
CHAPTER 2

LITERATURE REVIEW

2.1. Physiological Parameters

2.1.1. Rectal Temperature

Rectal temperature of the birds is mostly affected by rate of heat loss from the body, feed intake, nature of feed, feeding schedules and acclimatization. The results of Wilson (1949) indicated that the body temperature of adult chickens also varied with the environmental temperature. King (1956) reported that young chickens brooded at an environmental temperature of 40.5°C had higher rectal temperature than chicks reared at 32.2°C. Ward and Peterson (1973) found that rectal temperature was higher (42 °C) in broilers exposed to 33-35°C, than in birds maintained at 18-22°C (41°C). Ching and Ching (1992) reported elevated rectal temperature of broilers kept under 38°C of ambient temperature. High ambient temperature increased the rectal temperature (Deyhim and Teeter 1994; Salvador *et al.*, 1999). There was a significant increase in rectal temperature of birds kept under 90% relative humidity compared with 30 and 60% relative humidity groups (Gu *et al.*, 1999). Whereas no significant differences were observed in rectal temperature among humidity treatments when birds were kept under 30°C ambient temperature. Broiler chickens exposed to 39±1°C for 2 h at 44 d of age showed increased rectal temperatures (Altan *et al.*, 2000).

Body temperature was found to be reduced by at least 1°F in broilers exposed to high temperature by feed deprivation for 12 hours daily (Smith and Teeter, 1988). Feed and light restriction reduced the rate of increase of rectal temperature at 35-40°C (Francis *et al.*, 1991). Full fed chickens compared to feed withdrawal group showed significantly higher body temperature during acute heat

stress (Smith, 1992). Macleod *et al.* (1993) assessed the impact on thermoregulation in broiler breeder fowls which were fed freely, fed once daily, and fed recommended doses of feed and showed 0.7°C higher rectal temperature in all the free fed chickens at 23 and 28°C temperature. Other groups also indicated elevated rectal temperature but less than full fed birds. Teeter and Belay (1996) recommended fasting/starvation, a method for lowering the rectal temperature in broilers during heat stress.

Lott (1991) acclimatized broilers to 24°C, 35°C, 24°C cyclic temperature for 3 consecutive days and after acclimation, exposed them to a linear change in temperature from 24 to 41°C over 3 h with a constant 10°C dew point, starting at 0800 h on day 34 for trial 1 and on day 41 for trial 2. For the feed group, feed was removed from the birds at 0400 h and placed again at 0700 h. For the no feed group, feed was removed at 0700 h. whereas at 0800 h, feed was removed from all birds but water was accessible throughout heat exposure. Acclimated and unacclimated broilers given access to feed had similar body temperatures, but the acclimated broilers not receiving feed had a significantly lower final rectal temperature than the unacclimated group.

Acclimatized birds (24 to 35°C) had a high rectal temperature (42.3 versus 41.2°C) when housed at 24°C and a lower rectal temperature (44.2 versus 44.6°C) when exposed to 35°C than the unacclimatized controls. Rectal temperature in the 24°C and 35°C environments increased linearly. Acclimatized birds exhibited increased body temperature when housed in thermoneutral environment and lower body temperature when exposed to high ambient temperature distress. Heat dissipation mechanisms require greater than 12 h to restore normal body temperature; or the bird's set point was elevated when exposed to cycling high ambient temperature distress (Teeter *et al.*, 1992). Inoue *et al.* (1995) reported the effects of short time heat exposure on rectal temperature (T_r). In experiment 1, broilers were exposed to 38°C for 3 h everyday for 7 days. Rectal temperature and leg-shank skin surface temperature (T_s) also increased more rapidly and was higher

after the 2nd day, but their stable levels became slightly lower on the 7th day. In experiment 2, broilers acclimated in experiment 1 and unacclimated broilers were exposed to 41°C for 3 h. Both acclimated and unacclimated broilers, Tr and Ts increased with increase in exposure time.

From the above information it can be concluded that increase in the rectal temperature may be related to environmental temperature and relative humidity. On the other hand feeding practices and acclimatization also affect the thermoregulation physiology of the broilers. Studying the interaction of these parameters may help to find strategies in order to reduce thermal load on the birds and improve their performance.

2.1.2. Respiration Rate

Respiration rate of birds is affected by various factors such as age, breed, sex, acclimatization and environmental conditions particularly ambient temperature and relative humidity. The measurements of respiration rate in White Leghorn x Rhode Island crossbred hens kept in cages were taken at 6 air temperatures at equal intervals between 2°C and 32°C ($\pm 1^\circ\text{C}$) (Arieli *et al.*, 1980). Response of respiratory rate to air temperature in the two acclimatization groups at 2°C to 18°C air temperature was 30 to 35 respiration per minute whereas, in summer acclimatized hens exposed to 26-32°C was 5 fold higher than basal value.

Pure breeds and crosses of different strains have been shown to differ in physiological manifestations in response to environmental variations but in either case increased environment temperature resulted in increased respiration. Physiological responses to the temperature of 40-45°C in a new breed of fowl (Leghorn x Bedouin) indicated that respiratory rate increased from a normal rate of 11-26 breaths per minute to 238 and 293 breaths per minute at 42°C and 45°C, respectively. However, normal respiration rates were observed that at ambient temperature upto 35°C (Arad *et al.*, 1975). Arad and Marder (1982) studied the thermoregulation of Sinai and its crosses with other breeds of fowl. Sinai breed exhibited a shallow high frequency panting compared to the others. Zhou and

Yamamoto (1997) reported rapid increase in respiration due to exposure of broilers to high temperature (38°C).

Adaptation reduced respiration rate in desert Sinai and White Leghorn fowls. Complete acclimatization in respiration rate required 7 to 10 days in white and brown strains of layers (Sykes and Al-Fataftah, 1981) whereas the mean daily respiration rate was higher at 35°C than at 23°C in commercial laying hens. Inoue *et al.* (1995) exposed broilers to 38°C for 3 h everyday for 7 days in experiment 1, and observed that respiration rate increased rapidly after the exposure, with the rate being higher after the 2nd day compared to the 1st day. Whereas in experiment 2, broilers acclimated in experiment 1 and unacclimated broilers were exposed to 41°C for 3 h. The start of panting was earlier and reached a higher level in acclimated broilers as compared to unacclimated broilers.

