, and it is harvested immaturely with moisture content of about 70%, sweetcorn both yellow and white can be easily differentiated from white, green maize because of higher sugar content due to the recessive transmutations stalling the conversion of sugar to starch, thus, sugar content accounts for up to 20% of dry matter, while it also accounts for about 3% in dent maize, when harvested at the green maize stage (Pajic, 2007).

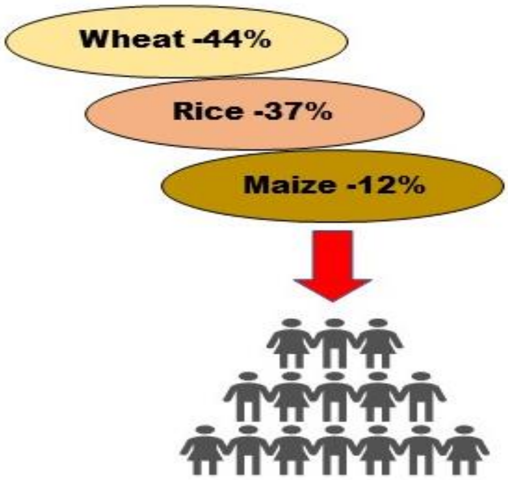


Fig. 1.2 A simplified diagram of the world's most consumed staple food crops form total of 157.2kg/person/year, with maize ranking third after wheat and rice. (Micronutrient Initiative, 2004).

Sweetcorn has even become commercially processed for canned and frozen mixed with other vegetables food products (Pollak and White, 1995). If used in the canning process, sweetcorn is harvested mechanically, so that the kernels are cut off the ear, canned and heat-treated for preservation (Henry and Kettlewell, 2012). Additionally, the fruit is cut off sweetcorn and further packaged into plastic bags and sold as fresh green mealies (Henry and Kettlewell, 2012).

Furthermore, dry grain maize can be processed for flour or semolina, mainly used for making couscous (Mali, Senegal, Togo), it is also used for porridges or pastes in countries like Togo, Ivory Coast, Nigeria, and Benin (Mestress *et al.*, 1990). Kernel and pericarp might be removed with the intention of ornamental decoration; the endosperm and kernel may be milled (or pounded) into maize flour, grits, or meal. In regions like Central America and Mexico, maize is changed into appetising products through the nixtamalization process which describes the preparation of maize by soaking in and cooking in basic or alkaline solution, mostly limewater [Ca $(OH)_2$], followed by the wash and afterwards hulled (Guzmán-de-Pena, 2010). The nixtamalization process is said to get rid of up 9 – 1005 of aflatoxins found in mycotoxin-contaminated maize (Guzmán-de-Pena, 2010), then after the steeping process for 10-12 hours. The ready cooked maize grain is then washed and milled into dough, which in turn is made into thin sheets in a pan, cut or extruded to make tortilla chips, maize chips, and other snack products (Katz *et al.*, 1974). All these processing procedures vary amongst regions and cultures (Rooney and Serna-Saldivar, 1987).

1.4 Maize botanical classification

These types are classified according to the traits of kernel endosperm (e.g., tissue surrounding the embryo which delivers food for the seed's growth). Maize is a tall about (1 – 4m) long and grown annually as a grass crop. It forms a seasonal root system bearing single most erect (clum) which is composed of nodes and internodes, even though other maize varieties form extended sideways branches (tillers). Maize has moderate varieties shorter than tropical and sub-tropical varieties. Maize leaves are broad and single leaf forms at each node in two opposite ranks (Esau, 1977). The outermost layer can be separated in cross-sections of the stalk. Having complex vascular bundles that appear scattered throughout the parenchyma tissue that creates the rest of the cross-section, though organised in a loosely form towards the middle (Esau, 1977).

Numerous authors (Purseglove, 1972; Paliwal, 2000; Darral *et al.*, 2003) identified different popular and most common maize types which are flint, floury, dent, popping, sweet, as well as waxy types.

- Flint maize: this type has kernels that are distinguished by their high percentage of hard endosperm all around small centre and is primarily grown in Latin America and Europe for food utilisation.
- Floury maize: It is grown primarily in Andean region and its endo sperm is made of soft starch, which makes it easy to grind and process into food stuff.
- Dent maize: most common type grown for grain and silage, it is grown primarily in USA. On each side and base of the kernel is a hard endosperm. The rest of the kernel is filled with soft starch, which when the grain begins drying the soft starch at the top part of the kernel contracts, depression on the kernel seed is caused resulting in a dent.
- Popping maize: these types of kernels are described by hard endosperm, for its proportion is higher than in other maize kernel types. Popping maize is not grown on a viable scale compared to the other types of maize, nevertheless, pop maize kernels are consumed worldwide as snacking.
- Sweetcorn: mainly grown for green ears (sweetcorn). The ears are mostly harvested immaturely roughly around 18-20 days post pollination when kernels still have moisture content of about 70%. Sugar content is much higher in the developing grain of sweetcorn due to the mutations caused by recessive alleles blocking the transformation of sugar into starch.

- Waxy maize: these kernels have almost all amylopectin as their starch slightly than the usual 70% amylopectin and 30% amylose. Waxy type is form part of food in some parts of East Asia and for some industrial purposes; it is said to produce starch that is like tapioca.

The appearance of kernels is attributed to the composition of the endosperm and the quantity and quality of endosperm as part of the fruit (George and Dickerson, 2008).

The Hidatsa Indians have at least nine classes of maize seed colours with five essentials that form part of daily human food: white, yellow, blue, pink, purple, and red (Wilson, 1917) which have been identified to be rich in carotenoids, anthocyanidins (about 325 mg/100g), that includes cyanidin derivatives (75% - 90%), peonidin derivatives (15% - 20%) and pelargonidin derivatives (5% - 10%) (Zhao *et al.,* 2009; Scott and Eldridge, 2005; de la Parra *et al.,* 2007; Romeo-Bastida *et al.,* 2018; Moreno *et al.,* 2005). Additionally, the Pueblo and Zuñi renowned black and connect the colours with the cardinal directions (Cushing, 1920). It was believed in ancient times that the seed colour had a significance meaning: the four main colours, named yellow signified the north, blue for signified the west, red signified the south, and white signified the east. It was also believed that black (very dark purple) signified the underworld while other colours were meant for the above (Underhill, 1946). Red coloured maize is identified as the sweeter cultivar because of the red colour which is probably mixed thoroughly with the rest of the crop (Johannessen *et al.,* 2018).

Seed colour serves as an important classification standard for maize consumers in Africa, where white maize is preferred over yellow maize. Nevertheless, 90% of maize produced globally is of the yellow type, while white maize continues to prevail in Africa entailing over 90% of the total maize crop produced, furthermore, contributes more than 30% of the world's white maize production (Khumalo *et al.,* 2011; McCann, 2005). Sweetcorn rich in carotenoids have now become the greatest popular type of maize especially in USA and China and, it is also most consumed as fresh maize in the world. Sweetcorn is ranked third amongst consumed vegetables in the USA and comes after canned tomatoes and frozen potatoes (Lucier and Dettmann, 2008). Maize stands out as the most diverse grain. Its diversity is due to human and nature alterations of the species which resulted in various types of maize.

1.5 Maize enhancement for nutritional quality

White maize is void of vitamin A and, a poor source of quality protein, therefore, maize has been biofortified with provitamin A and minerals to improve its nutritional quality (FAO, 1992; Johnson, 2000). Maize has been targeted for biofortification, since it is one of the six most- consumed staple foods therefore, drastic measures had to be taken to enhance with provitamin A (carotenoids) by means of conventional agricultural techniques as part of an international effort to combat Vitamin A Deficiency (VAD) (Tanumihardjo, 2008; HarvestPlus Brief, 2006).

Biofortification is defined as a strategy to intensify staple food crops with nutrients by means of agronomic approaches, conventional and mutation breeding, and genetic engineering (Hotz., 2013). Micronutrient malnutrition is also posing a serious threat to humanity and, livestock and, therefore, enhancing maize with micronutrients, more especially copper (Cu), iron (Fe), manganese (Mn) and zinc (Zn) by increasing micronutrient concentrations in the soil, could become a means to reduce such nutrient deficiency respectively (Gupta *et al.,* 2008).

Intensive usage of soils has caused a variety of challenges, among them a decline in soil micronutrients (Gupta *et al.,* 2008). The supplementation of the soil with micronutrient fertilizers has been perceived as the easiest approach for biofortification (Cakmak, 2008). Ortiz-Monasterio *et al.* (2008) reported that over 2 billion people across the world suffer from Zn deficiency (over 30% of the globe's residents). As a result, zinc deficiency has various, serious effects on the human body, resulting in stunted growth and becoming prone to infectious diseases. Iron deficiency has been found to be the root source of anaemia (Mabesa *et al.,* 2013), which is observed by a decrease in the number of red blood cells or the amount of haemoglobin in the blood, caused by inadequate dietary intake, or even poorly absorbed iron from food (NHLBI, NIH, 2014).

Because of the health incidents, maize has been enhanced to combat low crop yield (Salem and El-Gizawy, 2012) through fertilizer applications containing micronutrients specifically Cu, Fe, Mn, and Zn by means of simple fertilizer application which was discovered to positively affect food quality (Gupta *et al.,* 2008). In 2003, Pereira Filho reported an escalated green maize production, such that traditional producers of dry grain maize, bean, and coffee bean are abandoning those traditional crops with the optimism of exploring green maize production, seeing that it has been attaining more consumer acceptance. While maize as a mature dry grain is relatively well-studied, little literature seems to be written on fresh maize studies (Qwabe *et al.*, 2011), especially sweetcorn furthermore, the information about fresh ear yield remains unsubstantiated (Silva *et al.*, 2010). Nonetheless, van Averbeke *et al.* (1998) reported that there had been damages appearing in fresh, white maize seedlings caused by birds in smallholder irrigation schemes in the Eastern Cape province, and this predicament enforced a strategic solution of transplanting green maize seedlings to establish crops in otherwise less favourable conditions for direct seeding, and, preventing threat from birds on emerging seedlings.

Another study done on fresh maize was subjecting Bt green maize under different water harvesting techniques using two concentrations of commercial rooting. The technique involved water harvesting in the basins and mounds by using rooting to provide an advanced crop yield, in terms of permitting the accumulation of water in the plant structure resulting in ears with enhance commercial traits (Nascimento *et al.,* 2018). According to other literature, Vanaja *et al.* (2015) investigated the CO_2 impact on C4 plants by growing three maize genotypes in OTC to further study growth and yield response at eCO_2 conditions. These genotypes were DHM-117, Varun and Harsha were grown at an ambient CO_2 of 390 ppm and at eCO_2 of 550 ppm. It was found that at 550 ppm, there was an enhanced grain yield of 46% in genotypes DHM-117, Varun had a yield of 61% and Harsha sustained a massive grain yield of 127% at maturity.

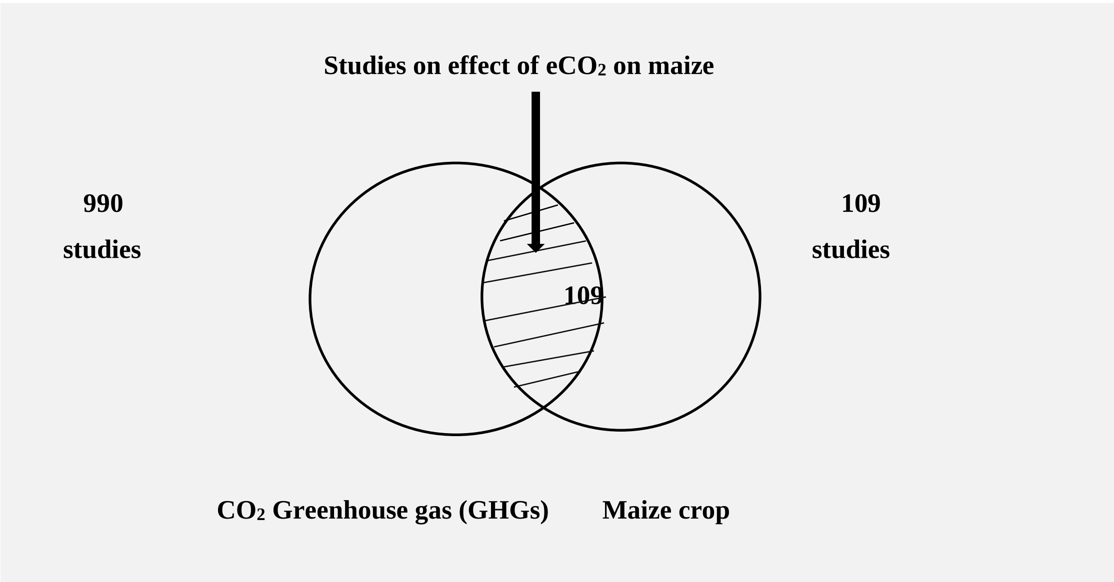


Fig. 1.3 Numbers of studies on eCO2 on plants and on maize between (1991–2020) according to the Web of Science. The shaded area represents studies on the eCO2 effect in dry grain maize.

Furthermore, Maroco *et al.* (1999) planted maize plants in plexiglass chambers, observed maize response to higher light intensity and CO_2 maintained at ambient or three times ambient CO_2 concentrations. The investigation showed that at elevated CO_2 of 1100 ppm had increased the photosynthetic rate by 15% better than maize plants grown at 350 ppm CO_2. Results obtained pointed out maize improvements by 20% stimulation of total biomass as well as 23% stimulation on leaf area at 30 days after emergence (DAE). This biomass stimulation was said to be intensified by the high CO_2 grown plants to maintain higher photosynthetic rates under higher light conditions. Other eCO_2 studies conducted on open top chambers (OTC) on mature maize crop have shown a great potential in terms of crop improvements.

Table 1.1: Studies carried out by various authors on the effect of elevated CO_2 concentrations (eCO_2) on dry maize (aCO_2 =…., open top chambers (OTC))

CO_2 concentrations	Method /methodology	Reference
aCO_2 390 – eCO_2 550 pp Growth environment	▪ Infrared gas analyser (IRGA). Copper tubing fitted with solenoid valve, rotameters to regulate CO_2 gas supply, sensors for measuring temperature and humidity	Vanaja *et al.*, 2006; 2015 (India)
aCO_2 µL L^{-1} and 3 times eCO_2 1100 µL L^{-1} High light temperature 3 $\pm$ 0.3^{0}C/20 $\pm$ 0.1^{0}C Growth environment – Plexiglass chambers (PC Relative humidity 75 - 85%	▪ Model 2006 AP, Valtronics, Concord, Calif., USA ▪ Solenoid valve that maintains the target CO_2 level	Maroco *et al.*, 1999 (USA)
aCO_2 380 - eCO_2 560 mmol /mol^{-1} Growth environment (OTC)	▪ PVC pipes CO_2 blowers to the OTC ▪ Infrared carbon dioxide analyser (WMA - 4 PP Systems, Harvehill MA), data logger to store data	Bunce 2014 (USA)
eCO_2 550 - 750 µmol/mo and aCO_2 control Growth environment (OTC)/ Plexiglass chambers	▪ F2000IAQ CO2 analyser (Control Technology Co. Ltd. China) ▪ Regulator inlet valves ▪ PVC air blowers to the OTC	Xie *et al.*, 2018 (China)
aCO_2 390 µmol/mol eCO_2 450 – 550 µmol/mol Growth environment (OTC)	▪ Pipe with pinholes to supply CO_2 ▪ Infrared gas analyser monitors to measure CO_2 gas	Meng *et al.*, 2014 (China)

Worldwide eCO_2 concentration in the atmosphere are escalating; hence, there is a need to investigate the effect of eCO_2 on the production of crops, external and internal quality parameters. The abundance of CO_2 in the atmosphere can, on the one hand, be taken advantage of, in order to produce improved vegetables and fruits. As Kimball (1983; 1993) and Poorter (1993) reported that, eCO_2 improves plant growth and development as had been investigated and more than 100 plant species by subjecting them to eCO_2 concentrations.

1.6 Influence of eCO_2 enrichment on vegetables and crops and their response to eCO_2

1.6.1 Effects of eCO_2 concentrations on plants enriched with nitrogen fertilization

According to Taub and Wang (2008) the concentration of nitrogen in plant tissue is frequently reduced when plants are grown under elevated CO_2 concentrations. Bunce (2014) described the effect of nitrogen application under eCO_2 under field conditions at the South Farm of the Beltsville Agricultural Research Centre, in Beltsville, Maryland, USA, conducted as part of two-year rotation routine (2004, 2006, 2008, and 2010), the author found an increased in ear fresh mass of maize; on the contrary, fresh ear mass was significantly declined with nitrogen application under the ambient CO_2.

Bunce (2014) also established that with nitrogen application of (0.5, 0.75, 1.0, 1.25, 1.5, and 2.0) kg/ha as recommended amount, was able to enhance maize stem and leaf dry mass respectively, as well as an increase in the maize leaf area index under eCO_2 concentrations compared to the nitrogen application under normal ambient CO_2. Bunce (2014) also established that nitrogen application under such eCO_2 conditions, enhanced maize leaf area index compared with nitrogen application to plants grown under normal, ambient CO_2.

According to Taub and Wang (2008), an increase in nitrogen increases plant use efficiency, thereby enhancing biomass; the authors attributed this effect to a rise in photosynthetic nitrogen use efficiency (PNUE) under eCO_2 conditions. Shimono and Bunce (2009) reported that the uptake of nitrogen by C3 rice roots plays an important role in enhancing grain yields under eCO_2 concentrations. It has been frequently recognised that an increase in nitrogen input eventually leads to an increase in yield under eCO_2 concentrations (Rogers *et al.,* 1996; Kimball *et al.,* 2002; Kim *et al.,* 2003). The yield due to eCO_2 per unit of nitrogen declines with additional of nitrogen input (Smart *et al.,* 1998; Kimball *et al.,* 2002; Kim *et al.,* 2003). According to Kimball *et al.* (2002) and Kim *et al.* (2003), eCO_2 was postulated to increases root mass as it enhances photosynthetic rate and, thereby growth; moreover, nitrogen uptake can also be increased due to a larger root mass with an increased root surface that meets soil nutrients to absorb them.

1.6.2 Effects of eCO_2 on germination, growth, and flowering

Climate change impacts particularly on vegetables grown in greenhouse are seemingly difficult to predict due to the vast number of imaginable scenarios (Bisbis *et al.,* 2018). It was also found that due to assessment of the regional and global climate simulations, that assumptions to some possible influences on greenhouse vegetable production can be attempted, and possible subsequent shift of quality that could possibly become to reality in the near future (Bisbis *et al.,* 2018). Bhattacharya *et al.* (1985) determined the effect of CO_2 at early stages of sweet potato. At the advanced stages of growth, variations between plants subjected to elevated CO_2 and those grown under ambient CO_2 greatly declined (Singh and Jasrai, 2011), because the compensation point of photosynthesis was probably not reached under eCO_2, hence, the CO_2 compensation point should differ between C3 and C4. Plants grown under eCO_2, display enhanced growth when CO_2 is elevated, and this growth is linked with lower transpiration rate and enhanced photosynthesis (Springer and Ward, 2007).

A meta-analysis conducted by Jablonski *et al.* (2002) revealed that plant reproduction and development are affected by eCO_2, such that an increased number in flowers (+19%), fruits (+18%) as well as seeds (+16%) was recorded on plants grown under eCO_2. Numerous authors (Edwards *et al.,* 2001; Hussain *et al.,* 2001; Thurig *et al.,* 2003; Mohan *et al.,* 2004; Zavaleta, 2006) found in some instances, eCO_2 can alter the developmental processes, such as germination and, leaf formation (Ainsworth *et al.,* 2006), the on-set of flowering and of senescence (Rae *et al.,* 2006). Ward *et al.* (2000) explained that eCO_2 concentrations may change plant strength towards adverse environmental conditions, mostly via modifications in the onset of flowering and through changed plant size at flowering, which, in turn, impacts resources accessible for the reproduction process.

An altered plant size was observed at the onset of flowering probably because plants were grown under eCO_2 (Reekie and Bazzaz, 1991; Wand *et al.,* 2000), such responses have been found to totally affect the reproductive output in annuals (Ward *et al.,* 2000). Additionally, delayed flowering of species grown at eCO_2 means that plants may not have completed their cropping cycle by the time the growing season ends (Ward and Kelly, 2004). On the contrary, a delayed flowering ensures the extension of vegetative growth and expands availability of resources for the reproduction which, therefore, improves the fitness during years when the growing season is elongated or rather in regions where the growing season is endless (Roux *et al.*, 2006). It was found that under eCO_2, maize does not exhibit consistent patterns in the response to flowering time, such that Hesketh and Hellmers (1973) stated noticeably delayed flowering of maize at eCO_2, while Leakey *et al.* (2006) stipulated that there were no changes in flowering time of maize under such conditions.

1.6.3 Effects of eCO_2 enrichment on leaf colour and size

Adishesha *et al.* (2017) discovered that the leaf area per plant at varying growth phases of maize was severely increased by eCO_2 and elevated temperature regimes. Additionally, the assimilation rate improved under eCO_2 treatments, such that a higher CO_2 absorption rate was discovered due to an increased in intercellular CO_2 concentration, which clearly suggests that the chloroplast was substrate limited under aCO_2 conditions.

Adishesha *et al.* (2017) also elucidated that there is a considerable amount of information affirming that the assimilation rate increases significantly, when the plants are exposed to eCO_2 concentrations. Thus, the higher chlorophyll concentration in the leaf due to eCO_2 treatments (1.500 ppm eCO_2 treatment) and an elevation in temperature results in increased assimilation.

1.6.4 Effects of eCO_2 enrichment and photosynthesis

Reports by several authors (Long *et al.,* 2006; Ainsworth *et al.,* 2008; Albert *et al.,* 2011; Dieleman *et al.,* 2012; Xu *et al.,* 2013, 2015; Huang and Xu, 2015; Pandey *et al.,* 2015; Kimball, 2016) demonstrated presence of the little documentation on maize available on the interactions attributed to elevated CO_2 and plant responses to the environmental stress factors. The positive impact of elevated CO_2 had only a minor stress effect on plants, demonstrated at various plant growth stages (Sicher and Bunce, 2015), nonetheless, cellular damage of oxidation (e.g., lipid peroxidation, protein oxidation) has been demonstrated (Geissler *et al.,* 2010). Such damage occurs due to stress-generated reactive oxygen species (Geissler *et al.,* 2010; Mishra *et al.,* 2013; Zinta *et al.,* 2014; AbdElgawad *et al.,* 2015).

Poorter *et al.* (1997) and Wang (1998) stipulated the photosynthetic rate is increased by eCO_2; thus, plant growth is stimulated and an increase in the C:N ratio results in most plant species. Turner and Brittain, (1962) contended that the CO_2 transition into organic compounds is initiated by the Rubisco enzyme situated in the chloroplast stroma in C3 plants. The CO_2 concentration in the stroma has been estimated to be 10 ppm, closer to the Michaelis constant (K_m) for the fixation reaction of CO_2 (Goldworthy, 1968; Sicher and Bunce, 2015).

The initial step of the Calvin cycle involves one molecule of CO_2 reacts with a molecule of ribulose-1,5-bisphosphate (RuBP) to form two molecules of 3-phosphoglycerate, and, therefore, a 3-carbon compound (the C3 species derives originates its name from this 3-carbon molecule). This reaction is said to be broken down by the RuBP carboxylase/oxygenase, called Rubisco enzyme. It is also postulated that Rubisco functions as an oxygenase. The last function induces a sequence of physiological events in which O_2 uptake with its dependence on light is linked to CO_2 evolution, thus, a process is called photorespiration, which happens in opposite direction to photosynthesis, which subsequently causes loss of CO_2 that has been fixed by the Calvin Cycle (DaMatta *et al.*, 2010).

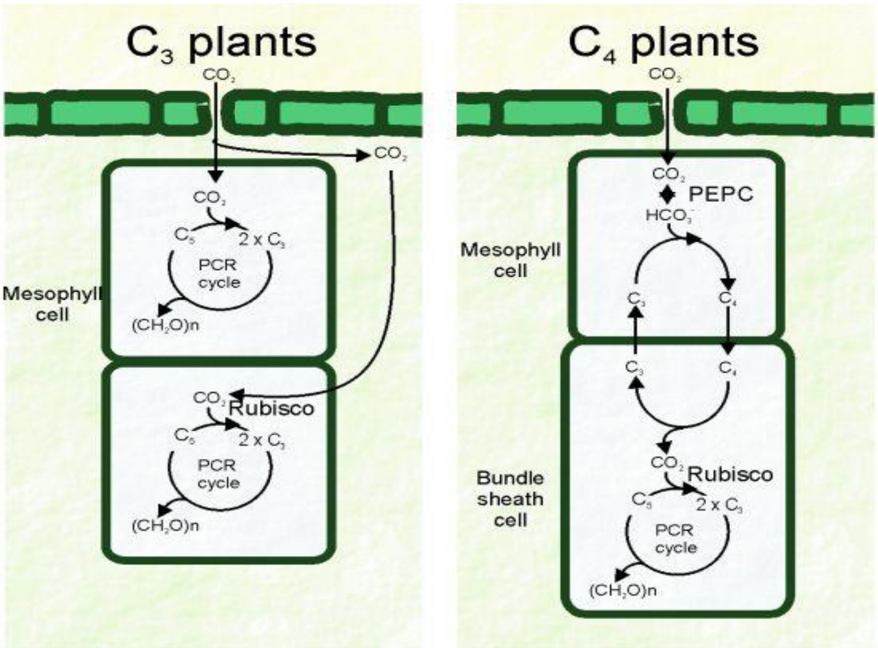


Fig. 1.4 A simplified schematic diagram depicting carbon fixation pathways in C3 and C4 plants. Abbreviations: C3, three-carbon organic acids; C 4, four-carbon organic acids; C5, ribulose-1,5-bispphosphate; PCR, Photosynthetic Carbon Reduction Cycle; PEPC, phosphoenolpyruvate carboxylase; Rubisco, Ribulose-1,5-biophosphate/oxygenase (Kranz anatomy), (Lara and Andreo, 2011).

C4 plants, such as maize, sorghum, and sugarcane (Fig. 3), utilise a series of enzymes that start by combining CO_2 (HCO_3^-) with 3-carbon molecule (phosphoenolpyruvate, PEP), thereby yielding oxaloacetate, with 4-carbon compound. The reaction is broken down by PEP carboxylase (PEPCase), with higher affinity for CO_2 than Rubisco and are in shot of oxygenation activity (DaMatta *et al.*, 2010). Products from the initial reaction of PEPCase are C4 acids, which at a later stage are decarboxylated in the presence of Rubisco; as a result, stomatal closure due to CO_2 usually does not delay the rate of photosynthesis in maize and other C4 plants and, hence, the photosynthetic rate is induced by eCO_2.

Rubisco is an oxygenase that prevents the activity of carboxylase. Vodnik *et al.* (2005) revealed that maize and other C4 plants, such as sorghum and sugarcane, rely on the carboxylase enzyme Rubisco, i.e., phosphor(enol) pyruvate carboxylase (PEPCase) to break down. The first products from the reaction of PEPCase are C4 acids, which are later decarboxylated in the surrounding area of Rubisco (Turner and Brittain, 1962). Consequently, the fixation rate of CO_2, the rate of the whole plant growth, and the yields of C4 plants are only slightly, or not at all, influenced by eCO_2 concentrations, unlike C3 plants. Nonetheless, both, C3 and C4 display stomatal closure as a response mechanism towards eCO_2 which has a significant plant–water use (Bunce, 2004).

Since the concentrations of CO_2 are sustained, slight stomatal closure due to eCO_2 should not hinder the rate of photosynthesis in maize and other C4 plants (Sage, 1999); hence, the rate of growth in maize is likely to be positively affected by CO_2 enrichment (Vodnik *et al.*, 2005). Additionally, eCO_2 concentration can increase the performance of C4 plants as they can close stomatal after sufficient CO_2 has been captured, while transpiration is halted due to closed pores, thereby increasing the plant's water use efficiency (Owensby *et al.*, 1996). It is also clear that eCO_2 encourages stomatal closing, thus, improving water use efficiency and protecting against drought stress and, in turn, ozone uptake, thereby reducing the impact of ozone stress (AbdElgawad *et al.*, 2016).

This points out that stress-alleviation influences due to eCO_2 cannot entirely be credited to maximised antioxidant defences. A crucial, alternative process probably involved in the effect reduction of oxidative stress, due to high CO_2, is photorespiration. Possibly eCO_2 stimulates carboxylation, carried out by Rubisco, and decrease reactive oxygen species (ROS) development (relaxation hypothesis; Long and Drake, 1991; Booker *et al.*, 1997; Ainsworth *et al.*, 2008; Zinta *et al.*, 2014; AbdElgawad *et al.*, 2015). At current CO_2 levels in the atmosphere, C4 plants especially dicotyledons usually require small amount of water than C3 plants due to the higher CO_2 uptake levels and huge stomatal resistance to water loss (Ehleringer *et al.*, 1997) put C4 plants at an advantage to thrive under drought conditions.

1.6.5 Impact that eCO_2 has on plant biomass and yield

It is postulated that the rate of difficulty in which the process of crop simulations in relation to allocation of biomass relies on plant's capability to allocate carbon sinks, such as roots, stems, leaves, as well as grain and fruit (Tubiello and Ewert, 2002). Several authors (Kramer, 1981;

Mauney *et al.*, 1979; Sionit *et al.*, 1982) reported an increased biomass production in several plants under CO_2. According to Mortensen (1994), eCO_2 in the range of 335 to 800-900 ppm increases yield of several vegetable crops, such as lettuce, carrots, and parsley by 18%, 19% and 17%, respectively. Reeves *et al.* (1994) reported that when treated with eCO_2 concentration, dry matter production of sorghum by 13.5% and soybean by 23.8%, was observed, while seed yield for sorghum increased by 17 .5% and of soybean by 34.7%.

Torbert *et al.* (2004), showed that eCO_2 improved total biomass accumulation of grain sorghum by an average of 30% and of soybean by average of 40%. Similarly, Vanaja *et al.* (2015) reported an increase in biomass of various maize cultivars grown under eCO_2, moreover, this increase ranged from 47% Varun, 34% Harsha to 32% DHM-117. Additionally, variations in magnitude of response to eCO_2 by maize is likely due to the source and sink relationships and influenced by crop management and an effect of the crop's water and nutritional status, the time of exposure, temperature, pot size and light intensity (Sage, 1994; Drake *et al.*, 1997).

1.7 Postharvest phytochemical parameters of maize grown under eCO_2 concentrations

1.7.1 Impacts of eCO_2 on kernel carotenoid, sugars, and acidity concentration

Dhami *et al.* (2018) reported that the effect of eCO_2 concentrations on leaf carotenoid of Arabidopsis and chlorophyll concentration on leaves of Arabidopsis differs between species. The C: N ratio increases in plants with an elevation in ambient CO_2 causing concomitant changes in elemental composition (Loladze, 2002; Myers *et al.*, 2014), isoprenoids (Sun *et al.*, 2013; Way *et al.*, 2013), flavonoids (Gaufo *et al.*, 2014; Karowe and Grubbo, 2011), anthocyanins (Tallis *et al.*, 2010), as well as alkaloids (Ziska *et al.*, 2008; Singh and Agrawal, 2015) in various plants/ plant parts. Elevated CO_2 improves absorption by maximising foliar soluble sugars, starch, as well as amino acids (Moore *et al.*, 1998; Florian *et al.*, 2014; Noguchi *et al.*, 2015). Hence, it is reasonable to assume that eCO_2 concentrations will enhance the production of isoprenoid substances needed for carotenogenesis (Dhami *et al.*, 2018) in maize.

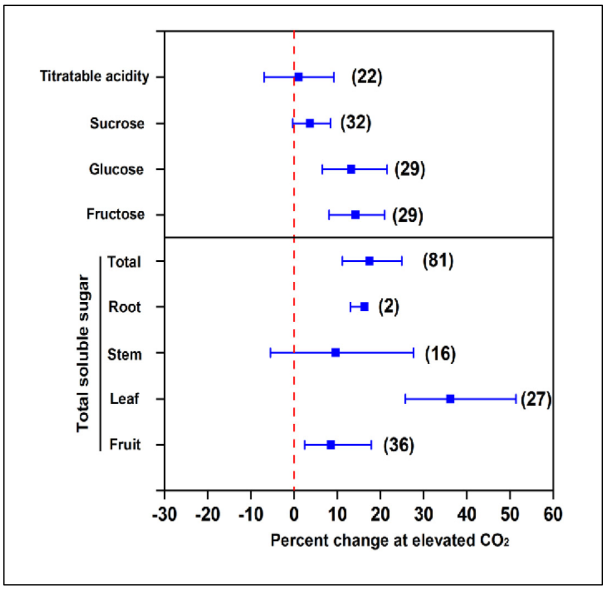


Fig. 1.5 The effect of eCO2 on the soluble sugar and acidity concentrations in vegetables. Data represents mean of percentage change with 95% confidence intervals (indicated by errors bars) for eCO2 when compare with aCO2. (Dong et al., 2018).

The rate of CO_2 assimilation and photosynthesis must be preserved in plants because they influence the carotenogenic process which can be altered from within the leaves to better suit the current environmental and developmental alterations, yet upholding carotenoid-derived production that points out to metabolites liable for homeostasis maintenance (Jin *et al.*, 2016; Ritz *et al.*, 2000; Wang *et al.*, 2015). It has also been revealed that strawberry fruit grown under eCO_2 levels contained higher concentrations of flavour compounds than those grown at normal aCO_2 levels (Balasooriya *et al.*, 2017), indicating that yellow maize grown under eCO_2 levels is likely to be even darker yellow colour due to an intensification of carotenoid accumulation.

Findings by Vodnik *et al.* (2005), indicated a decrease in leaf chlorophyll concentration in maize (leaves and fruit) grown under eCO_2 conditions compared with plants grown under low CO_2 conditions. Additionally, Vodnik *et al.* (2005) stipulated a decrease in foliar of neoxanthin, lutein, β-carotene and total carotenoids under eCO_2 which was not prominent on plants grown under aCO_2. Findings on tomato plants indicate that eCO_2 has a positive impact on tomato fruit quality (Dong *et al.*, 2018), as the concentration of sugars e.g., fructose, glucose and total soluble sugars analysis indicated that eCO_2 enhances tomato quality Fig. 4, and more likely the taste, (14.7%, by utilizing meta-analysis, n = 24), showed a positive of tomato (Islam *et al.*, 1994; Behboudian and Tod, 1995; Li *et al.*, 2007; Zhang *et al.*, 2014) towards eCO_2 conditions. Apparently, it was pointed out by Khan *et al.* (2013) that eCO_2 is also said to increase soluble sugar and fibre build-up in tomato cv. "Eureka" by hastening its maturity.

Long *et al.* (2004) discovered that the increased CO_2 fixation under eCO_2 conditions encourages the synthesis of triose phosphates in leaves ultimately transforming into C6 carbohydrates, e.g., glucose, fructose, and sucrose. Moreover, an increase in carbon availability, is likely to result in an increase in supply of defence (antioxidant) molecules and is frequently held largely accountable for better protection against oxidative damage under eCO_2 conditions (antioxidant notion) (AbdElgawad *et al.*, 2016).

Reports from previous studies on CO_2 enrichment point, however, points out that eCO_2 has only a small or no effect on antioxidant concentration, with even a possible decline in antioxidant levels (Erice *et al.,* 2007; Farfan-Vignolo and Asard, 2012; Mishra *et al.*, 2013). Dong *et al.* (2016) performed a meta-analysis of all vegetables including mostly consumed vegetables (lettuce, tomato, and potato) and uncovered that eCO_2 increased the sugar concentration in leaves by e.g., glucose by 13.2%, fructose by 14.2%, sucrose by 3.7% (at P = 0.07) and total soluble sugar by 17.5%.

1.7.2 Impacts of eCO_2 on starch and protein (amino acids) concentration

Starch is the most important storage carbohydrate in most plants, and its concentration is positively enhanced in plants grown under eCO_2 conditions (Araujo *et al.,* 2008). Spring wheat grown under eCO_2 of 550 and 700 ppm had higher concentrations of starch, sucrose, glucose, total non-structural carbohydrates (TNCs), free amino acids, soluble protein and less fructose and nitrogen (Chen *et al.,* 2010). Because plants show improved growth at eCO_2, related to declined transpiration and improved photosynthesis, this phenomenon often gives rise to the concentration of non-structural carbohydrate (sugars and starches) inside the leaf tissues (Curtis and Wang, 1998; Long *et al.*, 2004; Teng *et al.*, 2006). Furthermore, Smith *et al.* (2005) postulated that the increase in starch possibly contributes to instantaneously enhanced levels of sucrose.

It was postulated that eCO_2 concentrations is liable for increasing photosynthesis in plants, which positively leads to improved growth, above-ground biomass, and yield (Ainsworth and Long, 2005; van der Kooi *et al.,* 2016). Nevertheless, a negative effect was identified to have adverse outcomes for the quality of crops species, bringing about a massive decline in a pool of nutrients such as protein concentrations of food crops (Fernando *et al.,* 2015; Broberg *et al.*, 2017). Plant proteins are most vitally significant for the macromolecules necessary for growth and developmental stages which includes biological reactions, structural, cellular, and membrane transport systems (Balasooriya *et al.,* 2017). In lettuce grown under eCO_2, it was found that the taste was potentially enhanced through the increase in soluble sugar accumulation by 27.1% (n = 18, meta-analysis), but a decline in the protein concentration by

5.6% under eCO_2 (n =12, meta-analysis) was also reported, which indicated decrease in nutritional value as proven by numerous studies (Jin *et al.*, 2009; Baslam *et al.*, 2012; Pérez-López *et al.*, 2015; Becker and Kläring, 2016).

1.7.3 Impacts of eCO_2 on mineral concentration

Several authors (Ziska and Bunce, 1997; Wand *et al.*, 1999; Ghannoum *et al.*, 2000) established evidence that growth of C4 plants is increased under eCO_2 concentrations, nonetheless, the fundamental mechanisms of response are largely unexplainable (Vodnik *et al.*, 2005). It is, furthermore, important that a well-established root system together with good-irrigation and nutritional practices also contributes to good development of green maize, because the roots are responsible for absorbing water and mineral salts from the soil to maximise plant productivity (Kluthcouski and Stone, 2003; Vieira and Santos, 2005) under eCO_2 conditions.

Results by Dong *et al.* (2018) provided evidence of a decline in nutrients in leafy vegetables, fruit vegetables, stem vegetables, and root vegetables, minerals such as Mg, Fe, and Zn by 9.2%, 16.0%, and 9.4%, respectively, while concentrations of P, K, S, Cu, and Mn remained unchanged under eCO_2. Idso and Idso (2001); Fierro *et al.* (1994) recommend compensating mineral deficiencies associated with eCO_2 by 900 ppm concentration but to a minimal amount to lessen chemical toxicity and losses associated with quality and quantity of vegetable crops more specifically for zinc (Rengel *et al.*, 1999).

1.8 Summary and conclusions

Green mealies are crops consumed by a vast number of people across the world. It is also produced in small-holder irrigation systems (SIS) in South Africa, specifically in the Eastern Cape Province. Green mealies, particularly yellow cultivars, are a good source of vitamin A and B, of protein, starch, and minerals as well as antioxidant compounds. Maize is known globally as a crop of high adaptability to different environmental conditions, is worldwide in high economic value. Maize, as dry grain ranks the second-most consumed crop in the world. In Sub-Saharan Africa and Latin America, maize is often consumed as green mealies and as dry grain for process the kernels and more than 1.2 billion people and is liable for 30-50% of small income households' particularly in Eastern and Southern Africa.

