FACTORS INFLUENCING BREAST MEAT YIELD IN BROILERS

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DECLARATION

I hereby certify that this research is the result of my own investigation. Where use was made of others, it has been duly acknowledged in the text. The results in this dissertation have not been submitted, in whole or in part, for a degree at any other university.

Signed: ……………………………….. Date:

I hereby release this thesis for examination in my capacity as supervisor.

Signed: ………………………………… Date:

I hereby release this thesis for examination in my capacity as co-supervisor.

Signed: ………………………………… Date:

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ABSTRACT

The increased demand for breast meat of broiler chickens has challenged researchers to investigate management techniques that could be used to increase the production of this valuable commodity. Two experiments were conducted in this study; the first investigated the effect of early feeding of newly hatched broilers on breast meat yield (BMY) at market weight, and the second focused on improving BMY of broilers exposed to short daylengths by feeding higher than conventional levels of dietary protein. In the first experiment, of the 528 eggs set in the incubator, half were placed, at day 18, in hatching trays containing a commercial broiler starter feed whilst the others (the held group) were hatched conventionally. Six chicks from both fed and held groups were sampled at nine-hour intervals from the time that the first chicks hatched for a subsequent period of 36 h. After measuring their body weight these chicks were euthanised and dissected in order to measure their breast and yolk weights. Body protein, lipid and water contents were measured on each chick. At day 21, six birds from the fed and held groups were sampled, and body weight, breast weight and body protein content were measured. The yolk sac weight for fed birds was significantly reduced compared to that of held birds ($P < 0.001$). Both fed and held birds had the same breast weight at hatch, but at day 21 the mean breast weight of the fed birds was significantly heavier than of held birds ($P < 0.05$). The birds that were removed first from the hatcher had a reduced breast muscle weight compared to those that were removed last. In the second experiment, a total of 3200 day-old broiler chicks were reared in eight light-tight rooms. Four lighting regimens (12L: 12D, 16L: 8D, 20L: 4D and 24L: 0D) were randomised between rooms, with each light treatment being replicated twice. Each room was divided into four pens and 100 chicks in each pen received one of four dietary protein treatments. At day 35, three birds from each pen were sacrificed so that measurements could be made of breast, thigh, drum and wing weight, and carcass chemical composition. Breast weight increased as daylengths increased except in birds that were fed low protein diet (143 g protein/kg feed). High levels of dietary protein increased breast weight in birds on all other daylengths except for those on 12 h which showed a reduced breast muscle weight when dietary protein content was increased. The results of the first trial suggests that breast meat yield could be improved if newly hatched chicks are offered feed immediately after hatch, however the hypothesis that this increment was due to an overall increase in body protein content in the first few days after hatching could not be corroborated. The results further showed that held birds do not withdraw nutrients from breast muscle to maintain growth at hatch, this is because birds that stayed longer in the hatchery without feed showed increased breast muscle weight compared to those that were removed first. The second trial could not identify a suitable feeding programme to overcome the problem of a lower breast muscle weight that results from the use of 12 h of lighting compared to that when long daylengths are used. Highest breast weight was obtained when birds were fed high protein diet at 20 h. More research is required to further investigate the combined effect of early feeding and daylength on breast meat yield in broiler chickens.

CHAPTER 1

GENERAL INTRODUCTION

Poultry meat production has been increasing over the past years due to an increased consumption rate of leaner meat. The demand for poultry meat has been increasing because it is an affordable protein source relative to other meat sources. Broiler production in South Africa is increasing at a reasonable rate. For example in 2009 the broiler production per week was 0.8 % more than that of 2008 (SAPA, 2010a). In terms of consumption South Africa is dominated by poultry meat followed by red meat. The SAPA (2010b) reported the consumption for poultry meat in 2010 was 33.0 kg per person per annum and that for beef was 17.7 kg per person per annum.

The increased demand for chicken meat has challenged geneticists to focus on producing broilers with a fast growth rate over the past 50 years (Gous *et al*., 1999), and through genetic selection, five week-old broilers weighing more than 2 kg have been produced (Cobb, 2005). Havenstein *et al*. (2003) reported that at 42 days of age, the body weight for 2001 broiler strain (Ross 308) was 3.94 and 4.62 times heavier than that of 1957 strain (ACRBC) when fed 1957 and 2001 diets, respectively. Other than rapid growth rate, genetic selection has reduced the amount of food required to produce a given quantity of edible meat and amount of fat content (Havenstein *et al*., 1994). The current focus is to produce broilers with increased breast meat yield in order to complement the shift in consumer preference from a whole bird to further-processed products (Ewart, 1993). So in response to this demand intense selection pressure has been applied to select broilers with increased breast meat yield (BMY) (Le Bihan-Duval *et al*., 1998).

The success of genetic selection to increase growth rate in broilers indirectly highlights the importance of the embryonic and post-hatch developmental stages as they now represent a significant phase of the growing period (Lilburn, 1998). The performances in the early stages of life are known to affect performance at harvest (Moore *et al*., 2005); therefore, the development of early feeding strategies is becoming more of a requirement rather than an option. *In ovo* feeding of embryos and early feeding of hatched chicks are the possible strategies that can be used to enhance growth and development of broiler chicks at early stages of the growing period.

At hatch, the yolk sac is the primary nutrient source for the birds (Noy & Sklan, 1998; El-Husseiny *et al*., 2008). The yolk sac decreases exponentially post-hatch, therefore, nutrients from the yolk sac can no longer meet the nutrient requirements of the growing chicks soon after hatch (Noy & Sklan, 1998). This does not only compromise the early growth of chicks but also reduce the development of the digestive tract and immune response (Noy & Sklan, 1998). Immediate access to feed of hatched chicks may overcome these consequences (Sklan *et al*., 2000). Breast meat yield has been increased in the early-fed birds compared to delay-fed birds, and this behaviour was associated with increased activity of satellite cell proliferation in early-fed birds (Noy & Sklan, 1999).

The bird's performance after hatch would depend on its ability to consume feed which may vary depending on the growth phase, chemical composition, nutrient requirements and environmental conditions. Breast meat yield has been found to be reduced in birds exposed to short daylengths and this was associated with reduced feed intake in these birds which probably caused insufficient consumption of dietary protein (Brickett *et al*., 2007). It is hypothesized in the current study that feeding increased levels of dietary protein to these birds will overcome this, since high protein diets increased BMY in broilers (Kidd *et al*., 2004).

Previous research has shown that nutritional and lighting manipulation could improve BMY. This study was designed firstly to determine whether early feeding would improve BMY and to determine whether the delay-fed birds withdraw the nutrients from the breast muscle to maintain growth post-hatch. It would be expected that birds that had early access to feed would have heavier BMY and those that were delayed would show a reduced BMY as they spent more time in the hatchery without feed. Secondly, this study was designed to determine whether feeding high levels of dietary protein to birds exposed to shorter daylengths would increase the BMY.

CHAPTER 2

LITERATURE REVIEW

2.1 INTRODUCTION

In the past years the poultry industry aimed to produce broilers with increased growth rates and this resulted in reduced market age. Currently, the poultry industry is aiming to produce broilers with improved feed conversion efficiency and increased breast muscle weight in response to the shift in consumer preference from whole to processed product. Breast muscle is considered the most valuable chicken portion in the further processing market due to its large size in which even small differences in BMY could have a significant impact on economic returns. The increased demand of breast muscle has challenged researchers to investigate techniques that could be used to increase the yield of this muscle. Genetic selection has managed to produce broilers with increased breast muscle weight, however, nutritional and environmental factors can still affect the growth of this muscle.

It is important to understand the growth pattern of the breast muscle as this will assist in the determination of how nutritional and environmental factors affect the growth of the muscle and how these factors can be manipulated in order to meet the increased market demand and improve profitability. This review focuses on the genetic, nutritional and environmental factors that affect BMY in broilers.

2.2 MUSCLE GROWTH AND DEVELOPMENT

Muscles are important for all movement in the body including peristaltic movement of food through the gastrointestinal tract (GIT). Development of skeletal muscle begins during embryogenesis in which mononucleated myoblasts fuse to form myotubes which mature into myofibres (Schultz & McCormick, 1994). Skeletal muscle growth during embryogenesis is mainly due to an increase in the muscle fibre number which stops shortly after hatch. Thereafter, muscle growth is mainly due to an increase in size of myofibre (Campion, 1984). Myofibre number does not increase after hatch because myonuclei are post-mitotic and cannot synthesize DNA (Moss & Leblond, 1971). The myofibre size increases simultaneously with that of myofibre DNA content. This occurs through the donation of nuclei by mitotically active satellite cells situated between myofibre plasma lemma and the covering basement membrane (Mauro, 1961; Campion, 1984). Satellite cells proliferate then donate nuclei to the growing fibre which fuse into the multinucleated fibres to provide the genetic machinery for the age-related increases in myofibre size (Mozdziak *et al*., 1994). The high percentage of satellite cells that are present at hatch decreases to less than 5 % of the total myofibre towards the end of growth period (Hawke & Garry, 2001) and they become largely quiescent (Schultz *et al*., 1978). Thereafter, satellite cells can be activated and re-enter the cell cycle when there are stress signals such as muscle injury (Schultz *et al*., 1986; Velleman, 2007). Satellite cell mitotic activity at hatch is crucial for muscle development both at hatch and at maturity (Mozdziak *et al*., 1997). Therefore it is important to manage environmental and nutritional changes accordingly in order to prevent alterations in satellite cell proliferation which may cause the muscle not to reach its potential size.

The breast muscle growth has the same growth pattern as body weight (Scheuermann *et al.*, 2003) which starts with a lag phase during the early stages, followed by an exponential phase and final phase with reduced growth rate but increased muscle weight (Sussman, 1960; Batt, 1980). The maximum growth rate for breast meat weight is reached approximately four days after that of body weight (Scheuermann *et al.*, 2003).

2.3 FACTORS AFFECTING BROILER BREAST MEAT YIELD

Genetic, nutritional and environmental factors play a crucial role in growth performance of broilers. Therefore, it is important to understand how these factors can be manipulated in order to improve the chemical and physical composition of the breast muscle in such a way that allows the bird to reaches its potential.

2.3.1 Genetic selection for increased breast meat yield

Genetic selection has been instrumental in the last half century in the improvement of broiler performance to meet the requirements of the market. The increase global demand for cheaper and healthier meat has challenged the breeding companies to select for increased growth rate, which in turn resulted in a reduced market age (Chambers *et al*., 1981). Emmans & Kyriazakis (2000) reported that from 1950 to 2000, the broiler growth has increased from 25 grams per day to 100 grams per day. An increase in BMY has also been achieved through genetic selection (Le Bihan-Duval *et al*., 1998; Le Bihan-Duval *et al*., 2001). Le Bihan-Duval *et al*. (1998) reported that BMY can be improved by selection since the heritability for the trait ranges between 0.53 and 0.65. This was supported by Berri *et al*. (2001) who reported that broilers that were selected for increased breast weight had 61 % higher breast weight compared to an unselected control strain. Selecting for increased BMY has been found to be negatively correlated (-0.39) to abdominal fat (Zerehdaran *et al*., 2004). This shows that selecting for higher BMY will concurrently reduce fat content of the carcass hence leaner meat will be produced and market requirements will be met. On the other hand, selecting for breast weight has been accompanied by increased incidence of leg disorders, which could be due to that the frame structure could not support the over-developed body (Wilson, 1990). Selecting birds for increased BMY has been reported in the study of Le Bihan-Duval *et al*. (1999) to affect the quality of meat. These authors reported that the meat from the experimental line of birds selected for increased BMY was pale in colour compared to that from control line birds. In contrast, Berri *et al*. (2001) did not support the idea that selection has a negative effect on meat quality and this was because the chemical composition of the breast muscle was almost unmodified by selection.

As genetic selection for broilers with fast growth rate is expected to continue in the near future, the market age will be reduced further, and therefore introducing feed in hatcheries will not be optional because by that time the first week of life will be representing more than 20 % of the growing period. Moreover, the incidence of physiological stress and muscle abnormalities will be more evident, therefore the breeding program that is currently used will need to be modified in such a way that it takes the biochemical and morphological alterations into consideration to minimize these problems.

2.3.2 Effect of nutritional strategies on breast meat yield

The short growing cycle of broilers calls for the development of continuous feeding systems to supply nutrients to the developing embryo, feed to newly hatched chicks within the hatchery and a pre-starter diet at placement (Noy & Uni, 2010). *In ovo* feeding and immediate feeding post-hatch are processes that can be used to supply nutrients to embryos and newly hatched chicks.

