Broiler Breeder Genetic Strain, Age, and Egg Size Effects on Egg Characteristics, Saleable Chick Production, Egg and Residual Yolk Sac Fatty Acid Composition, and Broiler Performance

by

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ABSTRACT

Three trials were conducted to determine the effects of genetic strain, flock age, and egg size on egg, chick and broiler quality. Hatching eggs from two modern strains (Cobb 500 and Ross 308), and three egg sizes (small, medium, and large) were collected at three flock ages (29, 45, and 59 wk). Egg characteristics, hatchability, egg and yolk sac fatty acid profiles, and broiler performance to 6 wk of age were assessed. The combined effect of strain by age produced the most differences on egg characteristics, hatchability, and broiler performance. The fatty acid profiles in eggs and yolk sacs were greatly affected by flock age, the youngest flocks differed from the other ages. This could explain the lowest broiler performance observed at this parent flock age. Only oleic acid was greater in yolk sacs than in eggs. Linolenic acid content in medium eggs was correlated to that in yolk sacs.

DEDICATION

For God, my strength and refuge For my family, my motivation and courage For Lance, my angel and twin soul

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1. GENERAL INTRODUCTION

THE CANADIAN BROILER HATCHING EGG INDUSTRY

During the late 1990's and early 2000's the hatching egg industry in Canada benefited from a steady growth in broiler production, with annual increases above the 4% target for the industry (CBHEMA, 2001). However, over the past few years, unstable market conditions have had a negative impact in the Canadian hatching egg industry: In 2003 production declined 0.6% from 2002 levels (CBHEMA, 2003).

In 2004, the first incidence of Highly Pathogenic Avian Influenza (HPAI) in Canada in 20 years hit the Frazer Valley in British Columbia; as a consequence, almost half the broiler breeders in the province were destroyed and the Canadian hatching egg production declined 3.4% from 2003 (CBHEMA, 2004). In 2005, efforts to recover from the HPAI outbreak secured a 2.0% industry growth (CBHEMA, 2005).

In 2006, there were approximately 300 farmers who produced broiler hatching eggs in Canada. The average size for a broiler breeder farm was 15,000 birds with the laying cycle beginning at 26 wk of age, and each bird laying about 150 hatching eggs during its entire production cycle (CBHEMA, 2006).

Hatching egg producers are paid based on the number of saleable chicks. The current price is 35.90¢ per saleable chick. These chicks are then shipped to broiler farms for the 6 wk grow out period (CBHEMA, 2006). In Alberta, the broiler breeder cycle is divided into three main periods: Rearing (0 to 20-22 wk of age), pre-lay (22 to 26-27 wk of age) and, laying (27 to 60 wk of age); the broiler breeders are usually kept for a period of 60 wk (Alberta Broiler Hatching Egg Industry, 2006).

KEY ASPECTS INVOLVED IN BROILER CHICK PRODUCTION

Hatching Egg Characteristics

At oviposition, a broiler hatching egg is composed of albumen (58.5%), yolk (31%) and shell/shell membranes (10.5%) (Robinson et al., 2003). In general, albumen, yolk and shell weights increase with flock age as egg weight increases (Ahn et al., 1997), however, differences in proportion of components as a result of genetic selection have been reported (Cahaner et al., 1986).

The major constituent of albumen is water (88% of total weight) with proteins being the major solid component (around 11%) (Ahn et al., 1997). Unlike the albumen, the yolk contains approximately 50% solids and its major constituents are lipids (32%) and proteins (16%) (Vieira and Moran Jr., 1999). The eggshell is primarily composed of minerals with a high content of calcium carbonate (94%) (Nakano et al., 2003). Other elements such as magnesium, phosphorus and micro minerals are present in smaller proportions (Schaafsma et al., 2000). The shell membranes have an organic matrix that contains proteins as the major constituent with small amounts of carbohydrates and lipids (Nakano et al., 2003).

<u>Albumen.</u> Albumen quality has been related to embryo viability. Some researchers have put forth the hypothesis that albumen characteristics may influence the movement of gases and nutrients from the albumen to the developing embryo (Brake et al., 1997; Peebles et al., 2000b). Albumen quality (height and viscosity) is greater in eggs laid by younger hens (Williams, 1992; Lapão et al., 1999; Tona et al., 2004a). A degradation in albumen viscosity (albumen liquefaction) occurs during egg storage (Hurnik *et al.*, 1978). Associated with this decline in albumen quality is a reduction in the barrier to gaseous diffusion (Meuer and Baumann, 1988). Brake et al. (1997) suggested

that, in young flocks, eggs should be stored for a longer time in order to reduce albumen viscosity and improve the ability of oxygen to move through the albumen to the growing embryo.

At oviposition, the proteins of the albumen, which include lysozyme, avidin, conalbumin and ovomucin, act as antimicrobials (Wang and Shelef, 1991). The antimicrobial properties of egg albumen have been long reported as powerful defence mechanisms protecting the developing embryo (Korotkova, 1956; Garibaldi, 1960; Clavijo et al., 2006).

<u>Yolk.</u> Lipids are the main component of the yolk and the main energy source for the developing chicken embryo, which derives more than 90% of its energy requirements from the oxidation of yolk fatty acids (Romanoff, 1960). The amount and distribution of lipid components in the egg yolk influence the yolk sac nutrient reserves in the chick (Vieira and Moran Jr., 1998; Vieira and Moran Jr., 1999). The yolk sac is a well-vascularised set of membranes that entirely surround the yolk after 5d of incubation; it's function is to absorb nutrients from the yolk and to transfer them to the embryo (Romanoff, 1960). At about 19d of incubation the yolk sac begins to be withdrawn into the embryo's body cavity through the navel. By the time of hatching the yolk has been entirely drawn into the abdominal cavity and the navel is closed. The remnant of the yolk sac gradually disappears during early postembryonic life (Romanoff, 1960).

<u>Albumen and Yolk Proportions</u>. Proportions of yolk and albumen in broiler breeder eggs are affected by egg weight (Peebles et al., 2000b; Joseph and Moran Jr., 2005a) and flock age (Suarez et al., 1997). Vieira and Moran, Jr. (1998) showed that smaller eggs have a greater proportion of yolk at the expense of albumen. It has been reported that as egg size and flock age increase there is an increase in albumen fraction and a decrease in yolk fraction (Ar and Meir, 2002; O'Dea et al., 2004).

Eggshell. The eggshell performs a dual function during embryonic development: It must be strong enough to contain the egg nutrients required for embryonic growth and protect the embryo during incubation, but also have adequate porosity to facilitate gas exchange (Barnett et al., 2004). During incubation avian embryos "breathe" using a specialized membrane (the chorioallantois) located directly beneath the shell membrane. The chorioallantois is composed of a placentalike rich capillary vascular plexus which carries out gas exchange with the ambient atmosphere through small pores (holes) in the eggshell (Tazawa, 1971).

<u>Shell Quality</u>. Specific gravity is the industry standard used to assess shell quality. This process involves immersing eggs into salt solutions with different specific gravities; with the solution in which an egg floats indicating the specific gravity of the egg (Bennett ,1992). Eggs with thinner shells float in solutions with lower specific gravities (less salt); eggs that float in a salt solution with a specific gravity of less than 1.080 are considered thin-shelled (Roque and Soares, 1994). Eggshell strength as an estimate of eggshell quality can be determined using diverse techniques described by De Ketelaere et al. (2002). The relationship between shell color and shell quality is still unclear (Joseph et al., 1999).

Shell quality (thickness) decreases as hens age (Peebles et al., 2000b) and egg size increases (Roque and Soares, 1994). This decrease in eggshell quality has been associated with lower hatchability (Narushin and Romanov, 2002). McDaniel et al. (1979) found a direct relationship between low specific gravity and low hatchability in older flocks that had a greater number of eggs that felt below a specific gravity value of 1.080.

Standard Incubation conditions.

Hatching eggs are incubated under specific temperature, relative humidity (RH), ventilation and turning conditions (Tullet, 1990). The optimum incubation temperature for poultry species is between 37 and 38°C (French, 1997). In commercial hatcheries, the RH in the incubator (0 to 18 d of incubation) is approximately 58-60% RH. After transfer of eggs to the hatcher (18 to 21 d of incubation) the humidity is increased to 65% RH (Smith, 2000). Egg turning is completed by wild birds during incubation and has been shown to be of critical importance from 3 to 7 d of incubation (Deeming, 1989). Hatching eggs in commercial incubators are held in a position 45° to the horizontal and are alternated to the left or right of the horizontal hourly.

Egg Weight Loss During Incubation

During incubation, the avian embryo is dependent on gas exchange through the eggshell. Oxygen enters into the egg and CO₂ and water vapour pass out through the pores of the shell. Excess metabolic water generated during embryonic development (Wangensteen and Rahn, 1970b) is almost entirely lost via water diffusion through the shell (Wangensteen and Rahn, 1970a). Since egg weight loss is related to initial egg weight and eggshell characteristics (shell structure, shell membrane structure); egg moisture loss during the incubation period can be an indicator of eggshell quality (Tona et al., 2001). Egg weight loss is also affected by incubation conditions such as temperature, relative humidity and air velocity (Ar, 1991). Although Tullet and Burton (1982) have reported egg weight losses ranging from 7% to 16% during incubation, Tona et al. (2001) reported that optimum hatchability in broiler breeder eggs was obtained when relative egg weight loss was between 10.9% to 11.1% by d 18 of incubation.

The percentage of egg weight loss affects hatchability and day-old chick quality. In the case of excessive water loss, dehydration of the chorioallantoic membrane may affect gas permeability and critical gas exchange during the final days of incubation (Tona et al., 2001). In contrast, eggs that do not lose enough water do not have an optimum volume of air in the air cell; this may restrict the ability of the embryo to initiate pulmonary respiration after internal pipping (Ar, 1991).

Variations in egg weight loss have been reported between different flock ages. Reis et al. (1997) found that eggs produced by older hens (48 to 50 wk) tended to lose less weight (as a percentage) when compared to eggs from younger birds (32 to 34 wk). Conversely, O'Dea et al. (2004) reported an increase in percentage egg weight loss as flocks aged (from 37, 45 and 52 wk old flocks). These differences may be due to the different genetic strains used in the experiments: Reis et al. (1997) used eggs from Avian breeders while O'Dea et al. (2004) used eggs from Cobb breeders.

Tona et al. (2004b) compared two types of Hubbard parent stocks (L-type and Etype) differing in growth potential. The L-type broiler breeder has high reproductive performance but the broiler offspring reach market weight later than those of standard heavy (S-type) breeders; the E-type was selected to create a balance between growth, reproduction and liveability. The highest egg weight loss after 18 d of incubation was reported in the L-type (10.3%), and the lowest loss in the E-type (9.7%). These values fall slightly below the optimum range previously reported by the same author (Tona et al., 2001); this fact encourages the hypothesis that eggs from modern strains may need different incubation conditions to optimize egg weight loss.

Indicators of Successful Hatching Egg Production Efficiency

Fertility. Success or failure of eggs to hatch is determined by two main factors: fertility and embryonic mortality (Kuurman et al., 2003). Fertilization is defined as the successful union of a spermatozoon and an ovum (the egg), and results in the merging of genetic material from the hen and the rooster. Fertilization is determined by numerous factors some of which include genetics, seasonal variations, flock age, sperm quality, sexual behaviour and artificial insemination techniques (Brillard, 2003). Fertility is significantly related to hen age and is usually higher in peak production flocks when compared to old flocks. The expected fertility for modern commercial strains varies according to flock age: From 25 to 30 wk of age the expected fertility is approximately 94%; fertility increases to 97% from 31 to 45 wk and by the end of the production cycle (51 to 60 wk) it decreases to 92% (Aviagen, 2003). Fasenko et al. (1992) reported an approximate 10% reduction in fertility from 31 to 54 wk of age in Indian River broiler breeders. Similar results were reported by Elibol et al. (2002) and Zakaria et al. (2005) using a modern broiler breeder strain (Ross 308).

<u>Hatchability</u>. Hatchability can be defined in terms of total percentage hatchability (number of eggs that hatch / total number of eggs set x 100) or hatchability of fertile eggs (number of eggs that hatch / number of fertile eggs set x 100). As with fertility, hatchability standards vary according to the age of the breeders. In Canada, hatching egg producers reported an average total hatchability of 82% in 2006 (CBHEMA, 2006).

Several factors affect hatchability of broiler hatching eggs. Physical damage to the eggshell disrupts the protective barrier of the egg and may create modes of entry for bacteria (Narahari et al., 2000). Hairline cracks (when the shell is broken, but the shell membranes remain intact) have been reported to decrease hatchability (Barnett et al., 2004). Egg size is one of the factors influencing hatchability, and lower hatchability for larger eggs has been reported by McNaughton et al. (1978). Lawrence et al. (2004) using 43 wk old flocks, and Kumpula and Fasenko (2004) using 28 and 30 wk old flocks, also reported lower hatchability in large eggs from the same genetic strain. This was due to higher rates of late embryo mortality and a greater percentage of culled chicks at hatch. With respect to flock age, Tona et al. (2001) reported lower hatchability and higher rates of embryo mortality in older (> 40 wk) than in peak production modern broiler breeder flocks. The lowest embryo mortality and the highest total hatchability were observed around 40 wk of age. Tona et al. (2001) concluded that the lower hatchability in eggs from younger breeders (27 wk of age) may be due to the egg composition of smaller eggs. Smaller eggs have low eggshell porosity, thicker shell membranes and more viscous albumen (which may impair the gas exchange to and from the embryos). For the older flocks, that produce larger eggs, the authors hypothesized that the lower hatchability may be due to a combined effect of high embryonic heat production and an inability to effectively remove this heat from the setter.

<u>Embryonic Mortality.</u> In chicken eggs, embryo mortality is not distributed uniformly but is relatively high during the first and third wk of incubation (Jassim et al., 1996). Mortality is high in the early stages (2 to 4 d of incubation) as physiological and genetic events lead to death of non-viable embryos (Christensen, 2001). In the late stages of incubation, mortality is high as the hatching process is complex and physically demanding. During this time the embryo must convert from chorioallantoic membrane respiration to pulmonary respiration, and is vulnerable to hypoxia (Menna and Mortola, 2002).

Embryonic mortality is affected by strain. Scott and Mackenzie (1993) compared broiler breeders (selected for egg and meat production) with table egg breeders (selected for egg production) and concluded that early embryonic mortality was higher

and heart defects more frequent in embryos from broiler breeders than in those from table egg breeders. Suarez et al. (1997) reported a variation in percentages of embryonic mortality from 6.2% to 14.4% between five different Arbor Acres broiler breeder strains (not specified), with the highest percentage characterized by a greater incidence of malpositioned embryos.

Breeder flock age influences embryonic mortality as well. An increase in late embryo mortality from 3.8% to 6.6% was reported by Elibol et al. (2002) when comparing broiler breeder flocks at 31 and 52 wks of age. The authors hypothesized that embryos from older flocks have more difficulties exiting the shell than embryos from younger flocks. Overall embryo mortality in Arbor Acres broiler breeders at 29, 41, and 52 wk of age was 10%, 5.8%, and 8.8% respectively (Suarez et al. 1997). These results offer a support to the higher hatchability results reported by Tona et al. (2001) in 40 wk old flocks.

In relation to egg size, Reinhart and Hurnik (1984) reported higher percentages of malformations in embryos from extra large eggs (69.6 g) than from other egg groups when analysing eggs from the same commercial broiler breeders. In a more recent report, Kumpula and Fasenko (2004) found higher late embryonic mortality from large eggs (6.1%) when compared to small eggs (1.8%) from two modern broiler breeder strains at similar flock ages (28 and 30 wk).

Keirs et al. (2004) documented the incidence of embryonic anomalies in an extensive analysis of hatch residue from broiler hatching eggs over a 21-year period (from December 1979 to December 2000) and concluded that even though hatchability remained similar (83% to 84 % average), embryonic anomalies increased from 0.29% to 2.54%. This might be an indicator that embryos from modern strains are facing different challenges during the incubation process than embryos from 20+ years ago. Although abnormal positions (malpositions) of avian embryos within the egg are one of the factors

contributing to failure to hatch, the incidence of malpositions did not differ significantly between embryos from six modern broiler breeder strains (Arbor Acres Regular X Arbor Acres Classic, Arbor Acres Yieldmaster X Arbor Acres Feather-Sexable, Cobb X Cobb, Hubbard X Hubbard, Ross X Ross, and Avian Main X Avian 24 K) (Wilson et al., 2003).

Incubation length and the hatch window. In commercial operations the goal is to remove chicks from hatchers after about 504 hours of incubation (21 d). However, all chicks do not hatch at the same time and the period of time (the hatch window) between the hatching of the first and the last chick is between 24 to 36 hours (Vieira et al., 2005). Because of the variability in time of hatching this also results in high variability in the number of hours spent between hatching and placement in the broiler house (Vieira et al., 2005). An extended hatch window can cause chick quality issues as the early hatching chicks are left in the hatcher and may become dehydrated while waiting for other chicks to hatch. Therefore, a compressed hatch window is optimum for chick quality. Under commercial conditions, chicks receive their first access to feed 36 to 48 hours after hatching. During this interval between hatching and placement, chick body weight decreases significantly (Pinchasov and Noy, 1993; Sklan et al., 2000).

Incubation length is affected by egg size. Recent research has reported that lighter eggs hatch earlier (Wilson, 1991; Kumpula and Fasenko, 2004) and also have a longer hatch window than heavy eggs (Vieira et al., 2005).

Flock age also affects incubation length. Reinhart and Hurnik (1984) reported the shortest incubation length (492 h) in eggs from a young breeder flock (33 vs. 50 wk), at the lowest setter humidity (45 vs. 57%) and the smallest egg size (59.3 vs. 69.6 g). Results from Hudson et al. (2004) contradict these results. At 480 h of incubation, 24.6% of hatched chicks were from 41 wk old hens while only 10% of hatched chicks were from 29-wk-old -hens. It should be noted that the flock ages in the two studies were different,

and that in the case of Hudson et al. (2004) the high percentage of hatched chicks was from a flock at peak production age.

<u>Chick Quality</u>. At the hatchery chick quality / health is evaluated by subjective visual characteristics. A quality chick is alert and active, free of any physical abnormalities or injuries, and has a closed navel. In terms of broiler production, chick quality is evaluated by post-hatching growth, feed conversion, thermoregulatory ability, susceptibility to stress and liveability (Decuypere and Michels, 1992). High quality day-old chicks are the final product of the hatchery and the starting material for broiler farmers. In Alberta, it is accepted that approximately 2% of chicks shipped from the hatchery will be of suboptimal quality; because of this 102 chicks are included in each box of 100 chicks paid for by broiler producers (Alberta Broiler Hatching Egg Industry, 2006).

Decuypere and Michels (1992) reported that good hatchability does not necessarily mean good chick quality, viability and growth potential. In their research Decuypere et al. (2001) considered that pre-incubation factors (physical quality of the egg, embryo development at oviposition and egg storage) and incubation factors (incubation conditions and spread of hatch) play an important role in determining the quality of the newly hatched chick.

In the broiler industry larger chicks are preferred by broiler producers. O'Dea et al. (2004) reported differences in day-old chick weight due to genetics. Not surprisingly, the heaviest chicks were from a modern strain selected for high breast yield, followed by chicks from a modern strain selected for the whole bird market, with the lightest chicks coming from a broiler breeder strain unselected from 1978.

The influence of egg size on chick weight as an indicator of chick quality has been extensively examined, with heavier chicks hatching from larger eggs (Wiley, 1950; Pope and Schaible, 1957; Guill and Washburn, 1973; Wilson, 1991). In the above

mentioned reports, egg size was mainly determined by flock age with older hens producing heavier eggs (and heavier chicks).

Although older breeder flocks produce a greater number of heavier chicks (Reis et al., 1997; Suarez et al., 1997) the percentage of high quality chicks was lower in older (45 wk) compared to younger flocks (35 wk of age) (Tona et al., 2004a).

There is some research questioning the quality of large chicks. Kumpula and Fasenko (2004) reported that when small, medium and large eggs from the same flock were compared, although the larger eggs produced heavier chicks, chicks from the large eggs had higher cull rates.

