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EFFECT OF ACUTE HEAT STRESS ON WHITE LEGHORN HENS WITH OR WITHOUT ACTIVE SHELL DEPOSITION AND SOME ATTEMPTS TO OVERCOME THE DETRIMENTAL EFFECT OF HEAT STRESS ON EGG SHELL QUALITY

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By

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ABSTRACT

EFFECT OF ACUTE HEAT STRESS ON WHITE LEGHORN HENS WITH OR WITHOUT ACTIVE SHELL DEPOSITION AND SOME ATTEMPTS TO OVERCOME THE DETRIMENTAL EFFECT OF HEAT STRESS ON EGG SHELL QUALITY

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This study was conducted with Single Comb White Leghorn (SCWL) laying hens to establish a procedure to evaluate the effect of acute heat stress on shell quality without endangering the hens. This research also included attempts to overcome the deterimental effect of heat stress on egg shell quality.

SCWL hens kept at 21°C were stressed at 35°C for 1, 2, 4, and 6 hours. Control hens were in the cabinet at ambient temperature (23°C) for the same length of time. Hens were either 46 (young) or 61 (old) weeks of age. Eggs were collected for 3 days before each hen was in the cabinet and for 3 days afterward. The difference between before and after values for % shell of egg weight, weight of shell, and Ca/mm^2 of the shell were determined for each hen. Heat at 35°C for 4 hours caused lower shell quality on the first day following the stay in the heat chamber than those from hens stressed for 1 or 2 hours, while shells laid on the second and third day were equivalent to values before heat stress.

At least 4 hours of 35°C, acute heat stress, were required to reduce shell quality significantly. Heat-stressed hens with an egg in the uterus laid eggs prematurely, or laid lesser quality shells. This was noted when the water was not available during egg shell formation. The minimum decline in the shell quality was observed when both feed and water were available during the stay in the chamber (during egg shell deposition). Blood PCO_2 and HCO_3^- significantly ($P \leq .01$) declined in heat stressed hens.

There was no improvement in egg shell quality due to the use of analgesic drugs such as aspirin (in feed or water) or acetaminophen (in water). Vitamin C had no significant effect on preventing the rise in body temperature or the decline in shell quality due to the acute heat stress. The inclusion in the water of high concentration of KCl and $CaCl_2$ increased the incidence of shell defects.

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1. Introduction

As domestic egg production becomes more mechanized, increasing numbers of shells get broken as eggs move from the hen to consumer.

Egg shell breakage is related to shell quality, or shell strength, which refers to the ability of egg shells to withstand externally applied forces without cracking or breaking.

Hamilton (1982) reported that shell breakage is estimated to cost Candian egg producers about $10 million annually and United States producers about $100 million. In addition to the economic losses, there is the loss of a high quality foodstuff for humans. During summer, heat waves cause hens to lay egg with lesser quality shells, thereby increasing egg breakage. This work was conducted with Single Comb White Leghorn (SCWL) laying hens to establish a procedure to evaluate the effect of acute heat stress on shell quality without endangering the hens. This research also included attempts to overcome the detrimental effect of heat stress on egg shell quality.

2. Review of Literature

2.1 Egg Formation

2.1.1 The Structure and the Role of the Oviduct

The fowl oviduct consists of five regions, (1) the infundibulum or funnel, which receives the ovum (yolk) when it is ovulated; (2) the magnum or albumen secreting region (also water and salts are added to the albumen); (3) the isthmus, in which the shell membranes are formed; (4) the shell gland, or uterus, where the shell is formed around the membranes and pigment (due to the secretion of porphyrins) added to the shell and (5) the vagina, which takes no part in the formation of the egg but may be involved in the expulsion of the egg. The duration of time for which the egg stays in the infundibulum, magnum, and isthmus for chickens are 1/4 - 1/2 hr., 2-3 hr., and about 1 1/4 hr., respectively. The average stay of the ovum in the shell gland is 20-26 hr. (Fig.1) (Sturkie, 1986).

2.1.2 Structure and Composition of the Egg Shell

There are three components of fowl egg shells. They are (from the inside to the outside: (a) two proteinaceous membranes that are rich in cystine and contain hydroxyproline and hydroxylysine, (b) the true shell, and (c) a proteinaceous cuticle (Fig. 2). The true or calcified shell consists of the mammillary and the pallisade or spongy layer. The true shell is composed of 2% organic material (a series of layers of

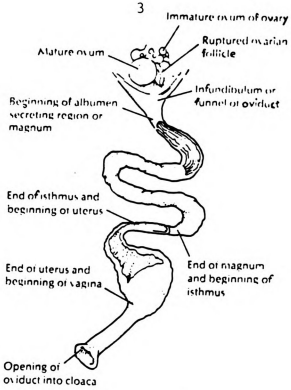

Figure 1. Reproductive tract of the laying hen (Sturkie, 1986).

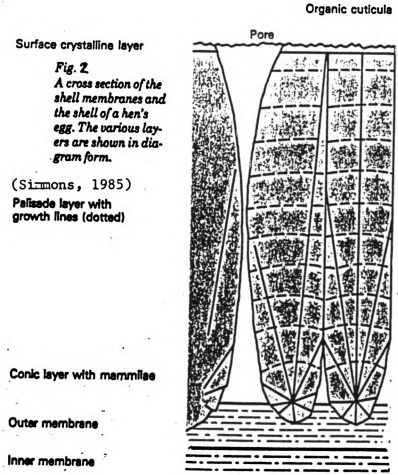

Fig. 2
A cross section of the shell membranes and the shell of a hen's egg. The various layers are shown in diagram form.

(Simmons, 1985)

protein plus acid mucopolysaccharide on which calcification takes place). And the remainder (98%) is mostly crystalline calcium carbonate in the form of calcite with small quantities of magnesium, phosphate, citrate, and traces of sodium and potassium. The beginning of true shell formation occurs in the isthmus (Sturkie, 1986).

2.2 Some of the Factors That Influence Egg Shell Quality

2.2.1 The Various Techniques and Devices That Are Used to Evaluate Shell Quality

McNally (1965) used shell weight and the number of cracked eggs to evaluate shell quality. True shell expressed as % of egg weight, shell thickness and specific gravity of eggs also were used (De Andrade et al., 1976). Also breaking strength, shell deformation (Belyavin and Boorman, 1980) and shell calcium (Atteh and Leeson, 1983) have been used. Meyer et al. (1973) reported that scanning electron micrographs revealed that most of the changes responsible for differences in shell breaking strength apparently take place in the palisade layer, while the mammillary layer remains approximately constant. Also they found a poor correlation between breaking strength and total shell thickness. They suggested that breaking strength is a superior measure of egg shell quality because it takes into consideration both the thickness and the concentration of the palisade layer around the crystalline mammillary portion of the egg shell. However egg shell thickness, egg shell weight and egg specific gravity were found to be correlated

with crack incidence (Anderson et al., 1970; McNally, 1965; Wells, 1967).

Belyavin and Boorman (1980) studied the relationship between the cuticle layer and shell quality. They found that the cuticle made a significant contribution to shell thickness and removal of it reduced shell strength when measured directly. Blake et al. (1985) found that percentage of amino acids in the dried membrane (outer shell membrane) was significantly less for old hens (75 weeks) that laid eggs of lower specific gravity as compared to those of younger hens (43 weeks). They suggested using shell membrane as a determinant of shell quality. Castaldo and Maurice (1988) provided that there is a relationship between phospholipid content of the chicken shell gland and egg shell strength. They indicated that weak egg shells produced towards the end of lay were laid by hens whose shell gland contained less phosphatidylcholine and a lower ratio of phosphatidylcholine: phosphatidyl-ethanolamine.

2.2.2 Nutrition

2.2.2.1 Effect of Minerals on the Hardness of Hen's Egg Shells:

Much of the egg shell quality research has emphasized the role of calcium, since approximately 98% of the shell is composed of calcium carbonate. Dietary phosphorous has become a focus of attention lately, and as a result, the nutritional strategy has shifted to include optimizing both

dietary calcium and phosphorus relative to the age and strain of the bird (Klingensmith and Hester, 1983).

Hurwitz (1978) indicated that a low dietary intake of calcium leads to a reduction in the thickness of the shell. The magnitude of this reduction is not proportional to the reduced dietary level, resulting in use of medullary bone calcium. Ousterhout (1981) suggested that egg shell quality may be improved by dietary manipulations of calcium levels, but this effect may be temporary (Hamilton and Cipera, 1981). Ousterhout (1981) reported that when protein was decreased by 1% while calcium was increased by 5% at 12 week intervals, the rate of the decline in shell quality with increasing age was slower than if a diet with average protein (16.5%) and calcium (3.5%) was used.

Atteh and Leeson (1983) studied the effect of increasing dietary magnesium level from 0.17 to 0.77% and calcium level from 3 to 4.2% for laying hens during 70 weeks. They indicated that increasing dietary magnesium level had no significant effect on calcium retention, although there was a significant ($P < 0.01$) reduction in the percentage of magnesium retained. Also they found that egg shell calcium content was significantly ($P < 0.01$) reduced and shell magnesium significantly ($P < 0.05$) increased in response to increase in dietary magnesium content. Hurwitz and Bar (1965) reported increased calcium absorption during shell formation. Alcock and MacIntyre (1962) explained the relationship between calcium and magnesium by reporting that

the presence of a high concentrations of calcium competing for the same absorptive site with magnesium may be responsible for the decreased trend in magnesium retention noted as dietary calcium level increased. Holder and Huntly (1978) studied the influence of added manganese, magnesium, zinc, and calcium level (in diet) on egg shell quality. They used laying hens (23 weeks old), which were fed practical basal diets with calcium 2.5 or 3.5%, each alone or with magnesium 1000 mg/kg added as $MgCO_3$, or manganese 65 mg/kg as $MnSO_4$, or zinc 130 mg/kg as ZnO. They found that more Ca in the diet increased shell thickness but not significantly. They also indicated that with added Mn, shell thickness was greater than without added Mn and significantly greater than with added Mg or Zn. Leach and Gross (1983) observed that manganese deficiency in the laying hen resulted in decreased egg shell thickness. They noted that the chemical analysis of the organic matrix of the eggs shells showed a decrease in hexosamine and hexuronic acid content consistent with the known role of manganese in polysaccharide synthesis.

Klingensmith and Hester (1983) investigated the relationship among available levels of dietary phosphorus (0.2, 0.3, and 0.4%) and the production of soft-shell (SS) and shell-less (SL) eggs in White Leghorns of a commercial strain. They found that egg specific gravity was significantly higher among hens consuming the 0.2% available phosphorus diet when compared to hens consuming the 0.3 or

0.4% available phosphorus diets. Also they indicated that the level of available phosphorus in this study did not significantly affect the production of SS or SL eggs.

Costello and Odom (1984) determined percent mineral ash, sodium (Na) and potassium (K) in tibia bone and egg shells of SCWL laying hens. They found significant negative correlations ($P < 0.05$) between percent bone ash and percent egg-shell ash for both mineral electrolytes. This indicated that Na and K in tibia bone can be used in egg shell formation. Sauveur and Mongin (1971) indicated that potassium is one of the ions involved in the electrolyte exchange which takes place between the egg and the shell gland tissue during shell formation.

There was a lower concentration of plasma calcium and magnesium in hens which just laid either soft-shell (SS), shell-less (SL) eggs or hard-shell (HS) eggs when compared to hens with SS/SL or HS eggs in their uteri. And the concentration of plasma phosphate was higher in birds that laid SS/SL eggs than in birds that laid HS eggs (Hester et al., 1980). Parsons and Combs (1981) reported that the concentrations of ionized calcium peak (5.67 mg%) 4 hr. after oviposition, then decrease significantly during the period of shell calcification (4.85 mg%). There was no detectable difference in total (ionized plus un-ionized) plasma calcium at the comparable periods. Feeding a calcium-deficient diet to laying hens causes a significant

decrease in plasma ionized calcium concentrations (Taylor et al., 1962).

2.2.2.2 Effect of Calcium Source on Egg Shell Strength

Egg shells from the hens fed the diets containing oyster shell, pulverized egg shell and limestone grit were significantly better than egg shells from hens fed diets containing pulverized or ground limestone as the calcium source (March and Amin, 1981; Meyer et al., 1973).

When no shell was being formed, only about 40% of the dietary calcium was being absorbed; then, this rate gradually increased to 72.4% during the late stages of egg shell formation (Hurwitz, 1970). Roland (1972) observed that the lowest serum calcium levels occurred during the early morning hours (4:00 A.M. & 8:00 A.M.) in hens receiving pulverized limestone. These results indicate that hens do not have the ability to resorb sufficient bone calcium to maintain high blood calcium levels, and that dietary calcium is absorbed at a faster rate during active egg shell deposition. Therefore, blood calcium levels can be maintained during the early morning hours due to the presence of calcium carbonate particles in the intestine (Meyer et al., 1973). Also, Kuhi et al. (1977) indicated that the larger particles allow a release of calcium to the gut at a rate that is optimum for maximum intestinal absorption during egg shell formation.

2.2.2.3 Effect of Vitamin D_3

Vitamin D_3 is hydroxylated in the liver to 25-hydroxy vitamin D_3 (25-OH-D_3) which is further hydroxylated in the kidney to the active form 1, 25-dihydroxyvitamin D_3 (1, 25$(OH)_2D_3$). It is involved in the synthesis of vitamin D-dependent calcium-binding protein for the calcium transport system in intestine (Christakos et al., 1979) and in avian shell gland (Bar and Hurwitz, 1975).

The high level (600 I.U.) of 25-hydroxy cholecalciferol in combination with oyster shell or limestone (for 4 weeks), or feeding 1α-OH-D_3 (5μg/kg diet) to laying hens for 22 weeks resulted in significantly improved shell breaking strength and tibial calcification (the hens were 62, 74, or 40 weeks of age) (McLoughlin and Soares, 1976; Soares et al., 1988). Soars et al. (1988) also reported that hens producing eggs with superior egg shell quality have a higher concentration of circulating 1, 25 $(OH)_2$-D_3 than hens producing poor quality egg shells. However, there was no improvement in shell quality (egg shell calcification or shell strength) by injection of vitamin D_3 in hens 72 and 50 weeks of age fed a 7.9% Ca diet or when the birds fed gelatin capsules containing 1,000 IU of 25-OH-D_3 each day for three days (Potts and Washburn, 1977; McLoughlin and Soares, 1976). Castillo et al. (1979) did not find a relationship between the plasma 1, 25-$(OH)_2$-D_3 and the changes in medullary bone during the egg-laying cycle.

2.2.2.4 Importance of Time of Feed Intake

Since hens stop eating food at night when the lights go out, and this occurs during the early stages of shell calcification, Simkiss (1961) and Taylor (1970) believed that crop and the skeletal system can supply additional calcium during the calcification process. Roland et al. (1973) found that much more calcium was metered from the crop during the early hours of the night than the late hours, and most of the calcium was used by midnight. They suggested that the crop was not able to provide a constant source of calcium to the small intestine for the shell calcification process.

The role of bone calcium in shell calcification was studied by Lennards and Roland (1981). This study was to determine if hens could store enough calcium in their bones to maintain shell quality when fed a 2 or 3 day supply of calcium in 1 day, followed by a calcium deficient diet the next 1 or 2 days. They found that egg specific gravity decreased on the day following consumption of the 0.4% calcium diet regardless of whether they consumed diets containing a 6.4% calcium or 9.4% calcium the day before. They concluded that hens could not maintain shell calcification for even one day with only skeletal calcium, even though there is enough calcium in the skeletal system to calcify several eggs.

Farmer et al. (1983) studied the importance of time of calcium intake on shell quality in broiler hens using three

groups of breeders. The control group was fed a diet containing 3.1% calcium. The other two groups were restricted to the same diet except that the diet contained only 0.4% calcium and the hens were intubated with 3 gm of calcium at 0800 or 1600 hr daily. All hens received the same total amount of calcium. The results of this experiment indicated that the broiler hens given most of their calcium at 0800 hr could not maintain egg specific gravity at levels comparable to that of the controls. However, hens intubated at 1600 hr showed no drop in shell weight or specific gravity. Thus, the most important time for the hens to receive calcium was during the afternoon when shell calcification was ongoing. Farmer and Roland (1982) indicated that for hens to receive maximum benefit from calcium supplementation they not only need calcium but also feed with calcium. They found that hens fed a low (0.08%) calcium diet and intubated with calcium (about 3.5 gm calcium) at 1700 hr. had significantly higher specific gravity than hens deprived of all feed for 24 hr. and intubated with the same asmount of calcium at the same time. Farmer and Roland (1982) also suggested that hens cannot absorb calcium efficiently from the digestive tract or mobilize skeletal calcium in the absence of the other nutrients. This is because they found that the hens fed the low calcium diet had significantly better shells than fasted hens. And the increase in shell weight and specific gravity was much greater than could be accounted for by the calcium

in the 0.08% calcium diet. On the same subject, Junqueira et al. (1984) found that hens fed 0.4% total phosphorus in the afternoon produced eggs with higher specific gravity as compared with hens fed 1.4% total phosphorus during the entire experiment. They concluded that dietary phosphorus level has an influence on eggshell quality when the hens is forming the egg shell.

2.2.2.5 Effect of Drugs and Hormones

Reserpine, in the diet, in all but one case (Wilkinson, 1961) was reported not to affect the egg shell thickness of chickens (Robertson et al, 1964; Robertson and Francis, 1965 and Heywang, 1965) or turkeys (Casey et al., 1963).

Aspirin at 0.1 or 0.2 percent in diet of laying chickens had no significant effect on egg shell thickness (Thomas et al., 1966).

Feeding chlortetracycline or oxytetracycline (Eoff et al., 1962) or penicillin (Gabuten and Shaffner, 1954) increased % shell and egg specific gravity or decreased the percentage of cracked eggs.

Sulfanilamide decreased shell quality (Becker and Bearse, 1962).

Feeding dry thyroid to chickens increased egg shell weight (Asmundson and Pinsky, 1935). Feeding thyroprotein (iodinated casein) increased egg shell quality (Savage et al., 1952). Although Berg and Bearse (1948) did not find a significant effect on egg shell thickness due to the

iodinated casein. Thiouracil, an antithyroid product, has been reported to decrease egg shell thickness (Berg and Bearse, 1948). Subsequently, Berg and Bearse (1951) and Hunt and Attken (1962) did not observe a significant effect on egg shell thickness or egg specific gravity when they used thiouracil. Feeding thyroxin (0.02% of the diet) or stilbesterol (0.2 mg/hen/day), resulted in an increase in egg specific gravity (Gabuten and Shaffner, 1954). These authors found that testosterone propionate (0.2 mg/hen/day) decreased the egg specific gravity. Polin and Sturkie (1957) indicated that parathyroidectomy resulted in the production of soft-shelled or shell-less eggs.

2.2.3 Effect of Disease

New Castle, infectious bronchitis and chronic respiratory disease reduced egg shell thickness (Quinn et al, 1956, Crinion, 1969 and Carson et al., 1954).

2.2.4 Effect of Physiology of the Bird

2.2.4.1 Interval between Oviposition

The amount of shell deposited is a linear function of time in the shell gland after plumping (Burmester et al., 1939). The length of time in the shell gland can influence the amount of shell deposited and, consequently, the shell strength. Ephedrin sulfate delayed oviposition of a chicken consistently laying shell-less eggs and allowed deposition of egg shell calcium (Polin and Sturkie, 1955). However,

when ephedrine was injected into normal hens, egg shell weight was decreased (Sturkie et al., 1954). Choi et al. (1981) have shown that the longer the interval between eggs the more shell is deposited on the egg. This influence is more noticeable if management, feeding, and lighting are disrupted and if an egg is held in the uterus too long so that the next egg in the clutch does not get sufficient shell (Siegel et al., 1978).

2.2.4.2 Effect of Acid Base Balance

Winget and Smith (1959) showed that an increase of H^+ ion concentration favours calcium liberation from the vitelline-calcium complex. Winget et al. (1958) indicated that it is likely that calcium drawn from the blood by the uterus is found in ionic form and that its immediate and local course is calcium complexed with proteins. Hodges (1965) found that a relatively low uterine pH makes the dissociation of protein-Ca complex easier. Taylor and Hertelendy (1961) showed that a pH increase from 7.55 to 7.87 decreases blood ionic calcium by 20%.

2.2.4.3 Frightening of Chickens or Foreign Objects in Uterine Tissue

Frightening of chickens has resulted in a decreased shell thickness (Stiles and Dawson, 1961). Also foreign objects, such as a thread, placed in the uterine tissue results in SL eggs (Jaap and Muir, 1968).

2.2.5 Effect of Age of the Birds

Peterson (1965) has proposed that the amount of Ca the hen absorbs and retains and the skeletal Ca available for shell calcification decrease with age. The studies by Roland et al. (1975) and Roland (1980) indicated that the decline in shell strength with age was not due to a decline in the ability to absorb or mobilize Ca, but was due to an increase in egg size with progressing age without a concomitant increase in shell weight (Ousterhout, 1981). Also Roland et al. (1975) and Roland (1980) showed that the amount of shell deposited did not decrease but remained reasonably constant or slightly increased with age. Roland (1979) found that an egg that had the greater increase in size with increasing age had the greater decline in shell quality. This was (Roland et al., 1978) due to an increase in egg weight at a faster rate than the increase in shell weight, which resulted in a decrease in the amount of shell to cover the egg.

However, Mehner and Torges (1967) indicated that egg breaking strength can be improved after a laying pause in old hens.

Ousterhout (1981) observed that a reduction in the dietary protein level of aging hens can result in a decreased egg size without increasing shell strength.

2.2.6 Effect of the Management

Hamilton et al. (1979) reported that although factors such as genetic potential, diet, temperature, and age of the flock may be fixed, management can be an important factor to the extent of shell breakage. They found that as the number of hens in a cage is increased, the chance that the egg will be damaged before it reaches the collection belt is increased. Also, they indicated that disruption of laying hens can result in breakage of an egg shell in the uterus. Yannakopoulos and Morris (1979) found an improvement in shell strength, when an ahemeral light cycle (28 hours) was used at a time near the end of lay.

2.2.7 The Effect of Heat Stress

2.2.7.1 Effect of Heat Stress on Feed Intake and Shel Quality

Mueller (1959) studied the effect of environmental temperature (85°F or 30°C) on feed intake, calcium balance, and serum calcium levels and to find out if differences in these characteristics explain the effect of environment on shell thickness. He used S.C.W. Leghorns (from 174 to 450 days of age). The results of his experiment indicated that high environmental temperature reduces shell thickness, blood calcium levels and feed intake. The lower calcium intakes at high temperature were offset by better calcium retention so that all the birds retained some quantity of calcium. Also the correlation between calcium intake and shell thickness was small and insignificant. This indicates

that hens with high and low calcium intakes produced eggs of about same shell thickness.