2.3.2.1 *In ovo* feeding

High glucose level is critical for late embryonic development, for the enhancement of the hatching process such as clearing the shell, and post-hatch development of broilers until feeding commences (John *et al*., 1988; Christensen *et al*., 2001). During this period the maintenance of glucose homeostasis is dependent upon the amount of glucose held in reserves in the liver (Elwyn & Burszstein, 1993). Glucose is generated from glycogen in the liver through gluconeogenesis from protein first mobilized from albumen and then muscle (John *et al*., 1988). Glycogen reserves are withdrawn as embryos go through the hatching process, and begin to replenish when the newly hatched chicks access exogenous feed (Rosebrough *et al*., 1978). Insufficient glycogen will force the embryo to mobilize more muscle protein toward glucogeneogenesis, and consequently early muscle growth and development is compromised (Vieira & Moran, 1999). Therefore administering carbohydrates into the amniotic fluid of embryos in the late term of incubation not only improves glycogen reserves, but also increased hatching weight and relative breast meat weight by 5 to 6 % and 6 to 8 % respectively, and this advantage may last until day 25 (Uni *et al*., 2005). A body weight difference of 2 g at hatch may result in a 50 to 60 g of increase in body weight at 25 days of age (Uni *et al*., 2005). The increased body weight at hatch is associated with a more developed GIT which functions as that of a two day-old immediately fed chick (Uni & Ferket, 2004; Foye *et al*., 2007). Other than administering carbohydrates, use of beta-hydroxy-beta-methylbutyrate has been found to increase BMY as this is a leucine metabolite which decreases chicken mortality and prevent excessive muscle proteolysis (Uni *et al*., 2005; Foye *et al*., 2006; 2007).

The introduction of *in ovo* feeding to commercial operations is questionable due to the large number of chicks hatched per day. Moreover, it is still considered expensive and difficult to practice but with the development of the equipment this might be a possibility in the future. The benefits of the technique cannot be ignored; therefore, more research is required to determine whether the advantages observed from *in ovo* feeding are sufficiently relevant to justify the cost and changes in systems to be implemented commercially.

2.3.2.2 Early feeding of broiler chicks

In commercial hatcheries, turkey poults are placed into brooders 36 h post hatching and this is when the majority has emerged from the shell (Moran, 1990). The holding period further increases when post-hatch treatments such as sexing, vaccination and transportation to farms are practiced (Pinchasov & Noy, 1993). It is common to find a reduced performance in chicks that hatch first in a batch because of the increased length of the holding period (Pinchasov &

Noy, 1993; Noy & Sklan, 1999; Sklan *et al*., 2000). Because of the early market weight that broiler chicks obtain (Havenstein *et al*., 1994; 2003), feeding broiler chicks immediately after hatch is recommended because the first week post hatching represents approximately 20 % of the rearing period (Lilburn, 1998). In addition, the increased body weight at hatching in the early fed bids resulted in an increased body weight at slaughter age (Halevy *et al*., 2000).

Hatched chicks are dependent on the yolk sac for nutrients for the first three days post-hatch (Donaldson, 1967; Noy *et al*., 1996; Noy & Sklan, 1999; Gonzales *et al*., 2003; Juul-Madsen *et al*., 2004). The residual yolk sac becomes internalized in the abdominal cavity just before hatch and at this stage it contains approximately 50 % lipid which can help sustain the chick during the first days of life (Noy *et al*., 1996). However the nutrients contained in the yolk sac seem to be insufficient to support both growth and maintenance, especially due to the broilers' high genetic potential for growth. Yolk sac weight decreases exponentially during the first four days post-hatch (Noy *et al*., 1996; Noy & Sklan, 1998; Bigot *et al*., 2003) and it is then negligible because it is approximately 1 g or less (Noy *et al*., 1996; Noy & Sklan, 1998). Thereafter, chicks are supposed to consume exogenous feed for further growth. Many studies (Noy & Sklan, 1997; Noy & Sklan, 1999; El-Husseiny *et al*., 2008) reported that early feeding of chicks result in rapid utilization of the yolk sac compared to when chicks were starved. This rapid absorption may be due to stimulated growth of the GIT stimulating anti-peristaltic activity (Noy & Sklan, 1999). In contrast, Bigot *et al.* (2003) did not find any difference in yolk sac resorption between early- and late-fed chicks in the first three days post-hatch, but at day four yolk sac weight was slightly higher in the early-fed group compared with the delay-fed chicks, 1.13 ± 0.13 g and 0.075 ± 0.07 g respectively. The difference between these experiments may be that the delay-fed birds in the study of Bigot *et al*. (2003) had access to water six hours before feeding, which may have enhanced the intestinal growth hence yolk sac was utilised in the same rate as those that had early access to feed.

Starving chicks for 48 h resulted in a decreased intestinal absorption area thereby limiting nutrient uptake capacity, thus contributing to a decreased growth potential later in life. Chicks that have access to feed immediately after hatch have an increased body weight and body weight gain, and this advantage lasts until market age (Halevy *et al*., 2000; Sklan *et al*., 2000; Bigot *et al*., 2003). Sklan *et al.* (2000) reported that early-fed chicks were 2 to 3.5 g heavier than held birds, when removed from the incubator. Similarly, Bigot *et al*. (2003) reported a 36 % increase in body weight in early-fed birds 48 h post hatch, and a 25 % loss in body weight in chicks starved for 48 h compared to those fed immediately, at day six of the experiment. Delayed feeding of chicks can decrease body weight at a rate of 0.14 to 0.17 g/h, therefore within 48 h birds can lose 6 to 9 g body weight (Sklan *et al*., 2000) which is 7 % (Bigot *et al*., 2003) to 7.8 % (Noy & Sklan, 1999) of the hatching body weight. Feeding chicks after 24 h of starvation did not have any effect on body weight (Juul-Madsen *et al*., 2004). In contrast, Bigot *et al*. (2003) found that early-fed birds were 15 and 30 % heavier than delay-fed birds at day one and two of age respectively.

The reduced body weight at day 21 in starved chicks may be associated with poor feed conversion and depression of feed intake (El-Husseiny *et al*., 2008). Feeding chicks after 2 d of starvation post-hatch increased feed consumption but it remained insufficient to correct the consequences of delayed feeding (Halevy *et al*., 2000; Bigot *et al*., 2003). These differences in body weight between fed and non-fed chicks remained significant until harvest (Halevy *et al*., 2000). Bigot *et al*. (2003) reported that even after held chicks were fed, the reduction in feed intake was still recorded at three and six days of age. Giving water instead of feed to newly hatched chicks may be an alternative, but body weight would not increase as much as those given feed (Noy & Sklan, 1999). The decreased body weight in delayed fed chicks was accompanied by a lower relative breast muscle weight (El-Husseiny *et al*., 2008), and this was associated with reduced satellite cell proliferation (Halevy *et al*., 2000; El-Husseiny *et al*., 2008). Breast muscle in fed chicks reached 15.5 g/100 g BW, whereas in the starved chicks it comprised <14 g/100 g BW at day 41 (Halevy *et al*., 2000). Immediate placement and feeding of broiler chicks can increase breast muscle weight (Halevy *et al*., 2000; Juul-Madsen *et al*., 2004; El-Husseiny *et al*., 2008) by 4 to 10 % between day 21 and slaughter period (Noy & Sklan, 1999). Therefore, the impact of early nutrition on satellite cell mitotic activity is very important for the determination of ultimate breast meat yield (Halevy *et al*., 2000). In contrast, Mozdziak *et al*. (2002) reported that any improvement in meat yield through early nutritional supplement did not occur through a satellite cell pathway and that there was no compensatory response in the satellite cell population following re-feeding of starved turkey poults. This was believed to be due to an increased feed intake of feed containing hydrated low fat, highly digestible protein and carbohydrate which decreased local expression of factors that stimulate satellite cell mitotic activity. Early fed chicks showed an increased pectoralis major muscle weight from day one, whereas in delay fed chicks the increase was observed from day two (Bigot *et al*., 2003). In the same trial, by day four of age, pectoralis major muscle weight was increased by about 2.5 and 5 fold in delayed and early fed chicks respectively.

Danisman & Gous (2007) showed that regardless of the strain, breast meat yield was below potential in the first two days post-hatch (Figure 2.1), when the allometric relationship between BMY and body protein weight was determined. These birds were not fed while they were in the hatchery, so it is possible that they withdraw nutrients from the breast muscle to sustain growth during the holding period and this resulted in reduced BMY.

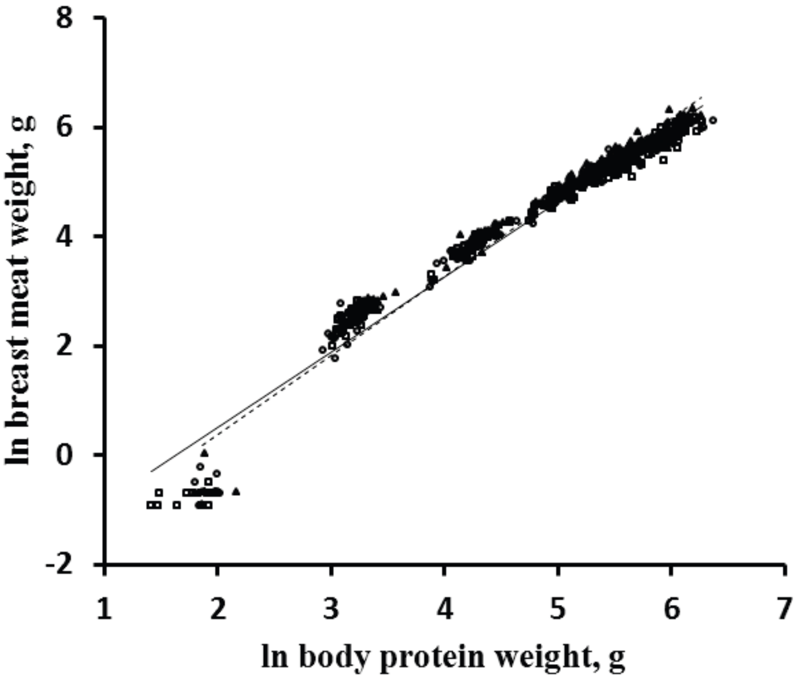

Figure 2.1 Allometric relationship between *ln* breast meat yield and *ln* body protein weight for three broiler strains namely, Cobb, Ross 308 and Ross 788 (from Danisman & Gous, 2007).

The immune system for chicks at hatch is not fully developed, and delayed feeding reduces the development of the system resulting in increased susceptibility to diseases, and consequently increased mortality (Dibner *et al*., 1998).

2.3.2.3 Nutrient density and broiler performance

Dietary protein intake has a major effect on the quality of broiler meat, muscle development, and on carcass fat content (Smith & Pesti, 1998; Corzo *et al*., 2005). An adequate dietary protein to energy ratio is critical to maximise broiler performance and economic returns. Feed cost constitutes approximately 70 % of the total production cost (Kamran *et al*., 2004), with protein being one of the most expensive inputs. Due to the high cost of protein sources, it was suggested that reducing dietary protein will reduce input cost and nitrogen waste. However, the reduction in dietary protein is associated with decreased meat yield and increased fattening, due to an increased energy intake in an attempt for the bird to meet its requirement for the limiting nutrient (Gous *et al*., 1990). This compensation is only possible if the bird is not constrained by its gut capacity, in which case it will achieve its potential body protein growth. An oversupply of nutrients reduces feed intake since a small concentrated amount consumed is sufficient to meet the bird's requirements (Gous *et al*., 1990). Oversupply of nutrients more than the bird requires may increase feed cost because birds would consume feed to meet the requirements for the limiting nutrients (Kidd *et al*., 2005). Therefore, it is important to match nutrient density of the diet with changing nutrient requirements as the bird ages.

An increase in dietary protein causes an increase in body weight without an increase in feed intake; hence feed conversion efficiency is improved (Smith & Pesti, 1998; Dozier *et al*., 2006). Similarly, feeding high levels of dietary protein would increase BMY as compared to low protein diet, and the amount of lipid deposited in the body would be reduced (El-Fiky *et al*., 2007; Danisman, 2009). Dietary protein concentration should be high, especially immediately post-hatch (Kemp *et al*., 2005). Smith & Pesti (1998) reported that birds that were fed a 24 % crude protein (CP) pre-starter diet had significantly improved breast muscle weight as compared to those fed a 16 % CP diet at day 53. They further reported an increased feed intake in birds fed 16 % protein compared to those fed 24 % protein. The increased feed intake was due to insufficient supply of dietary protein, which resulted in an excess in energy intake and consequently an increase in abdominal fat. These findings are in agreement with those of Kidd *et al*. (2004), who found that chicks fed on a highly nutrient dense diet had significant increased BMY of 20.5 % at slaughter compared to those fed on medium (19.4 %) and low (17.5 %) dietary CP. In the same study, birds fed on low protein diets had reduced body weight and increased feed conversion. Similar results were reported by Corzo *et al*.

(2005) where BMY showed a significant reduction of 0.6 and 0.5 % at 42 and 56 days of age respectively in birds fed low protein diets.