Garcia *et al.* (1992) conducted study on seven groups of 10 (5 male and 5 female) 7-week-old fowls: group I and II (control) subject to normal rearing conditions; III, deprived of food for 20 h; IV, exposed to 40°C for 2 h; V, exposed to 40°C for 4 h; VI, food-deprived for 20 h and exposed to 40°C for 2 h; VII, food-deprived for 20 h and exposed to 40°C for 4 h. They found that food-deprived birds had smaller changes in respiration rate than those subjected to normal rearing conditions.

2.2. Physical Parameters

2.2.1. Weight Gain/Body Weight

High ambient temperature adversely affects the weight gain in poultry birds (Smith and Oliver, 1972; Swain and Farrell, 1975; Sabry *et al.*, 1978; Jovanovic *et al.*, 1995; Deyhim *et al.*, 1995; Yahav *et al.*, 1996; Sands and Smith, 1999). Body weights were lower at high temperature in the studies of Elahi *et al.* (1985); Pardue *et al.* (1985); Li *et al.* (1986); Nakamura *et al.* (1992); Macleod *et al.* (1993); Njoya and Picard, (1994); Geraert *et al.* (1996). The differences in weight gain between birds in cooled and uncooled houses have been reported to range from 101.5 g to 385 g at different densities (Al-Zujajy *et al.*, 1978). Body

weight and weight gain in early heat-stressed birds were not affected when they were exposed to heat stress later in life (Arjona *et al.*, 1988). May (1995) also found that early heat exposure (36.1 °C for 24 h) in environmental chambers did not affect body weight gain under later heat exposure to 37.8°C for 6.5 h in trial 1 at 47 days and 8 h in trial 2 at 42 days. Blood alkalosis limits growth rate of broiler chicks reared under chronic heat stress (32°C), however, respiratory alkalosis and weight gain depressions can partially be alleviated by feeding management (Teeter *et al.* 1985).

Datta *et al.* (1984) reported that group of broilers provided with a cooler environment during hot and dry season exhibited the highest body weight. Lacey (1996) introduced a cooling system during summer with the help of foggers in curtain-sided broiler houses which proved very useful for the heat loss, and were economical as well, on the basis of more weight gain. Srivastava *et al.* (1980) compared the efficiency of cooling systems such as exhaust fan, fogging system plus a ceiling fan and an evaporative cooling system with no cooling system regarding the performance of broilers during summer months. Body weight was found maximum in the evaporative cooling system and the lowest in control group in trials conducted under hot and dry as well as hot and humid atmospheres. Broilers with air coolers or foggers showed significantly higher body weight over the control (Sharma and Gangwar, 1985). Cooling efficiency declined in the order, air cooler, fogger and exhaust fans with better overall efficiency in the dry as opposed to humid part of the summer. Hence there is a need to reduce heat production instead of evaporative cooling during summer when temperature and humidity both rises.

Feed withdrawal during heat stress did not show any adverse effect on weight gain (Smith, 1992; Spinu and Degen, 1993) or body weight (Savic *et al.*, 1993; Zakia *et al.*, 1995) in birds. Smith (1994) reported that 23 h light (L): 1 h dark (D) or 16L:8D had no effect on body weight gain in broilers from 22 to 49 days of age. Wong *et al.* (1993) found non significant difference between body weight of continuous (23L:1D) and intermittent (1L:3D) lighting schedules for birds at four

and seven weeks of age. Similar findings were reported by Zakia *et al.* (1995) who reported that food and light withdrawal had no effect on body weight gain. Zhong *et al.* (1995) found no difference in mean body weights between full fed and restricted males and combined broiler at 49 day of age. Females subjected to skip-a-day feeding from 7-13 d of age fully recovered in body weight irrespective of dietary protein fed, during the 1st 7 days after skip-a-day feeding (Santoso, 1995). Plavnik (1998) reported that early growth restriction did not affect weight gain. Whereas, Fontana *et al.* (1992) found that early feed restriction in broilers resulted in significantly ($P<0.05$) lower mean body weight than controls. Deaton (1995) determined the effect of early feed restriction on broiler performance, neither females nor males could overcome a 17 or 18% weight reduction. Renden *et al.* (1996) applied four lighting treatments on broilers, T₁ (23L: 1D), T₂ (16L: 2D: 1L:2D), T₃ (23L: 1D for day 1-7 and 16L:8D for day 8 to 14) with light period increased by 2 hr/week during days (15 to 35) and T₄ (23L:1D for days 1 to 7, 16L:8D for day 8 to 14, 16L: 3D: 2L: 3D for days 15 to 21, 16L: 2D: 4L: 2D for day 22 to 28, 16L: 1D: 6L: 1D for days 29 to 35 and 23L:1D there after) were provided. Body weight was greater in T₁ as compared to T₃ and T₄ at 21 days.

Gonzales *et al.* (1998b) reported the effect of quantitative feed restriction (0, 10, 20, 30, 40 and 50%) from 8 until 14 days post-hatching on performance of male broilers during winter and summer. Feed restricted birds showed compensatory gain only until 3 weeks after restriction. At 49 days, body weight and weight gain was lower in feed-restricted birds, but feed:gain ratio was improved than birds fed *ad libitum*. Su *et al.* (1999) conducted two trials. In the 1st trial, 2686 broilers were offered 2, 3 or 4 meals per day or consumed feed *ad libitum* from 5 to 34 days of age. In trial 2 (n = 2846), birds were restricted to achieve 25, 50 or 75% of predicted growth for 5 or 7 days, starting at 5, 7 or 9 days of age. *Ad libitum*-fed birds served as controls. In trial 1, fewer meals per day resulted in lower body weight. In trial 2, earlier restriction, and longer duration and more severe level of restriction were all associated with lower body weight. Therefore, it can be concluded that weight gain

is directly related to the method (feed withdrawal, intermittent feeding, skip a day feeding, two or three meals per day, decreasing the energy or protein levels in the poultry ration) and duration of feed restriction.