Mueller also found a negative correlation between serum calcium level and shell thickness. This indicated that hens having high serum calcium levels produced eggs with thinner shells than hens having low serum calcium levels. He concluded that it is unlikely that the reduction in shell thickness at high temperature was caused by the concomitant reduction in calcium intake or serum calcium levels. He also indicated that the positive correlation between calcium intake and serum calcium levels may be due to increased calcium absorption in the digestive tract when the calcium intake increases. Campos et al. (1960) studied the effect of heat stress on egg quality. They found a reduction in shell thickness and also a marked reduction in feed consumption from heat stress. De Andrade et al. (1976) subjected two groups of S. C. W. Leghorn pullets to either a 21 or 32°C environmental temperature. The birds were fed either a typical corn-soy laying diet (15% protein, 2.8% Ca, 0.57% avail. P, 2,915 Cal. M.E./gm plus vitamins and minerals) or a High Nutrient Density (HND) diet (20% more of all nutrients except energy which was increased only 10%). They found that food consumption, egg weight, specific gravity of eggs, shell thickness, and % shell decreased from heat stress. The use of (HND) diet partially alleviated the effects of heat stress on egg weight but there was no significant improvement in specific gravity due to the

significant increase in egg weight with the (HND) diet. This was supported by Hurwitz and Griminger (1962) who reported that egg specific gravity was affected by change in egg weight and will therefore fail to reveal true changes in shell quality when changes in egg weight take place. De Andrade et al. (1976) observed that there was an increase in shell thickness observed with HND diet under heat stress; yet it was inferior to the control diet at 21°C. However, De Andrade et al. (1977) found that High Nutrient Density diet did not improve specific gravity, % shell or shell thickness under heat stress. They added that the calcium consumption was increased in all environments by feeding (HND) diet. Vasquez and Teeter (1986) used the force feeding (70, 85, 100 and 115% of thermoneutral (24°C) ad lib. consumption. This was to study the effect of heat stress (35°C) on egg quality. They indicated that heat stress depressed egg weight (3.2%), shell weight (7.4%), shell thickness (9.3%) and specific gravity (1%). Increasing feed consumption of heat-stressed layers by force feeding increased egg weight (2.6%), shell weight (0.4%), shell thickness (0.7%) and specific gravity (0.2%). They also observed that there was a linear increase of hen mortality with intake. These authors also noticed that increasing feed consumption of the heat stressed hens enabled partial recovery of egg quality. There was a beneficial effect from adding fat to the diet of broilers during heat stress, as a means of reducing the

Specific Dynamic Action (SDA) of the diet (Dale and Fuller, 1980). The latter authors indicated that with constant high temperature stress, the situation is so severe as to override completely any beneficial effect due to the reduced SDA of the diet. Pastro et al. (1969) found that the body temperature increased in lysine deficiency.

2.2.7.2 Effect of Heat Stress on Egg Shell Quality

Campos et al. (1960) studied the effect of heat stress (100°F or 37°C) for 24 hours using a fast rise (4°F or 2.2°C/hr) and a slow rise (5°F for 2.8°C/24 hrs) in air temperature on Rhode Island Reds, W. Leghorns and New Hampshires (8 to 15 months of age). They found that the rate at which temperature rises above 70-80°F influences the quality of eggs laid and the amount of feed consumed, but not egg production. They also reported that the rate of temperature change alone did not exert marked adverse effect on egg size or albumin quality, since in other tests employing fast (3.5°C/hr.) rises in temperature to 37.5°C for a duration of only 6 hours, no effect was observed. They also indicated that, of the egg characters considered in their experiment, shell thickness was found to be more sensitive to rises in ambient temperature than egg production. The degree of reduction was greatly influenced by the prevailing temperature when shell deposition was taking place. Also, they noticed a marked reduction in feed consumption with fast rises in temperature when compared to

that in case of slow rises. Miller and Sunde (1975) showed that the increase in the environmental temperature decreased rigidity of the egg shell, egg weight and feed consumption, but egg production was not markedly affected over an extended period of time (up to 90 days). They indicated that constant high temperature (32°C) resulted in lower shell deformation (0.018 vs. 0.023 mm) than acyclic (over 10 hours) hot temperature (26-38°C). Arima et al. (1976) studied the effect of weekly constant environmental temperatures on egg shell quality using hens of two age groups. They found that neither 14 months old nor young 8 months old hens showed a marked change in egg weight at environmental temperatures up to 32.5°C. However, egg weight decreased to about the same degree in both age groups subjected to 35.0°C. They also found that egg shell strength gradually decreased when the environmental temperature in both age groups was increased to 29.5°C and higher. The decline in shell strength was more severe in older than in younger hens.

Wolfenson et al. (1979) subjected mature laying hens (10 to 15 months of age) to ambient temperature (34 to 40°C) sufficient to maintain body temperature of 43°C for periods of 6 to 7 hrs during the day (from 10.00 to 16.00 hr) or the night (from 20.00 to 03.00 hr). There was no reduction in total daily food consumption. The effect of heat stress during the day was mostly on egg shell quality. The heat

stress at night caused a significant decrease in egg production.

Laying hens exposed to a 24 hr linear temperature cycle ranging from 26.7 to 35°C had a significantly lowered egg shell breaking strength and a significantly thinner shell than the hens exposed to the temperature cycles of 21.1 to 35°C or 15.6 to 35°C (Deaton et al., 1981). White Leghorn hens 21, 25, and 33 weeks of age had decreased feed consumption, egg production (except for the hens 21 weeks of age), egg weight, and egg shell thickness from 35°C stress (after a 3-day test period) (Tanor et al., 1984). Increases in energy and calcium intake partially helped to maintain normal egg production, egg weight, and egg shell deformation.

2.2.7.3 Effect of Heat Stress on Physiology of the Birds and Shell Quality

2.2.7.3.1 The Effect on Pulse Rate, Blood Pressure, Body Temperature and Oxygen Consumption

Thornton (1962) observed that an increase in environmental temperature to 35°C brought about a rise in body temperature of both control hens and those given 44 mg ascorbic acid/kg diet. Body temperature increased more in control hens than in hens fed vitamin C. Thornton also found that the control and ascorbic acid supplemented hens exhibited an initial decrease in body temperature when the ambient temperature was increased followed by the increase in body temperature. Also, there was a marked reduction in

oxygen uptake (in case of the decreased body temperature) by each group followed by a higher oxygen uptake as the body temperature increased. S. C. W. Leghorn hens moved from 21°C to 35°C and from 5°C to 35°C environmental temperatures had slowly decreasing pulse rate and blood pressure, and increasing respiratory rate, body temperature, and oxygen consumption; the latter decreased later in the exposure period (Harrison and Biellier, 1969). Also, oxygen consumption was significantly reduced at 35.0°C for 13-month old S. C. W. Leghorn hens, but not for 15 and 18-month old birds. Pullets subjected to an abrupt change in temperature from 21 to 35°C laid eggs with lower specific gravity of eggs (with no change in egg weight) within the first day of exposure to 35°C (Harrison and Biellier, 1969). Panting caused the birds to develop a respiratory alkalosis compensated for by renal excretion of calcium bases. This change in calcium metabolism was the reason for the rapid change in specific gravity. All parameters approached a plateau or acclimated functional level within 12 to 24 hours following exposure of the hens to the increase in ambient temperature.

2.2.7.3.2 The Effect of Heat on the Blood and Acid-Base Balance

Adult male turkeys subjected to 32.2 or 37.8°C had blood tests at 0, 1, 7, 14, 21 and 28 days of stress (Parker and Boone, 1971). There was a low red blood cell count due to hemodilution beginning at 14 days of 37.8°C stress. But

not until 28 days at 32.2°C did hemodilution occur. Hemodilution occurred on day 14 for the 37.8°C treatment, and it continued throughout the remainder of the study. Blood pH was the same for both heat treatments. Most of the changes in criteria became constant during the 37.8°C stress at between 14 and 21 days. Acclimatization to the heat occurred generally between 7 and 14 days. Stress at 37.8°C (for 21 or 28 days) proved to have long lasting deleterious effects. Kohne and Jones (1975a) studied the effect of the acute hyperthermia of 49°C for 165 minutes on PCO_2, pH and plasma electrolytes in non-laying female turkeys to avoid the effect of egg shell deposition on the blood parameters measured. The pH and PCO_2 of the turkeys did not change significantly during the first 60 minutes of the experimental period, but as the ambient temperature reached 43°C, the venous blood pH increased significantly (from 7.40 to 7.69) and the PCO_2 decreased significantly (from 54.5 to 16.3 mmHg). Also, there was a significant decrease in plasma sodium, total calcium, magnesium and inorganic phosphorus from the 43°C heat, while plasma potassium was significantly increased. In another study, Kohne and Jones (1975b) exposed turkey hens to stepwise increases in ambient temperature from 21 to 25 to 30 to 35°C at two week intervals. They studied the effect of heat stress on venous blood pH, PCO_2 and plasma electrolytes during the second week after each increase in ambient temperature. There was a significant decrease in plasma sodium and a significant

increase in plasma potassium as the ambient temperature rose to 30°C. Also, there was a significant increase in PCO_2 from 52.9 mmHg at 21°C to 62.5 mmHg at 25°C and then declined at a significant rate to 56.2 mmHg at 30°C and 53.7 mmHg at 35°C. There was no significant difference in venous pH. The shell thickness or egg size was not significantly correlated to any of the plasma electrolytes measured.

The relationship between PCO_2 and egg shell quality was studied by Mongin (1968) who indicated that the thinning of egg shells in hot weather was due to hyperventilation of the lungs with a resultant loss of CO_2. Mongin and Lacassagne (1966) reported that hyperventilation due to high temperature causes the bird to lower its blood carbon dioxide tension and thus its blood bicarbonate level. Gutowska and Mitchell (1945) indicated that the bicarbonate ions are the source of shell carbonate. Hodges and Lorcher (1967) found that when ^{14}C bicarbonate ions were injected into the blood there was relatively little labelling of the shell.

2.2.7.3.3 Effect of Heat Stress on the Hormones and Metabolism

High temperature decreased thyroxin production in laying hens (Bell and Freeman, 1971). The reduction in egg weight under heat stress may be due to a reduction in calcium metabolizing hormones because of a negative effect on the pituitary and/or hypothalamus (Miller and Sunde,

1975). Less follicle stimulating hormone may account for a decreased ovum weight that results in lower egg weight.

Edens (1976) showed that injection of a 5% Na^+ solution was followed by a significantly greater increase in body temperature during exposure to 45°C than in controls (using 3-week old chickens) at the same temperature. Edens also reported that corticosterone may have a function in survival during heat exposure. One of the crucial functions of corticosterone is the maintenance of phosphorus and calcium concentrations in the blood, thus protecting against cardiovascular failure. Adult roosters exposed to 42.2°C survived longer than controls when they were fed thiouracil (inhibitor of thyroid hormone synthesis) and less than the controls when they received thyroxine (Fox, 1980).

Goto et al. (1979) studied the effect of heat stress (32°C) on the concentration of carbonic anhydrase which is one of the key factors that is involved in egg shell formation. Concentrations of carbonic anhydrase in uteri and kidney tissue of hens were significantly increased from 0.33 and 1.85 to 0.43 and 2.31 Eu/mg after high temperature (32°C) exposure (Goto et al., 1979). The activity of this key enzyme in shell formation was significantly decreased from 1.00 to 0.61 Eu/mg in whole blood. They indicated that the concentration of carbonic anhydrase of the uteri of hens that laid normal shell eggs was significantly higher than that of hens laying eggs with highly pimpled shells. Also,

egg shell thickness was significantly decreased as the pimpling of egg shell was increased.

2.2.7.3.4 Effect of Heat on Heat Tolerance

Randall and Hiestand (1939) and Randall (1943) found that panting depended on rectal temperature,whether the bird was heated from outside, or internally, indicating that the stimulation of the panting center was central, and did not depend on skin temperature. Hutchinson and Sykes (1953) used 3 cocks and 3 hens to study the effect of hot environment on acclimatization of the fowls. They found that twenty-four daily exposures for 4 hr., or for shorter periods, significantly improved heat tolerance at 37.2°C. According to Arieli et al. (1979) metabolic and insulative modifications are involved in seasonal acclimatization which is slow and gradual in the fowl. Arieli et al. reported in 1980 that at 32°C the adjustment potentials of vasomotor function and metabolic rate are exhausted in hens. Vasomotor function reached a maximal value and the metabolic rate its minimum. They suggested that 32°C probably represents an upper limit for homeothermy in the hen. Sykes (1983a, b) exposed some birds for up to 4 hrs daily to an ambient temperature that brought about a steep rise in body temperature. Unacclimatized birds showed a rise of 4°C in 1 hour; whereas, those fully acclimatized showed a rise of only 1.5°C in 4 hours.

2.2.8 Alleviating Thermal Stress in Poultry

2.2.8.1 Dietary Manipulation

De Andrade et al. (1977) studied the effect of elevated temperatures of 21 and 31°C constant and 26.7-35.6°C cyclic on egg production and shell quality. The hens in each environment were fed either a typical corn-soy laying diet or High Nutrient Density (HND) diet that contained 25% more of all nutrients except energy which was increased 10%. During heat stress, the HND diet increased egg production and egg size in comparison to the typical diet but not egg specific gravity, shell thickness or % shell.

Vitamin C may be one of the nutrients that should be added to poultry diets for hens subjected to high environmental temperature. Hunt and Aitken (1962a) indicated that blood ascorbic acid (AA) was significantly depressed in Leghorn hens maintained at 35°C, when compared to hens maintained at 21°C. Also, there was a significant decline in plasma AA 4 hours after heat stress at 42°C and AA levels returned to preheat values within 24 hours (Nathan et al., 1976). In contrast, Kechik and Sykes (1978) observed a significant increase in plasma AA in fowls exposed to 36°C for three hours.

Thornton (1960) found that dietary ascorbic acid at 44 mg/kg diet) was effective in maintaining shell thickness during periods of increased environmental temperatures when the calcium level was 2.5 or 3.0% in the diet. Thornton and Deeb (1961) showed that iodinated casein in the diet was

accompanied by increased blood AA and that the addition of thiouracil to the diet decreased vitamin C blood level. Apparently, the metabolic rate of the chicken is associated with its ability to synthesize ascorbic acid. Thornton (1962) reported that 44 mg ascorbic acid/kg diet had no effect on the body temperatures of laying hens maintained in a moderate environment (24.4 to 26.1°C). Elevating the ambient temperature to 37.8°C resulted in significantly lower body temperatures in AA-supplemented hens as compared to controls. Lyle and Moreng (1968) found that ascorbic acid supplementation significantly limited the increase in body temperature of Leghorns exposed to 29°C for one week. There were no significant differences in the cloacal temperatures of chicks 4 weeks of age supplemented with 1000 ppm ascorbic acid, when compared with nonsupplemented chicks maintained at 22°C or acutely exposed to 43°C, according to Pardue et al. (1985).

Ahmad et al. (1967) studied the effect of adding AA at 44 mg/kg diet on progressive heat stress using 13, 15 and 18 months old S. C. W. Leghorns. AA did not maintain egg shell thickness of the 13-month old hens. The 15-month old hens being fed ascorbic acid maintained shell thickness at 29.4°C but not at 35.0°C. There was an adverse effect on shell thickness due to supplementation of ascorbic acid in the diet of hens 18 months of age. Only in the 18-month old hens was there a significant reduction in body temperature (42.09°C for control vs. 41.79°C for treated group) due to

AA suplementation. Shell deformation or percentage of cracked eggs from 10 or 13 months old hens was not influenced using 100 or 500 mg AA/kg diet at 32.2 or 33.3°C environmental temperature (Isnak and Sykes, 1974). Oluyemi and Adebanjo (1979) studied the effect of AA at 50, 100, 150 and 200 mg/kg diet on egg shell quality of 30- and 62-weeks old hens held at 22.8 to 36.9°C. The shell thickness of the younger birds was depressed from feeding AA. This trend appeared to be reversed in the older hens in which AA was beneficial at dietary concentrations of 100, 150 or 200 mg/kg.

Deetz and Ringrose (1976) found that heat stress increased potassium excretion from the body. They suggested that 0.4% potassium in the diet is adequate for meeting potassium requirements at temperatures of 26.7, 32.2 and 37.8°C. They recommended that 0.6% dietary potassium may be a more suitable level for assurance against a potassium imbalance at elevated temperature.

Blood pH of 7.40 was greater in panting birds kept at 32°C than in heat-stressed nonpanting (7.28) birds or those kept at 24°C (7.28) (Teeter et al., 1985). Adding 0.3 or 1% ammonium chloride to diets decreased blood pH ($P < .01$) to 7.19. Also, adding calcium chloride (0.5, 1%) to a mixture of 1% NH_4Cl plus 0.5% $NaHCO_3$ lowered the dietary Na:Cl ratios. The blood pH of panting birds declined linearly with lowered dietary Na:Cl ratio, but not to the extent of the 1% NH_4Cl level.

Rossoff (1974) indicated that acetaminophen (N-acetyl-p-aminophenol) increased growth rate and feed utilization in chickens. Acetaminophen is analgesic, antipyretic and is the active metabolite of phenacetin. It was used at levels of 0.5 gm/kg of feed for cockerels and 2.0 gm/kg of feed for hens. Oluyemi and Adebanjo (1979) studied the effect of aspirin (acetylsalicylic acid) on egg shell quality from hens kept at 22.8 or 36.9°C environmental temperature. Aspirin at 0.15 and 0.20% of diet gave better shell thickness (mm) for eggs from young hens (30 weeks old) than levels of 0.05 and 0.10%. There was no significant effect due to aspirin on shell thickness when fed to old hens (62 weeks of age). Gilbert et al. (1982) indicated that daily ingestion of up to 400 mg aspirin for 3 weeks, or 750 mg aspirin for 2 weeks had no effect on egg production, oviposition time or bird behavior.

2.2.8.2 Water Treatment for Heat Stress

Balnave and Scott (1986) studied the effect of adding mineral salts to drinking water on egg quality. They added the following minerals to municipal town water: NaCl(250 mg/l), KCl (40 mg/l), $CaCl_2$ (120 mg/l), hydrated $MgSO_4$ (300 mg/l), anhydrous $CuSO_4$ (200 mg/l) and $NaNO_3$ (350 mg/l). The minerals were added to the drinking water over a 6-week period during which birds were allowed free access to food and water. The inclusion in the water of low concentrations of all the mineral salts substantially increased the

incidence of shell defects particularly those supplements with chloride ion. The increase in shell defects was not associated with the pH of the water. In all cases water consumption was reduced by between 10 and 20% as compared to intake by birds receiving no added minerals. Also, the effect was not related to a decreased feed intake. They suggested that the adverse shell effects noted from the NaCl and KCl supplements may be due to the increased intake of chloride. Balnave and Yoselewitz (1987) also noted a significant linear increase in egg-shell defects by 60 week-old laying hens given increasing concentrations of NaCl in drinking water (up to 600 mg/l).

The addition of 1000 ppm AA to the drinking water of cockerels, 4 weeks of age, for 4 hours resulted in a significant increase in plasma AA (Pardue et al., 1984).

Market-age broilers were given Banamine at 0, 1.32, 5.26, and 10.52 mg/l as a treatment for heat stress (35°C for 8 hours) (Oliver and Birrenkott, 1982). Banamine is a nonsteroidal anti-inflammatory drug thought to interfere with prostaglandin production at the level of cyclo-oxygenase (Ferreira, 1979). There was no effect of Banamine on peripheral venous prostaglandin F level. However, Banamine at 10.52 mg/l resulted in less survival time than the level of 5.26 mg/l, 242 vs 337 minutes, respectively. The authors presumed that Banamine may be exerting an effect through its analgesic activity.

2.2.8.3 Using Other Measures

Oluyemi and Adebanjo (1979) tried to combat thermal stress (22.8 to 36.9°C) in 30 and 62-week old laying hens with the following treatments: 1) control; 2) forced ventilation at the rate of 9 cfm/layer; 3) drinking water at 0°C between 1200 to 1800 hr; 4) dunking in cold water (0°C) at 1300 to 1400 hr; 5) dunking in water at 4°C at 1300 to 1400 hr; 6) feeding only at night time, 1800 to 0600 hr under artificial lighting (10 lux); 7) feeding confined to day time (0600 to 1800 hr), with artificial lighting (10 lux). Egg size of older hens was significantly ($p < .05$) heavier with night feeding whereas the younger layers laid at a higher rate but produced lighter eggs. The shells of the older birds were thicker with night feeding and drinking cold water (0°C) than with any other treatment.

3. Experimentation

A total of 256 Single Comb White Leghorn (SCWL) hens forty six and sixty one weeks of age were used in this study. The objectives of the experiments were mainly: 1. To find the minimum time of exposure to heat stress that can result in an obvious reduction in shell quality; 2. To study the effect of heat stress (35°C) on egg shell quality during egg shell formation, and to study the importance of allowing feed and/or water during that time. 3. To overcome the effect of heat stress on egg shell quality during the time of egg shell formation. In the last part of the study, the lighting program was modified so that the light was on from 12 midnight to 4 p.m. This was done by changing one hour every two days. The shift in time was done to change the time of egg shell formation (night time for hens) to match the regular day time.

The experiments were conducted at the Department of Animal Science, Michigan State University. The hens came from a commercial flock of SCWL hens kept at the Poultry Teaching and Research Center. The birds were kept in single cages of wire floored batteries in a room kept at 21°C. In this room, birds were supplied with feed and water ad

libitum, and artificial light was on a schedule of 16:8, (light:dark) hours.

An environmental chamber with room for six hens in single cages was used to study the effect of heat stress on egg shell quality. Air was provided from an external source and dispersed through the chamber by a fan. Artificial light was provided so that the hens could see the feed and/or the water during the experiment. In some experiments no feed and water were supplied during the time the hens were in the heat chamber. An electric heater was used to provide the required heat. It took 30 to 60 minutes for the temperature to reach 35°C. When this temperature was attained, the birds started panting and continued panting to the end of the experiment. The hens and treatments were randomly distributed within the environmental chamber to reduce possible position influences.

Three eggs were collected from each hen before and after each treatment to measure egg quality (egg weight; shell weight; % shell of egg weight; mg calcium per unit surface area, Ca/mm^2 (Carter, 1975)). The change in egg quality (due to the treatment) was measured by the difference between the average measurements of egg quality for the three eggs before treatment vs the measurements on each of the three eggs immediately following the treatment. For example, if the average of the egg weight for the three eggs before treatment was 60 grams and the egg weight for the first day following the stay in the chamber was 58

grams. The change in egg weight due to the treatment will be (58-60) - 2 grams. Eggs were collected and weighed daily. After weighing the eggs and removing their contents, the shells were carefully washed, dried, and weighed. Percent shell (weight of dried shell plus membranes/total egg weight x 100) was determined. For some experiments a model 158 Corning Blood Gas System was used for determining the pH, PCO_2 (partial pressure of carbon dioxide) and HCO_3^- (bicarbonate) activities of whole blood immediately after each sample was drawn from the wing vein of each bird.

3.1 Experiment 1

This experiment was designed to study the effect of acute heat stress (35°C) for 1, 2, 4, and 6 hours on egg quality. SCWL hens, approximately 61 weeks of age, were used after one month of acclimatization at 21°C. During this period, the birds were fed a commercial laying diet (14.0% crude protein, 3.2% calcium and total phosphorus of 0.55%). They received 15 hours artificial light per day. Following the acclimatization period, a total of 12 birds for each treatment were subjected to 35°C for 1, 2, or 6 hrs., and 24 birds for 4 hours. Another group of hens was used as a control in the environmental chamber for each of the same lengths of time at the chamber temperature (23°C). During the experiment in the environmental chamber, only water was available. After being used in the heat chamber, the hens were kept at 21°C for two weeks, at least, before

being used again. This two week period was to avoid acclimatization to the heat. Wolfenson et al. (1979) reported that one week after exposure to 34 to 40°C for 6 to 7 hours was enough for recovery from the high temperature exposures.