Both amino acid and energy requirements change daily but not in a same proportion, the amino acids increase faster than that of energy. Therefore, it is important to adjust nutrients at a correct period so that the supply will be efficient (Kidd *et al*., 2005). Highly digestible nutrients such as protein are recommended to be fed to newly hatched chicks to accommodate the immature digestive system and also as an aid in meeting the requirements (Longo *et al*., 2007). A high amino acid to energy ratio is required for the fast growing strains (Bartov & Plavnik, 1998). However, excessive and non-balanced supply of amino acids can increase energy cost due to the fact that excess amino acids become toxic and nitrogen needs to be excreted from the body in the form of uric acid (Moran *et al*., 1992).

Dietary energy plays an essential role in protein deposition; therefore, an insufficient supply will constrain protein deposition (Gous *et al*., 1990). Energy supply is as important as that of dietary protein throughout the birds' life. Highly digestible dietary energy sources such as sucrose are recommended for broiler chicks in the first week of life to maximise performance during this time (Plavnik *et al*., 1997). Molenaar *et al*. (2009) found that increased levels of dietary energy in pre-starter feed did not increase growth post hatch. This may be due to immature digestive tract and reduced secretion of enzymes such as lipase, trypsin and amylase that are responsible for fat digestion and absorption (Sklan & Noy, 2000; Longo *et al*., 2007). Feed intake decreased and feed conversion improved when energy levels were increased in the diet (Vieira *et al*., 2006).

2.3.3 Environmental factors affecting breast meat yield

The correct management of environmental conditions will aid to maximise flock performance. It also ensures that the bird's health and welfare are not compromised**.** Environmental temperature, light intensity and photoperiod are the factors that need to be controlled in the broiler house to optimise broiler's performance.

2.3.3.1 Environmental temperature

High ambient temperature is one of the major factors that can hinder poultry meat production. Exposing broilers to high temperatures has been associated with reduced growth rate and meat yield due to reduced feed intake in an attempt to reduce metabolic heat production to maintain body homeostasis (Cahaner & Leenstra, 1992; Temim *et al*., 2000; Deeb & Cahaner, 2001; Pope & Emmert, 2002). Al-Batshan & Hussein (1999) using cyclic temperature and Temim *et al*. (2000) using constant temperature reported that birds reared in hot (32 to 34 °C) temperatures had a reduced breast meat weight compared to those reared at moderate temperature (22 to 24 °C), but abdominal fat was not affected (Al-Batshan & Hussein, 1999). Heat tolerance in broilers decreased with an increase in body weight (Cahaner & Leenstra, 1992), meaning birds were able to tolerate high temperatures better in the first three weeks of life. Similar results were reported by Geraert *et al*. (1996) who showed that feed intake was reduced by 14 % between two to four weeks of age and 24 % between four to six weeks when birds were exposed to 32 °C, which resulted in reduced body weight gain. Heat exposure impairs performance mostly during the growing and finishing phase (Rosa *et al*., 2007); therefore, from three weeks to slaughter the ambient temperature should be reduced to be between 18 and 21 °C.

The ability to tolerate heat in broilers can be increased by means of genetic selection, but this may lead to reduced growth potential under normal temperatures (Mathur & Horst, 1994). Moreover, the heritability of heat tolerance is low and it is negatively correlated with growth rate (El- Gendy & Washburn, 1995). Birds that are selected for high growth rate have the reduced ability to cope with heat stress (Deed & Cahaner, 2001). Heat stress in broilers could be reduced by selecting for the naked-neck gene (Na) because it reduces feather cover by 20 % in the heterozygous (Na/na) birds and 40 % in homozygous (Na/Na) as compared to their fully-feathered counterparts (Cahaner *et al*., 1993; Deeb & Cahaner, 1999). The loss of feathers from the neck increases the effective surface of heat dissipation and increases the sensible heat loss from the neck, hence body temperature is stabilised and consequently performance can be improved (Yahav *et al*., 1998; Deeb & Cahaner, 1999). The naked-neck gene is preferred in the heterozygous (Na/na) form compared to the homozygous (Na/Na) form because the latter may be a disadvantage when broilers are exposed to low temperatures because birds might consume more feed to meet the increased energy requirements for thermoregulation (Yahav *et al*., 1998). This genotype (Na/Na) may also cause excess heat loss during the early days of life hence growth would be compromised (Deeb & Cahaner, 1999). In addition, when this gene is in the homozygous state it is known to compromise hatchability (Merat, 1986).

Naked-neck birds have a higher growth rate and meat yield compared to fully-feathered birds under high temperatures (Cahaner *et al*., 1993). This can be associated with high feed intake which resulted in high body weight gain and feed conversion efficiency in the naked-neck birds while the body temperature is low (Cahaner *et al*., 1993; Yahav *et al*., 1998). High temperatures decreased body weight, body weight gain and carcass parts in both naked-neck and fully-feathered birds but the effect was more pronounced in fully-feathered birds (Deeb & Cahaner, 1999). Deeb & Cahaner (2001) reported that at day 53, the BMY for the heterozygous naked-neck birds was lowered by 11.8 % at high temperature (30 °C) than that of birds maintained at comfortable temperature (25 °C), whereas in the normally feathered birds the difference was 21.4 %. These findings were in agreement with the results of Ajang *et al*. (1993) and Cahaner *et al*. (1993) who reported that BMY for naked-neck birds was 6.4 to 17 % significantly more than that of the normal chickens respectively. The higher BMY in the naked-neck (Na/na) birds maybe due to larger protein availability for muscle production since less feathers are produced (Ajang *et al*., 1993; Cahaner *et al*., 1993). In contrast, Raju *et al*. (2004) did not find any differences between naked-neck birds and their counterparts in terms of breast muscle protein, body weight, feed intake and feed conversion efficiency. The difference between these studies may be due that in the study of Raju *et al*. (2004) the environmental temperature was characterised by cyclic diurnal temperatures compared to other studies where the temperature was constant.

The naked-neck gene has also been associated with reduced fat deposition in the breast muscle. This was attributed to the reduced insulation that these birds have, causing the birds to use more energy to thermo-regulate and consequently less fat is deposited (Raju *et al*., 2004). Moreover, the advantage of naked-neck broilers over fully-feathered birds might be expected to play more role since the ambient temperatures that are currently considered to be normal are expected to reduce broiler growth in the near future, since birds are continually selected for high growth rate and breast yield (Deeb & Cahaner, 2001).

Other than selecting for the naked-neck gene, increased dietary protein intake has also been investigated to determine whether it would compensate for the consequences of high temperature on the performance of the fully-feathered birds. Cahaner & Leenstra (1992) reported that, irrespective of the temperature, high dietary protein increased breast meat weight but there was no interaction between temperature and protein. Cahaner *et al*. (1995) found that the growth rate and meat yield of the fast growing strain was reduced when these birds were given high protein diets when exposed to high temperatures. In addition, the study of Cheng *et al*. (1997) suggested that a crude protein intake higher than NRC recommendations should not be used to grow broilers exposed to temperatures greater than

26.6 °C because a higher protein concentration in the feed did not allow the birds to compensate for the reduced feed intake. The reduced performance in birds fed high protein diets at high temperature may be associated with the high heat increment from protein metabolism, which prevented the birds from eating the feed; hence the performance was reduced (Cheng *et al*., 1997). In contrast, Temim *et al*. (1999; 2000) reported that the reduced BMY for fully feathered broilers subjected to high temperatures can be increased by feeding increased levels of dietary protein which will compensate for the reduced FI (Temim *et al*. 1999; 2000). Temim *et al*. (1999) comparing 20 and 25 % CP, and Temim *et al*. (2000) feeding different CP levels ranging from 10 to 33 % found that high levels of dietary protein slightly improved the performance of broilers exposed to high temperature (22 vs. 32 °C), but this was not sufficient to aid broilers to withstand hot conditions. Rahman *et al*. (2002) reported that feeding low protein diets in hot climates resulted in poorer feed conversion and body weight gain and suggested slightly less than recommended protein (21 vs. 23 % CP) should be used during the hot humid season (in Bangladesh) for optimal growth. Moreover, heat production was increased in birds fed a low protein diet due to increased feed consumption as broilers were eating to meet protein requirements. Further research is required to determine whether increased levels of dietary protein would increase the performance of the naked-neck broilers reared in hot temperatures.

Feeding increased levels of dietary energy to broilers exposed to high temperatures to increase body weight gain has been proposed since energy has a low heat increment. However, the breast muscle weight was not affected (Yalcin *et al*., 1998; Raju *et al*., 2004; Ghazalah *et al*., 2008). In contrast, Al-Batshan & Hussein (1999) reported an increased performance and BMY when broilers were fed increased levels of dietary energy but these broilers were exposed to moderate temperatures, and this was not the case in hot (26 to 34 °C) cyclic temperatures. The increased level of dietary energy intake was also associated with increased fat deposition which is not favoured by consumers (Yalcin *et al*., 1998; Al-Batshan & Hussein, 1999). Zaman *et al*. (2008) reported that feeding low CP with high energy improved breast weight when temperatures ranged from 32 to 39 °C. The high intake of dietary energy enhanced protein deposition since it was not a limiting factor (Cheng *et al*., 1997). The above studies contradict each other in terms of the optimum dietary protein levels that should be fed to birds in high temperatures; therefore further research is needed to investigate the optimum protein needed to maximise production under these conditions. Feeding high protein diets during the cooler hours of the day and high energy diets during the warmer time of the day reduced body temperature and growth (De Basilio *et al*., 2001). The reduced growth suggests that feeding high protein diets during cooler hours does not compensate for the reduced growth during warmer hours. Therefore, one balanced feed should be formulated to meet the requirements of the bird. Pope & Emmert (2002) reported that feeding diets with reduced amino acids every other day to broilers may reduce heat stress in broilers exposed to high temperatures without affecting growth performance. It may be expensive and difficult to practice this feeding system in commercial farms because sets of diets will have to be formulated and more labour will be required to change the feed every day.

Yalcin *et al*. (2001) reported that feed restriction during the warmest part of the day reduces heat stress and mortality, and increases feed efficiency in broilers, however, body weight was not affected. The lack of effect on body weight may be due to the birds' ability to consume feed during the cooler period to compensate growth (Arjona *et al*., 1988). The number of hours for feed withdrawal must be reviewed because prolonged feed restriction might reduce growth rate hence delay market weight. Exposing five-day old chicks to high temperature (36 °C) for 24 h increases broiler's ability to cope with heat stress later in life since these birds will have low body temperature at both low and high environmental temperature, and it also improves feed efficiency (De Basilio *et al*., 2001; Yalcin *et al*., 2001) but this conditioning does not affect body weight (Yalcin *et al*., 2001). Early feed restriction of 60 % of the daily *ad libitum* consumption at days four, five and six of age and exposure to high temperature (36 °C) until day 21 improves heat tolerance and weight gain when birds experience heat stress later in life (Zulkifli *et al*., 2000; Liew *et al*., 2003; Abu-Dieyeh, 2006).

2.3.3.2 Light intensity

Light intensity is considered important in broiler production as it affects the growth performance of chicks. Commercial broilers are mostly reared under artificial lighting; which presents the advantage of adjusting light intensity and wavelength to optimize broiler performance (Kristensen *et al*., 2007). Natural light has high intensity that can increase up to 100 000 lux whilst chicks are recommended to receive light intensities between 10.8 to 64.8 lux (Barrot & Pringle, 1951; Cherry & Barwick, 1962). Light intensities beyond this range will reduce growth performance in birds, and for optimal growth birds should receive 20 lux (Thery, 2001). During the first week of life broiler chicks do not have a light source preference due to the immature nature of the visual cortex (Rogers, 1993). Use of fluorescent light over incandescent light is recommended to rear broiler chickens because of potential energy saving (Kristensen *et al*., 2007) and improved performance to market age (Zimmermann, 1988). Rearing broilers under incandescent light needs to be monitored because two week old broilers prefer a high (100 clx) light intensity compared to six week old broilers which prefer low (5 clx) intensity (Kristensen *et al*., 2007). Reduction of light intensity to 0.25 foot candles (fc) increases feed intake and therefore body weight (Downs *et al*., 2006; Kristensen *et al*., 2007). However the increased feed consumption may cause excessive deposition of abdominal fat which reduces carcass quality of the chicken. Reduced light intensity increases wing and leg yield at the expense of the breast meat (Downs *et al*., 2006). High light intensity reduces leg disorders (Downs *et al*., 2006) due to increased walking and standing behaviours (Kristensen *et al*., 2007).

2.3.3.3 Management of photoperiod

Commercial broilers are commonly reared under continuous or near continuous lighting for optimal growth and performance due to maximised feed intake. However, continuous lighting has been associated with increased physiological stress due to reduced sleeping hours hence animal welfare is compromised (Lien *et al*., 2007). The new EU welfare for birds kept for meat production stipulates a 24 h cycle, with a day and night lighting schedule of more than 6 h of darkness, in which 4 h should not be interrupted (European Commission, 2000); hence the current practice of rearing birds on long days may be required to change. Therefore, attention has to be paid to other lighting schedules in order to optimise production without compromising welfare.