Time of hatching and the duration the chick spends in the hatcher post-hatching can affect chick quality. When a chick hatches early and is kept in the hatcher for an extended period of time, this can lead to a significant reduction in chick weight due to dehydration and utilization of yolk sac reserves (Reinhart and Hurnik, 1984; Wyatt et al., 1985). Joseph and Moran Jr. (2005b) reported smaller yolk sac reserves in early hatched chicks (longer hatcher holding time) than in late hatched chicks (shorter hatcher holding time).

Methods Used to Determine Chick Quality

The quality of the newly hatched chick is important not only as an estimator of survival but also as an indicator of potential growth performance (Meijerhof, 2005). However, the common parameters used to establish chick quality are based on subjective observation of the general appearance of the chick at hatching (Tona et al., 2005). Over the past 10 years as hatchery automation has replaced hatchery workers, there is less opportunity for visual assessment of the chicks. Both quantitative and qualitative factors have been studied as potential estimators of chick quality.

<u>Quantitative parameters of chick quality</u>. Chick body weight is the most commonly used quantitative measure. Research dating back to the 1950's has studied the association between chick weight and post-hatch growth performance with contradicting results. Egg size relates to chick weight at hatching but the initially strong correlation changes during the growth period to the point where in some studies, chick weight at hatching is not a good indicator of broiler body weight (BW) at slaughter (Kosin et al., 1952; Bray and Iton, 1962; Gardiner, 1973; Shanawany,1987). Other studies present data showing a strong relationship between hatched chick weight and final broiler weight (Tindell and Morris, 1964; Merrit and Gowe, 1965; Proudfoot and Hulan, 1981; Sklan et al., 2003). Although easily recorded, day-old chick weight has limited value as an indicator of chick quality as it is more strongly correlated with egg weight rather than chick growth potential (Meijerhof, 2005).

Another parameter used to measure chick quality is chick length (crown to rump measurements) (Hill, 2001). Lawrence et al. (2004) examined chick length (measuring length from the tip of the beak to the end of the middle toe) as a quantitative measure of chick quality. They reported that chick length (as well as chick weight) was greater in chicks hatched from large versus small eggs (collected on the same day from one commercial breeder flock) and that BW at 14 d was significantly greater in chicks that were longer at hatching. Accordingly, Wolanski et al (2006) reported that chick length and shank length (measurement of the tibiotarsus from the top of the hock joint to the bottom of the footpad) correlate more strongly with BW at 14 d than hatch weight. In addition, Molenaar and Reijrink (2006) reported that longer chicks at hatching had more developed organs at 7 d (significant differences in heart, liver and spleen weights) than short chicks.

<u>Qualitative parameters of chick quality</u>. Different scoring methods have been previously described to provide qualitative measures of chick quality (Boerjan, 2002,

2005; Tona, 2003; Meijerhof, 2005). One of the most comprehensive lists of physical characteristics used to evaluate chick quality was provided by Tona et al. (2003) (Table 1-1). To summarize these characteristics: a day-old chick of good quality should be dry and clean, alert, standing and willing to move. It should be free of deformities, and have a normal conformation of the legs and toes. The navel should be completely healed and dry, with no yolk sac remnants protruding from the navel.

Many of the methods of evaluating chick quality can provide an indication of growth potential. Chick length seems to be a quality parameter that is related with development and broiler performance (Meijerhof 2005, 2006). The ultimate determination of chick quality is whether or not the hatchling grows at an acceptable rate and survives in order to be shipped for processing.

Broiler Performance

Consumer demand for white meat has influenced genetic selection in the poultry industry to increase growth potential and improve meat yield. Through application of quantitative genetics, progress in growth rate, as well as reductions in feed conversion (FC) and in the number of days needed to reach market BW have been achieved (Havenstein et al., 1994, 2003a). These genetic changes have, however, negatively affected the reproductive efficiency of broiler breeders (Siegel and Dunnigton, 1985; Pollock, 1999; Joseph and Moran Jr., 2005a) and some characteristics of the broiler offspring such as embryonic visceral development (Havenstein et al., 2003b; Mikec et al., 2006) and chick immunologic status (Yunis et al., 2000; Cheema et al., 2003a, 2003b).

Environmental Management and Influence on Health. In commercial poultry production, chicks should be placed in a barn environment that permits the birds to

achieve optimum performance in growth rate, uniformity, feed efficiency and meat yield, without compromising the health and welfare of the birds. To attain these objectives, broiler producers must control temperature, ventilation, air quality, and stocking density in the barn.

During the first days of life, a chick still reacts as a poikilotherm (an animal that is unable to regulate its own body temperature; thus the body temperature fluctuates according to ambient temperature). As a consequence, if ambient temperature is not optimum (about 32°C) a drop or an increase in body temperature can lead to cold or heat stress. Previous research indicated that Ross 208 chicks hatching from older flocks (60 wk of age) were more resistant to cold, while chicks hatching from younger flocks (25 wk of age) were more resistant to heat (Weytjens et al., 1999). The authors hypothesized that these differences in heat tolerance ability were due to variation in thermoregulating hormone levels (thyroid hormones) (Weytjens et al., 1999).

Appropriate ventilation and stocking density in the chicken barn should be controlled to prevent the accumulation of ammonia produced by the decomposition of organic matter. Ammonia levels above the recommended 25 ppm cause irritation of various mucous membranes and predispose the birds to diseases as a consequence of airsaculitis, tracheitis, and keratoconjuntivitis (Kristensen and Wathes, 2000). A significant reduction in BW at 7 weeks of age was reported by Miles et al. (2004) in Ross 508 broilers exposed to 75 ppm versus 25 ppm of atmospheric ammonia (2,920 g vs. 3,202 g). In the same report, cumulative mortality at 7 weeks of age was significantly greater in broilers from the 75 ppm ammonia treatment than in those from the 25 ppm ammonia group (13.9 % vs. 2.8%). These facts confirm the importance of providing the birds with adequate environmental conditions not only to prevent welfare and health related issues but also to maximize production.

<u>Nutrition.</u> The time between hatching and access to feed has proven to be of great importance for the normal development of the gastrointestinal tract of the chick (Reinhart and Hurnik, 1984; Wyatt et al., 1985). A delay in access to feed has negative consequences on chick weight (Joseph and Moran Jr., 2005b). Access to feed and water in the first hours after hatching is crucial to the early performance of chicks (Careghi et al., 2005) as it enhances a rapid development of the gastrointestinal tract and associated organs (Uni et al., 1998), and stimulates the transport and utilization of the yolk sac contents (Noy and Sklan, 2001). Feed is a major component of the total cost of broiler production. Modern broiler rations are formulated to give the correct balance of energy, protein and amino acids, minerals, vitamins and essential fatty acids, to allow optimum growth and performance (Aviagen, 2007). About 85 to 90% of the increase in broiler growth rate over the past five decades has been brought about by genetic selection, with nutritional improvements contributing the remaining 10 to 15% of the change (Havenstein et al., 2003a).

Feed conversion (FC). FC is the ratio of grams of feed consumed by the broiler to increase its BW by 1 g. The current expected FC at 42 d of age under standard commercial conditions for the Ross bird is 1.72 (Aviagen, 2007). As a result of constant genetic selection, FC falls by 0.1 every five years, with the target of reaching 1.5 by 2010 in the Cobb bird (Cobb-Vantress, 2007).

When analyzing broilers from 40+ years ago, different results regarding the effects of egg size on FC were reported. Proudfoot et al. (1982) reported better efficiency in chicks from larger eggs but Morris et al. (1968), Guill and Washburn (1973), and Wyatt et al. (1985) reported no differences in FC between egg weight groups. In modern broilers, Hulet et al. (2007) concluded that chicks that hatched from eggs laid by older breeder flocks (57 wk) have significantly greater cumulative FC values compared with chicks from younger flocks (29 wk of age).

<u>Broiler mortality</u>. A daily mortality of 0.1% is considered normal in commercial broiler production, however, broiler producers have reported average mortalities of up to 2% during the first wk of the grow out period.

Research results are not consistent regarding the effects of egg size and flock age on broiler mortality. Older reports (Morris et al., 1968) found no relationship between egg size and mortality, but more recent reports (Vieira and Moran Jr., 1998) have reported an increase in mortality in chicks hatched from small eggs. Likewise, when studying broilers from 30 + years ago, McNaughton et al. (1978) reported higher mortality rates in chicks hatched from younger breeder flocks, but more recently, Peebles et al. (1999a) reported opposite results. According to Havenstein et al. (2003a) most of the changes in chick mortality rates for the modern strains are probably due to an improved growing environment.

RELATIONSHIP BETWEEN EGG YOLK COMPONENTS AND THE RESIDUAL YOLK SAC OF THE HATCHED CHICK

Yolk Utilization by the Embryo

The egg yolk provides the developing embryo with nutrients from the start of vascularization (formation of the first blood vessels and blood cells) and formation of the yolk sac around 2 d of incubation through hatching (Romanoff, 1960). The yolk sac is a well vascularised membrane that entirely surrounds the yolk by 5 d of incubation. The main functions of the yolk sac are to produce blood, and to absorb nutrients from the yolk and transfer them to the embryo (Romanoff, 1960).

At about 19 d of incubation the yolk sac begins to be withdrawn into the embryo's body cavity through the navel. By the time of hatching, the yolk sac (whose weight has been reduced by about 60% over the course of incubation) has been entirely drawn into the abdominal cavity and the navel is closed. This residual yolk sac (RYS) is absorbed after hatching (Romanoff, 1960). In post-hatch chicks, the yolk sac content is utilized preferentially for growth of the small intestine and also as a supply of energy (Noy and Sklan, 1999).

The last 7 d of the incubation period are distinguished by intense lipid metabolism and rapid embryo growth (Romanoff, 1960). It is during the last wk of incubation that the most important changes in yolk fatty acid composition (Noble and Connor, 1984; Noble et al., 1986), rapid absorption of yolk lipids, and accumulation of lipids in embryonic tissues (Peebles et al., 1999b) occur.

It has been estimated that more than 90% of the total energy requirements of the chicken embryo are provided by the β -oxidation of fatty acids derived from yolk lipids (Romanoff, 1960). Lipid utilization by the avian embryo is a complex process directed by the requirements of particular embryonic tissues for specific fatty acids (Speake et al., 1998b).

The yolk sac contents enable the nutritional adaptation of the newly hatched chick from the embryonic environment to independent life. A direct positive relationship has been reported between the amount of nutrients provided by the yolk sac and the subsequent performance of the broilers (Vieira and Moran Jr, 1999).

Composition of the Egg Yolk

A typical chicken egg contains about 30% yolk, of which half is water and half is solid mater. Of the total solids contained in the yolk, 60% is comprised of lipids and 30%

of protein (Ahn et al., 1997). The lipids in the yolk are mainly very low density lipoproteins (93%) which can be divided in different lipid classes: Triacylglycerol (TAG) (67%), phospholipids (PL) (25%), free cholesterol (FC) (up to 5%), and cholesteryl ester (CE) and free fatty acid (trace amounts) (Speake et al., 1998a). The relative proportions of these lipid classes are very similar across domestic avian species and are not easily manipulated by dietary, physiological or pharmacological means (Speake et al., 1998a).

Speake et al. (1998a) described a general FA composition of the TAG, PL and CE of the yolk of eggs laid by hens maintained on commercial diets. Palmitic ($C_{16:0}$) and oleic ($C_{18:1(n-9)}$) acids are the major components of all the three lipid classes (approximately 25% and 40% of total FA respectively) whereas the major polyunsaturated fatty acid (PUFA) is linoleic acid ($C_{18:2(n-6)}$) (15%). The presence of linolenic acid ($C_{18:3(n-3)}$) tends to be very low (1%).

The long-chain PUFA of the n-6 and n-3 series account for about 3% and 2%, respectively, of the total yolk fatty acids (Nielsen, 1998).

Factors Affecting the Fatty Acid Composition of Egg Yolk. Information on the factors affecting the proportions of egg yolk components from broiler breeder hens is sparse. Vieira and Moran Jr. (1998) conducted an analysis of eggs from four different broiler breeder strains (not specified by authors) each strain having a different age (36, 40, 42, and 45 wk of age) and reported significant differences in percentage yolk weight with greater amount of yolk in the 40 wk old flock (33.3%) than in the other three flocks (results ranged from 31.6% to 32.4% of egg weight). Significant differences were reported for the total lipid content of the yolk: the 40 and 42 wk old birds laid eggs with greater amounts of total lipids (60.8% and 59.1% respectively) than the 36 and 45 wk old birds (56.9% and 56.3% respectively). Although statistically significant, the differences

are small perhaps because all the ages from which eggs were analyzed were close to the peak production age.

The composition of the whole egg can be altered by genetic selection. Cahaner et al. (1986) studied broiler breeder hens from lines selected for reduced abdominal fat (high-fat vs. low-fat), and reported an association between high abdominal fat and an increase in yolk weight, yolk dry matter and yolk fat content.

Results reported by Latour et al. (1998) explained some differences in the FA content of eggs from Arbor Acres broiler breeders at 36, 51 and 64 wk of age. Hen age altered palmitic ($C_{16:0}$), stearic ($C_{18:0}$), and palmitoleic ($C_{16:1(n-7)}$) acid concentrations: palmitic and stearic acids were higher in the yolks of eggs from old hens (51 and 64 wk of age), than in eggs from young hens (36 wk old). The opposite proportion was observed for palmitoleic acid. A complete description of the FA profile was not provided in this experiment.

Extensive research in laying hens has been conducted with respect to the factors affecting the FA composition of eggs. Nielsen (1998) reported no significant differences in total lipid, total cholesterol and phospholipids contents in egg yolk between young (21 wk) and old (57 wk) hens. However he did report that the contents of arachidonic $(C_{20:4(n-6)})$ and docosahexaenoic (DHA, $C_{20:6(n-3)})$ acids were 20% to 25% higher in egg yolks from the young hens.

Manipulation of the hen's diet is the main factor affecting the composition of the egg. Peebles et al. (2000a) showed a greater percentage yolk weight at the expense of the albumen in hens that were fed a diet with high energy level (467 peak Kcal/hen-day) than in hens fed a diet with low energy level (430 peak Kcal/hen-day).

Gao and Charter (2000) analyzed table eggs from six different Canadian sources (three from hens receiving regular commercial diets, and three from hens under special

diets). Although the ingredients or inclusion rates of the diets were not specified (these data are needed in order to interpret the egg FA content), the levels of linolenic $(C_{18:3(n-3)})$, arachidonic $(C_{20:4(n-6)})$, and DHA $(C_{20:6(n-3)})$ differed between eggs, meaning that long chain PUFA are more likely affected by dietary changes than other FA.

Conjugated linoleic acid (CLA), which is a mixture of isomers of linoleic acid $(C_{18:2(n-6)})$, has been reported to have potential anti-cancer, anti-atherogenic, anti-adipogenic, anti-diabetogenic, and anti-inflammatory properties, as a result, numerous research has been conducted in an effort to supplement human's diet with CLA through enriched table eggs (Wahle et al., 2004). Dietary addition of CLA has been reported to increase palmitic ($C_{16:0}$), stearic ($C_{18:0}$), SFA, and long chain n-3 PUFA, and to decrease palmitoleic ($C_{16:1(n-7)}$), oleic ($C_{18:1(n-9)}$), total MUFA, and long-chain n-6 PUFA (Alvarez et al., 2004, 2005; Muma et al., 2006).

Not only the ingredient composition but also the source of fat used in the hen's diet alter the FA composition of eggs (Grobas et al., 2001). Diets based on corn and animal fats or vegetable oils, which are high in SFA and linoleic acid ($C_{18:2(n-6)}$), increase the deposition of linoleic acid and arachidonic acid ($C_{20:4(n-6)}$) in the egg (linoleic acid is the precursor of arachidonic acid) (Ajuyah et al., 2003; Pardio et al., 2005, Muma et al., 2006). On the other hand, addition of fish oil and certain oilseeds (flax seeds) to the hen's diet, promotes the deposition of long-chain n-3 PUFA, (van Elswyk, 1997) but decreases long-chain n-6 PUFA (Alvarez et al., 2004) in the egg.

Composition of the Residual Yolk Sac

Data regarding the composition of RYS of newly hatched broiler chicks is limited. A dated report from Entenman et al. (1940) on newly hatched chicks (~35 g) showed that the RYS represented approximately 14% of the chick BW, and that it was composed of 11% lipids. In more recent research, using day-old broiler chicks from the Shaver-Starbro line, Jamroz et al. (2004) reported that the RYS was composed of approximately 15% lipid and 26% protein. Deeming (2005) reported that the RYS of day old Ross chicks represented 9.0% of total BW. Different results were reported by Mikec et al. (2006) using the same Ross strain of chicks: the RYS was approximately 14% of the total BW. It is important to clarify that the age of the broiler breeders or the egg weight were not specified in either of these experiments. A complete description of the FA profile of the RYS of the newly hatched chick from modern broiler strains has not been reported.

Similar to what was described for egg yolk FA composition, the description of FA content of the RYS of chicks from 43 wk old Single Comb White Leghorns hens has been provided by Latour et al. (2000). The main FA present in the RYS according to Latour et al. (2000) were palmitic ($C_{16:0}$, 19.8%), stearic ($C_{18:0}$, 9.7%), oleic ($C_{18:1(n-9)}$, 44.1%) and linoleic ($C_{18:2(n-6)}$, 17,2%).

Factors Affecting the Fatty Acid Composition of the Residual Yolk Sac. The effect of breeder flock age on the FA composition of RYS from newly hatched chicks has been reported by Latour et al. (1998). In that research, they compared RYS from chicks from an Arbor Acres flock at 36, 51 and 64 wk of age. The results showed no variation in palmitoleic ($C_{16:1(n-7)}$) acids in RYS even though flock age affected the palmitoleic content of fresh eggs.

The FA composition of the RYS can be affected by variations in the hen's diet. Multiple authors have conducted research in laying hens, not only analyzing the FA content of RYS (Latour et al., 2000), and embryonic tissues (Peebles et al., 1999b; Surai and Sparks, 2001), but also in other tissues from the newly hatched chick (Cherian and Sim, 1997). Results from Cherian et al. (2002) showed that although the total fat content

of the egg yolk was not affected by different diets, the fatty acid content of the hen's hepatic and muscular tissues was significantly altered by the inclusion of CLA in the diet. In addition, the SFA content in liver as well as the CLA content in the thigh and breast muscles increased in birds fed CLA. Similarly, hens fed different diets produced chicks that had different FA composition of tissues (Cherian et al., 2005). An increase in dietary CLA produced an increase in liver, plasma, adipose tissue, heart, and brain levels of SFA and stearic acid ($C_{18:0}$); as well, a decrease in MUFA and oleic acid ($C_{18:1(n-9)}$) were observed. These results show that changes in the yolk fat proportions, produced by maternal diet manipulation also affect the composition of FA in the embryo.

Relationship Between Fresh Egg Yolk and Residual Yolk Sac Fatty Acids

In their 1998 research Latour et al. showed some variations between the FA content of fresh egg yolk and RYS of day old chicks. The eggs and chicks analysed were from an Arbor Acres flock at 36, 51 and 64 wk of age. The significant differences that were reported were: 1) Palmitic acid ($C_{16:0}$) concentrations were higher in RYS (26.4%) than in fresh yolks. 2) A pronounced decrease (~3.5 times) was observed in the level of palmitoleic acid ($C_{16:1(n-7)}$) in the RYS from 36 wk old chicks when compared to the fresh eggs from the same age (no differences for that FA were observed for the 51 or 64 wk old flocks). 3) The levels of oleic ($C_{18:1(n-9)}$) and arachidonic ($C_{20:4(n-6)}$) acids were higher in RYS compared to fresh eggs from the 36 wk old flocks.

Objectives

The first objective of the research presented in this thesis was to evaluate the effects of broiler breeder genetic strain, flock age, and egg size on the following

economically important production parameters in the broiler industry: Hatching egg characteristics, fertility, hatchability, saleable chick production, and broiler growth and performance.