3.2. Experiment 2

The experiment was designed to study the effect of heat stress (35°C) on the first egg following the exposure of young hens 46 weeks of age to 6 hours of heat stress during the day time (without active shell deposition). During the acclimatization period (about a month), the birds were fed a laying diet which was formulated to ensure an adequate intake of all nutrients, recommended by N. R. C. (1984) (Table 1.a).

There was a period of recovery from heat stress for 3 to 4 weeks before hens were used again in the heat chamber. In this experiment the temperature of 35°C was reached in 30 minutes. A total of 12 hens were heat-stressed, and 12 were in the chamber without heat. Ambient temperature was about 23°C. During the stay in the chamber only water was available. Egg quality was measured as described in experiment 1. Also in this experiment, feed consumption was determined for each hen over the next 24 hours after leaving the chamber. Body temperature was taken for each bird as they were placed into the chamber and when they were removed. Body temperature was read directly from a

thermometer inserted into the rectum to a depth of six centimeters and held in place for two minutes prior to a reading (Harrison and Biellier, 1969).

Table 1.a The percentage composition of the diet fed to the hens used in the experiments from 2 to 10.

Ingredients	%
Ground yellow corn	63.5
Soyabean meal (44%)	23.36
Corn oil	2.27
Dicalcium phosphate	1.19
Ground limestone	9.00
Iodized salt	0.366
Vitamin and mineral mix (1)	0.250
DL Methionine	0.072
Total	100.00
Calculated values	
crude protein, %	15.87
calcium, %	3.75
available phosphorus, %	0.35
metabolizable energy, K cal/gm	2.848
lysine, %	0.836
ether extract (fat), %	4.87
methionine + cystine, %	0.61

<Supplies per kg diet>

(1) Vitamin A, 11,013 IU; vit D_3, 3304 ICU; vit E, 16.5 IU; vit K_1, 2.2 mg; vit B_2, 5.5 mg; pantothenic acid, 16.5 mg; Niacin, 55.1 mg; choline chloride, 330.0 mg; vit B_{12}, 0.014 mg; biotin, 0.06 mg; vit B_1, 1.10 mg; vit B_6, 1.65 mg; folic acid, 0.55 mg; Mn, 75 mg; Se, 0.1 mg; Zn, 75 mg; Fe, 37.5 mg; Cu, 0.75 mg; I, 0.75 mg.

3.3 Experiment 3

The purpose of this experiment was to study the effect of 35°C for 6 hours on egg quality of hens stressed at the time of egg shell formation and when there was no egg shell formation. The photoperiod was from 0700 to 2300 hr. The effect of heat stress on egg quality during the time of active shell deposition was studied during the hours of 1600 to 2300. The effect of heat stress on egg quality from hens without active shell deposition was studied at the time between 0800 and 1600 hr. During the stay in the chamber only water was available. Feed comsumption and body temperature were determined.

3.4 Experiment 4

Heat for 4 or 6 hours or a stay in the chamber without heat was applied (during the time of egg shell formation) to forty eight hens in 2 x 2 factorial design. During the time the hens were in the chamber only water was available. Body temperature was determined just before and after hens were in the chamber. Feed intake was determined 24 hours after the hens were in the chamber.

3.5 Experiment 5

This experiment was designed to study the effect of feed and/or water, heat or no heat during egg shell formation. A total of 12 birds for each treatment was used. Four hours exposure time in the chamber was used. Hens were palpated

for an egg in the uterus, and only those with an egg were used in this experiment.

3.6 Experiment 6

SCWL hens approximately 61 weeks of age were used after a 30-day period of acclimatization to 21°C. They were used to study the effect of drugs and chemicals to alleviate the effect of 35°C heat for 4 hours during egg shell formation. Feed consumption, body temperature, water intake and drug consumed were determined as well as egg quality. Only birds actively depositing shells were used in the heat chamber. At least 3 to 4 weeks were allowed after heat stress before birds were used in another experiment.

Six treatments were used in this experiment consisting of a control diet, (table 1), which was supplemented with vitamin C at 50 or 150 mg/kg; aspirin, 0.2%; potassium bicarbonate, 0.9% or sodium bicarbonate, 0.75% (by proportionate displacement of all ingredients). Hens were fed the experimental diets for at least 3 days before they were placed into the environmental chamber and stressed at 35°C. During the heat stress, only water was available. Egg weight,shell weight and % shell were measured. Also body temperature before and after treatment and feed consumption over the next 24 hours for each hen were also determined.

3.7 Experiment 7

In this experiment, three of the last six treatments were duplicated. These treatments were: the control diet, aspirin

at 0.2%, and potassium bicarbonate at 0.9%. Shell thickness was added to the previous egg quality measurements. Blood samples from the brachial vein were taken at the end of the 4 hours in the heat chamber, using a heparinized syringe (Corning Medical, Corning Glass Works, Medfield, Massachusetts 02052, USA). The pH, PCO_2 and HCO_3^- activities of whole blood were determined. In addition, body temperature of each hen was taken at the time of the blood sample.

3.8 Experiment 8

This experiment was designed to compare the blood pH, PCO_2, and HCO_3^- of hens stressed at 35°C compared to the blood of hens in the chamber at 21°C. Twelve hens were used for heat study and 10 hens for the no heat study. The blood samples were taken only from a bird with an egg in the uterus. During heat stress only water was available.

3.9 Experiment 9

This experiment was carried out (during the time of egg shell formation) to determine if the effect of heat stress could be counteracted by drugs and minerals in the water. In this experiment one of the following compounds was added to tap water (control): 1.6 gm KCl/l (or 0.839 gm K/l); 8.30 gm $CaCl_2$/l (or 2.996 gm Ca/l); 1.083 gm aspirin/l; 0.267 gm acetaminophen/l, and 4.81 pH buffer solution. The buffer solution was made of 1.2 gm KOH/l and citric acid in deionized water. It contained 0.833 gm K/l of water. A pH meter, Model

825 (Fisher Scientific) was used to adjust the pH of the buffer solution. Aspirin was provided as sodium acetyl-salicylate (Miles Inc., Elkhart, IN 46515 USA). Acetaminophen is product of Mead Johnson Nutritionals, A Bristol Myers Company, Evansville, IN 47721 U.S.A. The following measurements were taken: egg weight, shell weight, % shell, body temperature, feed intake/day. Also water and drug intake during the heat treatment were measured. There was no feed available during the heat of 35°C for 4 hours.

3.10 Experiment 10

In this experiment, five different treatments were used during heat (35°C) at the time of egg shell formation for four hours (Table 1.b). Each hen was given orally 2 equal doses of each of KCl and acetaminophen. The first dose was given orally in a volume of 5 ml distilled water about 2 hours before hens went into the environmental chamber. The second dose was given in the water allowed during heat stress. A dose of 5 mg Vitamin C/hen was given orally two-hours before providing carbonated water during heat stress, or in addition to the oral dose of the acetaminophen. Carbonated water was provided in the commercial form for human consumption which contains corn syrup. In this experiment both water and feed were available during heat stress. Feed consumption, water intake, drug intake, body temperature, egg weight, shell weight, and % shell were determined in this experiment.

Table 1.b Treatments used in an attempt to alleviate the effect of heat at 35°C for 4 hrs. during egg shell formation.

Treatment number	Compounds that are used	gm/hen (2 hrs before)	gm/l water allowed (during heat stress)
1	None in tap water (control)	0	0
2	KCl (level 1)	0.286 KCl (or 0.15 K)	1.907 KCl (or 0.999 K)
3	KCl (level 2)	0.572 KCl (or 0.30 K)	3.814 KCl (or 1.998 K)
4	carbonated water + vit C	carbonated water + 5 mg vit C	carbonated water
5	acetaminophen + vit C	0.096 acetamino-phen + 5 mg vit C	0.640 acetamino-phen

Statistical Analysis

Data collected (for each treatment in each of the three stages) were subjected to analysis of variance and simple linear regression using the Stat View 512+ program by Brain Power Inc., 24009 Ventura Boulevard Calabases, CA 91302 for the Macintosh computer. Means were further subjected to Fishers (Protected) least significant difference (FPlsd). Analysis of covariance was used for some data, according to Steel and Torrie (1980).

4. RESULTS

4.1. Experiment 1. Effect of heat (35°C) or no heat treatments for 1,2,4, and 6 hrs on egg quality using SCWL hens 61 weeks of age

4.1.1. The Effect on the Change (gm) in Egg Weight

Hens held in the heat chamber at ambient temperature for 1, 2, 4, or 6 hours laid eggs averaging 0.32 gm more than the 3 eggs they laid before going into the chamber. On the other hand, the hens stressed at 35°C for 1, 2, 4 or 6 hours laid eggs that averaged 0.218 gm less than the three each hen laid before being heat stressed (Table 2.a). There was no consistent trend for egg weight to decline with increasing hours of heat (Fig. 1). The loss in egg weight was as great after 1 hour of heat stress as that after 6 hours. Furthermore, the eggs laid by hens stressed for 4 hours actually weighed more than their controls, or those laid by hens in the chamber for 4 hours but not heat stressed (Fig. 1). The average gain or loss in egg weight for the hens in the chamber at various times, whether heat stressed or not, is provided in tables 2.a, b, and c. No definite trend for a loss of egg weight associated with acute heat stress is noted. Linear regression analysis (Table 6) showed that there was no significant reduction in egg weight with increasing hours of the exposure to the heat (Fig. 2) or no heat (Fig. 3) on the 1st day following the

Table 2.a. Effect of heat or no heat treatments for 1, 2, 4, and 6 hrs. on the change (gm) in egg weight using SCWL hens 61 weeks of age.

hrs. in chamber and days following the stay in the chamber	No heat		Heat (35°C)		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
1hr.						
day 1	11	0.394	11	-1.42	22	-0.513
day 2	8	0.031	10	-1.012	18	-0.548
day 3	10	2.325	10	-0.714	20	0.806
2hrs.						
day 1	9	-0.422	8	-0.951	17	-0.671
day 2	10	0.345	11	1.077	21	0.729
day 3	10	-0.36	8	0.571	18	0.054
4hrs.						
day 1	22	0.15	20	0.23	42	0.188
day 2	16	0.467	22	0.465	38	0.466
day 3	22	-0.288	21	0.073	43	-0.111
6hrs.						
day 1	9	0.854	10	-1.374	19	-0.318
day 2	12	-0.08	10	-0.333	22	-0.195
day 3	9	1.407	9	-0.886	18	0.261
overall average of change	148	0.322	150	-0.218	298	0.05

(1) Change = The egg weight for a hen on the 1st (or 2nd or 3rd) day following the stay in chamber - Average of 3 eggs weights for the same hen before being in the chamber.

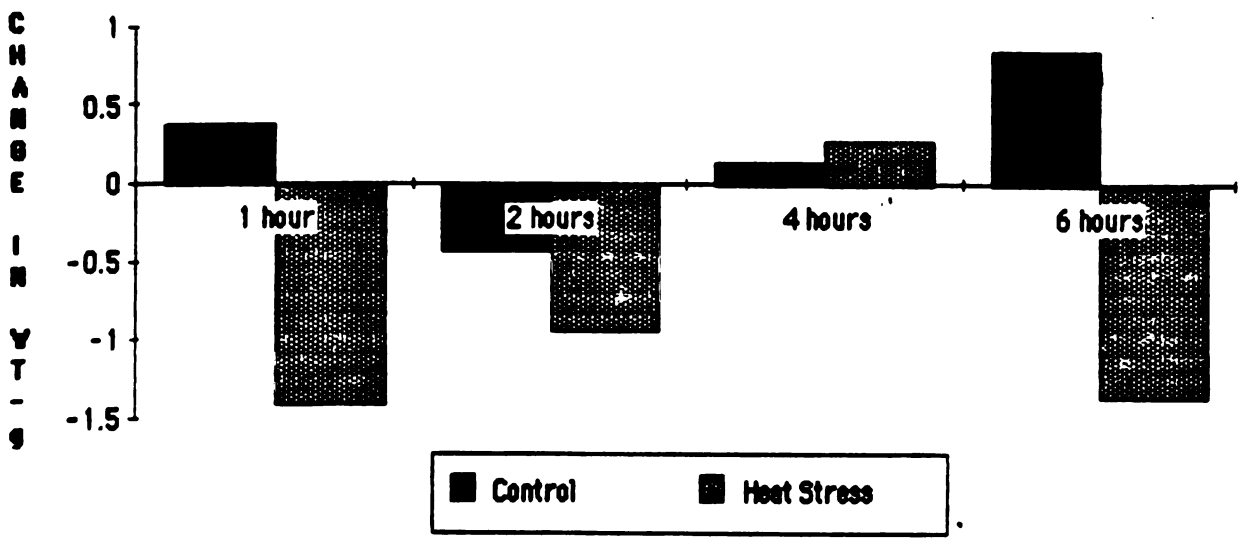

Figure 1: Effect of heat or no heat treatments for 1,2,4 and 6 hrs on the change (gm) in egg weight on the 1st day following the stay in the chamber

Table 2.b. Effect of heat or no heat treatments and the hours of exposure to these treatments on the change (gm) in egg weight

hrs. in chamber	No heat		Heat (35°C)		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
1hr.	29	0.96	31	-1.061	60	-0.084
2 hrs.	29	-0.136	27	0.326	56	0.087
4 hrs.	60	0.074	63	0.26	123	0.169
6 hrs.	30	0.646	29	-0.863	59	-0.096
overall AV. of change	148	0.322	150	-0.218	298	0.05

Table 2.c. Effect of heat or no heat treatments and the days following the stay in chamber on the change (gm) in egg weight.

days following the stay in the chamber	No heat		Heat (35°C)		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
day 1	51	0.226	49	-0.66	100	-0.208
day 2	46	0.222	53	0.163	99	0.19
day 3	51	0.509	48	-0.187	99	0.172
Overall AV. of change	148	0.322	150	-0.218	298	0.05

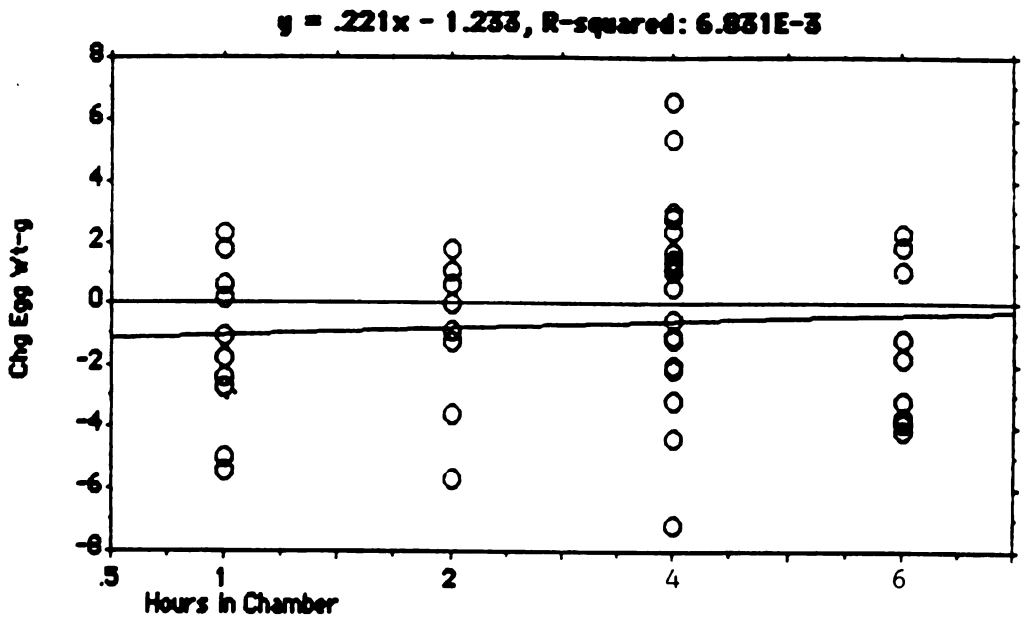

Figure 2: Effect of heat stress (35°C) for 1,2,4 and 6 hrs on the change (gm) in egg weight on the 1st day following the stay in the chamber

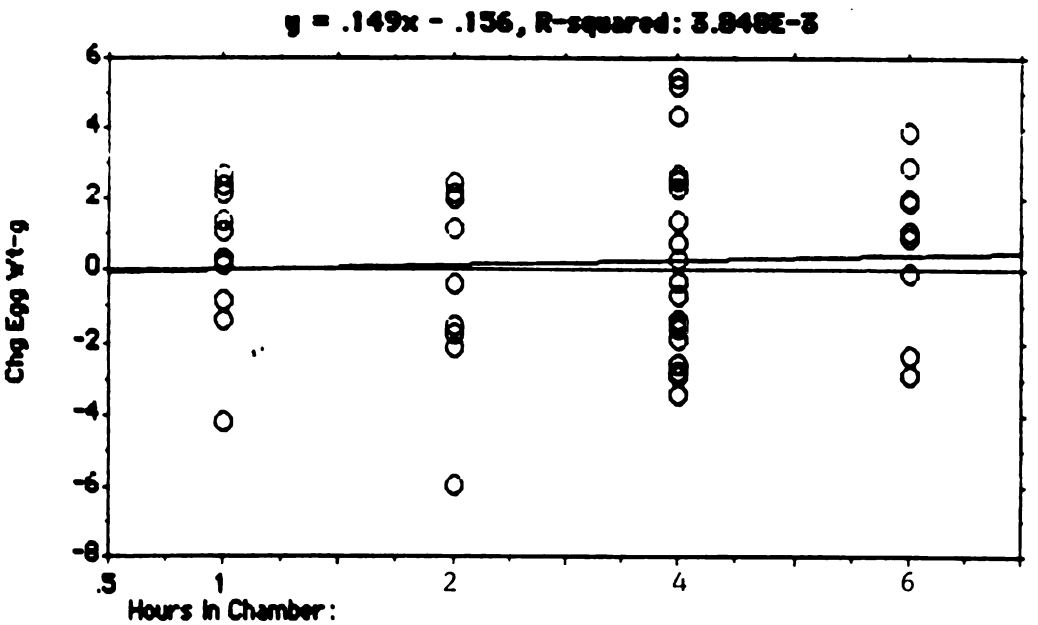

Figure 3: Effect of no heat treatments for 1,2,4 and 6 hrs on the change (gm) in egg weight on the 1st day following the stay in the chamber

stay in the chamber. Covariance analysis showed that there was no difference in the change in egg weight between heat or no heat treatments at the various times on the 1st day following the treatment.

4.1.2. The Effect on the Change (gm) in Shell Weight

Average change in shell weight is shown in Table 3.a, b, and c. There was a reduction of 0.131 gm in shell weight due to heat stress (35°C) regardless of the hours of the exposure to that heat or the days following the stay in the chamber. There was a reduction of only 0.014 gm in shell weight due to no heat treatments (Table 3.b). Four hours heat stress caused the maximum reduction in shell weight (-0.172 gm) and the reduction due to 4 hrs and 6 hrs heat stress was about the same (-0.172 vs. -0.146 gm) (Table 3.6). Most of the reduction in shell weight was on the 1st day following the stay in the chamber (regardless of the hours in the chamber or the temperature) (Table 3.c). There was a reduction in shell weight of 0.198 gm on the 1st day following the stay in the chamber vs. a reduction of 0.046 or an increase of 0.026 gm on the second and the third day, respectively. There was an obvious trend for shell weight to decline with increasing hours of heat (Fig. 4). Maximum reduction in shell weight was on the 1st day following heat stress for 6 hours (0.486 gm). And there was a reduction of only 0.141 gm due to the stay in the chamber for 6 hours but without heat stress (Table 3.a). Linear regression analysis

Table 3.a. Effect of heat or no heat treatments for 1, 2, 4, and 6 hrs. on the change (gm) in shell weight using SCWL hens 61 weeks of age.

hrs. in chamber and days following the stay in the chamber	No heat		Heat (35°C)		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
1hr.						
day 1	11	-0.105	11	-0.206	22	-0.156
day 2	8	-0.17	10	-0.016	18	-0.084
day 3	10	0.086	10	0.045	20	0.065
2 hrs.						
day 1	9	-0.044	8	-0.181	17	-0.109
day 2	10	-0.129	11	-0.065	21	-0.095
day 3	10	-0.144	8	-0.068	18	-0.11
4 hrs.						
day 1	22	-0.064	20	-0.351	42	-0.20
day 2	16	0.129	22	-0.197	38	-0.06
day 3	22	0.091	21	0.026	43	0.059
6 hrs.						
day 1	9	-0.141	10	-0.486	19	-0.323
day 2	12	0.018	10	0.101	22	0.055
day 3	9	-0.042	9	0.123	18	0.041
overall AV. of change	148	-0.014	150	-0.131	298	-0.073

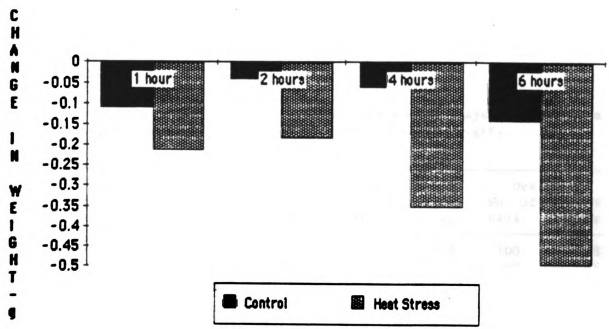

Figure 4: Effect of heat or no heat treatments for 1,2,4 and 6 hrs on the change (gm) in shell weight on the 1st day following the stay in the chamber

Table 3.b. Effect of heat or no heat treatments and the hours of exposure to these treatments on the change (gm) in shell weight.

hrs. in chamber	No heat		Heat (35°C)		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
1 hr.	29	-0.057	31	-0.064	60	-0.061
2 hrs.	29	-0.108	27	-0.10	56	-0.104
4 hrs.	60	0.045	63	-0.172	123	-0.066
6 hrs.	30	0.0016	29	-0.146	59	-0.071
overall AV. of change	148	-0.014	150	-0.131	298	-0.073

Table 3.c. Effect of heat or no heat treatments and the days following the stay in chamber on the change (gm) in shell weight.

days following the stay in the chamber	No heat		Heat (35°C)		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
day 1	51	-0.083	49	-0.318	100	-0.198
day 2	46	-0.0080	53	-0.079	99	-0.046
day 3	51	0.05	48	0.0014	99	0.026
overall AV. of change	148	-0.014	150	-0.131	298	-0.073