Increased hours of lighting have been found to increase feeding time hence increased body weight and this advantage lasts until end of experiment (Ingram *et al*., 2000; Downs *et al*., 2006; Lien *et al*., 2007). Ingram *et al.* (2000) reported a significant decrease in body weight of 2 % due to a 4 % decrease on feed intake in birds that were exposed to 12 h compared to those on 23 h. However, feed conversion efficiency was improved for birds exposed to 12 h. These findings are in agreement with results reported by Classen (2004) who found that birds on 20 h were 85 g heavier than those on 12 h due to reduced total feed intake in birds subjected to 12 h (3.27 kg/ bird) compared to those on 20 h (3.54 kg/bird) but had improved feed efficiency and reduced mortality. Ozkan *et al.* (2006) observed a reduced body weight in birds on 16 h compared to those on 24 h until day 21. Thereafter growth compensation occurred since there were no significant differences in body weight at day 42 except for numerically lower body weight in 16 h birds (2569 g) than 24 h birds (2641 g). Birds exposed to short (12 h) daylengths had reduced BMY compared to those on long (20 h) daylengths at day 35 (Brickett *et al*., 2007; Lien *et al.*, 2007). However, drum, back and wing yield were heavier (Brickett *et al*., 2007).

The positive relationship between daylength and feed consumption may be age-dependent. Body weight gain increased by 10 g/ h of lighting (Lewis & Morris, 2006), and feed intake increased from 8 g (Lewis *et al*., 2009a) to 15 g (Lewis & Morris, 2006) per hour of lighting until day 21; thereafter, body weight gain decreased by about 6 g/ h for birds given more than 12 h (Lewis & Morris, 2006). The authors further reported that at day 49, the body weight for birds that were exposed to daylengths longer than 12 h was not significantly affected but those on 8 h showed a reduced performance compared to those above 12 h (Lewis & Morris, 2006). This suggests that longer daylengths could be used during the first three weeks of the growing age. Thereafter, 12 h daylengths could be used to obtain optimal performance. This will not only improve bird's performance but also reduce electricity costs. The total mortality from 0 to 49 days was higher in birds reared on long daylengths compared to those on short daylengths (Lewis & Morris, 2006). The studies of Lewis & Gous (2007) and Lewis *et al.* (2008) did not find any significant effects of lighting on feed intake, growth and feed conversion efficiency when birds were subjected to 8, 16 or increasing (8 to 16 h) photoperiods for both males and females until day 21. However, between day 22 and 42 the birds on 8 h and increasing (8 to 16 h) lighting had higher body weight gains and feed conversion efficiency as compared to those on 16 h. The reduced body weight of birds on 16 h may be due to reduced feed intake compared to those on 8 h which ate about half of their daily feed intake during the dark period while those on 16 h consumed about 10 % of their feed during the dark period. The increased weight gain of birds on 8 h can be associated with lower energy expenditure during the dark period (Lewis & Gous, 2007). The increased feed intake in birds that were transferred from 8 to 16 h may be due to the birds continuing to eat in the dark even when they were transferred to 16 h. This suggests that it may not be necessary to grow birds on long daylengths since they are able to consume sufficient feed by eating in the dark. However, it may be necessary to revise the levels of dietary protein to increase the BMY which was reduced in birds reared on short daylengths. Lewis *et al.* (2009a) reported that body weight and feed intake increased with increasing daylengths until day 21, but between day 22 and 35 body weight increased by 50 g/ 1 h increase in daylength up to 7 h, and beyond 7 h no significance was observed. The feed intake increased by 90 g/ 1 h increase in daylength until 4 h, beyond that it was minimally affected.

Other studies have investigated the use of other lighting schedules such intermittent lighting and step-up or step down lighting. Scott (2002) did not find any significance in body weight between birds on constant photoperiods (16 or 23 h) and those on intermittent (5 L: 1 D) lighting until day 21, but at day 35 the body weight for birds on 16 h (1771 g) was significantly heavier than those on 23 h (1735 g) but not significantly heavier than those on intermittent lighting (1752 g). Rahimi *et al.* (2005) did not find any significant differences between intermittent (1 L:3 D) and constant (23 h) lighting in terms of feed intake but feed conversion ratio was improved for birds on intermittent lighting compared to birds subjected to constant lighting. El-Fiky *et al.* (2007) who showed a reduced body weight and feed intake in birds subjected to intermittent lighting (4 L: 8D) compared to those on 23 h. In contrast, Lewis *et al.* (2010) noted a reduced body weight gain and feed intake in birds that were given 18 h compared to those given intermittent (1 L: 2 D) lighting by day 21; however, at slaughter no significant differences were found in feed intake, body weight, feed conversion efficiency or BMY between the lighting treatments. Mortality was higher in birds given 23 h than those in intermittent lighting. The difference between these studies may be due to different lighting hours used for intermittent lighting in each experiment. Overall, these studies suggest that it is better to use constant lighting compared to intermittent light because no advantage of using intermittent lighting was observed at 35 days of age, however, the advantage of saving electricity cannot be ignored.

Downs *et al.* (2006) showed that birds that were exposed to a decreasing-increasing (DEIN) lighting programme (going from decreasing to increasing quantity of light) had suppressed body weight in the early growth phase compared to those on constant lighting. On day 15 and 35 body weight was 36.3 g and 68.1 g less, respectively for DEIN birds. The decreased body weight was due to reduced feed intake which was 2.2 % less than those on 23 h at day 35. At day 56, there was no significant differences in body weight and feed intake among the treatments, but DEIN birds were numerically heavier (+ 31.8 g) and had consumed more feed (+ 86. 3 g) than those on 23 h. This indicates that growth compensation occurred in the last two weeks when birds were transferred back to 23 h. In addition, DEIN birds yielded larger legs at an expense of breast yield which was 0.44 % less than in the 23 h group. Schwean-Lardner *et al*. (2007) reported a rapid growth and intermediate BMY in birds that were given step-up lighting compared to those on constant 14 and 23 h, while birds on 14 h had significantly heavier drum and thigh yields compared to those subjected to other lighting programmes. Further, there were no significant differences in feed conversion ratio for step-up and 23 h birds, and it was significantly reduced in birds on 14 h.

2.4 CONCLUSIONS

Birds given early access to feed post-hatch have been shown to have an increased BMY due to an increased proliferation of satellite cell in the breast. The reduced BMY in birds which have delayed access to feed after hatch could be due to a deficiency in the nutrients in the yolk sac to meet the requirements for early growth. This would cause the birds to withdraw nutrients from the breast muscle to maintain growth until feeding commences, hence breast muscle growth would be compromised. Therefore, it is possible that giving hatched chicks early access to feed could increase the BMY.

The studies reviewed in this chapter show that the breast meat growth is compromised when birds are reared on short daylengths. It is hypothesised that the reduced breast muscle weight in such conditions is due to insufficient consumption of dietary protein. This suggests that there might be an interaction between dietary protein and photoperiods. Increased levels of dietary protein increases BMY, therefore, it is possible that the reduced BMY in broilers grown in short daylengths may be increased by feeding increased levels of dietary protein.

CHAPTER 3

THE EFFECT OF EARLY OR DELAYED FEEDING ON BREAST MEAT YIELD OF GROWING BROILERS

3.1 INTRODUCTION

In commercial hatcheries, chicks are removed when the majority has hatched. There is sometimes a considerable delay between the time the chick hatches until it is placed in the broiler house with access to feed and water. During this time early growth could be compromised and body weight at slaughter would be reduced (Noy & Sklan, 1999; Sklan *et al*., 2000). The most severely affected are those chicks that hatch first. Even though chicks obtain nutrients from the yolk sac after hatch, these nutrients are utilised at a slow rate due to the immature development of the intestine (Noy & Sklan, 1999; Bigot *et al*., 2003; El-Husseiny *et al*., 2008).

Immediate access to feed in the hatching trays has been associated with increased breast muscle weight at hatching and at slaughter age (Halevy *et al*., 2000; Sklan *et al*., 2000; Bigot *et al*., 2003; El-Husseiny *et al*., 2008). This has been attributed to a more rapid utilisation of the yolk sac, due to increased intestinal mechanical (anti-peristaltic) activity, and to the more rapid development of the microvilli in the small intestine (Noy *et al*., 1996; Noy & Sklan, 1998; Noy *et al*., 2001). Early feeding has also been shown to increase satellite cell proliferation (Mozdziak *et al*., 2002; Halevy *et al*., 2003) with a concomitant increase in muscle growth.

In the study of Danisman & Gous (2007), breast muscle weight in chicks held without food before being removed from the incubator appeared to fall below the potential in the first few days of life, the potential being described by the allometric relationship between breast muscle and body protein weight. This was not observed in other muscle parts (thigh, drum and wing). However, this difference in BMY was not observed later in the broiler's life. This suggests that the breast muscle was used to provide nutrients to the chicks at the time of hatching as nutrients from yolk sac were not sufficient.

The aim of this study was to determine whether immediate access to feed of hatched chicks would improve breast muscle weight both at hatch and day 21. A secondary aim was to evaluate if chicks given delayed access to feed withdraw nutrients from breast muscle to maintain growth.

3.2 MATERIALS AND METHODS

3.2.1 Study site

This trial was conducted in the brooder room at Ukulinga research farm, University of KwaZulu-Natal in June 2010.

3.2.2 Incubation and treatment groups

Eggs from Cobb 500 breeders housed at Ukulinga farm, were incubated. The eggs were randomly allocated to four setting trays using 132 eggs per tray and two trays per incubator (total of 528 eggs). The incubator was set at 37.7 °C temperature and 60 % relative humidity, and turned through 90° every hour, for 18 days. At day 18, the eggs in each incubator were transferred at random to one of the two hatching trays one of which was fitted with small Perspex feeders in which a crumbled commercial broiler starter feed was placed (Meadow Feeds, Pietermaritzburg).

Artificial lighting was used continuously in each incubator, commencing on day 18 of incubation. Water trays were placed in the incubator from day 18 to minimize evaporation from chicks.

3.2.3 Hatching and chick selection at hatch

It was assumed that the chicks would hatch over a period of 36 h starting at 20.25 days, and ending at 21.75 days after setting. Three groups of chicks were sampled: those that hatched first (group A, identified as hatching from day 20.25 to 20.5), those that hatched in the medium time (group B, hatching on day 21) and those that hatched last (group C, from day 21.5 to 21.75). The hatch window for the first and the last of these periods was 6 h ensuring that at least 60 chicks were sampled per period, this being, for purposes of the trial, the number of chicks required from each group. Only about 1 h was required to collect sufficient chicks in the medium period. At the end of the 6 h hatch window period for group A and 1 h for group B, a further 50 chicks from each group were removed and placed in a brooder room and were reared to 21 days. In each interval, all chicks that had cleared the shell were removed from the hatching tray but chicks that were still in their shells were not assisted. This did not take longer than 15 minutes before the next sampling period.

Within each of the hatching-time groups, chicks were placed into designated compartments in a separate hatching tray that had been divided into four compartments using Perspex dividers, two of which were supplied with Perspex feeders. Each group (A and B) had 'fed' and 'held' chicks. Group C chicks were removed from the incubator after the final six-hour window period without being transferred first to separate hatching compartments, and, together with all other chicks, were placed into brooder cages keeping groups A, B and C, and fed and held groups separated and identified. In each group the held chicks were given food only at the end of 36 h after the onset of hatching

Six of the first chicks to hatch from the held and fed compartment of group A and B were removed within the hatching time period and sacrificed immediately to measure chick weight, breast meat weight, yolk sac weight and body protein content. Thereafter, six chicks were removed at nine hour intervals after the start of each sampling time period and were slaughtered and measured in a similar manner as those removed at the start of each sampling period. This continued until end of the incubation period (Table 3.1). Group A was sampled at 0, +9, +18, +27 and +36 h after the start of the hatching period, and group B was sampled at +18, +27, and +36 h after commencement of hatching. Group C had only one sampling period before all chicks were transferred to the brooder cages, however, only held chicks were sampled as the birds in the fed group did not get a chance to feed, and as a consequence, no differences were expected between the two groups.

Table 3.1 Sampling of fed (F) and held (H) chicks hatched within 36 h (summary)

Group	A		B		C
Hatching day (d)	20.25-20.5		21		21.5-21.7
Treatment	F	H	F	H	H
Sampling time (h)					
0	6	6	0	0	0
9	6	6	0	0	0
18	6	6	6	6	0
27	6	6	6	6	0
36	6	6	6	6	6
Total	30	30	18	18	6

The sampled birds were slaughtered by cervical dislocation, weighed and the abdomen cut open to remove the yolk sac. The yolk sac was weighed and discarded, and then the actual body weight was obtained by subtracting the yolk sac weight from the initial body weight. The breast muscle with bone was removed, weighed, and put back with the whole bird into a labelled plastic for mincing. Each bird was minced with feathers and placed in a labelled plastic bag, and stored in the freezer for chemical analysis. Each sample was freeze dried, then analysed for crude protein (CP) content using Kjeldahl Nitrogen method (AOAC, 1990).