Fat supplementation improved the weight gain (Rotter *et al.*, 1987) in broilers. Birds fed highest energy level showed the most rapid weight gain when dietary energy levels (2600, 2900, and 3200 Kcal ME/kg) in isonitrogenous diets were fed to broilers (Junqueira *et al.*, 1999). Oliveira *et al.* (2000) found that weight gain linearly improved as dietary metabolizable energy increased. Korver *et al.* (1998) in their first trial fed chickens on one of 5 diets from 3 days of age: a low-energy diet containing 7% cellulose (ME 2714 Kcal/kg), or high-energy diets containing about 7% of tallow, maize oil, safflower oil or fish oil (3302 Kcal/kg each). Whereas in a second experiment, 3-day-old chickens were fed on one of 8 isoenergetic diets containing tallow as the sole supplemental fat source, or tallow plus 2% maize oil, 1, 1.5 or 2% fish oil, or fish meal providing 1, 1.5 or 2% supplemental oil. Fish oil at 7% of the diet did not improve weight gain. Fish oil diets improved weight gain in experiment 2 relative to the other diets. However, no difference between weight gain among the treatments was observed when four levels of dietary flax seed oil in combination with animal tallow were fed to give a total of 6% added fat in the diets (Olomu and Baracos, 1991). Latour *et al.* (1994) found that broiler chicks fed nonisocaloric diets with 0, 3 or 7% added lard showed non-significant difference in their body weights. Peebles *et al.* (1997) reported that body weight was not affected by diet when broiler chickens were provided either 0, 3, or 7% added lard in starter diets through 10 d of age (S1), followed by either 3 or 7% added dietary lard through 21 d of age (S2). As scientists have observed contradictory findings regarding supplementation of fat in ration, therefore response of broilers fed fat supplemented ration during summer was needed to be addressed.

2.2.2. Feed Consumption

Heat stress has been reported to depress the feed intake of chickens (Bray and Gesell, 1961; Swain and Farrell, 1975; Andrade *et al.*, 1976; Cowan and

Michie, 1978; Daniel and Balnave, 1981 and Tanor *et al.*, 1984). High ambient temperature reduced feed intake in broilers (Kutlu and Forbes, 1993; Jovanovic *et al.*, 1995; Sands and Smith, 1999). Correlations between body temperature and gain, feed consumption, and feed conversion ratio were negative and non-significant for most periods in the 21°C environment. In the 32°C environment, body temperature was significantly correlated with gain, feed consumption, and feed conversion ratio after 7 d of heat stress (Cooper and Washburn, 1998). Plavnik and Yahav (1998) also found that feed intake from 6 to 8 wk of age (after acclimatization to the various temperatures) was significantly ($P<0.05$) depressed by the high environmental temperature (35 and 25-35°C) compared to values observed at 25 and 30°C.

Feed deprivation resulted in reduced total feed intake (Smith and Teeter, 1988) in broiler. Feed consumption exacerbates bird heat content rise when environmental conditions limit sensible heat loss (Wiernusz and Teeter, 1993). It was also reported that feed consumption was elevated in broilers kept under the thermo neutral zone (24°C) than those housed at 32 to 35°C. Suprunov *et al.* (1995) distributed 240 day old broilers into 4 groups, and fed on standard balanced diets during June and August 1994. Control chicks (group 1) had free access to water and feed while group 2 had free access to water and feed in granulated form; groups 3 and 4 (from 4 days of age) were subjected to 2-h feeding periods alternating with 2-h non-feeding periods with access to water and in group 4 were subjected to 2-h feeding periods alternating with 2-h non-feeding periods with free access to water and feed in granulated form. Group 4 showed the lowest feed intake during the trial. Buyse *et al.* (1996) found a similar body weight as that of continuous light and intermittent light was applied whereas, intermittent lighting reduced feed intake. Restricted feeding adversely affected feed consumption (Bhat and Banday, 2000). Whereas, Smith, (1994) observed that photo-schedules of 23 h light (L): 1 h dark (D) or 16L: 8D had no effect on feed intake. Gonzalez *et al.*, (2000a) also reported non-significant effect of feed consumption when 0 or 25% feed reduction was

applied from 7 to 21 days. Above review showed that feed consumption varied with method of feeding used.

Birds previously acclimatized to cycling temperature heat distress (24 to 35°C) for two 24-h cycles were observed by Teeter *et al.*, (1992) to have 24% lower feed consumption than birds previously housed at 24°C and experiencing their first heat distress exposure. The results indicate that feed intake plays a significant role in the acclimatization process; that bird acclimatization to heat distress has metabolic effects independent of feed consumption; and that either the heat-distressed, acclimatized bird has the capacity to shift exothermic processes from the heat-distressed to cooler time periods; that heat dissipation mechanisms require greater than 12 h to restore normal body temperature; or the birds' set point is elevated when exposed to cycling high ambient temperature distress. Similarly Wiernuz (1998) also reported that fat addition in ration may affect the process of acclimatization regarding feed intake. High fat ration had the potential to negate acclimation effect by obligating the bird to high levels of heat production.

Olomu and Baracos (1991) studied the effect of feeding flax seed oil on the performance of broilers. Four levels of dietary flax seed oil were fed in combination with animal tallow to give a total of 6% added fat in the diets. Feed consumption was not significantly different among the treatments. Similarly, Latour *et al.* (1994) found no difference in feed consumption due to 0, 3 or 7% added lard. Whereas Junqueira *et al.*, (1999) studied the effect of three dietary energy levels (2600, 2900, and 3200 Kcal ME/kg) in isonitrogenous diets on broiler chicks. Birds fed the highest energy level showed more rapid weight gain and greater feed intake than their counterpart.

Jaffar *et al.* (1996) kept the chickens (control, group 1) under normal temperature of 21-28°C and relative humidity (RH) of 39 to 85%. Groups 2, 3 and 4 were reared at 29° to 42°C temperature and RH of 45 to 89%. Groups 1 and 2 were fed on the same diets containing ME 12.6 MJ/kg and 22 and 20% CP during the starting and finishing periods of rearing, respectively. Group 3 was fed on a high-

energy and high-protein diet with ME 13.5 and 13.0 MJ/kg, and 24 and 23% CP, in the starting and finishing periods. The starter and finisher diets for group 4 contained 24.0 and 23.5% CP and ME 14.0 and 13.5 MJ/kg. Feed intake was 3123, 3060, 2890 and 2760 gm per bird in groups 1,2,3 and 4 respectively. Results indicate that feed consumption decreased with high ambient temperature whereas at high ambient temperature, increasing the energy content of diet further decreased the feed intake referring towards reduced energy requirements of bird during high ambient temperature. This indicates that bird's feed intake is dependent on its energy requirements.