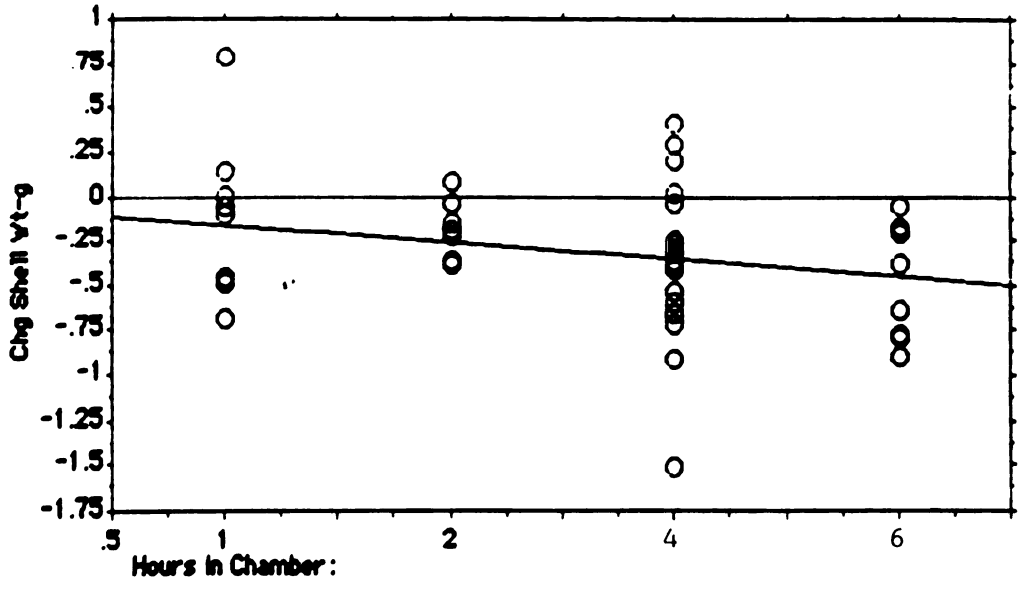

Figure 5: Effect of heat stress (35°C) for 1,2,4 and 6 hrs on the change (gm) in shell weight on the 1st day following the stay in the chamber

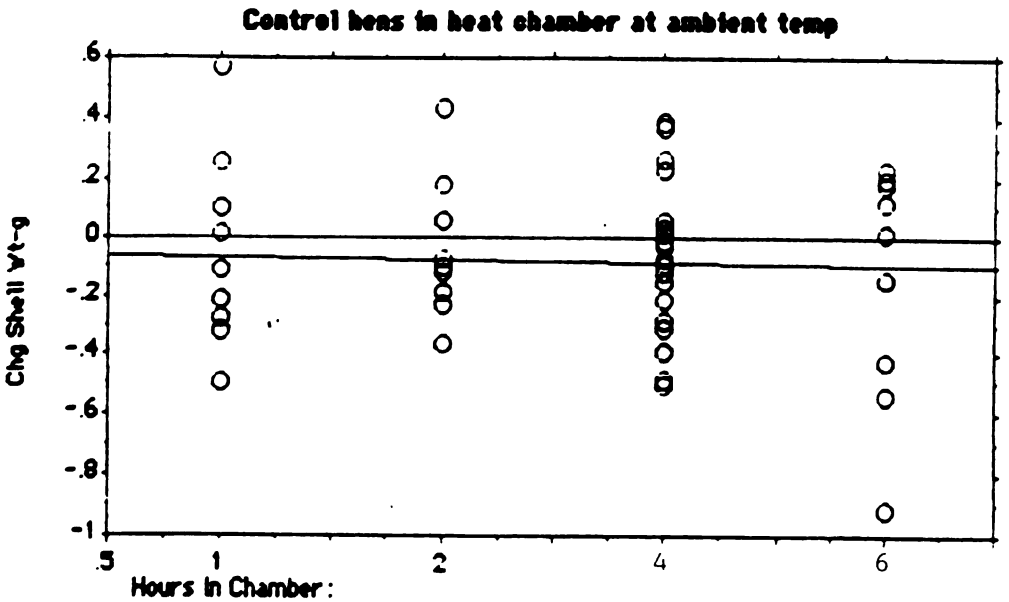

Figure 6: Effect of no heat treatments for 1,2,4 and 6 hrs on the change (gm) in shell weight on the 1st day following the stay in the chamber

(Table 6) showed that there was a borderline significant ($P = .05$) reduction in shell weight with increasing hours of heat (Fig. 5) but there was no effect due to the hours when there was no heat (Fig. 6 and Table 6). Covariance analysis showed that there was a highly significant ($P < .01$) reduction in shell weight between heat or no heat treatments at the various times on the 1st day following the treatment. Table 3.c showed that there was a reduction of 0.318 gm in shell weight due to heat stress compared to a reduction of only 0.083 gm due to no heat treatments on the 1st egg following the stay in the chamber. Covariance analysis also showed that there was no significant difference between the effect of heat or no heat treatments at the various times on the shell weight in case of the second or third egg following the stay in the chamber. This indicates that the effect of 35°C heat stress (for periods up to 6 hours) on the shell weight lasts for only one day.

4.1.3. The Effect on the Change in % Shell

The average value of change in % shell are shown in tables 4.a, b, and c. There was a reduction of 0.197% in % shell for hens held in the chamber with heat (35°C) for 1, 2, 4 or 6 hours but there was a reduction of only 0.121% when there was no heat (Table 4.a). The % shell results supported the previous results for shell weight in that most of the effect was on the 1st egg following the stay in the chamber (Table 4.c). Regardless of the hours in the

Table 4.a. Effect of heat or no heat treatments for 1, 2, 4, and 6 hrs. on the change (gm) in % shell using SCWL hens 61 weeks of age.

hrs. in chamber and days following the stay in the chamber	No heat		Heat (35°C)		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
1hr.						
day 1	11	-0.238	11	-0.023	22	-0.13
day 2	8	-0.296	10	0.112	18	-0.069
day 3	10	-0.229	10	0.223	20	-0.0030
2 hrs.						
day 1	9	0.0033	8	-0.16	17	-0.074
day 2	10	-0.26	11	-0.265	21	-0.262
day 3	10	-0.191	8	-0.196	18	-0.193
4 hrs.						
day 1	22	-0.135	20	-0.679	42	-0.394
day 2	16	-0.229	22	-0.46	38	-0.363
day 3	22	0.148	21	-0.0090	43	0.071
6hrs.						
day 1	9	-0.349	10	-0.604	19	-0.483
day 2	12	0.045	10	0.207	22	0.119
day 3	9	-0.027	9	0.116	18	0.044
overall AV. of change	148	-0.121	150	-0.197	298	-0.159

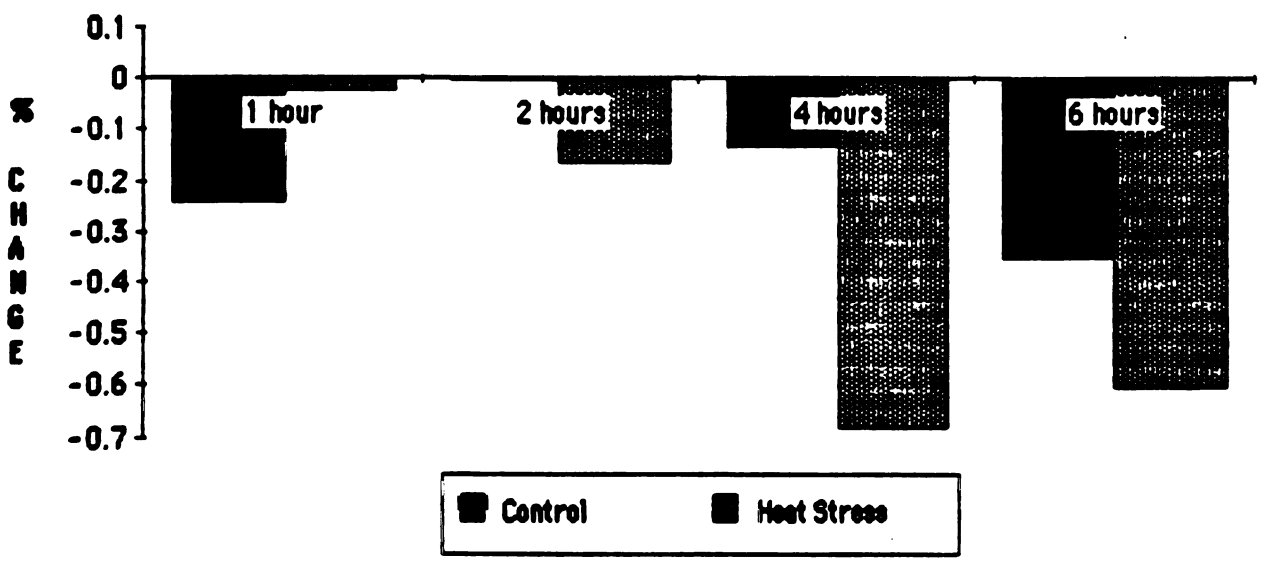

Figure 7: Effect of heat or no heat treatments for 1,2,4 and 6 hrs on the change in % shell on the 1st day following the stay in the chamber

Table 4.b. Effect of heat or no heat treatments and the hours of exposure to these treatments on the change in % shell.

hrs. in chamber	No heat		Heat (35°C)		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
1 hr.	29	-0.251	31	0.10	60	-0.07
2 hrs.	29	-0.154	27	-0.213	56	-0.183
4 hrs.	60	-0.056	63	-0.379	123	-0.222
6 hrs.	30	-0.095	29	-0.101	59	-0.098
overall AV. of change	148	-0.121	150	-0.197	298	-0.159

Table 4.c. Effect of heat or no heat treatments and the days following the stay in chamber on the change in % shell.

days following the stay in the chamber	No heat		Heat (35°C)		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
day 1	51	-0.17	49	-0.432	100	-0.299
day 2	46	-0.176	53	-0.185	99	-0.181
day 3	51	-0.023	48	0.031	99	0.0033
overall AV. of change	148	-0.121	150	-0.197	298	-0.159

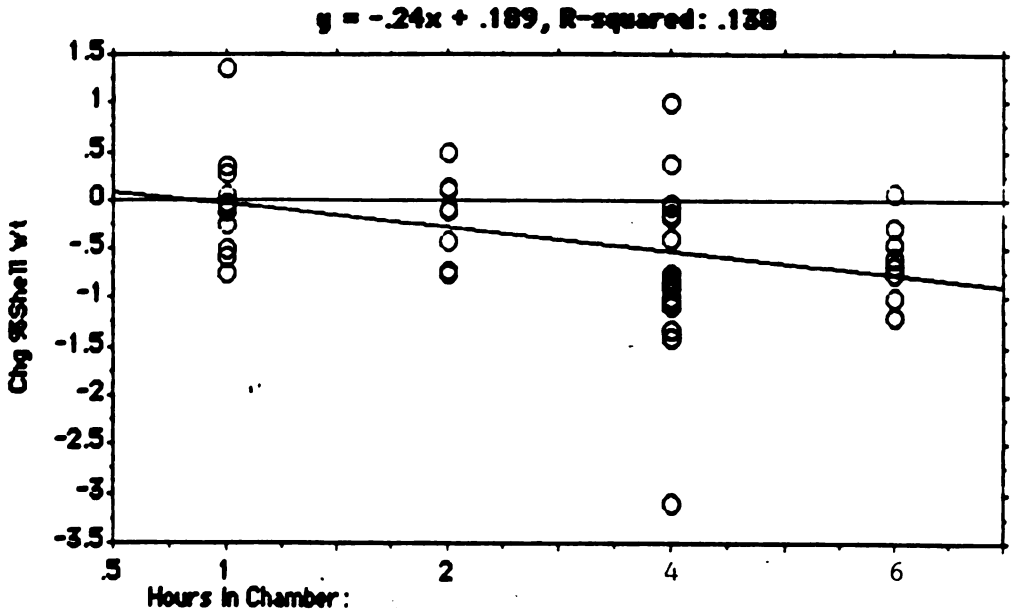

Figure 8: Effect of heat stress (35°C) for 1,2,4 and 6 hrs on the change in % shell on the 1st day following the stay in the chamber

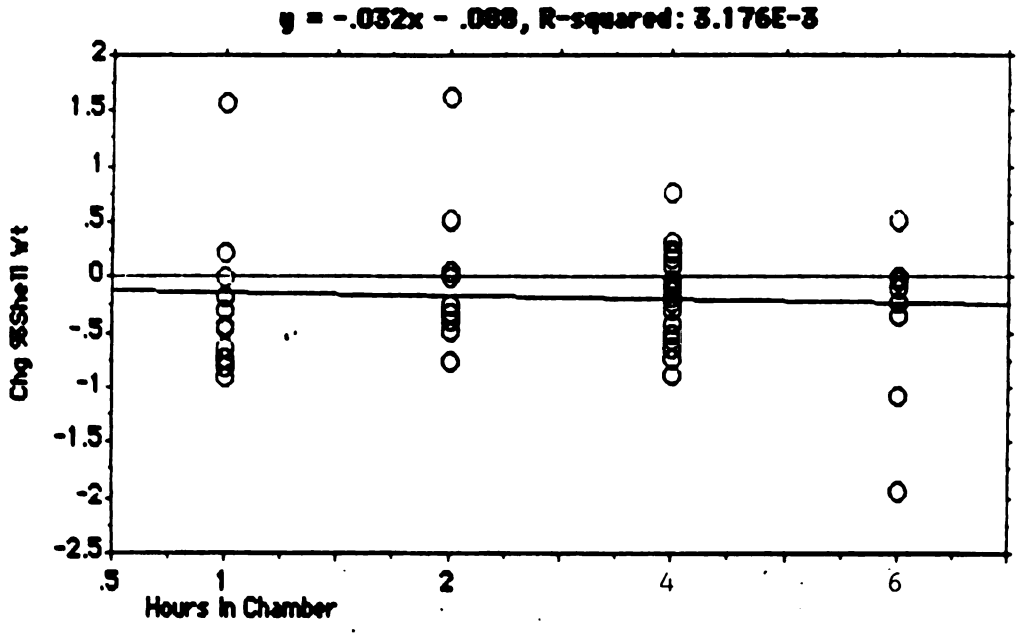

Figure 9: Effect of no heat treatments for 1,2,4 and 6 hrs on the change in % shell on the 1st day following the stay in the chamber

chamber, there was a reduction of 0.299% in shell percentage in case of the 1st egg following the stay in the chamber and a reduction of only 0.181% or almost no change in case of the second or the third egg following the stay in the chamber (Table 4.C). The trend for % shell to decline with increasing hours of stay in the chamber was not clear for the overall average of all the 3 days together (Table 4.b); but the effect of hours of the exposure to heat was definite on the 1st day (Fig. 7). There was a reduction of 0.679 and 0.604% shell for 4 and 6 hours, respectively, and there was a reduction of only 0.023 and 0.16% shell for 1 and 2 hours heat stress, respectively (Table 4.a). Linear regression analysis (Table 6) indicated that there was a highly significant reduction in % shell with increasing hours of heat (Fig. 8) but there was no effect in case of no heat (Fig. 9). Covariance analysis indicated that there was a significant reduction in % shell in case of heat when compared to no heat treatments at various times on the 1st day following the stay in the chamber. There was no difference between heat or no heat treatments on the % shell on the second or third day following the stay in the chamber.

4.1.4. The Effect on the Change in mg Ca/mm^2

The average change in mg Ca/mm^2 for the hens in the chamber at various times, whether heat stressed or not, is provided in table 5.a, b and c. Hens held in the heat

chamber at ambient temperature for 1, 2, 4, or 6 hours laid eggs averaging 0.0054 mg Ca/mm^2 less than the 3 eggs they laid before going into the chamber. However, the hens stressed at 35°C for 1, 2, 4 or 6 hours laid eggs that averaged 0.0069 less than the three each hen laid before being heat stressed (Table 5.a.). There was a significant decline in Ca/mm^2 starting with 4 hours of heat stress (Fig. 10). The results of the change in mg Ca/mm^2 supported the results of both shell weight and % shell in that the effect of heat stress was obvious only on the 1st day following the treatment (Fig. 11) and there was no effect from staying in the chamber without the heat treatments (Fig. 12). The decline in mg Ca/mm^2 of the shell was 0.022 and 0.021 mg Ca/mm^2 for 4 and 6 hours exposure to heat stress (35°C) vs. a reduction of 0.0065 and 0.0068 mg Ca/mm^2 for 1 and 2 hours exposure to the same temperature (Table 5.a).

The overall results of this experiment showed that: 1) four or six hours exposure to heat stress for only one day was enough to reduce the egg shell quality significantly for one day (1st day following the treatment), 2) there was no significant effect on egg weight due to this experiment, 3) there was a high correlation between egg shell quality parameters (shell weight, % shell and Ca/mm^2) (Table 5.d).

Table 5.a. Effect of heat or no heat treatments for 1, 2, 4, and 6 hrs. on the change in mg Ca/mm^2 of the egg shell using SCWL hens 61 weeks of age.

hrs. in chamber and days following the stay in the chamber	No heat		Heat (35°C)		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
1hr.						
day 1	11	-0.0074	11	-0.0065	22	-0.0070
day 2	8	-0.016	10	0.0008	18	-0.0069
day 3	10	-0.0034	10	0.0040	20	0.0003
2 hrs.						
day 1	9	-0.0004	8	-0.0068	17	-0.0034
day 2	10	-0.0083	11	-0.0075	21	-0.0079
day 3	10	-0.0076	8	-0.0076	18	-0.0076
4 hrs.						
day 1	22	-0.0043	20	-0.022	42	-0.013
day 2	16	-0.0070	22	-0.011	38	-0.0092
day 3	22	-0.0055	21	0.0025	43	-0.0027
6 hrs.						
day 1	9	-0.011	10	-0.021	19	-0.016
day 2	12	0.0017	10	0.0069	22	0.0040
day 3	9	0.0010	9	0.0089	18	0.0094
overall AV. of change	148	-0.0054	150	-0.0069	298	-0.0062

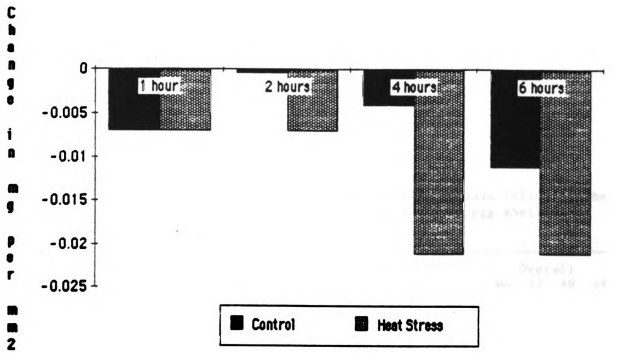

Figure 10: Effect of heat or no heat treatments for 1,2,4 and 6 hrs on the change in mg Ca/mm^2 on the 1st day following the stay in the chamber

Table 5.b. Effect of heat (35°C) or no heat treatments and the hours of exposure to these treatments on the change in mg Ca/mm^2 of egg shell.

hrs. in chamber	No heat		Heat (35°C)		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
1 hr.	29	-0.0085	31	-0.0008	60	-0.0045
2 hrs.	29	-0.0056	27	-0.0073	56	-0.0064
4 hrs.	60	-0.0055	63	-0.011	123	-0.0082
6 hrs.	30	-0.0022	29	-0.0047	59	-0.0034
overall AV. of change	148	-0.0054	150	-0.0069	298	-0.0062

Table 5.c. Effect of heat or no heat treatments and the days following the stay in chamber on the change in mg Ca/mm^2 of egg shell

days following the stay in the chamber	No heat		Heat (35°C)		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
day 1	51	-0.0054	49	-0.016	100	-0.011
day 2	46	-0.0067	53	-0.0046	99	-0.0056
day 3	51	-0.0044	48	-0.0002	99	-0.0023
overall AV. of change	148	-0.0054	150	-0.0069	298	-0.0062

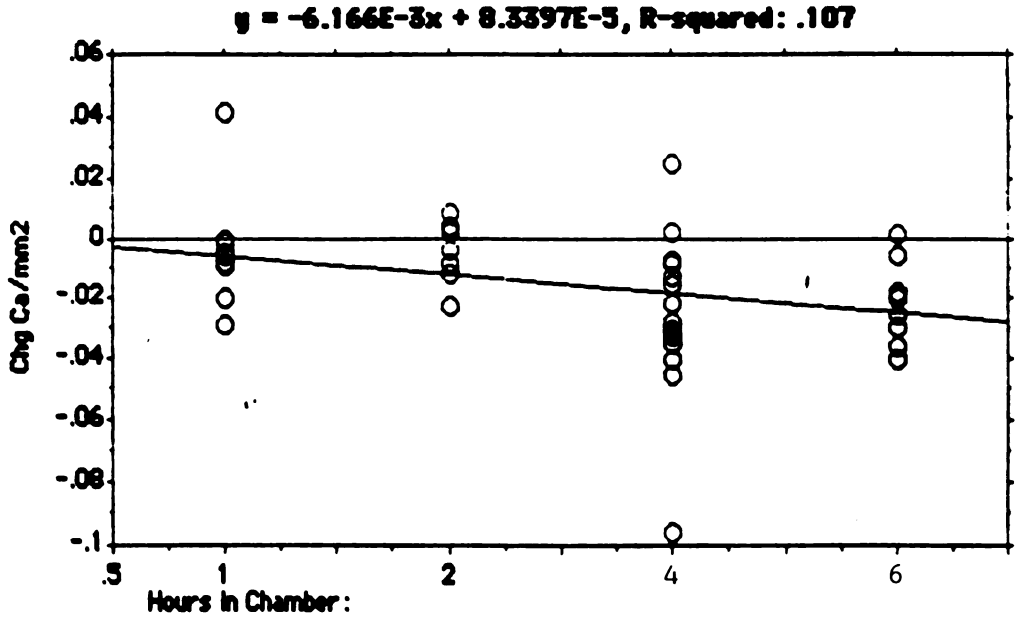

Figure 11: Effect of heat stress (35°C) for 1,2,4 and 6 hrs on the change in mg Ca/mm^2 on the 1st day following the stay in the chamber

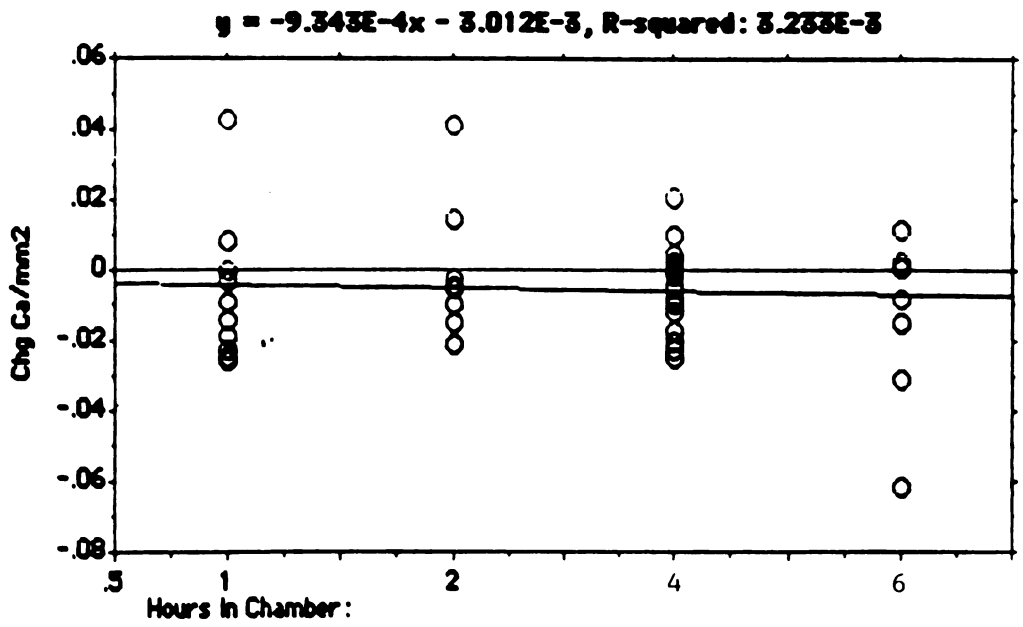

Figure 12: Effect of no heat treatments for 1,2,4 and 6 hrs on the change in mg Ca/mm^2 on the 1st day following the stay in the chamber

Table 5.d. The correlation between egg shell quality parameters used in experiment 1.