C3 and C4 plant species are affected by rising concentrations of CO_2 in the atmosphere as this crease directly alters leaf morphology and plant biochemistry. Elevated CO_2 levels have been shown to greatly affected C3 crops, like sugar beet, potato, as well as winter wheat. It is expected that the prevalence of the CO_2 in the atmosphere will rise to 700 ppm by the end of the current century and this could possibly result in a rise in the Earth's surface temperatures by 2^o to 4^oC to such an extent that it will pose as threat to plant production and productivity. Therefore, enhanced carbon sequestration under eCO_2 by sweetcorn plants could result in enhancing growth, development, and post-harvest, the presence of certain phytochemical compounds.

Enrichment with CO_2 might play a significant role in the growth and development of plants. With adequate application of nitrogen under eCO_2, in grains, an increase in fresh ear mass, leaf area index and leaf dry mass is observed; however, other shortcomings such as lack of N under eCO_2 can result in fresh ear mass reduction. It is known that the eCO_2 enrichment maximises root mass, simply by improving photosynthetic rate, and thereby plant growth, leading to an increased nitrogen uptake by the roots due to an increased root surface allowing better uptake of soil nutrients; thus, absorption can occur more effectively.

Elevated CO_2 positively impacts the flowering duration as it increases the number of flowers. Elevated CO_2 is said to alter development processes such as germination, leaf formation, the onset of flowering and senescence, furthermore, eCO_2 concentration can alter the plant vigour towards adverse environmental conditions, frequently via modifications in the onset of flowering and through changed plant size at flowering, which, in turn, influences resources accessible for the reproduction process. Though eCO_2 has positive impacts on plants, shortcomings also prevail to an extent that delayed flowering can occur leading to non-completed life cycle of the plant in a growing season.

Studies also revealed an increase in leaf area of dry grain maize under eCO_2 and higher temperature regimes. It has also been postulated that the leaf chlorophyll concentration will be enhanced by eCO_2 concentrations. The higher CO_2 concentration also induces the rate of photosynthesis in both, on C3 and C4 plants. The Rubisco enzyme, situated within the chloroplast stroma, acts as an oxygenase that constantly prohibits carboxylase activity; thus, Rubisco and phosphor(enol) pyruvase carboxylate (PEPCase), which form the initial reactions of photosynthesis process. In tall plants, carbon is almost assimilated through the lessening of pentose phosphate cycle, often termed the Calvin cycle. Basically, research has found that photosynthesis is encouraged by eCO_2 levels.

In recent studies on the effects of CO_2 enrichment of maize, the focal point has always been on dry grain mass and no studies have been conducted on sweetcorn as green mealies.

Sweetcorn cultivars are said to have a great potential as a vegetable consumed at the green stage when the caryopsis, the botanical fruit, is immature. There is a need to investigate the effect of carbon elevation in the atmosphere on sweetcorn because the crop contains a higher amount of phytochemicals, such as carotenoids, sugars, starch, protein, and minerals than dry grain maize.

Because maize, as a C4 plant, has the capacity to absorb a great quantity of CO_2, more so than other starchy C3 crops (potato, beans, and wheat), conducting a glasshouses study using instrumentation to measure and monitor CO_2, seeks to validate recent work. Additionally, elevated CO_2 concentrations are likely to have a substantial influence on green maize plant growth and metabolism, particularly colour development and the fruit's sugar concentration. Moreover, recommendations from previous studies do not focus on green mealies cultivars. Instruments used for the experiments from previous CO_2 studies were not clearly indicated for the accuracy of results, albeit the outcomes show a great potential towards addressing the atmospheric eCO_2 concentration predicament.

In conclusion, there is very little information available on the effects of eCO_2 on fresh sweetcorn growth and development, and on the post-harvest traits of fresh sweetcorn. Investigations with combined information of temperature and eCO_2 levels towards fresh maize germination, growth and development, more specifically fresh maize yield, biomass, quality, and phytochemical quality remain scarce or are lacking. The quality of a horticultural commodity is determined by its size and external appearance, both factors essential for consumer acceptance. Therefore, there is a need to study the effect of eCO_2 and elevated temperature, alone and in combination, at various stages of development to understand how climatic change may impact especially the morphological, chemical, and nutritional qualities of sweetcorn. From previous CO_2 investigations done on maize crop, the focus was mainly directed towards improving dry grain maize. A gap remains when it comes to investigations concerned with responses towards morphological, development, and phytochemical parameters of sweetcorn as green mealies for which a rise in level GHGs remains a predicament.

Literature cited

AbdElgawad H.; Farfan-Vignolo E. R.; De Vos D. and Asard H. (2015). Elevated CO_2 mitigates drought and temperature-induced oxidative stress differently in grasses and legumes. Plant Science. Vol. 231: 1–10.

AbdElgawad H.; Zinta G.; Beemester G. T. S.; Janssens I. A. and Asard H. (2016). Future Climate CO_2 Levels Mitigate Stress Impact on Plants: Increases Defense or Decreased Challenge? Plant biotechnology, Journal of Frontiers in Plant Science Vol. 7: 556.

Acock B. and Allen L. H. Jr. Office of Energy Research, US Department of Energy Washington, DC (1985).

Adishesha K.; Janagoudar B. S. and Amaregouda A. (2017). Response of maize (*Zea mays* L.) genotypes to elevated carbon dioxide and temperature regimes. International Journal of Chemistry Studies. Vol. 5(5): 2448 – 2456.

AGRA (Alliance for a Green Revolution in Africa). (2014). Africa agriculture status report 2014: Climate change and smallholder agriculture in sub-Saharan Africa. Nairobi: AGRA.

Ainsworth E. A. and Long S. P. (2005). what have we learned from 15 years of free-air CO_2 enrichment (FACE)? A meta-analytic review of the responses of photosynthesis, canopy properties and plant production to rising CO_2. New Phytol. Vol. 165(2): 351-371.

Ainsworth E. A.; Rogers A.; Vodnik L. O.; Walter A. and Schurr U. (2006). The effects of elevated CO_2 concentration on soybean gene expression. An analysis of growing and mature leaves. Plant Physiology. Vol. 142: 135–147.

Ainsworth E. A.; Beier C.; Calfapietra C.; Ceulemans R.; Durand-Tardif M. and Farquhar G. D., *et al.* (2008). Next generation of elevated CO_2 experiments with crops: a critical investment for feeding the future world. Plant Cell Environment. Vol. 31: 1317–1324.

Albert K. R.; Mikkelsen T. N.; Michelsen A.; Ro-Poulsen H. and Van Der Linden, L. (2011). Interactive effects of drought, elevated CO_2 and warming on photosynthetic capacity and photosystem performance in temperate heath plants. Journal of Plant Physiology. Vol. 168: 1550–1561.

Albuquerque C. J. B.; Von-Pinho R. G. and Silva R. (2008). Performance of experimental and commercial maize hybrids for green maize production. Ciênc. Agrotechnology. Vol. 32(2): 69-76.

Allen L. H. Jr; Bisbal E. C.; Campbell W. J. and Boot K. L. (1990). Science Society of Florida Proceedings Vol. 49: 124-131.

Alexander R. J. (1987). Maize dry milling: processes, products, and applications. In: Watson S. A. and Ramstad P. E., editors. Maize: chemistry and technology. St. Paul, Minn.: American Association of Cereal Chemistry. (pp 351–76).

Andrews T. J. and Lorimer G. H. (1987). Rubisco: Structure, mechanisms, and prospects for improvement. In 'The Biochemistry of Plants: A Comprehensive Treatise. Vol: 10, Photosynthesis'. (Eds). MD Hatch and NK Boardman. Academic: New York. (pp. 131–218).

Antoniali S.; Santos N. C. B. and Nachiluk K. (2012). Organic green maize: Production and post-harvest. Research & Technology. Vol. 9(2): 1-6. Portuguese.

Araujo R. C.; Pires A. V.; Susin I.; Mendes C. Q.; Rodrigues G.H.; Packer I. U. and Eastridge M. L. (2008). Milk yield, milk composition, eating behaviour, and lamb performance of ewes fed diet containing soybean hulls replacing coast cross (Cynodon species) hay. Journal of Animal Science. Vol. 86(12): 3511-3521.

Bancy, M. M. (2000). The influence of climate change on maize production in semi-humid and semi-arid areas of Kenya Journal of Arid Environments. Vol. 46: 333-334.

Barros J. F. C. and Calado J. G. A. (2014). Culture of maize. Soil Technology and Cultures, Agricultural Basics and General Agriculture Fundamentals. Évora: University of Évora; Portuguese.

Balasooriya H. N.; Dassanayake K. B.; Tomkins B.; Seneweera S. and Ajlouni S. Impacts of Elevated Carbon Dioxide and Temperature on Physicochemical and Nutrient Properties in Strawberries. Journal of Horticultural Science and Research ISSN: 2578-6598.

Baslam M.; Garmendia I. and Goicoechea N. (2012). Elevated CO_2 may impair the beneficial effect of arbuscular mycorrhizal fungi on the mineral and phytochemical quality of lettuce. Annals of Applied Biology. Vol. 161: 180–191.

Bauer H. and Martha P. (1981). The CO_2 Compensation Point of C3 Plants – A Re-Examination. I. Interspecific Variability. *Z. Pflanzenphysiol. Bd.* 103.S. (pp 445-450).

Becker C. and Kläring H. P. (2016). CO_2 enrichment can produce high red leaf lettuce yield while increasing most flavonoid glycoside and some caffeic acid derivative concentrations. Food Chemistry. Vol. 199: 736–745.

Behboudian M. H. and Tod C. (1995). Postharvest attributes of ‘Virosa’ tomato fruit produced in an enriched carbon dioxide environment. Horticultural Science. Vol. 30: 490–491.

Berry J. and Bjorkman O. (1980). Photosynthetic response, and adaptation to temperature in high plants. Annual Reviews of Plant Physiology. Vol. 31: 491-543.

Bhattacharya N. C.; Biswas P. K.; Bhattacharya S.; Sionit N. and Strain B. R. (1985). Crop Science Vol. 25: 975-981.

Bisbis M. B.; Gruda N. and Blanke M. (2018). Potential impacts of climate change on vegetable production and product quality – a review. J. Clean. Prod. Vol. 170: 1602–1620 https://doi.org/10.1016/j.jclepro.2017.09.224.

Biology of maize. (2011). Retrieved from http://dbtbiosafety.nic. in/guidelines/maize.pdf.

Booker F. L.; Reid C. D.; Brunschön-Harti S.; Fiscus E. L. and Mille J. E. (1997). Photosynthesis and photorespiration in soybean [Glycine max (L.) Merr.] chronically exposed to elevated carbon dioxide and ozone. Journal of Experimental. Botany. Vol. 48: 1843–1852.

Broberg M.; Högy P. and Pleijel H. (2017). CO_2-induced changes in wheat grain composition: meta-analysis and response functions. Agronomy. Vol. 7: 32.

Brown W. L. and Darrah L. L. (1985). Origin, adaptation, and types of maize. National Maize Handbook. The Maize Crop NCH-10. Cooperative Extension Service, Iowa State University of Science and Technology and the United States Department of Agriculture cooperating.

Bunce J. A. (2004). Carbon dioxide effects on stomatal responses to the environment and water use by crops under field conditions. Oecologia. Vol. 140: 1–10.

Bunce James S. (2014). Maize Growth Response to Elevated CO_2 Varies with the Amount of Nitrogen Applied American Journal of Plant Sciences. Vol.: 5: 306 – 312 (http://www.scirp.org/journal/ajps).

Calzadilla A.; Zhu T.; Rehdanz K.; Tol R. S. J. and Ringler C. (2013). Economy wide impacts of climate change on agriculture in Sub-Saharan Africa. Ecological Economics. Vol. 93: 150-165.

Cakmak I. (2008). Enrichment of cereal grains with zinc: agronomic or genetic biofortification, Plant Soil. Vol. 302(1-2): 1-17.

Chen J.; Xu W.; Burke J. J. and Xin Z. (2010). Role of phosphatidic acid in high temperature tolerance in maize. Crop Science. Vol. 50: 506-515.

Curtis P. and Wang X. (1998). A meta-analysis of elevated CO_2 effects on woody plant mass, form, and physiology. Oecologia. Vol. 113: 299–313.

DaMatta F. M.; Grandis A.; Arenque B. C. and Buckeridge M. S. (2010). Impacts of climate changes on crop physiology and food quality. Food Research International Vol. 43: 1814-1823.

Darrah L. L.; McCullen M. D. and Zuber M. S. (2003). Breeding, genetics, and seed maize production. Chapter 2. In: P. J. White, L. A. Johnson, eds, Maize: Chemistry and technology, Edition 2nd. American Association of Cereal Chemist, Inc. St. Paul, Minnesota, USA, (pp 35-68).

Dale E. and Niernberger F. F. (1982). Economic models of dry maize milling: part I-model design and system specifications. Assoc. Oper. Millers Tech. Bull, (pp 3994–3402).

De la Parra C.; Saldivar S. O. S. and Liu R. H. (2007). Effect of processing on the phytochemical profiles and antioxidant activity of maize for production of masa, tortillas, and tortilla chips, Journal of Agriculture Food Chemistry. Vol.: 55: 4177-4183.

De Temmerman L. and Hacour A. Guns M. (2002). Changing climate and potential impacts on potato yield and quality 'CHIP': introduction, aims and methodology. European Journal of Agronomy. Vol. 17: 233–242.

Dickerson George W. Extension Horticulture Specialist. Speciality Maizes, Guide H-232, pp 1-4. (2008). Cooperative Extension Service, College of Agriculture and Home Economics. New Mexico State University.

Dieleman W. I.; Vicca S.; Dijkstra F. A.; Hagedorn F.; Hovenden M. J. and Larsen K. S., *et al.* (2012). Simple additive effects are rare: a quantitative review of plant biomass and soil process responses to combined manipulations of CO_2 and temperature. Global Change in Biology. Vol, 18: 2681–2693.

Dhami Namraj; Tissue Davi T. and Cazzonelli Christopher I. (2018). Leaf Dependent Response of Carotenoid accumulation to elevated CO_2 in Arabidopsis. Archives of Biochemistry and Biophysics.

Dong J.; Gruda N.; Lam S. K.; Li X. and Duan Z. (2018). Effects of Elevated CO_2 on Nutritional Quality of Vegetables: A Review. Frontiers Plant Science. Vol. 9: 924.

Dowswell C. R.; Paliwal R. I. and Cantrell R. P. (1996). Maize in the third world. United State of America; Westvie Press.

Drake B. G.; Gonzalez-Meler M. A. and Long S. P. (1997). More efficient plants: a consequence of rising atmospheric CO_2? Annual Review in Plant Physiology. Vol. 48: 609-639.

Edwards G. E. and Walker D. A. (Eds.) (1983.) C3, C4: mechanisms, and cellular and environmental regulation of photosynthesis, Blackwell Scientific, Oxford.

Edwards G. R.; Clark H. and Newton P. C. D. (2001). The effects of elevated CO_2 on seed production and seedling recruitment in a sheep-grazed pasture. Oecologia. Vol. 127: 383–394.

Edwards G. E.; Franceschi V. R. and Voznesenskaya E. V. (2004). Single-cell C4 photosynthesis versus the dual-cell (Kranz) paradigm. Annual Review in Plant Biology Vol. 55: 173-196.

Ehleringer J. R.; Cerling T. E. and Helliker B. R. (1997). C4 photosynthesis, atmospheric CO_2, and climate. Oecologia. Vol. 112: 285-299.

Erice G.; Irigoyen J. J.; Sánchez-Díaz M.; Avice, J. C. and Ourry, A. (2007). Effect of drought, elevated $CO_{2,}$ and temperature on accumulation of N and vegetative storage proteins (VSP) in taproot of nodulated alfalfa before and after cutting. Plant Science. Vol. 172: 903–912.

Esau K. (1977). The Stem: Primary state of growth. Chapter 16. In: Anatomy of see plants, Edition 2. John Willey & Sons, Inc. New York, (pp 257-294).

FAO of the United Nations (FAO) (1992). Maize in Human Nutrition. Rome, Italy.

FAO (2006). World agriculture towards 2030/2050, Interim Report. Food and Agricultural Organization of the United Nations. FAO, Rome, Italy.

FAO (2016). Climate Change and Food Security: Risks and Responses; FAO: Rome, Italy.

FAOSTAT, (2009). Online at http://faostat.fao.org.

FAOSTAT (2016). Food Balance Sheet, Available: http://faostat.fao.org/site/345/default.aspx.

Fanadzo M.; Chiduza C. and Mnkeni P. N. S. (2010). Comparative performance of direct seeding and transplanting green maize under farmer management in small scale irrigation: A case study of Zanyokwe, Eastern Cape, South Africa. African Journal of Agricultural Research. (pp. 524-531).

Farfan-Vignolo E. R. and Asard H. (2012). Effect of elevated CO_2 and temperature on the oxidative stress response to drought in *Lolium perenne* L. and Medicago sativa L. Plant Physiology in Biochemistry. Vol. 59: 55–62.

Farnham D. E.; Benson G.O. and Pearce R. B. (2003). Maize perspective and culture. In: White, P. J., Johnson, L. A., (eds.) Maize: Chemistry and technology, Second Edition, (pp 1-33). American Association of Cereal Chemists, Inc. St. Paul, MN.

Fernando N.; Panozzo J.; Tausz M.; Norton R.; Fitzgerald G. and Khan A., *et al.* (2015). Rising CO_2 concentration altered wheat grain proteome and flour rheological characteristics. Food Chem. Vol. 170: 448–454.

Fierro A.; Gosselin A. and Tremblay N. (1994). Supplemental carbon dioxide and light improved tomato and pepper seedling growth and yield. Horticultural science. Vol. 29: 152–154.

Fleisher D.; Timlin D.; Reddy K.; Reddy V.; Yang Y. and Kim S. (2011). "Effects of CO_2 and temperature on crops: lessons from SPAR growth chambers," in Handbook of Climate Change and Agroecosystems, Volume 1 of ICP Series on Climate Change Impacts, Adaptation, and Mitigation, eds D. Hillel and C. Rosenzweig (London: Imperial College Press), (pp 55–86).

Florian A.; Timm S.; Nikoloski Z.; Tohge T.; Bauwe H.; Araújo W.L. and Fernie A.R. (2014). Analysis of metabolic alterations in Arabidopsis following changes in the carbon dioxide and oxygen partial pressures, Journal of Integrative Plant Biology. Vol. 56(9): 941-959.

Francesco N. T. and Frank E. (2002). Simulating the effects of elevated CO_2 on crops: approaches and applications for climate change. European Journal of Agronomy. Vol. 18: 57-74.

Frank Hamilton Cushing: Zuñi Breadstuff, Indian Notes and Monographs (1920). Vol.: 8: 36-37.

Fritsche-Neto R. and Silva P. S. L. Selection index for maize cultivars with dual aptitude: Maize and green maize. Bragantia. (2011). Vol. 70(4): 781-787.

Gardner H. W. and Inglett G.E. (1971). Food products from maize germ: enzyme activity and oil stability. Journal of Food Science. Vol. 36: 645–8.

Geissler N.; Hussin S. and Koyro H. W. (2010). Elevated atmospheric CO_2 concentration enhances salinity tolerance in Aster tripolium L. Planta. Vol. 231: 583–594.

Ghannoum O.; von Caemmerer S.; Ziska L. H. and Conroy J. P. (2000). The growth response of C4 plants to rising atmosphere CO_2 partial pressure: a reassessment. Plant Cell Environment. Vol. 23: 931–942.

Ghannoum O. (2009). C4 photosynthesis and water stress. Annals of Botany. Vol. 103: 635–644.

Ghannoum O.; Evans J.R. and von Caemmerer, S. (2011). Nitrogen and water use efficiency of C4 plants. In: Raghavendra, A.S. & Sage, R.S. (Eds.) C4 Photosynthesis and Related CO_2 Concentrating Mechanisms, Springer Science+Business Media B.V., Dordrecht, The Netherlands, (pp.129-146).

Gilbert Livingstone Wilson: Agriculture of the Hidatsa Indians: An Indian Interpretation, Univ. of Minnesota Studies in the Social Sciences No. 9, Minneapolis.

Goldworthy A. (1968). Comparison of the kinetics of photosynthetic carbon dioxide fixation in maize, sugarcane, and tobacco, and its relation to photorespiration. Nature. Vol. 217: 62.

Goufo P.; Pereira J.; Figueiredo N.; Oliveira M. B. P. P.; Carranca C.; Rosa E.A.S. and Trindade H. (2014). Effect of elevated carbon dioxide (CO_2) on phenolic acids, flavonoids, tocopherols, tocotrienols, gamma oryzanol and antioxidant capacities of rice (Oryza sativa L.), Journal of Cereal Science. Vol. 59(1): 15-24.

Gruda N. (2005). Impact of environmental factors on product quality of greenhouse vegetables for fresh consumption. Critical Review in Plant Science. Vol. 24: 227–247.

Gupta U.; Wum K. and Liang S. (2008). Micronutrients in soils, crops, and livestock, Earth Science Front. Vol. 15(5): 110-125.

Guzmán-de-Pena Doralinda (2010). The Destruction of Aflatoxins in Maize by Nixtamalización (PDF). In M. Rai; A. Varma (eds.). Mycotoxins in Food, Feed and Bioweapons. Berlin Heidelberg: Springer-Verlag, pp 39-49. Retrieved April 12, 2019.

HarvestPlus Brief (2006). HarvestPlus: breeding crops for better nutrition. Washington DC, USA.

Hatch M. D. (1987). photosynthesis a unique blend of modified biochemistry, anatomy, and ultra-structure. Biochimica et Biophysica Acta. Vol. 895: 81–106.

Henry R. and Kettlewell P. General Grain Quality (December 2012). Springer Science & Business Media. ISBN:9789400915138, (pp 97).

Hesketh J. and Hellmers H. (1973). Floral initiation in four plant species growing in CO_2-enriched air. Environmental Control in Biology. Vol. 11: 51–53.

Himali N.; Balasooriya Kithsiri B.; Dassanayake Bruce Tomkins; Saman Seneweera and Said Ajlouni. Journal of Horticultural Science and Research (2017), Vol. (1): 19 – 29.

Högy P. and Fangmeie A. (2009). Atmospheric CO_2 enrichment affects potatoes: 2. Tuber quality traits. European Journal of Agronomy. Vol. 30: 85–94.

Hotz C. (2013). Biofortification, In Benjamin Caballero (Eds), Encyclopaedia of Human Nutrition, 3rd edition. Academic Press, Waltham.

Huang B. and Xu Y. (2015). Cellular and molecular mechanisms for elevated CO_2 regulation of plant growth and stress adaptation. Crop Science Vol. 55: 1405.

Hussain M.; Kubiske M. E. and Connor K. F. (2001). Germination of CO_2-enriched Pinus taeda L. Seeds and subsequent seedling growth responses to CO_2 enrichment. Functional Ecology. Vol. 15: 344–350.

Idso S. B. and Idso K. E. (2001). Effects of atmospheric CO_2 enrichment on plant constituents related to animal and human health. Environmental and Experimental Botany. Vol. 45: 179–199.

IPCC, 2007: Climate Change (2007). Impacts, Adaptation and Vulnerability. Contribution of Working Group II to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change, M.L. Parry, O.F. Canziani, J.P. Palutik of, P.J. van der Linden and C.E. Hanson, Eds., Cambridge University Press, Cambridge, UK, (pp 976).

IPCC, 2014: Climate Change (2014): Synthesis Report. Contribution of Working Groups I, II and III to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change [Core Writing Team, R.K. Pachauri, and L.A. Meyer (Eds.)]. IPCC, Geneva, Switzerland, (pp 151).

Inglett G. E. (1970). Maize: Culture, Processing, Products. Major Feed and Crops in Agriculture and Food Science Series. 369 Seiten, 73 Abb., 65 Tab. The AVI Publishing, Inc., Westport, Connecticut.

Islam M.; Matsui T. and Yoshida Y. (1994). Effects of carbon dioxide enrichment on acid invertase and sugar concentration developing tomato fruit. Environmental Control Biology. Vol. 32: 245–251.

Khan I.; Vanaja M.; Sathish P. and Vagheera P. (2019). Impact of Elevated CO_2 on Two Successive Generations of CO_2 Responsive Maize Genotype. Agriculture Research Vol. 9: 310-315.

Jablonski L. M.; Wang X. and Curtis P. S. (2002). Plant reproduction under elevated CO_2 conditions: a meta-analysis of reports on 79 crop and wild species. New Phytologist. Vol.: 156: 9–26.

Jin C.; Du S.; Wang Y.; Condon J.; Lin X. and Zhang Y. (2009). Carbon dioxide enrichment by composting in greenhouses and its effect on vegetable production. Journal of Plant Nutrition Soil Science. Vol. 172: 418–424.

Jin H.; Li M.; Duan S.; Fu M.; Dong X.; Liu B.; Feng D.; Wang J. and Wang H. B. (2016). Optimization of Light Harvesting Pigment Improves Photosynthetic Efficiency, Plant Physiology. Vol. 172(3): 1720-1731.

Johannessen Carl L.; Wilson Michael R. and Davenport William A. (2018). The Domestication of Maize or Event? Article in Geographic Review–July 1970. https://www.researchgategate.net/publication/272540549.

Johnson L. A. (2000). Maize: the major cereals of the Americas. In: Kulp K. Ponte J.G. (eds) Handbook of cereal science and technology, 2nd edition. Dekker Inc, New York. (pp. 38).

Karowe D.N. and Grubb C. (2011). Elevated CO_2 increases constitutive phenolics and trichomes but decreases inducibility of phenolics in Brassica rapa (Brassicaceae), Journal of Chemistry Ecology. Vol. 37(12): 133240.

Katz S. H. and Hediger L. A. Valleroy (1974). Traditional Maize Processing Techniques in the New World. Traditional alkali processing enhances the nutritional quality. https://www.researchgate.net/publiccation/6057975.

Kessler J. R. and Armitage A. M. (1993). Effects of carbon-dioxide, light, and temperature on seedling growth of begonia x semperflorens-cultorum. Journal of Horticultural Science. Vol. 68(2): 281-287.

Khan I.; Azam A. and Mahmood A. (2013). The impact of enhanced atmospheric carbon dioxide on yield, proximate composition, elemental concentration, fatty acid, and vitamin C contents of tomato (Lycopersicon esculentum). Environ. Monit. Assess. Vol. 185: 205–214.

Khumalo T. P.; Schönfeldt H. C. and Vermeulen H. (2011). Consumer acceptability and perceptions of maize meal in Giyani, South Africa. Dev. South. Africa. Vol. 28: 271-281.

Kimball Bruce A. (1983). Carbon Dioxide and Agricultural Yield: An Assessment and Analysis of 430 Prior Observation. Agronomy Journal. Vol. 75: 75, 779.

Kimball Bruce A. (1993). Ecology of Crops in Changing CO_2 Concentration. Journal of Agricultural Meteorology. Vol. 48 (5): 559 – 566.

Kimball Bruce A.; Kobayashi H. and Bindi M. (2002). Responses of agricultural crops to free-air CO_2 enrichment. Advances in Agronomy Vol. (77): 293 – 386.

Kimball Bruce. A. (2016). Crop responses to elevated CO_2 and interactions with H2O, N, and temperature. Current Opinion in Plant Biology. Vol. 31: 36–43.

Kim H. Y.; Lieffering M.; Kobayashi K.; Okada M.; Mitchell M. W. and Gumpertz M. (2003). Effects of free- air CO_2 enrichment and nitrogen supply on the yield of temperature paddy rice crops. Field Crops Research. Vol. 83: 261 – 270.

Kluthcouski J. and Stone L. F. (2003). Main factors that interfere with the root growth of annual crops, with an emphasis on potassium. Piracicaba: Embrapa Rice and Beans. Agronomic Information. 103. Portuguese (Accessed August 13, 2018) Available: http://www.ipni.net/publication/iabrasil.nsf/0/8bd1bb43e687652283257aa20 05bee87/$file/page5-11-103.pdf.

Kramer P. J. (1981). Carbon dioxide concentration, photosynthesis, and dry matter production, Bioscience. Vol. 31: 29-33. https://doi.org/10.2307/1308175.

Lal R. (2004). Soil carbon sequestration impacts on global climate change and food security. Science. Vol. 304(5677): 1623-1627.

Lara M.V.; Casati P. and Andreo C.S. (2002). CO_2 concentration mechanisms in Egeria densa, a submersed aquatic species. Physiologia Plantarum. Vol. 115: 487-495.

Lara M.V. and Andreo, C.S. (2005). Photosynthesis in non-typical C4 species in: Pessarakli, M. (Ed.), Handbook of Photosynthesis: Second Edition” CRC press, Taylor & Francis Group. Boca Ratón, FL, USA, (pp 391-421).

Lara M. V. and Andreo C. S. (2011). C4 Plants Adaptation to High Levels of CO_2 and to Drought Environments. Abiotic Stress in Plants – Mechanisms and Adaptations, Prof. Arun Shanker (Ed.), ISBN: 978-953-307-394-1, In Tech, Available from http://www.intechopen.com/books/abiotic-stress-in-plantsmechanisms-and-adaptations/c4-plants-adaptation-to-high-levels-of-co2-and-to-drought-environments.

Lawlor D.W.; Kontturi M. and Young, A.T. (1989). Photosynthesis by flag leaves of wheat in relation to protein, ribulose bisphosphate carboxylase activity and nitrogen supply. Journal of Experimental Botany. Vol. 40: 43–52.

Leakey A. D. B.; Uribelarrea M.; Ainsworth E. A.; Naidu S. L.; Rogers A.; Ort D. R. and Long S. P. (2006). Photosynthesis, productivity, and yield of maize are not affected by open-air elevation of CO_2 concentration in the absence of drought. Plant Physiology. Vol. 140: 779–790.

Li F.; Wang J.; Chen Y.; Zou Z.; Wang X. and Yue M. (2007). Combined effects of enhanced ultraviolet – B radiation and doubled CO_2 concentration on growth, fruit quality and yield of tomato in winter plastic greenhouse. Frontiers in Biology China. Vol.: 2: 414-418.

Lin E.; Wei Xiong; Hui Ju; Yinlong Xu; Yue Li; Liping Bai and Liyong Xie. (2005). Climate change impacts on crop yield and quality with CO_2 fertilization in China. Philosophical Transaction B. The Royal Society Publishing London Book of Biology Science. Vol. 360 (1463): 2149–2154.

Liu R. H. (2004). Potential synergy of phytochemicals in cancer prevention: mechanism of action, Journal of Nutrition. Vol. 134: 3479s-3485s.

Loladze I. (2002). Rising atmospheric CO_2 and human nutrition: toward globally imbalanced plant stoichiometry? Trends in Ecology and Evolution. Vol. 17(10): 457-461.

Long S. P. and Drake B. G. (1991). Effect of the long-term elevation of CO_2 concentration in the field on the quantum yield of photosynthesis of the C3 sedge, *Scirpus Olneyi* Plant Physiology. Vol. 96: 221–226.

Long S.P.; Ainsworth E.A.; Rogers A. and Ort D. R. (2004). Rising atmospheric carbon dioxide: plants FACE the Future. Annual Review of Plant Biology. Vol. 55: 591–628.

Long S. P.; Ainsworth E. A.; Leakey A. D.; Nösberger J. and Ort D. R. (2006). Food for thought lower-than-expected crop yield stimulation with rising CO_2 concentrations. Science. Vol.: 312: 1918–1921.

Lucier G. and Dettmann R. L. (2008). Vegetables and melons situation and outlook yearbook, in: U. Economic Research Service (Eds.) Washington DC.

Mabesa R.; Impa S.; Grewal D. and Johnson-Beebout S. (2013). Contrasting grain-Zn response of biofortification rice (Oryza sativa L.) breeding lines to foliar Zn application, Field Crop Research. Vol. 149: 223-233.

Maroco Joâo P.; Gerald E. Edwards and Maurice S. B. (1999). Photosynthetic acclimation of maize to growth under elevated levels of carbon dioxide. Planta© Springer Verlag Vol. 210: 115 – 125.

Mouney J. R.; Guinn K. E.; Fry E. and Hesketh J. D. (1979). Photosynthesis Vol. 13: 260-266.

McCann J. (2005). Maize and grace. Africa's Encounter with a New World Crop, 1500–2000. Cambridge: Harvard University Press. 2005. Pp. xiii, 289.The American Historical Review. Vol. 111(5): 1640–1641.

Meng F.; Zhang J.; Yao F. and Hao C. (2014). Interactive Effects of Elevated CO_2 Concentration and Irrigation on Photosynthetic Parameters and Yield of Maize in Northeast China. PLOS ONE. Vol. 9(5): e98318.

Mestres C.; Louis-Alexandra A.; Matencio F. and Lahlou A. (1990). Dry – Milling properties of maize, Cereal Chemistry. Vol. 68: 51-56.

Micronutrient Initiative. (2004). Fortification handbook: vitamin and mineral fortification of wheat flour and maize meal. Available from: http://www.iaom.info/indlinks/Fort_handbook.pdf. Accessed Aug 2009.

Mishra A. K.; Rai R. and Agrawal S. (2013). Individual and interactive effects of elevated carbon dioxide and ozone on tropical wheat (Triticum aestivum L.) cultivars with special emphasis on ROS generation and activation of antioxidant defence system. Indian Journal of Biochemistry of Biophysics. Vol. 50: 139–149.

Mohan J. E.; Clark J. S. and Schlesinger W. H. (2004). Genetic variation in germination, growth, and survivorship of red maple in response to sub ambient through elevated atmospheric CO_2. Global Change Biology. Vol. 10: 233–247.

Moore B. D.; Cheng S. H.; Rice J. and Seemann J. R. (1998). Sucrose cycling, Rubisco expression, and prediction of photosynthetic acclimation to elevated atmospheric CO_2, Plant Cell Environment. Vol. 21(9): 905-915.

Moretti C. L.; Mattos L. M.; Calbo A. G. and Sargent S. A. (2010). Climate changes and potential impacts on postharvest quality of fruit and vegetable crops are view. Food Res. Int. Vol. 43: 1824–1832.

Mortensen L. M. (1994). Effects of elevated CO_2 concentrations on growth and yield of eight vegetable species in a cool climate. Science Horticulture. Vol. 58: 177–185.

Moureaux C.; Debacq A.; Bodson B.; Heinisch B. and Aubinet M. (2006). Annual net ecosystem exchange by a sugar beet crop. Agricultural and Forest Meteorology. Vol. 139: 25–39.

Moureaux C.; Debacq A.; Hoyaux J.; Suleau M.; Tourneur D.; Vancutsem F.; Bodson B. and Aubinet M. (2008). Carbon balance assessment of a Belgian winter wheat crop (Triticum aestivum L.). Global Change Biology. Vol. 4: 1–14.

Myers S. S.; Zanobetti A.; Kloog I.; Huybers P.; Leakey A. D.; Bloom A. J.; Carlisle E.; Dietterich L. H.; Fitzgerald G.; Hasegawa T.; Holbrook N. M.; Nelson R. L.; Ottman M. J.; Raboy V.; Sakai H.; Sartor K. A.; Schwartz Seneweera S.; Tausz M. and Usui Y. (2014). Increasing CO_2 threatens human nutrition, Nature. Vol. 510(7503): 139-42.

Nascimento Ana Marinho do; da Costa Franciscleudo Bezerra; da Silva Jéssica Leite; Brito Marcos Eric Barbosa; Gadelha Tatiana Marinho; Frade Luciano Jonatas Gomes; de Andrade Silva Luderlândio; dos Santos Anderson Formiga; da Silva Kátia Gomes; da Silva Márcio Santos and de Sousa Sátiro Larissa. (2018). Post-Harvest Quality of Bt Green Maize Produced in Different Water Harvesting Techniques with the Application of Rooting. Journal of Experimental Agriculture International. Vol. 27(1): 1-13. Article no. JEAI.44250. ISSN: 2457-0591. (Past name: American Journal of Experimental Agriculture, Past ISSN: 2231-0606).

NDA (National Department of Agriculture). n.d. Maize profile. Available at http://www.nda.agric.za/docs/FactSheet/maize.htm [accessed 19 June 2016].

NIH (National Heart, Lung, and Blood Institute). What is iron-deficiency anaemia? www.nhlbi.nih.gov. 26 March 2014. Archived from the original on 16 July 2017. Retrieved 17 July 2017.

Noguchi K.; Watanabe C. K. and Terashima I. (2015). Effects of Elevated Atmospheric CO_2 on Primary Metabolite Levels in Arabidopsis thaliana Col-0 Leaves: An Examination of Metabolome Data, Plant Cell Physiology. Vol. 56(11): 2069-78.

Nuss E.T. and S. A. Tanumihardjo. (2010). Maize: a paramount staple crop in the context of global nutrition. Comprehensive. Reviews in Food Science. Food Safety. Vol. 9: 417–436.

OECD (2006). “Section 3 - Maize (*Zea mays* subsp. mays)”, in Safety Assessment of Transgenic Organisms, Volume 1: OECD Consensus Documents, OECD Publishing, Paris.

Ortiz-Monasterio J.; Palacios-Rojas N.; Meng E.; Pixley K.; Trethowan R. and Pena R. (2007). Enhancing the mineral and vitamin content of wheat and maize through plant breeding, Journal of Cereal Science. Vol. 46(3): 293-307.

Owensby C. E.; Ham J. M.; Knapp A.; Rice C. W.; Coyne P. I. and Auen L. M. (1996). Ecosystem-level responses of tallgrass prairie to elevated CO_2. In "Carbon Dioxide and Terrestrial Ecosystems" (G. W. Kock and H. A. Mooney, eds.), (pp 147-162). Academic Press, San Diego.

Pajic Z. (2007). Breeding of maize types with specific traits at the maize research institute Zemune polje. Genetika Vol. 39: 169-180.

Pandey P.; Zinta G.; AbdElgawad H.; Ahmad A.; Jain V. and Janssens I. A. (2015). Physiological and molecular alterations in plants exposed to high CO_2 under phosphorus stress. Biotechnology Advances Vol. 33: 303–316.

Paliwal R. L. (2000). Maize Types. In: R. L. Paliwal, G. Granados, H. R. Lafitte, A. D. Vlollc, eds. Tropical maize: Improvement and Production. Food and Agriculture Organization of the United Nations Rome, (pp 39-43).

Pereira Filho I. A. (Ed.). O cultivo do milho-verde. Brasília: Embrapa Informação Tecnológica, (2003). (pp 204).

Pérez-López U.; Miranda-Apodaca J.; Lacuesta M.; Mena-Petite A. and Muñoz-Rueda A. (2015). Growth and nutritional quality improvement in two differently pigmented lettuce cultivars grown under elevated CO_2 and/or salinity. Scientia Horticulturae. Vol. 195: 56–66.

Pollak L. M. and White P. J. (1995). Maize as a food source in the United States: Part I; Historical and current perspectives. Cereal Foods World. Vol. 40: 749 – 754.

Poorter H. (1993). Interspecific variation in the growth response of plants to an elevated ambient of CO_2 concentration. CO_2 and Biosphere, Kluwer Academic Publishers, Dordrecht, Netherlands, Vegetation Vol. 104/105: 77 – 97.

Poorter H.; Roumet C. and Campbell B.D. (1996). Interspecific variation in the growth response of plants to elevated CO_2: A search for functional types. In Carbon Dioxide, Populations, and Communities (eds C. Körner & F.A. Bazzaz), (pp 375–412). Academic Press, New York.

Poorter H.; Berkel V. Y.; Baxter R.; Hertog J. D.; Dijkstra P. and Gifford R. *et al.*, (1997). The effect of elevated CO_2 on the chemical composition and construction costs of leaves of 27 C3 species. Plant Cell Environment. Vol. 20: 472-482.