2.2.3. Water Consumption

High ambient temperature has been known to increase water intake of birds (Khadi *et al.*, 1988; Deyhim and Teeter, 1991; Kutlu and Forbes, 1993). Water acts as heat sink to lower the body temperature (Smith and Teeter, 1989). Heat stressed birds dissipate over 80% of their heat production via evaporative cooling (Wiernusz and Teeter, 1993).

May and Lott (1992) reared the broilers on litter and housed at a constant 24°C for several days before cyclic temperatures were started. Broilers were then maintained at 24°C (control) or with a 24-35-24°C cycle for 3 days. Broilers that were 5, 6 or 7 weeks old consumed as much feed or water on day 1 of the cycle as on the succeeding days. Water intake increased during the 12 h when the temperature was maximum. At 7 weeks, water intake was greater for broilers on the cyclic temperature for each 6-h period except for the period of temperature decrease immediately preceding the minimum temperature. The birds exposed to the 3 days of cyclic temperatures consumed more water than controls during a subsequent exposure to temperatures up to 40.8°C. The data showed that the increased water intake and decreased feed intake observed due to high cyclic temperatures arose from changes that occurred during some times of the day and no changes occurred during other times.

Lott (1991) studied the effect of feed intake on water consumption of acclimated and unacclimated broilers during heat exposure. Male broiler chicks, 100 per trial, were raised as one group to 29 days for Trial 1 and 36 days for Trial 2. The birds were moved to environmental chambers and 50 per trial were acclimated by being subjected to 3 consecutive days of 24°C, 35°C, 24°C cyclic temperature. After acclimation, the birds were exposed to a linear change in temperature from 24 to 41°C over 3 h with a constant 10°C dew point, starting at 0800 h on Day 34 for Trial 1 and on Day 41 for Trial 2. The feeding schedule for the day of the heat exposure was as follows. For the feed group, feed was removed from the birds at 04:00 h and replaced at 07:00 h. For the no feed group, feed was removed at 07:00 h. At 0800 h, feed was removed from all birds but water was accessible throughout heat exposure. Acclimated and unacclimated birds given access to feed for 1 h before a heat exposure consumed 60 and 50 ml of water per broiler, respectively, during the heat exposure. However, for broilers not receiving feed, the water consumption was 58 and 30 ml per broiler, respectively.

Above results indicate that temperature, feeding management, and acclimatization affects the water intake as well as process of thermoregulation of broilers hence study of water consumption may be helpful to understand birds responses towards feeding trials during summer.

2.2.4. Feed Efficiency

It is well documented that high ambient temperature adversely affects the efficiency of feed utilization in birds (Plavnik and Yahav, 1998; Temim *et al.*, 2002). Whereas short sub-lethal heat stress did not affect feed efficiency (Ernst *et al.*, 1984). Air cooler improved feed conversion efficiency by 4.3 to 9.7 percent (Wang *et al.*, 1995). Al-Zujajy *et al.* (1978) reared chicks for 56 days in two broiler houses. One house was provided with two air coolers. Feed consumption in the cooled (21.2 - 29.5°C) house and in hot, dry conditions (30.1 - 39.9°C) was studied. Feed utilization was clearly better in cooled house than under conventional conditions. Feed efficiency from 6 to 8 weeks of age (after acclimatization to the

various temperatures) was significantly ($P < 0.05$) depressed by the high temperature (35 and 25-35°C) compared with values observed at 25 and 30°C (Plavnik and Yahav, 1998). Thermal conditioning slightly enhanced the feed efficiency (Basilio *et al.*, 2001). As discussed above it is clear that high ambient temp reduces feed efficiency, which can be controlled by cooling the air, but some times it becomes more expensive to use cooling devices.

Broilers subjected to time limited feeding and receiving feed for 5 hours daily during cooler hours showed better-feed conversion ratios than those of control group. Early feed restriction improves the feed conversion (Deaton, 1995). Fontana *et al.* (1992) induced feed restriction in all experimental groups by providing chicks with 40 Kcal ME/bird/day, commencing at four days of age. Male chicks were fed restricted diet for 7 (Experiment 1 and 2) or six days (Experiment 3 and 4). Whereas, broiler females received restricted diets for five days (Experiment 1). *Ad libitum* feeding was resumed after the feed restriction periods, and continued through the conclusion of the experiments at 49 (Experiment 1 and 2) or 28 (Experiment 3 and 4) days of age. Broilers provided *ad libitum* access to feed for the entire experimental period served as the controls in each study. Feed conversion ratios for feed restricted broilers were significantly lower at 28 (Experiments, 1 through 4) and 49 (Experiments 1 and 2) days of age than for birds consuming feed *ad libitum*. Buyse *et al.* (1996) reported that broilers under intermittent lighting showed improved feed efficiency than those kept under continuous lighting. Zulkifli and Fauzi (1996) found that feed restricted chickens (9-17 hrs) showed improved feed conversion efficiency than those fed *ad libitum* in response to the high ambient temperature (36°C from 12-170 hrs). Lower feed conversion ratio for female and combined sex broilers for the restricted than full fed broilers was reported by Zhong *et al.* (1995). Whereas Sheila *et al.* (1993) found that broilers on restricted feeding programs exhibited compensatory growth at 21, 35 and 45 days of age equivalent to *ad libitum* programs. Moreover, feed efficiency was not affected in feed restricted (1.68) and *ad libitum* fed (1.73) broilers.