	Chg. % shell wt.	Chg. Ca/mm^2	Chg. shell wt.
Chg % shell Wt.	1		
Chg Ca/mm^2	.949	1	
Chg Shell Wt.	.747	.839	1

4.2. Experiment 2. Effect of heat (35°C) or no heat treatments for 6 hrs at the time between 0800 and 1600 hr (without active shell deposition) on the change in egg quality on the 1st day following the stay in the chamber using SCWL hens 46 weeks of age

4.2.1. The Effect on the Change (gm) in Egg Weight

The average change in egg weight is shown in Table 7. There was a reduction of 2.043 gm in egg weight in case of heat stress and 0.866 gm in case of no heat. The reduction in egg weight due to heat was not significant.

4.2.2. The Effect on the Change (gm) in Shell Weight

There was almost no change (+0.03) in shell weight (Table 7) due to no heat for 6 hours, but the shell weight of the 1st egg following the heat stress (35°C) was 0.283 gm less than average value of the the 3 eggs laid before being heat stressed. The reduction in shell weight due to heat stress was significant ($P < .05$).

Table 6. Regression analysis of hours[1] in heat chamber and the change in egg parameters for heat or no heat treatments on the first egg following the treatments in experiment 1.

Egg parameters	Regression equation	Correlation coefficient	Significance level (p)
1. Egg weight			
a. with heat	y = 0.221x - 1.233	0.083	0.572
b. without heat	y = 0.149x - 0.156	0.062	0.665
2. Shell weight			
a. with heat	y = -0.097x - 0.066	0.269	0.062
b. without heat	y = -0.007x - 0.065	0.026	0.857
3. % Shell			
a. with heat	y = -0.24x + 0.189	0.371	0.009
b. without heat	y = -0.032x - 0.088	0.056	0.695
4. Ca/mm^2			
a. with heat	y = -0.006x + 0.00008	0.327	0.022
b. without heat	y = -0.0009x - 0.003	0.057	0.692

[1] Hours = 1, 2, 4, and 6 hours.

Table 7. Effect of heat (35°C) or no heat treatments for 6 hrs. at the time between 0800 and 1600 hr. (without egg shell deposition) on the change in egg quality on the first day following the stay in the chamber using SCWL hens 46 weeks of age.

Egg quality Parameters	No heat (9 hens) Average of change	Heat (35°C) (10 hens) Average of change
1. egg weight (gm)	-0.866[a]	-2.043[a]
2. shell weight (gm)	0.03[a]	-0.283[b]
3. % shell	0.184[a]	-0.20[a]
4. mg Ca/mm^2	0.0051[a]	-0.0076[a]

a, b values followed by different letters in the same row are significantly different ($P \leq .05$).

4.2.3. The Effect on the Change in % Shell

The values for the change in % shell are shown in Table 7. The reduction in % shell (-0.20%) due to heat stress was not significant when compared to the change in % shell in case of no heat (+0.184%).

4.2.4. The Effect on the Change in Ca/mm^2

The change in mg Ca/mm^2 of the egg shell (Table 7) was not significant between heat (-0.0076 mg Ca/mm^2) and no heat (+ 0.0051 mg Ca/mm^2). There was a high correlation between egg shell quality parameters (Table 8).

Table 8 . The correlation between egg shell quality parameters in experiment 2.

	Chg. % shell wt.	Chg. Ca/mm^2	Chg. Shell wt.
Chg. % shell Wt.	1		
Chg. Ca/mm^2	.969	1	
Chg. Shell Wt.(g)	.982	.98	1

4.2.5. The Effect on the Feed Intake

The amount of feed intake (Table 9) 24 hours following the heat stress (35°C) for 6 hours (74 gm) did not differ significantly than the amount of feed intake 24 hours following the stay in the chamber without heat (85 gm).

4.2.6. The Effect on the Change in Body Temperature

There was a significant increase in body temperature due to heat stress (+0.79°C) when compared to no heat treatment (+0.11°C) (Table 9).

Table 9. The relationship between heat (35°C) or no heat treatment for 6 hours at the time between 0800 and 1600 hr (without egg shell formation) and both of feed consumption and the change in body temperature due to these treatments.

	Heat (10 hens.)	No Heat (9 hens.)
Feed Intake (gm) 18 hours following the stay in chamber	74[a]	85[a]
Change in body temp. (°C) due to the stay in the chamber	+0.79[a]	+0.11[b]

a, b values in the same row differently superscripted are significantly different ($P \leq .05$).

The overall results of this experiment showed that the effect of heat stress (35°C) for 6 hours at day time, when shell deposition was not occuring, in case of young hens (46 weeks of age) was significantly effecting only the shell weight of many factors analyzed. This effect may be due to the significant increase in body temperature rather than the difference in feed intake at that time.

4.3. Experiment 3. Effect of heat (35°C) or no heat for 6 hrs at the time between 0800 and 1600 hr (without shell deposition) and during egg shell formation (between 1600 and 2300 hr) on the change in egg quality for the 1st egg following the stay in the chamber using SCWL hens 46 weeks of age

4.3.1. The Effect on the Change (gm) in Egg Weight

There was a reduction of 1.408 gm in egg weight for the treatments when there was no shell formation (No SF) due to stay in the chamber for 6 hours and there was a reduction of only 0.255 gm for the treatments during egg shell formation (SF). Regardless of heat or no heat treatments, the difference between those values was significant. Regardless of the time of the stay in the chamber, heat treatments caused a reduction of 1.197 gm in egg weight vs. a reduction of only 0.27 gm for no heat treatments (Table 10). The difference between the last two values was not significant. LSD analysis (Table A.1.a Appendix) showed that there was no significant difference between the change in egg weight due to heat (-1.899 gm) and no heat (-0.863 gm) at (No SF) time. Also there was no difference between the values of the change in egg weight due to the temperature during egg shell formation time (Table 10.A).

4.3.2. The Effect on the Change (gm) in Shell Weight

Table 10.B shows that the egg shell for the first egg laid after being in the chamber decreased 0.683 gm due to the heat stress but it decreased only 0.317 gm due to the stay in the chamber for 6 hours without heat. The difference between those two values was significant. There

Table 10. Effect of heat (35°C) or no heat for 6 hrs. at (No SF) time vs. (SF) time on
A. The change in egg weight (gm) on the 1st day following the stay in the chamber using SCWL hens 46 weeks of age.

treatments	(No SF) time		(SF) time		overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
No heat	9	-0.863^{a}	8	0.397^{a}	17	-0.27^{a}
Heat	10	-1.899^{a}	13	-0.656^{a}	23	-1.197^{a}
Overall AV. of change	19	-1.408^{a}	21	-0.255^{b}	40	-0.803

B. The change in shell weight (gm)

No heat	9	0.03^{a}	8	-0.707^{b}	17	-0.317^{a}
Heat	10	-0.283^{a}	13	-0.99^{b}	23	-0.683^{b}
Overall AV. of change	19	-0.135^{a}	21	-0.882^{b}	40	-0.527

C. The change in % shell

No heat	9	0.181^{a}	8	-1.259^{b}	17	-0.496^{a}
Heat	10	-0.142^{a}	13	-1.577^{b}	23	-0.953^{a}
Overall AV. of change	19	0.011^{a}	21	-1.456^{b}	40	-0.759

D. The change in mg Ca/mm^2

No heat	9	0.0051^{a}	8	-0.04^{b}	17	-0.016^{a}
Heat	10	-0.0076^{a}	13	-0.051^{b}	23	-0.032^{a}
Overall AV. of change	19	-0.0016^{a}	21	-0.047^{b}	40	-0.025

a, b values followed by different letters in the same column or row are significantly different ($P \leq .05$). Values followed by different letters in the overall average column or row are significantly different ($P \leq .05$).

was a highly significant reduction in shell weight (-0.882 gm) due to the stay in the chamber during egg shell formation when compared to the stay in the chamber at (No SF) time (-0.135 gm). LSD analysis (Table A.1.b) showed there was no significant differences between the values for heat and no heat at (SF) or at (No SF) time. But there was a significant reduction in shell weight due to the stay in the chamber during egg shell formation regardless of the temperature.

4.3.3. The Effect on the Change in % Shell

The values for % shell were -0.953% for heat and -0.496% for no heat. The difference between these values was not significant. There was a highly significant reduction of 1.456% shell for (SF) time treatment when compared to an increase of 0.011% shell for (No SF) time treatment (Table 10.c). LSD analysis (Table A.1.c.) indicated that the values of % shell (-1.259 and -1.577%) obtained from the treatments during egg shell formation for no heat and for heat treatments, respectively, were significantly different from those (0.181 and -0.142%) obtained from the stay in the chamber during (No SF) time. And there was no difference between the values due to the temperature during (No SF) time (0.181 vs. -0.142%) or at the time of egg shell deposition (-1.259 vs -1.577%) (Table 10.c).

4.3.4. The Effect on the Change in mg Ca/mm^2

The change in mg Ca/mm^2 is presented in Table 10.D. The reduction (-0.032 mg Ca/mm^2) in mg Ca/mm^2 of the egg shell due to heat treatments was not significantly different from that (-0.016 mg Ca/mm^2) for no heat treatments. There was a highly significant reduction in % shell (-0.047%) due to the treatments during egg shell formation when compared to that (-0.0016%) during (No SF) time. LSD analysis (Table A.1.d) showed that there was a significant reduction in Ca/mm^2 for the values obtained from hens in the chamber for 6 hours during egg shell formation (with or without heat) when compared to those values obtained from the (No SF) time treatments (Table 10.D). The high correlation between egg shell quality parameters was supported also by this experiment (Table 11).

Table 11. The correlation between egg shell quality parameters used in experiment 3.

	Chg. % shell wt.	Chg. Ca/mm^2	Chg. Shell wt.
Chg. % shell Wt.	1		
Chg. Ca/mm^2	.995	1	
Chg. Shell Wt.(g)	.956	.975	1

4.3.5. The Effect on the Feed Intake

There was a significant reduction ($p = 0.03$) in feed intake due to both of heat stress (69.1 vs. 79.6 gm) and (SF) time treatments (68.5 vs. 79.1 gm) when compared to no heat or (No SF) time treatments (Table 12). LSD analysis (Table A.1.e) showed that there was no significant reduction in feed intake due to heat at (No SF) time (74 vs 85 gm) but there was a significant reduction in feed intake due to (SF) time treatments (68.5 vs. 79.1 gm) when compared to no heat and (No SF) time treatments.

4.3.6. The Effect on the Change in Body Temperature

Heat stress caused a highly significant increase (0.76°C) in body temperature when compared to no heat treatments (-0.15°C) Table 12. During egg shell formation, the body temperature increased 0.28°C after the stay in the chamber for 6 hours. The increase in body temperature after (No SF) time treatments was 0.47°C. There was no significant difference between these values (0.28 and 0.47°C). LSD analysis (Table A.1.f) showed that the change in body temperature at (No SF) or (SF) time due to heat was significantly different than that for no heat treatments (Table 12). There was no significant difference in the change in body temperature between (No SF) time and (SF) time treatments when there was heat (35°C) (0.79 vs. 0.73°C) or when there was no heat (-0.45°C vs. 0.11°C) during the stay in the chamber.

Table 12. Effect of heat (35°C) or no heat during egg shell formation and during (No SF) time for 6 hours on both of feed consumption and the change in body temperature.

	Feed Intake (gm) 18 hours following the stay in the chamber			The change (°C) in body temperature		
	(No SF) time	(SF) time	overall AV.	(No SF) time	(SF) time	overall AV.
No heat	85^{a} (9 hens)	73^{ab} (8 hens)	79.6^{a} (17 hens)	0.11^{a} (9 hens)	-0.45^{a} (8 hens)	-0.15^{a} (17 hens)
Heat	74^{ab} (10 hens)	66^{bc} (13 hens)	69.1^{b} (23 hens)	0.79^{b} (10 hens)	0.73^{b} (13 hens)	0.76^{b} (23 hens)
Overall AV. of change	79.1^{a} (19 hens)	68.5^{b} (21 hens)	73.6 (40 hens)	0.47^{a} (19 hens)	0.28^{a} (21 hens)	0.37 (40 hens)

a,b,c values followed by different letters within the same parameters (Feed intake or body temperature) are significantly different ($P \leq .05$). Values followed by different letters in the overall average row or column within the same parameters are sigificantly different ($P \leq .05$).

4.4. Experiment 4. Effect of heat (35°C) or no heat for 4 or 6 hours (during egg shell formation) on egg quality using SCWL hens 46 weeks of age

4.4.1. The Effect on the Change (gm) in Egg Weight

Heat stress did not reduce egg weight significantly, but 6 hours of stay in the chamber, regardless of the temperature, caused significantly less egg weight than did the 4 hours stay (Table 13.A). The average values of the

change were 0.081 and 0.857 gm for heat and no heat, respectively. The reduction in egg weight due to 6 hours was 0.255 gm and there was an increase in egg weight of 1.187 gm due to 4 hours stay in the chamber. The value of the change in egg weight due to the stay in the chamber for 6 hours was less than the value obtained after 4 hours in the heat chamber. These values averages 0.397 and -0.656 gm for no heat and heat treatments respectively, for 6 hours and 1.224 and 1.146 gm for no heat and heat treatments respectively for 4 hours.

4.4.2. The Effect on the Change (gm) in Shell Weight

The reduction in shell weight due to heat stress (-0.807 gm) or due to 6 hours stay in the chamber, regardless of the temperature (-0.805 gm), was not significant when compared to no heat (-0.629 gm) or 4 hours stay in the chamber (-0.641 gm) (Table 13.B).

4.4.3. The Effect on the Change in % Shell

There was no significant difference between the values of the change in % shell due to the hours of the stay in the chamber or due to the temperature (Table 13.C).

4.4.4.. The Effect on the Change (mg) Ca/mm^2 of the Shell

The results of the Ca/mm^2 were similar to those for shell weight and % shell in that there was no differences

Table 13. Effect of heat (35°C) or no heat for 4 and 6 hrs. (during egg shell formation) on
A. The change in egg weight (gm) on the 1st day following the stay in the chamber using SCWL hens 46 weeks of age.

treatments	4 hours		6 hours		overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
No heat	10	1.224	8	0.397	18	0.857
Heat	9	1.146	13	-0.656	22	0.081
Overall AV. of change	19	1.187	21	-0.255	40	0.43
B. The change in shell weight (gm)						
No heat	10	-0.566	8	-0.707	18	-0.629
Heat	9	-0.723	13	-0.865	22	-0.807
Overall AV. of change	19	-0.641	21	-0.805	40	-0.727
C. The change in % shell						
No heat	10	-1.092	8	-1.128	18	-1.108
Heat	9	-1.282	13	-1.577	22	-1.456
Overall AV. of change	19	-1.182	21	-1.406	40	-1.30
D. The change in mg Ca/mm^2						
No heat	10	-0.034	8	-0.04	18	-0.037
Heat	9	-0.04	13	-0.051	22	-0.047
Overall AV. of change	19	-0.037	21	-0.047	40	-0.042

N.B: The significant difference in the change in egg weight between 6 hours and 4 hours stay in the chamber was due to significant difference only between (no heat 4 hours) and (heat 6 hours) treatments.

between the values due to the temperature or the time of the stay in the heat chamber (Table 13.D).

4.4.5. The Effect on the Feed Intake

Table 14 shows that there was a reduction in feed intake (24 hours following the stay in the chamber) due to hours of the stay in the chamber (68.5 gm for 6 hours vs. 72.89 gm for 4 hours) and due to the temperature (69.0 gm for heat vs. 72.6 gm for no heat). The difference between these values was not significant.

4.4.6. The Effect on the Change in Body Temperature

The difference between the change in body temperature for 4 hours and 6 hours stay in the chamber, regardless of the temperature, was not significant (0.27°C for 4 hours vs. 0.28°C for 6 hours). There was a highly significant increase in body temperature (0.72°C) due to heat stress when compared to no heat (-0.26°C) (Table 14).

There was a reduction in body temperature of 0.11 and 0.45°C for 4 hours and 6 hours stay in the chamber without heat. The increase in body temperature due to heat stress was about the same for both 4 hours and 6 hours exposure to 35°C heat stress (0.70 vs. 0.73°C).

The overall results of this experiment indicates that both 4 and 6 hours stay in the chamber during egg shell formation have about the same effect on egg shell quality. The reduction in shell quality due to stay in the chamber

with heat or without heat for same hours was about the same. This study also showed that both of 4 and 6 hours to heat stress caused a reduction in shell quality when compared to the three eggs each hen laid before being heat stressed. This reduction in shell quality appears to be due to the absence of feed during the stay in the chamber (during egg shell formation).

Table 14. Effect of heat (35°C) or no heat for 4 and 6 hours (during egg shell formation) on the feed intake and the change in body temperature of SCWL hens 46 weeks of age.

	Feed Intake (gm)			The change (°C) in body temperature		
	20 hrs after chamber	18 hours after chamber				
	4 hours	6 hours	overall AV.	4 hours	6 hours	overall AV.
No heat	72 (10 hens)	73 (8 hens)	72.6 (18 hens)	-0.11[a] (10 hens)	-0.45[a] (8 hens)	-0.26[a] (18 hens)
Heat	73.9 (9 hens)	65.6 (13 hens)	69.0 (22 hens)	0.70[b] (9 hens)	0.73[b] (13 hens)	0.72[b] (22 hens)
Overall AV. of change	72.89 (19 hens)	68.5 (21 hens)	70.6 (40 hens)	0.27[a] (19 hens)	0.28[a] (21 hens)	0.28 (40 hens)

a, b values followed by different letters are significantly different ($P \leq .05$). Values followed by different letters in the overall average row or column are significantly different ($P \leq .05$).

N. B. 1) The overall average of feed intake/hen/day in hens room before hens go to the heat chamber was 107 gm.

2) There was no significant difference between the values of feed intake due to temperature and hours of stay in the chamber.

4.5. Experiment 5. Effect of feed and/or water under heat (35°C) or no heat for 4 hours during egg shell formation on egg quality using SCWL hens 46 weeks of age

4.5.1. The Effect on the Change (gm) in Egg Weight

The average values of the change in egg weight are shown in Table 15.1. Almost all the values increased after the stay in the chamber except for no water and no feed treatment under heat stress (-0.123 gm). There was no significant difference in the change in egg weight due to water (+ water or no water), feed (+feed or no feed) or due to the temperature (+ heat or no heat).

4.5.2. The Effect on the Change (gm) in Shell Weight

Egg shell weight for the first egg laid following the stay in the chamber with heat (35°C) for 4 hours was 0.709 gm less than the three each hen laid before being heat stressed (Table 15.2.c). There was a reduction of 0.484 gm in shell weight of hens kept in the chamber with no heat (Table 15.2.c). The difference between these values (-0.709 and -0.484 gm) was significant. The feed did not have a significant effect on egg shell weight (Table 15.2.d). There was a reduction of 0.602 gm in shell weight when the feed was not available during the stay in the chamber and a reduction of 0.568 gm when the feed was available. The difference between the values of the change in shell weight

Table 15. Effect of providing feed and/or water during heat stress (35°C) or no heat for 4 hrs. at the time egg shell formation is occurring on:
1. The change (gm) in egg weight using SCWL hens 46 weeks of age

Treatments	No feed				+ Feed					
	No heat		Heat		No heat		Heat		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	Av. of change	No of hens	AV. of change
No water	9	0.976	6	-0.123	9	0.744	7	1.206	31	0.748
+ water	10	1.224	9	1.146	7	0.099	7	1.170	33	0.952
Overall AV. of change	19	1.106	15	0.638	16	0.462	14	1.188	64	0.853

Table 15.2.a. The change (gm) in shell weight
(The effect of the temperature, feed and water)

No water	9	-0.472	6	-0.673	9	-0.649	7	-0.981	31	-0.677
+ water	10	-0.566	9	-0.723	7	-0.17	7	-0.447	33	-0.50
Overall AV. of change	19	-0.522	15	-0.703	16	-0.439	14	-0.714	64	-0.586

N.B: Analysis of variance of the change in egg weight and shell weight is shown in Tables A.2.a and A.2.b (Appendix).

Table 15.2.b. The change (gm) in shell weight
(The effect of water and feed)

treatments	No feed		+ Feed		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
No water	15	-0.553	16	-0.794	31	-0.677
+ Water	19	-0.641	14	-0.309	33	-0.50
Overall AV. of change	34	-0.602	30	-0.568	64	-0.586

Table 15.2.c. The change (gm) in shell weight
(The effect of the temperature and water)

treatments	No heat		+ Heat		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
No water	18	-0.561	13	-0.839	31	-0.677
+ Water	17	-0.403	16	-0.602	33	-0.50
Overall AV. of change	35	-0.484	29	-0.709	64	-0.586

Table 15. 2.d. The change (gm) in shell weight
(The effect of the temperature and feed)

treatments	No heat		Heat		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
No feed	19	-0.522	15	-0.703	34	-0.602
+ Feed	16	-0.439	14	-0.714	30	-0.568
Overall AV. of change	35	-0.484	29	-0.709	64	-0.586

when the water was not available during the stay in the chamber (-0.677) and when the water was available (-0.50) was significant. When the water was available, there was a significant difference in the change in shell weight between (No Feed) and (+Feed) treatments (-0.64 vs. -0.309 gm) but when the water was not available it did not make any difference in the change in shell weight whether the food was available or not (-0.794 vs. -0.553 gm) (Table 15.2.b). The minimum reduction in shell weight (-0.17 gm) was when both of the water and feed were available and there was no heat stress. The maximum reduction in shell weight (-0.981 gm) was when the water was not available during heat stress (35°C) and the feed was available.

4.5.3. The Effect on the Change in % Shell

There was a significant reduction due to heat treatments when compared to no heat treatments (-1.197 vs. -0.881% shell) (Table 15.3.d). Regardless of the effect of water or the temperature on % shell, the feed did not have a significant effect on the change in % shell (-1.088% for no feed vs. -0.952% shell for feed treatments) (Table 15.3.d). When the water was not available during the stay in the chamber for 4 hours, there was a significant reduction of 1.14 % shell when compared to the change in % shell for hens provided with water (-0.915%). As detected for the change in shell weights, when water was available there was a significant difference between the values of the change in %

Table 15.3.a. The change in % shell due to the effect of the temperature, feed and water

Treatments	No feed				+ Feed				Overall	
	No heat		Heat		No heat		Heat			
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	Av. of change	No of hens	AV. of change
No water	9	-0.908	6	-1.058	9	-1.069	7	-1.599	31	-1.14
+ water	10	-1.092	9	-1.282	7	-0.303	7	-0.803	33	-0.915
Overall AV. of change	19	-1.005	15	-1.193	16	-0.734	14	-1.201	64	-1.024

N.B: Analysis of variance of the change in % shell is shown in Table A.2.c (Appendix).