Purseglove J. W. (1972). Tropical Crops: Monocotyledons Band 1 und 2, 1. Aufl. 1972. Longman Scientific and Technical New York. Food / Nahrung/ Vol. 19(5-6): 519-519.

Ragasa C., Dankyi A.; Acheampong P.; Wiredu A. N.; Chapoto A; Asamoah M. *et al.,* (2013). Patterns of Adoption of Improved Maize Technologies in Ghana. International Food Policy Research Institute, Working paper 36.

Rae A. M.; Ferris R.; Tallis M. J.; and Taylor G. (2006). Elucidating genomic regions determining enhanced leaf growth and delayed senescence in elevated CO_2. Plant, Cell & Environment. Vol. 29: 1730–1741.

Rengel Z.; Batten G. D. and Crowley D. E. (1999). Agronomic approaches for improving the micronutrient density in edible portions of field crops. Field Crops Research. Vol. 60: 27–40.

Reekie E. G. and Bazzaz F. A. (1991). Phenology and growth in four annual species grown in ambient and elevated CO_2. Canadian Journal of Botany. Vol. 69: 2475–2481.

Reeves D. W.; Rogers H. H.; Prior S. A.; Wood C. W. and Runion G. B. (1994). Journal of Plant Nutrition. Vol. 17: 939-1954.

Ritz T.; Damjanovic A.; Schulten K.; Zhang J. P. and Koyama Y. (2000). Efficient light harvesting through carotenoids, Photosynthesis research. Vol. 66(1-2): 125-44.

Rogers G. S.; Milham P. J.; Gillings M. and Conroy J. P. (1996). Sink strength may be the key to growth and nitrogen responses in N-deficient wheat at elevated CO_2. Australian Journal of Plant Physiology. Vol. 23: 253–264.

Rogers H. H.; Peterson C. M.; McCrimmon J. N. and Cure J. D. (1987). Agronomy Journal Vol. 105: 100.

Romeo-Bastida C. A.; Chávez Gutiérrez M.; Bello-Pérez L.A.; Abarca-Ramírez E.; Velazquez G. and Mendez-Montealvo G. (2018). Rheological properties of nanocomposite-forming solutions and film based on montomorillonite and maize starch with different amylose content, Carbohydr. Polym. Vol. 188: 121-127.

Rooney L. W. and Serna-Saldivar S. O. (1987). Food uses of whole maize: Chemistry and technology. St Paul., Minn., USA, Am. Association of Cereal Chemistry.

Roux F.; Touzet P.; Cuguen J. and Le Corre V. (2006). How to be early flowering: an evolutionary perspective. Trends in Plant Science. Vol. 11: 375–381.

Ruth Murray Underhill: Work-a-Day Life of the Pueblos (U. S. Indian Service, Phoenix Indian School Printing Dept., 1946.

Sage R. F.; Pearcy R. W. and Seemann J. R. (1987). The nitrogen use efficiency of C3 and C4 plants. III. Leaf nitrogen effects on the activity of carboxylating enzymes in Chenopodium album (L.) and Amaranthus retroflexus (L.). Plant Physiology Vol. 85: 355–359.

Sage R. F. (1994). Acclimation of photosynthesis to increase atmospheric CO_2, the gas exchange perspective. Photosynthesis Research. Vol. 39(3): 351-368.

Sage R. F.; Wedin D. A. and Li M (1999). The biogeography of C4 photosynthesis: patterns and controlling factors. In: Sage RF, Monson RK (eds) C4 plant biology. Academic, New York.

Sage R. F. (2002). Variation in the k(cat) of Rubisco in C3 and C4 plants and some implications for photosynthetic Performance at High and Low Temperature> Journal of Experimental Botany Vol. 53: 609-620.

Sage R. F. and Kubien D. S. (2003). Quo Vadis? An ecological perspective on global change and the future of C 4 plants. Photosynthesis Research. Vol. 77: 209–25.

Salem H. and El-Gizawy N. (2012). Importance of micronutrients and its application methods for improving maize (*Zea mays* L.) yield grown in clayey soil, American-Eurasian Journal of Agriculture and Environmental Science. Vol. 12(7): 954-959.

Scott C. E. and Eldridge A. L. (2005). Comparison of carotenoid content in fresh, frozen, and canned maize, Journal of Food Anals. Vol. 18: 551-559.

Serna-Saldivar S. O.; Gomez M. H. and Rooney L. W. (2000). Food uses of regular and specialty and their Dry- Milled. In Arnel R. H. (ed.) Specialty Maize 2nd edition.

Shashidara C. K. (2008). Early generation testing for combining ability in maize (*Zea mays* L). MSc. Thesis. University of Agricultural Science. Dharward- 580 005.

Shava, S.; O'Donoghue R.; Krasny M. E. and Zazu C. (2009). Traditional food crops as a source of community resilience in Zimbabwe. International Journal of African Renaissance. Vol. 4: 1-21.

Shimono Hiroyuki and Bunce James A. (2009). Acclimation of nitrogen uptake capacity of rice to elevated atmospheric CO_2 concentration. Annals of Botany. Vol. 103: 87 – 94.

Shukla G. N.; Kumar A.; Jha A.; Singh N.; Sharma P. and Singh J. *et al.*, FICCI PwC knowledge Report: Maize Vision 2022; 2013.

Sicher R. C. and Bunce A. J. (2015). The impact of Atmospheric CO_2 Concentrations on the Responses of Maize and Soybean to Elevated Growth Temperatures. R. Mahalingam (ed.), Combine Stresses in Plants.

Sihlobo W. (2014). South African maize market structure and East African export opportunities under the spotlight. SA Graan/Grain August 2014, (pp 88–89). Available at http://www.grainsa.co.za. [accessed 19 June 2016].

Sionit N.; Hellmers H. and Strain B. R. Agronomy Journal (1982). Vol. 74: 721-725.

Silva P. S. L.; Silva K. M. B.; Silva P. I. B.; Oliveira V. R. and Ferreira J. L. B. (2010). Green Ear Yield and Grain Yield of Maize Cultivars in Competition with Weeds[1]. Planta Daninha, Viçosa-MG, Vol. 28(1): 77-85.

Singh A. M. Agrawal. (2015). Effects of ambient and elevated CO_2 on growth, chlorophyll fluorescence, photosynthetic pigments, antioxidants, and secondary metabolites of Catharanthus roseus (L.) G Don. grown under three different soil N levels, Environmental Science Pollution Research. Vol. 22(5): 3936-46.

Smale M. and Jayne T. (2003). Mazie in Eastern and Southern Africa: "Seeds" of Success in Retrospect. Environment and Production Technology Division, International Food Policy Research Institute, 2033 K Street, N. W. Washington, D.C. 20006 U.S.A. EPTD Discussion Paper No. 97.

Smart D. R.; Ritchie K.; Bloom A. J. and Bugbee B. B. (1998). Nitrogen balance for wheat canopies (Triticum aestivum cv. Veery 10) grown under elevated and ambient CO_2 concentrations. Plant, Cell and Environment. Vol. 21: 753 – 763.

Smith A. M.; Zeeman S. C. and Smith S. M. (2005). Starch degradation. Annual Review of Plant Biology. Vol. 56: 73–89.

Smith B. (2006). The farming handbook. Pietermaritzburg: University of KwaZulu-Natal Press.

South Africa (2013). Millennium Development Goals – Country Report 2013, Pretoria, South Africa.

Springer Clint J. and Ward Joy K. Flowering time and elevated atmospheric CO_2. © The Authors (2007). Journal compilation © New Phytologist (2007). Vol. 176: 243-255.

Sreedevi Shankar K.; Vanaja M. and Jyothi Lakshni N. (2015). Effect of elevated atmospheric CO_2 concentration on nutrient quality of different maize genotypes. Scholars Journal of Agriculture and Veterinary Sciences. Vol. 2(1A): 9-12.

Sun Z.; Niinemets U.; Huve K.; Rasulov B. and Noe S. M. (2013). Elevated atmospheric CO_2 concentration leads to increased whole-plant isoprene emission in hybrid aspen (Populus tremula x Populus tremuloides), New Phytologist. Vol. 198(3): 788-800.

Suyker A. E.; Verma S. B.; Burba G. G.; Arkebauer T. J.; Walters D. T. and Hubbard K. G. (2004). Growing season carbon dioxide exchange in irrigated and rain fed maize. Agricultural and Forest Meteorology. Vol. 124: 1–13.

Suyker A. E.; Verma S. B.; Burba G. G. and Arkebauer T. J. (2005). Gross primary production and ecosystem respiration of irrigated maize and irrigated soybean during a growing season. Agricultural and Forest Meteorology. Vol. 131: 180–190.

Tallis M. J.; Lin Y.; Rogers A.; Zhang J.; Street N. R.; Miglietta F.; Karnosky D. F.; De Angelis P.; Calfapietra C. and Taylor G. (2010). The transcriptome of Populus in elevated CO_2 reveals increased anthocyanin biosynthesis during delayed autumnal senescence, New Phytol. Vol. 186(2): 415-28.

Tanumihardjo S. A. (2008). Food-based approaches for ensuring adequate vitamin A nutrition. Comprehensive Revolution of Food Science, Food Safety. Vol. 7: 373-381.

Taub D. R. and Wang X. (2008). "Why Are Nitrogen Concentrations in Plant Tissues Lower under Elevated CO_2? A Critical Examination of the Hypotheses," Journal of Integrative Plant Biology, Vol. 50 (11): 1365- 1374.

Tcherkez G. G. B.; Farquhar G. D. and Andrews T. J. (2006). Despite slow catalysis and confused substrate specificity, all ribulose bisphosphate carboxylases may be nearly perfectly optimized. Proc Natl Academy of Science. USA Vol. 103: 7246–7251.

Teng N.; Wang J.; Chen T.; Wu X.; Wang Y. and Lin J. (2006). Elevated CO_2 induces physiological, biochemical, and structural changes in leaves of Arabidopsis thaliana. New Phytologist. Vol. 172: 378–378.

Thurig B.; Körner C. and Stocklin J. (2003). Seed production and seed quality in a calcareous grassland in elevated CO_2. Global Change Biology. Vol. 9: 873–884.

Thompson A. M.; Brown R. A.; Rosenberg N. J.; Izaurralde R. C. and Benson V. (2005). Climate change impacts for the conterminous USA: an integrated assessment. Part 3. Dryland production of grain and forage crops. Climate Change. Vol. 69: 43–65.

Torbert H. A.; Prior S. A.; Rogers H. H. and Runion G. B. Field Crop Research Vol. 83 (2004): 57-67.

Turner J. S. and Brittain E. G. (1962). Oxygen as a factor in photosynthesis. Biological Reviews of Cambridge Philosophical Society. Vol. 37: 130-170.

United Nations (1992). United Nations Framework Convention on Climate Change. FCCC/INFORMAL/84.

Usman J.; Zalkuwi J.; Bakari U. M. and Hamman M. (2015). Economics of white maize production in fufore local government area of Adamawa State, Nigeria, International Journal of Scientific Research and Management. Vol. 3(2): 2159-2166.

Vanaja M.; Maheswari M.; Jyothi Lakshmi N.; Sathish P.; Yadav S. K.; Salini K.; Vagheera P.; Vijay Kumar G. and Abdul Razak. (2015). Variability in Growth and Yield Response of Maize Genotypes at Elevated CO_2 Concentration. Advances in Plants & Agriculture Research. Vol. 2.

van Averbeke W.; M'marete C. K.; Igodan C. O. and Belete A. (1998). An investigation into food plot production at irrigation schemes in Central Eastern Cape. WRC Report 719/1/98. Water Res. Commission, Pretoria, South Africa.

van der Kooi C. J.; Reich M.; Löw M.; De Kok L. J. and Tausz, M. (2016). Growth and yield stimulation under elevated CO_2 and drought: a meta-analysis on crops. Environmental and Experimental Botany. Vol. 122: 150–157.

Vieira E. L. and Santo S. C. M. G. (2005). Plant stimulant in the growth and initial development of the cotton root system in rhizotrons. V Brazilian Cotton Congress. Embrapa Cotton; (Accessed on August 20, (2018). Available: http://www.cnpa.embrapa.br/prod utos/algodao/publicacoes/trabalhos_cba5/ 161.pdf.

Vodni D.; Pfanz H.; Wittman C.; Maèek I.; Kastelec D.; Turk B. and Batiè F. (2002). Photosynthetic acclimation in plants growing near carbon dioxide spring, Phyton; Annales Rei Botanicae "Responses of Plant Metabolism to Air Pollution and Global Change". Vol. 42: 239-244.

Vodnik D.; Šircelj H.; Kastelec D.; Maček I.; Pfanz H. and Batič F. (2005). The Effects of Natural CO_2 Enrichment on the Growth of Maize. Journal of Crop Improvement Vol. 13: 193 – 212.

Dreamstime. All rights reserved. Copyright © 2000-2021. https://www.dreamstime.com/maize-plant-diagram-infographic-elements-parts-corn-plant-anthers-tassel-corn-ears-cobs-roots-stalks-maize-plant-image137290027 (accessed 25/3/2020)

Wand S. J. E.; Midgley G. F.; Jones M. H. and Curtis P. S. (1999). Responses of wild C4 and C3 grass (Poaceae) species to elevated atmospheric CO_2 concentration: a test of current theories and perceptions. Global Change Biology. Vol. 5: 723–741.

Wang M.; Xie B.; Fu Y.; Dong C.; Hui L, Liu G. and Liu H. (2015). Effects of different elevated CO_2 concentrations on chlorophyll contents, gas exchange, water use efficiency, and PSII activity on C3 and C4 cereal crops in a closed artificial ecosystem, Photosynthesis research. Vol. 126(2-3): 351-62.

Ward J. K. and Kelly J. K. (2004). Scaling up evolutionary responses to elevated CO_2: lessons from Arabidopsis. Ecology Letters. Vol. 7: 427–440.

Ward J. K.; Antonovics J.; Thomas R. B. and Strain B. R. (2000). Is atmospheric CO_2 a selective agent on model C3 annuals. Oecologia. Vol. 123: 330–341.

Way D. A.; Ghirardo A.; Kanawati B.; Esperschutz J.; Monson R. K.; Jackson R. B.; Schmitt P. and Kopplin Schnitzler J. P. (2013). Increasing atmospheric CO_2 reduces metabolic and physiological differences between isoprene- and non-isoprene-emitting poplars, New Phytol Vol. 200(2): 534-46.

Weatherspoon D. and Reardon T. (2003). The rise of supermarkets in Africa: implications for agri-food systems and the rural poor. Development Policy Review Vol. 21: 333–356.

Wilson Gilbert Livingstone: Agriculture of the Hidatsa Indians: An Indian Interpretation, University of Minnesota Studies in the Social Sciences No. 9, Agriculture of the Hidatsa Indians: an Indian interpretation.

Whistler R. L. (1970). Industrial uses of maize starches. In: G. E. Inglett, editor. Maize: culture, processing, products major fred and food crops in agriculture and food series. Westport, Connectictut.: Avi Publishing, (pp 171-94).

Xiaojin Xie; Renying Li; Yaohong Zhang; Shuanghe Shen and Yunxuan Bao. (2018). Effect of Elevated [CO_2] on Assimilation, Allocation of Nitrogen and Phosphorus by Maize (*Zea mays* L.). Communications in Soil Science and Plant Analysis, Vol. 49(9): 1032–1044.

Yeoh H. H.; Badger M. R. and Watson L. (1980). Variations in K_m (CO_2) of ribulose-1,5-bisphosphate carboxylase among grasses. Plant Physiology. Vol. 66: 1110–1112.

Young K. J. and Long S. P. (2000). Crop ecosystem responses to climatic change: maize and sorghum. In: Reddy KR and Hodges HF (Eds.), Climate change and global crop productivity, CABI International, Oxon, United Kingdom, (pp. 107-131).

Zavaleta E. S. (2006). Shrub establishment under experimental global changes in a California grassland. Plant Ecology. Vol. 184: 53–63.

Zinta G.; Abdelgawad H.; Domagalska M. A.; Vergauwen L.; Knapen D. and Nijs I. *et al.*, (2014). Physiological, biochemical, and genome-wide transcriptional analysis reveals that elevated CO_2 mitigates the impact of combined heat wave and drought stress in Arabidopsis thaliana at multiple organization levels. Global Change in Biology. Vol. 20: 3670–3685.

Ziska Lewis H. and Bunce James. A. (1997). The role of temperature in determining the stimulation of CO_2 assimilation at elevated carbon dioxide concentration in soybean seedlings. Physiologia Plantarum Vol. 100: 126-132.

Ziska L.; Panicker S. and Wojno H. (2008). Recent and projected increases in atmospheric carbon dioxide and the potential impacts on growth and alkaloid production in wild poppy (Papaver setigerum DC.), Climatic Change. Vol. 91(3-4): 395-403.

Zhao X. Y.; Zhang C.; Guigas Ma Y.; Corrales M.; Tauscher B. and Hu X. S. (2009). Composition, antimicrobial activity, and antiproliferative capacity of anthocyanin extracts of purple maize (*Zea* mays L.) from China, Europe Food Research Technology. Vol. 228: 759-765.

Chapter 2

Diurnal variations in CO_2 concentration and soil moisture in pot-grown sweetcorn (*Zea mays* L. var. *saccharata*)

Abstract

The current escalation in the atmospheric CO_2 as part of the current climate change has resulted in altered precipitations patterns, causing a decline in crop production and yield. Investigations on plant responses to elevated CO_2 concentration are plentiful, but little is known on the effect of elevated CO_2 under greenhouse conditions. To simulate slightly enhanced CO_2 conditions, the effect of mycelium CO_2 bags, installed above maize plants grown in pots in a temperature-controlled greenhouse, resulting in an ambient CO_2 (aCO_2) of ~430 ppm and eCO_2 of ~500 ppm concentrations. The concentrations of CO_2 varied with distance from the mycelium CO_2 bags, while soil moisture did not differ between treatments and the control.

There were delays in the days to emergence for a cultivar 'Assegai' resulting from the sowing in the winter season followed by lodging of maize plants which ultimately lead to the death of a control under aCO_2. Subsequently, 'Assegai' treatment had no data available for a control to be compared with on growth and development, while the effect of eCO_2 on STAR7719 treatment was positive. The effect of eCO_2 on days to emergence (DTE) for 'Assegai' and 'STAR7719' treatments were significantly different at ($P<0.003$) with 'Assegai' having mean of 12.11 and 'STAR7719' with a mean of 7.9 under eCO_2, while, under aCO_2 the means for 'Assegai' and 'STAR7719' were 25.44 and 9.22. Leaf area for 'STAR7719' was not significant different with means of 179 under eCO_2 over 174.3 under aCO_2. Leaf number per plant and plant height were both significant under eCO_2 at ($P<0.001$). Significant differences were also found in days to tasselling, anthesis and silking at ($P<0.001$). Fresh and dry above ground biomass were highly significant at ($P<0.005$ and 0.009). Root fresh and dry mass were also significant at ($P< 0.002$ and 0.018). the ear mass and fresh ear length were significant at ($P< 0.05$ and 0.013). Nevertheless, fresh cob mass was not significant at ($P=0.140$), however, a tendency to increase was recorded.

2.1 Introduction

The current increase in atmospheric CO_2 is predicted to exceed 1100 ppm by the 21st century (Booth *et al.,* 2017). This catastrophic climate change would likely result in temperature rise of 2 to 4^{o}C by the year 2100 (Lin *et al.,* 2005). Nevertheless, carbon dioxide's natural availability in the atmosphere forms part of the Earth's carbon cycle (Prasad *et al.*, (2003). Additionally, CO_2 is a good plant nutrient with its enrichment projected to have positive impact on crop growth and productivity (Prasad *et al.,* 2003). Such growth of crops is due to exposure to elevated carbon dioxide (eCO_2) concentrations from a type of CO_2 fertilization, leading to an increased biomass production and higher yield (Mortensen, 1987).

According to Muller *et al.* (2011) the influence of climate change on agricultural crop production and productivity differs from one place to another, and from crop to crop (Tubiello *et al.,* 2002). One of the most significant changes in recent decades is the atmospheric CO_2 which played a significant role in global warming. This has led to a stimulation in research studies to determine the probable effects of eCO_2 on agricultural productivity of crops as well as natural ecosystems (Dahlaman *et al.*, 1985). Previous research on the plant response to eCO_2 has been conducted in various systems, including growth chambers, greenhouses, controlled environmental chambers, phytotrons, open-top chambers (OTCs), and free air carbon dioxide enrichment (FACE) facilities (Vanaja *et al.,* 2006).

Atmospheric CO_2 enrichment has been investigated for over a century, particularly for studying the effect of CO_2 gas supplementation on plant growth and development (Drake *et al.*, 1985; Enoch and Kimball, 1986; Schulze and Mooney, 1993; Uprety *et al.*, 2000). Many studies have failed to accurately maintain the CO_2 concentration within the chambers, simply because of technical limitations and the fact that in an enclosed structure the typical environment is not identical to open field conditions with free airflow (Kimball *et al.,* 1997).

Studies have further outlined the limited root growth, in such experiments, as plants are placed in pots in controlled environment chambers (Vanaja *et al.*, 2005). Consequently, attempts have been implemented to employ structures that mimic natural conditions, such as that of open fields, while maintaining eCO_2 throughout the experimental duration which lead to the development of OTCs (Vanaja *et al.*, 2005). The aim of this chapter, is therefore, to compare the environmental conditions between two glasshouses, one with ambient 430 ppm CO_2 (aCO_2) and an adjacent one with enhanced elevated 500 ppm CO_2 (eCO_2). Elevated CO_2 was produced in the one glasshouse through the use of the mycelium CO_2 powered generator bags.

2.2 Materials and methods

2.2.1 Growing conditions and plant material

The experiment was carried out at the University of KwaZulu-Natal, Pietermaritzburg, in two adjacent glasshouses. The glasshouses were well sealed with duct tape (OMEGA - Tapd48W 48m x 25mm WHITW Duct Tape). The CO_2 enrichment was supplemented before the emergence of the seedlings by using Mycelium CO_2 powered generator bags. These bags were meant to maintain CO_2 concentrations at ~500 ppm while, an aCO_2 was at ~430 ppm for the controls. The Mycelium CO_2 Generator bags are a patented strain of mycelium producing CO_2 gas without producing fruiting bodies (Because Nature Mycelium CO_2 Generator bags, Windell Hydroponics.co.za, Cape Town, SA). The Mycelium CO_2 Generator bags fully colonise, in so doing giving off CO_2 gas to the sweetcorn plants while taking in O_2 which is produce as a by-product by the plants. The effectiveness of these bags in supplementing the CO_2 concentration in the glasshouse was monitored with the intention to record potential, effects on sweetcorn growth and development as well as biomass.

Agrometeorological equipment was used to monitor the concentration of CO_2 in the glasshouses continuously throughout the growing season. The monitoring system consisted of a closed path system, developed to measure and control the CO_2 (LI-84OA, Li-Cor, Lincoln, Nebraska USA) concentrations in the glasshouses. Data was collected and stored in a data-logger (CR1000, Campbell Science, Logan Utah USA). A CO_2/H_2O calibration was performed using a dew point generator (LI-610) Li-Cor, with CO_2 and H_2O scrubs. The air intake of the LI-84OA was controlled by using solenoid valves (SCE210C093, Diya Valves International, RSA) that switched between the glasshouses every 15 min.

The environmental variables in both glasshouses such as the temperature and relative humidity (RH) were measured with agrometeorology tools. The solar radiance for both glasshouses was natural and ranged between 200-425 W/m^2 depending on a day's photoperiodic light. Prior to planting, the physicochemical properties of soil from three soil samples were analysed (Tables 2.3, 2.4, and 2.5.) as well as soil moisture percentages for the two sets of four consecutive weeks (Appendices 8 and 9) recorded.

Seeds were sown in 30 cm diameter (10 litre) plastic pots in both glasshouses. Two locally obtained hybrid cultivars used were, 'Assegai' and 'STAR 7719' sweetcorn from Starke Ayers. Each glasshouse had 18 plants where each cultivar replicated 9 times in 3 rows. The spacing between the rows was 15cm respectively. Sweetcorn plants were irrigated at regular intervals of two to three times a week to prevent stress and to prevented diseases that exist when plants are excessively wet. Biological weed control was done without application of herbicides. Soil moisture percentages were recorded weekly to determine if there were any significant differences in the water use efficiency (WUE).

2.2.2 Soil analysis

Table 2.1 Soil fertility of the used potting medium mixed with local soil was analysed from three samples (A, B, C) at the ARC-Cedara, Pietermaritzburg as described by (Manson and Roberts, 2000), and the physicochemical properties shows the pH of the soil was not acidic and required no lime application.

Sample ID	Lab number	Sample density g/mL	P gm/L	K mg/L	Ca mg/L	Mg mg/L	Exch. Acidity cmol/L	Total cations cmol/L	Acid sat. %	pH (KCl)	Zn mg/L	Mn mg/L	Cu mg/L
A	F8548	0.82	84	669	1949	639	0.04	16.74	0	5.16	17.9	18	2.7
B	F8549	0.78	82	639	1854	620	0.05	16.03	0	5.18	17.7	15	2.9
C	F8550	0.82	81	629	1901	627	0.03	16.29	0	5.19	16.6	17	3.0

Table 2.2 Nutrient and lime recommendations after the three soil samples (A, B, C) have been analysed

Sample ID	**Lab Num.**	**NITROGEN**		**PHOSPHORUS**			**POTASSIUM**			**LIME**				**ZINC**
		Yield target t/h	**Req. N Kg/h**	**Sample soil test mg/L**	**Target soil test mg/L**	**Req. P kg/ha**	**Sample soil test mg/L**	**Target soil test mg/L**	**Req. K kg/ha**	**Sample acid sat. kg/ha**	**PAS %**	**Req. Lime t/ha**	**Lime type**	**Zinc fert. Req.?**
A	F8548	12.0	200	84	12	20	669	120	0	0	20	0	-	No
B	F8549	12.0	200	82	12	20	639	120	0	0	20	0	-	No
C	F8550	12.0	200	81	12	20	629	120	0	0	20	0	-	No

Mineral nutrients supplementation was applied to the soil according to local fertiliser practices based on soil analysis results (Table 2.4). Recommended nitrogen 60 kg N/ha and minor and micronutrients such as calcium magnesium nitrate doses were applied to maize plants (30 kg $CalMgNO_3$/ha) as a second dose for the reason that these micronutrients leach during irrigation. Lime was not recommended based on the soil's power of hydrogen as described by (Sørensen, 1909) not being acidic and having the average pH of 5,17 from the analysed samples.

Table 2.3 The physicochemical properties of the soil in which the soil organic matter and nitrogen had very small percentage, and the soil is a heavy clay loam type.

Sample ID	Lab number	Mid-infrared Estimates		
		Org. C %	N %	Clay %
A	F8548	3.4	0.25	19
B	F8549	4.6	0.35	15
C	F8550	4.0	0.35	18

2.3 Results

GenStat® 18th edition (VSN International, Hemel Hempstead, UK) was used to conduct the statistical analysis through the analysis of variance (ANOVA), and Fisher's protected LSD which revealed significant differences between the selected sweetcorn cultivars 'Assegai' and 'STAR7719' in terms of water use efficiency under different CO_2 growing conditions. In (appendices 8 and 9) for the eight consecutive weeks on water use efficiency indicated significant differences at $P<0.001$ (appendix 10) for cultivar by CO_2 interaction and CO_2 by cultivar interaction per week was also significant at $P<0.001$ for the initial four weeks. Furthermore, significant differences were also recorded at $P<0.001$ for the cultivar by CO_2 interaction and CO_2 by cultivar interaction per week on the second set of four weeks.

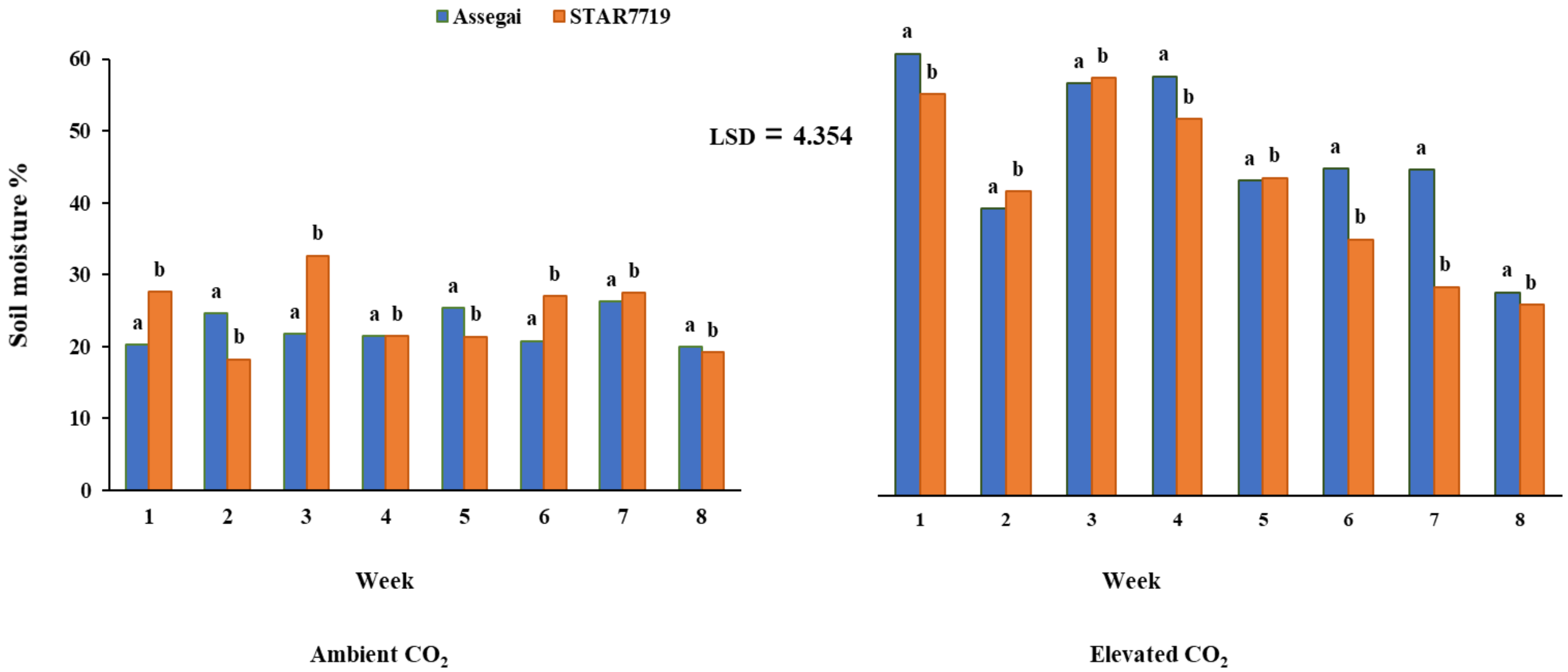


Fig 2.1 Mean in soil moisture percentage measured for eight consecutive weeks under ambient 430 ppm CO2 and elevated 500 ppm CO2 concentrations for sweetcorn cultivars 'Assegai' and 'STAR7719' in two glasshouses. Letters 'a' indicates 'Assegai' and 'b' indicates 'STAR7719'.

(F = 5.15; df = 8; P<0.001)

It can be observed from the statistical results that eCO_2 enrichment effect was smaller with rather enhanced significant effect on the whole crop's water use, these findings slightly agree with that of Conley *et al.* (2001) which in their study, they observed smaller effect from eCO_2 concentrations with significant effect on plant water use. The treatments under eCO_2 had slightly increased in absorbing water from the soil and faster growth resulting in taller maize plants. On the contrary, it was recounted that eCO_2 enhanced the ability to avoid drought stress sustaining soil-water, yielding little internal water shortages that are due to the soil-water content being exhausted (Wall *et al.,* 2001).

2.4. Assimilation response of sweetcorn to aCO_2 and eCO_2 concentrations at bottom height of 50cm

2.4.1 Response of maize plants to eCO_2 during cooler and hot days

2.4.1.1 Overcast day with lower temperatures

On an overcast day sweetcorn response to eCO_2 in glasshouse with aCO_2 in the early morning hours, shows that the temperature shown in Fig. 2.2 at 19,1^{o}C had little influence on maize plants to assimilate eCO_2 at the concentration of 431 ppm in the presence of 14,5 W/m^2 solar radiance. However, during mid-morning hours, the temperature gradually increased to 24,45^{o}C, in the presence of enhanced solar radiance of 113,5 W/m^2, enabled the sweetcorn plants to slightly assimilate 9 ppm from a concentration ranging between 442 to 439 ppm in very low amounts with a slight significant difference between the mid-morning and midday hours in eCO_2 concentrations owing to the lower temperatures and low light intensity.

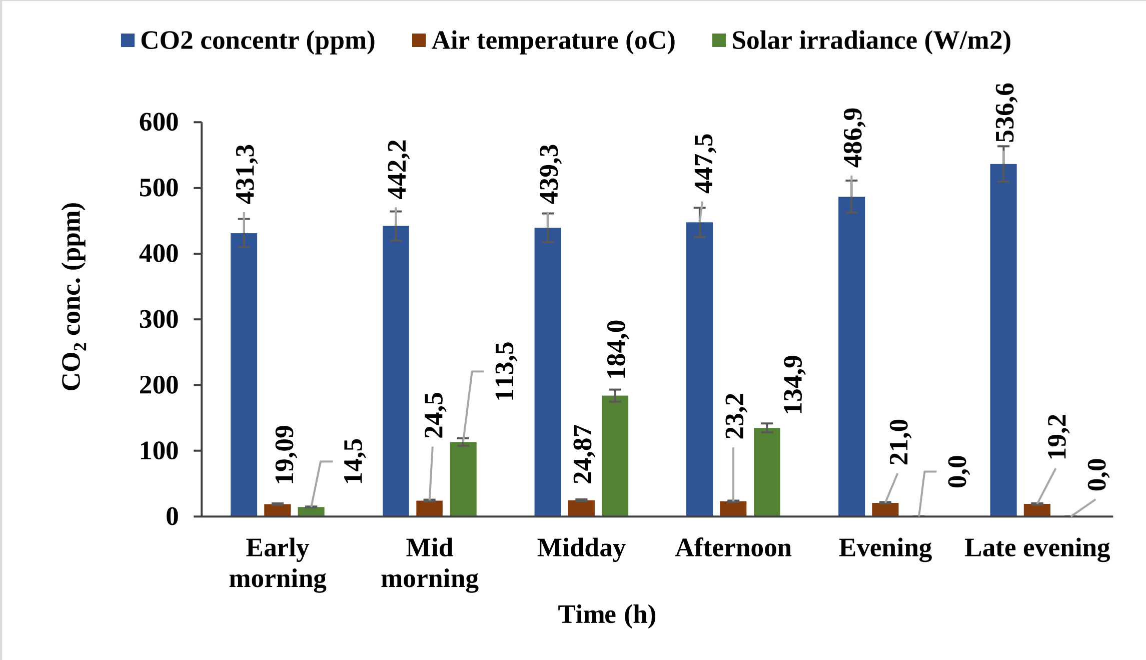


Figure 2.2 Sweetcorn plants treatments grown under eCO_2 concentration in a cooler day under the influence of air temperature (oC) and solar radiance (W/m2) in glasshouse with aCO_2.

In the midday hours, there was a slight assimilation of eCO_2 at least in small amounts due to little effect of solar radiance at 180,0 W/m^2 and temperature at 24,87^{o}C. Lower temperature as well as low light intensity had little or no effect on the sweetcorn plants to assimilate eCO_2 concentration, and gradually resulted in slightly higher concentrations in CO_2 due to maize plant respiration and temperature declining from 20,96 to 19,18^{o}C, as well as eCO_2 elevating from 487 to 536 ppm. A correlation between temperature and solar radiance is quite a noticeable characteristic of plant's response to eCO_2. Having lower temperature and lower solar radiance restricted maize plant's ability to assimilate and absorb CO_2 in higher amounts, although there was a reduction in CO_2 between mid-morning and midday hours.

2.4.2 Response of maize plants to aCO_2 in hot and cooler days

2.4.1.2 Sunny day temperatures

On a sunny day the relative variations in CO_2 concentrations are due to the availability of solar radiance and higher temperatures. In the early morning and mid-morning hours, the temperature illustrated in Fig. 2.3 varied significantly with values between 23,1 to 34,7^{o}C, and solar radiance with values varying significantly between 77,4 to 312,2 W/m^2 which resulted in sweetcorn plants assimilating an amount of 18 ppm of eCO_2 in generous amounts from concentrations between 458 to 439 ppm with a noticeable significant decline. Solar radiance increased by 56,6 W/m^2 from 312,2 to 368,8 W/m^2 as temperature from 34,7 to 39,3°C by 4,56 °C increased from mid-morning to midday encouraging sweetcorn plants to assimilate and absorb eCO_2 in higher concentrations (Fig. 2.3) between 439 to 412 ppm to intensely photosynthesise causing a considerable decline in CO_2 concentration.

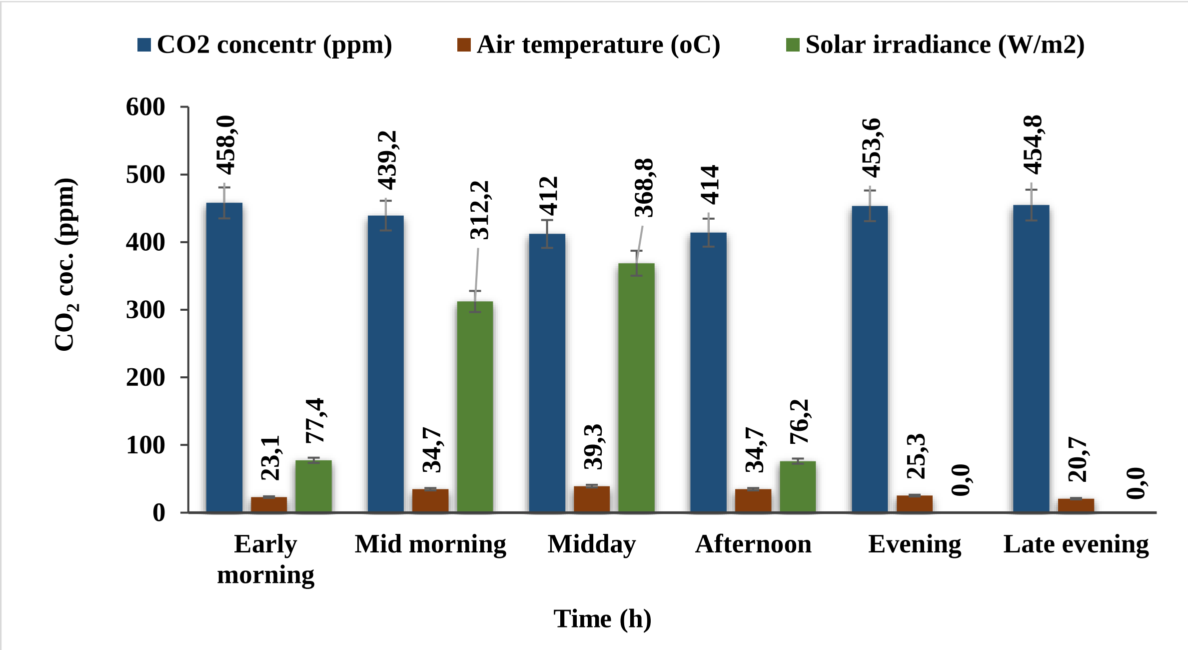


Fig. 2.3: Sweetcorn plants treatments grown under eCO2 concentration in a hot day under the influence of air temperature (oC) and solar radiance (W/m2) in glasshouse with eCO2.