A second objective was to establish if genetic strain, flock age, and egg size are factors that affect the fatty acid content of hatching eggs and residual yolk sacs,

A third objective was to examine the relationship between fatty acid content in the hatching egg and fatty acid content in the RYS of the newly hatched chick.
TABLE

Parameters	Assessment
Activity	Activity is assessed by laying the chick on its back to determine how quickly it returned to its feet. A quick spring back onto its feet was regarded as good, but trailing back onto its feet or remaining on its back was assessed as weak.
Down and	The chick body was examined for dryness and cleanness. It was
appearance	regarded as normal if it is dry and clean. If it is wet or dirty or both (which can be a source of contamination) then it is not good.
Retracted yolk	The chick was put on its back obliquely on the hand palm until abdominal movement totally stopped. The height of its abdomen was estimated. The consistency of the abdomen to touch was then estimated. If the height of abdomen was estimated to be higher and harder to touch than normal, then the yolk was regarded as large and consistent.
Eyes	The chick was put on the legs, and its eyes were observed. The state of brightness and wideness of the gape of the eyelids were estimated
Legs	The chick was put on its feet to determine if it remained upright well. The toes were examined for their conformation. If the chick remained upright with difficulty, articulations of the knees were examined to detect signs of inflammation or redness or both.
Navel area	Navel and surrounding areas were examined for closure of the navel and its coloration. If the color was different from the skin color or the chick, then it was regarded as bad.
Remaining	Observation of the navel area allowed estimation of the size of
membrane	any remaining membrane. The size of any remaining membrane was classified as very large, large, or small.
Remaining yolk	Observation of the navel area allowed estimation of the size of any remaining yolk. The size of any remaining yolk was classified as very large, large, or small.

Table 1-1. Assessment of different parameters for determining chick quality(obtained from Tona et al., 2003).

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2. BROILER BREEDER GENETIC STRAIN, AGE AND EGG SIZE AFFECT HATCHING EGG CHARACTERISTICS, HATCHABILITY, SALEABLE CHICK PRODUCTION AND BROILER PERFORMANCE

INTRODUCTION

Because of consumer demand for white meat, primary breeder companies have shifted the focus of genetic selection to increase breast muscle meat production. It has been reported that this selection has had a negative impact on visceral development (Havenstein et al., 1994, 2003; Mikec et al., 2006) and immune function (Yunis et al., 2000; Cheema et al., 2003a, 2003b) in broiler offspring. There is limited research comparing egg characteristics, hatchability and broiler chick health and performance. In one of the few recent studies, Joseph and Moran (2005) compared two strains selected for growth rate and BW with a third strain selected for high breast meat yield. They found that egg components were different between the strains but hatchability and hatched chick organ weights were similar. The specific genetic strains compared were not identified.

Because broiler chicks are produced by parent flocks whose egg production period spans 30 wk or more, research into the effects of flock age on egg components, hatchability, and broiler growth has always been of interest. In 2004, Tona and colleagues examined eggs and offspring from a Cobb flock at 35 and 45 wk of age. In general, they found that chicks from the percentage of poor quality chicks was higher at the old (45 wk) than at the young age (35 wk of age).

In contrast to genetic strain and flock age, few studies have been conducted looking at the effect of egg size independent of flock age. While it is known that egg size increases with flock age (Reis et al., 1997; Suarez et al., 1997; O'Dea et al., 2004), it is not well documented what effect egg size at each flock age has on egg components, hatchability and broiler growth parameters. Lawrence et al (2003) and Kumpula and Fasenko (2004) reported lower hatchability and a higher incidence of culled chicks in eggs that were larger than average.

In Canada hatching egg producers are paid based on saleable (not hatched) chicks, therefore, the study of factors that influence the production of high quality chicks is of great interest. As the demand for poultry meat increases, the impact of genetics, flock age, and egg size in the broiler production process will gain importance. The objective of this study was to determine the effects of modern genetic strains, flock age and egg size on hatching egg characteristics, fertility, hatchability, saleable chick production, and broiler performance. It was hypothesized that 1) The Cobb strain (which has been genetically selected for high meat yield) will have higher embryo mortality, poorer hatched chick quality, but better broiler performance than the Ross strain (which has been genetically selected for the further processing market). 2) Eggs from young flocks will produce smaller chicks, but those chicks will have the same growth performance as broilers from the older flocks. 3) Average size eggs will at each age hatch the most saleable chicks compared to smaller or larger eggs.

MATERIALS AND METHODS

The experimental protocol was approved by the Faculty of Agriculture, Forestry and Home Economics, Animal Policy and Welfare Committee at the University of Alberta, in accordance with the guidelines set forth by the Canadian Council on Animal Care (1993).

Egg Collection

A total of 2,736 hatching eggs produced by two modern commercial broiler breeder strains (Cobb 500 (C) and Ross 308 (R), n= 1,368 eggs/strain) were obtained from a commercial hatchery using the same breeder flocks at 29, 45 and 59 wk of age. The C strain has been genetically selected for high white meat yield (particularly breast muscle) while the R stain has been designed for the further processing sector (whole bird market).

At each collection time the average egg weight for each strain was determined by randomly weighing 72 eggs/strain. Based on the average egg weight, eggs were collected according to three weight ranges: Small (S), Medium (M) and Large (L). Eggs from the same weight category were within \pm 1.5 g of each other and were selected in the following way: the M eggs were \pm 1.5 g from the average egg weight, the S eggs were 3.0 to 6.0 g lighter than the average and the L eggs were 3.0 to 6.0 g heavier than the average and the L eggs were transported to the Alberta Hatching Egg Producers Hatchery.

Egg Characteristics

For each flock age, thirty eggs within each strain/weight group were randomly selected to determine egg characteristics. These eggs were weighed, numbered, and

specific gravity assessed (SG) by the flotation method (Bennett, 1992) using different saline solutions with densities from 1.064 to 2.000. Eggs were then broken open, and wet eggshell and wet yolk weights immediately recorded. Albumen weight was calculated by subtracting wet yolk and wet shell weights from the total egg weight. The eggshells and yolks were dried at 65°C for 3 d in a Despatch V Series Heat Processing Unit¹ and the dry weights measured.

Incubation and Hatching

Remaining eggs were numbered, individually weighed and candled. Any eggs with hairline cracks were replaced with spare eggs from the same strain and weight category. Settable eggs (n= 2,376) were randomly divided into groups of 18 eggs for the purpose of statistical replication (n= 22 groups/strain/egg weight). Each group of 18 eggs was randomly allocated (to account for possible environmental differences within the incubator) into a 5,000 egg capacity Jamesway² single stage setter and incubated for 18 d at a dry bulb and wet bulb temperature of 37.5°C and 29.4°C respectively.

At 7 d of incubation all eggs were removed from the setter and candled. Any egg not containing a viable embryo was removed and broken open to assess fertility, and then replaced with an artificial plastic egg. If fertile, the day of embryonic death was estimated. At 18 d of incubation the eggs were removed from the setter, individually weighed, and transferred to a 5,000 egg capacity Jamesway hatcher where they were incubated for an additional 3.5 d at a dry and wet bulb temperature of 35.2°C and 29.4°C respectively. One group of 18 eggs per each strain/egg weight category was randomly selected and these eggs placed into pedigree hatch baskets (122 cm x 35 cm) so that

¹ Despatch Industries Inc. 8860 207 St. Minneapolis, MN. 55044

² Jamesway Incubator Company Inc., Cambridge, ON, Canada, N1R 7L3

hatching time could be assessed and the chick could be identified back to the egg from which it hatched. The pedigree hatch baskets were checked at 476, 484, 492, 500, 508 and 515 h of incubation. External pipping (embryo's beak through the shell) and hatching (chick free of shell and down dry) times were recorded.

Hatchability and Saleable Chick Production

After 21.5 d of incubation, all hatched chicks were counted, and chick quality was visually assessed according to commercial hatchery standards. Chicks that were abnormal, weak, had unhealed navels or red hocks were considered unsaleable. Hatchability was calculated based on saleable chicks only. All saleable chicks were individually weighed, neck tagged³ and randomly allocated to one of two replicate groups for each strain/egg weight. All unhatched eggs were broken open to determine the approximate day of embryonic death and embryonic mortality grouped into three categories: Early (1 to 7 d of incubation), mid (8 to 14 d of incubation) and late (15 to 21 d of incubation).

Broiler Performance

At each flock age, groups of saleable chicks in the same strain/egg weight category were randomly placed in six environmental rooms⁴ in which temperature, humidity and ventilation were strictly controlled to be the same. Each room was divided into two floor pens (n= 12 pens/age) such that the stocking density was 0.07 m2 /bird (0.75 ft2/bird). The number of chicks placed in each pen varied between trials (29 wk and 59 wk of breeder flock age – 119 chicks/pen; 45 wk of breeder flock age – 109

³ Avery Dennison Mark III Swiftach Tagging Gun. 150 North Orange Grove Blvd. Pasadena, CA

⁴ Dimensions: 4.4 m (14.44 ft) deep x 3.9 m (12.8 ft) wide = 17.16 m² (184.7 ft²)

chicks/pen) due to differences in the number of hatched saleable chicks. However, stocking density was held constant across the three trials by adjusting the pen area. The broilers were reared on wood shaving litter under standard commercial conditions for 42 d with a photoperiod of 23L:1D. Water and feed were provided ad libitum. Birds were fed a standard pelleted broiler starter (1 to 14 d), grower (15 to 28 d) and finisher (29 to 41 d). The ingredient composition and nutrient content for each of the diets are listed in Table 2-1. During the rearing period, daily mortality, weekly feed consumption and individual BW at 21 and 41 d were recorded. Feed conversion (FC), determined at 21 and 41 d, was corrected according to the number of birds that died. Possible cause of death was determined by dissection of the birds.

Statistical Analysis

The experimental design was a $2 \times 3 \times 3$ factorial arrangement with strain (C and R), age (29, 45 and 59 wk of age), and egg weight (S, M, and L) as main effects. Because average egg weight was different for each strain at each age, egg size groups were different. As a consequence, egg size was used as a covariable (as a nested factor in strain and age) to eliminate the variability due to egg size categories.

The statistical model for the nested analysis of variance was as follows:

$$Y_{ijkl} = \mu + G_i + A_j + S_k(G_iA_j) + G_iA_j + \varepsilon_{ijkl}$$

Where Y_{ijkl} = characteristic that was measured, µ= overall mean, G_i = main effect of genetic strain, A_j = main effect of flock age, $S_k(G_iA_j)$ = effect of egg size nested in the main effects of genetic strain and flock age, G_iA_j = interaction between genetic strain and flock age, and ε_{ijkl} = random error term. The experimental units (EU) differed according to the parameter that was measured: for hatching egg characteristics the EU was each egg. For incubation parameters the EU was each group of 18 eggs. For broiler performance the EU was each pen of broilers, except for BW which was each bird.

All percentage data were subjected to angular transformation to stabilize variances (arc sine square root percentage transformation) prior to statistical analysis. All data were analyzed using the MIXED model of SAS (SAS Institute, 2002), and the probability level set at P<0.05. When the model indicated significance, the means were separated using the LSMEANS procedure of SAS.

RESULTS

Hatching Egg Characteristics

<u>Breeder Strain</u>. Genetic strain did not influence egg weight or dry yolk weight (Table 2-2).

Because there was an interaction effect on the other hatching egg characteristics, the main effect of strain will not be discussed for these variables.

<u>Breeder Flock Age</u>. Average egg weight differed between flock ages with the 29 wk old flocks laying the lightest eggs and the 59 wk old flocks producing the heaviest eggs. As flock age increased, the proportion of dry yolk in the eggs significantly increased at the expenses of albumen and shell.

Because there was an interaction effect on the other hatching egg characteristics, the main effect of flock age will not be discussed for these variables. <u>Breeder Strain and Flock Age Interaction</u>. Shell quality, as estimated by SG, was significantly affected by the interaction of strain by age. It is of interest to point out that all egg categories had a SG equal or lower to 1.080, which has been reported as the minimum SG indicating good shell quality (Bennett, 1992; Roque and Soares, 1994). For C eggs, SG increased from 29 to 45 wk and then decreased to 59 wk, and for R eggs, SG decreased from 29 to 45 wk, increasing again at 59 wk. In relating specific gravity to percent dry shell weight, C eggs followed a similar pattern: Shell weight increased from 29 to 45 wk; however, in the R flock, percent dry shell was not related to SG and the highest percent of dry shell was observed in eggs with high and with low SG.

Albumen weight, as a percentage of the total egg weight, decreased as flock age increased in both strains.

Fertility, Hatchability and Saleable Chick Production

<u>Breeder Strain</u>. Significant differences between strains were only observed for early embryo mortality (Table 2-3). The R strain had significantly higher early embryo mortality (6.2%) than the C strain (4.0%). Breeder strain effects on all other parameters during incubation were not significant.

Because there was an interaction effect on fertility, total hatchability and mid embryo mortality, these parameters will not be discussed.

<u>Breeder Flock Age</u>. Percentage egg weight loss, late embryo mortality and percentage of culled chicks were significantly higher in the 45 wk old flocks than in the younger or older flocks; as a consequence, hatchability of fertile eggs was significantly lower in the 45 wk old flocks. Flock age had no effect on early embryo mortality.

Because there was an interaction effect on all other incubation parameters, the main effect of flock age will not be discussed for these variables.

<u>Breeder Strain and Flock Age Interaction</u>. Fertility, total hatchability and mid embryo mortality were affected by the interaction of strain by age. The R strain showed higher fertility in the youngest flock age and significantly decline thereafter. In contrast, fertility in the C strain was higher at 45 and 59 wk than at 29 wk of age. Total hatchability mirrored the results for fertility.

The C*45 wk group had higher mid embryo mortality than all other interactions.

Pipping and Hatching Times

No significant differences between genetic strains or flock ages were observed in pipping or hatching time (Table 2-4).

Chick and Broiler Body Weights

<u>Breeder Strain</u>. Because there was an interaction effect on broiler body weights, the main effect of breeder strain will not be discussed for these variable.

<u>Breeder Flock Age</u>. Chick weight at hatching and 41 d BW were affected by breeder flock age (Table 2-5). Chick weight increased significantly as the breeder flock aged. The highest BW at market age was observed in the broilers produced by the 45 and 59 wk old flocks with the greatest overall weight gain observed in broilers from the 45 wk old flocks.

<u>Breeder Strain and Flock Age Interaction</u>. Early weight gain was greater in the C*45 wk and C*59 wk than in the other groups, in turn, the lowest early weight gain was

observed in broilers from the C*29 wk group. As a result, BW at 21 d was greater in the C*45 and C*59 birds, and the C*29 birds had the lowest 21 d BW.

Feed Conversion

<u>Breeder Strain</u>. Late feed consumption (21 to 41 d of age) was greater for the R broilers than for the C broilers (Table 2-6). Because there was an interaction effect on the other broiler performance parameters, the main effect of breeder strain will not be discussed for these variables.

<u>Breeder Flock Age</u>. Significant differences in early FC were observed between flock ages with the chicks hatching from the 45 wk old flock having greater efficiency at 21 d of age. Chicks hatching from the 59 wk old flocks had poorer late FC value. indicating that broiler produced by older flocks are less efficient than broilers produced by young or peak production flocks.

<u>Breeder Strain and Flock Age Interaction</u>. Early feed consumption was greater in the C*59 wk than in the other groups, in turn, the lowest early feed consumption was observed in broilers from the C*29 wk group. When relating these results with BW at 21 d it was not surprising to observe that the heaviest and lightest broilers were the C*59wk and the C*29 wk respectively. Total FC was affected by the interaction breeder strain * flock age. The highest FC was observed in the R*59 wk and the lowest FC in the C*45 wk birds.

Broiler Mortality

<u>Breeder Strain</u>. There was no effect of strain on broiler mortality (Table 2-7).

<u>Breeder Flock Age</u>. Chicks hatched from young hens had higher first wk mortality than chicks hatched from the other flock ages. The lowest late and total mortality were observed in the broilers produced by the oldest flocks.

<u>Breeder Strain and Flock Age Interaction</u>. There was no effect of the interaction on broiler mortality (data not shown).

DISCUSSION

Hatching Egg Characteristics

Because a positive correlation between flock age and egg weight has been reported in several studies since the early 50's (Wiley, 1950; Tona et al., 2004) it was not surprising to find that average egg weight increased as the flocks aged. The increase in the proportion of dry yolk as flocks aged comply with the data reported by Suarez et al. (1997), but disagree with O'Dea et al. (2004) who did not find significant differences between 37 wk, 45 wk and 53 wk old flocks. In the latest report, the differences may be due to a smaller sample size used by O'Dea et al. It is well known that the egg yolk provides the necessary nutrients for the developing embryo (Romanoff, 1960), thus, the result that eggs from younger hens have a smaller proportion of dry yolk (and perhaps less nutrient content as a percentage of the egg) may have a negative effect in the embryonic development and posthatch performance of these chicks.

The fact that all egg groups had SG equal to or lower than 1.080, which was reported in the early 90's as the minimum SG indicating good shell quality (Bennett, 1992; Roque and Soares, 1994) may indicate that shells from eggs of modern strains have lower quality than shells from eggs of strains from 15+ years ago. It should be clarified that egg holding time prior to testing the SG was unknown. Longer egg storage would increase air cell size and make the eggs more buoyant. However, these data

suggest shell quality, as estimated by SG, of eggs from modern strains should be reexamined. It was expected that eggs from the C and R young flocks would have greater shell weights as previous research has shown that young flocks have a higher percentage of shell than older flocks (Peebles et al., 2000). This held true for the R strain but not for the C strain. Eggshell quality is a determinant factor for gas exchange and moisture loss during incubation (Wangesteen et al., 1970), and defective shell quality has been associated with higher percentage of egg weight loss (Reis et al., 1997; Peebles et al., 2001) and low hatchability (Narushin and Romanov, 2002).

In relating specific gravity to percent wet and dry shell weight there appears to be no relationship. For example, although the strain by age interaction with the highest specific gravity (R*29= 1.080) had the highest percent dry shell (9.2%), the interaction that produced one of the lowest specific gravities (R*45= 1.069) also shared the highest percent dry shell (9.2%) This data brings into question the validity of using specific gravity as an indicator of good shell quality.

It was observed that, as a percentage of the total egg weight, albumen weight decreased and wet yolk weight increased as the flocks aged. These results are in disagreement with Vieira and Moran Jr. (1998) who reported that smaller eggs from commercial broiler breeders have a greater proportion of yolk in relation to albumen, however, in their study, the above mentioned authors did not report flock age or genetic strain and their results were only based on egg size.

Fertility, Hatchability and Saleable Chick Production

The greater fertility observed in young birds from the R strain agree with previous research on the same strain by Elibol et al. (2002) and Zakaria et al. (2005) who

reported a decline in fertility as flock age increased (from 31 to 52 wk and from 34 to 59 wk, respectively). The above mentioned authors concluded that breeders from the R strain had better reproductive capacity earlier in the production cycle. In contrast, fertility in the C strain was higher at 45 and 59 wk than at 29 wk of age. It is important to clarify that the breeder flocks that were analyzed were reared by different producers, and that management practices could have had an effect on fertility. It should not be concluded that the differences observed in fertility are the consequence of genetic strain.

Ultimately, the interaction did not influence the hatchability of fertile, but flock age did. Unexpectedly the flocks at 45 wk had the lowest hatch of fertile. This result was not expected because it has been previously reported that eggs laid by hens in the middle of their production cycle have better chick production parameters than eggs from younger or older hens (Tona et al., 2001). Further examination of incubator conditions identified that the temperature in the setter during the last 2 wk of incubation for this flock age was higher than recommended (average 38.4 C vs. recommended 37.5 C). It is well known that high temperatures during incubation have a negative impact on egg weight loss (Tullet, 1990), that high rates of egg weight loss increase embryo dehydration (Tullet and Burton, 1982) and, as a consequence, embryonic mortality increases and hatchability and chick quality decrease (Reis et al., 1997; Peebles et al., 2001).

The higher early embryo mortality in the R strain could be a consequence of variations in early embryonic metabolism for this genetic line. Hamidu et al. (2007) reported significant differences in daily gas exchange between Ross 308 and Cobb 500 eggs (from the same flock age and egg weight) during the first 4 d of incubation.

It was surprising to observe the greatest percentage egg weight loss, late embryo mortality and culled chicks in the 45 wk old flocks. These results were not expected, and the increased incubator temperature described above is provided as an explanation. The C*45 wk interaction had higher mid embryo mortality than all other interactions. When referring this fact to the results observed for egg weight loss at transfer at 45 wk of age, it could be hypothesized that embryos from the C flock were more deeply affected by the heat stress (and dehydration) suffered in the incubator than embryos from the R strain at the same age.