3.2.4 Sampling and measurements to day 21

At day 21, six chicks from each group of fed and held groups were sacrificed by cervical dislocation to measure body weight, breast weight (with bone) and body protein content. Each bird was de-feathered in a rotatory drum, minced using a mincing machine and placed in a closed bottle, and stored in the freezer (-20 °C) until chemical analysis was performed. Each sample was freeze dried, milled and then analysed for crude protein (CP) content with a LECO FP2000 Nitrogen analyser using the Dumas combustion method (AOAC, 2003)

The same starter feed used in the hatchery was given to birds until day 21. Both fed and held birds had *ad libitum* access to water and feed from 36 h post-hatch. Feed intake and body weight were measured weekly. Mortality was monitored and recorded daily.

3.2.5 Experimental design and statistical analysis

The three hatching-time periods used, with fed and held chickens in each period, were replicated in two incubators. The 25 chicks from each of the six treatments were transferred and allocated randomly to two brooder cages where their food intake and growth was monitored at seven day intervals to 21 days. Data were analysed using unbalanced analysis of variance to test for the effect of early feeding on bird’s performance (FI, BW, FCE and BMY) and to obtain treatment means, and simple linear regression was used to evaluate the response of breast muscle weight to time of hatch (Genstat, 14th ed., 2011.). Differences in mortality between fed and unfed chicks were determined using the Chi-square test (Wright, 2005).

3.3 RESULTS

3.3.1 Body, breast and yolk sac weight for held and fed birds at hatch (0 to 36 h)

The mean body weight, breast weight and yolk sac weight for held and fed birds at hatch are given in Table 3.2. There was no significant interaction between early feeding and sampling period for body weight and breast weight but it was significant for yolk sac weight. Early feeding did not significantly affect body weight ($P > 0.05$). Regression analysis showed that sampling time significantly affected the body weight, where birds that were sampled first had a lower body weight compared to those sampled last ($b = 0.0247 \pm 0.0247$ g/h post-hatch, $P = 0.034$). Breast meat weight did not vary due to early feeding ($P > 0.05$). A significant variation was observed between birds that were sampled at the beginning and at the end of the incubation period ($b = 0.00524 \pm 0.00133$ g/h post-hatch, $P = 0.003$), in which birds that were sampled first had lower breast weight than those sampled last. The significant interaction between early feeding and sampling time for yolk sac weight showed that birds that were sampled last and had early access to feed had the most reduced yolk sac weight as compared to those held but sampled at a same time ($P < 0.05$).

3.3.2 Body protein content for held and fed birds at hatch (0-36 h)

The mean body protein content for held and fed birds removed from hatcher at a nine-hour interval is given in Table 3.2. There was no significant interaction between early feeding and sampling time for body protein content. Early fed birds had significantly higher body protein content as compared to held birds ($P < 0.05$). Body protein content was not significantly ($P > 0.05$) different between sampling time.

3.3.3 Percentage ratio of breast weight as a proportion of body protein weight (BrW/BPW) or body weight (BrW/BW) for held and fed birds at hatch (0-36 h)

Mean breast weight as a proportion of body protein weight or body weight for held and fed birds sampled at a nine-hour interval are presented in Table 3.2. The interaction between early feeding and sampling time was not significant for BrW/BPW ratio ($P > 0.05$) but it was significant for BrW/BW ratio ($P < 0.05$). Mean BrW/BPW ratio was significantly affected by early feeding; where the ratio for fed birds was lower than that of held birds ($P < 0.05$).

Sampling time had a significant effect on BrW/BPW, as birds that were removed first from the incubator had a lower ratio compared to those removed last (b= 0.0508 ± 0.0256 g/g* h^{-1} hatch, P = 0.05). The significant interaction between early feeding and sampling time for BrW/BW showed that fed birds maintained the same BrW/BW ratio throughout the hatching period, whereas the ratio for held birds increased with sampling time ($P < 0.05$).

3.3.4 Body, breast and yolk sac weight for held and fed birds from three hatching groups, at hatch

The mean body weights, breast weights and yolk weights for held and fed birds hatched at three different times are presented in Table 3.3. The interaction between early feeding and time of hatch (group) was not significant for any of the measured variables at hatch ($P > 0.05$). Body weight at hatch was not influenced by early feeding or time of hatch ($P > 0.05$). Breast meat weight did not vary due to early feeding ($P > 0.05$). Significant variation was observed between the birds that hatched at the beginning and at the end of the incubation period where birds that hatched first had lower breast weights than those that hatched towards the end of the incubation period (b= 0.00524 ± 0.00133 g/h hatch, $P < 0.020$). The fed birds had significantly reduced yolk sac weight compared to held birds at hatch ($P < 0.05$). Time of hatch was not significant for yolk sac weight ($P > 0.05$).

3.3.5 Body protein content for held and fed birds from three hatching groups, at hatch

The mean body protein content for held and fed birds removed from three hatched groups are given in Table 3.3. There was no significant interaction between early feeding and time of hatch for body protein content ($P > 0.05$). Early fed birds had significantly higher body protein content as compared to held birds ($P < 0.05$). Body protein content was not significantly different between sampling time ($P > 0.05$).

3.3.6 Percentage ratio of breast muscle weight as proportion of body protein weight (BrW/BPW) or body weight (BrW/BW) from three hatching groups, at hatch

The mean BrW/BPW and BrW/BW ratio for held and fed birds hatched at different times are given in Table 3.3. The interaction between early feeding and time of hatch was not significant for both BrW/BPW and BrW/BW ratio at hatch ($P > 0.05$). The BrW/BPW ratio was significantly affected by early feeding ($P < 0.05$), where BrW/BPW ratio for fed birds was lower than that of held birds. The time of hatch had a significant effect on BrW/BPW ratio as birds that hatched first (group A) had lower BrW/BPW ratio as compared to those that hatched last ($b = 0.0508 \pm 0.0256$ g/g* h^{-1} hatch, $P = 0.026$). Early feeding did not influence BrW/BW at hatch ($P > 0.05$). Time of hatch significantly affected BrW/BW ratio where group A had a lower yield compared to other groups that hatched later ($P < 0.05$).

Table 3.2 Mean body, breast muscle and yolk sac weight (g), body protein content (g/kg), and percentage ratio of breast muscle weight as a proportion of body weight (Br/BW) or body protein weight (Br/BPW) for held (H) and fed (F) birds at hatch (0 to 36 h)

Sampling time (h)	Body weight			Breast weight			Yolk sac weight			Body protein content			BrW/BPW			BrW/BW		
	F	H	Mean	F	H	Mean	F	H	Mean	F	H	Mean	F	H	Mean	F	H	Mean
0	39.2	39.8	39.5	1.46	1.31	1.38	8.12	9.08	8.60	134	123	129	28.3	26.7	27.5	3.73	3.28	3.51
9	38.1	37.2	37.6	1.40	1.34	1.37	7.18	6.90	7.04	137	127	132	26.9	28.3	27.6	3.68	3.59	3.64
18	38.1	38.6	38.3	1.38	1.44	1.41	7.32	7.69	7.50	134	131	133	27.2	28.6	27.9	3.62	3.73	3.68
27	39.5	39.5	39.5	1.45	1.49	1.47	5.95	6.90	6.42	138	135	137	26.7	28.1	27.4	3.68	3.77	3.73
36	41.2	39.9	40.6	1.53	1.56	1.55	5.25	7.79	6.52	135	128	132	27.7	30.7	29.2	3.72	3.91	3.82
Mean	39.5	39.0		1.46	1.46		6.76	7.67		136	129		27.4	28.5		3.69	3.66	
RMS		8.98			0.0265			3.58			90.1			9.01			0.0929	
P-value		0.769			0.377			0.047			0.634			0.317			0.041	
Main effect										P-values								
Early feeding		0.804			0.648			0.008			0.002			0.008			0.421	
Sampling time (h)		0.034			0.003			0.029			0.123			0.05			0.001	

RMS: Residual mean square; n= 48 for fed, n= 54 for held; n= 12 for 0 h, n= 12 for 9 h, n= 24 for 18 h, n= 24 for h, n= 30 h

Table 3.3 Mean body, breast and yolk sac weight (g), body protein content (g/kg), and percentage ratio of breast muscle weight as a proportion of body protein weight (BrW/BPW) or body weight (BrW/BW) at hatch for held (H) and fed (F) birds from three hatching groups

	Body weight			Breast weight			Yolk sac weight			Body protein content			BrW/BPW			BrW/BW		
Group	F	H	Mean	F	H	Mean	F	H	Mean	F	H	Mean	F	H	Mean	F	H	Mean
A	38.8	38.7	38.8	1.43	1.41	1.42	6.68	7.12	6.90	137	131	134	27.0	28.0	27.5	3.68	3.65	3.67
B	39.9	39.7	39.8	1.47	1.49	1.48	6.41	7.78	7.10	134	129	132	27.6	29.3	28.5	3.68	3.75	3.72
C	*	39.3	39.3	*	1.57	1.57	*	8.97	8.97	*	127	127	*	31.6	31.6	*	4.01	4.01
Mean	39.4	39.2		1.45	1.49		6.55	7.96		136	129		27.3	29.6		3.68	3.80	
RMS		9.40			0.0292			3.94			89.7			8.93			0.102	
P-value		0.907			0.651			0.253			0.903			0.533			0.480	
Main effect									P-values									
Early feeding		0.804			0.648			0.008			0.002			0.008			0.421	
Group		0.237			0.02			0.066			0.174			0.026			0.042	

RMS: Residual mean square; Group: A= hatched first, B= hatched at medium time, C= hatched last; n= 48 for fed, n= 54 for held; n= 60 for group A, n= 36 for group B, n= 24 for group C;*= birds were not sampled because they were not fed

3.3.7 Body weight gain, feed intake and feed conversion efficiency for held and fed birds to day 21

The mean body weight gain, feed intake and feed conversion efficiency for held and fed birds from group A, B and C at day 21 are presented in Table 3.4. There were no significant interactions between early feeding and group (time of hatch) for any of the measured variables (P > 0.05). Body weight gain for held birds was significantly lower as compared to that of fed birds (P < 0.05). Feed intake and feed conversion efficiency at day 21 were not significantly affected by early feeding (P > 0.05). Body weight gain, feed intake and feed conversion efficiency at day 21 was not significantly different between the different hatching periods (P > 0.05).

Table 3.4 Mean body weight gain (g), feed intake (g) and feed conversion efficiency (g gain/ g feed) for fed (F) and held (H) birds from three hatched groups at 21 days.

	Body weight gain			Feed intake			Feed conversion efficiency		
Group	F	H	Mean	F	H	Mean	F	H	Mean
A	816	797	807	1082	1099	1091	755	726	740
B	845	801	823	1080	1072	1076	784	748	766
C	*	780	780	*	1047	1047	*	745	745
Mean	831	793		1081	1073		769	740	
RMS		908			1865			1209	
P-value		0.424			0.579			0.852	
Main effect					P-values				
Early feeding		0.013			0.671			0.079	
Group		0.125			0.252			0.371	

RMS: Residual mean square; Group: A= hatched first, B= hatch at medium time, C= hatched last; n= 18 for fed, n= 12 for held; n= 12 for group A, n= 12 for group B, n= 6 for group C; *= birds were not sampled because they were not fed

3.3.8 Breast weight, body protein content, and percentage ratio of breast muscle weight as a proportion of body protein weight (BrW/BPW) or body weight (BrW/BW) for fed and held birds from three hatched groups at day 21

The mean breast weight and body protein content for fed and held birds, at day 21 of age, hatched from three groups are given in Table 3.5. There was no significant interaction between early feeding and group (time of hatch) for both breast weight and body protein content at day 21 ($P > 0.05$). Breast weights for fed birds were significantly heavier than those of held birds ($P < 0.05$), but the body protein content was significantly ($P < 0.05$) higher for held birds. When breast weight was regressed against time of hatch, the breast weight decreased with time (b= -11.7 ± 4.08 g/h, P = 0.008), where chicks that hatched first (Group A) had heavier breast weight compared those that hatched last. Body protein content remained constant over all hatching times.

There was no significant interaction between early feeding and group (time of hatch) for both BrW/BPW and BrW/BW ($P > 0.05$) (Table 3.5). The BrW/BPW ratio was significantly ($P < 0.05$) affected by early feeding as early-fed birds had higher ratio compared to delayed-fed birds. The BrW/BPW yield was not significantly affected by time of hatch (P = 0.061). The BrW/BW ratio was not significantly affected by early feeding ($P > 0.05$) but it was significantly affected by time of hatch where the ratio for birds that hatched first was higher than those hatched last (b= -0.845 ± 0.227 g/g* h^{-1} hatch, $P < 0.001$).

3.3.9 Mortality

Mortality was not significantly affected by early feeding ($P < 0.05$); both fed and held had approximately 1.3 and 0.84 % mortality, respectively and it was observed in the first week of life.