A slight increase in eCO_2 concentration occurred in the afternoon hours as the temperature declines from midday 39.26°C to afternoon with 34.7°C accompanied by a decrease in solar radiance in midday with 368.8 W/m^2 to 76,2W/m^2 in the noon. In the evening hours significant declines in temperature and absence of light intensity, elevated the CO_2 concentration due to respiration from maize plant releasing CO_2 as a by-product. On a sunny day, the solar radiance as well as higher temperatures positively influence the assimilation of eCO_2 by maize plants, consequently, photosynthesis is enhanced. There is correlation between temperature and solar radiance, a noticeably enhanced assimilation in CO_2 is strongly influenced by an increased in temperature as well as solar radiance.

2.4.2.1 Overcast day with lower temperatures

In a cooler day sweetcorn plant's response to ambient CO_2 (aCO_2) in chamber 9 had significantly different result from because sweetcorn plants depicts lower assimilation and absorption of aCO_2 concentration of 434 to 434 ppm in Fig. 2.4 in a glasshouse with natural environment, between the early morning and mid-morning a relatively low temperature of 19,7 ^oC had limited effect on the sweetcorn crop to assimilate aCO_2, seeing that the concentration of aCO_2 had very little or no change during a significantly low solar radiance of 9,2 W/m^2 (Fig. 2.4). Clearly, there a significant contrast is observed in the assimilation of aCO_2 during hot and cooler days in the presence of lower air temperature and solar radiance.

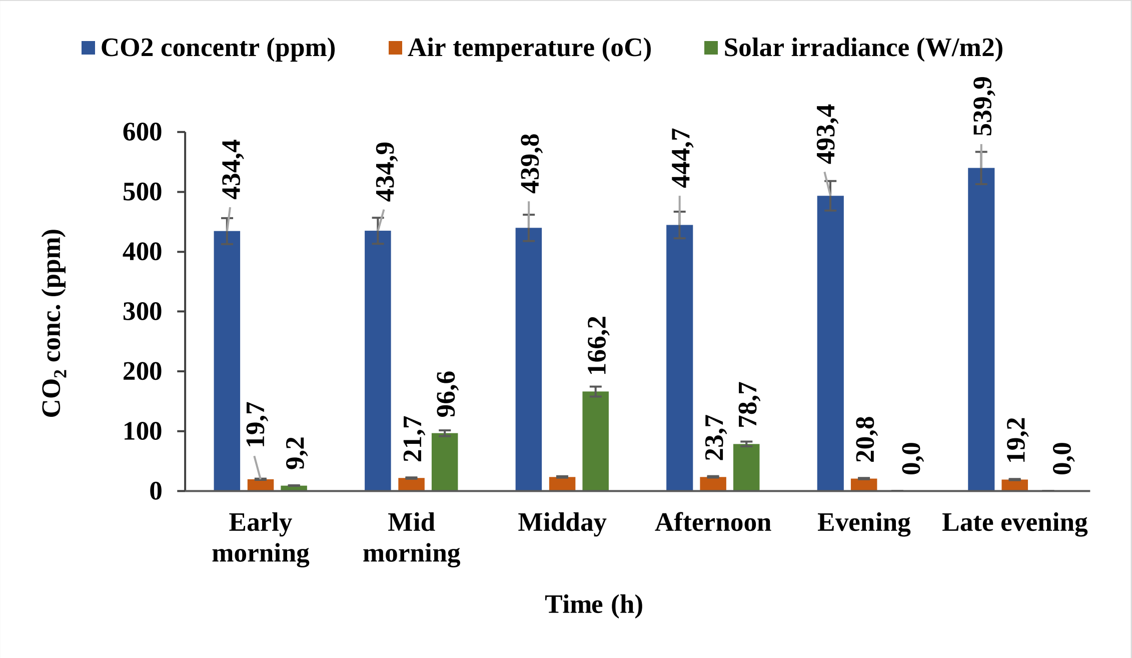


Figure 2.4 Sweetcorn plants controls grown under aCO_2 concentration in a cooler day under the influence of air temperature (^oC) and solar radiance (W/m^2) in glasshouse with aCO_2.

A slight rise in the air temperature from mid-morning, midday and afternoon with temperatures varying from 21,7; 23,6; and 23,7^oC, furthermore, the presence of solar radiance exemplified in Fig. 2.4 varying from 96,6; 166,2; and 78,7 W/m^2 in the same sets of hours had no significant effect on the assimilation and absorption of aCO_2 by sweetcorn crop, seeing that the concentrations from midday to the afternoon seemed to have slightly increased instead of declining. This scenario shows an importance of higher air temperature as well as higher solar radiance for maize plants to assimilate CO_2 are significantly important. Subsequently, as a C4 plant, sweetcorn exhibits a positive response to CO_2 assimilation in the presence of higher temperature and solar radiance (Ghannoum *et al.*, 2000). Throughout the evening hours, a decline in the air temperature and the absence of solar radiance had no effect on the assimilation of aCO_2 by maize crop, but an increase in the aCO_2 concentration was observed a little higher from evening to late by 46 ppm produced as a by-product by maize crop.

2.4.2.2 Sunny day temperatures

During a hot day a relatively different assimilation of CO_2 was observed in which sweetcorn plants response differently to ambient CO_2 (aCO_2) in a chamber with natural environment. In the early sunrise hours, the air temperature in Fig. 2.5 differed significantly from the mid-morning air temperature, influencing the photosynthesis process under the ambient CO_2 (aCO_2) concentration that went from 446 ppm declining significantly to 423 ppm in between the early and early morning hours. As the air temperature improved from mid-morning with a range of 32,1 to 36,4°C towards mid-morning hours and declined to 28,9 towards the afternoon, the solar radiance was significantly enhanced from 73,2 to 244,3 W/m^2 enabling the sweetcorn controls to assimilate and absorb aCO_2 in significantly small amount that ranged from 416 to 426 ppm.

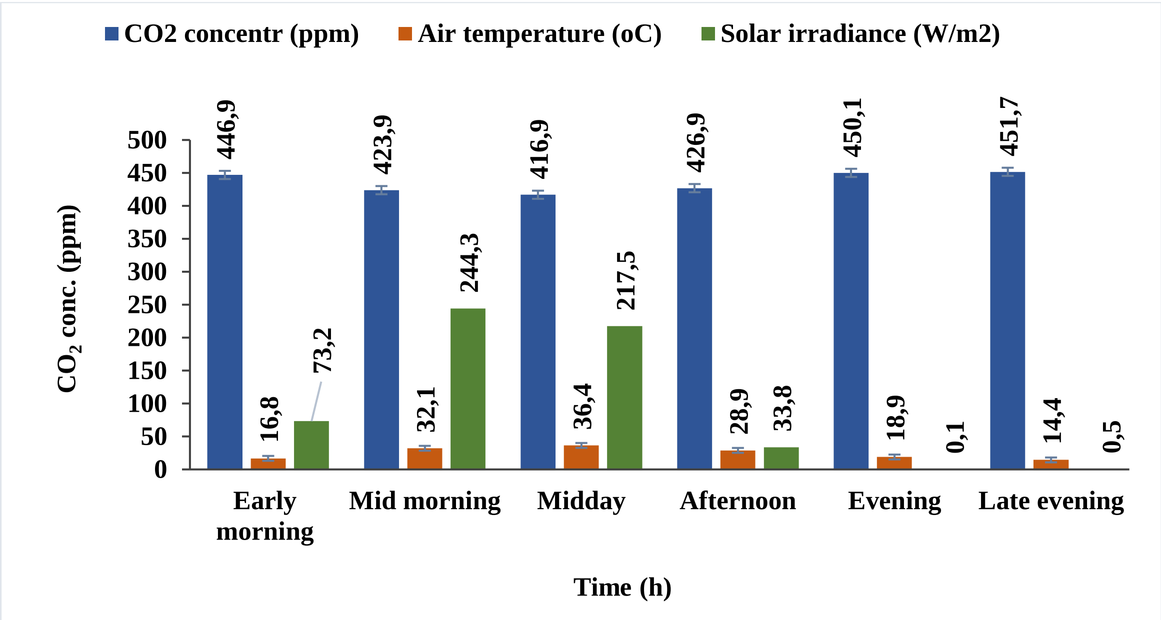


Fig. 2.5 Sweetcorn plants controls grown under eCO_2 concentration in a hot day under the influence of air temperature (°C) and solar radiance (W/m2) in glasshouse with aCO_2.

Likely so, Ghannoum *et al*, (2011) reasoned that at lower temperatures of 25 to 28°C, the photosynthesis rate in C4 plants is less efficient than in C3 plants photosynthesis grown under conditions with limiting light. Seemingly, in the afternoon and during the evening hours, it is observed that during the absence of light declining from 217,5 to 33,8 W/m^2 as well as a decline in air temperature from 28,9 to 18,8^{o}C, sweetcorn plants do not photosynthesise, and subsequently leading to the production of CO_2 as a by-product due to respiration which slightly increased by 25 ppm.

There is seemingly a strong correlation between the air temperature and solar radiance, simply because their association encourage the rise in the CO_2, therefore, the higher the air temperature and the solar radiance, the slightly higher the CO_2, and the assimilation of CO_2 by sweetcorn plants is increased. The solar radiance is too considered a high paramount component because it improves photosynthesis under elevated CO_2 conditions especially in C4 plants grown under high solar radiance, while effect it is not so prominent when grown under low solar radiance (Ghannoum *et al.,* 2000).

2.5 Assimilation response of maize plants grown under eCO_2 concentration in a glasshouse with eCO_2 at three different heights (50 cm, 100 cm and 150 cm) variations

2.5.1 Elevated CO_2 at height 50 cm below CO_2 mycelium bags

The CO_2 tube used for measuring CO_2 at different heights below the Mycelium CO_2 bags was used to measure the eCO_2 to determine the exact concentration of eCO_2 since it is a gas heavier than air. The eCO_2 gas ranged from 430 to 500 ppm, whereas the aCO_2 concentration ranged from 400 to 430 ppm. The tube was placed at different heights for 24 hours to determine the rate of assimilation and absorption by sweetcorn plants. At 50 cm height below the Mycelium CO_2 bags, the concentration of CO_2 in the early and mid- morning hours was between 443,7 and 418 ppm, maize plants actively assimilated CO_2 seeing that they were able to sequestrate a concentration of 25 ppm and resulted in a significant decline in the CO_2 concentration.

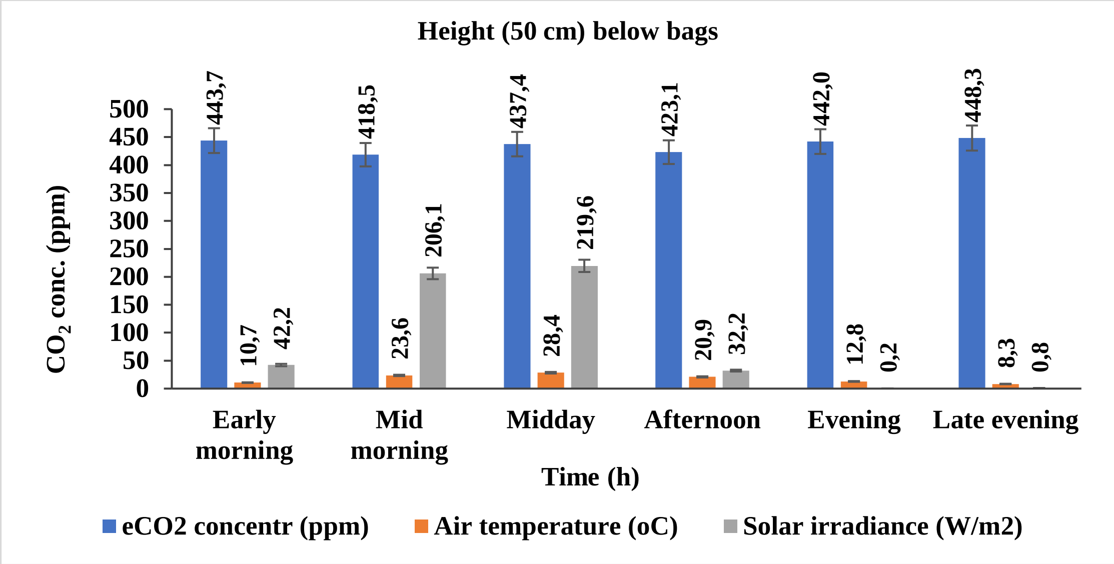


Fig. 2.6 Elevated CO_2 variations in different times of a day at height 50 cm below Mycelium CO_2 generator bags.

Elevated CO_2 significantly increased by 18,9 ppm from mid-morning to midday which had a concentration of 437 ppm. There was no decline in the eCO_2 concentration during the process of photosynthesis at midday, nevertheless, from midday to afternoon there was a slight assimilation of CO_2 in the presence of maximum air temperature between 28 to a minimum of 20,9°C, and solar radiance with light intensity varying between 219,6 and 32,2 W/m^2 respectively. It is observed from the data that in the afternoon hours the concentration of CO_2 was elevated to 423 ppm and continued to rise reaching a concentration of 448 ppm in the late evening hours. Because of the very low temperature and solar radiance, the assimilation in CO_2 by maize plants was not so active enough to decline the CO_2 effectively.

2.5.2 Elevated CO_2 at height 100 cm below CO_2 mycelium bags

In the early morning hours, the rate of eCO_2 concentration at height 100 cm below the Mycelium bags were found to have decreased significantly from the concentration of 455 to 465 owing to the raised temperatures from 16,3 to 33,1°C and the solar radiance from 38,1 to 209,1 W/m^2 at midday. Sweetcorn plants assimilated 21 ppm CO_2 in early morning to midday hours, simply because the air temperature rose by 14,5°C and solar radiance by 156,5 W/m^2 encouraging the sequestration of eCO_2. A further significant decline during midmorning and midday hours was observed when sweetcorn plants subsequently sequestrated about 6,7 ppm of CO_2 since temperature increased by 2,3°C and was elevated to 33,1°C and solar light at

209,1 W/m^2 respectively. In the afternoon and evening hours, CO_2 was elevated from 444 to 465 ppm since light was entirely absent and decline temperatures. Clearly, there is a correlation between air temperature and solar radiance variables to prolong carbon sequestration by plants.

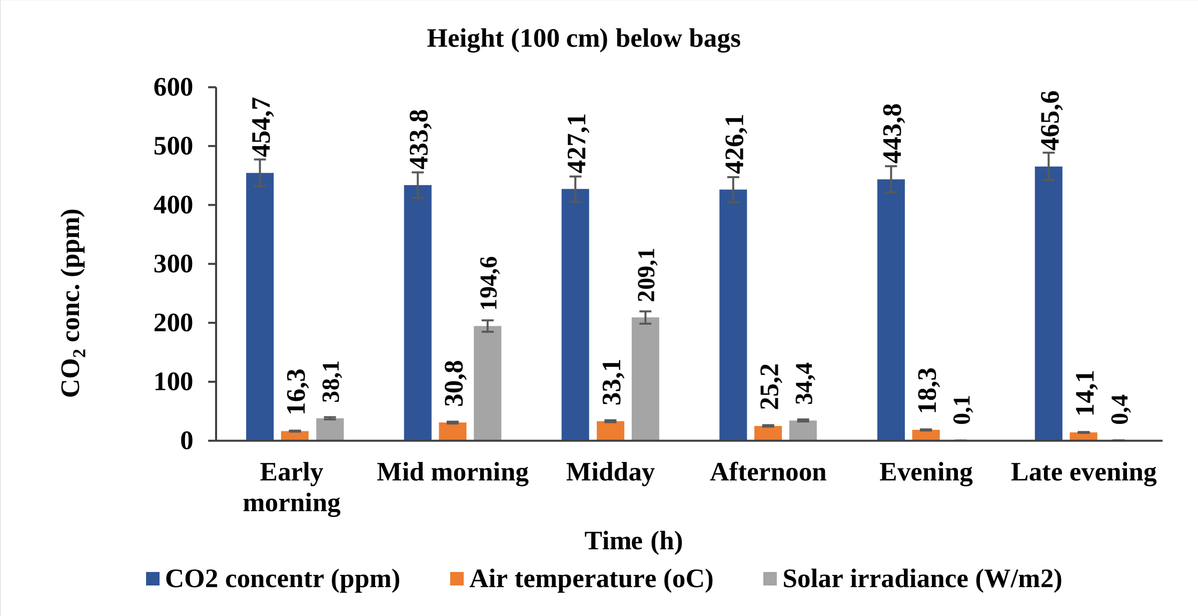


Fig. 2.7 Elevated CO_2 variations in different times of a day at height 100 cm below Mycelium CO_2 generator bags

Light intensity depends on the availability of air temperature to encourage the assimilation of CO_2 by plants, for which their decline or absence slow down the assimilation of CO_2 by sweetcorn plants. Sage (1994) and Drake *et al.* (1997), reported that alterations may arise due to the duration of exposure, species, cultivars, the presence of solar radiation, air temperature, nutrient supplementation as well as water stress and plant pot size, furthermore, the source for the observed growth improvement in C4 plants grown under eCO_2 remains unclear as in C3 plants (Maroco *et al.*, 1999).

2.5.3 Elevated CO_2 at height 150 cm below CO_2 mycelium bags

At height 150 cm below the Mycelium CO_2 bags, the concentration of eCO_2 during early morning hours was at concentration of 496 ppm with air temperature and solar radiance sitting at 22,6 ^oC and 52,3 W/m^2 respectively. A significant decline in CO_2 gas is seen through the manner in which sweetcorn plants assimilates eCO_2 during the day, and it indicates a significant decrease in the concentration towards mid-morning as well as midday hours and is quite similar in pattern to the assimilation that occurred at a height of 100 cm below the

Mycelium bags. The amount of eCO_2 assimilated between early morning and mid-morning was 62 ppm, displaying a significant decline in the CO_2 concentration. Furthermore, a prolonged significant decline in CO_2 concentration was observed during midday with temperature at 28,5°C and solar radiance at 176,7 W/m^2 with which 5 ppm was assimilated and absorbed.

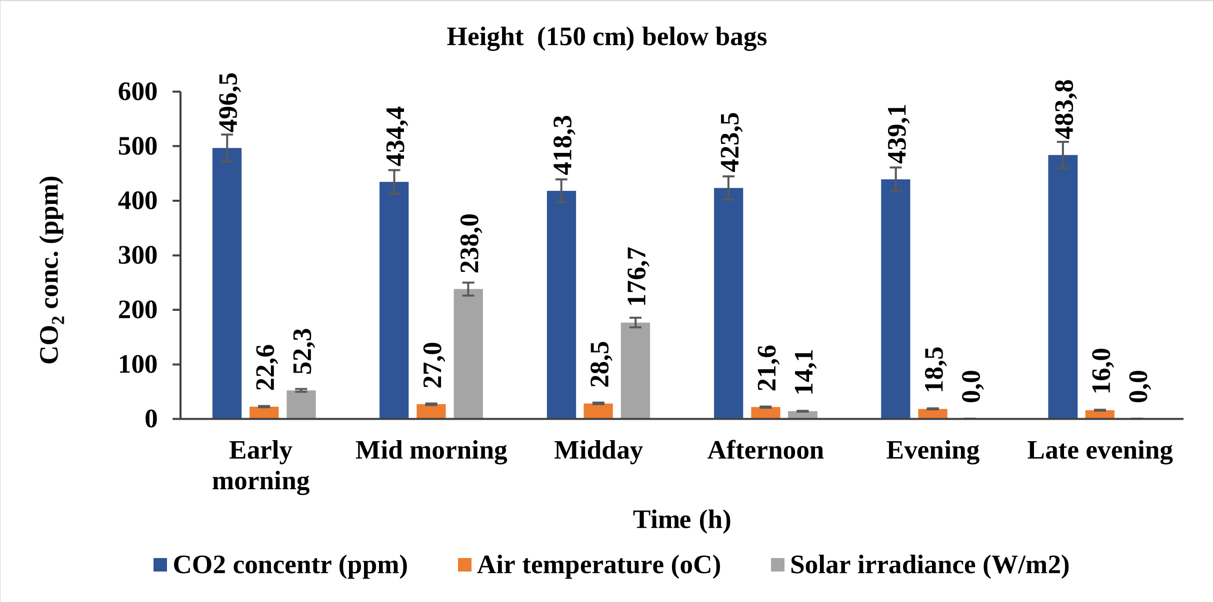


Fig. 2.8 Elevated CO_2 variations in different times of a day at height 150 cm below mycelium CO_2 generator bags

Meanwhile, in the afternoon hours slight accumulation in eCO_2 was observed when sweetcorn plants were photosynthesising due to declines in temperature and solar radiance. In the evening hours, the concentration rate of CO_2 was greatly enhanced from 439 to 484 by 45 ppm, with temperature sitting at 16°C. Reicosky (1989), reported that there were limited data in regard to natural CO_2 concentration patterns in agricultural systems said to have the strongest sinks for CO_2. Clearly the assimilation of eCO_2 at 150 cm height below the Mycelium CO_2 bags is higher than the assimilation and absorption occurring at 100 and 50 cm heights below the CO_2 bags, simply because, CO_2 is heavier than air with its atomic mass being 44,01 g/mol bigger than that of oxygen (O_2) with atomic mass of 32 g/mol, compelling the CO_2 gas to sink at the bottom and get sequestrated to the soil. Nevertheless, limited results are found in individual crops as well as in trials conducted in small fields, and considerably portray dramatic diurnal progressions of CO_2 concentrations with considerable higher amounts above the crop canopy than with enhanced concentrations (Reicosky 1989).

2.6 Conclusion

The diurnal trends of CO_2 concentrations in glasshouses showed a relatively strong reliance on solar radiation and higher temperature varying between 25-35^oC day and night, while relative humidity (RH) was maintained between 65 to 85%. Higher solar radiance during the midday intensified the assimilation of CO_2 leading to declined concentration as little as 418 ppm at a height of 150 cm below mycelium CO_2 bags and later showed a gradual increase on sunset as well as the absence of solar radiation. On overcast days with limited solar radiation, CO_2 concentration was not reduced intensely due to the combined effect of plant and soil respiration as well as reduced photosynthesis. The effect of irrigation was generally not significant, possibly signifying suitable ventilation within root absorption in CO_2 concentrations at height of 150 cm below the mycelium CO_2 bags affected by soil management practice. The results obtained in glasshouse conditions and CO_2 enhancement from the mycelium bags, however, agree with findings from model predictions stipulated by (Allen *et al.*, 1971; Waggoner, 1969) in which water management practices and carbon sequestration from the soil would not be adequate to be efficiently utilised as natural fertiliser mostly due to the rapid loss of CO_2 to the atmosphere resulting from turbulent mixing with other atmospheric gases.

Literature cited

Alle L. H., Jr.; Jensen S. E. and Lemon E. R. (1971). Plant response to carbon dioxide under field conditions: Simulation. Science. Vol. 173: 256-258.

Booth B. B. B.; Harris G. R.; Murphy J. M.; House J. I.; Jones C. D.; Sexton D.; Sitch S. (2017). Narrowing the range of future climate projections using historical observations in atmospheric CO_2. Journal of Climate. Vol. 30(8): 3039–3053.

Conley M. M. ; Kimball B. A.; Brooks T. J.; *et al.* (2001). CO_2 enrichment increases water-use efficiency in sorghum. *New Phytologist* Vol. 151: 407–412.

Concise Oxford Dictionary, (1999). Concise Oxford Dictionary. Oxford University Press.

Dahlaman R. C.; Strain B. R.; and Rogers H. H. (1985). Research on the response of vegetation to elevated carbon dioxide, Journal of Environmental Quality (USA), Vol. 14(1).

Drake B. G.; Rogers H. H.; and Allen L. H. (Jr). (1985). Methods of exposing plants to elevated carbon dioxide in Direct effects of increasing carbon dioxide on vegetation, edited by B. R. Strain and J. D. Cure (DOE/ER-0238, United States Dept of Energy, Washington, DC).

Drake B. G.; Gonzalez-Meler M. A.; Long S. P. (1997). More efficient plants: a consequence of rising atmospheric CO_2? Annual Review of Plant Physiology. Plant Molecular Biology. Vol. 48: 609-639.

Enoch H. Z. and Kimball B. A. (1986). Carbon Dioxide enrichment of greenhouse crops: volume I, Status and CO_2 source and volume II, Physiology, Yield, and Economics (CRC Press, Boca Raton, FL, USA).

Ghannoum O.; Evans J. R. and von Caemmerer S. (2011). Nitrogen and water use efficiency of C4 plants. In: Raghavendra, A.S. & Sage, R.S. (Eds.) C4 Photosynthesis and Related CO_2 Concentrating Mechanisms, Springer Science+Business Media B.V., Dordrecht the Netherlands, (pp129-146) Ghannoum, O.; von Caemmerer, S. C.

Ghannoum O.; von Caemmerer S.; Ziska L. H. and Conroy J. P. (2000). The growth response of C4 plants to rising atmospheric CO2 partial pressure: a reassessment. Plant, Cell and Environment Vol. 23: 931–942.

Grodzinski B. Plant nutrition and growth regulation by CO_2 enrichment. BioScience. 1992; Vol. 42(7): 517– 527.

https://www.esalq.usp.br/lepse/imgs/conteudo-thumb/LI-84OA-$CO_2$$H_2$O-Gas-Analyzer.pdf.

https://www.licor.com/documents/287pss2fnkcwoOox29gh.pdf-file.

https://grndwork.com/wp-content/uploads/2015/01/GroundWork-EquipSpec-CampbellScientific.CR1000.Datalogger. pdf.

https://www.licor.com/enu/products/gas-analysis/LI-610.

https://www.campbellsci.com/ec150.

Kimball B. A.; Pinter P. J. Jr.; Wall G. W.; Garcia R. L.; LaMorte R. L.; Jak P. M. C.; Frumau K. F. A.; and Vugts H. F. (1997). Comparisons of responses of vegetation to elevated carbon dioxide in free air and open top chamber facilities in Advances in Carbon Dioxide Research, edited by L. H. Allen (Jr); M. B. Kirkham; D. M. Olszyk; and C. E. Whitman (Am. Soc. Agron. Crop Science Society of America and Soil Science Society of America, Madison, WI, USA), (pp 113-130).

Lin E.; Wei Xiong; Hui Ju; Yinlong Xu; Yue Li; Liping Bai and Liyong Xie. (2005). Climate change impacts on crop yield and quality with CO_2 fertilization in China. Philosophical Transaction B. The Royal Society Publishing London Book of Biology Science. Vol.360 (1463): 2149–2154.

Maroco J. P.; Edwards G. E.; and Ku M. S. B. (1999). Photosynthetic acclimation of maize to growth under elevated levels of carbon dioxide. Planta Vol. 210: 115-125.

Manson A. D. and Roberts V. G. (2000). Analytical Methods Used by the Soil Fertility Services Section; KZN Agri-Report No. N/A/2001/4; KwaZulu-Natal Department of Agriculture and Rural Development, Pietermaritzburg, KwaZulu-Natal.

Mortensen L. M. (1987) Review: CO_2 enrichment in greenhouses. Crop Responses. Scientia Horticulturae. 1987. Vol. 33: 1–25.

Muller C.; Cramer W.; Hare W. L.; Lotze-Campen H. (2011) Climate change risks for African agriculture. P Natl Acad. Sci USA Vol. 108: 4313–4315.

Reicosky D. C. (1989). Diurnal and seasonal trends in carbon dioxide concentrations in maize and soybean canopies as affected by tillage and irrigation. Agricultural and Forest Meteorology. Vol. 48: 285-303 Elsevier Science Publishers B.V., Amsterdam - Printed in The Netherlands.

Sage R. F. (1994). Acclimation of photosynthesis to increasing atmospheric CO_2: the gas exchange perspective. Photosynth Res. Vol. 39: 351-368.

Schulze E. D. and Mooney H. A. (1993). Design, and execution of experiments on CO_2 enrichment, ecosystems research report No. 6 (Commission of the European Communities, Brussels, Belgium).

Sørensen S. P. L. Biochemistry Zeit. 1909, Vol. 21: 131–199.

Tubiello F.; Rosenzweig C.; Goldberg R.; Jagtap S.; Jones J. (2002). Effects of climate change on US crop production: simulation results using two different GCM scenarios. Part I: wheat, potato, maize, and citrus. Clim Res. Vol. 20: 259–270.

Uprety D. C.; Garg S. C.; Tiwari M. K. and Mitra A. P. (2000). Crop responses to elevated CO_2: Technology and research (India study), Global Environmental Research (Japan), Vol. 3: 155.

Vanaja M.; Maheswari P.; Ratnakumar P. and Ramakrishna Y. S. (2006). Monitoring and controlling of CO_2 concentrations in open top chambers for better understanding of plants response to elevated CO_2 levels. India Journal of Radio & Space Physics. Vol. 35: 193-197.

Waggoner P. E. (1969). Environmental manipulation for higher yields. In: J. D. Eastin, F.A. Haskins, C. Y. Sullivan, and C. H. M. van Bavel {Editors), Physiological Aspects of Crop Yield. Am. Soc. Agron., Madison, WI, (pp. 343-373).

Wall G. W.; Brooks T. J.; Adam R.; *et al.* (2001). Elevated atmospheric CO_2 improved sorghum plant water status by ameliorating the adverse effects of drought. New Phytologist Vol. 152: 231–248.

Windell Hydroponics.co.za, www.hydroponics.co.za.

Chapter 3

Effect of artificially elevated CO_2 (eCO_2) on morphology of sweetcorn (*Zea mays* L. var. *saccharata*)

Abstract

As a response to global warming, greenhouse gases (GHGs) have significantly increased in the atmosphere, possibly reaching over 700 ppm at times. Elevated CO_2 (eCO_2) has been reported to encourages growth and development of agricultural crops through positive spin-offs that facilitate plant growth, resulting in increased biomass. The aim of the present study was to investigate whether slightly elevated CO_2 concentration of between 430-500 ppm will induce growth and development as well as early reproductive development in the sweetcorn cultivars Assegai and STAR7719. Sweetcorn was grown in a glasshouse in the South African winter season in two well-sealed glasshouses adjoining each other. One of the glasshouses contained elevated CO_2 through the use of 'Mycelium CO_2 generator bags'. Nine sweetcorn plants were grown under ambient (400-430 ppm) as well as under elevated (430-550 ppm) CO_2 concentrations. The performance of the plants under the two growing environments was evaluated. Our study reports on the lodging of Assegai control under aCO_2 experiencing lodging and gradual plant death due to very low temperature affected plant growth. Results showed significant differences in DTE were found in Assegai and STAR7719 under aCO_2 had means of 25.44 and 9.77 and under eCO_2 was 12.11 and 7.89. STAR7719 plants displayed a significant difference in days to tasselling, anthesis, and silking between control and eCO_2 plants (57, 67 and 65) days after emergence under eCO_2 with significant effect from eCO_2 for STAR7719 whereas, Assegai plants had lodged, and data capture was discontinued. Treated plants STARR7719 had a slightly higher leaf number and plant height were taller, contributing to enhanced above ground biomass significant (0.1 cm over 0.08 cm and 1.81cm over 1.23cm). The CO_2 treatment enhanced fresh ear mass (219 g over 163 g and ear length 19.8 g over 13.2 g) significantly. Total plant biomass and root mass were also found to be significantly enhanced by the CO_2 treatment. Additionally, fresh, and dry root mass were significantly increased under eCO_2 conditions. Elevating CO_2 levels to approximately 500 ppm positively affected sweetcorn growth and development (biomass, reproduction, ear mass and size) resulting in shortened growing season.

3.1 Introduction

Major, rapid changes in climate have resulted from the accumulation of greenhouse gasses (GHGs), in the Earth's atmosphere; this has been, and is, aligned with an elevation in global temperature, altering world precipitation patterns (Forster *et al.,* 2007; Vanaja *et al.*, 2015). Emissions of methane (CH_4) and nitrous oxide (N_2O) from land on which crops are grown, are viewed as a threat to the ozone layer, and, therefore, the reduction of these emissions particularly from agricultural activities, is essential (Forster *et al.,* 2007). Burning of fossil fuel, extensive wildlife has also contributing to a CO_2 enrichment in Earth's atmosphere, so that CO_2 concentrations are predicted to reach 550 ppm by 2050, with the probability of exceeding 700 ppm by the end of the 21st century (IPPC, 2007). This CO_2 concentration is likely to affect the production and productivity of agricultural crops, hence, impacting world food security (Vanaja *et al.,* 2015).

Thomson *et al.* (2005) envisaged that altered climatic conditions will cause a decline in yield of all food crops, and these changes in climate will continue over the next 50 years. To the contrary, the climate change that has occurred thus far might offer some benefits to agricultural production, simply because it might result in positive spin-offs for the production of certain crops (i.e., increased biomass and possible early maturity). Certain aspects of climate change are projected to stimulate growth and development of productivity of particular crops, e.g., C3 were reported to have enhanced growth of 17%-29% and C4 crops had growth increase of 6%-10% (Thomson *et al.,* 2005; Kimball, 1983; Allen *et al.*, 1987). Elevated CO_2 (eCO_2) was observed to positively improve yield in maize (Vanaja *et al.,* 2015; Brown and Rosenberg, 1999; Parry *et al.,* 2004). It has, therefore, been termed "CO_2 fertilization" (Philips *et al.,* 1996; Thomson *et al.,* 2005), due to its positive impact on plant growth and yield. Similarly, Foster (2016) emphasized that growers had known long ago that an increase in CO_2 boosts plant productivity. Long *et al.* (2004) found that CO_2 enrichment is liable to raise net photosynthetic rates of plants and, therefore, plant productivity and yield.

Plenty of greenhouses are supplied by CO_2 that is delivered by truck in liquid form popularly known as 'pure CO_2'; other methods of enhancing CO_2 in the plant environment include carbon-capture technology by CO_2 Solutions, a cost-effective technique used to capture CO_2 from on-site combustion of natural gas and fossil fuel (Blom *et al.,* 2002). Enhanced plant tolerance to environmental stresses can be increased through soluble sugars, antioxidants, as well as exudates (Drake *et al.,* 2011; Huang and Xu, 2015). Plants can emit exudates liquids from the roots, these particular secretions impact the rhizosphere close to the roots to hinder development of damaging microbes, thus promoting plant growth (Badri and Vivanco, 2009). Fierro *et al.* (1994) found that an eCO_2 concentration of 900 ppm through additional solar radiation (ambient + 100 $\mu mol^{-2}s^{-1}$ photosynthetically active radiation (PAR) increased yield of tomato and pepper by 15% and 11%, respectively.

Sweetcorn (*Zea mays* L. var. *saccharata*) is a horticultural vegetable with a dramatic increase in consumption in the USA, Brazil, Canada, and Europe, both as a fresh vegetable and as processed food (Williams, 2012). Farmers are attracted to this commodity because it is early maturing and harvest can be mechanized (Johnson, 1991). Sweetcorn originates from consequential mutations of the Peruvian maize race Chullpi in which the endosperm is luminous indicating that starch is partially converted into sugar (Boutard, 2012). Sweetcorn is available in two colours white and yellow, with yellow sweetcorn being the most preferred type by consumers, particularly because of enhanced Vitamins A and C concentrations in yellow sweetcorn (Gangaiah, 2008). Coskun *et al.* (2006) described the nutritional composition of sweetcorn has about 221 g carbohydrates, 3.35 g protein and 10 g lipids.

Both, C3 and C4 plants have been identified as crops likely to be directly affected by rising atmospheric CO_2 levels. Alternatively, in morphological structure and biochemical processes in both plant types are likely to occur under eCO_2, particularly changing the plant's carbon to nitrogen (C/N) balance. This phenomenon of an altered C/N ratio under eCO_2 has been described in several agricultural crops, such as maize, tomato, potato, and winter wheat (Islam *et al.*, 1994; Behboudian and Tod, 1995; Li *et al.*, 2007; De Temmerman *et al.*, 2002; Högy and Fangmeier, 2009). Increased atmospheric CO_2 levels have been found to enhance yield in C3 plants ranging from 17% - 29%, while in C4 plants an increase of 6% - 10% under artificially enhanced CO_2 conditions has been reported (Kimball, 1983). Unlike C3 plants, C4 plants are accustomed to capture radiant energy from the sun, while their stomata are closed, thereby lessening photorespiration; C4 plants are, due to their ability to suppress the oxygenation reaction of Rubisco (photorespiration), also better adapted to stress conditions, such as water, heat, and temperature stress (Lara and Andreo, 2011).

It has, therefore, been hypothesised that eCO_2 concentrations are likely to result in increased photosynthesis in plants, which will lead to better growth, higher above-ground biomass, and higher yield (Ainsworth and Long, 2005; van der Kooi *et al.*, 2016). Since maize is a C4 plant, photosynthesis is not hindered by elevated CO_2, and, thus, yields will increase under elevated

CO_2 conditions, as the stomatal conductance improves the plant's water–use efficiency, thereby permitting photosynthesis to carry on, even under water-scarce conditions (Ghannoum *et al.,* 2000; Leaky, 2009; Long *et al.*, 2004). Additionally, Ghannoum *et al.*, (2011) postulated that C4 plants have a higher water use efficiency than C3 plants, simply because, firstly, a higher photosynthetic rate per unit leaf area and, secondly, a lower stomatal conductance, permitting C4 plants to capture CO_2 better, and, more significantly, having the capacity to separate CO_2 intake from the Calvin cycle by storing an intermediate CO_2 containing product (malate in the mesophyll cells) and transporting it into the bundle sheath cells. Here, the biochemical pathway of producing sugars from CO_2 and H_2O occurs.

This study is intended to investigate growth and development of sweetcorn under eCO_2 conditions by cultivating sweetcorn cultivars under eCO_2 and to identify, if this eCO_2 will result in enhanced yield and/or quality of sweetcorn. Research on eCO_2 has focused on standard maize, dry grain maize, which is used in manufacturing many foods types. A rigorous scientific investigation on Web of Science discovered that over the past 30 years out of the 990 research papers that have dealt with CO_2 effects on crop plants, 109 with maize and 109 on eCO_2 effects on maize. Google Scholar also identified 791 maize publications over the same time period only 1280 focused on eCO_2 enrichment of maize. Additionally, sites such as Science Direct and Scopus discovered that out of 2678, research papers available 2361 dealt with maize and 2278 on eCO_2 enrichment in maize. Similarly, SciELO discovered that out of all research including CO_2 622 papers 1008 dealt with maize research and 127 papers reported on eCO_2 enrichment in maize. Sweetcorn is regarded as a horticultural commodity that plays an important nutritional role. The increase in sweetcorn production over the past 9 years from 15.2% to 27.8% (FAOSTAT, 2018) and in consumption over the same period (FAOSTAT, 2018), outlines the importance of this commodity, so that the effect of eCO_2 on sweetcorn warrants investigation.

The study is, therefore, aiming to examine the effects of artificially eCO_2 on the morphology of sweetcorn (*Zea mays* L. var. *saccharata*). The objectives were: to determine the response of (1) plant height, leaf area, (2) above ground biomass and root fresh and dry mass, (3) yield (number of fresh cobs per plant), (4) individual cob mass and total cob mass per plant to ambient CO_2 (aCO_2) *versus* eCO_2 conditions.