Pipping and Hatching Times

The lack of significance in these results differ from previous research by Reinhart and Hurnik (1984) in commercial broiler breeders (strain not specified) who reported that eggs from the youngest breeder flock age (33 vs. 50 wk) had the shortest incubation period. They also differ from results published by Hamidu et al. (2007) who reported shorter pipping time for Ross 308 eggs than for Cobb 500 eggs (from the same flock age and set weight). However, It is important to note that in the current research, even though the hatching time means for strain, age and interactions are close together, the large SEM reveals that the hatching window was spread. The lack of significance could be a consequence of a large SEM for hatching time (5.9 h for strain, 7.3 h for age and 10.3 h for the interaction).

Chick and Broiler Body Weights

It has long been reported that bigger eggs (in this case, eggs laid by older hens) hatch bigger chicks (Wiley, 1950; McNaughton et al., 1978), hence, it was expected that chick weight increased significantly as the breeder flocks aged. Chick BW has traditionally been used to measure chick quality and as a predictor of broiler BW at market. In the present research, the highest BW at market age was obtained by the broilers from the 45 and 59 wk old flocks, which were statistically the same irrespective

of the fact that the chick weights at hatching were different. These results agree with previous research (Kosin et al., 1952; Bray and Iton, 1962; Gardiner, 1973; Shanawany, 1987) that show that BW at hatching does not accurately predict BW at slaughter age. It is important to point out that the chicks hatching from the young flocks did not catch up in final BW with the chicks from the older ages, demonstrating that these chicks, regardless of the strain, do not have the same potential for growth as chicks from older flocks. The lower chick BW could be a reflection of the smaller proportion of yolk in eggs from the younger flocks.

The greatest 21 d BW observed in the C*45 and C*59 birds was expected because the C strain has been selected for faster growth and greater meat yield. Similar results were reported by Joseph and Moran Jr. (2005). However, and for the same reason, it was not expected that the lowest 21 d BW observed would be in the C*29 wk broilers. These results indicated that that chicks from young C strain parents do not grow as well. The reason for this is unknown, but may be related to nutrient deposition in the egg.

Feed Conversion

Early feed consumption was greater in the chicks from the C*59 wk parents than in chicks from all other groups. In turn, the lowest early feed consumption was observed in broilers from the C*29 wk group. When relating these results with BW at 21 d it was not surprising to observe that the heaviest and lightest broilers were the C*59 wk and the C*29 wk respectively. Because all the chicks (irrespective of strain, egg size or flock age) were grown out under the same barn conditions, the question that arose was that chicks hatching from young C flocks may require different management practices during the first days of life for them to be able to perform to their optimum. Broilers produced by old parent flocks were less efficient than broilers produced by young or adult flocks as reflected by late FC. These results are in agreement with results reported by Hulet et al. (2007). The highest total FC was observed in the broilers from the R*59 wk parent flock while the lowest FC was in the broilers from the C*45 wk parent flock. The genetic selection for high meat yield in the C strain could be the reason why the broilers produced at older ages could still show a good production performance.

Broiler Mortality

The higher first wk mortality observed in chicks hatched from young hens agrees with results reported by McNaughton et al. (1978), and Vieira and Moran Jr. (1998) but are opposite to results reported by Peebles et al. (1999) who reported high first wk mortality in chicks hatched from 64 wk old flocks. The observation that it was the youngest flocks that had the highest mortality may be related to the fact that eggs from these flock ages also had smaller yolks and smaller chicks at hatching.

SUMMARY AND CONCLUSIONS

In the current study, most of the significant effects on egg, chick, and broiler production parameters were due to the interaction of breeder strain * flock age. This means that when aiming at improving saleable chick production, both parameters, genetic strain and flock age should be accounted for variations in the final outcome.

It is important to highlight that chicks produced by the youngest breeder flocks had lower quality / performance than chicks produced at older ages. This would be an important fact for broiler producers to be aware of as chicks from the young flocks may require a higher level of care. The hypothesis that average size eggs would have the best quality and would hatch the most saleable chicks compared with smaller or larger eggs could not be tested because the effect of egg size was nested in the main effect of breeder strain. However, the interactions that included broilers produced by peak production flocks (average egg weight) had better growth performance (higher final BW and lower total FC) than the other interactions.

IMPLICATIONS FOR FUTURE RESEARCH

Results from the current research are valuable for the hatching egg and broiler industry since they provide information on modern broiler breeder strains that are commonly used in western Canada. The results contained in this thesis could provide the hatching egg industry with answers to the variability observed between hatching eggs from different strains at different flock ages. For example, should the variations in fertility observed in the young C and R flocks be considered when shipping hatching eggs for incubation?. Irrespective of strain or flock age, fertility should always be tested at the farm level (e.g. by breaking open non-settable eggs (dirty, misshapen, small, or collected from the floor)) to prevent shipping unfertile eggs to the hatchery which not only present a risk for contamination but also take over space in the incubators. If the broiler industry is made aware that chicks produced by young breeder flocks may require a higher level of care, the implementation of different management practices could help to decrease early mortality and to enhance production performance.

Future research should study different incubation conditions (e.g. temperature, humidity, air velocity) for eggs from different strains and parent flock ages, in order to
determine whether or not eggs should be incubated in a different way to increase the number of saleable chicks.

Likewise, it is recommended that research analyzing different conditions in the broiler barn (e.g. temperature, stock density) should be performed. Specific environment conditions should be examined in order to improve the livability of broiler chickens from different strains and young parent flocks.

Research that could lead to a greater understanding of why chicks from young parent flocks do not have the same livability and growth potential than chicks from older parent flocks should be performed. Because a number of factors could be involved, research on embryonic metabolism, digestive system development, immunological status, and endocrine system regulation (specifically heat regulation) is advised. Based on the results obtained from basic science, applied research could be performed to cope with the physiological needs of chicks produced by young parent flocks.

TABLES

Ingredient	Starter diet	Grower diet	Finisher diet
	دوانی دو بیر نا نام با کر ور ایک ای دو ایک دو بو ای ور ور ای دو بیر ور	%	
Wheat, hard, grain	42.93	53.22	58.03
Soybean, meal, dehyd	26.87	16.23	15.10
Corn, yellow, grain	18.00	18.00	15.00
Fat, vegetable	3.77	3.36	4.13
Fish, meal menhaden	3.00	5.00	3.51
Calcium carbonate	1.50	1.05	1.07
Dicalcium phosphate	1.55	1.00	1.08
Salt, plain (NaCl)	0.42	0.34	0.36
L-Lysine	0.23	0.15	0.15
DL-Methionine	0.23	0.10	0.09
L-Threonine	0.05	0.10	0.03
Broiler Micronutrient Premix ¹	0.50	0.50	0.50
Vit. E (5000 IU/Kg)	0.30	0.30	0.30
Choline chloride	0.50	0.50	0.50
Avizyme 1302	0.05	0.05	0.05
Amprolium	0.05	0.05	0.05
Zn-bacitracin	0.05	0.05	0.05
Dietary component			
M. E. poultry (Kcal/Kg)	3,067.51	3,152.00	3,196.00
Choline (mg/Kg)	1,490.09	1,328.24	1,254.90
Protein, crude (%)	23.00	20.15	19.00
Fat, crude (%)	5.73	5.59	6.19
Fiber, crude (%)	2.47	4.28	2.51
Calcium (%)	1.10	0.90	0.85
Phos. Available (%)	0.50	0.45	0.42
Methionine (MET) (%)	0.60	0.46	0.42
MET + CYS	0.97	0.79	0.75
Lysine (%)	1.35	1.10	1.01
Tryptophan (%)	0.29	0.25	0.24
Threonine (%)	0.87	0.80	0.67
Isoleucine (%)	1.12	0.96	0.92
Valine (%)	1.18	1.02	0.97
Arginine (%)	1.46	1.19	1.11
Sodium (%)	0 23	0.21	0.21

Table 2-1. Ingredient percentages and calculated composition of the standard	d
broiler rations fed in the present experiment (starter, grower and finisher diet	ts)

¹Supplied the following per Kg of finished feed: Vitamin A, 10,000 IU; cholecalciferol, 2,500 IU; α-tocopherol acetate, 35 IU; iron, 100 mg; zinc, 80 mg; manganese, 70 mg; niacin, 65 mg; d-pantothenic acid, 14 mg; copper, 8.5 mg; riboflavin, 5 mg; pyridoxine, 4 mg; thiamine, 2 mg; menadione, 2 mg; folic acid, 0.8 mg; iodine, 0.5 mg; biotin, 0.18 mg; selenium, 0.1 mg; and vitamine B₁₂, 0.02 mg

	n¹	Egg weight (g)	Specific gravity	Albumen weight (%)	Wet yolk weight (%)	Dry yolk weight (%)	Dry shell weight (%)
Strain ²							
С	270	64.3	1.074	60.8	29.7	15.4	8.8
R	270	62.7	1.073	60.2	30.0	15.6	8.9
SEM ³		1.4	0.001	0.2	0.2	0.1	0.1
Flock age							
29 wk	180	55.8°	1.074	63.6ª	27.4 [°]	14.0 ^c	9.0 ^ª
45 wk	180	64.8 ^b	1.073	59.4 ^b	30.6 ^b	15.7 [⊳]	9.0ª
59 wk	180	69.8 ^a	1.074	58.6 ^b	31.5ª	16.8 ^ª	8.5 ^b
SEM		1.7	0.001	0.3	0.3	0.2	0.1
Strain*age							
C * 29 wk	90	53.8	1.069 ^d	63.3ª	27.8 ^c	13.9	8.7 ^{bc}
C * 45 wk	90	67.6	1.078 ^{ab}	60.2 [⊳]	30.1 ^b	15.5	8.9 ^b
C * 59 wk	90	71.3	1.075 ^{⊳c}	58.6 ^c	31.3ª	16.7	8.6 ^c
R * 29 wk	90	57.8	1.080 ^a	63.9 ^ª	26.8 ^d	14.0	9.2 ^ª
R * 45 wk	90	61.9	1.069 ^d	58.2°	31.3ª	15.8	9.2ª
R * 59 wk	90	68.3	1.072 ^{cd}	58.6°	31.7ª	16.8	8.3 ^d
SEM		2.4	0.001	0.4	0.3	0.2	0.1

Table 2-2. Effects of genetic strain, flock age and the interaction of strain*age on hatching egg characteristics

^{a-d}Means within the same column with different superscripts differ significantly (P \leq 0.05) ¹Number of eggs ²C= Cobb 500, R= Ross 308 ³Standard error of mean

	n¹	Fert ² (%)	Total hatch ³ (%)	Hatch fertile⁴ (%)	Weight Ioss⁵ (%)	Early dead ⁶ (%)	Mid dead ⁷ (%)	Late dead ⁸ (%)	Culls ⁹ (%)
Strain ¹⁰			······································	· · · · · · · · · · · · · · · · · · ·			--	`	
С	33	88.7	70.4	81.7	13.3	4.0 ^b	1.7	8.3	3.3
R	33	88.0	70.1	82.0	14.4	6.2ª	1.6	7.5	1.7
SEM ¹¹		0.6	1.8	1.9	0.1	0.4	0.3	1.0	0.6
Flock age									
29 wk	22	86.7 ^b	72.8 ^a	86.0 ^a	12.6 [⊳]	5.2	1.0 ^b	6.2 [⊳]	1.1 ^b
45 wk	22	91.5ª	64.4 ^b	72.8 [♭]	16.4 ^a	4.9	2.8 ^a	13.8ª	4.9 ^a
59wk	22	86.9 ^b	73.6ª	86.7ª	12.6 [⊳]	5.2	1.3 [⊳]	3.7 ^b	1.5 [⊳]
SEM		0.7	2.2	2.3	0.1	0.6	0.4	1.3	0.7
Strain*age									
C * 29 wk	11	76.7°	64.7 ^b	88.0	12.8	4.8	0.6 ^b	5.2	1.2
C * 45 wk	11	95.0 ^a	64.9 ^b	69.9	15.4	3.3	3.8ª	15.2	6.8
C * 59 wk	11	94.4ª	81.5ª	87.0	11.8	4.0	0.8 ^b	4.5	2.0
R * 29 wk	11	96.8ª	80.8ª	84.0	12.5	5.6	1.3 [⊳]	7.3	1.1
R * 45 wk	11	88.1 [⊳]	63.8 ^b	75.7	17.4	6.4	1.8 [⊳]	12.3	3.1
R * 59 wk	11	79.3°	65.6 ^b	86.3	13.4	6.4	1.7 [⊳]	2.8	1.1
SEM		1.0	3.1	3.3	0.1	0.8	0.6	1.8	1.0

Table 2-3. Effects of genetic strain, flock age and the interaction of strain*age on fertility, hatchability, egg weight loss, embryonic mortality, and culled chicks

^{a-c}Means within the same column with different superscripts differ significantly (P≤0.05) ¹Number of experimental units, each experimental unit = 18 eggs

²Fertility (%) = (number of fertile eggs/number of eggs set)*100

³Total hatchability (%) = (total number of chicks hatched/number of eggs set)*100
 ⁴Hatch of fertile (%) = (total number of chicks hatched/number of fertile eggs set)*100
 ⁵Weight loss at 18d of incubation (%) = [(egg weight at setting – egg weight at transfer) / egg weight at setting] * 100

⁶Early dead (%) =(number of embryos that died between 1 to 7 d of incubation/number of eggs set)*100

⁷Mid dead (%) =(number of embryos that died between 8 to 14 d of incubation/number of eggs set)*100

⁸Late dead (%) =(number of embryos that died between 15 to 21 d of incubation/number of eggs set)*100

⁹Culls (%) = (number of chicks culled at hatching/number of set eggs)*100

¹⁰C= Cobb 500, R= Ross 308

¹¹Standard error of mean

	n ¹	Piping ² time (hr)	Hatching ³ time (hr)
Strain⁴			
С	162	492.3	502.1
R	162	491.9	502.5
SEM⁵		1.5	5.9
Flock age			
29 wk	108	491.9	501.4
45 wk	108	493.4	502.5
59 wk	108	490.9	500.3
SEM		1.8	7.3
Strain*age			
C * 29 wk	54	494.4	503.9
C * 45 wk	54	492.3	502.1
C * 59 wk	54	490.1	500.3
R * 29 wk	54	489.4	499.2
R * 45 wk	54	494.6	502.8
R * 59 wk	54	491.6	500.4
SEM		2.6	10.3

Table 2-4. Effects of genetic strain, flock age and the interaction of strain*age on pipping time and hatching time

¹Number of eggs set ²Beak of embryo punctured through shell ³Chick was completely out of shell and down was dry ⁴C= Cobb 500, R= Ross 308 ⁵Standard error of mean

na an a	Chick weight ¹ (g)	21 d weight (g)	41 d weight (g)	Early weight gain ² (g)	Late weight gain ³ (g)	Overall weight gain⁴ (g)
Strain⁵						
С	44.2 (2082) ⁶	785.3 (2001)	2492.2 (1947)	741.3°	1707.9	2447.9
R	42.5 (2082) ⁶	768.5 (1999)	2479.0 (1949)	726.0 ^b	1709.1	2436.3
SEM ⁷	1.2	5.5	15.9	4.4	13.6	15.1
Flock age						
29 wk	38 4° (1428)	737 2 ^b (1374)	2438 9 ^b (1326)	699 1 ^b	1703 7	2400 2°
45 wk	$43.6^{b}(1308)$	$790.3^{a}(1252)$	$2525.9^{a}(1215)$	746 7 ^a	1735.0	2482.3 ^a
59 wk	48.1 ^a (1428)	803 1 ^a (1378)	2492 1 ^a (1357)	755 1 ^a	1686.9	2443 9 ^b
SEM	1.4	6.7	19.5	5.4	16.5	18,4
0 4 · • •						
Strain*age						
C * 29 wk	37.3 (714)	/16.8° (690)	2411.2 (665)	679.5°	1694.4	2373.9
C * 45 wk	46.4 (654)	813.8° (621)	2559.5 (606)	767.4ª	1745.7	2513.1
C * 59 wk	48.9 (714)	825.2ª (692)	2505.9 (677)	776.3ª	1679.8	2457.0
R * 29 wk	39.4 (714)	757.6 ^⁰ (683)	2466.6 (661)	718.2 ^⁰	1709.0	2427.2
R * 45 wk	40.9 (654)	766.8 [°] (629)	2492.2 (609)	725.9 [⊾]	1725.4	2451.3
R * 59 wk	47.3 (714)	781.0 ^b (687)	2478.3 (679)	733.7 ^b	1697.3	2431.0
SEM	2.0	9.5	27.5	7.6	23.4	26.0

Table 2-5. Effects of genetic strain, flock age and the interaction of strain*age on average chick and broiler body weights and weigh gains

^{a-c}Means within the same column with different superscripts differ significantly (P≤0.05) ¹Weight at hatching

²Early weight gain = 21 d weight – chick weight ³Late weight gain = 41 d weight – 21 d weight

⁴Overall gain = 41 d weight – chick weight

⁵C= Cobb 500, R= Ross 308

⁶Number of observations

⁷Standard error of mean

	n¹	Early feed ² (g/bird)	Early FC ³	Late feed ⁴ (g /bird)	Late FC⁵	Total feed (g /bird)	Total FC ⁶
Strain ⁷							
С	18	1035.1	1.40	3115 <i>.</i> 6⁵	1.83	4150.6	1.70 ^b
R	18	1021.9	1.41	3190.7 ^a	1.87	4212.5	1.73ª
SEM ⁸		6.8	0.01	20.3	0.01	23.9	0.01
Flock age							
29 wk	12	983.4 [°]	1.41 ^a	3101.4	1.83 ^b	4084.8 ^c	1.70 ^b
45 wk	12	1026.3 [⊳]	1.38 [♭]	3175.2	1.83 ^b	4201.6 ^b	1.69 [⊳]
59 wk	12	1075.6ª	1.42ª	3182.7	1.89ª	4258.3ª	1.74 ^a
SEM		8.3	0.01	24.9	0.02	29.3	0.01
Strain*age							
C * 29 wk	6	961.1 ^d	1.41	3102.7	1.84	4063.8	1.71 ^b
C * 45 wk	6	1048.4 ^b	1.37	3128.5	1.80	4176.9	1.66°
C * 59 wk	6	1095.7 ^a	1.41	3115.5	1.86	4211.1	1.71 ^b
R * 29 wk	6	1005.7°	1.40	3100.1	1.81	4105.8	1.69 ^{bc}
R * 45 wk	6	1004.3°	1.39	3222.0	1.87	4226.3	1.70 ^{bc}
R * 59 wk	6	1055.6 [⊳]	1.43	3249.9	1.92	4305.5	1.77 ^a
SEM		11.8	0.01	35.1	0.02	41.4	0.02

Table 2-6. Effects of genetic strain, flock age and the interaction of strain*age on early, late and total feed consumption, and feed conversion (FC)

^{a-d}Means within the same column with different superscripts differ significantly (P≤0.05) ¹Number of experimental units; each experimental unit= pen of 119 broilers (29 wk & 59 wk) or 109 broilers (45 wk)

²Early feed = cumulative feed intake/bird from 1 d to 21 d

³Early FC = FC from 1 d to 21 d. FCR= g feed / g weight gain

⁴Late feed = cumulative feed intake/bird from 22 d to 41 d

⁵Late FC = FC from 22 d to 24 d. FCR= g feed / g weight gain

⁶Total FC = g total feed / g total weight gain

⁷C= Cobb 500, R= Ross 308

⁸Standard error of mean

		Wk1	Early	Late mortality ³	Total mortality
	n ¹	mortality (%)	mortality ² (%)	(%)	(%)
Strain⁴					
С	18	0.5	3.9	2.6	6.5
R	18	0.6	4.0	2.4	6.4
SEM⁵		0.2	0.3	0.4	0.5
Flock age					
29 wk	12	1.0 ^a	3.8	3.4 ^a	7.2 ^a
45 wk	12	0.5 ^b	4,3	2.8ª	7.1 ^a
59 wk	12	0.1 ^b	3.5	1.5 [⊳]	5.0 ^b
SEM		0.2	0.4	0.4	0.6

Table 2-7. Effects of genetic strain and flock age on early, late and, total broiler mortality

^{a-b}Means within the same column with different superscripts differ significantly (P≤0.05) ¹Number of experimental units; each experimental unit= pen of 119 broilers (29 wk & 59 wk) or 109 broilers (45 wk)

²Early mortality = cumulative mortality between 1 d to 21 d

³Late mortality = cumulative mortality between 22 d to 41 d ${}^{4}C$ = Cobb 500, R= Ross 308

⁵Standard error of mean

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3. EGG YOLK AND NEWLY HATCHED CHICK YOLK SAC WEIGHT AND FATTY ACID CONTENT AS AFFECTED BY BROILER BREEDER GENETIC STRAIN, AGE AND EGG SIZE

INTRODUCTION

A typical chicken egg contains about 30% yolk, of which half is water and half is solid. Of the total solids in the yolk 60% is comprised of lipids and 30% of protein (Ahn et al., 1997). Through genetic selection the composition of the whole egg can be altered, thus, in broiler breeder lines selected for abdominal fat, an association between high abdominal fat and an increase in yolk weight, yolk dry matter and yolk fat content has been reported (Cahaner et al., 1986).