Olomu and Baracos (1991) studied four levels of dietary flax seed oil in combination with animal tallow to give a total of 6% added fat in the diets and found that feed efficiency was not significantly different among treatments. Peebles *et al.* (1997) studied the effect of 0, 3, or 7% added lard, in starter diets through 10 d of age. They found no effect on feed conversion ratio due to treatments between 21 and 42 d when birds were fed 3 or 7% added lard in the diet. Korver *et al.* (1998) fed the chickens isoenergetic diets containing tallow as the sole supplemental fat source, or tallow plus 2% maize oil, 1, 1.5 or 2% fish oil, or fish meal providing 1, 1.5 or 2% supplemental oil. It was observed that fish oil diets improved feed efficiency. Crespo and Esteve-Garacia (2001) reported that broilers fed sunflower and linseed oils presented better values of feed efficiency. Above review can be summarized in a way that feed efficiency is associated with environmental temperature, level of restriction and composition of feed regarding its energy and protein ratio and method of restriction.

2.3. Protein and Energy Requirements

Metabolizable energy (ME) requirements decrease with increasing temperature above 21°C (Daghir, 1995) as a result of reduced requirements of maintenance because energy requirements during summer are reduced up to 15 % than winter. Use of high energy ration for broilers has become quite common in warm regions. However, this practice should be accompanied by raising the level of the most critical amino acids (McNaughton and Reece, 1984) in the birds. This practice may lead to increased heat load and reduced survival of birds during summer. Fuller and Rendon (1977) found that broilers fed a diet in which 33% of the ME was supplied by fat, consumed 10% more both ME and protein contents, and also gained more weight than chicken fed low fat ration. Addition of fat to the diet appears to increase the energy value of other feed constituents (Mateos and Sell, 1981). Fat has also been shown to decrease the rate of food passage in the gastrointestinal (GI) tract (Mateos *et al.*, 1982) and thus increase nutrient utilization. Wilson *et al.* (1980) reported that high environmental temperature caused an

increase in food passage in white Pekin ducks. Dietary fat therefore, help counteracting the effect of high temperature.

Balnave and Oliva (1990) reported that methionine requirement of 3-6 week old broilers kept at a constant 30°C, or at cycling temperature of 25-30°C reduced compared with broilers kept at 21°C. Hurwitz *et al.* (1980) suggested a method of estimating the protein and amino acid requirements which takes into account the reduced rate of production at high temperature. A mathematical model was used, which evaluates the amino acids requirements on the basis of the sum of the maintenance requirements per Kcal, or as percentage of diet increase as environmental temperature increases, above the optimum range of growth. Then as the temperature of 28-38°C approached, there was a decline in amino acid requirements (arginine, leucine and the sulphur amino acid requirement). Sinurat and Balnave (1985) reported that the food intake and growth rate of broilers in a cyclic temperature (25-35°C) were improved not only by increasing the dietary ME, but also by reducing the amino acid: ME ratio during the finishing period. Similar findings were reported by Waldroup *et al.* (1976) in which they found that growth rate and feed utilization of heat stressed broilers was significantly improved when diets were formulated to minimize excess of amino acids. From the above discussion it can be concluded that high energy and low protein diets may be the best choice during summer. Present study was therefore conducted to further explore the addition of fat in ration under different feeding methods.

2.4. Leg Abnormalities

Leg abnormalities are mostly associated with increased growth rate. Over weight birds may become more susceptible to leg abnormalities. Broilers have the ability to maintain their growth even in adverse environmental condition. Hence there is a tendency that along with mortality, leg abnormalities may occur in non-restricted birds due to gaining more weight under stress full environment.

Reduction in leg abnormalities under the influence of intermittent lighting has been reported by Buckland *et al.* (1976), who applied four lighting regimes: (1)

continuous light (5 to 10 lux); (2) an intermittent system of 1 h of light (5 to 10 lux) and 3 h of darkness; (3) an intermittent system of 1 h of light (5 to 10 lux) and 3 h of darkness interrupted by 13 h of regular light (5 to 10 lux); (4) as with (3) but with low intensity light (less than 1 lux) during the 13 h period while the intermittent system continued, in 2400 broilers. Birds grown on continuous light had more leg abnormalities than those grown on any of the intermittent regimes. Influence of managerial factors on the incidence and severity of leg abnormalities was studied by Wilson *et al.* (1984) who reported that intermittent illumination (IL) resulted in significantly fewer and less severe leg abnormalities than birds under continuous illumination. Bird's activity was greater under IL and was deemed to be at least partially responsible for the stronger legs recorded among broilers under this management practice. Intermittent lighting programmes have invariably reduced the incidence of leg disorders in broilers (Buckland *et al.*, 1973, 1974; Buckland, 1975; Simons, 1982, 1986; Wilson *et al.*, 1984; Simons and Haye, 1985; Ketelaars *et al.*, 1986; Classen and Riddell, 1989; Renden *et al.*, 1991, 1996).

Changing the ratio of photoperiod: scotoperiod from commonly used 23 h light: 1 h dark to a more natural light:dark pattern may limit expression of skeletal deformities (FAWC, 1992; Sorensen *et al.*, 1999). Step-up lighting programmes which operate on the same principle as early feed restriction are also used to control leg abnormalities in broilers. Chicks are kept on a 23L:1D schedule for the first 2 days of life, to make them learn to find feed and water. Then a 6 hour photoperiod is employed until 21 days of age, to reduce feed intake and growth rate. This is then stepped up, abruptly or gradually, to a 23 hour photoperiod until slaughter. The increasing day length is thought to stimulate increased anabolic steroid production, causing acceleration in growth rate (Classen, 1992). Classen and Riddell (1989), Classen *et al.* (1991) and Riddell and Classen (1992) found that step-up program reduced the incidence of skeletal disorders in broilers by up to 50%, with no difference in body weight at 42 days compared with continuous lighting. Whereas,

increased leg disorders in the birds kept under continuous lighting than interrupted lighting schedules have been reported by Renden *et al.* (1996).

Hulan and Proudfoot (1987) found that cooler ambient temperature increase (linearly) the incidence of curly toes, angular deformity, enlarged hocks and total leg abnormalities but resulted in a linear decrease in tibial dyschondroplasia (TD). They also reported that dietary energy had no significant effect on leg abnormalities except for curly toes, the incidence of which decreased with increasing dietary energy.