3.2 Materials and methods

3.2.1 Growing conditions and plant material

The experiment was carried out at the University of KwaZulu-Natal, Pietermaritzburg, in two adjacent glasshouses, well-sealed from each other using duct tape (as described in Diurnal chapter 2). Glasshouse environmental conditions such as air temperature between 25-35^{o}C, relative humidity (RH) ranging between 60-80% and solar radiance ranging between 200-425 W/m^2 were not altered during the experimental period. One glasshouse had an aCO_2 concentration of ~430 ppm, while the other glasshouse was maintained at an eCO_2 of approximately 500 ppm through use of 'Because Nature Mycelium CO_2 Generator Bags' (Because Nature Mycelium CO_2 Generator, Windell Hydroponics.ac.za, Cape Town, SA). These bags contain a patented strain of mycelium that colonises the bag and gives off CO_2 gas in exchange for O_2 gas without fruiting. Sweetcorn hybrid seeds of the Assegai and STAR 7719 cultivar locally obtained hybrids from Starke Ayres (Pietermaritzburg, South Africa). Seeds were sown in 30 cm diameter plastic pots filled with locally obtained, heavy black clay-loam from Ukulinga Research Farm, Pietermaritzburg, UKZN. Each glasshouse (4 x 3,5m) had two cultivars with nine replications (18 plants per glasshouse, total of 36 plant) arranged in three rows in a randomized complete block design.

Soil samples were analysed by the Soil Fertility Laboratory at the ARC-Cedara before the soil was mixed with GROMOR® potting medium in a ratio of 3:2 to optimise soil structure, pH and fertility (as described in the descriptive chapter 2). The spacing between pots in rows as well as between rows was 15cm. Sweetcorn plants were irrigated two to three times weekly to prevent drought and to prevent disease development. Morphological parameters, such as days to emergence (DTE), total leaf area, leaf number per plant, plant height, days to tasselling, silking and anthesis under aCO_2 and eCO_2 conditions were recorded. Harvesting was initiated for 'STAR7719' from the 8th to the 21st of September for the treatment and the control. 'Assegai' plants experienced stunted growth, possibly due to the cold temperature, such that flowering was hindered. This shortcoming resulted in 'Assegai' not having a control to compare with. Total above ground fresh and dry biomass, number of ears per plant, fresh ear mass and length, fresh cob mass, root fresh as well as dry mass were, therefore, only recorded for 'STAR7719'.

3.3 Results

Data were analyzed statistically using GenStat®18th edition (VSN International, Hemel Hempstead, UK) software significantly evaluated using analysis of variance (ANOVA). Parameters analyzed were days to emergence (DTE), emergence percentage, plant height and branching, reproductive growth, fresh ear mass and ear length as well as total biomass. Differences in the means of analyzed parameters were determined using LSD's (Fisher's protected least significant difference test) at $P<0.05$, $P<0.01$ and $P<0.001$.

Under eCO_2 conditions, DTE was statistically difference at $P<0.003$ between treatment (fig.2.1). Under eCO_2 'STAR7719' took an average of 7.9 days to emergence, while 'Assegai' seeds emerged on average after 12.11 days. Under aCO_2, DTE were prolonged to 25.44 and 9.22 for 'Assegai' and 'STAR7719', respectively. The emergence percentage under eCO_2 were all 100% for both 'Assegai' and 'STAR7719', while under aCO_2 the emergence percentage was a 100% for only 'STAR7719' and 0.111% for 'Assegai'.

Additionally, emergence % (fig. 2.2) was highly significantly different at $P<0.001$ under eCO_2 versus aCO_2 conditions for 'Assegai', whereas no significant differences were found for 'STAR7719' between eCO_2 and aCO_2 conditions, both resulted in a 100% emergence. The number of DTE and emergence % (Table 2.1) depicts the effect of CO_2 on cultivar; DTE and emergence % were highly significantly affected by CO_2 $P<0.001$.

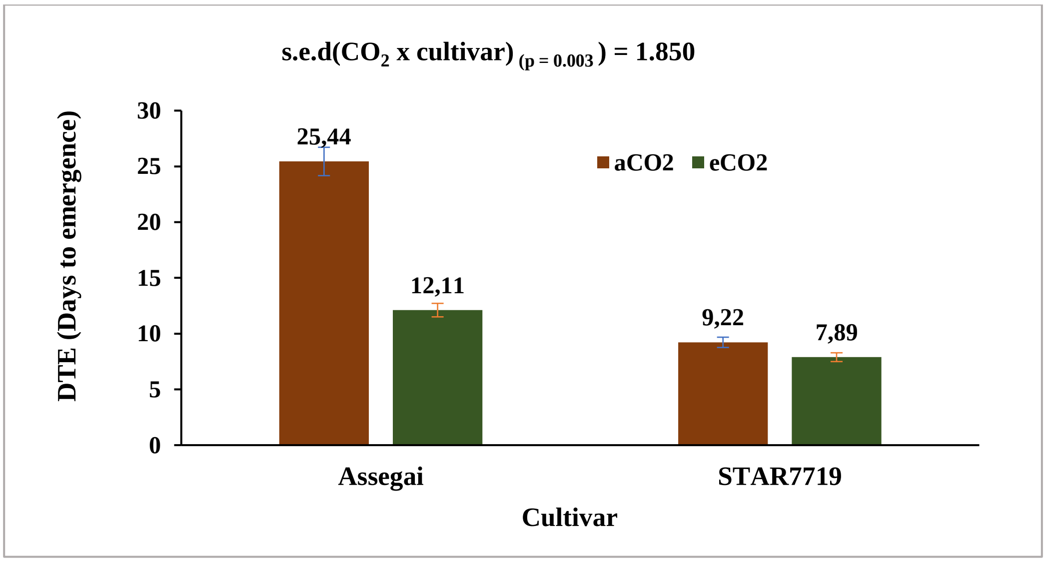


Fig. 3.1 Influence of eCO_2 on days to emergence (DTE) for sweetcorn cultivar 'Assegai' and 'STAR7719' under ambient ~430 ppm (aCO_2) and elevated ~500 ppm (eCO_2) CO_2 concentrations.

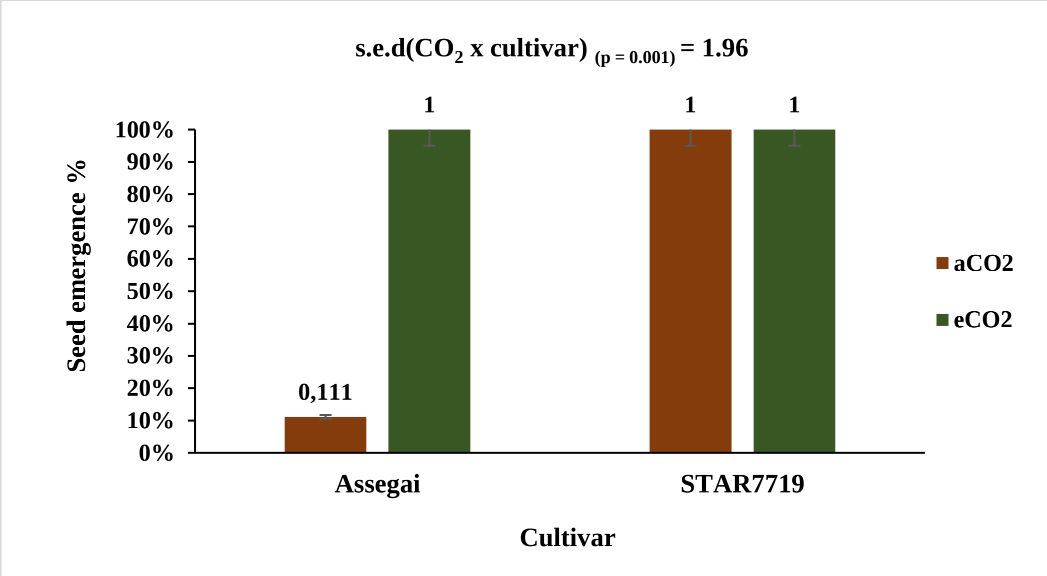


Fig. 3.2 Influence of CO_2 concentrations ambient, ~430 ppm, aCO_2) and elevated, ~500 ppm eCO_2) on emergence percentage of sweetcorn cultivars 'Assegai' and 'STAR7719'.

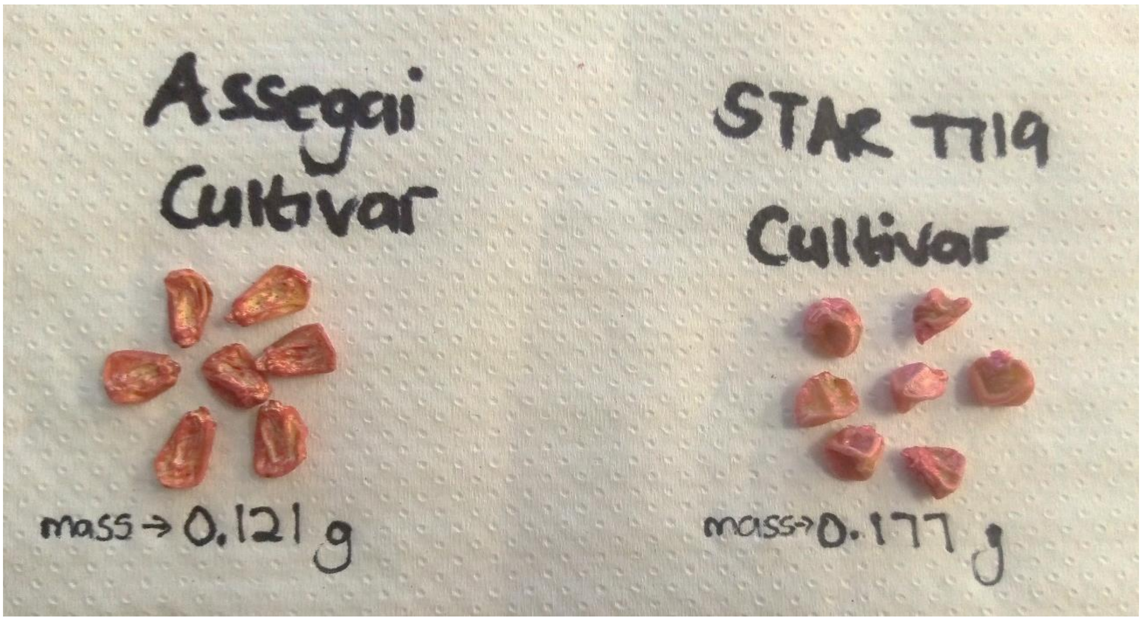


Fig. 3.3 Sweetcorn supersweet hybrid seeds cultivars: Assegai (left) with shrunken and flat endosperms and STAR7719 (right) with bulgy and shrunken endosperms.

Table 3.1 Days to emergence and seed emergence percentage of sweetcorn 'Assegai' and 'STAR7719' under aCO_2 (~430 ppm) and eCO_2 (~500 ppm).

Parameter	**Assegai**		**STAR7719**		**Significant Differences**		
	aC 2	**eC(**	**aC(**	**CO2**	**C**	**CO_2 CO_2**	**C x**
Days to emergenc	25.4	12.1	9.2	7.89	0.001*:	0.001***	0.003**
Mean	13.(						
LSD	5.3:						
Emergenc %	11.	10(	10(	100	0.001*:	0.001*** 0.001***	
Mean	77.						
LSD	7.9						

*, **, *** indicates that the parameter is significant at $P<0.05$, $P<0.01$ or $P<0.001$ respectively, C indicates Cultivar, and ns = non-significant under concentrations ~430 ppm (aCO_2) *versus* ~500 ppm (eCO_2).

Leaf area, plant height and leaf number per plant measured for 'STAR7719' (fig 2.4) indicate that there was no significant difference between treatment and control in leaf area, whereas the number of leaves per plant was significantly higher under eCO_2 (mean of 179 leaves per plant) compared with aCO_2 (mean of 174.3 leaves per plant). Number of leaves per was significantly enhanced by eCO_2 with a mean of (9.56) over a mean of (8.33) under aCO_2, at $P<0.001$. Plants were significantly taller under eCO_2 (mean values of 180.9 cm) over 123.4 cm under aCO_2. Days to tasselling, anthesis and silking (fig. 2.5) were highly significantly shortened ($P<0.001$) by eCO_2 of 56.89, 64.22, and 67.67 days over 67.33, 74.22 and 75.33 days under aCO_2, respectively.

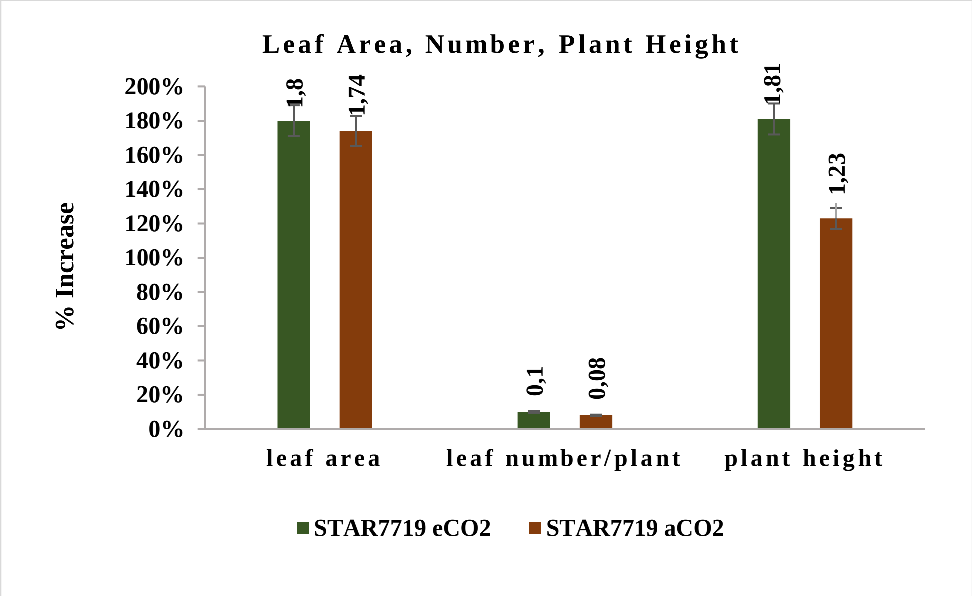


Fig. 3.4 Increase in plants growth parameters of maize cultivar 'STA7719' under elevated CO2 (~500 ppm) compared with ambient CO2 (~430 ppm).

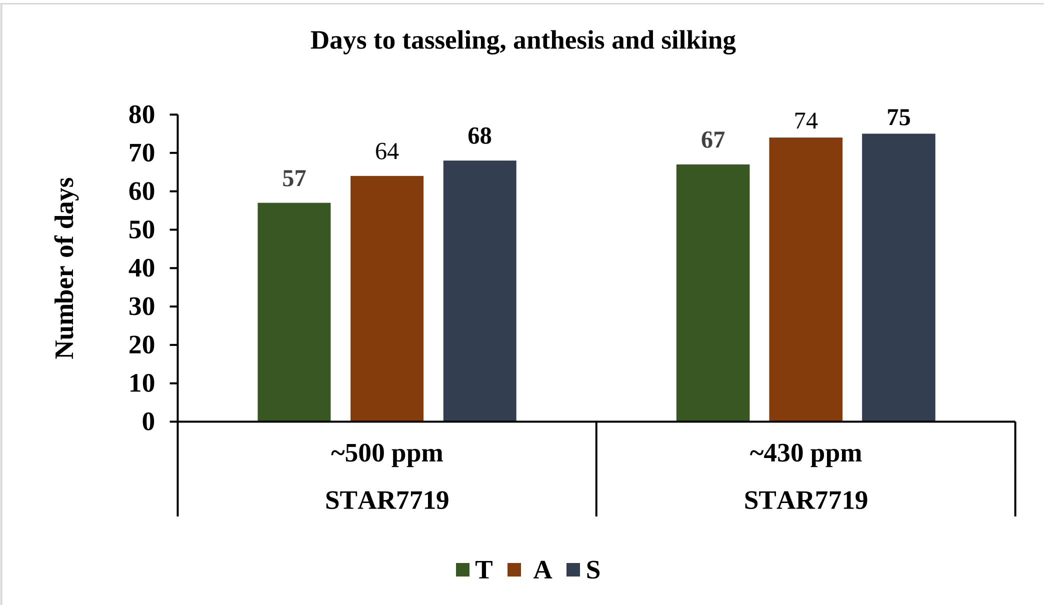


Fig. 3.5 Number of days to tasselling (T), anthesis (A) and silking (S) of cultivar 'STAR7719' under ambient (~430 ppm aCO2) and elevated ~500 ppm eCO2) concentrations.

Table 3.2 Plant growth and reproduction under two CO_2 regimes (~430 ppm (aCO_2) and ~500 ppm (eCO_2) of sweetcorn cultivar STAR7719.

	STAR7719				
Parameters	**aCO_2**	**eCO_2**	**Significant Difference**	**Mean**	**LSD**
Leaf area (mm^2)	174.3	179.8	0.786^{ns}	177.1	44.64
Plant height (cm)	123.4	180.9	0.001***	152.2	11.13
Leaf number/plant	8.33	9.56	0.001***	8.94	0.512
Days to tasselling	67.33	56.89	0.001***	62.11	1.680
Days to anthesis	74.22	64.22	0.001***	69.22	2.400
Days to silking	75.33	67.67	0.001***	71.50	2.400

*, **, *** indicate that the parameter is significant at $P<0.05$, $P<0.01$ or $P<0.001$, respectively, under ambient (~430 ppm, aCO_2) *versus* elevated (CO_2~500 ppm, eCO_2).

The influence of eCO_2 on above ground fresh and dry biomass (Table 2.3) was highly significant, resulting in an above ground fresh mass of 522g under eCO_2, while under aCO_2 a fresh mass of 334g was recorded. Above ground dry mass was also higher under eCO_2 (139.4g) compared with aCO_2 (76.4g). Elevated CO_2 further significantly enhanced root fresh and dry mass (Table 2.3). No significant differences were found in fresh cob mass; nevertheless, significant differences were found in individual ear mass. Ears were highly significantly ($P<0.01$) longer (19.8 cm) under eCO_2 than under aCO_2 (13.2 cm).

Table 3.3 Above ground biomass, fresh cob mass and length parameters under ~430 ppm (aCO_2) and ~500 ppm (eCO_2) of sweetcorn cultivar STAR7719.

	STAR7719				
Plant parameter	**aCO_2**	**eCO_2**	**Significant Difference**	**Mean**	**LSD**
Ear mass (g)	163	219	0.056*	191.0	58.5
Fresh ear length (cm)	13.2	19.8	0.013**	16.5	4.78
Fresh cob mass (g)	104.2	134.1	0.140^{ns}	119.1	42.09
AG fresh mass (g)	334	522	0.005***	428	111.9
AG dry mass (g)	76.4	139.4	0.009***	107.9	42.25
Fresh root mass (g)	61.9	88.8	0.018**	75.4	20.93
Dry root mass (g)	8.77	13.93	0.002***	11.35	4.131

*, **, *** indicates that the term is significant at $P<0.05$, $P<0.01$ and $P<0.001$ respectively, where ns = non-significant at aCO_2 (~430 ppm) and eCO_2 (~500 ppm).

3.4 Discussion

A delayed seedling emergence in the supersweet 'Assegai' hybrid cultivar, agrees with findings by Emmalea (2018), who described supersweet (*sh2*) hybrids to have a relatively low field emergence, due to seeds having low food reserves in the form of starch. Pedro *et al.* (2021) also outlined that hinderances such as susceptibility to cold stress and wet soils, contribute immensely to reducing sweetcorn emergence and seedling vigour. The environmental variables in both glasshouses such as the temperature (ranged from 25-35^{o}C) and relative humidity (RH) ranged from (60-80 %) were measured with agrometeorology tools during South African winter season. The solar radiance for both glasshouses was natural and ranged between 200-425 W/m^2 depending on a day's photoperiodic light. Prior to planting, the physicochemical properties of soil from three soil samples were analysed (Tables 2.3, 2.4, and 2.5, as described in descriptive chapter 2) as well as soil moisture percentages for the two sets of four consecutive weeks (Appendices 8 and 9) were recorded.

Due to cooler temperature, a delay of over one month in DTE and slow growth was found in 'Assegai' plants. Lodging in control 'Assegai' plants subsequently resulted in death of these plants. Emmalea (2018) stipulated that the supersweet hybrids contain the *sh2* gene and are characterised by shrunken endosperms (fig. 2.3). This feature is aligned with the modified carbohydrate synthesis in the high sucrose containing maize endosperm, endosperm starch levels at maturity.

During the time of sowing, germination and emergence could have also affected seed performance, resulting in slowed development due to the cooler winter season with low temperatures in the glasshouse with temperature between 19 and 24^{o}C during the day and 19^{o}C at night, seemingly causing seeds to remain dormant or emerge slowly. Wrinkled cultivars (supersweet hybrids cultivars, with enhanced sugar concentrations in the fruity) were reported to have a smaller amount of food than standard cultivars and, consequently, grow slower and are vulnerable to stand problems (Emmalea, 2018). Additionally, two plants of the 'Assegai' eCO_2 treatment lodged and died, hence, data were only recorded from the remaining, healthy plants. Under eCO_2 conditions 'STAR7719' emerged between 7 and 8 days after sowing, meanwhile, in control the emergence was visible between 8 and 10 days after sowing.

Emergence is governed by environmental conditions driving early embryo growth. Cell elongation was likely limited due to relatively low temperatures in the glasshouse during winter season (Wolfe, 1991); this could have affected, plant height (180.9 under eCO_2 and 123.4 under aCO_2), visible as relatively short maize plants in both eCO_2 as well as aCO_2 glasshouse but growth improved under eCO_2. Significant differences were found between the 'Assegai' eCO_2 treatment and the control, resulting in over a month of delay in seed emergence of the control compared with seed emergence of the treatment (figure 3.1). This led to lodging and, subsequently, no data being could be recorded for control plants. Significant differences were found in plant height and leaf number per plant for treatment compared with control (Table 3.2). As a C4 crop, Driscoll *et al*. (2006) reported maize to respond relatively well to CO_2 enrichment. These authors also reported that the epidermal cells of leaves were larger after six weeks of transferring plants to eCO_2 of 700 ppm compared with plants maintained under 350 ppm CO_2.

The two concentrations of CO_2 used in this experiment (~400 ppm for aCO_2 and Mycelium CO_2 powered generator bags with a concentration of ~500 ppm for eCO_2) resulted in significant differences in various vegetative and yield parameters (Table 3.2 and 3.3). Vanaja *et al.* (2006) elaborated that growing plants in controlled environment chambers can result in root growth restriction in pot-grown plants. Similarly, in the present study, the days to achieving certain phenological milestone (days to tasselling, anthesis and silking) were reduced. This allowed maize plants under eCO_2 to overcome adverse environmental conditions, while control plants succumbed to lodging (Table 3.3). Elevated CO_2 had significant positive effects on sweetcorn, increasing fresh as well as dry root mass; additionally, eCO_2 enhanced above ground fresh and dry biomass (Table 3.3). The advancement in growth and development of maize plant under eCO_2 lead to a shortened growing season due to earlier reproductive development (56-67 days to tasselling) (Table 3.3), indicating the potential to grow even two maize crops in one season.

Elevating CO_2 could, therefore, shorten the period to produce certain horticultural crops, allowing two sweetcorn crops to be produced in one season. At ~500ppm, individual cob mass and leaf area of individual plants were not significantly affected, however, the IPCC (2007) mentioned the possibility of a rise in CO_2 to 550 ppm, possibly even escalating to 700 ppm by the end of 21st century. Such an increase is likely to strongly affect growth parameters measured in this experiment, fast-forwarding growth and development of sweetcorn, as well as resulting in faster maturation of the crop.

Elevated CO_2 not only fast-forward ontogenetic development, but also produced tall plants (Table 3.2). The higher leaf number combined with unchanged chlorophyll concentration and above ground mass are possibly responsible for a higher photosynthetic rate (Lara and Andreo, 2011). The total biomass from data recorded in the present study was greatly enhanced except the for-fruit yield (Table 3.3) this was possibly due to plants being susceptible to a colder growing season with lower temperatures (Waqas *et al.,* 2021). Coupled with this are reports on declining maize yields declining under eCO_2, attributed to a strongly reduced metabolite transport coupled with a reduced photosynthetic activity (Foyer *et al.,* 2002). The effect of CO_2 enrichment in the environment plants are grown in has been debated in C4 plants, as these already possess a mechanism to concentrate CO_2 in the bundle sheath cells, such that C4 plants could be unaffected by eCO_2 concentration in the atmosphere (Lara and Andreo, 2011). This thinking, however, reflects ignorance and has resulted in little interest in investigation on C4 plants and their response to eCO_2 conditions, increasing the CO_2 to possibly 550 ppm needs to be investigated for its effects on sweetcorn production.

Ear mass was positively impacted by eCO_2, similar to ear length; however, cob mass was not significant enhanced (Table 3.3). This might have been due to the experiment being conducted in winter, as this limited Rubisco activity has been reported at cooler temperatures (Kubien *et al.*, 2003). Vanaja *et al.* (2015) reported maize yield being improved under eCO_2, as elevated CO_2 triggered the partitioning of biomass towards cobs or grains. Reddy *et al*. (2010) conducted a fifteen year-literature survey on the influence of eCO_2 among numerous C3, C4 and CAM plants by providing data on forty C3, two C4 as well as three CAM species. Several authors further found positive, significant responses of C3 plants to eCO_2 conditions in terms of photosynthetic acclimation, *i.e.,* a change in photosynthetic efficiency of leaves due to long exposure to eCO_2, while in C4 crops, such as *Sorghum* and *Panicum*, certain negative responses to eCO_2 have been reported. For CAM plants, such as pineapple, agave and kalanchoe, positive responses in plant growth and development were reported (Reddy *et al.*, 2012).

The outcome of the survey conducted by Reddy *et al.* (2010), portrayed little evidence of investigations on the response of C4 species to eCO_2. Vanaja *et al.* (2015), however, found a response of the C4 crop maize to eCO_2, which was solely attributed to the efficient partitioning of reproductive biomass towards the reproductive rather than towards vegetative parts. Furthermore, inconsistent reports on the response of C4 plants to eCO_2 and differences in photosynthetic responses to eCO_2 are attributed to plant species, differences in experimental techniques, the period of treatment, the age of the plant and the growing season (Sage, 2002).

3.5 Conclusion

In conclusion, the study shows evidence of the C4 crop sweetcorn to be directly affected by eCO_2. The investigation further showed that there is a tendency that sweetcorn assimilates more carbon under eCO_2. The current study provides, however, no proof that yields are enhanced under 500 ppm. Despite the two contrasted glasshouses only differing by 70 ppm significant differences in certain plant parameters were detected. It could also be possible that higher CO_2 concentrations (in a range of 430 to 700 ppm) result in more positive effect on sweetcorn plant growth and development and, therefore, in enhanced yield and recorded time to harvest. Some maize plants in the eCO2 glasshouse developed second ears, even though there were no kernels produced possibly due limited fertilization under the prevailing low temperature. Addition of N and $CaMgNO_3$ fertilizers might have resulted in enhanced development and contributed to higher yields.

Literature cited

Ainsworth E. A. and Long S. P. (2005). What have we learned from 15 years of free-air CO_2 enrichment (FACE)? A meta-analytic review of the responses of photosynthesis, canopy properties and plant production to rising CO_2. New Phytologist. Vol. 165(2): 351-371.

Bedri D. V. and Vivanco J. M. (2009). Regulation and function of root exudates. Plant, Cell & Environment. Vol. 32 (6): 666-81.

Behboudian M. H. and Tod, C. (1995). Postharvest attributes of 'Virosa' tomato fruit produced in an enriched carbon dioxide environment. Horticultural Science. Vol. 30: 490–491.

Blom T. J.; Straver W. A.; Ingratta F. J.; Khosla-OMAFRA S. and Brown W. (2002). Carbon dioxide in greenhouses. http://www.omafra.gov.on.ca/english/crops/facts/00-077.htm.

Boutard A. (2012), Beautiful Maize: America's Original Grain from Seed to Plate. Gabriola Island, Canada. New Society Publishers, (pp 224).

Brown R. A. and Rosenberg N. J. (1999) Climate change impacts on the potential productivity of maize and winter wheat in their primary United States growing regions. Climate Change. Vol. 41: 73–107.

Coskun M. B.; Yalcın. I. and Zarslan, C. O. (2006). "Physical properties of sweetcorn seed (*Zea mays* L. *saccharate Sturt*)". Journal of Food Engineering. Vol: 74(4), pp 523–528.

Creech R. G. (1956). Genetic control of carbohydrates synthesis in maize endosperm. Genetics. Vol. 52: 1175–1186.

De Temmerman L.; Hacour A. and Guns M. (2002). Changing climate and potential impacts on potato yield and quality 'CHIP': introduction, aims and methodology. European Journal of Agronomy. Vol. 17: 233–242.

Drake J. E.; Gallet-Budynek A.; Hofmockel K. S.; Bernhardt E. S.; Billings S. A. and Jackson R. B., *et al.* (2011). Increases in the flux of carbon belowground stimulate nitrogen uptake and sustain the long-term enhancement of forest productivity under elevated CO_2. Ecology Letters. Vol. 14: 349–357.

Driscoll S. P.; Prins A.; Olmos E.; Kunert K. J. and Foyer C. H. (2006) Specification of adaxial and abaxial stomata, epidermal structure, and photosynthesis to CO_2 enrichment in maize leaves. Journal of Experimental Botany. Vol. 57(2): 381-390.

Emmalea G. E. (2018). Poor stands and plant vigour in early planted fresh market sweetcorn. University of Delaware. https://sites.udel.edu/weeklycropupdate/?page_id=4749.

FAOSTAT, 2018. Crop Production Statistics. http://faostat.fao.org, Accessed: July 2018.

Fierro A.; Gosselin A. and Tremblay N. (1994). Supplemental carbon dioxide and light improved tomato and pepper seedling growth and yield. HortScience. Vol. 29: 152–154.

Forster P.; Ramaswamy V.; Artaxo P.; Berntsen T. Betts R. and Fahey D. *et al* (2007). Changes in atmospheric constituents and in radiative forcing. In: Climate change 2007: The Physical Science Basis. Contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change, Cambridge University Press, and (pp 129–234).

Foster M. Five reasons why CO_2 levels are controlled at night (2016). Edaphic Scientific blog, the latest in the world of environmental research & monitoring. https://ggs-greenhouse.com/blog/5-reasons-why-co2-levels-are-controlled-at-night.

Foyer C. H.; Vanacker H.; Gomez L. D.; Harbinson J. (2002). Regulation of photosynthesis and antioxidant metabolism in maize leaves at optimal and chilling temperatures. Plant Physiology and Biochemistry, Vol. 40: 659–668.

Gangaiah B. (2008). Agronomy-Kharif Crops: maize: division of agronomy, Indian Agricultural Research Institute. New Delhi. [http://nsdl.niscair.res.in/bitstream/], (Accessed September 2012).

Ghannoum O.; von Caemmerer S.; Ziska L. H. and Conroy J. P. (2000). The growth response of C4 plants to rising atmosphere CO_2 partial pressure: a reassessment. Plant Cell Environment. Vol. 23: 931–942.

Ghannoum O.; Evans, J. R. and von Caemmerer, S. (2011). Nitrogen and water use efficiency of C4 plants. In: Raghavendra, A.S. & Sage, R.S. (Eds.) *C4 Photosynthesis and Related CO_2 Concentrating Mechanisms,* Springer Science + Business Media B.V., Dordrecht, The Netherlands, (pp 129-146).

Gruda N. and Tanny J. (2014). "Protected Crops," in Horticulture: Plants for People and Places: Production Horticulture, Vol: 1, (Eds.) G. R. Dixon and D. E. Aldous (Dordrecht: Springer), (pp 327–405).

Högy P. and Fangmeier, A. (2009). Atmospheric CO_2 enrichment affects potatoes: 2. Tuber quality traits. European Journal of Agronomy. Vol. 30: 85–94.

Huang B. and Xu Y. (2015). Cellular and molecular mechanisms for elevated CO_2-regulation of plant growth and stress adaptation. Crop Science. Vol. 55: 1–20.

IPCC, (2007) Summery for policy makers, In: Solomon S. D., Qin D. Manning M. (Eds.), Climate Change, 2007: The physical science basis. Contribution of Working Group 1 to Fourth Assessment Report of the Intergovernmental Panel on Climate Change. Cambridge University Press, Cambridge, United Kingdom and New York, USA.

Islam M.; Matsui T. and Yoshida Y. (1994). Effects of carbon dioxide enrichment on acid invertase and sugar concentration developing tomato fruit. Environmental Control Biology. Vol. 32: 245–251.

Johnson L. A. (1991). Maize production, processing, and utilization. In: Handbook of Cereal Science and Technology. Vol. 1: 55-131. K. J. Lorenz, and K. Kulp. ed. New York, U.S.A: Marcel Dekker Inc.

Kimball Bruce A. (1983). Carbon Dioxide and Agricultural Yield: An Assessment and Analysis of 430 Prior Observation. Agronomy Journal. Vol. 75: 75, 779.

Kubien D. S.; von Caemmerer S.; Furbank R. T. and Sage R. F. (2003). C4 photosynthesis at low temperature: a study using transgenic plants with reduced amounts of Rubisco. Plant Physiology. Vol. 132: 1577–1585.

Lara M. V. and Andreo C. S. (2011). C4 Plants Adaptation to High Levels of CO_2 and to Drought Environments. Abiotic Stress in Plants – Mechanisms and Adaptations, Prof. Arun Shanker (Ed.), ISBN: 978-953-307-394-1, InTech, Available from http://www.intechop.en.com/books/abiotic-stress-in-plantsmechanisms-and-adaptations/c4-plants-adaptation-to-high-levels-of-co2-and-to-drought-environments.

Leakey A. D. B. (2009) Rising atmospheric carbon dioxide concentration and the future of C4 crops for food and fuel. Proceedings of the Royal Society B-Biological Sciences. Vol. 276: 2333–2343.

Li F.; Wang J.; Chen Y.; Zou Z.; Wang X. and Yue M. (2007). Combined effects of enhanced ultraviolet – B radiation and doubled CO_2 concentration on growth, fruit quality and yield of tomato in winter plastic greenhouse. Frontiers in Biology China. Vol. 2: 414–418.

Long S. P.; Ainsworth E. A.; Rogers A. and Ort, D. R. (2004). Rising atmospheric carbon dioxide: plants FACE the Future. Annual Review of Plant Biology. Vol. 55: 591–628.

Najeeb S.; Sheikh F. A.; Ahangar M. A. and Teli N. A. (2011). Popularization of sweetcorn (*Zea mays* L. var. *saccharata*) under temperate conditions to boost the socioeconomic conditions. Maize Genetics Cooperation Newsletter 85.

Nobel P. S. (1996). Responses of some North American CAM plants to freezing temperatures and doubled CO_2 concentrations: Implications of global climate change for extending cultivation. Journal of Arid Environments. Vol. 34: 187–196.

Parry M. L.; Rosenzweig C.; Iglesias A.; Livermore M. and Fischer G. (2004). Effects of climate change on global food production under SRES emissions and socio-economic scenarios. Global Environmental Change. Vol. 14: 53–67.

Phillips D. L.; Lee J. J. and Dodson R. F. (1996). Sensitivity of the US maize belt to climate change and elevated CO_2. 1. Maize and soybean yields. Agriculture Systems. Vol. 52: 481–502.

Revilla P.; Anibas C. M.; Tracy W. F. Sweet Maize Research around the World 2015–2020. Agronomy 2021, 11, 534.

Reddy, A.R., Rasineni, G.K., Raghavendra, A.S. (2010). The impact of global elevated CO_2 concentration on photosynthesis and plant productivity. – Current Science. Vol: 99, pp 46-57.

Sage R. F. (2002). Variation in the k(cat) of Rubisco in C3 and C4 plants and some implications for photosynthetic performance at high and low temperature Journal of Experimental Botany. Vol. 53: 609-620.

Thompson A. M.; Brown R. A.; Rosenberg N. J.; Izaurralde R. C. and Benson V. (2005) Climate change impacts for the conterminous USA: an integrated assessment. Part 3. Dryland production of grain and forage crops. Climate Change. Vol. 69: 43–65.

Vanaja M.; Maheswari P.; Ratnakumar P. and Ramakrishna Y. S. (2006). Monitoring and controlling of CO_2 concentrations in open top chambers for better understanding of plants response to elevated CO_2 levels. India Journal of Radio & Space Physics. Vol. 35: 193-197.

Vanaja M.; Maheswari M.; Jyothi Lakshmi N.; Sathish P.; Yadav S. K.; Salini K.; Vagheera P.; Vijay Kumar G. and Abdul Razak. (2015). Variability in Growth and Yield Response of Maize Genotypes at Elevated CO_2 Concentration. Advances in Plants & Agriculture Research. Vol. 2 (2): 00042.

van der Kooi C. J.; Reich M.; Löw M.; De Kok L. J. and Tausz M. (2016). Growth and yield stimulation under elevatedCO_2 and drought: a meta-analysis on crops. Environmental and Experimental Botany. Vol. 122: 150–157.

Waqas M. A.; Wang X.; Zafar S. A.; Noor M. A.; Hussain H. A.; Azher Nawaz M.; Farooq M. (2021). Thermal Stresses in Maize: Effects and Management Strategies. Plants 2021. Vol. 10: 293.

Windell Hydroponics.co.za, www.hydroponics.co.za.

Williams M. M. (2012). Agronomics and economics of plant population density on processing sweetcorn. Field Crops Research. Vol. 128: 55–61.

Wolfe D. W. (1991). Low temperature effects on early vegetative growth, leaf gas exchange and water potential of chilling-sensitive and chilling-tolerant crop species. Annals of Botany. Vol. 67: 205–212.

Zhang Y.; Duan B.; Qiao Y.; Wang K.; Korpelainen H. and Li C. (2008). Leaf photosynthesis of *Betula albosinensis* seedlings as affected by elevated CO_2 and planting density. Forest Ecology and Management. Vol. 255: 1937–1944.

Chapter 4

Does elevated CO_2 affect the nutritional and leaf pigment parameters of sweetcorn (*Zea mays* L. var. *saccharata*)? Mineral composition of fruit, leaf carotenoids and leaf chlorophylls

Abstract

Sweetcorn (*Zea mays* L. var. *saccharata*) is one of the most consumed fresh vegetable worldwide. Sweetcorn is an early maturing horticultural crop consisting of essential mineral nutrients and leaf pigments. Because of the climate change resulting in accumulated GHGs with CO_2 considered as the main culprit contributing to global warming, nevertheless, predicted climate change interference are projected to cause shortcomings in sweetcorn production because of negative influence on mineral nutritional quality of sweetcorn. The effect of elevated CO_2 on mineral absorption by sweetcorn has not been well documented. The aim of the study was to investigate the effect of elevated CO_2 on selected nutritional quality parameters of sweetcorn and the objective were to analyse the mineral composition of fruit and leaf pigments. Five fresh sweetcorn cobs were harvested for only 'STAR7719' cultivar and analysed in three kernel locations (top, middle and bottom cob). Analysed from 12 green leaf samples were carotenoids and chlorophyll pigments collected from 'Assegai' and 'STAR7719' under eCO_2 (500 ppm) and ambient CO_2 (430 ppm) concentrations. There we no significant difference in mineral nutrients for 'STAR7719'. No further significant differences occurred in leaf pigments under eCO_2. Further investigations of eCO_2 on mineral nutrition and leaf pigments of sweetcorn needs to be studied even further to comprehend the causes of declines in crop quality under eCO_2 concentrations higher than 550 ppm.

4.1 Introduction

The global atmospheric carbon dioxide concentration [CO_2] is likely to continue to escalate, thereby potentially straining plant biochemistry (Ezzati *et al.,* 2002; Caulfield and Black, 2004; Stoltzfus *et al.,* 2004; Tulchinsky, 2010; Chakraborty *et al.,* 2011). It is predicted that the future climatic conditions will impact food nutrient concentration, as well as the productivity of staple cereal crops (Högy *et al.,* 2009; Chakraborty 2013). This will impact on potential nutrient absorption from the soil. If this should be the case, the nutritional quality of such crops for human and animal consumption is likely be affected (Lönnerdal, 2002).