The egg yolk provides the developing embryo with nutrients from the start of vascularization and formation of the yolk sac, around 2 d of incubation, through hatching (Romanoff, 1960). At about 19 d of incubation the yolk sac starts withdrawal into the embryo's body through the navel. By the time of hatching this process should be complete and the navel closed (Romanoff, 1960). It has been estimated that more than 90% of the total energy requirements of the chicken embryo are provided by the β -oxidation of fatty acids derived from yolk lipids (Romanoff, 1960).

The lipids in the yolk are mainly (93%) very low density lipoproteins formed by different lipid classes: Triacylglycerol (67%), phospholipids (25%), free cholesterol (up to 5%), and trace amounts of cholesteryl ester and free fatty acids (Speake et al., 1998a). The relative proportions of these lipid classes are very similar across domestic avian

species and are not easily manipulated by dietary, physiological or pharmacological means (Speake et al., 1998a).

The major saturated fatty acids (SFA) palmitic ($C_{16:0}$), and stearic ($C_{16:0}$), as well as the monounsaturated fatty acids (MUFA) palmitoleic ($C_{16:1(n-7)}$) and oleic ($C_{18:1(n-9)}$) can be synthesized *de novo* in the maternal liver from dietary carbohydrate (Speake et al., 1998a). However, the incorporation of the long-chain polyunsaturated fatty acids (PUFA) linoleic ($C_{18:2(n-6)}$) and linolenic ($C_{18:3(n-3)}$) into the egg is dependant on the dietary provision of these PUFAs to the laying hen (Speake et al., 1998a). Corn and animal fats are the main source of linoleic acid ($C_{18:2(n-6)}$) (Pardio et al., 2005, Muma et al., 2006), while the addition of fish oil and certain oilseeds (flax seeds) to the hen's diet, promotes the deposition of linolenic acid ($C_{18:3(n-3)}$) in the egg (van Elswyk, 1997). The very long chain PUFAs arachidonic ($C_{20:4(n-6)}$) and docosahexaenoic or DHA ($C_{22:8(n-3)}$) acids can be synthesized by the maternal liver from linoleic ($C_{18:2(n-6)}$) and linolenic ($C_{18:3(n-3)}$) acids respectively (Ajuyah et al., 2003) or can be supplemented in the hen's diet (e.g. fish oil is rich in DHA) (van Elswyk, 1997).

In general, the main yolk FA in eggs laid by broiler breeder hens maintained on commercial diets are palmitic ($C_{16:0}$), oleic ($C_{18:1(n-9)}$), and linoleic ($C_{18:2(n-6)}$) acids (approximately 25%, 40%, and 15% of total FA respectively). Linolenic acid ($C_{18:3(n-3)}$) tends to be very low (1%). The long-chain PUFA of the n-6 and n-3 series (arachidonic, ($C_{20:4(n-6)}$), and DHA, ($C_{22:6(n-3)}$)) represent about 3% and 2% of the total FA, respectively (Nielsen, 1998).

Dietary manipulations are the main cause of variation in the FA composition of eggs, with most of the research being conducted in laying hens (Gao and Charter, 2000). As well, the composition of egg FA varies with breeder flock age. Palmitic ($C_{16:0}$) and stearic acid ($C_{18:0}$) concentrations were greater in eggs from older hens (51 wk and

64 wk of age) than in eggs from hens at 36 wk of age; while palmitoleic acid ($C_{16:1(n-7)}$) was present in the opposite proportion (Latour et al., 1998).

The lipids from the egg yolk undergo the most important changes during the last week of incubation (distinguished by intense lipid metabolism and rapid embryo growth) (Noble and Connor, 1984; Noble et al, 1986). It is during this period that rapid absorption of yolk lipids (mediated by the yolk sac membrane) and accumulation of lipids in embryonic tissues occur (Peebles et al., 1999). Lipid utilization by the avian embryo is a complex process directed by the requirements of particular embryonic tissues for specific fatty acids (Speake et al., 1998b). Of particular interest is the large content of DHA in the brain and retina, and the predominance of arachidonic acid in organs such as the heart, kidneys and liver (Speake and Deans, 2004). Phospholipids rich in DHA facilitate neurotransmission and photoreception (Salem et al., 2001), while arachidonic acid is involved in mechanisms such as heartbeat regulation (Pavoine et al., 1999).

In post-hatch chicks, the residual yolk sac (RYS) content is absorbed and utilized preferentially for growth of the small intestine and also as a supply of energy (Noy and Sklan, 1999). A direct positive relationship exists between the amount of nutrients provided by the RYS and the subsequent performance of the broilers (Murakami et al., 1992; Latour et al., 1994; Vieira and Moran Jr, 1999). Deeming (2005) reported that the RYS of day-old Ross chicks represented 9% of their total BW. Different results were reported by Mikec et al. (2006) in the same strain of chicks: RYS was approximately 14% of the total BW. However, the age of the broiler breeders or the initial egg weight were not specified by the authors. Jamroz et al. (2004) reported that the RYS of day-old broiler chicks from the Shaver-Starbro line, was composed of approximately 15% lipid and 26% protein.

In layer hens, the FA composition of the RYS of chicks is affected by variation in the diet and these values for 43 wk old Single Comb White Leghorns have been provided by Latour et al. (2000).

The FA content of RYS (Latour et al., 2000), embryonic tissues (Peebles et al., 1999; Surai and Sparks, 2001), and diverse tissues from the newly hatched chick (Cherian and Sim, 1993, 1997) have been reported. Results from Cherian et al. (2002) in laying hens showed an increase in SFA in the liver as well as an increase in conjugated linoleic acid (CLA- a mixture of isomers of linoleic acid, $(C_{18:2(n-6)})$) contents in the thigh and breast muscles in birds fed CLA. Additionally, when maternal dietary CLA was increased, liver, plasma, adipose tissue, heart, and brain tissues of the day-old chick had greater levels of SFA and stearic acid (C_{18:0}) as well as lower MUFA and oleic acid (C_{18:1(n-9)}) (Cherian et al., 2005).

When comparing newly hatched chicks from an Arbor Acres flock at 36, 51 and 64 wk of age, Latour et al. (1998) reported greater palmitic ($C_{16:0}$) concentrations in RYS than in fresh yolks. In the same study, a pronounced decrease (~3.5 times) in palmitoleic ($C_{16:1(n-7)}$) acid was observed in the RYS vs. the egg at 36 wk, however, no differences were observed for this FA at 51 or 64 wk of age. Noble et al. (1986) and Latour et al. (1996, 1998) have reported that there is a reduction in transfer of yolk lipids into the developing chick embryo in embryos from eggs produced by young flocks. As a result, fat metabolism may be compromised in these chicks, which, in turn, could predispose these chicks to higher mortality.

There is limited research on the effects of broiler breeder genetic strain, flock age, and egg size in the FA composition of the hatching egg and RYS. Research relating the FA content in hatching eggs to RYS of the day old chick of modern strains is limited.

Objectives

The first objective of this study was to determine the effects of genetic strain, flock age and egg size on the FA content in hatching eggs and in the RYS from newly hatched chicks. A second objective was to analyze the relationship between the FA content of eggs and the FA content of RYS at different ages and from different genetic strains and egg sizes.

Hypotheses

It was hypothesized that:

1) Eggs from the young flocks would have a higher content of PUFA than eggs from the other flock ages.

2) Chicks hatched from the young flocks would have a smaller RSY and the RYS would have a higher content of PUFA than that of chicks from the older flocks.

3) Correlations between egg FA and RYS FA composition would be significant.

MATERIALS AND METHODS

The experimental protocol was approved by the Faculty of Agriculture, Forestry and Home Economics, Faculty Animal Policy and Welfare Committee (FAPWC) at the University of Alberta in accordance with the guidelines set forth by the Canadian Council on Animal Care (1993).

Egg Collection

A total of 504 hatching eggs produced by two modern commercial broiler breeder strains (Cobb 500 (C) and Ross 308 (R), n= 252 eggs/strain) were obtained from a commercial hatchery using the same breeder flocks at 29, 45 and 59 wk of age. The C strain has been genetically selected for high white meat yield (particularly breast muscle) while the R strain has been selected for the further processing sector (whole bird market).

At each collection time the average egg weight for each strain was determined by weighing 72 eggs/strain. Based on the average egg weight, eggs were collected according to three weight ranges: Small (S), Medium (M) and Large (L). Eggs from the same weight category were within \pm 1.5 g of each other and were selected in the following way: the M eggs were \pm 1.5 g from the average egg weight, the S eggs were 3.0 to 6.0 g lighter than the average and the L eggs were 3.0 to 6.0 g heavier than the average and the L eggs were transported to the Alberta Hatching Egg Producers Hatchery.

Egg Sampling

At each of the flock ages 10 eggs within each strain x weight group were randomly selected, weighed and broken open. After recording wet yolk weight, the albumen and yolk from each egg were mixed with a manual blender¹, individual samples (~12 mL) were put into 15 mL plastic tubes, sealed and placed in a freezer (-18°C) for future gas chromatography analysis.

¹ MAHHB2CAN. Columbia, MO, USA, 65205.

Incubation and Hatching

Remaining eggs (n= 18 eggs/age x strain x weight) were numbered, weighed and candled; any eggs with hairline cracks were replaced with spare eggs from the same strain and weight category. Each treatment group of 18 eggs was incubated along with 2,376 additional eggs (these eggs were used for the research conducted in chapter 2 of this thesis). The eggs were randomly allocated (to account for possible environmental differences) within a 5,000 egg capacity Jamesway² single stage setter and incubated for 18 d at a dry bulb and wet bulb temperature of 37.5°C and 29.4°C respectively.

At 18 d of incubation the groups of 18 eggs were removed from the setter, individually weighed, and put into pedigree hatch baskets (122 cm x 35 cm). The baskets were placed into a 5000 egg capacity Jamesway hatcher and incubation continued for an additional 3.5 d at a dry bulb temperature of 35.2°C and a wet bulb temperature of 29.4°C.

At 21.5 d of incubation all hatched chicks from the pedigree baskets were weighed and euthanized by cervical dislocation. The RYS was dissected from each chick and wet weights of the carcass (yolk free body mass (YFBM)) and RYS were recorded. Each RYS (membrane and yolk sac content) was mixed with 5 mL of distilled water and stored in sealed 15 mL-plastic tubes in a freezer (-18°C) for future gas chromatography analysis. Ten RYS within each strain x age x size group were analyzed.

Lipid Analysis

Approximately 0.2 g of the hatching egg components (yolk + albumen) or RYS (membrane + content) were used to extract total lipid using the chloroform:methanol

² Jamesway Incubator Company Inc., Cambridge, ON, Canada, N1R 7L3

(2:1, vol/vol) modified Folch's method (1957). The lipid extracts were dried under nitrogen and methylated (converted to fatty acid methyl esters (FAME)) using a mixture of boron-trifluoride, hexane, and methanol (35:20:45, vol/vol/vol) according to the method of Metcalfe et al. (1961). The FAME were recovered with hexane prior to quantification by an automated gas chromatograph. The samples (5uL) were injected into a Varian 3400 Gas Chromatograph³ equipped with a Varian 8100 autosampler, on column SPI injector, and flame ionization detectors. The analytical column (a SGE BPX-70⁴) had a 30m x 0.25mm inside diameter x 0.25µm film thickness. The carrier gas, helium, was maintained at a flow rate of 1-1.5mL/min through the column. The initial column temperature (50°C) was maintained for 0.1 min and increased by 25°C/min to 170°C. At this point, the temperature was maintained for 1 min, then increased to 180°C at 2°C/min. A final temperature increase to 230°C occurred at 10°C/min where the sample was maintained for 3 min. The initial injector temperature of 70°C increased by 150°C/min to 230°C where it was held for 16 min. The detector temperature was set at 230°C. The calibration and identification of FA peaks was carried out by comparison with retention times of known authentic standards⁶. The FA results are presented as a percentage of total FA, and were determined using the Shimadzu EZChrom chromatography data system⁷.

Statistical Analysis

Each hatching egg yolk/albumen sample or RYS sample was the experimental unit for this research. The experimental design was a $2 \times 3 \times 3$ factorial arrangement with strain (C and R), age (29, 45, and 59 wk), and egg weight (S, M, and L) as the main

³ Varian Inc., Walnut Creek, CA, USA, 94598

⁴ SGE Inc., Austin, TX, USA, 78758

⁶ Standard 463, Nu Chek Prep, Elysian, MN, USA, 55612

⁷ Shimadzu Scientific Instruments Inc., Columbia, MD, USA, 21046.

effects. Egg weight ranges were different for each strain and for each age, thus, to eliminate the variability due to egg size categories, egg size was used as a covariable (as a nested factor in strain and age) when analyzing egg and chick components.

Because the exact ingredients of the commercial diets fed to the broiler breeders were not made available, the FA profiles were analyzed within each strain in a 3 x 3 factorial experimental design with age (29, 45, and 59 wk of age), and egg size (S, M, and L) as main effects. The statistical model for the analysis of variance within each strain was:

$$Y_{ijk} = \mu + A_i + S_j + A_i S_j + \mathcal{E}_{ijk}.$$

Where Y_{ijkl} = characteristic that was measured, μ = overall mean, A_i = main effect of parent flock age, S_j = main effect of egg size, A_iS_j = interaction between flock age and egg size, and ε_{ijkl} = random error term.

All percentage data were subjected to angular transformation to stabilize variances (arc sine square root percentage transformation) prior to statistical analysis. All data were analyzed using the mixed model procedure of SAS® (Proc Mixed) (SAS Institute, 2002) and the probability level set at P≤0.05. When the model indicated significance, the means were separated using the LSMEANS procedure of SAS®.

The association between the FA composition of the yolk/albumen sample and RYS was measured using the Pearson Correlation Coefficient (r). Values with an $r \ge 0.7$ were considered correlated, and with $r \ge 0.9$ highly correlated.

The FA content of the egg and the RYS were compared using the TTEST procedure of SAS®.

RESULTS

Egg, Chick, and Residual Yolk Sac Weights

<u>Breeder Strain</u>. The weight of the hatching egg and the associated chick weight, and RYS weight did not differ between strains (Table 3-1).

<u>Breeder Flock Age</u>. All parameters measured varied due to breeder flock age (Table 3-1). Since there was an interaction effect, yolk weight as affected by age will not be discussed. Average egg weight increased with each increase in breeder flock age. Chick weight at hatching reflected the egg weight at setting, and their correlation was positive and highly significant (r= 0.96, p<0.0001). Chick weight at hatching, as a percentage of the egg set weight, was about 70% for the 29 and 59 wk old flocks (which did not differ from each other), however, chicks hatching from the 45 wk old flocks were proportionally smaller (68%) than chicks from the other flock ages. As a percentage of the chick weight the RYS of the chicks that hatched from the young flocks were smaller than those from the older flock ages, which did not differ. In contrast, wet carcass weight as a percentage of the hatched chick weight was greater at 29 wk of age compared to the older flocks. The correlation between chick weight and RYS weight was positive and significant (r= 0.85, p<0.0001).

<u>Breeder Strain and Flock Age Interaction</u>. The only interaction observed was for yolk weight as a percent of the hatching egg. Percent yolk weight in the C eggs increased with each increase in age while the R eggs increased from, 29 to 45 wk but did not differ between 45 and 59 wk (Table 3-1).

Because maternal diet can influence FA composition of the eggs, eggs from the same flocks for both the C and R strains were examined at all flock ages to minimize dietary effects. Since the hatching eggs analyzed in the current study were obtained from commercial flocks, neither a sample of the diet nor the specific dietary composition was made available to the researchers.

Fatty Acid Composition of Yolk/Albumen Samples from the C Flock

The FA composition of eggs laid by C hens is presented in Table 3-2.

<u>Breeder Flock Age</u>. Flock age did not affect the proportions of total SFA, MUFA or PUFA of eggs laid by C hens. However, the p-values for SFA (0.0606) and MUFA (0.0562) approached significance; the trend for SFA was to increase and for MUFA to decrease with age. Individual FA were present in different proportions at different ages. Myristic (C14:0), palmitic (C_{16:0}), and linolenic (C_{18:3(n-3)}) acids increased from 29 to 45 wk of age, but oleic (C_{18:1(n-9)}) and arachidonic (C_{20:4(n-6)}) acids decrease at 59 wk of age. The proportions of other FA were not influenced by flock age.

Egg Size. There were significant differences in the proportions of total SFA, MUFA and PUFA. The M eggs had lower SFA and PUFA but higher MUFA than S or L eggs, which did not differ from each other. Individual FA were also affected by egg size: M eggs had lower percentages of the main FA than S or L eggs which shared the same FA composition; this result was observed for palmitic ($C_{16:0}$), stearic ($C_{18:0}$), palmitoleic ($C_{16:1(n-7)}$), linoleic ($C_{18:2(n-6)}$), arachidonic ($C_{20:4(n-6)}$) and DHA ($C_{22:6(n-3)}$). For oleic acid ($C_{18:1(n-9)}$) M eggs had a greater percentage of this FA compared to S or L eggs which did not differ. <u>Breeder Flock Age and Egg Size Interaction</u>. The only interaction effect was on palmitoleic acid ($C_{16:1(n-7)}$) with no particular pattern emerging. Eggs from the S*45 wk group had the largest amount of this FA (but not statistically different from S*59 wk or L*59 wk), while M*59 wk eggs had the lowest value of palmitoleic acid compared to all other egg groups (data not shown).

Fatty Acid Composition of Residual Yolk Sacs From the C Flock

The FA composition of the RYS of chicks hatched from the C eggs is presented in Table 3-3.

<u>Breeder Flock Age</u>. Except for stearic acid ($C_{18:0}$), all the FA in the RYS of C chicks were affected by flock age. Total SFA were greater at 45 than at 59 wk while SFA at 29 wk did not differ between the other flock ages. In contrast, total MUFA were greater at 59 than at 45 wk, and MUFA at 29 wk did not differ between the other ages. Total PUFA were greater at 45 wk than at 29 and 59 wk of age which, did not differ from each other.

Myristic acid ($C_{14:0}$) decreased with each increase in flock age. Palmitic ($C_{16:0}$), palmitoleic ($C_{16:1(n-7)}$), and linoleic ($C_{18:2(n-6)}$) acids decreased as flock age increased from 45 to 59 wk. Conversely, oleic acid ($C_{18:1(n-9)}$) increased as flock age increased from 45 to 59 wk. Arachidonic acid ($C_{20:4(n-6)}$) increased from 29 to 45 wk of age and did not change at 59 wk of age. Linolenic ($C_{18:3(n-3)}$) and DHA ($C_{22:6(n-3)}$) were lowest at 29 wk, highest at 45 wk and intermediate at 59 wk of age.

Egg Size. No significant effects in the FA content of RYS were observed for egg size.

<u>Breeder Flock Age and Egg Size Interaction</u>. The only significant interaction effect on the FA content of RYS was for palmitoleic acid ($C_{16:1(n-7)}$). Although no particular pattern was observed, RYS from the S*45 wk group had the largest amount of this FA (but not statistically different from L*29 wk or S*29), while M*45 and the three egg sizes interactions at 59 wk of age had the lowest value of this FA compared to the other egg groups (data not shown).