Table 3.5 Mean breast weight (g), body protein content (g/kg), and percentage ratio of breast weight as a proportion of body protein weight or body weight for held and fed birds from three hatched groups at day 21

	Breast weight			Body protein content			BrW/BPW			BrW/BW		
Group	F	H	Mean	F	H	Mean	F	H	Mean	F	H	Mean
A	194	173	183	156	162	159	136	131	133	21.2	21.2	21.2
B	186	165	175	144	153	149	143	132	138	20.5	20.3	20.4
C	*	159	159	*	161	161	*	121	121	*	19.5	19.5
Mean	190	165		150	159		139	128		20.9	20.3	
RMS	202			39.1			54.6			0.961		
P-value	0.919			0.529			0.327			0.710		
Main effect												
Feeding time	< 0.001			0.007			0.002			0.236		
Group	0.024			< 0.001			0.061			< 0.001		

RMS: Residual mean square; Group: A= hatched first, B= hatch at medium time, C= hatched last; n= 18 for fed, n= 12 for held; n= 12 for group A, n= 12 for group B, n= 6 for group C; *= birds were not sampled because they were not fed

3.4 DISCUSSION

One aim of this study was to evaluate early feeding as a means to improve breast muscle weight in broiler chickens. Early access to feed did not increase body weight or breast muscle weight of the chicks at hatch, but it significantly increased the breast muscle weight at day 21.

The lack of significant effect of early feeding on breast weight at hatch could be due to the fact that changes take place at a cellular level which cannot be measured with the scale used in this trial (Halevy *et al*., 2000). Halevy *et al*. (2000) and Sklan *et al*. (2000) only observed an effect of early feeding on body weight after 21 h of hatching, which could explain the lack of differences in body weight between fed and held birds in the current study as the birds were sacrificed within 9 h of hatching.

The reduced body weight in birds that were sampled first may be associated with increased yolk sac in these birds. This shows that these birds were slaughtered before they got the chance to utilise nutrients from the yolk sac compared to those that were sampled towards the end of the incubation period which had a chance to utilize nutrients from yolk sac, hence showed increased body weight. Time of hatch did not bring significant differences in body weight between birds that hatched first and those that hatched last, which contradicts the findings by Sklan *et al*. (2000) who reported that birds that hatched first had a reduced body weight compared to those that hatched last, and associated this behaviour with metabolism and evaporation of remnants of the amniotic and choriollantoic fluids in birds that hatched first. The evaporation in the current study could have been minimal due to water tray that was placed in the incubator during the hatching period.

The reduced yolk sac weight in birds that were removed towards the end of the incubation and had early access to feed has also been reported by previous studies (Noy *et al*., 1996; Noy & Sklan, 1998; El-Husseiny *et al*., 2008). These studies associated the reduced yolk sac weight with increased activity of the anti-peristaltic movements of the intestine in fed birds which enhanced yolk sac utilization. This could be the case in the current study, however, the intestinal growth was not measured, and therefore, this could be a subject for future study. In contrast, Bigot *et al*. (2003) did not find any difference between yolk sac weights of fed and held birds, and this could be due to access to water immediately post-hatch in both fed and held birds, which probably enhanced intestinal growth (Noy & Sklan, 1999) in both groups.

The lack of differences in feed intake and feed conversion efficiency at day 21 between fed and held birds in the current study is in contrast with earlier studies (Bigot *et al*., 2003; Juul-Madsen *et al*., 2004; El-Husseiny *et al*., 2008) which reported that delayed access to feeding reduced feed consumption throughout the experiment. The current study suggest that it is beneficial to early feed birds because feed intake remained the same between fed and held birds while body weight gain was increased in fed birds compared to held birds at day 21. Feeding birds after 36 h of starvation does not improve performance in held birds; this is because fed birds were significantly 10. 9 % (99 g) heavier in body weight than held birds at day 21 and the relative breast muscle weight was 25 g heavier than that of held birds.

The increased breast muscle weight in fed birds can be associated with increased proliferation of satellite cell at hatch (Halevy *et al*., 2000). The held birds did not withdraw nutrients from the breast muscle when they were starved as was hypothesised in the current study. This is because the breast muscle weight in held birds was not reduced as birds spent a longer time in the hatchery without feed; instead it was increased compared to those sampled first. Therefore the reduced breast muscle weight in held birds can be associated with the slow utilisation rate of yolk sac at hatch in which these birds were dependent on for nutrients. These results indicate that early access to feed of hatched chicks result in considerable performance benefits. Therefore, in commercial hatcheries this could be carried out by simply placing feed in the hatching trays. Hatching time did not significantly affects any bird's performance other than breast muscle weight which was increased in birds that were hatched first compared to those hatched towards the end of the incubation period at day 21, and this was associated with increased body protein in these birds. This suggest that birds in group A are older than those hatched last in terms of growth, and it could happen in future that time of hatch will need attention.

The reduced BrW/BPW ratio in fed birds at hatch was due to increased body protein weight in these birds while the breast muscle weight between fed and held birds remained similar. At day 21, this ratio was increased by 0.11 for fed birds and this could be associated with increased breast muscle weight in these birds at day 21. This increase suggests that the allometric relationship between breast muscle weight and body protein weight can be manipulated through early feeding. Therefore, birds with high BMY can be produced through giving hatched birds early access to feed.

Mortality for both fed and held birds was less than 1.5 % and this is in agreement with the findings by Noy & Sklan (1999) who reported a mortality of less than 2 % in all birds (fed and held) throughout the trial.

3.5 CONCLUSION

Early access to feed of hatched chicks increased BMY at day 21. The increased breast meat yield in early-fed birds was not associated with increased body protein weight; therefore it was hypothesised that it was due to increased activity of satellite cell proliferation. Availability of feed after 36 h of starvation does not compensate for the reduced early growth. Delayed-fed birds do not withdraw nutrients from the breast muscle to sustain growth after hatch. Determining the economic importance of early feeding would validate the advantage of using this feeding technique.

CHAPTER 4

THE EFFECT OF DIETARY PROTEIN INTAKE ON BREAST MEAT YIELD OF BROILERS REARED ON DIFFERENT DAYLENGTHS

4.1 INTRODUCTION

Breast meat yield has been shown to be reduced when birds are exposed to daylengths shorter than 20 h whereas most of other important biological parameters showed improved performance (Lewis *et al*., 2009a). Whilst continuous lighting increases BMY, it also increases physiological stress levels and mortality (Lewis *et al*., 2008). The new welfare regulations in the European Union stipulate that birds be reared with a minimum of 8 h of darkness, six of which must be un-interrupted within a 24-hour cycle (European Commission, 2000). As yet, no explanation has been given for the low BMY of broilers reared on short daylengths, but because of the welfare benefits of such a management procedure, it is worth finding a way of overcoming this hurdle.

The study by Danisman & Gous (2007) has shown that body protein content is positively associated with BMY and that this relationship is constant across genotypes and sexes. It is only when low protein feeds are offered to broilers that the relationship between body protein and breast meat yield changes (Danisman, 2009). Under these circumstances, excess energy is consumed by birds in their attempt to consume sufficient protein which results in an increase in fat deposition in the various parts of the body (Gous *et al*., 1990; Smith & Pesti, 1998) causing the allometric relationship of BMY with body protein to differ from that where high protein feeds are offered (Kemp *et al*., 2005). It is possible that the reduced feed intake of broilers reared on short daylengths (Ingram *et al*., 2000) may result in less dietary protein being consumed than is necessary to meet the potential body protein growth of broilers, and this could be the cause of the lower BMY in birds reared on short daylengths. Alternatively, the lower energy usage by birds on short daylengths would result in a surplus which would be deposited as body lipid (Lewis & Gous, 2007). Therefore, at the same body weight as broilers on long daylengths, broilers on short daylengths would have more body lipid and hence less body protein, and as a result, less breast meat.

The hypothesis tested in this trial was that BMY would improve in broilers reared on short daylengths if higher levels of dietary protein were fed. In order to test this hypothesis, feeds ranging in dietary protein content were fed to broilers reared under a range of daylengths, with growth rate, food intake, BMY and body protein content being measured.

4.2 MATERIALS AND METHODS

4.2.1 Study site and housing management

A total of 3200 day-old Ross 308 International broiler chicks were reared in 8 light-tight rooms. Each room was divided into four pens using a mesh fence as a barrier. Each pen was populated with 100 feather-sexed male or female chicks. On the first day all chicks received continuous lighting with the lighting treatments being implemented from the second day of age. Illumination was provided by 11W warm-white compact fluorescent lamps. Heating was provided initially using gas spot-brooders which were placed over two pens on either side of each room. The room temperature was set initially at 32 °C and it was decreased by 3 °C at seven days and then linearly to reach 20 °C by 21 days of age. The Animal Ethics Committee approved, in 2010, the use of broiler chickens for this study (reference number: 086/10/Animal).

4.2.2 Feed treatments and feeding procedure

The four experimental diets used in this trial were formulated to contain 85, 100, 115 and 130 % of the Aviagen recommendations for dietary CP (Aviagen, 2009). Two basal feeds (lower and higher protein levels) were mixed at a commercial mill (Table 4.1), and the chemical composition for these feeds is given in Table 4.2. The two intermediate levels of protein were produced by appropriately blending the high and low protein basal feeds (Table 4.3). The basal feeds were sampled after mixing for the analysis of apparent metabolisable energy (AME), using the method of Fisher and McNab (1987), and digestible amino acid content using a Waters amino acid analyzer (AOAC, 2003). The experimental diets were provided in a crumbled form for the first two weeks, and in a pelleted form until the end of the trial. Water was supplied in chick founts for the first week and in suspended bell drinkers thereafter. Feed and water were supplied *ad libitum* for the entire trial, and drinkers were cleaned daily.

Table 4.1 Ingredient composition (g/kg) for low and high protein basal starter and finisher feeds used in the trial

	Starter		Finisher	
	LP	HP	LP	HP
Maize	591	320	730	553
Wheat middlings			23.2	
Soybean full fat	300	300	202	300
Soybean 46	60.8	255		92.3
Fish meal 65		45.0		
L-lysine HCl	3.00	2.40	2.00	1.80
DL methionine	1.60	2.3	0.50	1.90
L-threonine	0.80	1.00	0.40	0.70
Choline chloride 60%	0.80		1.40	0.70
Vit+min premix	1.50	1.50	1.50	1.50
Limestone	17.1	12.8	18.1	17.0
Salt	2.00	1.20	2.00	2.20
Monocalcium phosphate	16.5	12.6	16.5	16.5
Sodium bicarbonate	2.40	1.80	2.50	2.00
Oil - sunflower	2.10	44.4		10.9

Table 4.2 Chemical composition for the high and low protein basal feeds used in the formulation (calculated)

	Starter		Finisher	
	LP	HP	LP	HP
AMEn chick (MJ/kg)	12.60	12.60		
AMEn adult (MJ/kg)			13.0	13.0
Crude protein (g/kg)	194	290	143	204
Lysine (%)	11.6	17.4	7.60	11.4
Methionine (%)	4.20	6.20	2.70	4.60
Methionine+cystine (%)	6.90	9.70	5.00	7.40
Threonine (%)	7.10	10.7	5.00	7.40
Tryptophan (%)	1.80	2.90	1.20	2.00
Arginine (%)	11.7	18.5	8.00	12.6
Isoleucine (%)	7.50	11.9	5.20	8.00
Leucine (%)	15.8	20.9	13.1	16.4
Histidine (%)	4.70	6.90	3.70	5.00
Phenylalanine (%)	7.90	11.7	5.70	8.40
Tyrosine (%)	6.10	9.50	4.40	6.60
Valine (%)	8.40	12.6	6.10	8.90
Calcium (%)	10.0	10.0	10.0	10.0
Avail. Phosphorous (%)	5.00	5.00	5.00	5.00

The amino acid composition was calculated on a digestible basis

Table 4.3 Treatment blending proportions and protein content (g/kg) of starter and grower feeds used

Treatment	HP	LP	Protein in starter	Protein in grower
1	0	100	194	143
2	33	67	226	164
3	67	33	258	184
4	100	0	290	204

4.2.3 Measurements

On arrival, all chicks were weighed per box to obtain their mean initial body weight. Thereafter a random sample of the birds in each pen was weighed at seven-day intervals and these values were used to calculate body weight gain (BWG). On the same day, feed remaining in the troughs was measured from which feed consumption was calculated as the difference between feed offered and rejected. Feed intake (FI) was corrected for mortality. Feed conversion efficiency (FCE) was calculated by dividing average daily gain by average daily feed intake. Mortality was recorded daily.