Iron and zinc have many functions in the human body, such that their lack lead to severe consequences; thus, deficiencies in these elements have a great impact on people's health and the economic development of countries (Hunt, 2005). The world-wide most-common nutritional disorder is iron deficiency, affecting close to 4 to 5 billion people, or 66 to 80% of the 8 billion world population, predominantly those from high-risk groups such as children, pregnant women, nursing mothers as well as elderly people (WHO, 2002; 2007; WFP 2007; UNICEF, 2007). The US Department of Agriculture 1994-1996 Continuing Survey of Food Intakes measured individually reported that, in men and women aged over 20 the mean daily intake of Zn was between 13.5 and 9.0 mg, 12.0 mg for men and women of 60 years and older. Women, as well as children of less than 1, 1-3 years and 4-5 years requiring Zn daily intakes from 6.6, 7.6, and 9.1 mg, respectively. Many countries are reported to have average intake of enough Zn, while other population groups are not fortunate enough in terms of adequate Zn intake which put people at risk especially in developing countries, as low Zn intake contribute to high risk, which include poverty, scarcity of food, and food preferences with low mineral concentrations (Maret and Sandstead, 2006). Another health-impairing factor, found to be the root source of anemia, is iron (Fe) deficiency (Mabesa *et al.*, 2013); anemia is observed as a reduced number of red blood cells or rather the amount of haemoglobin in the blood, linked closely to inadequate dietary intake, or even poorly absorbed iron from food (NHLBI 2014; NIH, 2014).

Ortiz-Monasterio *et al.* (2008) stated that over 2 billion people across the world are lacking zinc (Zn) in their diet (over 30% of the globe's population). Zinc deficiency has serious effects on human body, such as stunted growth and a weakened immune system, symptoms that are prevalent in many regions of the world. Further elements of major significance when lacking in a person's diet are calcium (Ca) and magnesium (Mg). Calcium, a divalent ion, has multiple functions in the life cycle of plants. The structural function of calcium is crucial in providing strength to cell walls and membranes and is, therefore, necessary for growth and development of roots and stems (White and Broadly, 2003; Lecourieux *et al.*, 2006). The functions of Ca and Mg in plant growth have been well documented, but still little is known about both these nutrients, especially about their impact on green mealies growth and their crop yield (Szczepaniak *et al.*, 2016).

Sweetcorn (*Zea mays* L. var. *saccharata*) is a versatile vegetable crop known to contain vitamin A along with β-carotene and other pigments, such as high levels of phenolic terpenoid pigment antioxidants such as lutein, xanthine and cryptoxanthin pigments, (USDA National nutrient data base), compound from the vitamin B-Complex, such as thiamine, niacin, pantothenic acid, folate, riboflavin, as well as pyridoxine (Kumar and Jhariya, 2013). The production of sweetcorn in countries, such as Argentina, has in recent years been gradually increasing; this crop is regarded as a horticultural crop of significantee nutritional value when consumed as fresh maize and for food industry (Bertolaccini *et al.,* 2010).

Sweetcorn, also termed ‘supersweet’, are group of maize cultivars that have been developed for over 25 years (ERS, 2007) with a high sucrose concentration in the caryopsis, is harvested at an earlier stage of fruit development (commonly three weeks after flowering, when at the early dough stage) than maize grown for dry grain (NDA, ARC-Grain Crops Institute, 1998). Moreover, sweetcorn serves as an essential source of protein, Vitamin A and B, iron, and other minerals (Badu-Apraku and Akinwale, 2011). According to NASS (2017), sweetcorn production and value ranks the second in terms of largest processing crop, retaining higher demand only exceeded by tomatoes. Moreover, sweetcorn processing for frozen and canned food totaled an amount of 2.5 million tons in 2015 with a value of $255.5 million (NASS, 2017).

To be consumed as vegetable, sweetcorn is typically harvested prior to physiological maturity, the fruit (botanically a caryopsis – a simple, dry fruit, monocarpellate without a suture and, therefore, not opening at maturity) is, due to being physiologically ‘pre-mature’, not able to germinate to produce a new plant. (Serna-Saldiva *et al.,* 2000). Meanwhile, Pereira Jr. (2003) reported such an escalation in green mealies production in South America, that traditional producers of dry grain maize, beans, and coffee beans are abandoning these traditional crops in favour of exploring green mealies production. Pereira Jr. (2003) also reported that fresh maize is receiving more acceptance by the consumers in favour of other crops.

Sweetcorn and green mealies are not only a dry staple food, but also a pharmaceutical crop, because DeFelice, (1994) discovered that maize has medicinal and nutraceutical properties, elements of the vitamin B-Complex, important in the maintenance of healthy skin and hair, a proper digestion and brain, and heart function, but also enhance the immune system as well as the thyroid gland activity (Kazerooni *et al.,* 2019). These compounds provide health benefits, when consumed and potentially prevent or even treat diseases. Maize kernels contain various macro- and micronutrients essential for the human metabolism the amounts of several important nutrients are inadequately present in the diet of people using maize as a major food source. For example, maize kernels are low in the elements iron, zinc, calcium, iodine, as well as in vitamin C (ascorbic acid) and in B vitamins.

Previous studies examining how increased atmospheric CO_2 concentrations affect crop growth and development, as well as crop nutrient concentration, have been hindered by only being able to use artificial growing environments, as these only allow small sample sizes and plants grown in controlled environmental chambers in relatively small pots, which limits root growth (Vanaja *et al.,* 2006). Plants grown in enclosed chambers and glasshouses under eCO_2 tend to have lower mineral concentrations, but the effect was not found to be significant, leading some researchers doubting a reduced mineral concentration in plants grown under eCO_2 under fields conditions (Loladze, 2014).

Various reports exist on lower zinc, iron, and protein concentrations in wheat (Manderscheid *et al.,* 1995; la Puente *et al.*, 2000), barley (Manderscheid *et al.,* 1995), and rice (Seneweera *et al.*, 1997) grown under ambient CO_2 (aCO_2, between 364-386 ppm) than under elevated (e) CO_2 (between 546-586 ppm) in outdoor, open-top chambers (OTC) or indoor, environmentally controlled growth chambers (Dietterich *et al.*, 2015). Furthermore, problems associated with plants grown in OTCs, controlled environmental chambers, greenhouses, as well as growth chambers pointed out the unlikely effect of eCO_2 in such chambers, simply because in enclosed structures the environment is not identical to the open field environment (Kimball *et al.,* 1997).

As a vegetable, sweetcorn forms an important part of the human diet, it is, therefore, vital to investigate the effect of eCO_2 on the nutritional concentration in sweetcorn and as 'green mealies,' as previous investigations have focused only on dry grain maize. Increased CO_2 concentrations may, however, affect nutrient uptake, distribution and, therefore, concentration in sweetcorn. The aim of the current study was therefore, to investigate the nutritional concentration of sweetcorn, as well as possible nutritional enhancements, as well as postharvest shelf life, of the sweetcorn cultivars 'Assegai' and 'STAR7719'. 'Assegai' was only sampled for leaf chlorophylls analysis and therefore, no data were available for fruit analysis since the control lodged resulting in plant death. The objectives of the study were to determine leaf carotenoid and chlorophyll concentrations as well as mineral nutrient concentration of Al, Fe, Mn, Zn, Cu, Ca, Mg, Na, P and K in fresh kernels of 'STAR7719' sweetcorn.

4.2 Materials and Methods

4.2.1 Plant material

Fresh green leaves were sampled for mineral analysis from sweetcorn plants in two glasshouses grown in 30 cm plastic pots. Each chamber had two local sweetcorn hybrids cultivars obtained from Starke Ayres (Pietermaritzburg, SA) (‘Assegai’ and ‘STAR 7719’) replicated nine times. Sowing took place in winter on the 1st of June 2021 into 30 cm plastic pots filled with soil from Ukulinga Research Farm, University of KwaZulu Natal, Pietermaritzburg. The soil type was a heavy clay. Pots were arranged in a randomized complete block design in three rows per cultivar on the floor of the adjacent glasshouses (as described in the descriptive chapter 2). The one glasshouse contained 10 ‘Because Nature Mycelium CO_2 generator bags’ (as described in the descriptive chapter 2) intended to raise CO_2 concentration from ~430 to 500 ppm, while the other glasshouse remains with an ambient CO_2 concentration between 400-430 ppm. Fresh sweetcorn cobs were harvested from a treatment and a control from the 8th to the 21st of September to determine mineral and nutritional compounds.

4.3 Laboratory analysis

4.3.1 Determination of mineral nutrients in fresh sweetcorn kernels

4.3.1.1 Material

Six fresh sweetcorn cobs from a treatment and a control were dried in the oven at 40°C for three days and milled into powder. Each maize powder was divided into three cob locations *i.e.,* (top, middle, bottom cob) part. Nutritional elements were analysed using ICP-OES (Al, Fe, Mn, Zn, Cu, Ca, Mg, Na, P and K) (Hunter, 1984) at the ARC-Cedara Analytical Plant Laboratory, Pietermaritzburg.

4.3.1.2 Methods

The crucibles used for ashing were stored overnight in an oven set at 110°C. On the following morning, the crucibles were cooled in a desiccator for 30 minutes and removed using pair of tongs. Crucible was recorded prior to placing the milled samples (0.5 g DM) into the crucible. The crucibles were placed in an oven, set at 110°C, for 2 hours. Then crucibles were removed using tongs, cooled in a desiccator for 30 min, and were weighed once again. The mass of the crucible plus the sample was recorded prior to be taking it to the furnace room for ashing.

The crucibles placed into the furnace at 450°C for 4 hours together with blanks. The furnace was allowed to cool off prior to working on the sample day. When the furnace was opened on the next day, crucibles with the ash were removed and taken for digestion.

Crucibles containing the ash were hydrated with few drops of distilled water and a 2 ml concentrated HCl were added to each sample. Samples were slowly evaporated to dryness in a water bath in a fume cupboard equipped with an extractor fan. A Fortuna Optifix dispenser, Poulten & Graf GmbH Fortuna®, Wertheim, Germany) was used to add 25 ml freshly prepared 1:9 HCl solution and stirred using a rubber policeman stirring rod, rinsing the rod in a beaker of distilled water in between each sample. Subsequently, the samples were filtered through Advantech filter paper (5B:90 mm diameter) into a clean rack of sample cups. The filtered sample was diluted with de-ionized water at a ratio 5:20 and further analysed by ICP-OES (PerkinElmer, Inc. Waltham, USA) or determination of elements.

4.3.2 Determination of carotenoid and chlorophyll concentrations in sweetcorn leaves

Chlorophyll and carotenoid pigments were determined in fresh twelve leaves sampled from sweetcorn plants in the morning hours (08: h30-09: h30). Fresh leaves were instantly placed into plastic pockets and transported in a cooler box straight to the laboratory. Fresh leaf material was extracted using acetone 80%. The concentrations of leaf chlorophylls and carotenoids were determined spectrophotometrically (Shimadzu UV-1800 UV-Vis Spectrophotometer, Japan), as described by Lichtenthaler and Buschmann, (2001) and Lichtenthaler (1987). Leaf tissue samples were cut and 1g subsamples, avoiding leaf margins, produced. The measured fresh leaf material was placed into centrifuge tubes and mixed with 4 ml 80% acetone. Tubes were left to stand for 10 minutes on ice covered with aluminium foil. After 10 min, samples were homogenized with mortar and pestle and transferred to centrifuge tubes. The paste remaining in the mortar was rinsed off with an additional 4 ml of 80% acetone and samples centrifuged for 5 min at 4000 rpm. Absorbance of supernatants were read at 663.2; 646.8; and 470 nm using 80% acetone as a blank, and, therefore, pigment concentrations in µg/mL were calculated using the equations as follows:

Table 4.1 Equations used to determine the concentrations (µg/ml) of pigments in which A = absorbance determined with a spectrophotometer, chlorophyll a (Ch-a), chlorophyll b (Ch-b), chlorophyll a+b (ch-a+b) and carotenoids (C-x+c) refer to relevant pigment concentration using acetone (80%) (Porra *et al.,* 1989; Lichtenthaler, 1987 and Lichtenthaler and Wellburn, 1983).

Solvent	Formulae (µg/mL)
80% acetone (v/v)	$C_a = 12.25\ A_{663.2} - 2.79\ A_{646.8}$ $C_b = 21.50\ A_{646.8} - 5.10\ A_{663.2}$ $C_{a+b} = 7.15\ A_{663.2} + 18.71\ A_{646.8}$ $C_{x+c} = (1000\ A_{470} - 1.82\ C_a - 85.02\ C_b) / 198$

4.4 Results

GenStat®18th edition (VSN International, Hemel Hempstead, UK) software was used to analyze data statistically through the application of the analysis of variance (ANOVA). The parameters analyzed were: leaf carotenoids and chlorophylls, as well as mineral nutrient concentrations in maize kernels. The differences in the means of these pigments and minerals, determined using LSD's (Fisher's protected test), revealed no significant differences at P<0.05. No effect from elevated CO_2 concentration of mineral nutrients of sweetcorn fruit was detected (Table 4.2).

The elevating CO_2 had no significant effect on both leaf pigments groups of both cultivars 'Assegai' and 'STAR7719' (Figure 4.1). Decline in leaf carotenoids and all leaf chlorophylls concentrations were recorded for treatments over controls (Table 4.3) under eCO_2 concentration. Mean chlorophyll a concentration in leaf tissues were 0.0069 µg/gFM under eCO_2 compared with 0.118 µg Chl a/gFM were determined under aCO_2. 'STARR7719' contained 0.11 µg Chl a/gFM under eCO_2, while 0.12 µg Chl a/gFM were determined under aCO_2. The chlorophyll b concentration of 'Assegai' averaged 0.096 µg/gFM under eCO_2 compared with 0.146 µgChl b/gFM under aCO_2 (Figure 4.1). The total leaf chlorophyll a+b of 'Assegai' leaves was lower under eCO_2 under than aCO_2, while carotenoid concentrations lower (Figure 4.1) than under aCO_2 concentrations. Meanwhile, in 'STAR7719' the total chlorophyll a+b was lower under eCO_2 than under aCO_2 concentration. The leaf chlorophyll a and b for cultivars 'Assegai' and 'STAR7719' was not significantly affected by CO_2 at P=0.17

and P=0.67. Similarly, CO_2 had no significant effect on total leaf chlorophylls a+b and total leaf carotenoids for cultivars ‘Assegai’ and ‘STAR7719’ at P=0.386 and P=0.702 (Table 4.2).

Table 4.2 Effect of eCO_2 on mineral nutrients in all of the cob (top, middle, and bottom) parts of sweetcorn (‘STAR7719’) cobs.

Parameters/ Cob	STAR 7719						
	aCO_2	**eCO_2**	**Cob**	**CO_2**	**Cob x CO_2**	**Mean**	**LSD**
N %	2.97	2.49	0.534^{ns}	0.188^{ns}	0.308^{ns}	2.73	1.179
Ca %	0.039	0.037	0.125^{ns}	0.945^{ns}	0.778^{ns}	0.04	0.071
Mg %	0.217	0.207	0.54^{ns}	0.285^{ns}	0.866^{ns}	0.21	0.025
K %	1.025	1.020	0.323^{ns}	0.229^{ns}	0.737^{ns}	1.04	0.134
P %	0.49	0.48	0.512^{ns}	0.535^{ns}	0.627^{ns}	0.49	0.03
Na mg/kg	21.9	25.4	0.200^{ns}	0.429^{ns}	0.201^{ns}	21.8	46.04
Fe mg/kg	48.8	49.9	0.480^{ns}	0.881^{ns}	0.326^{ns}	49.4	34.56
Zn mg/kg	59.6	54.7	0.747^{ns}	0.565^{ns}	0.526^{ns}	57.2	21.80
Cu mg/kg	1.33	1.63	0.749^{ns}	0.561^{ns}	0.450^{ns}	1.48	1.342
Mn mg/kg	26.0	24.5	0.099^{ns}	0.697^{ns}	0.073^{ns}	25.3	10.10
Al mg/kg	4.10	1.21	0.884^{ns}	0.222^{ns}	0.129^{ns}	2.65	5.443

ns indicates non-significant; CO_2 levels are ambient CO_2 (~430 ppm) and elevated CO_2 (~500 ppm) conditions.

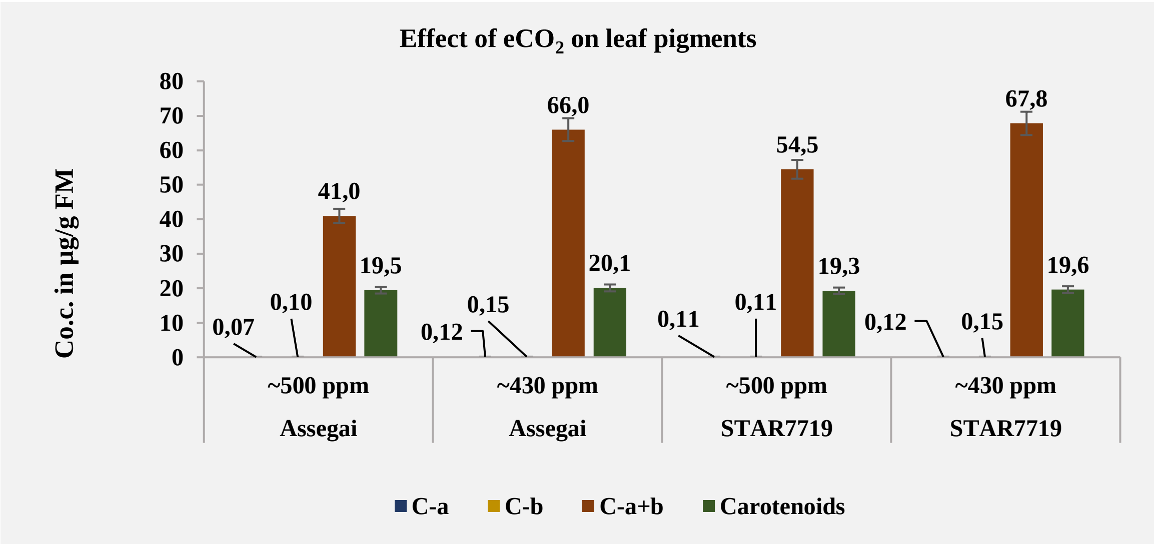


Fig. 4.1 Leaf chlorophyll a (Ca), chlorophyll b (Cb), total leaf chlorophyll a+b (Ca+b) and total leaf carotenoid (Cx+c) concentrations in sweetcorn cultivars ‘Assegai’ and ‘STAR7719’grown under control (430 ppm) and elevated (500 ppm) CO2 conditions. Error bars indicate pigment concentrations in µg/gFM.

Table 4.3. Effect of eCO_2 on leaf pigments of sweetcorn ('Assegai' and 'STAR7719') leaf tissues.

	Assegai		STAR7719						
Parameters	**aCO_2**	**eCO_2**	**aCO_2**	**eCO_2**	**C**	**CO_2**	**C x CO_2**	**Mean**	**LSD**
Chlorophyll a	0.118	0.069	0.12	0.11	0.12^{ns}	0.02^{*}	0.17^{ns}	0.103	0.037
Chlorophyll b	0.146	0.096	0.150	1.11	0.4^{ns}	0.02^{*}	0.67^{ns}	0.126	0.051
Chlorophyll a+b	66.0	41.0	67.8	54.5	0.27^{ns}	0.02^{*}	0.386^{ns}	57.3	21.85
Carotenoids x+c	20.10	19.46	19.60	19.2	0.35^{ns}	0.21^{*}	0.702^{ns}	19.60	1.208

ns indicates non-significant; C indicates cultivar; CO_2 levels are ambient CO_2 (~430 ppm) and elevated CO_2 (~500 ppm) conditions.

4.5 Discussion

The concentration of N% was not significant affected under eCO_2 with a mean of 2.49% compared to mean of 2.97% under aCO_2 conditions. Minerals Mg, K, and P were not significantly affected by eCO_2 and tended to be lower under eCO_2 compared with aCO_2 conditions (Table 4.3). Previous research in Poaceae species by Brown (1978) found that in C4 grasses, total nitrogen was produced less in leaves while more plant dry matter was produced per unit of nitrogen fertilizer (N, % of DM) applied than dry matter produced in C3 grasses under eCO_2, leading to hypothesis that nitrogen might be greatly utilised more efficiently in C4 plants. Similarly, Myers *et al.* (2014) reported that eCO_2 had no significant effect on the nitrogen content on maize plants. Furthermore, it is unclear, whether nitrogen restriction is contributed to C4 plants evolution, and, therefore, conclusions are that escalating CO_2 concentrations do not pose a threat to reduced nitrogen in C4 species simply because C4 plants are already saturated at currently available CO_2 concentrations (von Caemmerer and Furbank, 2003).

The leaf calcium concentration was not significantly influenced by eCO_2, with 0.037% N under eCO_2 and 0.039% N under aCO_2 conditions; these findings disagree, however, with Dong *et al.* (2018) who reported that leaf Ca increased to 8.2% under eCO_2, pointing out that there is neither an effect from dilution nor a transpiration limitation that could possibly explain the enhanced accumulation of Ca under eCO_2 in root, all vegetables (root, stem, leafy and fruit).

There were no significant differences in Fe, but under eCO_2, there was a tendency to an increased Fe concentration in all of the kernels (49.9 mg/kg kernel DM compared with 48.8

mg kernel DM/kg under aCO_2. Zinc concentration in kernels were also not significantly affected by eCO_2 (54.7 mg kernel DM/kg compared with 59.6 mg kernel DM/kg under aCO_2. Sodium, copper, manganese, and aluminium were also not significantly different between eCO_2 and aCO_2 condition (Table 4.3), although a tendency towards an increase in Na and Cu under eCO_2 was recorded. These findings agree with Dong *et al.* (2018), who discovered that minerals, such as Mg, Fe, and Zn, decreased in their concentrations by 9.2%, 16.05, and 9.4% of FM under eCO_2 conditions, whilst the concentrations of P, K, S, Cu, and Mn were maintained.

Additionally, the mineral nutrient on leafy vegetables (cabbage, Chinese cabbage, chives, fenugreek, Hongfengcai, lettuce, palak, spinach and oil sowthistle) declined in Fe concentration by 31.0% under eCO_2, as well as fruit and root vegetables, Fe concentration declined by 19.2% and 8.2%; however, the Zn concentration declined in both, fruit and root vegetables by 18.15, while stem vegetables (broccoli, celery, celtuce, Chinese kale, ginger, scalion), Fe concentration declined by 10.7% (Dong *et al.*, 2018). Previous investigations reported that mineral nutrient declined in concentration due to an eCO_2 dilution effect (Fangmeier *et al.*, 2002; Högy and Fangmeier, 2009; Loladze, 2014) or even the restricted transpirations (McDonald *et al.*, 2002). Other reviews identified a decrease in all mineral nutrient concentrations of grain crops under eCO_2 conditions (Loladze, 2014; Myers *et al.*, 2014), again pointing out that the decrease in minerals is particularly controlled by metabolic processes but it is due to the dilution effect from increased biomass achieved under eCO_2.

Elevated CO_2 had no significant effect on leaf pigments the maize cultivars of 'Assegai' and 'STAR7719', but reduced leaf pigments might have been due to possible changes in physiology because of a need for adaptability to eCO_2 concentrations (Houpis *et al.*, 1988). Several authors (Delucia *et al.*, 1985; Houpis *et al.*, 1988; Wullschleger *et al.*, 1992) have reported such frequent declines in leaf chlorophyll, particularly in young leaves in response to eCO_2. Meanwhile, Sgherri *et al.* (1998) found that the alfalfa plants accumulated greater chlorophyll concentrations and plants (fruit) had higher lipid to protein ratios when grown under eCO_2 levels.

Previous investigations (Kooij *et al.*, 1999; Bae *et al.*, 2004; Cheng *et al.*, 1998) have reported insignificant changes in leaf carotenoids as well as chlorophyll concentrations under eCO_2, with emphasis on the photosynthetic pigment pool being highly resilient to eCO_2 levels. This could also be interpreted as both, carotenoid and chlorophyll concentrations, not being only associated with the photosynthetic rate, but also with leaf development and carotenoid accumulation due to leaf age and leaf type (Dhami *et al.,* 2018). Van der Kooij *et al.* (1998) also reported that total chlorophyll, carotenoid and chlorophyll a/b ratio of *Arabidopsis thaliana* leaves were slightly affected by being grown under eCO_2. The findings of this study (Figure 4.1) of a decline in leaf chlorophyll agree with the findings by Sage *et al.,* (1989) and Tissue *et al.,* (1993), that leaf N percentage in Rubisco was relatively reduced in plants grown under eCO_2 levels. Other authors (Downton *et al*. 1980, Patterson and Flint 1982; DeLucia *et al.* 1985; Oberbauer *et al.* 1986) identified a reduced leaf chlorophyll concentration; however, enhanced growth in response to elevated CO_2 have been reported in (*Daucus carota*, *Gossypium hirsutum*, *Beta vulgaris, Abelmoschus esculentus, Brassica napus, Glycine max and P. macroloba* and *O. lagopus*) (Poorter, 1993).

4.6 Conclusions

Seemingly, eCO_2 plays a vital role in leaf pigment composition and fruit mineral nutrient composition. The adaptability and resilience of sweetcorn plants to eCO_2 conditions still needs to be explored further, using higher CO_2 concentrations, exceeding 700 ppm, such that higher leaf pigment as well as mineral nutrient concentrations could be observed. It is possible, that such eCO_2 positively impacts leaf pigments concentrations and mineral nutrient concentrations. There seems to be a scarcity of adequate literature focusing on eCO_2 impacts on leaf pigments, especially leaf carotenoids. Such investigation is, however, crucial for the greater understanding of the impact of eCO_2 the reduced carotenoid and chlorophyll concentrations in leaf tissues and in the reduced leaf mineral concentration. It is, therefore, necessary to extend the studies of eCO_2 on plants growth and their response, to such alteration, in order to fully understand how sweetcorn productivity (yield) and the quality of the produce will be altered under CO_2 enrichment.

Literature cited

Badu-Apraku B. and Akinwale R. (2011). Identification of early-maturing maize inbred lines based on multiple traits under drought and low N environments for hybrid development and population improvement. Canadian Journal of Plant Science. Vol. 91: 931–942.

Bae H. H.; Sicher R. (2004). Changes of soluble protein expression and leaf metabolite levels in Arabidopsis thaliana grown in elevated atmospheric carbon dioxide, Field Crop Research. Vol. 90(1): 61- 73.

Bertolaccini I.; Bouzo C. A.; Larsen N. and Favaro J.C. (2010). Species of the genus Euxesta Loew (Diptera: Ulidiidae (Otitidae)), pests of Bt sweetcorn in Santa Fe province, Argentina. Revistas de la Sociedad Entomologica Argentina. Vol. 69: 123-126.

Brown R. H. (1978). A difference in the nitrogen use efficiency of C3 and C4 plants and its implications in adaptation and evolution. Crop Science. Vol: 18, pp 93–98.

Chakraborty S. and Newton A. C. (2011). Climate change, plant diseases and food security: an overview. Plant Pathology. Vol. 60: 2−14.

Chakraborty S. (2013). Migrate or evolve: Options for plant pathogens under climate change. Global Change in Biology. Vol.19: 1985−2000.

Cheng S. H.; Moore B.; Seemann J. R. (1988). Effects of short- and long-term elevated CO_2 on the expression of ribulose-1,5-bisphosphate carboxylase/oxygenase genes and carbohydrate accumulation in leaves of Arabidopsis thaliana (L.) Heynh, Plant Physiology. Vol. 116(2): 715-23.

Caulfield L. E. and Black R. E. (2004). Comparative quantification of health risks : Global and regional burden of disease attributable to selected major risk factors (Eds Ezzati *et al.*, 2004). World Health Organization. https://apps.who.int/iris/handle/10665/42770.

DeFelice S. L. (2017). What is a True Nutraceutical? And – What is the Nature and size of the US Nutraceutical Market? 1994, http://www.fimdelice.org./ p2462.htm/.

Delucia E. H.; Sasek T. W. and Strain B. R. (1985). Photosynthesis inhibition after long-term exposure to elevated levels of atmospheric carbon dioxide. Photosynth. Research. Vol. 7: 175-184.

Dietterich L. H.; Zanobetti A.; Kloog I.; Huybers P.; Leakey A. D. B.; Bloom A. J.; Carlisle, E.; Fernando N.; Fitzgerald; G.; Hasegawa T.; Holbrook N. M.; Nelson R. L.; Norton R.; Ottman M. J.; Raboy V.; Sakai H.; Sartor K. A.; Schwartz J.; Seneweera S.; Usui Y.; Yoshinaga S. and Myers S. Impacts of elevated atmospheric CO_2 on nutrient content of important food crops. Scientific Data 2:150036.

Dong J.; Gruda N.; Lam S. K.; Li X. and Duan Z. (2018). Effects of Elevated CO_2 on Nutritional Quality of Vegetables: A Review. Frontiers in Plant Science. Vol. 9: 924.

Downton W. J. S.; 0. Bjorkman and C. S. Pike. (1980). Consequences of increased atmospheric concentrations of carbon dioxide for growth and photosynthesis of higher plants. In Carbon Dioxide and Climate. Ed. G.I. Pearman. Australian Research, Australian Academy of Science, Canberra, Australia. (pp 143-151).

Ezzati M., Lopez A. D., Rodgers A., Vander Hoorn S., Murray C. J. L. (2002). The Comparative Risk Assessment Collaborating Group: Selected major risk factors and global and regional burden of disease. The Lancet Vol. 360: 1347–1360.

Fangmeier A.; De Temmerman L.; Black C.; Persson K. and Vorne V. (2002). Effects of elevated CO_2 and/or ozone on nutrient concentrations and nutrient uptake of potatoes. Eur. J. Agron. Vol. 17: 353–368.

Högy P. and Fangmeier A. (2009). Atmospheric CO_2 enrichment affects potatoes: 2. Tuber quality traits. European Journal of Agronomy. Vol. 30: 85–94.

Houpis J. L. J.; Surano K. A.; Cowles S. and Shinn J. H. (1988). Chlorophyll and carotenoid concentrations in two varieties of Pinus ponderosa seedlings subjected to long-term elevated carbon dioxide. Tree Physiology. Vol. 4: 187-193.

Hunt J. M. (2005). The potential impact of reducing global malnutrition on poverty reduction and economic development. Asia Pacific Journal of Clinical Nutrition, Vol. 14: 10-38.

Hunter A. H. (1984). Soil Analytical Services in Bangladesh. BARI/Aids Consultancy Report. Contract Aid/388-005, Dhaka. Bangladesh. (pp. 1-7).

Inglett G. E. (1970). Maize: Culture, Processing, Products. Major Feed and Crops in Agriculture and Food Science Series. 369 Seiten, 73 Abb., 65 Tab. The AVI Publishing, Inc., Westport, Connecticut.

Kazerooni E. G.; Sharif A.; Nawaz H.; Rehman R. and Nisar S. (2019). Maize (Maize)-A useful source of human nutrition and health: a critical review. International Journal of Chemical and Biochemical Sciences (ISSN 2226-9614) Journal Home page: www.iscientific.org/Journal.html © International Scientific Organization.

Kimball B. A.; Pinter P. J.; Jr. Wall G. W.; Garcia R. L.; LaMorte R. L.; Jak P. M. C.; Frumau K. F. A. and Vugts H. F. (1997). Comparisons of responses of vegetation to elevated carbon dioxide in free air and open top chamber facilities in Advances in Carbon Dioxide Research, edited by L. H. Allen (Jr); M. B. Kirkham; D. M. Olszyk and C. E. Whitman (Am. Soc. Agron. Crop Science Society of America and Soil Science Society of America, Madison, WI, USA), (pp 113-130).

Kumar D. and Jhariya, A. N. (2013). Nutritional, medicinal, and economical importance of maize: A mini review. Research Journal of Pharmaceutical Sciences ISSN. 2319: 555x.

la Puente de L. S.; Perez P. P.; Martinez-Carrasco R.; Morcuende R. M. and del Molino I. M. M. (2000). Action of elevated CO_2 and high temperatures on the mineral chemical composition of two varieties of wheat. Agrochimica Vol. 44: 221–230.

Lichtenthaler H. K. and Wellburn, A. R. (1983). Determinations of total carotenoids and chlorophylls a and b of leaf extracts in different solvents, Biochemical Society of Transactions. Vol. 11: 591–592.

Lichtenthaler H. K. (1987). Chlorophylls, and carotenoids: pigments of photosynthetic membranes, Method Enzymol. Vol. 148: 350–382.

Lichtenthaler H.K. and Buschmann C. (2001). Chlorophylls and Carotenoids: Measurement and Characterization by UV-VIS Spectroscopy, Current Protocols in Food Analytical Chemistry (F4.3.1-F4.3.8), John Wiley & Sons, Inc.

Loladze I. (2014). Hidden shift of the ionome of plants exposed to elevated CO_2 depletes minerals at the base of human nutrition. Ecology/Epidemiology and global health. *eLife* 3:e02245.Research Article. DOI: 10.7554/eLife.02245.001.

Lönnerdal B. (2002). Phytic acid-trace element (Zn, Cu, Mn) interactions. International Journal of Food Science & Technology, v. 37, p. 749-758.

Mabesa R.; Impa S.; Grewal D. and Johnson-Beebout S. (2013). Contrasting grain-Zn response of biofortification rice (Oryza sativa L.) breeding lines to foliar Zn application, Field Crop Research. Vol. 149: 223-233.

Manderscheid R.; Bender J.; Jäger H. J. and Weigel H. J. (1995). Effects of season long CO_2 enrichment on cereals. II. Nutrient concentrations and grain quality. Agriculture, Ecosystems and Environment. Vol. 54: 175–185.

Maret W. and Sandstead H. H. (2006). Zinc requirements and the risks and benefits of zinc supplementation. Journal of Trace Elements in Medicine and Biology. Vol. 20: 3-18 PMid:16632171. http://dx.doi.org/10.1016/j.jtemb.2006.01.006 Research, 2013, Vol. 149: 223-233.

McDonald E. P.; Erickson J. E. and Kruger E. L. (2002). Can decreased transpiration limit plant nitrogen acquisition in elevated CO_2? Functional Plant Biology. Vol. 29: 1115–1120.

Myers, S. S., Zanobetti, A., Kloog, I., Huybers, P., Leakey, A. D., Bloom, A. J., *et al.* (2014). Increasing CO_2 threatens human nutrition. Nature Vol. 510(7503): 139–142.

National Agricultural Statistics Service (NASS), (2017) http://www.nass.usda.gov/.

NIH (National Heart, Lung, and Blood Institute). What is iron-deficiency anaemia? www.nhlbi.nih.gov. 26 March 2014. Archived from the original on 16 July 2017. Retrieved 17 July 2017.

Nuss E. T. and Tanumihardjo S. A. (2010). Maize: A Paramount Staple Crop the Context of Global Nutrition. Vol. 9: 417-436. Comprehensive Reviews in Food Science and Food Safety. Institute of Food Technologists®.

Oberbauer S. F.; B. R. Strain and N. Fetcher. (1985). Effect of CO_2 enrichment on seedling physiology and growth of two tropical tree species. Physiol. Plant. Vol. 65: 352-356.

OECD (2006). "Section 3 - Maize (ZEA MAYS SUBSP. MAYS)", in Safety Assessment of Transgenic Organisms, Volume 1: OECD Consensus Documents, OECD Publishing, Paris.

Ortiz-Monasterio J.; Palacios-Rojas N.; Meng E.; Pixley K.; Trethowan R. and Pena R. (2007). Enhancing the mineral and vitamin content of wheat and maize through plant breeding, Journal of Cereal Science. Vol. 46(3): 293-307.

Patterson D. T. and E. P. Flint. (1982). Interacting effects of CO_2 and nutrient concentration. Weed Science. Vol. 30: 389-394.

Pereira Filho I. A. (Ed.). (2003). Cultivation of green maize. Brasília, DF: Embrapa Information Technology, (pp 204). ISBN: 85-7383-204-5.

Porra R. J.; Thompson W. A. and Kreidemann P. E. (1988). Determination of accurate extinction coefficients and simultaneous equations for assaying chlorophylls a and b extracted with four different solvents: verification of the concentration of chlorophyll standards by atomic absorption spectrometry, Biochimica et Biophysica Acta (BBA) – Bioenergetics. Vol. 975: 384-394.

Poorter H. (1993). Interspecific variation in the growth response of plants to an elevated ambient CO_2 concentration. Vegetation, Vol. 104 (105): 77-97. J. Rozema; H. Lambers; S. C. Van de Geijn and M. L. Cambridge (Eds). CO_2 and Biosphere © Kluwer Academic Publishers. Printed in Belgium.

Sage R. F.; Sharkey T. D. and Seemann J. F. (1989). Acclimation of photosynthesis to elevated CO_2 in five C3 species. Plant Physiology. Vol. 89: 590-596.

Seneweera S. P. and Conroy J. P. (1997). Growth, grain yield and quality of rice (Oryza sativa L.) in response to elevated CO_2 and phosphorus nutrition. Soil Science and Plant Nutrition. Vol. 43: 1131–1136.

Serna-Saldivar S. O.; Gomez M. H. and Rooney L. W. (2000). Food uses of regular and specialty and their Dry- Milled. In Arnel R. H. (ed.) Specialty Maize 2nd edition.

Sgherri C. L. M.; Quartacci M. F.; Menconi M.; Raschi A. and Navari-Izzo F. (1998). Interactions between drought and elevated CO_2 on alfalfa plants. J Plant Physiology. Vol. 152: 118–124.

Stoltzfu R. J.; Mullany L. and Black R. E. (2004). In Comparative Quantification of Health Risks: Global and Regional Burden of Disease Attributable to Selected Major Risk Factors (eds Ezzati M.; Lopez A. D.; Rodgers A. and Murray C. J. L.) 1 (World Health Organization).

Sweetcorn for Processing, vegetables and melons outlook, Economic Research Service (ERS), USDA, (2007).

Szczepaniak W.; Grzebisz W.; Potarzycki J.; Łukowiak R. and Przygocka-Cyna K. (2016). The magnesium and calcium mineral status of maize at physiological maturity as a tool for an evaluation of yield forming conditions. Journal of Elementology. Vol. 21(3): 881-897. ISSN 1644-2296.

Tissue D. T.; Thomas R. B. and Strain B. R. (1993). Long-term effects of elevated CO_2 and nutrients on photosynthesis and Rubisco in loblolly pine seedlings. Plant Cell Environment. Vol. 16: 859-865.

Tulchinsky T. H. (2010). Micronutrient deficiency conditions: global health issues. Public Health Reviews. Vol. 32: 243–255.

[USDA]. United States Department of Agriculture. (2009). National nutrient database for standard reference. Available from: http://www.nal.usda.gov/fnic/foodcomp/search/. Accessed September and October 2009.

Vanaja M.; Maheswari P.; Ratnakumar P. and Ramakrishna Y. S. (2006). Monitoring and controlling of CO_2 concentrations in open top chambers for better understanding of plants response to elevated CO_2 levels. India Journal of Radio &Space Physics. Vol. 35: 193-197.

Van der Kooij T. A. W.; De Kok L. J.; Stulen I. (1999). Biomass production and carbohydrates content of *Arabidopsis thaliana* at Atmospheric CO_2 Concentrations from 390 to 1680 µl l^{-1}. Plant Biology. Vol. 1(4): 482-486. ISSN 1435-8603.