Correlation Between the FA Content of Eggs and Residual Yolk Sacs From the C Flock

<u>Breeder Flock Age</u>. When analyzing the correlation between the FA content of eggs and RYS for each flock age, statistical significance was observed at 59 wk for myristic ($C_{14:0}$), palmitic ($C_{16:0}$), palmitoleic ($C_{16:1(n-7)}$), DHA ($C_{22:6(n-3)}$), and total SFA (Table 3-4). However, the r value for these comparisons was well below 0.7 indicating a very low correlation between these FA in the egg and RYS.

<u>Egg Size</u>. Although statistical significance was observed for linolenic ($C_{18:3(n-3)}$) acid in the S and L groups, and for linoleic ($C_{18:2(n-6)}$) and total MUFA in the M group, the correlations for these FA were low (Table 3-4). The correlations between PUFA in egg and RYS was moderate for the M group with the only positive correlation being observed for linolenic ($C_{18:3(n-3)}$) in this group.

Comparison Between the FA Content of Eggs and Residual Yolk Sacs From the C Flock

<u>Breeder Flock Age</u>. When comparing the FA content of the egg vs. the RYS at each of the three flock ages, in almost all cases, the egg had significantly higher percent

FA than the RYS (Table 3-5). The exception to this was for myristic acid ($C_{14:0}$) for the 29 wk old flock, and for oleic acid ($C_{18:1(n-9)}$) and the total MUFA for all flock ages.

<u>Egg Size</u>. When comparing the FA content of the egg vs. the RYS for each egg size, again in almost all cases the egg had significantly higher FA percentages than the RYS (Table 3-6). The exceptions to this were, again, for myristic acid ($C_{14:0}$) for the M and L eggs, and for oleic acid ($C_{18:1(n-9)}$) and the total MUFA for all egg sizes.

Fatty Acid Composition of Yolk/Albumen Sample From the R Flock

The FA composition of eggs laid by R hens is presented in Table 3-7.

<u>Breeder Flock Age</u>. Flock age affected the proportions of total SFA, MUFA and PUFA of eggs laid by R hens. Eggs laid at 29 wk had lower SFA than eggs laid at 45 wk, with percentage of total SFA in eggs laid at 59 wk not differing from the other two ages. Total MUFA were lower in eggs laid at 45 wk than in eggs laid at 29 and 59 wk, which did not differ from each other. Total PUFA were higher at 29 and 45 wk than at 59 wk of age. All other FA (except for arachidonic acid ($C_{20:4(n-6)}$)) were significantly affected by flock age. Myristic ($C_{14:0}$), palmitic ($C_{16:0}$), stearic ($C_{18:0}$) and palmitoleic ($C_{16:1(n-7)}$) increased from 29 to 45 wk, with all but myristic acid ($C_{14:0}$) declining at 59 wk of age. Oleic acid ($C_{18:1(n-9)}$) mirrored total MUFA. The percent of linoleic acid ($C_{18:2(n-6)}$) was the same at 29 and 45 wk but decreased at 59 wk. Linolenic acid ($C_{18:3(n-3)}$) decreased with each increase in flock age, DHA ($C_{22:6(n-3)}$) decreased from 29 to 45 wk of age.

<u>Egg Size</u>. Egg size did not affect the proportions of total SFA, MUFA, PUFA or individual FA in eggs laid by R hens.

<u>Breeder Flock Age and Egg Size Interaction</u>. Three interactions were significant for the FA content of eggs laid by the R flock (data not shown). Although no particular pattern was observed for either FA, eggs from the M*29 wk, L*29wk and S*45 wk had the lower percentage of myristic acid ($C_{14:0}$) (but not statistically different from M*59 wk) than the other age by egg size interactions. Similarly, palmitoleic acid ($C_{16:1(n-7)}$) was lower in eggs from the M*29 wk, and L*29wk than in all other age by egg size interactions. The opposite was observed for linolenic acid ($C_{18:3(n-3)}$) which was greater in the S, M and L eggs of the 29 wk age interaction and lower in the S, M and L eggs of the 59 wk age interaction.

Fatty Acid Composition of Residual Yolk Sacs From the R Flock

The FA composition of the RYS of chicks hatched from the R flock is presented in Table 3-8.

<u>Breeder Flock Age</u>. The total SFA of RYS at 29 wk were lower than at 45 or 59 wk of age which did not differ from each other. No differences were observed in the total MUFA of RYS at different flock ages. Total PUFA decreased with each increase in flock age. The proportion of individual FA was affected by flock age. Myristic ($C_{14:0}$), stearic ($C_{18:0}$), and palmitoleic ($C_{16:1(n-7)}$) acids increased from 29 to 45 wk but did not increase further at 59 wk of age. The percentage of palmitic acid ($C_{16:0}$) increased from 29 to 45 wk, then decreased at 59 wk of age. Most PUFA (linoleic ($C_{18:2(n-6)}$), linolenic ($C_{18:3(n-3)}$), and DHA ($C_{22:6(n-3)}$) decreased from 29 to 45 wk, but only linoleic ($C_{18:2(n-6)}$) decreased further at 59 wk of age.

<u>Egg Size</u>. Egg size only influenced the linolenic acid ($C_{18:3(n-3)}$) content of RYS. This FA was greater in L than S or M eggs, which did not differ from each other.

<u>Breeder Flock Age and Egg Size Interaction</u>. The only interaction observed in the FA content of RYS was for palmitoleic acid ($C_{16:1(n-7)}$). RYS from the S, M and L egg size groups at 29 wk of age had lower palmitoleic acid ($C_{16:1(n-7)}$) than RYS from other interaction groups (data not shown).

Correlation Between the FA Content of Eggs and Residual Yolk Sacs From the R Flock

<u>Breeder Flock Age</u>. When analyzing the correlations between the FA content of eggs and RYS for each flock age, only linoleic ($C_{18:2(n-6)}$) and linolenic ($C_{18:3(n-3)}$) showed statistical significance. However, the FA in the egg and RYS had a low correlation (Table 3-9).

<u>Egg Size</u>. When comparing the FA content of eggs and RYS between the three egg sizes, only linolenic acid ($C_{18:3(n-3)}$) was correlated in S and M eggs. Although DHA ($C_{22:6(n-3)}$) in the M group, and stearic ($C_{18:0}$) and linolenic ($C_{18:3(n-3)}$) for the L group also showed statistical significance, these FA had low correlation (Table 3-9).

Comparison Between the FA Content of Eggs and Residual Yolk Sacs From the R Flock

<u>Breeder Flock Age</u>. When comparing the FA content of the egg vs. the RYS, significant differences occurred at each of the three flock ages for palmitoleic ($C_{16:1(n-7)}$), arachidonic ($C_{20:4(n-6)}$), and DHA ($C_{22:6(n-3)}$) (Table 3-10). At the three flock ages these three FA were greater in the egg than in the RYS. Total SFA, palmitic ($C_{18:0}$), and stearic ($C_{18:0}$) acids at 29 wk of age, and total PUFA at 45 and 59 wk of age were also greater in

the egg than in the RYS. The only FA that was significantly greater in the RYS than in the egg was myristic acid ($C_{14:0}$) at 29 wk of flock age.

Egg Size. When comparing the FA content of the egg vs. the RYS for each egg size, again significant differences were observed for each egg size for palmitoleic ($C_{16:1(n-7)}$), arachidonic ($C_{20:4(n-6)}$), and DHA ($C_{22:6(n-3)}$) (Table 3-11). These three FA were greater in the egg than in the RYS for each egg size group. Additional differences were observed in L eggs for palmitic ($C_{16:0}$), stearic ($C_{18:0}$), total SFA, and total PUFA; these FA were greater in the egg than in the RYS. Oleic acid ($C_{18:1(n-9)}$) and total MUFA were greater in the RYS than in the RYS.

DISCUSSION

Egg, Chick, and Residual Yolk Sac Weights

<u>Genetic Strain</u>. For the main effect of genetic strain it was expected that C would have larger eggs than R as this has been anecdotally reported by the hatching egg industry. Although numerically larger, the C eggs were not statistically different from the R eggs. Likely, because egg size did not differ, none of the other egg or chick weight parameters were affected by strain.

<u>Breeder Flock Age</u>. Previous research has clearly shown that average egg weight increases as breeder flocks age (Wiley, 1950; McNaughton et al., 1978), and that a positive and highly significant correlation between egg weight and chick weight exists (Guill and Washburn, 1973; Shanawany, 1987). The results of the current study support this. A result that was not expected, was that chicks produced by the 45 wk old flocks were the smallest (as a percentage of egg weight). In examining possible reasons for this, it was discovered that the temperature in the setter throughout the last two weeks of

incubation for this flock age was higher than recommended (average of 38.4 C vs. recommended 37.5 C). It is well known that high temperatures during incubation increase egg weight loss (Tullet, 1990) and can produce more water loss (dehydration) in the chicks (Tullet and Burton, 1982). It was thus hypothesized that the difference in percentage chick weight at hatching for the 45 wk old flocks may have been a consequence of chick dehydration rather than a flock age effect.

The positive and significant correlation between chick weight and RYS weight as well as the result of smaller RYS in chicks from younger flocks in the present study were in agreement with results reported by Burnham et al. (2001). Although in their research the broiler breeders were closer in age (26, 28 and 30 wk), Burnham and colleagues hypothesized that embryos from younger hens may have a relatively higher rate of yolk uptake later in the incubation period in an effort to compensate for possible deficiencies in yolk nutrient content. This could result in a smaller percent RYS at hatching (as reported in this thesis). The smaller RYS may contribute to poor chick quality in offspring produced by young breeders because these chicks may have a smaller reserve of energy for early development post hatching. Research answering these possibilities may produce further evidence why chicks from young flocks characteristically have suboptimal quality.

<u>Breeder Strain and Flock Age Interaction</u>. Just as reported by Suarez et al. (1997) in Arbor Acres breeders, this research also showed differences in the proportion of yolk in hatching eggs due to a strain and flock age interaction. Because the egg yolk provides the energy for the developing embryo (Romanoff, 1960) these differences may influence the embryo development and survival. The results from this study show that chicks from the C and R strains at 29 wk of parent flock age may have a disadvantage as the percentage of egg yolk is smaller compared to the other ages.

Fatty Acid Composition of Yolk/Albumen Sample From the C Flock

<u>Breeder Flock Age</u>. The results showing lower levels of palmitic ($C_{16:0}$) as well as a greater proportion of oleic ($C_{18:1(n-9)}$) acid in eggs from the younger age are in agreement with those of Latour et al. (1998) who compared eggs from 36, 51, and 64 wk old flocks. While the differences observed in the various other FA of eggs from the C strain at different parent ages could be due to dietary lipids (van Elswyk, 1997; Pardio et al., 2005; Muma et al., 2006), the inclusion of FA in different proportions by hens of different ages should not be discounted. Future research with diets of known composition during the entire production period of a flock would answer this question.

Egg Size. The greater proportion of MUFA observed in M eggs when compared to the S or L eggs was a consequence of the greater proportion of oleic acid ($C_{18:1(n-9)}$) observed for this egg size. Similarly, the larger proportions of total SFA and PUFA in the S and L eggs reflect the composition individual FA. No previous research comparing the composition of hatching eggs of different sizes has been completed. The physiological meaning of a greater percentage of oleic acid ($C_{18:1(n-9)}$) in average size eggs in unknown.

Fatty Acid Composition of Residual Yolk Sacs From the C Flock

<u>Breeder Flock Age</u>. The lower levels of some FA (myristic ($C_{14:0}$), palmitic ($C_{16:0}$), and palmitoleic ($C_{16:1(n-7)}$)) observed at the oldest flock age agree with those reported by Latour et al. (1998). However, the lower level of linoleic acid ($C_{18:2(n-6)}$) in RYS from chicks produced by 59 wk old parents disagrees with that reported by Latour et al. (1998). This result could be an effect of variations in the hen's diet (Cherian and Sim, 1997; Latour et al., 2000), or could reflect a greater need for this FA or a better ability for chicks from old flocks to metabolize this FA. Egg Size. There is no previous research on the effects of egg size on the FA composition of RYS. Data from the current research show that egg size at setting does not influence the FA profile of the RYS. The fact that the FA profile in eggs was affected by egg size showing greater percentage of oleic acid ($C_{18:1(n-9)}$) in M than in S or L eggs, and that this result was not mirrored by the FA in the RYS poses the question as to whether or not embryos from M eggs have different metabolism for this FA.

Correlation Between the FA Content of Eggs and Residual Yolk Sacs From the C Flock

The positive correlation observed for linolenic acid ($C_{18:3(n-3)}$) in the M group shows that the percentage of this FA in the RYS is dependant of its percentage in the egg. This is an interesting finding and could possibly be used to help improve chick quality. Linolenic ($C_{18:3(n-3)}$) acid is a known precursor of DHA ($C_{22:8(n-3)}$) (Speake and Deans, 2004) and DHA is essential for maintaining membrane integrity and facilitating neurotransmission and photoreception in the brain and retina (Salem et al., 2001). Thus, the author of this thesis hypothesizes that if linolenic ($C_{18:3(n-3)}$) acid is increased in the egg by dietary provision to the laying hen (e.g. by adding flax seeds) more linolenic ($C_{18:3(n-3)}$) acid will be made available in the RYS for absorption by the chick. Because under normal production conditions it is expected that most hens (regardless of flock age) would lay eggs of an M (average) size, the dietary supplementation with linolenic acid ($C_{18:3(n-3)}$) would likely benefit a high percentage of chicks.

Comparison Between the FA Content of Eggs and Residual Yolk Sacs From the C Flock

The percentage of myristic acid ($C_{14:0}$) was greater in the RYS than in the egg at 29 wk of age, and in M and L eggs. The greater content of oleic acid ($C_{18:1(n-9)}$) in the RYS at all parent flock ages and egg sizes is in agreement with that reported by Speake et al. (1998a) but no hypothesis was provided for this. The greater total MUFA observed in the RYS at all flock ages and egg sizes was a consequence of the greater amount of oleic acid ($C_{18:1(n-9)}$) present in the RYS. It was expected that during incubation most FA would be either oxidized to provide energy or incorporated into the embryo's tissues (Speake et al. ,1998a). Thus most FA were expected to be lower in the RYS vs. the egg. The differences observed for the FA listed above could be a result of different mechanisms affecting the oxidation of oleic ($C_{18:1(n-9)}$) and myristic ($C_{14:0}$) acids in these embryos. It was hypothesized that these two FA may play a more important role in post-hatching growth than in embryonic development.

Fatty Acid Composition of Yolk/Albumen Sample From the R Flock

<u>Breeder Flock Age</u>. As a consequence of the effect of flock age on most individual FA, total SFA, MUFA and PUFA were significantly different between flock ages. Some differences in percent of specific FA agree with those of Latour et al. (1998) who reported lower levels of stearic acid ($C_{18:0}$) in younger (36 wk) than older Arbor Acres breeders (51 wk and 64 wk), and with Nielsen (1998) who reported higher levels of DHA ($C_{22:6(n-3)}$) in eggs from young white Lohmann hens. Results from the current research however, disagree with Latour et al. (1998) who reported lower levels of palmitic ($C_{16:0}$) as well as a greater proportion of oleic ($C_{18:1(n-9)}$) acid in eggs from the younger age. In the current research these FA did not differ between the young and old flocks. As previously stated in this chapter it is important to note that proportions of longchain PUFA in eggs can be altered by manipulations of the hen's diet (van Elswyk, 1997; Pardio et al., 2005; Muma et al., 2006), hence, the results observed in the present experiment in these FA should be reviewed with this in mind.

<u>Egg Size</u>. No differences were observed for total or individual FA in eggs from different sizes for the R strain, and to the author's knowledge this is the first time data of this nature has been recorded.

Fatty Acid Composition of Residual Yolk Sacs From the R Flock

<u>Breeder Flock Age.</u> The lower levels of some FA (myristic (C14:0), palmitic (C16:0), and palmitoleic (C16:1(n-7))) observed in the RYS of chicks hatched from the young parent flock, as well as the lower level of linoleic acid (C18:2(n-6)) in the RYS of chicks hatched from the 59 wk old breeders disagree with those reported by Latour et al. (1998). Because the long-chain PUFAs of the RYS can be affected by variations in the hen's diet (Cherian and Sim, 1997; Latour et al., 2000), the changes observed in these type of FA should not be considered a singular effect of flock age.

Egg Size. Prior to reporting the data in this thesis there was no information on the effects of egg size on the FA composition of RYS. From this study, the only significant difference observed was for linolenic acid ($C_{18:3(n-3)}$), however, the author is sceptical if there is a biological meaning for this result. The role of linolenic ($C_{18:3(n-3)}$) acid as a precursor of DHA ($C_{22:6(n-3)}$) (Speake and Deans, 2004), which is essential for the proper functioning of the brain and retina (Salem et al., 2001) leads the author to hypothesize that chicks hatching from L eggs (which showed a greater percentage of linolenic ($C_{18:3(n-3)}$) in the RYS) may not have the ability to properly incorporate this FA into the tissues

during incubation. This later fact may affect post-hatch quality by impairing normal neurological development in these chicks.

Correlation Between the FA Content of Eggs and Residual Yolk Sacs From the R Flock

<u>Breeder Flock Age</u>. Although the correlation between the FA content of eggs and the RYS was statistically significant for linoleic acid (C18:2(n-6)) and linolenic acid (C18:3(n-3)), the correlation was low.

Egg Size. The positive correlation observed between the linolenic acid ($C_{18:3(n-3)}$) content of eggs and RYS in the S and M groups shows that the percentage of this FA in the RYS is dependant of its percentage in the egg. The biological importance of linolenic acid ($C_{18:3(n-3)}$) has been described previously in this chapter of the thesis, and the same hypothesis presented in the discussion of the C flock is presented for the R flock. If the hens are provided with a diet rich in linolenic acid ($C_{18:3(n-3)}$) (e.g. by adding flax seeds) more linolenic ($C_{18:3(n-3)}$) acid would be made available in the RYS for absorption by the chick. This could be used as a method of improving chick quality by enhancing the development of the neurological system including the retina. It is hypothesized that by this mean, neurological malformations and blindness could be prevented. Further research should analyze these possibilities.

Comparison Between the FA Content of Eggs and Residual Yolk Sacs From the R Flock

The lack of a significant difference between the oleic acid $(C_{18:1(n-9)})$ content of the egg vs. the RYS at all parent flock ages and in the S and M egg sizes is in disagreement
with that reported by Speake et al. (1998a), although the genetic strain was not specified by the authors. This FA was expected to be greater in the RYS than in the egg. According to that reported by Maldjian et al. (1995) in commercial R breeders up to 80% of the total palmitic ($C_{16:0}$) and stearic ($C_{18:0}$) acids that is transferred to the embryo during incubation is oxidised for energy. As a result, these FA were expected to be lower in the RYS vs. the egg, these was held true for the 29 wk group and for the L group. However, the remaining age and egg size groups did not show significant differences for palmitic ($C_{16:0}$) or stearic ($C_{18:0}$) acids. It was hypothesized by the author of this thesis that these results reflect an increased demand for energy (specially during the hatching process) and thus an increased oxidation of these SFA by embryos from the 29 wk and L groups. This later fact could be related to chick quality as these chicks would have a smaller energy reserve for post-hatching development than chicks from older parents or from smaller eggs. These chicks may be in the need of more careful management practices in the grow out barn (e.g. appropriate temperature, high quality feed) during the first day post-hatch to restore their possible energy deficiency.

SUMMARY AND CONCLUSIONS

In summary: 1) Percentage of FA in eggs were greatly influenced by breeder flock age (in both C and R strains). 2) Percentage of FA in RYS were greatly influenced by breeder flock age (in both C and R strains). 3) Percentage of FA in the RYS was not affected by egg size (in both C and R strains).

It was interesting to note that the composition of the RYS of chicks hatching from young hens was different than that of chicks hatching at older ages, and that myristic acid ($C_{14:0}$) was present in greater amount in the RYS of the chicks from young flocks (in

both strains). Whether or not this is a consequence of impaired fat metabolism in chicken embryos from young flocks leading to a decrease in the ability to elongate this FA from the egg remains a hypothesis.