At 35 days, three birds from each pen were sacrificed for measurement of physical and chemical characteristics. Body weight of each bird was recorded three times, once before being electrically stunned and exsanguinated, the second time after bleeding had ceased, and the third time after the feathers had been removed using a rotatory drum. Breast meat (with bone and without skin), thigh, drum and wing (with skin and bone) were dissected and weighed and then all parts of each bird, other than feathers, were sealed in separate plastic bags, marked and stored in a freezer until mincing took place. Each bird was minced twice using a medium size mincing machine, then a sample of approximately 500 g was taken from each carcass for subsequent chemical analysis. Each carcass sample was freeze-dried and then milled using a Retsch ultracentrifugal mill. The crude protein (CP) content of each milled sample was analysed with a LECO FP2000 Nitrogen analyser using the Dumas combustion method, approved (AOAC, 2003). Gross energy (GE) content was determined using a DDS isothermal CP 500 bomb calorimeter. Lipid (L) content was calculated from GE using the equation $L = -0.8754 + 0.04754*GE$ (University of KwaZulu-Natal, unpublished). To determine the ash content, samples were placed in a furnace for four hours at 550 °C (AOAC, 2003).

Because moisture may have been re-absorbed by the samples between being freeze-dried and being weighed for CP, GE and ash determinations, a duplicate sample of each milled carcass was weighed and placed in an oven at 90 °C overnight to determine the amount of accumulated moisture. A correction was made for this water uptake when calculating the protein, lipid and ash contents of the carcass.

4.2.4 Experimental design and statistical analysis

A factorial design was used, with main effects being dietary protein and lighting. Dietary protein had four levels containing 85, 100, 115 and 130 % of the Aviagen recommendations. Four lighting treatments 12L: 12D, 16L: 8D, 20L: 4D and 24L: 0D were randomised between rooms, with each lighting treatment being replicated twice. Data were analysed using analysis of variance to determine the significance of the main effects at the 5 % level. Simple linear regression with groups was used to evaluate the response to dietary protein content and daylength, using Genstat, 14^{th} ed. (2011). Birds were sexed at the hatchery but to allow for the maximum number of replications of the main effects (light and dietary protein), sex was not included in the statistical analysis and birds were treated as "as-hatched". The Chi-square test was used to determine the effects of photoperiods and dietary protein content on mortality (Wright, 2005).

4.3 RESULTS

4.3.1 Body weight gain, feed intake, feed conversion efficiency and mortality

The mean results for BWG, FCE and FI are presented in Table 4.4. There were no significant interactions between lighting programme and dietary protein content for any of the measured variables ($P > 0.05$). Body weight gain to day 35 was unaffected by both dietary protein content (b= 0.84 ± 1.12 g/g protein, $P = 0.459$) and light (b= 5.78 ± 5.46 g/h light, $P = 0.299$). Feed conversion efficiency increased significantly with dietary protein content up to day 21 only (b= 0.626 ± 0.266 g gain/g feed intake * g $protein^{-1}$, $P = 0.025$). Feed intake to day 35 was not influenced by light (b= 7.61 ± 7.18 g/h light, $P = 0.298$) or by dietary protein content (b= -0.41 ± 1.4 g/g protein, $P = 0.782$).

The birds on 24 h had a significantly ($P < 0.05$) higher mortality (5.58 %) compared with those on other lighting programmes which were not significantly different from each other. Dietary protein content did not significantly affect mortality ($P > 0.05$).

Table 4.4 Mean body weight gain (g), feed intake (g) and feed conversion efficiency (g gain/g feed intake) of birds fed four levels of dietary protein under four lighting treatments from day old until day 35

					Body weight gain					
			21					35		
Dietary protein (g/kg)	194	226	258	290	Mean	143	164	184	204	Mean
Light (h)										
12	941	866	845	942	899	1363	1363	1389	1450	1391
16	866	857	866	911	875	1311	1402	1405	1425	1386
20	889	883	914	935	905	1359	1397	1465	1579	1450
24	913	947	952	953	941	1440	1461	1406	1481	1447
Mean	902	888	894	935		1368	1406	1416	1484	
RMS			2534					29879		
P-value			0.771					0.999		
					Feed Intake					
			0-21					22-35		
Light (h)										
12	1200	1107	1006	1147	1115	2488	2363	2244	2492	1685
16	1129	1088	1057	1071	1086	2409	2401	2424	2357	1669
20	1159	1163	1117	1139	1144	2418	2467	2463	2490	1729
24	1190	1118	1131	1139	1145	2531	2503	2423	2453	1737
Mean	1169	1119	1078	1124		2462	2434	2389	2448	
RMS			4664					54780		
P-value			0.854					0.995		
					Feed conversion efficiency					
			21					35		
Light (h)										
12	750	746	799	788	770	548	576	619	583	582
16	730	749	780	811	768	544	584	579	604	578
20	731	725	781	783	755	562	565	593	633	588
24	732	809	805	799	786	569	583	579	601	583
Mean	736	757	791	795		556	577	592	605	
RMS			580					643		
P-value			0.354					0.504		

RMS: Residual mean square; n= 32

4.3.2.1 Breast meat yield

Mean breast muscle weights at 35 d from birds fed four levels of dietary protein under four lighting schedules are given in Table 4.5. There was a significant interaction between dietary protein content and lighting programme for breast muscle weight ($P < 0.001$). The breast weights for birds that were fed 164, 184 and 204 g protein/kg dietary protein content increased by 3.55 ± 1.63 g/ h light with increasing daylengths but those that were fed 143 g protein/kg feed had a reduced (b= -4.61 ± 3.05 g/h light) breast weight as lighting hours increased (Figure 4.1). Breast weight was increased when dietary protein content was increased under both short and long daylengths, except for those on 12 h which showed a reduced breast meat weight when high levels of protein were fed (Figure 4.1). Birds on 20 h of light and fed the highest protein content had significantly heavier breast weights compared to those on 12, 16 and 24 h (Table 4.5).

Birds on three of the four dietary protein levels had the same constant term and a positive slope when breast meat weight was regressed against daylength, but breast meat weight of birds on the lowest protein level (143 g protein/kg feed), although having the same constant term, declined as daylengths increased (Table 4.7 and Figure 4.1). When regressed against dietary protein content (Figure 4.2) breast meat yield increased in birds on 20 h of light but the slope either remained constant or decreased slightly for birds reared on any of the other lighting treatments.

The BrW/BPW ratio was not significantly affected by neither the interaction nor protein content or lighting programme ($P > 0.05$), where as BrW/BW ratio was significantly affected by the interaction of dietary protein content and lighting programme ($P = 0.029$) where the birds on 20 h of light fed highest protein diet showed significant higher yield compared to those on other treatments (Table 4.6).

4.3.2.2 Thigh weight

Mean thigh weights for broilers at 35 days are shown in Table 4.5. The interaction between feed and lighting programme was significant for thigh weight, which increased with increasing daylength when birds were fed dietary protein levels of 164, 184 and 204 g protein/kg feed, but birds on the lowest protein (143 g protein/kg feed) had reduced thigh weights as daylength was increased ($P = 0.045$). There was no significant effect of dietary protein content (b= 0.123 ± 0.0888 g/g protein, P = 0.154) or light (b= -0.0650 g/hr light ± 0.440, P = 0.884) on thigh weight.

When thigh weights were regressed against daylength, with dietary protein levels as the factor (or group), the resultant coefficients (constant term and slope) were not significantly different between protein levels (Table 4.7). However when these were regressed against dietary protein level, and daylength used as a group, birds on 20 h had a lower constant term than the other treatments and a positive slope, whereas the thigh weights declined on the other treatments as daylength increased.

3.3.2.3 Drum weight

There was no significant interaction between feed and lighting for drum weight (Table 4.5). Dietary protein content (b= 0.0595 ± 0.0717 g/g protein, P = 0.408) and lighting programme (b= -0.0245 ± 0.364 g/h light, P= 0.503) did not significantly affect the drum weight. The constant terms and slopes for drum weight were not significantly different (P > 0.05) between any of the treatments (Table 4.7).

4.3.2.4 Wing Weight

Mean wing weights for birds fed four levels of dietary protein content and reared under four lighting treatments to day 35 are given in Table 4.5. There was no interaction found between treatments for wing weight. Dietary protein content (b= 0.116 ± 0.0519 g/g protein, P= 0.0280) was significant for wing weights but not light (P > 0.05). Birds given high protein feed had heavier wing weights than those on low protein feed. The regression between wing weight and lighting programme or dietary protein content showed that neither the constant term nor the slope differed significantly between levels of the two factors (Table 4.7).

Table 4.5 Mean breast, thigh, drum and wing weights (g) of broilers at 35 d fed four levels of dietary protein and subjected to four lighting programmes.

	Feed protein content (g/kg)				
Light (h)	143	164	184	204	Mean
			Breast		
12	429	332	385	383	395
16	380	391	385	433	397
20	364	456	439	532	447
24	373	416	449	413	413
Mean	379	419	414	440	
RMS			3793		
P-value			0.004		
			Thigh		
12	132	122	118	126	123
16	121	127	122	122	123
20	112	132	133	147	131
24	114	125	123	121	121
Mean	119	126	124	129	
RMS			359		
P-value			0.323		
			Drum		
12	109	107	98.0	120	108
16	112	112	104	117	111
20	104	116	108	111	110
24	98.2	118	105	103	106
Mean	105	113	104	113	
RMS			251		
P-value			0.532		
			Wing		
12	87.0	86.7	80.8	94.9	86.3
16	86.4	91.6	87.0	95.8	90.2
20	85.5	87.4	90.8	94.6	89.6
24	81.0	95.8	86.7	90.8	88.6
Mean	83.9	90.4	86.3	94.0	
RMS			139		
P-value			0.816		

RMS: Residual mean square, n= 96

Table 4.6 Mean breast weight (g) as a proportion of body weight (g) (BrW/BW) or body protein weight (g) (BrW/BPW) of broilers fed one of four protein diets at different lighting programmes

	BrW/BPW				
Protein	143	164	184	204	Mean
Light (h)					
12	1.41	1.16	1.32	1.19	1.27
16	1.30	1.23	1.30	1.29	1.28
20	1.27	1.40	1.40	1.50	1.39
24	1.30	1.29	1.40	1.29	1.32
Mean	1.32	1.27	1.36	1.32	
RMS			0.0278		
P-value			0.16		
	BrW/BW				
Light (h)					
12	0.194	0.161	0.196	0.175	0.182
16	0.180	0.182	0.192	0.200	0.189
20	0.182	0.202	0.200	0.222	0.202
24	0.189	0.190	0.212	0.197	0.197
Mean	0.186	0.184	0.200	0.199	
RMS			0.000494		
P-value			0.026		

RMS: Residual mean square, n= 96

Table 4.7 Constant terms and regression coefficients (± standard error of the mean) for breast, thigh, drum and wing weights with dietary protein content and lighting as explanatory variate. Where no difference exist between levels of a factor only one value is given for the constant term and/ or regression coefficient, at day 35

Group = Feed protein

	Constant term				Regression coefficient			
Protein	143	164	184	204	143	164	184	204
Breast		354			-4.61		4.77	
		± 56.7					± 3.05	
Thigh		123					0.312	
		± 16.3					± 0.876	
Drum		125					-1.044	
		± 12.9					± 0.695	
Wing		100					0.093	
		± 9.67					± 0.107	

Group = Light

	Constant term				Regression coefficient			
Light	12	16	20	24	12	16	20	24
Breast	459	459	29.0	459	-0.445	-0.445	2.404	-0.445
		± 100					± 0.573	
Thigh	144	144	38.6	144	-0.110	-0.110	0.531	-0.110
		± 29.0					± 0.166	
Drum		89.0					0.113	
		± 25.7					± 0.147	
Wing		71.1					0.0930	
		± 18.7					± 0.107	

Table 4.8 Parameter estimates for the two-way interactions between dietary protein content and lighting programme on breast meat weightof broilers at day 35, compared with the reference level: Feed 204

Parameter	estimate	s.e.	t(88)[1]	t pr.[2]
Constant	354.1	56.7	6.25	<0.001
Light	4.77	3.05	1.56	0.122
Feed 143	115.1	80.1	1.44	0.154
Feed 164	-97.6	80.1	-1.22	0.226
Feed 184	-50.4	80.1	-0.63	0.531
Light x Feed 143	-9.38	4.32	-2.17	0.033
Light x Feed 164	3.13	4.32	0.72	0.471
Light x Feed 184	1.37	4.32	0.32	0.752

[1]n = 88: [2]pr. = Probability of an estimate

Table 4.9 Estimates of parameters for the two-way interactions between dietary protein content and lighting programme on breast weight of broiler at day 35, compared with the reference level: Light 12

Parameter	estimate	s.e.	t(88)[1]	t pr.[2]
Constant	459	100	4.58	<0.001
Feed	-0.445	0.573	-0.78	0.439
Light 16	-193	142	-1.36	0.177
Light 20	-430	142	-3.03	0.003
Light 24	-180	142	-1.27	0.209
Feed x Light 16	1.197	0.81	1.48	0.143
Feed x Light 20	2.849	0.81	3.52	<0.001
Feed x Light 24	1.209	0.81	1.49	0.139