Von Caemmerer, S., and Furbank, R. T. (2003). The C(4) pathway: an efficient CO (2) pump. Photosynthesis Research. Vol. 77: 191–207.

World Health Organization - WHO. The world health report 2002. Geneva, (2002). (pp248).

World Health Organization - WHO; World Food Programme; United Nations Children’s Fund - UNICEF. Preventing and controlling micronutrient deficiencies in populations affected by an emergency. Geneva, Switzerland: WHO. (2007). Disponível em: http://www.who.int/nutrition/ publications/ WHO_WFP_UNICEF statement.pdf> Acesso em: 22 ago. 2008.

Wullschleger S. D.; Norby R. J. and Hendrix D. L. (1992). Carbon exchange rates, chlorophyll content, and carbohydrate status of two forest tree species exposed to carbon dioxide enrichment. Tree Physiology. Vol. 10: 21-31.

Young K. J. and Long S. P. (2000). Crop ecosystem responses to climatic change: maize and sorghum. In: Reddy KR & Hodges HF (Eds.), Climate change and global crop productivity, CABI International, Oxon, United Kingdom, 9pp. 107-131).

Chapter 5

The effect of CO_2 on biochemical quality of sweetcorn (*Zea mays* L. var. *saccharata*): Phytochemical parameters

Abstract

Over the recent decades, atmospheric CO_2 has increased significantly, and concentrations are speculated to advance even further in future. As consumers have become very aware of vegetable quality, research has been geared to investigate the effect of environmental conditions due to higher atmospheric CO_2 levels on particular quality parameters of vegetables. A specific environmental factor is the escalated atmospheric carbon dioxide concentration, even though it can have a valuable contribution to crop and vegetable production and productivity, excessive accumulation might also yield negative effects on vegetables. The current study was undertaken with the aim to investigate the effect of elevated CO_2 on phytochemical quality parameters of sweetcorn. Maize plants (STAR7719) were established in two adjacent glasshouses, one under ambient CO_2 (aCO_2, ~430 ppm) and a further with elevated CO_2 (eCO_2, ~500 ppm) concentrations using of "Because Nature Mycelium CO_2 generator bags". At horticultural maturity, fresh cobs were harvested and analysed for ascorbic acid, carotenoids, total protein and total soluble solids (TSS) in three cob locations. Ascorbic acid and total soluble solids were significantly increased under eCO_2. Carotenoids and total proteins were not significantly higher under eCO_2, nevertheless, a tendency to increased levels under eCO_2 conditions was observed.

5.1 Introduction

Phytochemical concentrations are considered key components which define the value as food sources (DeLucia *et al.,* 2012). The term 'phyto' in the word 'phytochemical' is consequentially derived from the Greek word 'phyto' referring to plant. Phytochemicals are described as bioactive, non-nutrient chemical compounds stored in plants, such as in fruit, vegetables and whole grains, which are said to function in a manner that reduces the risk of chronic diseases (Liu, 2004). Many plant foods contain starch, the most common storage carbohydrate in plant; its concentration is said to be enhanced in plants grown under eCO_2 conditions (Araujo *et al.,* 2008). Studies have also postulated that there is an increase in phenolic concentrations in plants grown under eCO_2 conditions, and such a phenolic increase negatively affects insects feeding on plants (Hartley *et al.,* 2000; Gao *et al.,* 2008).

In the quest for an enhanced nutrient availability in crops, elevated CO_2 concentrations in the atmosphere have been hypothesised to improve the absorption of soluble sugars, starch, and amino acids (Moore *et al.,* 1998; Florian *et al.,* 2014; Noguchi *et al.,* 2015); moreover, it is likely that eCO_2 concentrations will also result in the higher concentrations of precursors of isoprenoid substances that are crucial for carotenogenesis (Dhami *et al.,* 2018), however, a major catastrophic predicament defined by Foster, (2007) was that of greenhouse (GHGs) gasses such as methane (CH_4) and nitrous oxide (N_2O) emissions from the land on which crops are produced through:

$$CH_4 + N_2O + 2H_2O \rightarrow 2NH_3 + CO_2 + H_2O \qquad \text{…………….equation (1)}$$

These greenhouses gasses pose a threat to the ozone layer, consequently declining all major crop yields (Thomson *et al.,* 2005). Nonetheless, numerous authors (Philips *et al.,* 1996; Brown and Rosenberg, 1999; Parry *et al.,* 2004; Thomson *et al.,* 2005) described the enhance atmospheric CO_2 levels as "CO_2 fertilization", purely for the reason that biomass and crop yield is effectively increased when grown under eCO_2 concentrations. Maize kernels lack essential phytochemicals such as vitamin C (ascorbic acid), and vitamin B; white maize, the preferred maize choice for human consumption in many parts of the world is obviously devoid of carotenoids and therefore, provitamin A (Nuss and Tanumihardjo, 2010). On the other hand, Adom and Liu (2002) determined that yellow maize has a higher total antioxidant activity, ranging from 181.42 ± 0.86 µmol/g vitamin C equiv/g grain) in between common grains such as rice ranging from 55.77 ± 1.62 µmol/g, wheat ranged from 76.70 ± 1.38 µmol/g as well as oats ranging from 74.67 ± 1.49 µmol/g. Health benefits of green mealies are, not only linked to basic nutrients, such as carbohydrates, vitamins, and minerals, but are also linked to phytochemicals, such as phenolics, which contribute to the nutritional composition of green mealies (Adom and Liu, 2002; Smith *et al.*, 2004). Therefore, the present climate change could be beneficial, such that it may improve crop productivity (Thomson *et al.,* 2005), by simply taking out greenhouse gases, such as methane (CH_4) and nitrous oxide (N_2O) and fixing them to the soil by means of carbon sequestration.

Sweetcorn, *Zea mays* L. *var saccharata*, is the consumed vegetable, is characterised by a higher concentration of sugar than grain maize. It is regularly grown for fresh food or processed into canned products (Zhang *et al.*, 2016). A similar produced, green mealies, is a commodity from the same genus, *Zea mays*, but is the immature form of grain maize. Both commodities contain many phytochemicals and mineral nutrients (Hu and Xu, 2011). Sweetcorn is harvested while the ears, have moisture percentage of about 70 to 80% (Albuquerque *et al.,* 2008; Fritsche-Neto and Silva, 2011). Additionally, sweetcorn is distinguished from maize by its higher sugar content due to a recessive transmutation stalling the conversion of sugar to starch; thus, the sugar content in sweetcorn accounts for up to 20% of its dry matter, while there is only about 3% sugar in grain maize when harvested at immature, 'green millies' stage (Pajic, 2007).

As a versatile vegetable crop, sweetcorn is known to contain provitamin A such as β-carotene, as well as carotenoids, such as lutein, xanthine and cryptoxanthin pigments, (USAD National nutrient data base), Kumar and Jhariya, (2013) reported the presence of B-vitamins such as thiamine, niacin, pantothenic acid, folates, riboflavin, as well as pyridoxine, contained in sweetcorn.

The German philosopher Ludwig Feuerbach once articulated: *"We are what we eat"* (Feuerbach, 1963) making consumers conscious of what is contained in the food eaten on a daily basis. Moreover, from previously conducted studies, maize grain comprises of about 72% starch, 4% lipids, and 10% protein (Inglett, 1970), providing energy of 1527 KJ/100 g (USDA Natl. Nutrient Database). The main maize carbohydrate and kernel component, is starch, amounting up to 72% of kernel dry mass weight (DM); since sugars vary from 1% to 3% with sucrose being the key element and maltose, glucose, fructose, and raffinose in minor quantities (Mertz 1970; Boyer and Shannon 1987).

It has been estimated that more than 900 million people count on maize as their staple food worldwide, more especially in Latin America, Africa and Asia (Shiferaw *et al.,* 2011). Unfortunately, white maize is the mostly preferred by vast populations even though they suffer from provitamin A deficiency (VAD) (Nuss and Tanumihardjo, 2010).

Elevated CO_2 has strong effects on C4 plant metabolism and, therefore, food production of such crops (Berry and Björkman, 1980; Heckathorn *et al.*, 1998, 2002). Such eCO_2 concentrations have a positive impact on strawberry fruit and the effect was likely to improve flavour compounds compared with fruit grown under normal aCO_2 levels (Balasooriya *et al.,* 2017). Elevated CO_2 was strongly on yellow maize could possibly be prolonged to even darker yellow colour, profoundly due to an intensification of carotenoid accumulation. In this study, the aim was to investigate the effect of eCO_2 on biochemical quality of sweetcorn cultivar

STAR7719. The objectives were to analyze phytochemical parameters of the cobs grown under a CO_2 and eCO_2 conditions with respect to: (1) fruit and leaf ascorbic acid, (2) fruit carotenoids, (3) fruit total soluble solids (TSS), and (4) fruit total proteins (amino acids).

5.2. Materials and Methods

5.2.1 Experimental set-up plant materials

The two glasshouses used were adjacent to each other. One glasshouse had a treatment and the other with a control sweetcorn cultivar STAR 7719 obtained locally from Starke Ayers and the sweetcorn plants had nine replicates. Sowing took place in winter season on the 1st of June 2021 in 30 cm plastic pots. Growth medium was taken from Ukulinga Research Farm, University of KwaZulu-Natal in Pietermaritzburg (SA), and the soil type was a heavy clay. Pots were arranged in randomized complete block design in three rows per cultivar. The plastic pots were placed on the floor in growing rooms. One growing room had 'Because Nature Mycelium CO_2 generator bags' (as described in a diurnal chapter two) intended to elevate the CO_2 concentration in the glasshouse (of approximately 500 ppm), meanwhile, the other chamber was kept at had an ambient CO_2 concentration (approximately 430 ppm). The sweetcorn cultivar STAR 7719, obtained locally from Starke Ayers®, was used in both glasshouses comprising nine sweetcorn plants. Sowing took place in the winter season on the 1st of June 2021 into 30 cm plastic pots, filled with heavy clay soil from Ukulinga Research Farm, University of KwaZulu-Natal, in Pietermaritzburg (SA). Pots were arranged in randomized complete block design in three rows per cultivar and placed on the floor in the growing rooms. Plants developed cobs that, once at the harvestable stage, were collected for analysis of fruit ascorbic acid, carotenoids, total protein (amino acids), and total soluble solids. Two rows of fresh sweetcorn kernels situated in the top, middle and bottom part of the cob were removed from five cobs for the determination of fruit characteristics mentioned above.

5.3 Laboratory analysis

All chemicals were purchased from Sigma Aldrich®

5.3.1 Determination of ascorbic acid concentration in sweetcorn kernels

The concentration of ascorbic acid was determined in fresh sweetcorn kernels (as described by Boonkasem *et al.*, 2015) with minor adjustments. The determination of ascorbic acid was analysed by sampling kernels from the top, middle and bottom section of the cob. Two rows from five cobs, were chopped into small pieces and weighed out to 2.0 g. The samples were homogenised using in mortar and pestle in 20 ml 3% metaphosphoric acid and transferred into centrifuge tubes, shaken for 30 min and centrifuged at 4000 rpm for 10 min. The supernatant of (1 mL) was transferred into glass tubes followed by addition of 3 mL 0.2 mM DCPIP (dichlorophenolindophenol). The supernatant was mixed for 15 s and immediately read spectrophotometrically (Shimadzu UV-1800 UV-Vis Spectrophotometer, Japan), at 515 nm.

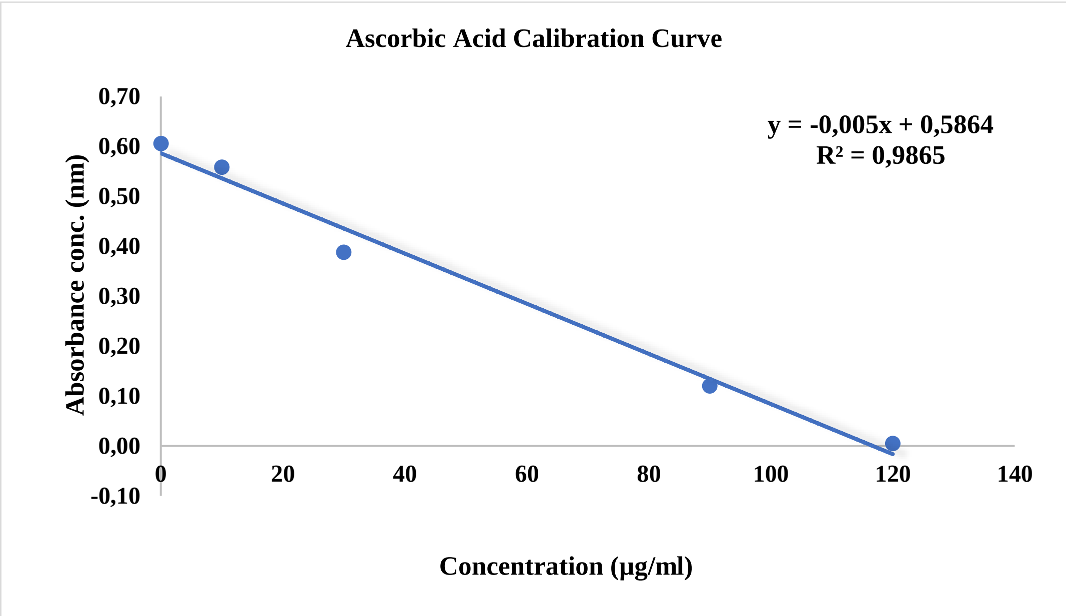


Fig. 5.1 Ascorbic acid calibration curve used for spectrophotometrical determination of ascorbic acid concentration. Ascorbic concentrations (µg/ml sample solution) were calculated using the equation y = 0.005x + 0.5864 with an R2 value of 0.9865, where ‘y’ was absorbance and ‘’x’ the concentration of the sample.

5.3.2 Determination of carotenoid and chlorophyll concentrations in sweetcorn kernels

Chlorophylls and carotenoids were determined in fresh sweetcorn kernels. Fresh kernel material was extracted using acetone 80% solvent. The concentrations of kernel carotenoids and chlorophylls were read spectrophotometrically (Shimadzu UV-1800 UV-Vis Spectrophotometer, Japan), and calculated using the method described by Lichtenthaler and Buschmann, (2001), an improved version of Lichtenthaler, (1987). Two rows of fresh kernels situated in the middle were removed from three cobs and 2.0 g were weighed out. The fresh samples were placed into centrifuge tubes and mixed with 4 ml acetone (80%). Tubes were left to stand on ice covered with aluminium foil for 10 min. Thereafter, the samples were homogenized with mortar and pestle into a very fine paste and transferred to centrifuge tubes. The paste remaining in the centrifuge tube was rinsed out with an additional 4 ml acetone and supernatant decanted into tubes, subsequently centrifuged for 5 min. Absorbance of supernatant samples were read at 663.2, 646.8, and 470 nm, using 80% acetone as a blank. Fruit carotenoid and chlorophyll concentrations were calculated using the equations as follows:

Table 5.1 Equations used to determine the concentrations (µg/ml) in fresh sweetcorn kernel tissue where A = sample absorbance chlorophyll a (Ch-a), chlorophyll b (Ch-b), chlorophyll a+b (ch-a+b) and carotenoids (C-x+c) using acetone 80% (Porra *et al.,* 1989; Lichtenthaler, 1987 and Lichtenthaler and Wellburn, 1983).

Solvent	Formulae (µg/mL)
80% acetone (v/v)	$C_a = 12.25\ A_{663.2} - 2.79\ A_{646.8}$ $C_b = 21.50\ A_{646.8} - 5.10\ A_{663.2}$ $C_{a+b} = 7.15\ A_{663.2} + 18.71\ A_{646.8}$ $C_{x+c} = (1000\ A_{470} - 1.82\ C_a - 85.02\ C_b) / 198$

5.3.3 Determination of total soluble solids (TSS) in sweetcorn kernels

The determination of TSS was achieved by weighing approximately 2 g of fresh kernels from the top, middle and bottom parts of fresh cob samples of the treatment and the control. The weighed fresh material was homogenised using mortar and pestle. Each sample was scooped with a spatula onto sterile gauze and juice squeezed on a refractometer (Bellingham +Stanley Digital Refractometer No. BU14006, Kent TN2 3EY, UK) to read TSS (in ^{o}Brix).

5.3.4 Determination of protein concentration in sweetcorn kernels

Protein was determined on five fresh sweetcorn cobs of the treatment and the control weighing between 99.3 to 158.7 g. Parts analysed were top, middle and bottom parts of the cobs. Five grams of fresh kernel were weighed out and cut into very small pieces, homogenised using mortar and pestle and transferred into the centrifuge tubes. Exactly 30 mL TRIS buffer (100 mM, pH of 7.5), was added to the samples which was centrifuged at 10000 rpm for 15 min at 2^{o}C. The supernatant was decanted and 100 µl subsamples were added to 5 ml Bradford reagent (Bradford, 1976). Samples were measured spectrophotometrically (Shimadzu UV-1800 UV-Vis Spectrophotometer, Japan) at absorbance of 590 nm. Protein concentrations were determined by comparison with a standard curve prepared with bovine serum albumin (BSA) (0.065, 2.125, 0.25, 0.5, 1, 2, 3, 4, 5, 6, 8, 10, 20, 40, 60, 80, 100, 150 and 200 mg) BSA per 100 ml deionised water.

A protein standard curve was plotted using bovine serum albumin (mg/ mL) against absorbance (Fig. 5.2). Protein concentrations (mg/ mL) were determined through Microsoft's Excel® software to give an equation of y = 0.2495x - 0.0122 with an R^2 of 0.9921. Protein concentration of 0.5 mg/mL was selected as standard for the determination of unknown sample concentration for protein in sweetcorn kernels.

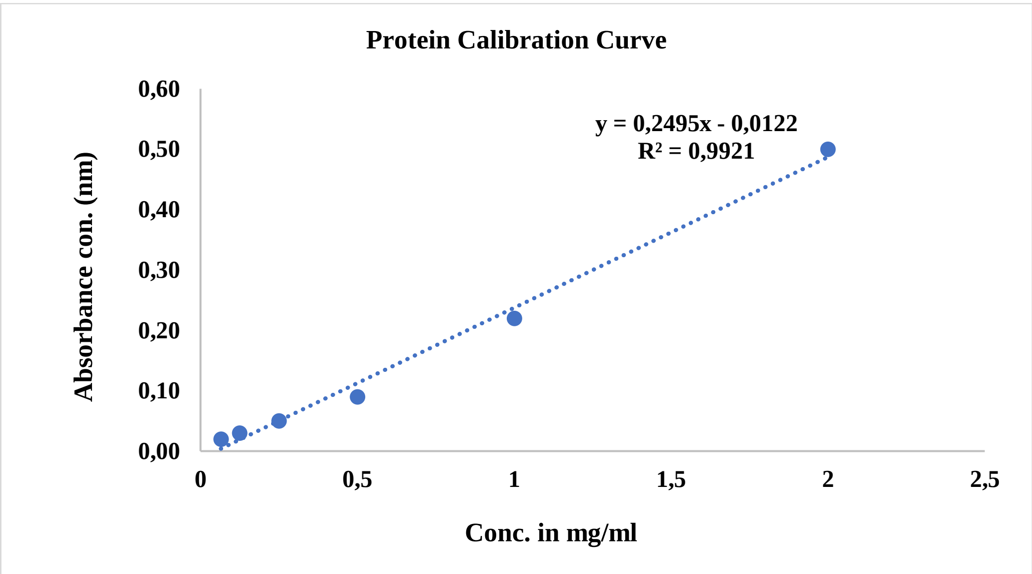


Fig. 5.2 Protein calibration curve using bovine serum albumin.

5.4 Results

Data were statistically analyzed using GenStat®18th edition (VSN International, Hemel Hempstead, UK) and analysis of variance (ANOVA) was performed. Fruit parameters analyzed were: ascorbic acid, total carotenoids, total protein and total soluble solids (TSS). The differences in the means for these phytochemicals were determined using Fisher's protected least significant difference test revealed significant difference at ($p<0.05$ and $p<0.01$).

Elevated CO_2 significantly affected fruit ascorbic acid with CO_2 concentration as well as TSS. Even though protein and carotenoids showed no significant differences, between control and treatment, both parameters in maize kernels of all three cob locations were enhanced under eCO_2. The P-values for ascorbic acid, carotenoids, protein and TSS indicate that ascorbic acid was highly significantly affected by locations ($P<0.001$; figure 4.3). Even though there was a tendency to an increase in carotenoids concentration given with the increase in CO_2 (figure 4.4), there were no significant differences ($P = 0.101$); additionally, protein concentration also tended to increase by the treatment (figure 4.5); however, differences were not significant ($P = 0.544$). A positive effect from eCO_2 on TSS (figure 4.6) yielded significant differences at ($P<0.01$).

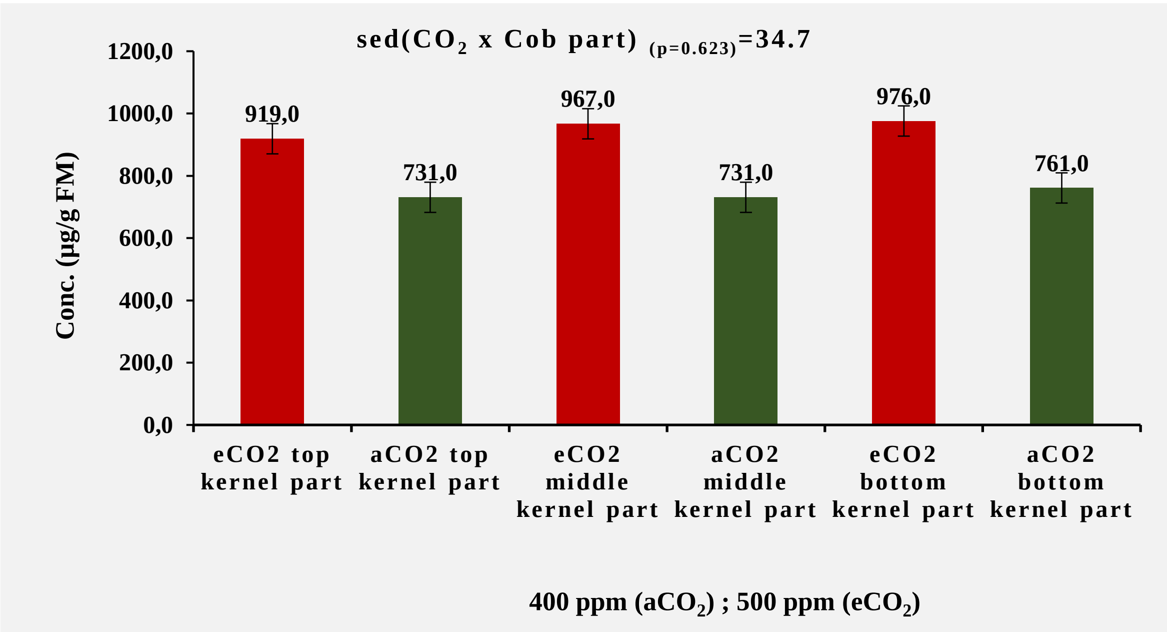


Fig. 5.3 Ascorbic acid concentration (µg/g FM) in maize kernels from three cob sections that were grown under ~400 ppm (aCO_2) and ~500 ppm (eCO_2).

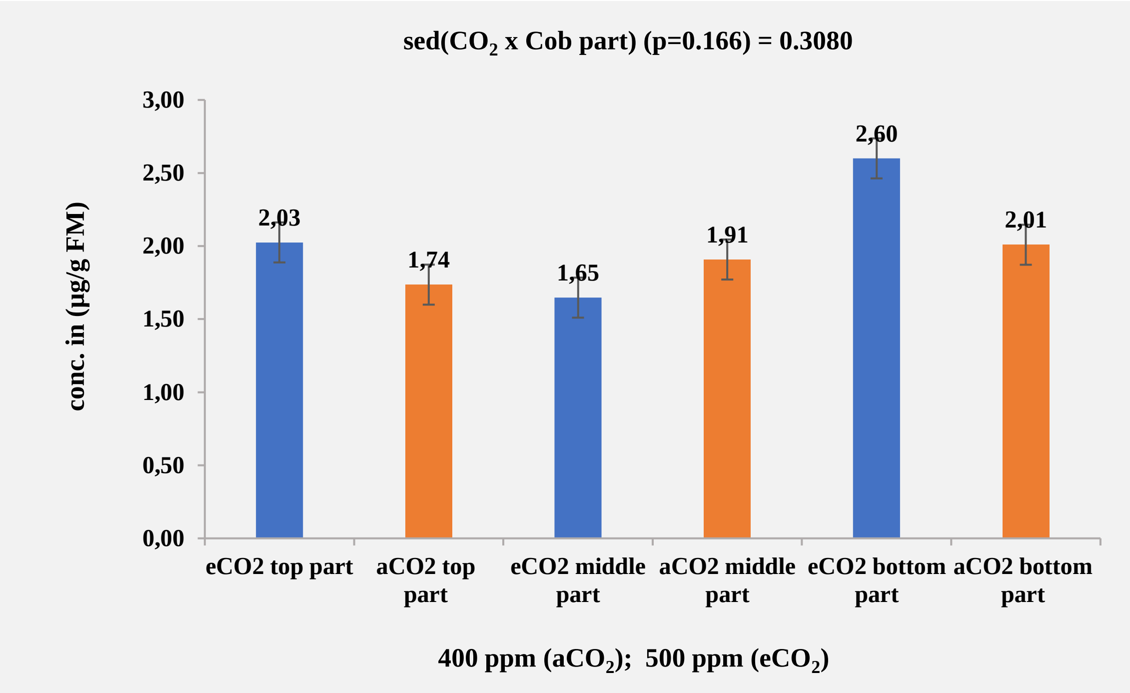


Fig. 5.4 Fruit pigments carotenoids and chlorophyll concentrations (mg/g FM) in three sections of fresh sweetcorn kernels grown under ~400 ppm (aCO2) and ~500 ppm (eCO2).

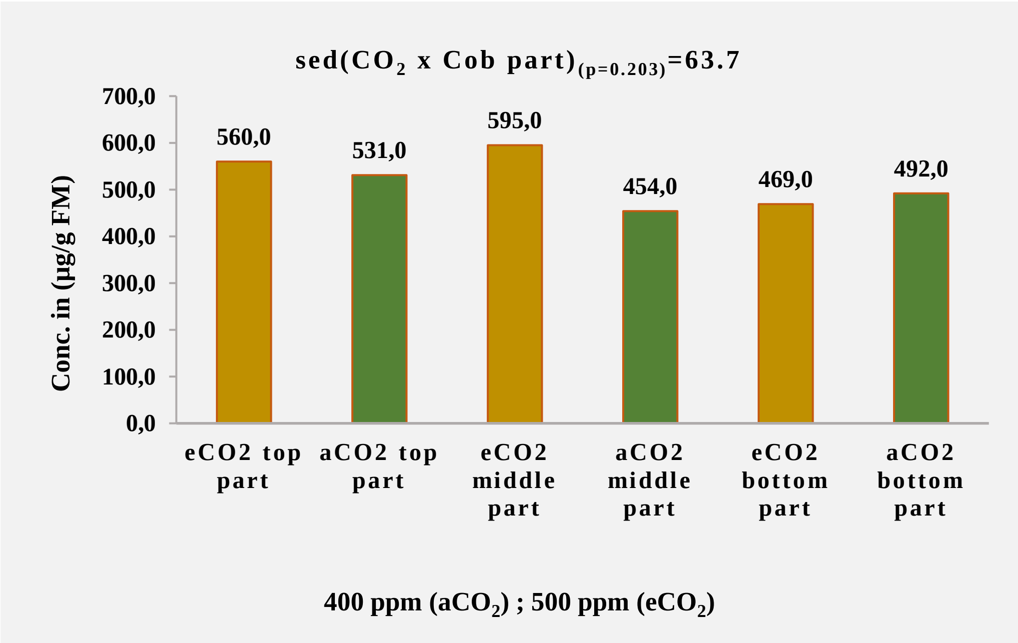


Fig. 5.5 Protein concentration (µg/g FM) in all of sweetcorn kernels in three sections grown under ~400 ppm (aCO_2) and ~500 ppm (eCO_2).

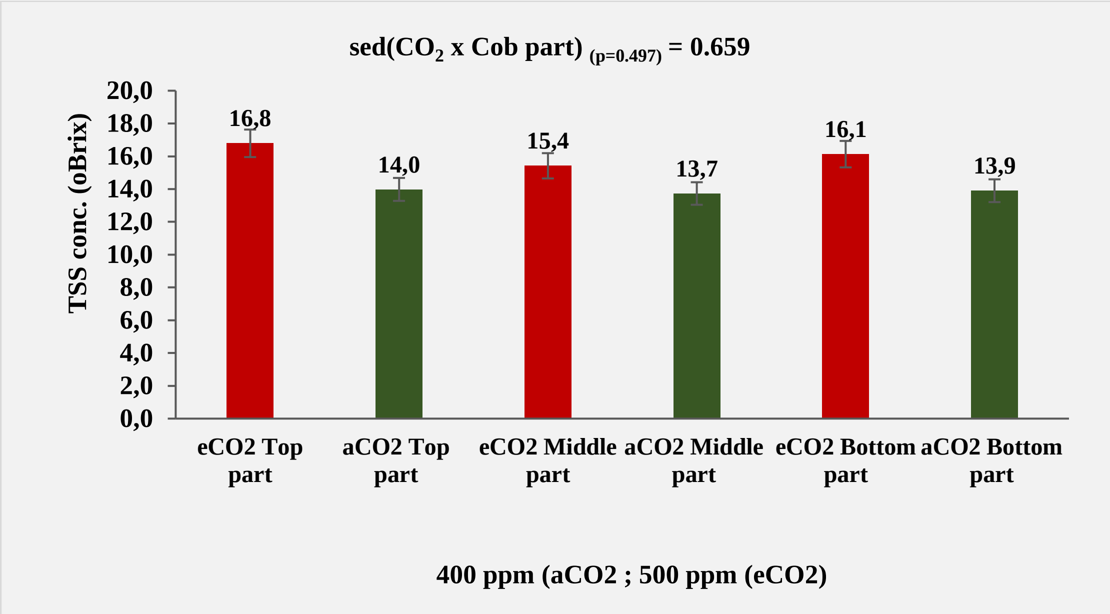


Fig. 5.6 TSS concentrations (oBrix) in three sections of fresh sweetcorn kernels grown under ~400 ppm (aCO_2) and ~500 ppm (eCO_2).

Table 5.2 Ascorbic acid, carotenoids, protein and TSS determined postharvest in sweetcorn kernels of aCO_2 and eCO_2.

			STAR7719				
Parameters	**aCO_2**	**eCO_2**	**Cob part**	**CO_2**	**Cob part x CO_2**	**Mean**	**LSD**
Ascorbic acid	741	954	0.234^{ns}	0.001^{***}	0.623^{ns}	847	72.5
Carotenoids	1.884	2.091	0.058^{ns}	0.259^{ns}	0.166^{ns}	1.987	0.643
Proteins	492	541	0.358^{ns}	0.196^{ns}	0.203^{ns}	517	132.8
TSS	13.87	16.11	0.244^{ns}	0.001^{***}	0.497^{ns}	14.99	1.376

*, **, *** indicate that the parameter is significant at $P < 0.05$, $P < 0.01$ or $P < 0.001$, respectively, where ns= non-significant. Cob part refers to the location of kernels on the maize cob. LSD values indicate significant differences between treatment and control under concentrations ~430 ppm (aCO_2) and ~500 ppm (eCO_2).

5.5 Discussion

Phytochemicals present in sweetcorn kernels were positively, as well as negatively, affected by the eCO_2 treatment. While ascorbic acid concentrations were enhanced under eCO_2. It could be possible, that a slight difference in distribution of ascorbic acid in the cob occurred due to eCO_2 and the effect was only visible in kernels localised at the bottom part of the cobs. Contrary to these results, eCO_2 was found to have no significant effect on titratable acidity in root, stem, leafy vegetables and fruit (Dong *et al.*, 2018). According to these authors, eCO_2 might have encouraged the transformation of fixed CO_2 to soluble sugars, resulting in a larger amount of sugars relative to organic acids consequently, fosters an enhanced sugar: acid ratio and, thus, vegetable of stronger taste (Wang and Bunce, 2004).

Total soluble solids (TSS) were positively enhanced by eCO_2 with a mean of 16.8 over 14.0 ^{o}Brix especially in kernels located at the top and bottom of cobs with a mean of 16.1 over 13.9 ^{o}Brix (figure 5.6) on the contrary, kernels located in the middle cob part had a slightly reduced TSS concentration. The results are in agreement with reports of meta-analysis by Dong *et al.* (2018), that total soluble sugars increase, specifically in leafy vegetables (consisting of organ of intense carbohydrate synthesis) increase under eCO_2 conditions by 36.2%. Furthermore, Jin *et al.,* (2009) postulated that the increase attributed to eCO_2 (1000-1500 ppm) was equivalent to an increase by 38-188% and the effect was particularly strong in Chinese cabbage leaves and 16-53% in oily sowthistle (*Sonchus oleraceus* L.) leaves. Similarly, Wang and Bunce (2004) reported that under eCO_2 (950 ppm) total soluble sugars in strawberry fruit were enhanced by 20% over the 350-ppm concentration under aCO_2. Similar results were reported for radish (*Raphanus sativus* L. *cv.* Mino) with a TSS increased by 13% and a 20% TSS increase in turnip (*Brassica rapa* L. *cv.* Grabe) under eCO_2 (1000 ppm) relative to aCO_2 (400 ppm) (Azam *et al.,* 2013).

Carotenoids only tended to be increased under eCO_2 (>1000 ppm) results contradicting the findings by Loladze *et al.* (2019) in a meta-analysis in which a significant decline in carotenoids concentrations in 44 plant cultivars mean response of -15% (95% CI, -26% to -6%), were reported under eCO_2 conditions due to the (dilution effect as carbohydrates are assimilated) or possibly (lower carotenoid synthesis). Loladze *et al.* (2019) further reported that plants and humans share a necessity for xanthophylls in their metabolism to shield against light-induced oxidative stresses; therefore, concentration of xanthophylls and other carotenoids decline under eCO_2 conditions.

Interestingly, protein concentrations decreased (figure 5.5) in all three cob locations; there was a tendency to increased protein under eCO_2 conditions (500 ppm). Findings from various authors (Loladze, 2014; Zhu *et al.*, 2018; Ujiie *et al.*, 2019) are in agreement with the current study demonstrating that, even though, elevated CO_2 concentrations enhance plant growth and yield, the protein content of the commodity tends decline due to enhanced allocation of sugars in expense of proteins, to these organs. This is commonly referred to as the 'carbon dilution effect' (Loladze, 2014; Zhu *et al.*, 2018; Ujiie *et al.*, 2019). Moreover, nitrogen is immobilization in vegetative tissues (Luo *et al.*, 2004), soil as well as decreased nitrate assimilation under eCO_2 (Bloom *et al.*, 2010).

On the other hand, in rice, a C3 plant, protein and nitrogen concentrations declined under eCO_2 not entirely due to the carbon dilution effect but was also due to differences in responses to the early emerging grains that were derived from early flowering and also the emerging of grains that was derived from late flowering under eCO_2 concentration (Zhang *et al.*, 2013). An eCO_2 concentrations is presumably not so likely to impact C4 crops since carbon uptake is reduced in C4 species under eCO_2 concentrations, furthermore, it becomes saturated under aCO_2 concentrations (Von Caemmerer and Furbank, 2003).

C4 crops hold great potential to provide nutrients for the health of humanity (Jobe *et al.* 2020); nonetheless, more effort is still required to comprehend nutrient flux control as well as maintaining homeostasis in C4 species, to confirm that C4 plants respond positively to eCO_2 in the coming decades. Medek *et al.* (2017) reported that C3 plants, such as wheat, rice and barley are likely to show a decline in grain protein between 7.8 in wheat, 7.6 in rice, and 14.1% in barley, when grown under eCO_2 (500-700 ppm) conditions by the middle of the century. Potato and vegetables are likely to contain 6.4% and 17.3% less protein at such high atmospheric CO_2 concentrations. Fruit crops will be most affected with an anticipated protein decline of 23% under these conditions. In legume species a protein decline of 3.5% is expected under the higher CO_2 conditions, with no significant effect on oily crops, while the same negative effect from eCO_2 was found in C4 plants (Medek *et al.* 2017).

Thus far, there is no literature clarifying whether the protein function is likely due to protein functioning as enzymic or as storage protein possibly causing decline in protein concentration directly linked to exposure to eCO_2 conditions. Furthermore, these seem little reporting in the scientific literature elaborating on the nutrient and phytochemical distribution of different effects on various kernels location in maize cobs under eCO_2 conditions. This might be very important for breeders and consumers alike, as difference cob parts are likely to diverge more and more in taste.

5.6 Conclusions

The C4 contribution of the C4 plants maize towards human nutrition is of vital importance, as it is a major crop, contributing to food security. The benefit that seems to arise in phytochemical concentrations of sweetcorn when the crop is grown under eCO_2 might even be enhanced when eCO_2 concentration will exceed 500 ppm. As an important environmental factor responsible for photosynthesis, the present study unfolded the ability of eCO_2 (at 500 ppm) to improve the accumulation of certain phytochemicals, such as total soluble solids (sugars and acids, including ascorbic acid), while negative effect was found on carotenoids and total protein registering a decline. Since there is limited published data regarding the eCO_2 effect on lowering carotenoid and protein concentrations in certain crops, it should be investigated, escalation of atmospheric CO_2 levels beyond 500 ppm positively or negatively affects the phytochemical composition of sweetcorn, consequently negatively affecting human and animal nutrition on a global scale.

Literature cited

Adom K. K. and Liu R. H. (2002). Antioxidant activity of grains, Journal of Agriculture Food Chemistry Vol. 50: 6182–6187.

Albuquerque C. J. B.; von-Pinho R. G. and Silva R. Performance of experimental and commercial maize hybrids for green maize production. Ciênc. Agrotechnology (2008). Vol. 32(2): 69-76.

Araujo R. C.; Pires A. V.; Susin I.; Mendes C. Q.; Rodrigues; G. H.; Packer I. U. and Eastridge M. L. (2008). Milk yield, milk composition, eating behaviour, and lamb performance of ewes fed diet containing soybean hulls replacing coast cross (*Cynodon sp.*) hay. Journal of Animal Science. Vol. 86(12): 3511-3521.

Azam A.; Khan I.; Mahmood A. and Hameed A. (2013). Yield, chemical composition and nutritional quality responses of carrot, radish and turnip to elevated atmospheric carbon dioxide. Journal of the Science of Food and Agriculture. Vol. 93: 3237–3244.

Badu-Apraku B. and Akinwale R. (2011). Identification of early-maturing maize inbred lines based on multiple traits under drought and low N environments for hybrid development and population improvement. Canadian Journal of Plant Science. Vol. 91: 931–942.

Balasooriya H. N.; Dassanayake; K. B.; Tomkins B.; Seneweera S. and Ajlouni S. (2017). Impacts of Elevated Carbon Dioxide and Temperature on Physicochemical and Nutrient Properties in Strawberries. Journal of Horticultural Science and Research ISSN: 2578-6598. Volume (1) 1.

Berry J. A. and Björkman O. (1980). Photosynthetic response and adaptation to temperature in higher plants. Annual Reviews in Plant Physiology Vol. 31: 491–543.

Biology of *Zea mays* (maize) (2011). https://geacindia.gov.in/resource-documents/biosafety-regulations/resource-documents/Biology_of_Maize.pdf.