A restriction of feed sufficient to produce a large decrease in growth rate has been shown to virtually abolish skeletal disorders (Riddell, 1983). Edwards and Sorensen (1987) fasted broilers for 8 hours a day, with the provision of *ad libitum* feed during the remainder of the day, during days 4 to 20 post hatching. The incidence of TD was consistently much lower than with normal feeding. A fasting period of at least 8 hours was needed to produce the maximal reduction of TD incidence although fasting did not have to occur every day. They suggested that fasting may have a similar mechanism to reducing the photoperiod, and that this mechanism may be the provision of intermittent periods of reduced growth during which the mobilization of adequate nutrients to the faster growing growth plates could occur. Quantitative feed restriction throughout the growth period has been shown to cause a proportional reduction in skeletal disorders, in addition to reduced growth rate (Duff and Thorp, 1985; Classen and Riddell, 1990; Robinson *et al.*, 1992).

Su *et al.* (1999) conducted two trials to find out effect of feeding methods on broiler performance. In the 1st trial they offered 2, 3 or 4 meals per day or fed *ad libitum* from 5 to 34 days. In trial 2 the birds were restricted to achieve 25, 50 or 75% of predicted growth for 5 or 7 days, starting at 5, 7 or 9 days of age. *Ad libitum* fed birds served as controls. In trial 1, fewer meals per day were associated with less tibial dyschondroplasia, less hock burn, better walking ability, lower body weight and better feed conversion. In trial 2, earlier restriction, and longer duration and

more severe level of restriction were all associated with less prevalence of tibial dyschondroplasia and better walking ability.

Robinson *et al.* (1992) restricted feed availability to maintenance levels during the second, third or fourth weeks of life in broilers and roasters. In all cases, the incidence of skeletal disorders was reduced. Restriction during the second week was the most effective. Disorders were reduced to one third of the incidence seen in full-fed controls. Market weight was delayed by only 2 to 3 days due to subsequent accelerated growth following the period of feed restriction. Feed restriction programs which reduced growth rate also consistently reduced the incidence of TD (Riddell, 1975; Poulos *et al.*, 1978; Edwards and Sorensen, 1987; Lilburn *et al.*, 1989; Su *et al.*, 1999) although there have been some contradictory findings (Poulos *et al.*, 1978). Su *et al.* (1999) concluded that meal feeding can beneficially affect the prevalence of leg weakness and that the major part of this effect is independent of changes in body weight. It was also concluded the early feed restriction decreased many aspects of leg weakness but the effects were mainly the result of decrease in body weight.

Rate of growth has long been shown to affect the incidence of leg disorders (Julian, 1997; Su *et al.*, 1999). Male broilers, which grow faster than females, have been reported to show about twice the incidence of skeletal disorders (Riddell and Springer, 1985; Classen and Riddell, 1989), as well as a poor walking ability (Kestin *et al.*, 1994). Kestin *et al.* (2001) reported that live weight and growth rate were important determinants of lameness in broilers. Keeping in view the above discussion it can be envisage that intermittent lighting significantly affected the leg abnormalities and intermittent lighting worked similar to intermittent feeding. Therefore various leg abnormalities were studied in this project to find out the relationship of leg abnormalities with different feed restriction systems during summer.

2.5. Mortality

Mortality during summer is mostly associated with heat stress and failure of thermoregulatory system of the birds. Arjona *et al.* (1988) investigated the effects of heat stress during the 1st week of life on subsequent mortality resulting from exposure to high environmental temperature ranging from 35.0 to 37.8°C for 8 h/day and feed restriction for 8 h/day on days 43, 44, and 45 just prior to marketing of broiler cockerels. Exposing birds to high environmental temperature at 5 days of age resulted in a significant decrease in mortality when birds were exposed to a high environmental temperature later in life. Cooper and Washburn (1998) reported that heat stress environment (32°C) did not affect mortality. Smith (1992) observed the effects of feed withdrawal and acclimation on livability in heat stressed broilers. Chicks were exposed or not, to ambient temperature of 38°C for 24 hours at 9 days of age. In experiment I, chickens were exposed to cycling temperature of 24° to 35° for the next 44 days. In experiment 2, chicken were exposed to ambient temperature of 24°C for 23 days followed by feed withdrawal for 0 to 24 hours and a rapid temperature increase to 37°C for the final 4 hours of feed withdrawal. Chickens on full feed had decreased livability. In a similar study birds were divided in to 3 groups that received either no food (starting 2 hours before the heat stress) no light or both were available during the hot period. Heat stressed birds with access to food and light showed 10% mortality (Zakia *et al.*, 1995). Mortality during thermal challenges was reduced by thermal conditioning and dual feeding (Basilio *et al.*, 2001).

Light treatments; continuous light, intermittent system of 1h light and 3 h darkness during the night with 13 h continuous light of the same intensity during the day, intermittent system of 1 h light and 3 h darkness during the full 24 h period, intermittent system of 1 h light and 1 h darkness from 1 to 2 weeks of age, 1 h light and 2 h darkness from 2 to 3 weeks of age and 1 h light and 3 h darkness from 3 to 14 weeks of age for the full 24 h, were applied by Buckland *et al.* (1973). Birds exposed to intermittent light tended to have lower mortality than those kept under

continuous light. Continuous lighting appeared to produce greater mortality from the "sudden death syndrome" than intermittent lighting. Prevalence was higher in cockerels than in pullets and deaths tended to occur most often at 3-4 weeks. Affected birds were heavier than the flock average (Ononiwu *et al.*, 1979). As incidence of sudden death syndrome is related to growth rate therefore growing broilers at slower rate may have reduced mortality (Bowes *et al.*, 1988).

Reduction in mortality rate has been observed due to feed restriction as compared to *ad libitum* fed birds. Lopez and Suarez (1989) reported that mortality was 4% in broilers given ration restricted by 30% during 4-5 week of age, 6.5% when ration was restricted by 20% for the same period, and 14.5% in broilers fed *ad libitum*. Fontana *et al.* (1992) studied the effect of early feed restriction on the mortality. Feed restriction was induced by providing chicks with 40 Kcal ME/bird/day, commencing at four days of age. Male chicks were fed restricted for 7 (Experiment 1 and 2) or six days (experiment 3 and 4), whereas broiler females were restricted for five days (Experiment 1). *Ad libitum* feeding was resumed after the restriction periods, and continued through the conclusion of the experiments at 49 (Experiment 1 and 2) or 28 (Experiment 3 and 4) days of age. Total pen body weights for restricted and *ad libitum* groups were similar at 49 days of age in experiment 1 and 2, which reflected a significant difference in mortality observed between the two groups. Broiler chicks subjected to feed restriction from 8th until 56th day of age with 8 h per day access to feed showed that mortality from ascites syndrome in the feed restriction groups was decreased by 88% (Arce *et al.*, 1995). Death rate in fully fed flocks was found to be higher at the age of 4 weeks than that of restricted-fed flocks (Tottori *et al.*, 1997).