[1]n = 88: [2]pr. = Probability of an estimate

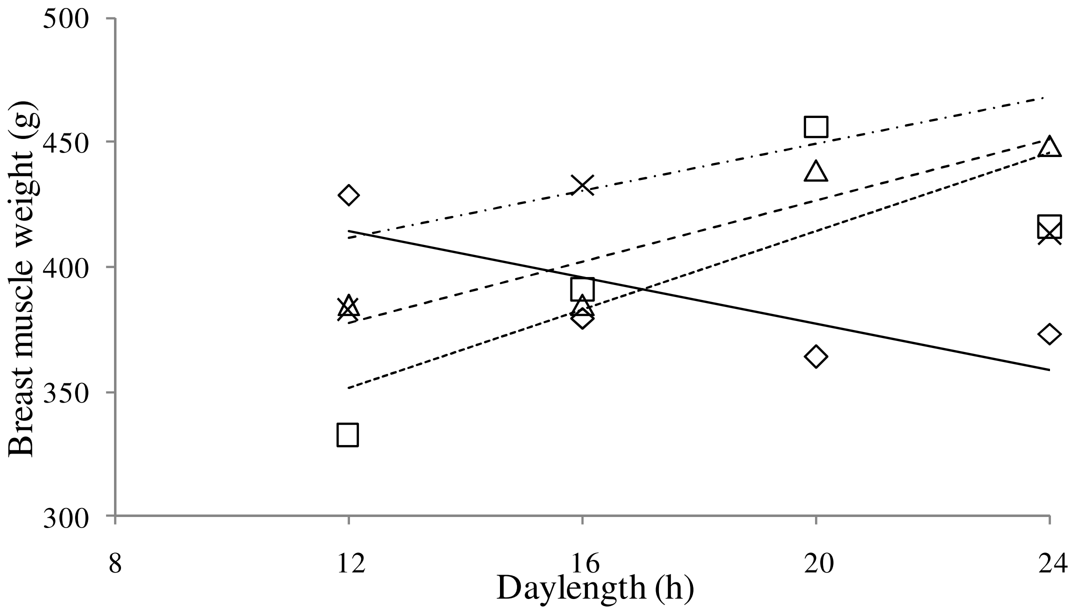

Figure 4.1 Response in breast meat weight under different daylengths separated between the four protein contents used**.** Protein 143_____◊; Protein 164………□; Protein 184------- Δ; Protein 204-.-.-.-.-.X

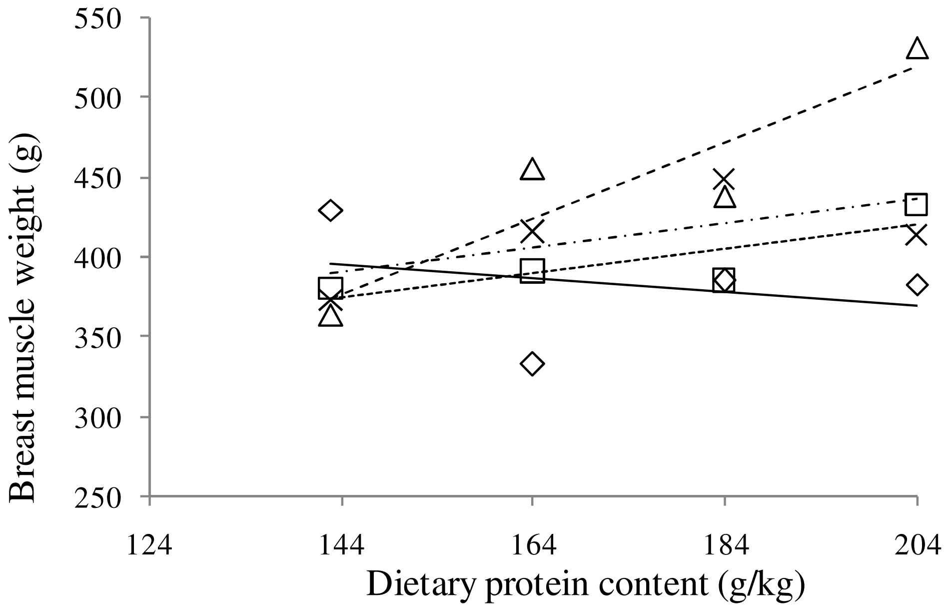

Figure 4.2 Response in breast muscle weight to increasing levels of dietary protein separated between the different daylengths used. Light 12____◊; Light 16…….□; Light 20------- Δ; Light 24-.-.-.-.-X

4.3.3 Body composition for birds fed different levels of dietary protein under different daylengths at 35 days of age

The body protein, lipid, moisture and ash content for birds fed four different levels of dietary protein and subjected to four lighting treatments are presented in Table 4.10. There was no significant interaction between lighting programme and dietary protein content for body protein, lipid, moisture or ash content ($P > 0.05$). Body protein content increased by 0.136 (± 0.038) g/kg * gprotein^{-1} as dietary protein content increased. The body lipid content decreased significantly by -0.696 (± 0.089) g/kg * g protein^{-1} as dietary protein content increased. The moisture content increased as dietary protein content increased (b= 0.540 ± 0.0923 g/kg *g protein^{-1}, $P < 0.001$). Lighting programme had no significant effect ($P > 0.05$) on any of the chemical components except for body protein content which increased with increasing daylength.

4.4 DISCUSSION

The lack of differences between the light programmes on BWG may be associated with lack of difference in feed intake for the same factor. This suggests that the birds on short daylengths either eat more during the daylight or they may have learnt to eat in the dark (Lewis *et al*., 2009a) to compensate for the growth limited by reduced lighting hours. This contradicts findings by Classen (2004) who reported increased body weight gain in birds reared on long daylengths at day 35 and associated this behaviour with increased feed intake. The similar feed intake between daylengths shows that feed consumption in birds is not only dependent on light duration but also on the first limiting nutrient.

Table 4.10 Mean content of body protein, lipid, moisture and ash (g/kg) for birds fed four levels of dietary protein on different lighting treatments to 35 days of age

	Feed protein content (g/kg feed)				
Light (h)	143	164	184	204	Mean
			Body protein content		
12	138	140	148	146	143
16	139	147	147	155	147
20	143	145	144	148	145
24	149	148	152	153	150
Mean	142	145	148	150	
RMS			71.2		
P-value			0.733		
			Body lipid content		
12	165	133	116	121	134
16	171	146	135	117	142
20	153	145	132	120	138
24	153	143	123	116	134
Mean	160	142	126	118	
RMS			403		
P-value			0.721		
			Body moisture content		
12	664	683	700	701	687
16	644	675	679	689	672
20	672	680	689	692	683
24	666	673	689	694	680
Mean	661	678	689	694	
RMS			426		
P-value			0.896		
			Body ash content		
12	24.8	26.8	26.3	26.1	26.0
16	26.3	25.8	26.0	26.6	26.2
20	26.9	26.0	26.7	25.7	26.3
24	28.4	26.3	26.1	26.1	26.7
Mean	26.6	26.3	26.3	26.1	
RMS			6.11		
P-value			0.685		

RMS: Residual mean square; n= 96

It was expected that feed intake between birds fed on low and high protein diets would differ but this was not the case. However, this can be used to explain the similar BWG between birds that were fed low and high protein diet. The similar BWG might be of different composition, this because the birds on low protein diet had increased body fat content compared to those on high protein diet. This could be the reason for the similar body weight between birds on low and high protein diets, the fat deposited probably contributed to body weight in birds fed low protein. Feeding high protein diets increased FCE at day 21 compared to birds fed low protein diets, but by day 35 birds fed low protein had similar feed efficiency as those fed high protein diets. Therefore, feeding high protein diets to young birds may enhance growth (Kemp *et al*., 2005), but from a commercial perspective this is not beneficial since both birds on low and high protein diets had similar body weight gains at slaughter.

Dietary protein content did not affect mortality and this agrees with earlier findings (Corzo *et al*., 2005; Kidd *et al*., 2005; Brickett *et al*., 2007) but contradict findings by Scott (2002) who reported that increased levels of protein were related to mortality. The difference between the current study and that of Scott (2002) could be that the protein levels used in the study of Scott (2002) were higher than those used in the current study, which possible caused metabolic disorders. The birds on 24 h daylengths had 5.58 % mortality which almost the same as that reported by Scott (2002) which was 5.66 % for birds on 23 h of light. The increased mortality could have been due to that birds on longer daylengths did not get to time sleep so physiological stresses were increased (Classen, 2004).

Feeding increased levels of dietary protein content to broilers exposed to short daylengths did not improve BMY. Birds on 12 h fed the high protein diet had a reduced BMY compared to those fed low protein diet. This shows that birds on high protein feed did not convert all the protein consumed into muscle. In this study birds reared on 20 h daylengths and high protein diets showed the highest BMY. This warrants more research to investigate potential ways of improving BMY on broilers reared on shorter daylengths. The results of this study suggest that compliance with the new EU welfare regulation (use of less than 18 h of light) with the current nutritional recommendations will reduce BMY. The decreasing breast weight as daylengths increased in birds fed dietary protein 143 could be associated with high activity (although this was not measured) in long daylengths which probably increased the demands for maintenance so the consumed feed was used for maintenance hence less energy was available for muscle growth. The breast muscle weight for birds reared on 16 h was slightly increased when high protein diets were fed; therefore, it would be interesting to determine protein diet that can give optimal breast muscle weight on 16 h.

Thigh weight was heavier when birds were fed high protein diet at 20 h of light. Drum weight was unaffected by dietary protein, but wing weights increased with the increasing levels of dietary protein. This contradicts the findings of Brickett *et al*. (2007) who reported that drum and wing weights at day 35 were heavier on low protein diet. It may be possible that the increased drum and wing weight in the study of Brickett *et al*. (2007) was not made of lean muscle only but intramuscular fat which was probably increased may have contributed to the weight.

Birds on low protein diet deposited more fat because there was not sufficient protein consumed hence the excess energy was deposited as fat. This is consistent with the findings by Bartov & Plavnik (1998), Smith & Pesti (1998), Kidd *et al*. (2004) and Corzo *et al*. (2005); however, their conclusion were based on the abdominal fat content. The birds that had increased body fat had reduced body protein content; therefore, less lean meat was produced.

4. 5 CONCLUSION

The increase in dietary protein content did not allow broilers reared on short days to match the BMY observed in those reared on long days as was assumed in the hypothesis. The BMY for birds reared on 12 h was not improved when these birds were fed high protein diets, and higher yields were obtain from birds fed high protein diets on 20 h. Further, feeding low protein diets to birds on long daylengths seem to further reduce BMY. Mortality will increase if daylengths more than 20 h are used.

CHAPTER 5

GENERAL DISCUSSION AND CONCLUSION

Breast muscle is the most valuable chicken portion in the further-processing market. Therefore, in order to meet the increased demand of this commodity, techniques that affect breast muscle growth needs to be modified. This study was planned to increase BMY through giving immediate access to feed to newly hatched chicks, and through feeding high protein diets to broilers reared on short daylengths.

The increased BMY through early feeding seems to be achievable at the present time (Chapter 3). The increased breast meat yield is probably due to increased activity of satellite cell proliferation. Feeding birds after 36 h of starvation does not improve; therefore, this study encourages the supply of feed to chicks during the hatching period to obtain chicks with high BMY at slaughter age. Since increased levels of dietary protein increase performance in broilers (El-Fiky *et al.*, 2007), it would be interesting in future to determine the effect of feeding high protein diets to newly hatched chicks immediately post-hatch. Birds that were held in the incubator without feed for a longer period showed an increase in breast muscle weight compared to those sampled first, this shows that these birds did not withdraw nutrients from the breast muscle to maintain growth. Therefore, the reduced BMY in held birds can be explained through the slow utilization of nutrients from the yolk sac.

The effect of feeding high protein diets to broilers reared on short daylengths on BMY was determined in Chapter 4. Consumption of increased levels of dietary protein did not complement the reduced breast muscle weight of birds exposed to short daylengths as birds on 12 h fed high protein diet had a reduced breast muscle weight compared to those on long daylengths fed the same diet. This suggests that when birds are subjected to 12 h daylengths, they convert the consumed protein to a certain extent. Birds on 16 h had increased breast muscle weight when they were fed high protein diet but the yield was not increased as in those exposed to longer daylengths (20 and 24 h). Therefore, it would interesting to determine the dietary protein level that will optimise breast muscle weight when birds are exposed to 16 h as this is within the EU welfare regulations stipulating photoperiod. Feeding low protein diets to birds on long daylengths also reduced breast muscle weight and this could be associated with high activity of birds responding to long hours of light. This resulted in high demand for maintenance hence less energy was available for protein deposition.

Increased breast meat weights were obtained when birds were given high protein diets in 20 h daylengths.

The results from both studies showed that even when birds are genetically selected for increased BMY, nutritional and environmental factors still influence the growth potential of the bird. This was observed when allometric relationship between breast muscle weight and body protein weight or body weight was altered when treatments of the first or second experiment were tested. Information obtained from both experiments can be used to meet the market demand for increased BMY; however, the high mortality observed in long daylengths (Chapter 4) cannot be ignored. Therefore, another strategy that will reduce physiological stress in birds exposed to long daylengths still needs to be investigated.

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