Bloom A. J.; Burger M.; Asensio J. S. R. and Cousins A. B. 2010. Carbon dioxide enrichment inhibits nitrate assimilation in wheat and Arabidopsis. Science, Vol. 328: 899–903.

Boyer C. D. and Shannon J. C. (1987). Carbohydrates of the kernel. In: Watson S. A. and Ramstad P. E. Editors. Maize: chemistry and technology. St Paul, Minn.: American Association of Cereal Chemists. (pp 253–72).

Bradford M. M. (1976). A rapid and sensitive for the quantification of microgram quantities of protein utilizing the principle of protein-dye binding. Analytical Biochemistry Vol. 72: 248-254.

Brown R. A. and Rosenberg N. J. (1999). Climate change impacts on the potential productivity of maize and winter wheat in their primary United States growing regions. Climate Change. Vol. 41: 73–107.

Cherno M. (1963). Feuerbach's "Man is what he eats": a rectification. Journal of History of Ideas. Vol: 24(3), pp 397–406. doi: 10.2307/2708215.

Dhami N.; Tissue D. T. and Cazzonelli C. I. (2018). Leaf dependent response of carotenoid accumulation to elevated CO_2 in Arabidopsis. Archives of Biochemistry and Biophysics. 2018 Jun 1;647:67-75. doi: 10.1016/j.abb.2018.03.034. Epub 2018 Mar 28.

DeLucia E. H.; Nabity P. D.; Zavala J. A. and Berenbaum M. R. Climate change: resetting plant-insect interactions. Plant Physiology 2012. Vol. 160: 1677-1685.

Dong J.; Gruda N.; Lam S. K.; Li X. and Duan Z. 2018. Effects of elevated CO_2 on nutritional quality of vegetables -A Review. Frontiers in Plant Science. Vol. 9.

Florian A.; Timm S.; Nikoloski Z.; Tohge T.; Bauwe H.; Araújo W. L. and Fernie A. R. (2014). Analysis of metabolic alterations in Arabidopsis following changes in the carbon dioxide and oxygen partial pressures, Journal of Integrative Plant Biology. Vol. 56(9): 941-959.

Fritsche-Neto R. and Silva P. S. L. (2011). Selection index for maize cultivars with dual aptitude: Maize and green maize. Bragantia 2011. Vol. 70(4): 781-787.

Forster P.; Ramaswamy V.; Artaxo P.; Berntsen T.; Betts R. and Fahey D. (2007). Changes in atmospheric constituents and in radiative forcing. In: Climate change 2007: The Physical Science Basis. Contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change, Cambridge University Press, and (pp 129–234).

Gao F.; Zhu S. R.; Sun Y. C.; Du L.; Parajulee M.; Kang L. and Ge F. (2008). Interactive effects of elevated CO_2 and cotton cultivar on tri-trophic interaction of *Gossypium hirsutum*, *Aphis gossyppii*, and *Propylaea japonica*. Environmental Entomology. Vol. 37: 29–37.

Hartley S. E.; Jones C. G.; Couper G. C. and Jones T. H. (2000). Biosynthesis of plant phenolic compounds is elevated atmospheric CO_2. Global Change Biology. Vol. 6: 497–506.

Heckathorn S. A.; Downs C. A.; Sharkey T. D. and Coleman J. S. (1998). The small, methionine rich chloroplast heat-shock protein protects photosystem II electron transport during heat stress. Plant Physiology Vol. 116: 439–444.

Heckathorn S. A.; Ryan S. L.; Baylis J. A.; Wang J. A.; Hamilton E. W. and Cundiff L. (2002). In vivo evidence from an Agrostis stolonifera selection genotype that chloroplast small heat-shock proteins can protect photosystem II during heat stress. Functional Plant Biology Vol. 29: 933–944.

Hu Q. P. and Xu J. G. (2011). Profiles of carotenoids, anthocyanins, phenolics, and antioxidant activity of selected colour waxy maize grains during maturation. Journal of Agricultural and Food Chemistry. Vol. 59(5): 2026–2033.

Inglett G. E. (1970). Maize: Culture, Processing, Products. Major Feed and Crops in Agriculture and Food Science Series. 369 Seiten, 73 Abb., 65 Tab. The AVI Publishing, Inc., Westport, Connecticut.

Jin C.; Du S.; Wang Y.; Condon J.; Lin X. and Zhang Y. 2009. Carbon dioxide enrichment by composting in greenhouses and its effect on vegetable production. Journal of Plant Nutrition and Soil Science. Vol. 172: 418–424.

Jobe T. O.; Zenzen I.; Rahimzadeh K. P. and Kopriva S. (2019). Integration of sulfate assimilation with carbon and nitrogen metabolism in transition from C3 to C4 photosynthesis. Journal of Experimental Botany. Vol. 70: 4211–4221.

Kumar D. and Jhariya A. N. (2013). Nutritional, medicinal, and economical importance of maize: A mini review. Research Journal of Pharmaceutical Sciences ISSN. 2319: 555x.

Liu R. H. (2004). Potential synergy of phytochemicals in cancer prevention: mechanism of action, Journal of Nutrition. Vol. 134: 3479–3485.

Loladze I. 2014. Hidden shift of the ionome of plants exposed to elevated CO_2 depletes minerals at the base of human nutrition. eLife, 3, e02245.

Loladze I.; Nolan J. M.; Ziska L. h. and Knobbe A. R., (2019). Rising atmospheric CO_2 lowers concentrations of plant carotenoids essential to human health: A Meta-Analysis. Molecular Nutrition Food Research.

Luo Y.; Su B.; Currie W. S.; Dukes J. S.; Finzi A.; and Hartwig U. *et al.* (2004). Progressive Nitrogen Limitation of Ecosystem Responses to Rising Atmospheric Carbon Dioxide. BioScience Vol. 54: 731–739.

Mertz E. T. (1970). Nutritive value of maize and its products. In: Inglett GE, editor. Maize: culture, processing, products. Westport, Conn.: Avi Publishing Company, Inc. (pp 350–9).

Moore B. D.; Cheng S. H.; Rice J. and Seemann J. R. (1998). Sucrose cycling, Rubisco expression, and prediction of photosynthetic acclimation to elevated atmospheric CO_2, Plant Cell Environ. Vol. 21(9): 905-915.

Noguchi K. and Watanabe C. K. and Terashima I. (2015). Effects of Elevated Atmospheric CO_2 on Primary Metabolite Levels in Arabidopsis thaliana Col-0 Leaves: An Examination of Metabolome Data, Plant Cell Physiology. Vol. 56(11): 2069-78.

Nuss E. T. and Tanumihardjo S. A. Maize: A Paramount Staple Crop the Context of Global Nutrition. Vol. 9, (2010). Comprehensive Reviews in Food Science and Food Safety. Institute of Food Technologists®.

OECD (2006), "Section 3 - Maize (ZEA MAYS SUBSP. MAYS)", in Safety Assessment of Transgenic Organisms, Volume 1: OECD Consensus Documents, OECD Publishing, Paris.

Pajic Z. (2007). Breeding of maize types with specific traits at the maize research institute Zemune polje. Genetika. Vol. 39: 169-180.

Parry M. L.; Rosenzweig C.; Iglesias A.; Livermore M. and Fischer G. (2004). Effects of climate change on global food production under SRES emissions and socio-economic scenarios. Global Environmental Change. Vol. 14: 53–67.

Phillips D. L.; Lee J. J. and Dodson R. F. (1996). Sensitivity of the US maize belt to climate change and elevated CO_2. 1. Maize and soybean yields. Agriculture Systems. Vol. 52: 481–502.

Shiferaw B.; Prasanna B.; Hellin J. and Banziger M. (2011). Crops that feed the world 6. Past successes and future challenges to the role played by maize in global food security. Food Security. Vol. 3: 307–27.

Smith C.W.; Betrán J.; Runge E. C. A. (2004). Maize: Origin, History, Technology, and Production, John Wiley, Hoboken N. J.; [Chichester], ISBN: 978-0-47-41184-0.

Thomson A. M.; Brown R. A.; Rosenberg N. J.; Izaurralde R. C. and Benson V. (2005). Climate change impacts for the conterminous USA: an integrated assessment. Part 3. Dryland production of grain and forage crops. Climate Change. Vol. 69: 43–65.

Ujiie K.; Ishimaru K.; Hirotsu N.; Nagasaka S.; Miyakoshi Y.; Ota M.; Tokida T.; Sakai H.; Usui Y.; Ono K.; Kobayashi K.; Nakano H.; Yoshinaga S.; Kashiwagi T.; Magoshi J. (2019). How elevated CO_2 affects our nutrition in rice, and how we can deal with it. PLoS One 14, e0212840.

[USDA]. United States Department of Agriculture. 2009. National nutrient database for standard reference. Available from: http://www.nal.usda.gov/fnic/foodcomp/search/. Accessed September and October 2009.

von Caemmerer S. and Furbank R. T. (2003). The C(4) pathway: an efficient CO (2) pump. Photosynthesis Research. Vol. 77: 191–207.

Wang S. Y. and Bunce J. A. 2004. Elevated carbon dioxide affects fruit flavour in field-grown strawberries (Fragaria×ananassa Duch). Journal of the Science of Food and Agriculture. Vol: 84, pp 1464–1468.

Zhang G.; Sakai H.; Tokida T.; Usui Y.; Zhu C. and Nakamura H. *et al.* (2013). The effects of free-air CO(2) enrichment (FACE) on carbon and nitrogen accumulation in grains of rice (Oryza sativa L.). Journal of Experimental Botany. Vol. 64: 3179–3188.

Zhang R.; Huang L.; Deng Y.; Chi J.; Zhang Y.; Wei Z. and Zhang M. (2016). Phenolic content and antioxidant activity of eight representative sweet maize varieties grown in South China. International Journal of Food Properties. Vol. 20(12): 3043–3055.

Zhu C.; Kobayashi K.; Loladze I.; Zhu J.; Jiang Q. and Xu X. *et al.* (2018). Carbon dioxide (CO_2) levels this century will alter the protein, micronutrients, and vitamin content of rice grains with potential health consequences for the poorest rice-dependent countries. Sci. Adv. 4, eaaq1012.

GENERAL DISCUSSION, CONCLUSION AND OUTLOOK

Introduction

Global climate change has resulted from the accumulation of greenhouse gases (GHGs) in the atmosphere, together with the subsequent rise in temperature resulting in changing rainfall patterns (Forster *et al.*, 2007; Vanaja *et al.*, 2015). The emissions of methane (CH_4) and nitrous oxide (N_2O) from the land on which crops are grown, are considered as threat to the ozone layer, such that reduction of these emissions specifically from agricultural activities is significantly important (Forster *et al.*, 2007). Amongst these emissions, carbon dioxide (CO_2) is considered the main culprit projected to have its concentrations reaching 550 ppm by 2050 (IPCC, 2007) which is likely to affect crop production and productivity of agricultural crops, and ultimately affect global food security (Vanaja *et al.*, 2015). Previous investigations studied the impact of climate change on C4 maize production and productivity by implementing the need for adaptation techniques to counter the harmful effect of climate change (Mulungu and Tembo, 2018). The rise in the production of *Zea mays* L. var. *saccharata* over the past 9 years between 15.2 to 27.8%, as well as consumption rate over the same number of years (FAOSTAT, 2018) revealed the significance of this horticultural comity, and therefore, the effect of elevated (eCO_2) warrants sweetcorn investigation.

The present study, therefore, was aimed at examining the effect of elevated CO_2 concentration on growth, development, and postharvest characteristics of sweetcorn. The use of environmental structures such as open top chambers (OTC), green houses, and open-air field in maintaining CO_2 in dry grain maize has been previously reported, nonetheless, documentations on the effect of eCO_2 on sweetcorn remains scarce.

The dissertation was divided into five sections. Each section is tackling effect of elevated CO_2 in particular quality parameters of *Zea mays* L. var. *saccharata*.

- **Section I:** Literature review on enhancement of the morphology and nutritional quality of vegetable crops through eCO_2 concentration
- **Section II:** Effect of artificially elevated CO_2 on maize plants growth and development under environmentally controlled conditions: Diurnal variations in CO_2 concentration ad soil moisture in pot-grow sweetcorn (*Zea mays* L. var. *saccharata*)

- **Section III:** Effects of artificially elevated CO_2 conditions on the morphology of sweetcorn (*Zea mays* L. var. *saccharata*)
- **Section IV:** Does elevate CO_2 affect the nutritional quality and leaf pigment parameters of sweetcorn (*Zea mays* L. var. *saccharata*): Mineral composition of fruit and leaf carotenoid and chlorophylls
- **Section V:** The effects of CO_2 on biochemical quality of sweetcorn (*Zea mays* L. var. *saccharata*): Phytochemical parameters

Discussion

The objective of the literature review (section 1) highlighted the enhancement of the morphology and nutritional quality of vegetable crops through eCO_2 levels, some of the previous investigations on the effect of eCO_2 were focused on maize as staple food grown in structures such as growth chambers, greenhouses, controlled environmental chambers, phytotron, OTC and free air carbon enrichment (FACE) facilities.

It has occurred (section II) that many studies have encountered failure in maintaining CO_2 concentration within enclosed structures due to technical limitations because the typical environment is not merely the same as the open field conditions in terms of the free air flow. The objectives of this present study were to determine if there were possible beneficial effect on size and the external appearance of sweetcorn to investigate the effect of eCO_2 sequestration to the soil on nutritional quality of sweetcorn. The current global rise in the atmospheric CO_2 causing climate change has generated uneven precipitations patterns resulting to a decline in crop production. Previously, research have been conducted on plant response to eCO_2 concentration, however, to our knowledge very little documentation on sweetcorn is known on the effect of eCO_2 under controlled environment. To mimic slightly optimised CO_2 conditions, the use of 'mycelium CO_2 generator bags' were installed above sweetcorn plants grown in pots in a temperature-controlled glasshouse, subsequently resulting in eCO_2 of ~500 ppm and ambient CO_2 (aCO_2) of ~430 ppm.

In section III, addressed the concentrations of CO_2 displayed different concentrations with distance from the 'mycelium CO_2 bags' (appendix 7) while soil moisture percentage (appendix 8 and 9) as well as the LSD (Fig. 2.1) did not differ significantly between treatments and controls. Due to cooler temperatures, growing sweetcorn in the winter season, resulted in sustained delays in days to emergence (DTE) (Fig. 3.1) for cultivar 'Assegai'.

The present study reports on 'Assegai' cultivar experiencing lodging which led to death of maize plants; however, leaf samples were collected for leaf pigment analysis before plant death. As a result, 'Assegai' data capture was discontinued because there was no control to be compared with on crop growth and development, while the effect of eCO_2 on 'STAR7719' treatment was positive. Further, significantly higher leaf number and plant height were recorded for 'STAR7719' at eCO_2 with mean 291 cm compared with 0.08 cm under aCO_2 and 1.18 cm compared with 1.23 cm under aCO_2 concentrations (Table 3.2). Elevated CO_2 also significantly enhanced fresh ear mass with means 210 g over 163 g under aCO_2 concentrations while the ear length recorded mean of 19.8 g under eCO_2 over mean of 13.2g under aCO_2 concentrations (Table 3.3) contributing to increased biomass. Total plant biomass as well as root mass were highly significantly enhanced under eCO_2 conditions.

Climate change has resulted to a rapid rise in various gases that affect crop production, the effects of CO_2 particularly on mineral absorption and leaf pigment concentrations of sweetcorn plant has not been well documented. Therefore, the aim of the recent study was to investigate the effect of eCO_2 on selected nutritional quality parameters of sweetcorn and the specific objective were to analyse the mineral composition of fruit and leaf pigments. While section IV discussed sweetcorn as an early maturing horticultural crop consisting of essential mineral nutrients and leaf pigments. Carotenoids and chlorophyll pigments were analysed from 12 green leaf samples collected from 'Assegai' and 'STAR7719' grown under eCO_2 (500 ppm) and ambient CO_2 (430 ppm) concentrations.

The nitrogen (N) concentration was not found significant under eCO_2, the mean values were lowered to 2.49% compared with 2.97% under aCO_2. Myers *et al.* (2014) is in agreement with the current findings on lower N%, the authors reported non-significant effect on N from maize plants subjected to eCO_2. Under eCO_2, minerals such as magnesium (Mg), potassium (K), and phosphorus (P) were found to be significantly lower compared to aCO_2 (Table 4.2). Brown (1978) found that in C4 grasses belonging to Poaceae species, total N fertilizer (N, % of DM) applied than dry matter that is produced in C3 grasses under eCO_2, led to a conclusion that N might be hugely utilised more efficiently in C4 grasses. While Dong *at el.* (2018) reported a significantly increased calcium (Ca) concentration of 8.2% in leafy vegetables under eCO_2, our results, however, are not in agreement with their findings in which Ca concentration under eCO_2 was significantly affected.

Additionally, the efficacy of eCO_2 on minerals had no significant effect on iron (Fe) concentration, nevertheless, a tendency to increased Fe concentration in all the kernels were achieved (Table 4.2). Furthermore, our findings agree with results by Dong *et al.* (2018) that minerals such as Fe, Mg, and Zn were significantly declined by 9.2%, 16.05%, and 9.4% of FM under eCO_2 concentrations, contrary to P, K, S, Cu, and Mn being maintained under eCO_2.

The same negative effect of eCO_2 was also observed in leaf pigments of maize cultivars 'Assegai' and 'STAR7719' with decreased carotenoids and chlorophyll concentrations (Table 4.3) it had been hypothesised that reduced leaf pigments might be due to possible changes in physiology because of a need for plant adaptability to eCO_2 concentrations (Houpis *et al.*, 1988). These current findings on declined leaf chlorophyll agree with findings by Delucia *et al.,* (1985); Houpis *et al.,* (1988); Wullschleger *et al.,* (1992) achieved a declined leaf chlorophyll in young leaves in response to eCO_2 conditions.

In section V the effect of eCO_2 on postharvest biochemical quality of sweetcorn was investigated because the consumers have now become fully aware of the nutritional quality of vegetables. Scarce research conducted on the effect of environmental conditions caused by higher atmospheric CO_2 conditions especially the quality parameters of vegetables. Excessive accumulation of CO_2 might result in negative effects on vegetables. The present study was aimed at investigating the effect of elevated CO_2 (eCO_2) on phytochemical quality parameters of sweetcorn. Maize plants established in two adjacent glasshouses were 'STAR7719', with eCO_2 of (500 ppm) maintained through using 'Mycelium CO_2 generator bags') and another with ambient CO_2 (aCO_2) of (430 ppm). Parameters such as ascorbic acid and total carotenoids, total protein and total soluble solids (TSS) were analysed from horticultural matured fresh cobs in both locations.

The concentration of ascorbic acid (fig 5.3) in the cob was significantly affected at ($P<0.001$) (Table 5.1) by eCO_2, it could be probably due to the effect visible only in kernels situated at the part of the cobs. On the contrary, Dong *et al.* (2018) reported no significant increase on titratable acidity in root, stem, leaves and fruit. According to Wang and Bunce (2004), eCO_2 might have resulted in transformation of fixed CO_2 to soluble sugars, leading to a larger amount of sugars accumulating relative to organic acids consequently, promotes an improved sugar: acid ratio thus resulting to much sweeter cobs.

Our study also showed that TSS was positively enhanced by eCO_2 in all the locations of the kernels (Table 5.1), while total carotenoids (fig. 5.4) declined significantly, and total protein (fig.5.5) was significantly declined under eCO_2 concentrations. Similarly, under eCO_2 TSS was significantly improved on strawberry (*genus Fragaria*) fruit (Wang and Bunce, 2004). Azam *et al.* (2013) further reported that eCO_2 enhanced the concentration of TSS in radish (*Raphanus sativus*) and turnip (*Brassica rapa* var. *rapa*). Even though total carotenoids and total protein were not significant, a tendency to increased concentrations were observed under eCO_2. Our current findings on protein decline agree with findings by Loladze (2014) and Zhu *et al.* (2018) who demonstrated that, even though eCO_2 increase plant growth and yield, the protein content of the commodity tends to reduce due to increased allocation of sugars in expense of proteins. This is commonly called 'carbon dilution effect'.

Conclusion

In conclusion, the research was intended to understand the effects of eCO_2 on sweetcorn cultivars and to evaluate the response of sweetcorn cultivars under eCO_2 conditions. This study was designed to enhance the nutritional value of sweetcorn and to promote food security, particularly, for rural households, where people still cultivate their own maize and consume as maize or as mealies. This study additionally provided a better understanding and is in agreement with previous investigations that had discovered positive response of C4 crops towards eCO_2 and subsequently, enhanced growth, development as well as yield. The current study was relevant with the intention to impact national and global agriculture. The findings from this study will assist with planning how commercial and subsistence farmers can produce sweetcorn in a sustainable and economic manner under eCO_2 conditions. This, in turn, will create more employment, since farmers will be looking into producing more sweetcorn a horticultural vegetable, as they obtain greater comprehension of sweetcorn response to eCO_2 conditions when they gain market share. From the experiment, the evidence pointed out that CO_2 enrichment had a positive impact on crop growth, development, and reproductive growth.

Outlook

According to a scientific viewpoint, the study revealed little knowledge on carbon sequestration in the soil by crops grown in South Africa and in Sub-Saharan Africa, therefore, there is a need to explore the effect of eCO_2 on maize specifically sweetcorn crop as one the mostly consumed horticultural commodity. This study will open opportunities for further research as well as inform policies for experienced and new researchers in different disciplines. Understanding the effect from the eCO_2, can be further extended to investigating the effect on growth hormones responsible for starch production possibly in concentrations higher than 700 ppm in horticultural crops. This multidisciplinary research will involve horticultural, agrometeorological, as well as plant breeding expertise so that strategies can be harnessed more efficiently by plant breeders in sequencing and identifying markers responsible for the for CO_2 sequestration and subsequently, producing hybrids with enhanced ability in the assimilation of CO_2. Additionally, researchers can further extend their assistance to farmers and rural households in optimising the growing of sweetcorn with increased kernel yield (edible part) and nutritional quality under eCO_2 conditions at a sustainable manner, while, creating a large germplasm pool of qualitative and quantitative genetic effects specifically for sweetcorn through breeding programs. This in turn will encourage production of hybrid seeds in the midst of climate change with enhanced ability to assimilate and sequestrate CO_2 in higher concentrations for enhanced nutritional benefits, while, maintaining food and nutritional security.

Literature cited

Delucia E. H.; Sasek T. W. and Strain B. R. (1985). Photosynthesis inhibition after long-term exposure to elevated levels of atmospheric carbon dioxide. Photosynth. Research. Vol. 7: 175-184.

Houpis J. L. J.; Surano K. A.; Cowles S. and Shinn J. H. (1988). Chlorophyll and carotenoid concentrations in two varieties of Pinus ponderosa seedlings subjected to long-term elevated carbon dioxide. Tree Physiology. Vol. 4: 187-193.

Dong J.; Gruda N.; Lam S. K.; Li X. and Duan Z. (2018). Effects of Elevated CO_2 on Nutritional Quality of Vegetables: A Review. Frontiers Plant Science, Vol. 9: 924.

FAOSTAT, (2018). Crop Production Statistics. http://faostat.fao.org, Accessed: July 2018.

Forster P.; Ramaswamy V.; Artaxo P.; Berntsen T.; Betts R. and Fahey D. (2007). Changes in atmospheric constituents and in radiative forcing. In: Climate change 2007: The Physical Science Basis. Contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change, Cambridge University Press, (pp 129–234).

IPCC. Climate Change: Synthesis Report. Fourth Assessment Report. Valencia: Intergovernmental Panel on Climate Change, IPCC; 2007.

Mulungu K. and Tembo G. (2018). Effects of weather variability on crop abandonment. Sustainability. Vol. 7: 2858-2870.

Myers S. S.; Zanobetti A.; Kloog I.; Huybers P.; Leakey A. D.; Bloom A. J.; Carlisle E.; Dietterich L. H.; Fitzgerald G.; Hasegawa T.; Holbrook N. M.; Nelson R. L.; Ottman M. J.; Raboy V.; Sakai H.; Sartor K. A.; Schwartz Seneweera S.; Tausz M. and Usui Y. (2014). Increasing CO_2 threatens human nutrition, Nature Vol.510(7503): 139-42.

Loladze I. (2014). Hidden shift of the ionome of plants exposed to elevated CO_2 depletes minerals at the base of human nutrition. Ecology/Epidemiology and global health. *eLife* 3:e02245. Research Article.

Vanaja M.; Maheswari M.; Jyothi Lakshmi N.; Sathish P.; Yadav S. K.; Salini K.; Vagheera P.; Vijay Kumar G. and Abdul Razak. (2015). Variability in Growth and Yield Response of Maize Genotypes at Elevated CO_2 Concentration. Advances in Plants and Agriculture Research. Vol. 2.

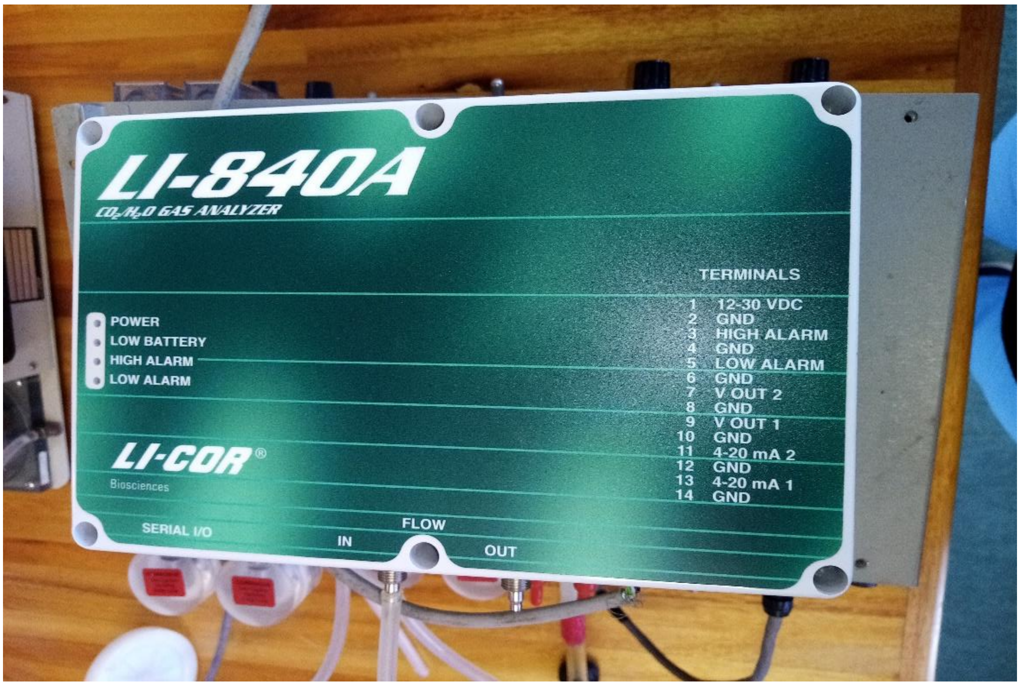
Wang S. Y. and Bunce J. A. (2004). Elevated carbon dioxide affects fruit flavour in field-grown strawberries (Fragaria×ananassa Duch). Journal of the Science of Food and Agriculture. Vol.84: 1464–1468.

Wullschleger S. D.; Norby R. J. and Hendrix D. L. (1992). Carbon exchange rates, chlorophyll content, and carbohydrate status of two forest tree species exposed to carbon dioxide enrichment. Tree Physiology. Vol.10: 21-31.

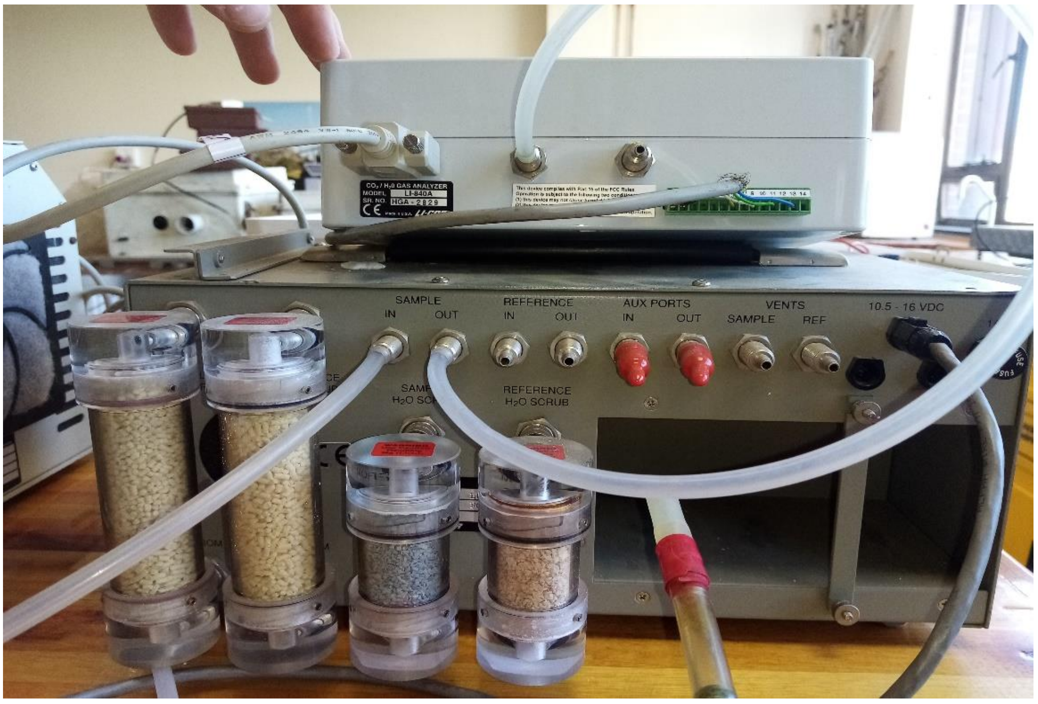
Zhu C.; Kobayashi K.; Loladze I.; Zhu J.; Jiang Q. and Xu X. *et al.* (2018). Carbon dioxide (CO_2) levels this century will alter the protein, micronutrients, and vitamin content of rice grains with potential health consequences for the poorest rice-dependent countries. Sci. Adv. 4, eaaq1012.

Appendix

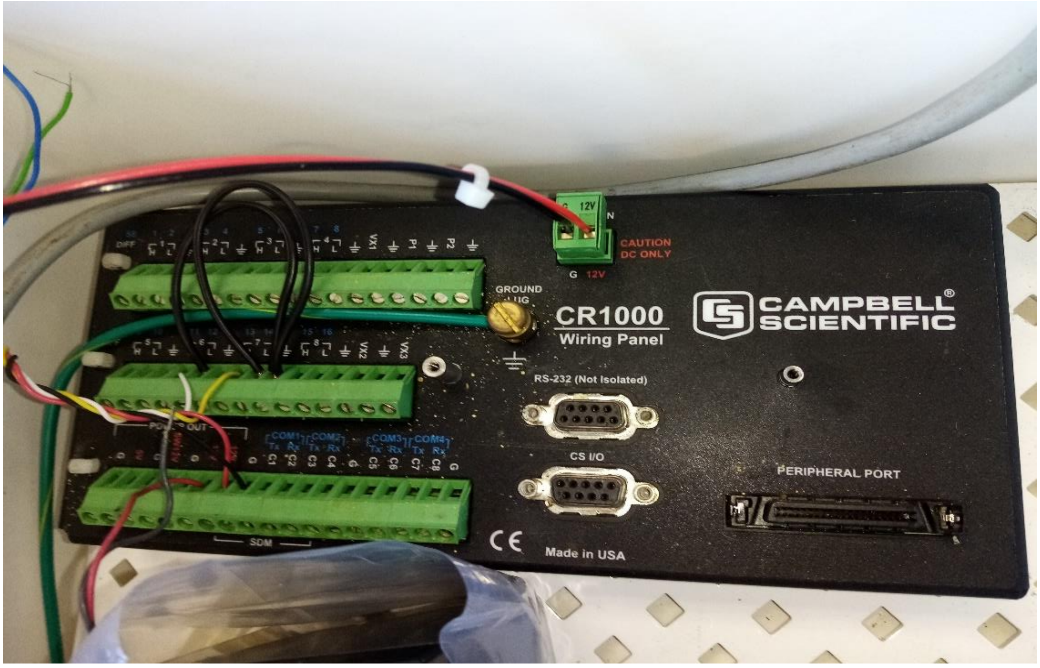
Appendix 1: LI-84OA, CO_2/H_2O gas analyser, USA



Appendix 2: LI-670, Control Air Flow, USA



Appendix 3: CR 1000-wiring panel, Data logger, Campbell Science, USA



Appendix 4: LI-610, CO_2/H_2O Portable Dew Point Generator, USA

Appendix 5: SCE210C093, Solenoid valve, Diya Valves International, RSA

Appendix 6: EC150, CO_2/H_2O Open-path gas analyser, Campbell Scientifics, USA

Appendix 7: Because Nature Mycelium CO_2 Generator, Windell Hydroponics.co.za, RSA

Appendix 8: Soil moisture percentage (%) in 10 L pots on a four-week interval. 'Assegai' and 'STAR7719' sweetcorn were grown in glasshouse under ambient (a) CO_2 (430 ppm) and elevated (e) CO_2 (500 ppm).

Reps	Cultivar	CO_2	Week 1	Week 2	Week 3	Week 4
1	Assegai	eCO_2	28,4	19,9	18,1	25,4
2	Assegai	eCO_2	29,7	16,7	26	25,5
3	Assegai	eCO_2	30	19,9	28,5	25,5
4	Assegai	eCO_2	22,3	11,9	22,3	30,1
5	Assegai	eCO_2	29,2	20,9	30,1	27,8
6	Assegai	eCO_2	29,5	18,3	26,8	19,7
7	Assegai	eCO_2	26,5	16,4	23,8	27.4
8	Assegai	eCO_2	23,9	17,8	27,6	23,7
9	Assegai	eCO_2	22,7	15,5	22,4	24,6
1	Assegai	aCO_2	27,1	16,5	30,0	24,1
2	Assegai	aCO_2	32,3	15,7	27,9	20,1
3	Assegai	aCO_2	28,4	21,8	20,4	27,9
4	Assegai	aCO_2	30,7	19,7	29,7	25,4
5	Assegai	aCO_2	26,9	16,9	29,3	27,4
6	Assegai	aCO_2	24,6	14,9	35,6	25,3
7	Assegai	aCO_2	23,0	18,6	30,1	22,5
8	Assegai	aCO_2	32,1	15,9	30,0	20,6
9	Assegai	aCO_2	26,4	13,8	28	18,2
1	STAR7719	eCO_2	19,4	18,8	27,5	24,1
2	STAR7719	eCO_2	25,7	21,5	26,8	20,1
3	STAR7719	eCO_2	25,4	16,2	24,1	19,8
4	STAR7719	eCO_2	20,6	18	23,4	25,4
5	STAR7719	eCO_2	26,1	19,9	23,9	27,4
6	STAR7719	eCO_2	26,9	22,2	30,5	26,5
7	STAR7719	eCO_2	23,6	13,9	18,1	22,5
8	STAR7719	eCO_2	23,2	18,5	25,4	20,6
9	STAR7719	eCO_2	28,9	17,6	28,9	20,1
1	STAR7719	aCO_2	28,1	18,9	32,6	19,7
2	STAR7719	aCO_2	29,7	21,3	34	20,3
3	STAR7719	aCO_2	31,2	20,8	35,8	24,6
4	STAR7719	aCO_2	21,1	15	29,5	22,7
5	STAR7719	aCO_2	29,5	19,2	33,5	19,6
6	STAR7719	aCO_2	27,1	16,1	34,5	24,3
7	STAR7719	aCO_2	26,2	14,2	29,4	19
8	STAR7719	aCO_2	31,8	19,3	34,8	20,8
9	STAR7719	aCO_2	24	19,4	29,8	22,5

Appendix 9: Soil moisture percentage (%) in 10 L pots. There were 9 replications for Assegai and STAR7719 in glasshouses 8 and 9, under eCO_2 (500 ppm) and aCO_2 (430 ppm).

Reps	Cultivar	CO_2	Week 5	Week 6	Week 7	Week 8
1	Assegai	eCO_2	19,9	0	0	0
2	Assegai	eCO_2	17,5	19,3	14,8	13,7
3	Assegai	eCO_2	20,8	18,6	16,9	12,7
4	Assegai	eCO_2	23,5	33	20,1	15,1
5	Assegai	eCO_2	16,3	16,2	23,3	10,2
6	Assegai	eCO_2	17,4	15,9	16,6	16,4
7	Assegai	eCO_2	19,9	30,8	34,3	13,2
8	Assegai	eCO_2	18,9	27,6	28,8	15,2
9	Assegai	eCO_2	18,.4	17,6	23,6	14,8
1	Assegai	aCO_2	16,1	26,3	16,1	19,0
2	Assegai	aCO_2	19,1	28,7	14,6	17,8
3	Assegai	aCO_2	18,3	23,7	16,7	0
4	Assegai	aCO_2	11,5	31,8	13,8	23,4
5	Assegai	aCO_2	19,4	29,4	19,2	19,8
6	Assegai	aCO_2	18,5	33,8	21	18,7
7	Assegai	aCO_2	19,6	25,6	25,9	16,8
8	Assegai	aCO_2	18,1	29,3	23,8	24,5
9	Assegai	aCO_2	19,1	26,1	21,5	20,3
1	STAR7719	eCO_2	14,3	15,6	18,9	10,9
2	STAR7719	eCO_2	16,4	14,3	17,7	10,8
3	STAR7719	eCO_2	19,8	11,6	6,6	12,4
4	STAR7719	eCO_2	25,3	16,7	12,1	13,2
5	STAR7719	eCO_2	22,1	15,1	14,2	7,7
6	STAR7719	eCO_2	20	19,9	11,2	8,9
7	STAR7719	eCO_2	17,7	16,8	10,6	13,4
8	STAR7719	eCO_2	18,8	17,1	11,1	12,8
9	STAR7719	eCO_2	19,7	13,2	11,8	14,4
1	STAR7719	aCO_2	22,7	26,9	21,8	18,99
2	STAR7719	aCO_2	22,3	32,6	27,8	17,7
3	STAR7719	aCO_2	30,7	31,3	31,3	19,9
4	STAR7719	aCO_2	19,8	28,4	25,8	20,6
5	STAR7719	aCO_2	20,1	24,4	24,6	23,3
6	STAR7719	aCO_2	24,4	30,5	32,8	18,8
7	STAR7719	aCO_2	18,8	23,1	26,8	19,8
8	STAR7719	aCO_2	19,5	26,8	27,8	17,1
9	STAR7719	aCO_2	14,2	19,4	28,8	16,9

Appendix 10: Analysis of variance for soil moisture percentage levels for sweetcorn grown under aCO_2 (430 ppm) and eCO_2 (500 ppm) for the eight consecutive weeks in two glasshouses.

Source	DF	SS	MS	Vr	F-value
Reps stratum	8	251.52	31.44	1.43	
Reps.*Units*stratum					
CO_2	1	971.63	971.63	44.18	<0.01
Cultivar	1	0.20	0.20	0.01	0.924
Week	7	2569.98	367.14	16.70	<0.001
CO_2. Cultivar	1	263.33	263.33	11.97	<0.001
CO_2.Week	7	1291.60	184.51	8.39	<0.001
Cultivar. Week	7	536.91	76.70	3.49	0.001
CO_2.Cultivar.Week	7	793.03	113.29	5.15	<0.001
Residual	248	5453.72	21.99		
Total	287	12131.93			

(*P<0.05, **P<0.01 and ***P<0.001) indicate significant difference in CO_2 by cultivar interaction in weeks between treatments and controls grown under aCO_2 (430 ppm) and eCO_2 (500 ppm) for the first four weeks.