Gonzales *et al.* (1998b) studied the effect of feed restriction (0, 10, 20, 30, 40 and 50%) from 8 until 14 days post-hatching, on total mortality in male broilers during winter and summer. *Ad libitum*-fed broilers had a higher total mortality rate (13.00% in winter vs. 3.67% in summer). All restricted groups had a lower mortality level in winter than the control group with a non-linear response influenced by feed

restriction. Birds submitted to 40% feed restriction showed the lowest mortality incidence (2.67%) followed by groups submitted to 30% (4.67%), 50% (5.33%) and 10% and 20% (7.67%) feed restriction. Birds fed *ad libitum* had the highest sudden death syndrome (SDS) incidence (3.67%, against 0.33, 1.00, 0.67, 1.33 and 1.33% at 10, 20, 30, 40, 50% feed restriction, respectively). Feed restriction at 30 to 40% for 7 days during the second week may decrease mortality in male broilers raised in winter without lowering productivity if the restriction is followed by at least 3 weeks of full feeding. Shah and Peterson (2001) reared broilers under three lighting program (23L:1D), on increasing photo period (IP) (from 14 to 23 L) and one decreasing photo period (DP) (from 23 to 14L) in a 42 days fattening period. Significant mortality was observed in birds reared under increasing photoperiod (2.8%) followed by (5.0%) in decreasing photoperiod. Mortality in male kept under (23L:1D) was higher (6.4%) than in female (3.9%) due to SDS.

Buckland *et al.* (1976) applied four lighting regimes: (1) continuous light (5 to 10 lux); (2) an intermittent system of 1 h of light (5 to 10 lux) and 3 h of darkness; (3) an intermittent system of 1 h of light (5 to 10 lux) and 3 h of darkness interrupted by 13 h of regular light (5 to 10 lux); (4) as with (3) but with low intensity light (less than 1 lux) during the 13 h period while the intermittent system continued, was studied in 2400 broilers of two commercial crosses over two trials. No effect on mortality was observed by the lighting treatments. Dorminey and Nakaue (1977) reported no effect on mortality when one-hour light and two hour dark (1L:2D) photo schedule was tested against continuous lighting. Similarly Renden *et al.* (1996) found no difference in mortality among treatments when he applied four lighting treatments (T₁) 23L:1D, (T₂) 16L:2D:1L:2D, (T₃) 23L:1D (day 1-7) and 16L:8D (day 8 to 14) with light period increased by 2 hr/week during days (15 to 35) and (T₄) 23L:1D (days 1 to 7), 16L:8D (day 8 to 14) 16L:3D:2L:3D (days 15 to 21), 16L:2D:4L:2D (day 22 to 28), 16L:1D:6L:1D (days 29 to 35) and 23L:1D there after.

In the above review lighting schedules were applied basically to restrict feeding in order to reduce growth rate, as the birds did not eat when there is no light (Buyse *et al.*, 1996). Hence the impact of intermittent feeding or intermittent lighting as a means of feed restriction on the mortality was highly dependent on the duration and intensity of restriction.

Effect of nutrition on mortality was studied by Kubena *et al.* (1972a) where broilers were provided diets with 1, 3, 7 and 10% added animal fat from 4 to 8 weeks of age. At 8 weeks of age the birds were exposed to heat (40.6 °C) for 2 h and death rates were recorded. The fat content of the diet did not affect mortality from heat prostration. The finding was significant as more concentrated rations are sometimes given to broilers in summer in an attempt to improve growth and feed utilization. Blair *et al.* (1975) subjected broiler chicks to an ambient temperature 2-3°C higher than normal and were fed various diets to determine the effects of dietary factors on mortality. The result showed that mortality was not influenced by cereal type (barley or wheat). However, dietary fat significantly affected the mortality rate, which decreased with the increase in the fat content of diet. High levels of protein and fat in the diet had a protective influence against mortality but the results, especially with pelleted diets, suggested that some other nutrients might be involved in the syndrome. Rotter *et al.* (1987) reported that wheat-soy diet supplemented with sunflower oil reduced mortality caused by sudden death syndrome (SDS) as compared to the same diet supplemented with tallow. There was a trend for the SDS occurrence to be lowest with the highest concentration of animal fat. Non-significant effect of dietary energy on mortality has been reported by Hulan and Proudfoot, 1987; Barbosa and Campos, 1996. Albuguerque *et al.* (2000) observed that mortality due to ascites was higher ($P < 0.05$) for males subjected to three different feeding programs and fed on energy rich ration.

2.6. Slaughter Characteristics

2.6.1. Dressing Percentage

Dressing percentage of broilers decreased when environmental temperature exceeded from 29°C (Al-Fataftah, 1987). The adverse effect of high ambient temperature on dressing percentage of broilers was also studied by Sokolowicz *et al.* (2000). Four groups of broilers were housed at temperatures (1) 34°C after hatching decreasing to 20° on day 42; (2) 10° higher than group 1 to day 21; (3) 10° higher during the last week and (4) 10° higher throughout. Dressing percentage was 76.6, 74.0, 75.0 and 70.6 for females and 76.5, 72.1, 73.9 and 69.6 for male broilers, respectively. The birds of group 4 exhibited the lowest dressing percentage than those of the other groups indicating that high ambient temperature resulted into reduced dressing percentage. Mizubuti *et al.* (2000) found that live weight, carcass weight and carcass yield, did not differ ($P < 0.05$) among three food restriction treatments (0, 6 and 12 h of fasting on alternating days) from 7 to 21 days of age. Oliveira *et al.* (2000) reported that carcass yield of broilers maintained under thermoneutral environment ($23.2 \pm 0.74^\circ\text{C}$) linearly reduced according to ME levels (3000, 3075, 3150, 3225 and 3300 Kcal ME/kg diet) from 22 to 42 days of age.