It was initially hypothesized that the FA content of the RYS of newly hatched chicks would be highly correlated with the FA content of the egg. However, except for linolenic acid ($C_{18:3(n-3)}$), the correlations between the initial FA content of the egg and the FA content of the RYS at hatching were weak or non existent. This means that the FA composition of the RYS was dependent upon the ability of the chick embryo to absorb the diverse FA from the egg, and not on the initial FA composition of the egg itself.

The correlation of linolenic acid ($C_{18:3(n-3)}$) for egg size (M for C and S, and M for R) was significant. Whether or not these results are related, the possibility of increasing the linolenic ($C_{18:3(n-3)}$) acid content in the egg via manipulations in the hen diet, may increase the amount of this FA in the RYS of chicks. This may increase the positive effects of DHA ($C_{22:6(n-3)}$) in the neurological development of chicks hatching for M eggs (which are expected to be the greater percentage in commercial flocks).

IMPLICATIONS FOR FUTURE RESEARCH

Results from the current research are a valuable start point to understand the differences in FA composition observed in hatching eggs from different strains and produced at different ages. To optimize the results presented in this thesis, it is suggested that future research should include the FA analysis of the diets fed to the birds, since it has been proved that dietary fat is one of the main factors affecting the FA composition of eggs and embryonic tissues. Future research on the FA composition of eggs and RYS from young broiler breeder flocks is advised.

TABLES

<u>, an ann an an</u>	Egg weight (g)	Yolk weight (% egg wt)	Chick weight (g)	Chick weight (% egg wt)	Carcass weight (% chick wt)	RYS weight (% chick wt)
Strain ¹						······································
С	64.5(162) ³	29.7(90)	45.2(114)	70.0	84.5	15.0
R	62.3(162)	30.0(90)	43.3(114)	69.3	86.1	13.5
SEM ²	1.5	0.2	1.3	0.5	0.8	0.8
Flock age						
29 wk	55.9 ^c (108)	27.4 ^c (60)	39.2 ^c (79)	70.1ª	89.8ª	10.2 ^b
45 wk	64.6 ^b (108)	30.6 ^b (60)	44.1 ^b (69)	68.2 ^b	82.7 ^b	16.1ª
59 wk	69.7 ^ª (108)	31.5°(60)	49.3°(80)	70.6ª	83.5 ^b	16.4ª
SEM	1.9	0.3	1.6	0.6	1.0	0.9
Strain*age						
C * 29 wk	54.1(54)	27.8°(30)	38.1(35)	70.4	90.4	9.6
C * 45 wk	68.3(54)	30.1 ^b (30)	46.9(35)	68.6	81.0	17.6
C * 59 wk	71.2(54)	31.3°(30)	50.5(44)	70.9	82.2	17.7
R * 29 wk	57.7(54)	26.8 ^d (30)	40.4(44)	69.9	89.3	10.7
R * 45 wk	60.9(54)	31.3 ^a (30)	41.4(34)	67.8	84.3	14.6
R * 59 wk	68.2(54)	31.7 ^a (30)	48.0(36)	70.3	84.9	15.2
SEM	2.7	0.3	2.3	0.8	1.4	1.3

Table 3-1. Effects of genetic strain, flock age and the interaction of strain*age on the percent weight of egg and chick components

^{a-d} Means within the same column with different superscripts differ significantly (P \leq 0.05) ¹C= Cobb 500, R= Ross 308

²Standard error of mean

³Number of observations

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Fatty		Flock ag	e				Egg size			
acid	29 wk	45 wk	59 wk	SEM ²	p-value	S³	M ⁴	۲ <u>ء</u>	SEM	p-value
C _{14:0}	0.24 ^b	0.46 ^a	0.47 ^a	0.03	<0.0001	0.44	0.35	0.39	0.03	0.3705
C _{16:0}	26.89 ^b	32.39 ^a	30.93 ^a	1.33	0.0166	32.11 ^ª	26.55 ^b	31.55^{a}	1.33	0.0073
C _{18:0}	10.32	10.64	10.34	0.48	0.8901	11.23 ^ª	9.05 ^b	11.03 ^a	0.48	0.0029
C _{16:1(n-7)}	4.02	4.51	4.18	0.21	0.2537	4.69 ^a	3.78 ^b	4.24 ^a	0.21	0.0109
C _{18:1(n-9)}	35.99^{a}	26.89 ^b	29.89 ^b	2.94	0.0329	25.93 ^b	38.74 ^a	28.11 ^b	2.94	0.0069
C _{18:2(n-6)}	13.44 ^b	16.08 ^a	14.70 ^b	0.65	0.0271	15.89^{a}	13.07 ^b	15.26^{a}	0.65	0.0069
C _{18:3(n-3)}	0.62 ^b	1.29 ^a	1.18 ^a	0.06	<0.0001	1.12	0.91	1.06	0.06	0.1369
$C_{20:4(n-6)}$	3.33ª	2.42 ^b	2.45 ^b	0.15	<0.0001	2.92 ^a	2.38 ^b	2.89 ^a	0.15	0.0223
$C_{22:6(n-3)}$	1.56	1.73	1.81	0.09	0.1836	1.84 ^a	1.47 ^b	1.79 ^a	0.09	0.0210
% Total	96.41	96.41	95.95			96.17	96.3	96.32		
ΣSFA^6	37.45	43.49	41.74	1.79	0.0606	43.78 ^a	35.95 ^b	42.97 ^a	1.79	0.0045
Σ MUFA ⁷	40.01	31.40	34.07	2.80	0.0562	30.62 ^b	42.52 ^a	32.35 ^b	2.80	0.0073
Σ PUFA [®]	18.95	.21.52	20.14	0.95	0.1409	21.77 ^a	17.83 ^b	21.0 ^a	0.95	0.0035

^{ad}Means within the same row with different superscripts within each treatment differ significantly (P≤0.05)

¹Fatty acids are given as a number of carbon atoms:number of double bonds, followed by the position of the first double bond relative to the end methyl carbon (C14:0⁼ Myristic; C16:0⁼ Palmitic; C18:0⁼ Stearic; C16:1(n-7)⁼ Palmitoleic; C18:1(n-9)⁼ Oleic; C18:2(n-6)⁼ Linoleic; C_{18:3(n-3)}= Linolenic; C_{20:4(n-6)}= Arachidonic; C_{22:6(n-3)}= Docosahexaenoic, DHA)

²Standard error of mean

³3 to 6 g lower than M eggs

average egg weight for that flock age

⁵3 to 6 g higher than M eggs

⁶Sum of saturated fatty acids ($\sum C_{14:0}$, $C_{16:0}$, $C_{18:0}$)

⁷Sum of monounsaturated fatty acids ($\sum C_{16:1(n-7)}$, $C_{18:1(n-9)}$) ⁸Sum of polyunsaturated fatty acids ($\sum C_{18:2(n-6)}$, $C_{18:3(n-3)}$, $C_{20:4(n-6)}$, $C_{22:6(n-3)}$) n= 10 yolk+albumen samples / flock age / egg size

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Fatty		Flock age					Egg size			
acid ¹	29 wk	45 wk	59 wk	SEM	p-value	S2	M³	L ⁴	SEM	p-value
C _{14:0}	0.51ª	0.43 ⁶	0.39 ^c	0.01	<0.0001	0.45	0.43	0.44	0.01	0.5726
C _{16:0}	25.84 ^a	26.70 ^a	23.52 ^b	0.79	0.0133	25.73	24.40	25.92	0.79	0.3605
C _{18:0}	8.40	8.77	8.12	0.28	0.2937	8.52	8.22	8.55	0.28	0.7069
C _{16:1(n-7)}	3.50 ^a	3.20 ^a	2.61 ^b	0.15	<0.0001	3.33	2.92	3.06	0.15	0.1440
C _{18:1(n-9)}	41.68 ^b	39.40 ^b	46.87 ^a	1.83	0.0183	41.48	45.10	41.37	1.83	0.2415
C _{18:2(n-6)}	14.08 ^a	14.74 ^a	11.76 ^b	0.55	0.0002	14.01	12.66	13.91	0.55	0.1502
C _{18:3(n-3)}	0.63 [°]	1.15 ^a	0.88 ^b	0.04	<0.0001	0.92	0.83	0.91	0.04	0.2945
C _{20:4(n-6)}	1.41 ^b	1.74 ^a	1.59 ^a	0.08	0.0065	1.61	1.49	1.63	0.08	0.3636
C _{22:6(n-3)}	0.25 ^c	0.65ª	0.55 ^b	0.03	<0.0001	0.47	0.48	0.51	0.03	0.1191
% Total	96.3	96.78	96.29			96.52	96.53	96.30		
ΣSFA⁵	34.75 ^{ab}	35.9ª	32.03 ^b	1.04	0.0270	34.70	33.05	34.91	1.04	0.3992
ΣMUFA ⁶	45.18 ^{ab}	42.6 ^b	49.48 ^a	1.72	0.0217	44.81	48.02	44.43	1.72	0.2572
ΣPUFA ⁷	16.37 ^b	18.28 ^a	14.78 ^b	0.68	0.0003	17.01	15.46	16.96	0.68	0.1703

^{a-e}Means within the same row with different superscripts differ significantly (P≤0.05)

¹Fatty acids are given as a number of carbon atoms:number of double bonds, followed by the position of the first double bond relative to the end methyl carbon (C_{14:0}= Myristic; C_{16:0}= Palmitic; C_{18:0}= Stearic; C_{16:1(n-7)}= Palmitoleic; C_{18:1(n-9)}= Oleic; C_{18:2(n-6)}= Linoleic; C_{18:3(n-3)}≈ Linolenic; C_{20:4(n-6)}= Arachidonic; C_{22:6(n-3)}= Docosahexaenoic, DHA)

²3 to 6 g lower than M eggs

average egg weight for that flock age

⁴3 to 6 g higher than M eggs

⁵Sum of saturated fatty acids ($\sum C_{14:0}, C_{18:0}, C_{18:0}$) ⁶Sum of monounsaturated fatty acids ($\sum C_{16:1(n-7)}, C_{18:1(n-9)}$) ⁷Sum of polyunsaturated fatty acids ($\sum C_{18:2(n-6)}, C_{18:3(n-3)}, C_{20:4(n-6)}, C_{22:6(n-3)}$)

n= 10 residual yolk sacs / flock age / egg size

Table 3-4. Pearson's correlation coefficient (r) between the percentage of fatty acids in the egg and the percentage of fatty acids in the residual yolk sac for a Cobb 500 flock at three flock ages and from three egg sizes

	p-value	0.0582	0.1361	0.2859	0.3525	0.2745	0.6821	0.0260	0.6953	0.1302	0.1701	0.2926	0.6986
	Ļ	-0.37	-0.29	-0.21	-0.18	-0.22	-0.08	0.43	-0.08	0.30	-0.27	-0.21	-0.08
lg size	p-value	0.2210	0.5193	0.1314	0.8177	0.0667	0.0085	<.0001	0.1639	0.6689	0.2644	0.0220	0.0001
Ec	M³	-0.24	-0.13	-0.30	0.05	0.36	0.50	0.73	-0.28	0.09	-0.22	0.44	0.67
	p-value	0.2967	0.6463	0.9056	0.6276	0.7846	0.9536	0.0362	0.2356	0.2297	0.7773	0.8266	0.8310
	S ²	-0.20	0.09	-0.02	0.10	0.05	0.01	0.40	-0.23	0.23	0.06	0.04	0.04
	p-value	0.0480	0.0139	0.9654	0.0351	0.1183	0.7792	0.1293	0.0861	0.0441	0.0350	0.2770	0.8090
	59 wk	-0.38	-0.47	-0.01	-0.41	-0.31	-0.06	-0.30	0.34	0.39	-0.41	-0.22	0.05
ck age	p-value	0.6409	0.9688	0.4984	0.1872	0.8137	0.3747	0.2295	0.6512	0.7631	0.8324	0.8638	0.4084
Flo	45 wk	0.09	-0.01	-0.13	0.26	0.05	0.17	0.23	-0.09	0.06	-0.04	0.03	0.16
	p-value	0.9890	0.0937	0.9598	0.2800	0.5994	0.4000	0.7004	0.1739	0.4479	0.2904	0.8609	0.7078
	29 wk	0.01	0.33	-0.22	0.01	0.11	0.17	0.08	-0.27	-0.15	0.21	0.03	0.07
Fatty	acid ¹	C _{14:0}	C _{16:0}	C _{18:0}	C _{16:1(n-7)}	C _{18:1(n-9)}	C _{18:2(n-6)}	C _{18:3(n-3)}	C _{20:4(n-6)}	C _{22:6(n-3)}	ΣSFA^{5}	Σ MUFA ⁶	Σ PUFA ⁷

¹Fatty acids are given as a number of carbon atoms:number of double bonds, followed by the position of the first double bond relative to the end methyl carbon (C_{14:0}= Myristic; C_{16:0}= Palmitic; C_{16:0}= Stearic; C_{16:1(n-7)}= Palmitoleic; C_{18:1(n-9)}= Oleic; C_{18:2(n-6)}= Linoleic; C_{18:3(n-3)}= Linoleic; C_{20:4(n-6)}= Arachidonic; C_{22:6(n-3)}= Docosahexaenoic, DHA) ²3 to 6 g lower than M eggs

³average egg weight for that flock age

⁴3 to 6 g higher than M eggs

⁵Sum of saturated fatty acids ($\Sigma C_{14:0}$, $C_{16:0}$, $C_{18:0}$) ⁶Sum of monounsaturated fatty acids ($\Sigma C_{16:1(n-7)}$, $C_{18:1(n-9)}$)

⁷Sum of polyunsaturated fatty acids (Σ C_{18:2(n-6)}, C_{18:3(n-3)}, C_{20:4(n-6)}, C_{22:6(n-3)}) n= 30 yolk+albumen samples – 30 residual yolk sacs / flock age - egg size

Fatty		29 wk			45 wk			59 wk	
acid ¹	Egg	RYS ²	p-value	Egg	RYS	p-value	Egg	RYS	p-value
C14:0	0.24	0.51	<0.0001	0.46	0.43	0.3626	0.47	0.39	0.0100
C _{16:0}	26.89	25.84	0.0991	32.39	26.70	0.0064	30.93	23.52	0.0006
C _{18:0}	10.32	8.40	<0.0001	10.64	8.77	0.0125	10.34	8.12	0.0019
C _{16:1(n-7)}	4.02	3.50	0.0250	4.51	3.20	<0.0001	4.18	2.61	<0.0001
C _{18:1(n-9)}	35.99	41.68	0.0013	26.89	39.40	0.0054	29.89	46.87	0.0003
C _{18:2(n-6)}	13.44	14.08	0.2121	16.08	14.74	0.0880	14.70	11.76	0.0013
C _{18:3(n-3)}	0.62	0.63	0.6929	1.29	1.15	0.0302	1.18	0.88	0.0004
C _{20:4(n-6)}	3.33	1.41	<0.0001	2.42	1.74	0.0003	2.45	1.59	<0.0001
C _{22:6(n-3)}	1.56	0.25	<0.0001	1.73	0.65	<0.0001	1.81	0.55	<0.0001
ΣSFA^3	37.45	34.75	0.0023	43.49	35.9	0.0072	41.75	32.03	0.0007
∑ MUFA ⁴	40.01	45.18	0.0020	31.40	42.6	0.0073	34.39	49.75	0.0003
Σ PUFA ⁵	18.95	. 16.37	0.0004	21.52	18.28	0.0080	21.35	15.42	<0.0001

Table 3-5. Comparison between the percentage of fatty acids in the egg and the percentage of fatty acids in the residual yolk sac for a Cobb 500 flock at three flock ages ¹Fatty acids are given as a number of carbon atoms:number of double bonds, followed by the position of the first double bond relative to the end methyl carbon (C_{14:0}= Myristic; C_{16:0}≂ Palmitic; C_{16:0}= Stearic; C_{16:1(n-7)}= Palmitoleic; C_{18:1(n-9)}= Oleic; C_{18:2(n-6)}= Linoleic; C_{18:2(n-6)}= Linol ²Residual yolk sac

³Sum of saturated fatty acids (Σ C_{14:0}, C_{16:0}, C_{18:0}) ⁴Sum of monounsaturated fatty acids (Σ C_{16:1(n-7)}, C_{18:1(n-9)}) ⁵Sum of polyunsaturated fatty acids (ΣC_{18:2(n-6)}, C_{18:3(n-3)}, C_{20:4(n-6)}, C_{22:6(n-3)})

n= 30 yolk+albumen samples - 30 residual yolk sacs / flock age

Fatty		S²			M³			L ⁴	
acid ¹	Egg	RYS ⁵	p-value	Egg	RYS	p-value	Egg	RYS	p-value
C _{14:0}	0.44	0.45	0.8433	0.35	0.43	0.0376	0.39	0.44	0.0376
C _{16:0}	32.11	25.73	0.0015	26.55	24.40	0.0410	31.55	25.92	0.0410
C _{18:0}	11.23	8.52	0.0002	9.05	8.22	0.0663	11.03	8.55	0.0663
C _{16:1(n-7)}	4.69	3.33	0.0001	3.78	2.92	0.0017	4.24	3.06	0.0017
C _{18:1(n-9)}	25.93	41.48	0.0008	38.74	45.10	0.0036	28.11	41.37	0.0036
C _{18:2(n-6)}	15.89	14.01	0.0879	13.07	12.66	0.2287	15.26	13.91	0.2287
C _{18:3(n-3)}	1.12	0.92	0.0238	0.91	0.83	0.0242	1.06	0.91	0.0242
C _{20:4(n-6)}	2.92	1.61	<0.0001	2.38	1.49	<0.0001	2.89	1.63	<0.0001
C _{22:6(n-3)}	1.84	0.47	<0.0001	1.47	0.48	<0.0001	1.79	0.51	<0.0001
Σ SFA ⁶	43.78	34.70	0.0008	35.95	33.05	0.0369	42.97	34.91	0.0045
Σ MUFA ⁷	30.62	44.81	0.0010	42.52	48.02	0.0047	32.35	44.43	0.0038
Σ PUFA [®]	21.77	. 17.01	0.0007	17.83	15.46	<0.0001	21.0	16.96	0.0011

Table 3-6. Comparison between the percentage of fatty acids in the egg and the percentage of fatty acids in the residual yolk sac for a Cobb 500 flock at three egg sizes 'Fatty acids are given as a number of carbon atoms:number of double bonds, followed by the position of the first double bond relative to the end methyl carbon (C_{14:0}= Myristic; C_{16:0}= Palmitic; C_{18:0}= Stearic; C_{16:1(n-7)}= Palmitoleic; C_{18:1(n-9)}= Oleic; C_{18:2(n-6)}= Linoleic; C_{18:3(n-3)}= Linolenic; C_{20:4(n-6)}= Arachidonic; C_{22:6(n-3)}= Docosahexaenoic, DHA)