Table 15.3.b. The change in % shell due to the effect of water and feed

treatments	No feed		+ Feed		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
No water	15	-0.968	16	-1.301	31	-1.14
+ Water	19	-1.182	14	-0.553	33	-0.915
Overall AV. of change	34	-1.088	30	-0.952	64	-1.024

Table 15.3.c. The change in % shell due to the effect of the temperature and water

treatments	No heat		Heat		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
No water	18	-0.988	13	-1.349	31	-1.14
+ Water	17	-0.767	16	-1.072	33	-0.915
Overall AV. of change	35	-0.881	29	-1.197	64	-1.024

Table 15. 3.d. The change in % shell due to the effect of the temperature and feed)

treatment	No heat		Heat		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
No feed	19	-1.005	15	-1.193	34	-1.088
+ Feed	16	-0.734	14	-1.201	30	-0.952
Overall AV. of change	35	-0.881	29	-1.197	64	-1.024

shell when feed was present (-0.553) as compared to when feed was not provided (-1.182) (Table 15.3.b). The minimum reduction of % shell (-0.303%) was when both feed and water were allowed when there was no heat during the hen's stay in the chamber (Table 15.3.a). The maximum reduction (-1.599% shell) was when there was no water but feed was provided during heat stress (Table 15.3.a).

4.5.4. The Effect on the Change in mg Ca/mm^2 of the Shell

The results of mg Ca/mm^2 supported the results of the other shell quality parameters. Heat treatments reduced the amount of Ca/mm^2 of the shell significantly when compared to no heat treatments (-0.038 vs. -0.027 mg Ca/mm^2) (Table 15.4.c). The presence of feed, regardless of allowable water or the temperature of the chamber did not have an effect on the change in mg Ca/mm^2 (-0.034 vs. -0.030 mg Ca/mm^2, Table 15.4.b). Absence of water during the hen's stay in the chamber caused a significant reduction in the amount of Ca/mm^2 of the shell (-0.036 mg) when compared to when there was water (-0.029 mg) (Table 15.4.b). In the absence of water during the stay in the chamber, providing feed or not did not make significant difference on the mg Ca/mm^2 (-0.042 vs. -0.03 mg Ca/mm^2) (Table 15.4.b).

The high correlation between egg shell parameters also was confirmed in this experiment (Table 16).

Table 15.4.a The change in mg Ca/mm^2 due to the effect of the temperature, feed and water

Treatments	No feed				+ Feed					
	No heat		Heat		No heat		Heat		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	Av. of change	No of hens	AV. of change
No water	9	-0.027	6	-0.035	9	-0.034	7	-0.052	31	-0.036
+ water	10	-0.034	9	-0.04	7	-0.009	7	-0.025	33	-0.029
Overall AV. of change	19	-0.031	15	-0.038	16	-0.023	14	-0.039	64	-0.032

N.B: Analysis of variance of the change in mg Ca/mm^2 of shell is shown in Table A.2.d (Appendix).

Table 15.4.b. The change in mg Ca/mm^2 due to the effect of the water and feed.

treatments	No feed		+ Feed		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
No water	15	-0.03	16	-0.042	31	-0.036
+ Water	19	-0.037	14	-0.017	33	-0.029
Overall AV. of change	34	-0.034	30	-0.03	64	-0.032

Table 15.4.c. The change in mg Ca/mm^2 due to the effect of the temperature and water.

treatments	No Heat		Heat		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
No water	18	-0.03	13	-0.044	31	-0.036
+ Water	17	-0.024	16	-0.034	33	-0.029
Overall AV. of change	35	-0.027	29	-0.038	64	-0.032

Table 15.4.d. The change in mg Ca/mm^2 due to the effect of the temperature and feed.

treatments	No heat		Heat		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
No Feed	19	-0.031	15	-0.038	34	-0.034
+ Feed	16	-0.023	14	-0.039	30	-0.03
Overall AV. of change	35	-0.027	29	-0.038	64	-0.032

Table 16. The correlation between egg shell quality parameters used in experiment 5.

	Chg. % shell wt.	Chg. Ca/mm^2	Chg. shell wt.
Chg % shell Wt.	1		
Chg Ca/mm^2	.989	1	
Chg Shell Wt.(gm)	.906	.954	1

4.5.5. The Effect on Shell-less Eggs (SL eggs)

Hens under heat stress produced shell-less eggs only when the water was not available (Table 17). Maximum number of the shell-less eggs was when both water and feed were not available during the heat stress (50% of the total eggs) (Table 17).

Table 17. Effect of feed and/or water under heat (35°C) or no heat for 4 hours (during egg shell formation) on the production of shell-less eggs.

	No heat				Heat			
	No water		+ water		No water		+ water	
	No. of SL eggs	% of total eggs	No. of SL eggs	% of total eggs	No. of SL eggs	% of total eggs	No. of SL eggs	% of total eggs
-Feed	0	0/9 (0%)	0	0/10 (0%)	6	6/12 (50%)	0	0/9 (0%)
+Feed	0	0/9 (0%)	0	0/7 (0%)	3	3/10 (30%)	0	0/7 (0%)

4.5.6. The Effect on Feed Intake

The values of feed intake 24 hours following the stay in the chamber are shown in Table 18.a, b, c, and d. There was no difference in the feed intake due to the temperature (83.83 gm for heat vs. 87.23 gm for no heat) (Table 18.d). There was a highly significant difference in the feed intake due to the presence of the feed during the stay in the chamber. The hens consumed more feed (100.2 gm) when there was feed than when there was no feed during the stay in the chamber (72.9 gm) (Table 18.b). When water was not available during the stay in the chamber the hens ate less food than when the water was available. The difference between the amounts of feed intake was highly significant (82.7 gm for no water vs. 88.5 gm for + water treatment) (Table 18.b).

Feed allowed during the stay in the chamber (with or without water) caused hens to consume significantly more food than when there was no feed available. When feed was not available during the stay in the chamber providing water did not make a difference in the subsequent feed intake (72.8 gm for no water vs. 72.9 for + water treatment) (Table 18.b). But when the feed was available during the time the hens were in the chamber, they consumed more feed in case of existence of the water than in its absence (109.6 vs. 92 gm).

Table 18. The effect of feed and/or water under heat (35°C) or no heat for 4 hrs. (during egg shell formation) on a. The feed intake 20 hrs after the hens were in the chamber (the effect of the temperature, feed and water).

Treatments	No feed				+ Feed					
	No heat		Heat		No heat		Heat		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	Av. of change	No of hens	AV. of change
No water	9	71.56	6	74.67	9	100.22	7	81.43	31	82.71
+ water	10	72.0	9	73.89	7	112.429	7	106.86	33	88.49
Overall AV. of change	19	71.79	15	74.2	16	105.56	14	94.14	64	85.69

N.B: Analysis of variance of the feed intake is shown in Table A.2.e (Appendix).

Table 18.b. The feed Intake due to the effect of water and feed.

treatments	No feed		+ Feed		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
No water	15	72.8	16	92.0	31	82.7
+ Water	19	72.9	14	109.6	33	88.5
Overall AV. of change	34	72.9	30	100.2	64	85.7

Table 18.c The feed intake due to the effect of the temperature and water

treatments	No heat		Heat		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
No water	18	85.9	13	78.3	31	82.7
+ Water	17	88.7	16	88.3	33	88.5
Overall AV. of change	35	87.2	29	83.8	64	85.7

Table 18.d. The feed intake due to the effect of the temperature and feed.

treatments	No heat		Heat		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
No feed	19	71.8	15	74.2	34	72.9
+ Feed	16	105.6	14	94.1	30	100.2
Overall AV. of change	35	87.2	29	83.8	64	85.7

When the feed was not available during the stay in the chamber, there was no effect due to the temperature on the subsequent feed intake (71.8 gm for no heat vs. 74.2 gm for heat). But when the feed was available, the hens consumed significantly more feed afterwards in case of no heat vs. heat stress (105.6 gm for no heat vs. 94.1 gm for heat treatment) (Table 18.d).

4.5.7. The Effect on the Change in Body Temperature

The change in body temperature due to 4 hours stay in the chamber during egg shell formation is presented in Tables 19.a, b, c, and d. There was a highly significant increase in body temperature due to heat (1.134°C) when compared to no heat treatment (-0.037°C) (Table 19.c).

Regardless of the effect of water and ambient temperature, there was no difference between the change in body temperature as influenced by the presence or absence of feed (0.60 vs. 0.40°C) (Table 19.b). There was a highly significant increase in body temperature when the water was not available during the stay in heat chamber (0.723°C) when compared to when the water was available (0.279°C) (Table 19.c). When the feed was available during the stay in the chamber, water reduced the increase in body temperature significantly (0.286°C) as compared to the lack of water (0.875°C) (Table 19.b). There was no difference in the change in body temperature between any of the other values in Table 19.b.

Table 19. The effect of allowing or not allowing of feed and/or water, heat (35°C) or no heat for 4 hrs., during the hens's stay in a chamber when they are actively depositing shells, on a. The change (°C) in body temperature due to the effect of the chamber's temperature, or the provision of water and feed.

Treatments	No feed				+ Feed					
	No heat		Heat		No heat		Heat		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	Av. of change	No of hens	AV. of change
No water	9	0.0	6	1.4	9	0.244	7	1.686	31	0.723
+ water	10	-0.11	9	0.7	7	-0.343	7	0.914	33	0.279
Overall AV. of change	19	-0.058	15	0.98	16	-0.013	14	1.30	64	0.494

N.B: Analysis of variance of the change in body temperature is shown in Table A.2.f (Appendix).

When water was not available during the stay in the chamber, heat caused a significant increase (1.554°C) in body temperature when compared to when the hens were not heat stressed (0.122°C). Also significantly increasing body temperature due to heat stress was when water was not available (1.554°C) when compared to when no water was available (0.794°C) (Table 19.c). Hens not stressed with 35°C in the chamber did not show a change in body temperature whether or not water was present (0.122°C vs. -0.206°C) (Table 19.c). This experiment revealed the importance of having both water and feed available during heat stress, especially during the time of egg shell formation. The results of this experiment also give an explanation for the reduction in shell quality obtained from the treatments during egg shell formation without heat stress when the feed was not available at that time. This conclusion is based on the fact that the best egg shell quality was when both of water and feed were available during the stay in the chamber. There was a negative correlation (-0.50) between the change in shell weight and the change in body temperature. This means that the increase in body temperature is one of the factors that cause a reduction in shell weight during heat stress, regardless of the effect of other factors such as feed or water.

Table 19.b. The change in body temperature due to the effect of the water and feed.

treatments	No feed		+ Feed		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
No water	15	0.56	16	0.875	31	0.723
+ Water	19	0.274	14	0.286	33	0.279
Overall AV. of change	34	0.40	30	0.60	64	0.494

Table 19.c. The change in body temperature due to The effect of the temperature and water.

treatments	No heat		Heat		Overall	
	No. of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
No water	18	0.122	13	1.554	31	0.723
+ Water	17	-0.206	16	0.794	33	0.279
Overall AV. of change	35	-0.037	29	1.134	64	0.494

Table 19.d. The change in body temperature due to the effect of the temperature and feed.

treatments	No heat		Heat		Overall	
	No of hens	AV. of change	No. of hens	AV. of change	No. of hens	AV. of change
No feed	19	-0.058	15	0.98	34	0.40
+ Feed	16	-0.013	14	1.30	30	0.60
Overall AV. of change	35	-0.037	29	1.134	64	0.494

4.6. Experiment 6. Effect of using some drugs and chemicals in the food to alleviate the effect of 4 hours of heat stress (35°C) on egg quality using SCWL hens 61 weeks of age actively forming egg shell

4.6.1. The Effect on the Change (gm) in Egg Weight

With the exception of giving the layers a diet containing 0.2% Aspirin, all the treatments caused a slight increase in egg weight. There was no significant difference in the change in egg weight due to the treatments. It was obvious that the heat stress (35°C) for 4 hours during egg shell formation does not affect the egg weight.

4.6.2. The Effect on the Change (gm) in Shell Weight

The Shell weight was not improved by any of the treatments (Table 20.1). The controls had a reduction in shell weight of 0.72 g. The minimum non-significant reduction in shell weight was for Vit C 50 mg/Kg diet (-0.52 gm). The maximum reduction was for $KHCO_3$ fed at 0.9% of the diet (-0.96 gm). There was no significant difference among any of the values due to the treatments. Heat stress reduced the shell weight in all cases.

4.6.3. The Effect on the Change in % Shell

There was no significant difference among any of the values for one of the treatments to alleviate the reduction in % shell. All the values of % shell for the 1st egg following the heat stress were less than the average of the three eggs each hen laid before being heat stressed.

Table 20.1. The effect of using vitamin C, aspirin, potassium bicarbonate and sodium bicarbonate in the feed to alleviate the effect of 4 hours of heat stress (35°C) during egg shell formation on egg quality using SCWL hens 61 weeks of age.

Treatments	No. of eggs	Shell-less eggs		AV. of the change in eggs with shells		
		No. of eggs	% of the total eggs	egg wt-g	shell wt-g	% shell
Control	6	2	33	2.64	-0.72	-1.53
Vit C - 50mg/ kg diet	6	3	50	0.55	-0.52	-0.93
Vit C - 150mg/ kg diet	7	4	57	2.22	-0.70	-1.45
Aspirin 0.2% of the diet	7	1	14	-0.11	-0.94	-1.44
$KHCO_3$ - 0.9% of the diet	6	1	17	1.57	-0.96	-1.63
$NaHCO_3$ - 0.76% of the diet	6	3	50	0.64	-0.56	-1.00
Overall average	38	14	37	1.17	-0.78	-1.38

N.B: There was no significant differences ($P > .05$) between the values of the change in egg shell quality parameters (egg wt., shell wt., or % shell) due to the treatments.

4.6.4. The Effect on Producing Shell-less Eggs

Many of the hens produced shell-less eggs in this experiment (Table 20.1). The minimum number of shell-less eggs was produced by hens fed $KHCO_3$ and Aspirin. The most shell-less eggs were laid by hens given Vit C at 150 mg/kg diet (Table 20.1).

4.6.5. The Effect on Feed Intake

Hens fed the commercial diet at all times consumed 69.5 gm feed per bird during the 24 hours after 4 hours of heat stress. In comparison those fed the diet containing $KHCO_3$ ate 64.6 gm, while those fed a diet with $NaHCO_3$ ate 79.3 gm/bird. There was no significant difference in feed intake from these treatments (Table 20.2).

4.6.6. The Effect on the Change in Body Temperature

The maximum increase in body temperature from 35°C heat stress was for hens given for vit C at 50 mg/kg diet treatment (0.53°C) and the minimum increase (0.13°C) was for 150 mg vit C/kg diet treatment (Table 20.2). There was no significant effect due to the treatments on the change in body temperature.

4.7. Experiment 7. Effect of using aspirin and potassium bicarbonate to alleviate the effect of heat stress (35°C) for 4 hours during egg shell formation on egg quality using SCWL 61 weeks of age

Because aspirin and potassium bicarbonate in the previous experiment gave the lowest number of shell-less

Table 20.2. The effect of vitamin C, aspirin, potassium bicarbonate and sodium bicarbonate on feed intake and the change in body temperature due to heat stress (35°C) for 4 hrs. during egg shell formation using SCWL hens 61 weeks of age.

Treatments	No. of hens laid eggs with shells	Feed intake (gm) 20 hrs. following the stay in the chamber	The change in body temperature (°C)
Control	4	69.5	0.38
Vit C - 50mg/ kg diet	3	75.7	0.53
Vit C - 150mg/ kg diet	3	79	0.13
Aspirin 0.2% of the diet	6	78	0.30
$KHCO_3$ - 0.9% of the diet	5	64.6	0.32
$NaHCO_3$ - 0.76% of diet	3	79.3	0.40

N.B: There was no significant difference ($P > .05$) between the values of feed intake or the change in body temperature due to the treatments.

eggs, these treatments were repeated to determine if the results could be duplicated.

4.7.1. The Effect on the Egg Quality

None of the egg quality parameters was improved by feeding aspirin or $KHCO_3$ (Table 21.1). Heat stress did not have a negative effect on egg weight except for control hens (-0.929 gm egg weight). There were no significant differences between the values for each parameter. Heat stress (35°C) for 4 hours during egg shell formation reduced egg shell quality (including shell thickness) for the 1st egg following the heat stress when compared to the average of the three eggs laid prior to heat stress. Aspirin or potassium bicarbonate in the diet did not prevent shell-less eggs.

4.7.2. The Effect on Feed Intake

The amounts of feed consumed 24 hours following 4 hours of heat stress (35°C) are shown in Table 21.2. The hens in the control group consumed 56 gm whereas the hens fed $KHCO_3$ in their diet consumed 69 gm. There was no significant difference in feed intake among the treatment groups.

4.7.3. The Effect on the Change in Body Temperature

The least increase in the body temperature was detected in control hens (0.48°C); whereas the most increase was for

Table 21.1. The effect of using Aspirin and Potassium bicarbonate in the feed to alleviate 4 hours of heat stress (35°C) during egg shell formation on egg quality using SCWL hens 61 weeks of age.

Egg quality parameters	Control	Aspirin 0.2% of the diet	$KHCO_3$ 0.9% of the diet
No. of eggs with shells	8	6	8
AV. of the change in			
egg weight (gm)	-0.93	0.66	1.01
shell weight (gm)	-0.87	-0.90	-0.61
% shell	-1.26	-1.48	-1.07
Shell thickness (mm)	-4.66	-5.08	-3.45
No. of shell-less eggs	2	3	4
% of shell-less egg of the total eggs	20	33	33

Table 21.2. The effect of feeding aspirin or potassium bicarbonate on feed intake and the change in body temperature due to 4 hours of heat stress (35°C) during egg shell formation using SCWL hens 61 weeks of age.

Treatments	No. of hens laid eggs with shells	Feed intake (gm) 20 hrs. following the stay in the chamber	The change in body temperature (°C)
Control	8	56	0.48
Aspirin 0.2%	6	61	0.92
$KHCO_3$ 0.9%	8	69	0.79

N.B: There was no significant difference ($P > .05$) between the values of the change in egg quality parameters, feed intake or the change in body temperature due to the treatments.

hens fed aspirin (0.92°C). There was no significant differences between any of the values due to the treatments.

4.7.4. The Effect on the Blood Parameters (pH, PCO_2 and HCO_3^-)

The results of the blood parameters, summarized in Table 21.3, reveal that the highest value of blood pH (7.4) was for hens fed $KHCO_3$ in their diet, and the minimum value (7.376) was for the control group. Blood pressure of the blood carbon dioxide (PCO_2) ranged between 37.3 mm Hg for aspirin treatment and 40.35 mm Hg for control group. The concentration of blood bicarbonate ions (HCO_3^-) ranged between 20.85 for aspirin group and 22.38 for control group. There was no significant difference between any of the values for each of the blood parameters due to the treatments.

4.8. Experiment 8. The difference between the effect of heat stress (35°C) for 4 hours during egg shell formation or no heat at that time on the blood parameters (pH, PCO_2 and HCO_3^-) of SCWL hens 61 weeks of age.

4.8.1. The Effect on the Blood pH

The pH was not significantly affected by 35°C for 4 hours during egg shell formation. The values were 7.35 for hens at ambient temperature (about 21°C) vs. 7.37 for those heat stressed for 4 hours (Table 22).

4.8.2. The Effect on Blood PCO_2

The blood PCO_2 activity in the hens kept in the chamber at 21°C was 54.18 mm Hg as compared to the significantly ($P < .01$) lower value of 40.89 mm Hg at the end of 4 hours exposure to 35°C (Table 22).

4.8.3. The Effect on Blood HCO_3^-

Heat stress (35°C) for 4 hours during egg shell formation decreased blood HCO_3^- significantly ($P \leq 0.01$) to 22.3 to 28.58 in hens at 21°C (Table 22).

Table 22. Effect of heat stress (35°C) for 4 hours on blood pH, PCO_2 and HCO_3^- during egg shell formation in SCWL hens 61 weeks of age

Group	Count	pH	PCO_2 (mm Hg)	HCO_3^- (m mol/l)
Control (at 21°C)	10	7.35^a	54.18^a	28.58^a
Heat (35°C)	12	7.37^a	40.89^b	22.32^b

a, b values in the same column differently supercripted are significantly different ($P \leq .01$).

4.9. Experiment 9. Effect on egg quality when using some drugs and minerals in the drinking water to alleviate the effect of 4 hours of heat stress (35°C) during egg shell formation of SCWL hens 61 weeks of age.

In this experiment, a small number of hens (4 hens) were used to just give an idea about how the hens will

respond to the chemicals which were used and the effect of these chemicals on egg shell quality during heat stress.

4.9.1. The Effect on the Change (gm) in Egg Weight

Egg weight was not affected by treatment. The results of the change in egg weight in this experiment confirm those of most of the previous experiments in that heat stress (35°C) up to 6 hours does not affect egg weight. The average egg weight was 1.23 to 3.28 gm higher of the treated groups as compared to the 3.19 gm higher weight of the control eggs (Table 23.1).

4.9.2. The Effect on the Change (gm) in Shell Weight

Hens treated with potassium chloride, calcium chloride, aspirin, acetaminohen, or a buffer solution laid eggs with less shell as a result of heat stress; their shell weights were -0.15 to -1.34 gm less than the shells laid just prior to heat stress. Hens not treated laid eggs with shells weighing -1.15 gm than prior to heat stress. There was no indications that any of the treatments acted to lessen the effect of heat stress on shell weight (Table 23.1).

4.9.3. The Effect on the Change in % Shell

The % shell was not improved by any of the treatments in drinking water and the effect of heat stress was to reduce the % shell for the first egg following the heat

Table 21.3. The effect of aspirin or potassium bicarbonate on the blood pH, blood PCO_2 and blood HCO_3 of hens, actively depositing shell, stressed for 4 hrs. at 35°C during egg shell formation.

Treatments	No. of hens laid eggs with shells	Blood pH	Blood PCO_2 (mm Hg)	Blood HCO_3
Control	8	7.38	40.35	22.38
Aspirin 0.2%	6	7.38	37.30	20.85
$KHCO_3$ 0.9%	8	7.40	37.71	21.99

N.B: There was no significant difference ($P > .05$) between the values of blood pH, PCO_2 or blood bicarbonate due to the treatments.

Table 23.1. The effect of using some drugs in the water to alleviate the effect of heat stress (35°C) for 4 hrs. during egg shell formation on egg quality using SCWL hens 61 weeks of age.

Treatments	No. of eggs with shells	AV. of the change in egg wt-g	shell wt-g	% shell	Shell-less eggs No. of eggs	% of the total eggs
Control	4	3.19	-1.15	-2.23	0	0%
KCl - 1.6gm/l	4	1.23	-1.15	-1.93	0	0%
$CaCl_2$ - 8.3gm/l	1	2.06	-0.64	-1.41	3	75%
Aspirin - 1.083gm/l	3	3.28	-0.15	-0.65	0	0%
Acetaminophen - 0.267gm/l	3	1.49	-1.10	-2.05	1	25%
Buffer solution - 4.81 pH	3	1.92	-1.34	-2.34	0	0%

N.B: There appeared to be no differences among the values of the change in each of egg quality parameters (egg wt., shell wt., or % shell) due to the treatments.

stress when compared to 3 eggs laid just before heat stress (Table 23.1).

4.9.4. The Effect on the Production of Shell-less Eggs

Calcium chloride at 8.3 gm/l caused 75% of the eggs laid to be shell-less. One of the 4 hens treated with acetaminophen laid a shell-less eggs and all of the other hens laid eggs with shells although they weighed less (except for aspirin) (Table 23.1).

4.9.5. The Effect on the Consumption of Feed, Water and the Drugs

Feed intake/hen ranged between 61.5 gm for buffer solution treatment and 83.5 gm for aspirin group as compared to 64.3 gm for controls. Water intake during the period of heat stress (4 hours) ranged between 44 ml for $CaCl_2$ group and 267.3 ml for acetaminophen group as compared to 194 ml for controls that were heat-stressed. There appeared to be no differences among the values for both of feed intake and water intake. In this experiment the consumption per hen was 168.3 mg K, 132 mg Ca, 249.7 mg aspirin, 71.4 mg acetaminophen and 113 mg K in case of KCl, $CaCl_2$, aspirin, acetaminophen and buffer solution treatments, respectively (Table 23.2).

4.9.6. The Effect on the Change in Body Temperature

Mean change in body temperature is shown in Table 23.2. All the values were higher after the heat stress than before

Table 23.2. The relationship between the drugs and both of the feed intake, water intake, drug intake and the change in the body temperature due to heat stress (35°C) for 4 hrs.

Treatments	Feed intake (gm) 20 hours following the heat stress		Water intake (ml) during heat stress		Drug intake (mg/hen) during heat stress		The change in body temperature (°C)	
	No. of hens laid eggs with shells	AV. of intake	No. of hens	AV. of intake	No. of hens	AV. of intake	No. of hens	Av.of change
Control	3	64.3	4	194.0	4	0	4	-0.08
KCl (1.6 gm/l)	3	65.3	4	210.0	4	168.3 (as K)	4	0.28
$CaCl_2$ (8.3 gm/l)	1	62.0	1	44.0	1	132.0 (as Ca)	1	0.70
Aspirin (1.083 gm/l)	2	83.5	3	227.0	3	249.7	3	0.30
Acetaminophen (0.267 gm/l)	3	69.0	3	267.3	3	71.4	3	-0.43
Buffer solution (4.81 pH)	2	61.5	3	141.3	3	113.0 (as K)	3	0.27

N.B: There appeared to be no differences among the values of feed intake, water intake, or the change in body temperature due to the treatment.

heat stress except for the control group in which there was almost no change (-0.08°C), and the acetaminophen group in which the body temperature decreased (-0.43°C). There was no indication that any of the treatments acted to keep body temperature from rising.

4.10. Experiment 10. Effect of using some drugs and minerals in the water during heat stress (35°C) for 4 hours in addition to an oral dose 2 hours before hens were placed into the environmental chamber on egg quality using SCWL hens 61 weeks of age

4.10.1. The Effect on the Change in Egg Quality

None of the treatments in this experiment prevented heat stress from reducing the egg quality parameters (egg weight, shell weight or % shell) (Table 24.1). Some of the values for egg weight increased after heat stress; whereas those of eggs from hens treated with KCl (Level 1) or carbonated water + vit C decreased about -0.50 gm. Changes in shell weight ranged between -0.55 gm for the control group and -1.44 gm for hens given carbonated water with vit C. The change in % shell was between -0.94% for KCl (Level 2) treatment and -2.20% for carbonated water with vit C. There was no significant difference ($P > .05$) between the values of any of these egg quality parameters due to treatments. The highest number of shell-less eggs (50%) was observed with birds receiving the KCl (Level 2) treatments (Table 24.1).

Table 24.1. The effect of using an oral dose of potassium chloride, carbonated water with vitamin C, or acetaminophen with vitamin C before heat stress in addition to that in the water during heat stress (35°C) to alleviate the effect of 4 hrs. of heat stress during egg shell formation in SCWL hens 61 weeks of age.

Treatments	No. of eggs with shells	AV. of the change in			Shell-less eggs	
		egg wt-g	shell wt-g	% shell	No. of eggs	% of the total eggs
Control	13	1.51	-0.55	-1.03	1	7.14
KCl (0.3 gm/hen)	10	-0.45	-0.94	-1.41	1	9.09
KCl (0.6 gm/hen)	5	0.07	-0.59	-0.94	5	50.00
Carbonated water + Vit C	2	-0.50	-1.44	-2.20	2	50.00
Acetaminophen + Vit C	4	0.82	-0.63	-1.12	2	33.33

4.10.2. The Effect on the intake of Feed, Water and Chemicals

Because both feed and water were available during the stay in the chamber, the maximum amount of feed consumed was 90.6 gm (for KCl Level 2 group), and the minimum amount was 55.5 gm (carbonated water with vit C) (Table 24.2). Each hen consumed 222.5 ml water that contained acetaminophen during the 4 hours of heat stress. Those provided carbonated water with vit C drank only 60.0 ml/hen. The controls drank 203 ml during that time. In this experiment each hen consumed a total of 325.3 mg K, 603.2 mg K and 238.5 mg acetaminophen for KCl (Level 1), KCl (Level 2) and acetaminophen groups, respectively.

4.10.3. The Effect on the Change in Body Temperature

Controls had an increase in body temperature of 0.52°C due to heat stress (Table 24.2). The hens treated with various chemicals in the drinking water had increases in body temperature of 0.23°C, for birds that consumed KCl (level 1), up to a maximum increase of 1.0°C for birds that consumed the carbonated water during the heat stress (Table 24.2). According to an ANOVA, no significant differences ($P > .05$) were detected for body temperatures due to any of the treatments (Table 24.2).

Table 24.2. The effect of potassium chloride, carbonated water with vitamin C, and acetaminophen with vitamin C on feed intake, water intake, chemical intake, and the change in body temperature due to 4 hours of heat stress (35°C) at the time of egg shell formation.

Treatments	No. of hens laid eggs with shells	Feed intake(g) 20 hours following heat stress	Water intake(ml) during heat stress	Chemical intake (mg/hen) during heat stress	The average of the change in body temp. (°C)
Control	13	80.6	203	0	0.52
KCl (0.3 gm/hen)	10	77.7	175	325.3 (as K)	0.23
KCl (0.6 gm/hen)	5	90.6	152	603.2 (as K)	0.32
Carbonated water + Vit C	2	55.5	60	0 5.0 (vit C)	1.00
Acetaminophen + Vit C	4	75.0	223	238.5 5.0 (vit C)	0.75

5. Discussion

The primary effect of acute heat stress on egg quality was to reduce shell weight and other measures involving shell. Egg weight was not significantly reduced. Wolfenson et al. (1979) reported that the exposure to heat stress (34 to 40°C) for 6 to 7 hours for only one day lowered shell weight ($P < .05$). The results of experiment 1 in this study showed that the exposure to 35°C for only 4 hours for just one day caused a significant reduction in egg shell quality as measured by shell weight, % shell and mg Ca/mm^2 of the shell. Furthermore, these results indicated, for the first time, that exposure of the hens to 35°C for 1 hour or 2 hours did not cause an effect on egg shell quality. Harrison and Biellier (1969) found that there was a decrease in egg shell quality (specific gravity) within the first day of exposure to 35°C but there was no change in egg weight. The results of our experiments confirm this. On the other hand, Vasquez and Teeter (1986) showed that heat stress (35°C) depressed both shell weight and egg weight. The reduction in shell quality due to heat stress was in agreement with that obtained by De Andrade et al. (1976 and 1977) who observed that % shell and other shell quality parameters such as specific gravity and shell thickness

decreased at the end of three weeks exposure to 32°C. The results of this experiment supported the results obtained by Wolfenson et al. (1979) in that shell quality which was decreased due to heat stress on the day after heating reverted to that of controls on the second and third day after the heat treatment.

The effect of heat stress (35°C) on the egg quality of hens 46 weeks of age (experiment 2) was also detected in hens 61 weeks of age; both laid eggs with less shell weight from heat stress for 6 hours, while their egg weight was not affected by that stress. Heat stress (35°C) for 6 hours during (No SF) time, when hens were not actively depositing shell, did not affect food intake over the next 24 hours. Wolfenson et al. (1979) had reported that there was no significant reduction in total daily feed consumption due to heat stress (34 to 40°C) for 6 to 7 hours. The effect of heat stress on egg shell weight was not mainly due to the reduction in feed intake but due to other changes in the body, as an example a rise in body temperature.

The significant increase in body temperature due to heat stress confirmed the earlier studies of Thornton (1962). Laying hens are more susceptible to any stress, i.e., no feed or the increase in the environmental temperature, during the time of egg shell formation than when there is no egg shell formation. As a result of the absence of feed during the hen's stay in the chamber there was a significant reduction in total daily feed consumption

if the hen is actively depositing shell. This did not occur with hens stressed with heat and not actively depositing shell. The reduction in feed intake at the time of shell deposition, even when there was no heat, indicated the importance of offering the feed during that time. This observation indicated that the reduction in shell quality of egg obtained from (SF) time experiments was not only due to heat stress but also the absence of feed at that time. Those not heat stressed also showed the reduction in shell quality from the absence of feed. The importance of the time of feed intake was indicated by Farmer et al. (1983) who concluded that the most important time for the hens to receive calcium was during the afternoon when shell calcification is initiated. Also, Former and Roland (1982) indicated that not only the calcium was important at the time of egg shell calcification but also the other nutrients (the whole feed) were also needed. The reduction in body temperature at the time of egg shell formation when there was no heat (Exp. 3) may be related to less ability in regulating the body temperature during egg shell formation, or to a lower metabolism from a sleep induced effect.

In addition to the importance of the feed at the time of egg shell deposition, water was also important at that time, regardless of the environmental temperature, to obtain the best egg shell quality. Oluyemi and Adebanjo (1979) observed that the shells of the old hens (62 weeks old) kept in an environmental temperature of 22.8 to 36.9°C were

improved with the offering of cold water (0°C); or feeding the hens during the time of egg shell formation. The study of Oluyemi and Adebanjo was different than the present study. In this study, various combinations of feed and water were used in a 2 x 2 x 2 factorial at the time of egg shell formation with 4 hours of heat stress (35°C). The best egg shell quality was obtained when both the water and feed were available at the time of egg shell formation with heat or without heat. The worst egg shell quality was when the water was not present and the feed was present during heat stress. This may be due to heat that is produced from the feed in addition to the heat stress. It was observed that hens during their stay in the chamber with heat (35°C) consumed about 22 gm feed during the 4 hours. Water was effective in reducing the increase in body temperature due to heat and its existence improved feed intake. Both water and feed were required for optimum shell quality.

There was no improvement in egg shell quality due to the use of analgesic drugs such as aspirin (in feed or water) or acetaminophen (in water). Oluyemi and Adebanjo (1979) showed that aspirin at 0.15 and 0.2% of the diet with environmental temperature of 22.8 to 36.9°C over a period of 5 weeks did not improve shell quality (shell thickness) in case of old hens (62 weeks old). In this experiment, 0.2% aspirin in the diet for at least 3 days before 4 hours of 35°C heat stress was not effective. The use of bicarbonate ions (as $NaHCO_3$ or $KHCO_3$) at 0.55% of the diet was also not

effective in improving shell quality. The concentration of Na and K was 0.21 and 0.35% of the diets. Possibly, bicarbonate ions or these minerals were not effective in alleviating the effect of heat stress on egg shell quality because feed was absent during heat stress. The presence of whole feed during egg shell formation is important. Ahmed et al. (1967) observed that the 15-month old hens being fed vitamin C (44 mg/kg diet) could not maintain shell thickness at 35°C. They also observed, and substantiated by our experiments that vitamin C had no significant effect on preventing a rise in body temperature due to heat.

Balnave and Scott (1986) reported that inclusion in the water of low concentrations of KCl (40 mg/l) and $CaCl_2$ (120 mg/l) substantially increased the incidence of shell defects. In this study, the inclusion in the water of high concentration of KCl (1.6 gm/l) and $CaCl_2$ (8.3 gm/l) also increased the incidence of shell defects. For example, $CaCl_2$ treatment gave the highest number of shell-less eggs. Balnave and Scott (1986) indicated that this effect may be due to the chloride ion and that the increase in shell defects was not associated with the pH of the water. Carbonated water did not have an effect on egg shell quality, and that may be due to the loss of CO_2 from the carbonated water during heat stress. Acetaminophen did not prevent the heat stress effect in these experiments, this may be because treatment was not long enough to give it a chance to be effective during the time of heat stress.

The effect of heat stress on blood bicarbonate ions PCO_2 was indicated by Mongin and Lacassagne (1966) who observed that the hyperventilation due to high temperature caused the hen to lower its blood carbon dioxide tension and thus its blood bicarbonate level. Gutowska and Mitchell (1945) indicated that the bicarbonate ions are the source of shell carbonate. The reduction in PCO_2 due to heat stress was observed also by Kohne and Jones (1975a) who studied the effect of acute hyperthermia (49°C for 165 minutes) on blood PCO_2 and pH using nonlaying female turkeys. They found that the venous blood pH and PCO_2 did not change significantly during the first 60 minutes of the experimental period, and not until the ambient temperature reached 43°C. The venous pH increased significantly (from 7.4 to 7.69) and the PCO_2 decreased significantly (from 54.5 to 16.3 mm Hg) during the experimental period. In this experiment the temperature was only 35°C and this temperature did not reduce the PCO_2 to the degree which was observed by Kohne and Jones (1975a). The significant reduction in blood HCO_3 ions may be one of the factors that caused the reduction in egg shell quality due to heat stress (even when both of feed and water were available during the treatments) when compared to no heat treatments.

The overall conclusion of all experiments indicates that egg shell quality can be improved during the hot environmental temperature by somehow keeping the body temperature of the hen and its blood bicarbonate ions

without change. In addition, offering feed and cold water together especially at the time of egg shell formation may help maintain egg shell quality.

6. SUMMARY AND CONCLUSION

During summer, increased temperature causes hens to lay egg with lesser quality shells, thereby increasing egg breakage. This study was conducted with SCWL laying hens to establish a procedure to evaluate the effect of acute heat stress on shell quality without endangering the hens. This research also included attempts to overcome the detrimental effect of heat stress on egg shell quality.

The results of the study could be summarized as follows:

1. Four or six hours exposure to heat stress (35°C) for only one day was enough to reduce egg shell quality (shell weight, % shell, and Ca/mm^2 of shell) significantly for only one day (1st day following the treatment). Generally, egg weight was not affected by heat stress in this study. There was a high correlation between shell quality parameters.
2. The effect of heat stress (35°C) for 6 hours when shell deposition was not occurring, in case of young hen (46 weeks of age) was significantly effected only the shell weight of many factors analyzed. This effect may be due to the significant increase in body temperature rather than the difference in feed intake at that time.

3. There was a significant reduction in shell weight due to the stay in the chamber (with or without heat) when feed was not available during egg shell deposition.
4. Minimum reduction in shell weight due to heat stress (-0.45 gm) was observed when both of feed and water were available during heat stress. There was a reduction of 0.98 gm shell weight for (+ Feed No water) treatment due to heat stress.
5. Supplementing the diet with vitamin C at 50 or 150 mg/kg; aspirin, 0.2%; potassium bicarbonate, 0.9% or sodium bicarbonate, 0.75% did not prevent heat stress (35°C) effect on egg shell quality.
6. Blood PCO_2 and HCO_3^- significantly ($P \leq .01$) declined in heat stressed hens.
7. The inclusion in the water (per liter) of 1.6 gm KCl; 8.30 gm $CaCl_2$, 1.083 gm aspirin (as sodium acetyl salicylate), 0.267 gm acetaminophen, or 4.81 pH buffer solution did not improve egg shell quality.
8. The high doses of KCl (0.3 or 0.6 gm/hen) or acetaminophen (0.239 gm with 5mg vitamin C/hen) did not prevent the effect of heat stress on egg shell quality. Also carbonated water with 5mg vitamin C/hen did not have a positive effect on egg shell quality.

The overall conclusion of this study indicates that egg shell quality can be improved during the hot environmental temperature by somehow keeping the body temperature of the hen and its blood bicarbonate ions static as the same as

before heat stress. In addition, offering feed and cold water together especially at the time of egg shell formation may help maintain egg shell quality.

BIBLIOGRAPHY

BIBLIOGRAPHY

Ahmad, M.M., R.E. Moreng, and H.D. Muller, 1967. Breed responses in body temperature to elevated environmental temperature and ascorbic acid. Poultry Sci. 46:6.

Alcock, N., and I. MacIntyre, 1962. Inter-relation of calcium and magnesium absorption. Clin. Sci. 22:185-193.

Anderson, G.B., T.C. Carter, and R.M. Jones, 1970. Some factors affecting the incidence of cracks in hen's egg shells. Br. Poultry Sci. 11:103-116.

Arieli, A., A. Meltzer, and A. Berman, 1979. Seasonal acclimatization in the hen. Br. Poultry Sci. 20:505-513.

Arieli, A., A. Meltzer, and A. Berman, 1980. The thermo-neutral temperature zone and seasonal acclimatization in the hen. Br. Poultry Sci. 21:471-478.

Arima, Y., F.B. Mather, and M.M. Ahmad, 1976. Response of egg production and shell quality to increases in environmental temperature in two age groups of hens. Poultry Sci. 55:818-820.

Asmundson, V.S. and P. Pinsky, 1935. The effect of the thyroid on the formation of the hen's eggs. Poultry Sci. 14:99-104.

Atteh, J.O., and S. Leeson, 1983. Influence of Increasing Dietary Calcium and Magnesium levels on Performance, Mineral Metabolism, and Egg Mineral content of laying hens. Poultry Sci. 62:1261-1268.

Balnave, D., and I. Yoselewitz, 1987. The relation between sodium chloride concentration in drinking water and egg-shell damage. Br. J. of Nutrition 58:503-509.

Balnave, D., and T. Scott, 1986. The influence of Minerals in drinking water on egg shell quality. Nutrition Reports International July 34(1):29-34.

Bar, A., and S. Hurwitz, 1975. Intestinal and uterine

calcium-binding protein in laying hens during different stages of egg formation. Poultry Sci. 54:1325-1327.

Becker, W.A., and G.E. Bearse, 1962. Sulfanilamide, specific gravity of eggs and an interaction. Poultry Sci. 41:198-200.

Bell, D.J., and B.M. Freeman, 1971. Physiology and Biochemistry of the domestic fowl. Academic Press Inc. (London) Ltd.

Belyavin, C.G., and K.N. Boorman, 1980. The influence of the cuticle on egg-shell strength. Br. Poultry Sci. 21:295-298.

Berg, L.R., and G.E. Bearse, 1948. The effect of iodinated casein and thiouracial on the performance of white Leghorn pullets. Poultry Sci. 27:653.

Berg, L.R., and G.E. Bearse, 1951. Effect of iodinated casein and thiouracial on the performance of laying birds. Poultry Sci. 30:21-28.

Blake, J.P., L.J. Kling, and W.A. Halteman, 1985. The relationship of the amino acid composition of a portion of the outer egg shell membrane to egg shell quality. Poultry Sci. 64:176-182.

Burmester, B.R., H.M. Scotte, and L.E. Card, 1939. Rate of egg shell formation in the hen. World's Poultry Congr. 7th ed. 99-101. (Poultry Sci. 61:2005, 1982).

Campos, A.C., F.H. Wilcox, and C.S. Shaffner, 1960. The influence of fast and slow uses in ambient temperature on production traits and mortality of laying hens. Poultry Sci. 39:119-129.

Carson, J.R., R.D. Eaton, and R.E. Luginbuhl, 1954. The effect of injections of an oil suspension of tetramycin on egg production and egg quality in hens affected with chronic respiratory disease. Poultry Sci. 33:589-596.

Carter, T.C., 1975. The hen's egg: Estimation of shell superficial area and egg volume, using measurements of fresh egg weight and shell length and breadth alone or in combination. Br. Poultry Sci. 16:541-543.

Casey, J.M., J.C. Gilbreath, and R.D. Morrison, 1963. Turkey reproduction as influenced by reserpine administered during growth. Poultry Sci. 42:637-641.

Castaldo, D.J., and D.V. Maurice, 1988. Phospholipid

content of the chicken shell gland and its relationship to egg shell strength. Poultry Sci. 67:427-433.

Castillo, L., Y. Tanaka, M.J. Wineleand, J.O. Jowsey, and H.F. Deluca, 1979. Production of 1-25- dihydroxy vitamin D and formation of medullary bone in the egg-laying hen. Endocrinology 104:1598-1601.

Choi, J.H., R.D. Mites, A.S. Arafa, and R.H. Harms, 1981. The influence of oviposition time on egg weight, shell quality, and blood phosphorous. Poultry Sci. 60:824-828.

Christakos, S., E.J. Friedlander, B.R. Frandsen, and A.W. Norman, 1979. Studies on the mode of action of calciferol. Development of a radio-immunoassay for vitamin D-dependent chick intestinal calcium-binding protein and tissue distribution. Endocrinology 104:1495-1503.

Costello, D., and T.W. Odom, 1984. The relationship between the concentration of sodium in bone and egg shell with percent mineral ash. Poultry Sci. 63:(Supplement 1).

Crinion, R.A.P., 1969. Egg quality following infectious bronchitis at an early age. Poultry Sci. 48:1799-1800.

Dale, N.M., and H.L. Fuller, 1980. Effect of diet composition on feed intake and growth of chicks under heat stress. Poultry Sci. 59:1434-1441.

DeAndrade, A.N., J.C. Rogler, and W.R. Featherston, 1976. Influence of constant elevated temperature and diet on egg production and shell quality. Poultry Sci. 55:685-693.

DeAndrade, A.N., J.C. Rogler, W.R. Featherston, and C.W. Alliston, 1977. Interrelationships between diet and elevated temperature (cyclic and constant) on egg production and shell quality. Poultry Sci. 56:1178-1188.

Deaton, J.W., F.N. Reece, J.L. McNaughton, and B.O. Lott, 1981. Effect of differing temperature cycles on egg shell quality and layer performance. Poultry Sci. 60:733-737.

Deetz, L.E., and R.C. Ringrose, 1976. Effect of heat stress on the potassium requirement of the hen. Poultry Sci. 55:1765-1770.

Edens, F.W., 1976. Body temperature and blood chemistry

responses in broiler cockerels given a single intravenous injection of Na+ or Ca++ and before acute heating episode. Poultry Sci. 55:2248.

Eoff, H.J., R.E. Davies, T.M. Ferguson, and J.R. Couch, 1962. Studies on the influence of terephthalic acid and broad spectrum antibiotics on egg production and egg shell coloration in caged layers. Poultry Sci. 41:1036-1041.

Gabuten, A.R., and C.S. Shaffner, 1954. A Study of the physiological mechanisms affecting specific gravity of chicken eggs. Poultry Sci. 33:47-53.

Gilbert, B., G.G. Mitchell, M.F. Davidson, K.F. Laughlin, B.O. Hughes, 1982. Effect of administering two prostaglandin synthetase inhibitors (indomethacin and aspirin) on egg production in the domestic fowl. Research in Veterinary Sci. 33:216-220.

Goto, K., G.C. Jr. Harris, and P.W. Waldroup, 1979. Relationship between pimpling of egg shells, environmental temperature and carbonic anhydrase activity of certain body tissues. Poultry Sci. 58:1014.

Gutowska, M.S., and C.A. Michell, 1945. Carbonic anhydrase in the calcification of the egg shell. Poultry Sci. 24:159-167.