²3 to 6 g lower than M eggs

³average egg weight

⁴3 to 6 g higher than M eggs

⁵Residual yolk sac

⁶Sum of saturated fatty acids (Σ C_{14:0}, C_{16:0}, C_{18:0}) ⁷Sum of monounsaturated fatty acids (Σ C_{16:1(n-7)}, C_{18:1(n-9})) ⁸Sum of polyunsaturated fatty acids (ΣC_{18:2(n-9)}, C_{18:3(n-3)}, C_{20:4(n-6)}, C_{22:6(n-3)})

n= 30 yolk+albumen samples - 30 residual yolk sacs / flock age - egg size

	p-value	0.5185	0.2164	0.0975	0.7947	0.1412	0.1235	0.1413	0.1829	0.4420		0.1669	0.1294	0.1111
	SEM	0.03	1.33	0.54	0.22	2.75	0.47	0.03	0.14	0.09		1.85	2.55	0.72
	[4	0.43	30.97	10.93	4.43	31.07	13.79	0.81	2.53	1.35	96.31	42.33	35.75	19.49
Egg size	M³	0.37	28.34	9.56	4.30	37.13	12.86	0.78	2.15	1.20	96.69	38.26	41.66	17.80
	S^2	0.38	27.90	9.53	4.16	38.27	12.42	0.71	2.30	1.26	96.93	37.81	42.65	17.47
	p-value	0.0055	0.0155	0.0004	0.0034	0.0253	0.0002	<0.0001	0.5125	<0.0001		0.0051	0.0352	0.0016
	SEM	0.03	1.33	0.54	0.22	2.75	0.47	0.03	0,14	0.09		1.85	2.61	0.72
	59 wk	0.43 ^a	28.94 ^b	10.19 ^b	4.25 ^b	37.41 ^a	11.43 ^b	0.56°	2.29	1.04 ^b	96.54	39.56 ^{ab}	41.95 ^a	16.17 ^b
Flock age	45 wk	0.45 ^a	31.91 ^ª	11.44 ^a	4.87 ^a	29.47 ^b	14,00 ^a	0.67 ^b	2.47	1.16 [°]	96.44	43.81 ^a	34.58°	19.35 ^a
	29 wk	0.30 ^b	26.35 ^b	8.39°	3.78 ^c	39.59 ^a	13.65 ^a	1.07 ^a	2.22	1.60^{a}	96,95	35.03 ^b	43.53 ^a	19.24ª
Fatty	acid ¹	C _{14:0}	C _{16:0}	C _{18:0}	C _{16:1(n-7)}	C _{18:1(n-9)}	C _{18:2(n-6)}	C _{18:3(n-3)}	$C_{20:4(n-6)}$	C _{22:6(n-3)}	% Total	ΣSFA ⁵	ΣMUFA	Σ PUFA ⁷

Table 3-7. Effects of flock age and egg size on the percentage of fatty acids of eggs from a Ross 308 flock

^{a-e}Means within the same row with different superscripts differ significantly (P≤0.05)

¹Fatty acids are given as a number of carbon atoms:number of double bonds, followed by the position of the first double bond relative to the end methyl carbon (C14:0= Myristic; C16:0= Palmitic; C18:0= Stearic; C16:1(n-7)= Palmitoleic; C18:1(n-9)= Oleic; C18:2(n-6)= Linoleic; $C_{18:3(n-3)}$ = Linolenic; $C_{20:4(n-6)}$ = Arachidonic; $C_{22:6(n-3)}$ = Docosahexaenoic, DHA) ²3 to 6 g lower than M eggs

³average egg weight for that flock age

⁴3 to 6 ğ higher than M eggs ⁵Sum of saturated fatty acids (Σ C_{14:0}, C_{16:0}, , C_{18:0}) ⁶Sum of monounsaturated fatty acids (Σ C_{16:1(n-7)}, C_{18:1(n-9)}) ⁷Sum of polyunsaturated fatty acids (ΣC_{18:2(n-8)}, C_{18:3(n-3)}, C_{20:4(n-6)}, C_{22:6(n-3)})

n= 10 yolk+albumen samples / flock age / egg size

	SEM p-value	0.03 0.6679	1.14 0.6814	0.47 0.7576	0.20 0.5379	2.52 0.7689	0.60 0.6751	0.04 0.0302	0.11 0.4061	0.04 0.0927		1.57 0.7332	2.35 0.7609	
	[_4	0.46	26.99	9.17	3.46	39.28	13.86	0.86 ^a	1.70	0.45	96.23	36.08	44.05	
Egg size	M³	0.43	26.74	8.92	3.44	40.30	13.24	0.75 ^b	1.64	0.45	95.91	36.08	44.05	
	S2	0.44	25.75	8.76	3.21	42.12	13.16	0.71 ^b	1.50	0.37	96.02	34.95	45.64	
	p-value	0.0156	0.0047	0.0257	<0.0001	0.2181	<0.0001	<0.0001	0.0751	0.0395		0.0045	0.2579	00000
	SEM	0.03	1.14	0.47	0.20	2.52	0.60	0.04	0.11	0.04		1.57	2.35	, I (
	59 wk	0.44 ^a	26.53 ^b	9.17 ^a	3.63 ^a	42.28	11.57°	0.56 ^b	0.74	0.35 ^b	95.27	36.15 ^a	46.19	
Flock age	45 wk	0.49 ^a	29.11 ^a	9.85 ^a	3.90ª	36.86	13.50 ^b	0.62 ^b	1.66	0.41 ^b	96.40	39.46 ^a	41.07	4.0.01
	29 wk	0.39 ^b	23.83 ^c	7.81 ^b	2.58 ^b	42.58	15.20 ^a	1.14 ^a	1.44	0.50^{a}	95.47	32.03 ^b	45.47	110
Fatty	acid ¹	C _{14:0}	C _{16:0}	C _{18:0}	C _{16:1(n-7)}	C _{18:1(n-9)}	C _{18:2(n-6)}	C _{18:3(n-3)}	C _{20:4(n-6)}	C _{22:6(n-3)}	% Total	ΣSFA ⁵	ΣMUFA ⁶	

Table 3-8. Effects of flock age and egg size on the percentage of fatty acids in the residual yolk sac of chicks hatched from a Ross 308 flock

^{a-b} Means within the same row with different superscripts differ significantly (P≤0.05)

Fatty acids are given as a number of carbon atoms:number of double bonds, followed by the position of the first double bond relative to the end methyl carbon (C_{14:0}= Myristic; C_{16:0}= Palmitic; C_{18:0}= Stearic; C_{16:1(n-7)}= Palmitoleic; C_{18:1(n-9)}= Oleic; C_{18:2(n-6)}= Linoleic; C_{18:3(n-3)}= Linolenic; C_{20:4(n-8)}= Arachidonic; C_{22:6(n-3)}= Docosahexaenoic, DHA)

²3 to 6 g lower than M eggs

³average egg weight for that flock age

¹3 to 6 g higher than M eggs

⁵Sum of saturated fatty acids (Σ C_{14:0}, C_{16:0}, C_{18:0})

⁶Sum of monounsaturated fatty acids ($\sum C_{16:1(n-7)}$, $C_{18:1(n-9)}$) ⁷Sum of polyunsaturated fatty acids ($\sum C_{18:2(n-6)}$, $C_{18:3(n-3)}$, $C_{20:4(n-6)}$, $C_{22:6(n-3)}$)

n= 10 residual yolk sacs / flock age / egg size

-atty			ЪО́Ч	ck age					Ш	ig size		
acid ¹	29 wk	p-value	45 WK	p-value	59 wk	p-value	S ²	p-value	M³	p-value	4	p-value
714:0	0.26	0.1969	0.13	0.5345	-0.37	0.0660	-0.02	0.9337	0.27	0.1692	0.25	0.2140
C16:0	-0.09	0.6494	0.10	0.6412	-0.14	0.4887	-0.09	0.6760	-0.01	0.9432	0.34	0.0866
C18:0	0.36	0.0665	0.04	0.8495	-0.21	0.2931	0.07	0.7404	0.09	0.6451	0.21	0.0113
C 16:1(n-7)	-0.04	0.8244	0.05	0.8129	-0.07	0.7391	-0.04	0.8380	-0.08	0.6818	0.48	0.1591
718:1(n-9)	-0.16	0.4193	0.09	0.6729	-0.15	0.4675	-0.19	0.3729	-0.07	0.7340	0.28	0.2895
J18:2(n-6)	-0.45	0.0192	0.12	0.5592	0.12	0.5500	-0.11	0.6077	0.13	0.5218	0.35	0.0708
℃ 18:3(n-3)	-0.56	0.0023	0.26	0.2019	0.37	0.0643	0.76	<.0001	0.70	<.0001	0.46	0.0165
℃20:4(n-6)	0.37	0.0566	-0.02	0.9231	-0.08	0.6955	0.01	0.9681	0.27	0.1783	-0.06	0.7518
J22:6(n-3)	0.34	0.0820	0.10	0.6382	0.07	0.7168	0.18	0.3976	0.51	0.0069	0.24	0.2262
∑ SFA ⁵	0.03	0.8611	0.08	0.7031	-0.19	0.3620	-0.05	0.8139	0.02	0.9249	0.32	0.1072
5 MUFA ⁶	-0.17	0.3952	0.09	0.6764	-0.15	0.4743	-0.19	0.3527	-0.06	0.7836	0.25	0.2010
5 PUFA7	-0.27	0.1698	0.12	0.5448	0.09	0.6784	-0.17	0.4276	0.15	0.4477	0.31	0.1151

Table 3-9. Pearson's correlation coefficient (r) between the percentage of fatty acids in the egg and the percentage of fatty acids in the residual yolk sac for a Ross 308 flock at three flock ages and from three egg sizes ¹Fatty acids are given as a number of carbon atoms:number of double bonds, followed by the position of the first double bond relative to the end methyl carbon (C_{14:0}= Myristic; C_{16:0}= Palmitic; C_{18:0}= Stearic; C_{16:1(n-7)}= Palmitoleic; C_{18:1(n-9)}= Oleic; C_{18:2(n-6)}= Linoleic; C_{18:3(n-3)}= Linoleic; C_{20:4(n-6)}= Arachidonic; C_{22:6(n-3)}= Docosahexaenoic, DHA) average egg weight for that flock age ²3 to 6 g lower than M eggs

n= 10 yolk+albumen samples - 10 residual yolk sacs / flock age - egg size

⁵Sum of saturated fatty acids ($\sum C_{14:0}, C_{16:0}, C_{18:0}$) ⁶Sum of monounsaturated fatty acids ($\sum C_{16:1(n-7)}, C_{18:1(n-9)}$) ⁷Sum of polyunsaturated fatty acids ($\sum C_{18:2(n-6)}, C_{18:3(n-3)}, C_{20:4(n-6)}, C_{22:6(n-3)}$)

¹3 to 6 g higher than M eggs

Fatty		29 wk			45 wk			59 wk	
acid ¹	Egg	RYS ²	p-value	Egg	RYS	p-value	Egg	RYS	p-value
C _{14:0}	0.30	0.39	0.0219	0.45	0.49	0.6510	0.43	0.44	0.7980
C _{16:0}	26.35	23.83	0.0177	31.91	29.11	0.1595	28.94	26.53	0.0866
C _{18:0}	8.39	7.81	0.0151	11.44	9.85	0.1011	10.19	9.17	0.0977
C _{16:1(n-7)}	3.78	2.58	<0.0001	4.87	3.90	0.0075	4.25	3.63	0.0165
C _{18:1(n-9)}	39.59	42.58	0.0639	29.47	36.86	0.0956	37.41	42.28	0.0677
C _{18:2(n-6)}	13.65	15.20	0.0842	14.00	13.50	0.4516	11.43	11.57	0.4558
C _{18:3(n-3)}	1.07	1.14	0.6309	0.67	0.62	0.1330	0.56	0.56	0.5388
C _{20:4(n-6)}	2.22	1.44	<0.0001	2.47	1.66	0.0013	2.29	0.74	0.0022
C _{22:6(n-3)}	1.60	0.50	<0.0001	1.16	0.41	<0.0001	1.04	0.35	<0.0001
Σ SFA ³	35.03	32.03	0.0130	43.81	39.46	0.1366	39.56	36.15	0.0897
∑ MUFA⁴	43.53	45.47	0.1292	34.58	41.07	0.1147	41.95	46.19	0.0802
Σ PUFA ⁵	19.24	18.75	0.2592	19.35	16.91	0.0344	16.17	14.79	0.0264

Table 3-10. Comparison between the percentage of fatty acids in the egg and the percentage of fatty acids in the residual yolk sac for a Ross 308 flock at three flock ages ¹Fatty acids are given as a number of carbon atoms:number of double bonds, followed by the position of the first double bond relative to the end methyl carbon ($C_{14:0}$ = Myristic; $C_{16:0}$ = Palmitic; $C_{18:0}$ = Stearic; $C_{16:1(n-7)}$ = Palmitoleic; $C_{18:1(n-9)}$ = Oleic; $C_{18:2(n-6)}$ = Linoleic; $C_{21:3(n-3)}$ = Linolenc; $C_{20:4(n-6)}$ = Arachidonic; $C_{22:6(n-3)}$ = Docosahexaenoic, DHA) ²Residual yolk sac

³Sum of saturated fatty acids (Σ C_{14:0}, C_{18:0}, C_{18:0}) ⁴Sum of monounsaturated fatty acids (Σ C_{18:1(n-7)}, C_{18:1(n-9)}) ⁵Sum of polyunsaturated fatty acids (ΣC_{18:2(n-9)}, C_{18:3(n-3)}, C_{20:4(n-9)}, C_{22:6(n-3)}) n= 30 yolk+albumen samples - 30 residual yolk sacs / flock age

Fatty		S ²			M ³			[4	
acid ¹	Egg	RYS ⁵	p-value	Egg	RYS	p-value	Egg	RYS	p-value
C _{14:0}	0.38	0.44	0.2725	0.37	0.43	0.2807	0.43	0.46	0.7940
C _{16:0}	27.90	25.75	0.2085	28.34	26.74	0.1639	30.97	26.99	0.0173
C _{18:0}	9.53	8.76	0.1973	9.56	8.92	0.2312	10.93	9.17	0.0116
C _{16:1(n-7)}	4.16	3.21	0.0047	4.30	3.44	0.0051	4.43	3.46	0.0004
C _{18:1(n-9)}	38.27	42.12	0.2202	37.13	40.30	0.1281	31.07	39.28	0.0189
C _{18:2(n-6)}	12.42	13.16	0.4455	12.86	13.24	0.7603	13.79	13.86	0.7803
C _{18:3(n-3)}	0.71	0.71	0.9547	0.78	0.75	0.0832	0.81	0.86	0.6835
C _{20:4(n-6)}	2.30	1.50	<0.0001	2.15	1.64	0.0018	2.53	1.70	0.0002
C _{22:6(n-3)}	1.26	0.37	<0.0001	1.20	0.45	<0.0001	1.35	0.45	<0.0001
Σ SFA ⁶	37.81	34.95	0.2090	38.26	36.08	0.1787	42.33	36.61	0.0146
Σ MUFA'	42.65	45.64	0.2803	41.66	44.05	0.1668	35.75	43.04	0.0262
Σ PUFA ⁸	17.47	16.22	0.1404	17.80	16.72	0.0637	19.49	17.52	0.0323

Table 3-11. Comparison between the percentage of fatty acids in the egg and the percentage of fatty acids in the residual yolk sac for a Ross 308 flock at three egg sizes ¹Fatty acids are given as a number of carbon atoms:number of double bonds, followed by the position of the first double bond relative to the end methyl carbon (C_{14:0}= Myristic; C_{16:0}= Palmitic; C_{18:0}= Stearic; C_{16:1(n-7)}= Palmitoleic; C_{18:1(n-9)}= Oleic; C_{18:2(n-6)}≃ Linoleic; C_{18:3(n-3)}= Linolenc; C_{28:4(n-6)}≂ Arachidonic; C_{22:6(n-3)}= Docosahexaenoic, DHA) ²3 to 6 g lower than M eggs

²3 to 6 g lower than M eggs ³average egg weight ⁴3 to 6 g higher than M eggs ⁵Residual yolk sac ⁵Sum of saturated fatty acids ($\sum C_{14:0}, C_{18:0}, C_{18:0}$) ⁷Sum of monounsaturated fatty acids ($\sum C_{18:2(n-5)}, C_{18:1(n-3)}, C_{20:4(n-6)}$) ⁸Sum of polyunsaturated fatty acids ($\sum C_{18:2(n-5)}, C_{18:3(n-3)}, C_{20:4(n-6)}$) ⁸Sum of polyunsaturated fatty acids ($\sum C_{18:2(n-5)}, C_{18:3(n-3)}, C_{20:4(n-6)}$) ⁸Sum of polyunsaturated fatty acids ($\sum C_{18:2(n-5)}, C_{18:3(n-3)}, C_{20:4(n-6)}$) ⁸Sum of polyunsaturated fatty acids ($\sum C_{18:2(n-5)}, C_{18:3(n-3)}, C_{20:4(n-6)}$)

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4. GENERAL DISCUSSION AND CONCLUSIONS

The Canadian chicken industry operates under a supply management system, which 'regulates domestic production and imports to ensure that the supply of a product matches the demand for it and that the prices paid to agricultural producers are steady over time and provide the producers with fair returns' (Poultry Marketplace, 2006). The study of factors related to chicken production efficiency is of great interest for the poultry industry which grows according to the increases in per capita chicken consumption. Annual per capita consumption of chicken in Canada increased from 24.9 Kg/person in 1996 to 31.8 Kg/person in 2006 (Chicken Farmers of Canada, 2007a).

Results from the 2007 Usage and Attitudes Survey revealed that boneless, skinless chicken breasts and regular chicken breasts are the most popular type of chicken purchased in Canada (78 and 71 % respectively) (Chicken Farmers of Canada, 2007b). Genetic selection in poultry is directed to satisfy the increasing demand for white meat.

Research studying the effects of genetic strain, flock age, and egg size in multiple poultry production parameters is available. However, most research has analyzed the effect of one factor at the time. Thus, the effects of genetic strain on hatching egg components (Suarez et al., 1997), embryo survival (Joseph and Moran Jr., 2005), and chick characteristics (Proudfoot et al., 1982) is available. Research comparing breeder flocks of different ages has generally been liked to eggs of different size, and has concluded that as hens age not only egg size increases (Roque and Soares, 1994), but also egg characteristics (Peebles et al., 2000), fertility (Elibol et al., 2002; Zakaria et al., 2005), and hatchability (Tona et al., 2001) change. These changes

associated with broiler breeder flock age affect broiler survival (McNaughton et al., 1978; Vieira and Moran Jr., 1998), and feed conversion rates (Hulet et al., 2007).

Research determining the effects of genetic strain, flock age and egg size in chick production parameters of modern broiler breeder strains has not been reported.

Variations in hatching egg components, specially those affecting the egg yolk, are important for the developing embryo. More than 90% of the total energy requirements of the chicken embryo is derived from yolk lipids (Romanoff, 1960), and the fatty acid content of the yolk sac in the newly hatched chick is directly related with the subsequent performance of the broilers (Vieira and Moran Jr., 1999).

The lipid composition of the hatching egg yolk has been previously reported (Nielsen, 1998), however, most research on the FA content of eggs has been performed in laying hens and has been specially aimed at the enrichment of eggs through manipulation of the hen's diet (Alvarez et al., 2004, 2005; Muma et al., 2006). Although the changes on FA between the egg and the yolk sac have been reported (Noble et al., 1986; Speake et al., 1998), no research comparing the FA of the egg vs. the FA in the RYS in modern strains is available.

After evaluating the effects of broiler breeder genetic strain, flock age, and egg size on diverse aspects involved with broiler production, this thesis concluded that most variations were a consequence of the interaction of breeder strain * flock age, and not a consequence of the main separate effects. In summary, hatching eggs from young parent flocks had smaller yolks than eggs laid at older ages. Fertility at the younger age was greater in R birds than C birds at the same flock age. The results on hatchability obtained from this research were affected by overheating of the incubators while incubating eggs from the 45 wk old flocks, however, it was observed that the heat stress affected the C embryos in a more severe way than it affected the R embryos (mid and late embryo mortality were greater in the C than in the R embryos). The chicks hatching from the young parent flocks had poorer performance during the grow out period with higher first wk mortality and lower final BW than the chicks from the other parent flock ages. The information provided by this thesis could have immediate applicability since the research was conducted on modern broiler breeder strains that are commonly used in Canada. The results here presented, could be used as a starting point to develop research aimed at determining appropriate incubation conditions for eggs produced by each strain and flock age. Similarly, barn conditions during the grow out period could be researched to maximize the performance of chicks produced by breeders of each strain and flock age.

After evaluating the effects of the same factors on the FA content of hatching eggs and RYS it was concluded that flock age produced most of the variations. Regardless of the strain, chicks hatching from younger hens had a different FA content in the RYS than chicks hatching at older ages. It was hypothesized by the author of this thesis that this fact could be related to the poor chick quality in chicks from young parent flocks that is commonly reported by broiler producers. Further research analyzing this fact is advised. The hypothesis if whether or not the embryos from younger hens absorb the fatty acids from the yolk in a different way than embryos at older ages should be tested. The positive correlation found between the linolenic acid ($C_{18:3(n-3)}$) content of eggs and RYS in the M group may provide an interesting opportunity to increase chick quality by supplementing the hen's diet with this FA and thus increasing its content in the RYS for post-hatch absorption.

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