Farmer, M., and D.A. Sr. Roland, 1982. Calcium utilization in the laying hen. Poultry Sci. 61:1378.

Farmer, M., D.A. Sr. Roland, and M.K. Eckman, 1983. Calcium metabolism in broiler breeder hens. 2. The influence of the time of feeding on calcium status of the digestive system and egg shell quality in broiler breeders. Poultry Sci. 62:465.

Ferreira, S.S., 1979. A discussion on the mode of action of non-steroidal anti-inflammatory drugs: Inhibition of prostaglandin synthesis. Prostaglandin systems, New York, NY: F. Berti and G.P. Velo, ed. Plenum Press (Poultry Sci. 61:1069, 1982).

Fox, T.W., 1980. The effects of thiouracial and thyroxin on resistance to heat shock. Poultry Sci. 59:2391.

Hamilton, R.M.G., 1982. Methods and factors that affect the measurement of egg shell quality. Poultry Sci. 61:2022-2039.

Hamilton, R.M.G., and J.D. Cipera, 1981. Effects of dietary

calcium levels during the brooding, rearing, and early laying period on feed intake, egg production, and shell quality of white Leghorn hens. Poultry Sci. 60:349-357.

Hamilton, R.M.G., K.G. Hollands, P.W. Voisey, and A.A. Grunder, 1979. Relationship between egg shell quality and shell breakage and factors that affect shell breakage in the field. World's Poultry Sci. J. 35:177-190.

Harrison, P.C., and H.V. Biellier, 1969. Physiological response of domestic fowl to abrupt changes of ambient air temperature. Poultry Sci. 49:1034-1045.

Hester, P.Y., E.K. Wilson, F.W. Pierson, and I. Fadijansha, 1980. Plasma inorganic phosphate, calcium, and magnesium levels of hens which laid soft-shell or shell-less eggs. Poultry Sci. 59:2336-2341.

Heywang, B.W., 1965. The effect of the tranquilizer reserpidine on caged white Leghorn layers during hot weather in a semi-arid subtropical climate. Poultry Sci. 44:1270-1273.

Hodges, R.D., 1965. The blood supply to the avian oviduct, with special reference to the shell gland. J. Anat. 99:485-506.

Hodges, R.D., and K. Lorcher, 1967. Possible sources of the carbonate fraction of egg-shell calcium carbonate. Nature, Lond. 216:609-610.

Holder, D.P., and D.M. Huntly, 1978. Influence of added manganese, magnesium, zinc, and calcium level on egg shell quality. Poultry Sci. 57:1629-1634.

Hunt, J.R., and J.R. Aitken, 1962a. Studies on the influence of ascorbic acid on shell quality. Poultry Sci. 41:219.

Hunt, J.R., and J.R. Aitken, 1962b. The effect of ammonium and chloride ions in the diet of hens on egg shell quality. Poultry Sci. 41:434-438.

Hurwitz, S., 1970. The role of the intestine in calcium homeostasis in the laying hen. Ann. Biol. Anim. Bioch. Biophys. 10:69-76.

Hurwitz, S., 1978. Calcium metabolism in birds. Chemical Zoology. X. Aves:273-306.

Hurwitz, S., and A. Bar, 1965. Absorption of calcium and phosphorous along the gastrointestinal tract of the laying fowl as influenced by dietary calcium and egg shell formation. J. Nutr. 86:433-438.

Hurwitz, S., and P.G. Griminger, 1962. Estimation of calcium and phosphorus requirements of laying hens by balance techniques. J. Sci. Food Agr. 13:185-192.

Hutchinson, J.C.D., and A.H. Sykes, 1953. Physiological acclimatization of fowls to a hot, humid environment. J. of Agri. Sci. (Cambridge) 43:294-322.

Ishak, T.K., and A.H. Sykes, 1974. Effect of dietary ascorbic acid on the performance of laying hens under warm environmental conditions. Br. Poultry Sci. 15:449-457.

Jaap, R.G., and F.V. Muir, 1968. Erratic oviposition and egg defects in broiler-type pullets. Poultry Sci. 47:417-423.

Junqueira, O.M., R.D. Miles, and R.H. Harms, 1984. Interrelationship between phosphorous, sodium, and chloride in the diet of laying hens. Poultry Sci. 63:1229-1236.

Kechik, I.T., and A.H. Sykes, 1978. Effect of hormones and acute environmental stress on plasma ascorbic acid in the fowl. Malaysian Applied Biology 7:105. (World's Poultry Sci. J. 42:107, 1986).

Klingensmith, P.M., and P.Y. Hester, 1983. The relationship of dietary levels of phosphorus to the production of soft-shelled and shell-less eggs. Poultry Sci. 62:1860-1868.

Kohne, H.J., and J.E. Jones, 1975a. Changes in plasma electrolytes, acid-base balance and other physiological parameters of adult female turkeys under conditions of acute hyperthermia. Poultry Sci. 54:2034-2038.

Kohne, H.J., and J.E. Jones, 1975b. Acid base balance, plasma electrolytes and production performance of adult turkey hens under conditions of increasing ambient temperature. Poultry Sci. 54:2038-2045.

Kuhi, H.J. Jr., D.P. Holder, and T.W. Sullivan, 1977. Influence of dietary calcium level, source, and particle size on performance of laying chickens. Poultry Sci. 56:605.

Leach, R.M., and J.R. Gross, 1983. The effect of manganese

deficiency upon the ultrastructure of the egg shell. Poultry Sci. 62:499-504.

Lennards, R.M., and D.A. Sr. Roland, 1981. The influence of time of dietary calcium intake on shell quality. Poultry Sci. 60:2106.

Lyle, G.R., and R.E. Moreng, 1968. Elevated environmental temperature and duration of post exposure ascorbic acid administration. Poultry Sci. 47:410.

March, B.E., M. Amin, 1981. Dietary limestone versus extra-dietary oyster shell as calcium supplements to different layer diets. Poultry Sci. 60:591-597.

Mc Loughlin, C.P., and J.H. Jr. Soares, 1976. A study of the effects of 25-hydroxycholecalciferol and calcium source on egg shell quality. Poultry Sci. 55:1400-1410.

McNally, E.H., 1965. The relationship of egg shell weight to cracked eggs. Poultry Sci. 44:1513-1518.

Mehner, A., and H.G. Torges, 1967. The effect of force moulting and of different temperatures on body weight, laying performance and egg quality of Leghorn pullets. World's Poultry Sci J. 23:261.

Meyer, R., R.C. Baker, and M.L. Scott, 1973. Effects of hen egg shell and other calcium sources upon egg shell strength and ultrastructure. Poultry Sci. 52:949-955.

Miller, P.C., and M.L. Sunde, 1975. The effects of precise constant and cyclic environments on shell quality and other lay performance factors with Leghorn pullets. Poultry Sci. 54:36-46.

Mongin, P., 1968. Role of acid-base balance in the physiology of egg shell formation. World's Poultry Sci. J. 24:200-230.

Mongin, P., and L. Lacassagne, 1966. Rythme respiratoire et physiology de la formation de la coquille de l'oeuf. Annls. Biol. Anim. Biochim. Biophys. 6:101-111.

Mueller, W.J., 1959. The effect of environmental temperature and humidity on the calcium balance and serum calcium of laying pullets. Poultry Sci. 38:1296-1301.

Nathan, D.B., E.D. Heller, and M. Perke, 1976. The effect of short heat stress upon leucocyte count. Plasma

corticosterone level, plasma and leucocyte ascorbic acid content. Br. Poultry Sci. 17:481.

N.R.C., 1984. Nutrient requirements of poultry. National Academy of Science. National Research Council U.S.A., Washington, DC.

Oliver, J.C., and G.P. Jr. Birrenkott, 1982. Response of Broilers to hyperthermic stress following treatment with a new cyclo-oxygenase inhibitor. Poultry Sci. 61:1069-1072.

Oluyemi, J.A., and A. Adebanjo, 1979. Measures Applied to combat thermal stress in poultry under practical tropical environment. Poultry Sci. 58:767-773.

Ousterhout, L.E., 1981. The effects of phased feeding protein and calcium on egg weight and shell quality with four strains of white Leghorn hens. Poultry Sci. 60:1036-1042.

Pardue, S.L., J.P. Thaxton, and J. Brake, 1984. Plasma Ascorbic acid concentration following ascorbic acid loading in chicks. Poultry Sci. 63:2492-2496.

Pardue, S.L., J.P. Thaxton, and J. Brake, 1985. Role of ascorbic acid in chicks exposed to high environmental temperature. J. of Applied Physiology 58:1511.

Parker, J.T., and M.A. Boone, 1971. Thermal stress effects on certain blood characteristics of adult male turkeys. Poultry Sci. 50:1287-1295.

Parsons, A.H., and G.F. Combs, 1981. Blood ionized calcium cycles in chicken. Poultry Sci. 60:1520.

Pastro, K.R., B.E. March, and J. Biely, 1969. Body temperature of chicks in response to lysine deficiency. Canadian J. of Physiology and Pharmacology 47:339-342.

Peterson, C.F., 1965. Factors influencing egg shell quality. World's Poultry Sci. J. 21:110-138.

Polin, D., and P.D. Sturkie, 1955. Prevention of premature oviposition and shell-less eggs with ephedrine. Poultry Sci. 34:1169-1170.

Polin, D., and P.D. Sturkie, 1957. The influence of the parathyroids on blood calcium levels and shell deposition in laying hens. Endrocrinology 60:778-784.

Potts, P.L., and K.W. Washburn, 1977. The effect of

supplemental calcium and vitamin D on line and strain differences in egg shell strength. Poultry Sci. 56:1067-1072.

Quinn, J.P., A.W. Brant, and C.H. Jr. Thompson, 1956. Effect of a naturally occurring outbreak of Newcastle disease on egg quality and production. Poultry Sci. 35:3-10.

Randall, W.C., 1943. Factors influencing the temperature regulation of Birds. The Amer. J. of Physiol. 139:56-63.

Randall, W.C., and W.A. Hiestand, 1939. Panting and temperature regulation in the chicken. The Amer. J. of Physiol. 127:761-767.

Robertson, R.H., and D.W. Francis, 1965. Egg quality factors as affected by calcium, ascorbic acid and reserpine levels of the ration. Poultry Sci. 44:1409-1410.

Robertson, R.H., D.W. Francis, and M.D. Finkner, 1964. Egg quality factors as affected by calcium, ascorbic acid and reserpine levels of the ration. Poultry Sci. 43:1356-1357.

Roland, D.A. Sr., 1972. Cause and prevention of poor egg shell quality. 31st Ann. Florida Paul. Inst. Proc.:13-17. (Poultry Sci. 52:949, 1973).

Roland, D.A. Sr., 1979. Factors influencing shell quality of aging hens. Poultry Sci. 58:774-777.

Roland, D.A. Sr., 1980. The ability of young and old hens to change shell deposition with sudden natural drastic changes in egg size. Poultry Sci. 59:924-926.

Roland, D.A. Sr., D.R. Sloan, and R.H. Harms, 1973. Calcium metabolism in the laying hen. 4. The calcium status of the hen at night. Poultry Sci. 52:351.

Roland, D.A. Sr., D.R. Sloan, and R.H. Harms, 1975. The ability of hens to maintain calcium deposition in the egg shell and egg yolk as the hen ages. Poultry Sci. 54:1720-1723.

Roland, D.A. Sr., C.E. Putman, and R.L. Hilburn, 1978. The relationship of age on ability of hens to maintain egg shell calcification when stressed with inadequate dietary calcium. Poultry Sci. 57:1616-1621.

Rossoff, I.S., 1974. Handbook of veterinary drugs. New York: Springer Publishing Company.

Sauveur, B., and P. Mongin, 1971. Etude comparative du fluide uterin et de l'albumen de loeuf in utero chez la poule. Ann. Biol. Anim. Biochem. Biophys. 11:213-224.

Savage, J.E., C.W. Turner, H.L. Kempster, and A.G. Hogan, 1952. The effects of vitamin B and thyroprotein or egg production, egg weight, shell quality and hatchability. Poultry Sci. 31:22-31.

Siegal, P.B., J.H. vanMiddelkoop, and P.R.K. Reddy, 1978. Comparisons of frequencies and egg shell characteristics of broken and intact eggs within divers populations of chickens. Br. Poultry Sci. 19:411-416.

Simkiss, K., 1961. Calcium metabolism during avian reproduction. Biol. Rev. 36:321-367.

Simons, P.C.M., 1985. Egg shell quality can sometimes be improved. Poultry Misset Jan. 6-9.

Soares, J.H. Jr., M.A. Ottinger, and E.G. Buss, 1988. Potential Role of 1,25 Dihydroxycholecalciferol in Egg shell calcification. Poultry Sci. 67:1322-1328.

Steel, R.G.D., and J.H. Torrie, 1980. Principles and procedures of statistics, second edition. New York: McGraw-Hill Book Company.

Stiles, P.G., and L.E. Dawson, 1961. The effect of physical disturbance, sound and light, on the incidence of blood spots and other egg quality factors. Poultry Sci. 40:189-195.

Sturkie, P.D., 1986. Avian Physiology, 4th edition. New York: Springer-Verlag.

Sturkie, P.D., H.S. Weiss, and R.K. Ringer, 1954. Effects of injections of acetylcholin and ephedrine upon components of the hens egg. Poultry Sci. 33:18-24.

Sykes, A.H., 1983 a. Heat stress and acclimatization. World's Poultry August:12-13. (World's Poultry Sci. 43:33, 1987).

Sykes, A.H., 1983 b. Heat stress and heat acclimatization of poultry. Australian Poultry Husbandry Research Foundation, Camden, N.S.W. (World's Poultry Sci. 43:33, 1987).

Tanor, M.A., S. Leeson, and J.D. Summers, 1984. Effect of heat stress and diet composition on performance of white Leghorn hens. Poultry Sci. 63:304-310.

Taylor, T.G., 1970. How an egg shell is made. Scientific Amer. 222:88.

Taylor, T.G., and F. Hertelendy, 1961. Changes in the blood calcium associated with egg shell calcification in the domestic hen. 2. Changes in the diffusible calcium. Poultry Sci. 40:115-123.

Taylor, T.G., T.R. Morris, and F. Hertelendy, 1962. The effect of pituitary hormones on ovulation in calcium deficient pullets. Vet. Rec. 74:123. (Avian physiology, 4th edition. New York: Springer-Verlag, 1986).

Teeter, R.G., M.O. Smith, F.N. Owens, S.C. Arp, S. Sangiah, and J.E. Breazile, 1985. Chronic heat stress and respiratory alkalosis: occurrence and treatment in broiler chicks. Poultry Sci. 64:1060-1064.

Thomas, J.M., H.S. Nakaue, and B.L. Reid, 1966. Effect of increasing dietary levels of acetyl-salicylic acid on performance and cecal microbial counts of white Leghorn pullets. Poultry Sci. 45:1313-1317.

Thornton, P.A., 1960. The effect of dietary calcium level on the efficiency of ascorbic acid in maintenance of egg shell thickness at increased environmental temperatures. Poultry Sci. 39:1401-1406.

Thornton, P.A., 1962. The effect of environmental temperature on body temperature and oxygen uptake by the chicken. Poultry Sci. 41:1053-1060.

Thornton, P.A., and S.S. Deeb, 1961. The influence of thyroid regulators on blood ascorbic acid levels in the chicken. Poultry Sci. 40:1063.

Vasquez, R., and R.G. Teeter, 1986. Feed intake effects on egg quality of nonstressed and heat stressed layers. Animal Sci. Research Report, Oklahoma Agricultural Experiment Station MP-118:77-81. (Nut. Abs. and Rev. 57:53, 1987).

Wells, R.G., 1967. Egg shell strength. I. The relationship between egg breakage in the field and certain laboratory assessments of shell strength. Br. Poultry Sci. 8:131-139.

Wilkinson, W.S., 1961. Effects of level and source of calcium and the addition of antibiotics, vitamin C, zinc and reserpine in laying hen rations. Poultry Sci. 40:1470.

Winget, C.M., A.H. Smith, and G.N. Hoover, 1958. Arterio-venous differences in plasma calcium concentration in the shell gland of the laying hen during shell formation. Poultry Sci. 37:1325-1328.

Winget, C.M., and A.H. Smith, 1959. Dissociation of the calcium protein complexe of laying hen's plasma. Am. J. Physiol. 196:371-374.

Wolfenson, D., Y.F. Frei, N. Snapir, and A. Berman, 1979. Effect of diurnal or nocturnal heat stress on egg formation. Br. Poultry Sci. 20:167-174.

Yannakopoulos, A.L., and T.R. Morris, 1979. Effect of light, vitamin D and dietary phosphorus on egg shell quality late in the pullet laying year. Br. Poultry Sci. 20:337-342.

APPENDIX

APPENDIX

Table A.1. LSD analysis of the change in
a. egg weight due to 6 hours exposure to heat stress (35°C) at day time and during egg shell formation (night time).

4	3	2	1	AV. Change	Treat No.
0.397	-0.656	-0.863	-1.899	in egg wt.	
	(1.913)	(2.068)	(2.0197)	0.397	4
	1.053	1.260	+2.296		
		(1.846)	(1.791)	-0.656	3
		0.207	1.243		
			(1.956)	-0.863	2
			1.036		
				-1.899	1

Table A.1.b. LSD analysis of the change in shell weight

2	1	4	3	AV. Change	Treat No.
0.03	-0.283	-0.707	-0.99	in shell wt.	
	(0.403)	(0.426)	(0.380)		
	0.313	+0.737	+1.02	0.03	2
		(0.416)	(0.369)		
		+0.424	+0.707	-0.283	1
			(0.394)		
			0.283	-0.707	4
				-0.99	3

Note: treat 1 = heat at day time
treat 2 = no heat at day time
treat 3 = heat at night time
treat 4 = no heat at night time
\+ = significant ($p < 0.05$)
( ) = LSD value at 5% level of significant

Table A.1.c. LSD analysis of the change in % shell

2 0.181	1 -0.142	4 -1.259	3 -1.577	AV. Change in % shell wt.	Treat No.
	(0.694)	(0.734)	(0.655)		
	0.323	+1.44	+1.758	0.181	2
		(0.717)	(0.636)		
		+1.117	+1.435	-0.142	1
			(0.679)		
			0.318	-1.259	4
				-1.577	3

Table A.1.d. LSD analysis of the change in mg Ca/mm^2

2 0.0051	1 -0.0076	4 -0.040	3 -0.051	AV. Change in Ca/mm^2	Treat No.
	(0.0219)	(0.0231)	(0.0206)		
	0.0127	+0.0451	+0.0561	0.0051	2
		(0.0226)	(0.0200)		
		+0.0324	+0.0434	-0.0076	1
			(0.0191)		
			0.011	-0.04	4
				-0.051	3

Table A.1.e. LSD analysis of the feed intake (gm) within 24 hours following the stay in the chamber.

2 85.22	1 73.6	4 73.25	3 65.62	AV. of Feed. Intake on 24hrs. following the stay in the chamber	Treat No.
	(0.328) +11.62	(0.347) +11.97	(0.310) +19.60	85.22	2
		(0.339) +0.35	(0.300) +7.98	73.6	1
			(0.321) +7.63	73.25	4
				65.62	3

Table A.1.f. LSD analysis of the change (°C) in body temperature

1 0.79	3 0.73	2 0.11	4 -0.45	AV. Change in body temp.	Treat No.
	(0.266) 0.06	(0.290) +0.68	(0.299) +1.24	0.79	1
		(0.273) +0.62	(0.284) +1.18	0.73	3
			(0.307) +0.56	0.11	2
				-0.45	4

Table A.2. Analysis of variance (3 factors) of the change in a. egg weight due to providing feed and/or water during heat stress (35°C) or no heat for 4 hours at the time egg shell formation is occurring.

Source:	df:	Sum of Squares:	Mean Square:	F-test:	P value:
Water (A)	1	.679	.679	.135	.7146
Feed (B)	1	9.0300E-6	9.0300E-6	1.7965E-6	.9989
AB	1	4.701	4.701	.935	.3377
Temperature (C)	1	.123	.123	.024	.8764
AC	1	2.585	2.585	.514	.4763
BC	1	7.14	7.14	1.421	.2383
ABC	1	.164	.164	.033	.8575
Error	56	281.474	5.026		

Table A.2.b. Analysis of variance of the change in shell weight.

Source:	df:	Sum of Squares:	Mean Square:	F-test:	P value:
Water (A)	1	.735	.735	4.481	.0387
Feed (B)	1	.034	.034	.208	.6499
AB	1	1.301	1.301	7.936	.0067
Temperature (C)	1	.911	.911	5.557	.0219
AC	1	.01	.01	.058	.8101
BC	1	.061	.061	.374	.5432
ABC	1	1.3125E-4	1.3125E-4	8.004E-4	.9775
Error	56	9.183	.164		

Table A.2.c. Analysis of variance of the change in % shell.

Source:	df:	Sum of Squares:	Mean Square:	F-test:	P value:
Water (A)	1	1.294	1.294	3.691	.0598
Feed (B)	1	.313	.313	.892	.349
AB	1	3.773	3.773	10.762	.0018
Temperature (C)	1	1.826	1.826	5.209	.0263
AC	1	9.6914E-5	9.6914E-5	2.7646E-4	.9869
BC	1	.461	.461	1.316	.2561
ABC	1	4.676E-3	4.676E-3	.013	.9085
Error	56	19.631	.351		

Table A.2.d. Analysis of variance of the change in mg Ca/mm^2.

Source:	df:	Sum of Squares:	Mean Square:	F-test:	P value:
Water (A)	1	1.288E-3	1.288E-3	3.671	.0605
Feed (B)	1	2.274E-4	2.274E-4	.604	.4405
AB	1	4.053E-3	4.053E-3	10.755	.0018
Temperature (C)	1	2.278E-3	2.278E-3	6.032	.0172
AC	1	1.2467E-5	1.2467E-5	.033	.8563
BC	1	3.7519E-4	3.7519E-4	.996	.3227
ABC	1	2.2575E-8	2.2575E-8	5.9905E-5	.9939
Error	56	.021	3.7685E-4		

Table A.2.e. Analysis of variance of the feed intake (gm) within 24 hours following the stay in the chamber.

Source:	df:	Sum of Squares:	Mean Square:	F-test:	P value:
Water (A)	1	1852.758	1852.758	9.035	.004
Feed (B)	1	11513.996	11513.996	76.904	1.0E-4
AB	1	1401.544	1401.544	9.361	.0034
Temperature (C)	1	364.589	364.589	2.435	.1243
AC	1	140	140	.935	.3377
BC	1	838.355	838.355	5.6	.0214
ABC	1	202.846	202.846	1.355	.2494
Error	56	8384.286	149.719		

Table A.2.f. Analysis of variance of the change (^{o}C) in body temperature.

Source:	df:	Sum of Squares:	Mean Square:	F-test:	P value:
Water (A)	1	4.578	4.578	15.292	3.0E-4
Feed (B)	1	.254	.254	.851	.3602
AB	1	.293	.293	.979	.3267
Temperature (C)	1	23.423	23.423	78.332	1.0E-4
AC	1	.583	.583	1.948	.1683
BC	1	.232	.232	.776	.3823
ABC	1	.16	.16	.536	.4673
Error	56	16.746	.299		