2.6.2. Organs Weight

Environmental temperature higher than 29°C resulted into decreased percentages of liver, heart, edible yield and carcass weight of broilers (Al-Fataftah, 1987). Heating the broiler chicks (36°C for 6-10 hours per day) from day old till 4 weeks of age resulted into reduced adrenal and thyroid weight (Kutlu and Forbes, 1993). Weights of the spleen, bursa fabricii, thymus and the ratio of their weights to body weight were significantly lower when broilers were subjected to heat stress at 28°C and 32-36°C at 65-80% relative humidity than those of the control group kept at 20-22°C and a relative humidity of 50-60% (Zhang *et al.*, 1998). Broiler chickens in group one were reared at 33°C from day 1, which was reduced to 18°C by day 49 (optimum temperature), whereas group 2 chickens were reared at 10°C higher than

the optimum temperature throughout the rearing period. Overall, adrenal gland weights were higher in group 2 than group 1 (Sokolowicz and Herbut, 1998).

Feed restriction resulted in greater relative adrenal mass but the difference decreased with the time (Freeman *et al.*, 1981). Willis *et al.* (1996) reported that when feed was withdrawn from male broilers at 0, 6, 12, 18 or 24 h before processing, liver weights (g) were reduced significantly with the extended feed withdrawal periods. Which indicate that even a short-term restriction may result in reduction of organ weights. Plavnik and Yahav (1998) studied the effect of early growth restriction (EGR) induced by feed restriction and found that relative heart, liver, and breast meat weights at 8 weeks of age were decreased significantly in the EGR chicks.

Conditioning at an early age ($36\pm 1^{\circ}\text{C}$, 70 to 80% RH for 24 h at the age of 5 in group 1, or 5 and 7 days in group 2) resulted into a significant decrease in heart weight (Yahav *et al.*, 1996). Heart to lungs ratio decreased in broiler stocks conditioned to 36°C for 24 h at 5 d of age or feed restricted (food withdrawn 2 h before the hot period and chicks fed between 17.00 and 08.00 h) during heat stress (Yalcin *et al.*, 2001). Basilio *et al.* (2001) reported that liver and gizzard were significantly heavier in dual feeding program and thermal conditioning.

Inclusion of lard (0, 3 or 7%) in the non-isocaloric diet did not affect body, heart, spleen or pancreas weight of broilers whereas liver weight was suppressed by the inclusion of lard in the diet (Latour *et al.*, 1994). Spleen and thymus weights were not affected by the inclusion of different fat sources. Whilst, the weight of the bursa was higher ($P<0.05$) at 3 and 7 weeks of age in chickens fed on linseed oil (rich in n-3 polyunsaturated fatty acids) than those fed beef tallow (rich in saturated fatty acids) or 1:1 mixture of both sources for 7 weeks (Zaki and Hady, 1995). Peebles *et al.* (1997) studied the effects of added lard in starter diets on organ weights. Broiler chickens received either 0, 3, or 7% added lard in starter diets through 10 d of age (S1), followed by either 3 or 7% added dietary lard through 21 d of age (S2). The effects of the S1 and S2 diets on relative liver weight were

inconsistent over time and were not influenced by sex. Oliveira *et al.* (2000) found no effect of dietary ME levels (3000, 3075, 3150, 3225 and 3300 Kcal ME/kg diet) on absolute and relative weights of thigh, drumstick, legs, breast with bone, heart, liver, gizzard, proventriculus, lungs, intestine, abdominal fat and feathers of broilers from 22 to 42 days of age maintained under thermoneutral environment ($23.2\pm 0.74^{\circ}\text{C}$).

In the above review it is clear that temperature, thermal conditioning, feed restriction, and addition of fat may affect relative or absolute weights of various body organs. However response of broilers regarding length and weight of alimentary tract and relative weight of different body organs under hyperthermic conditions by providing various feeding therapies still has not been studied much. Therefore, this experiment is expected to explore the changes in this respect.

2.6.3. Abdominal Fat Pad

Exposure of broilers to high temperature (exceeding 29°C) increased the abdominal fat (Al-Fataftah, 1987). Temim *et al.* (2000) also found that high ambient temperature increased the abdominal fat proportion in broilers. Plavnik and Yahav (1998) reported that relative abdominal fat pad was significantly lower in chickens kept at $25:35^{\circ}\text{C}$ than in those kept at 25 and 35°C , when chickens were placed in temperature-controlled chambers at 25, 30, 35°C and a diurnal cyclic temperature $25-35^{\circ}\text{C}$ from 4 to 8 wk of age. These results indicate that although high temperature resulted in increased abdominal fat yet cyclic temperature may alter the results.

Feed restriction (60% of *ad libitum*) did not affect abdominal fat pad size (Sheila *et al.*, 1993). Early feed restriction (7 to 14 d of age or 8 to 16 d of age) with 90, 75, and 60% restriction of previous 24-h feed consumption of full-fed also showed no effect on abdominal fat (Deaton, 1995). Mizubuti *et al.*, 2000 reported that percentage of abdominal fat did not differ ($P < 0.05$) among three feed restriction treatments (0, 6 and 12 h of fasting on alternating days from 7 to 21 days of age). Phase-feeding (PF relative to an NRC), was studied by Pope and Emmert,

(2002) lowering the amino acid requirements predicted with PF linear regression equations by 10% (PF10), increasing the slope of the linear regression equation by 15% (PF15). No differences ($P < 0.05$) in abdominal fat were observed when birds were fed PF, PF10, or PF15 diets. Whereas Zhong *et al.* (1995) found that abdominal fat was lower for female and combined sex broilers for the restricted than full fed broilers. Skip-a-day feeding resulted in decreased abdominal fat (Santoso, 1995). Gonzalez *et al.* (2000a) also reported that feed restriction reduced the abdominal fat pad in broilers. Hence, it could be concluded that method of restriction and duration of restriction are the basic factors responsible for abdominal fat content of broilers.

Abdominal fat weight in the females at 7 weeks of age was 63.5 g, accounting for 3.2% of the body weight, and those of the males and the females at 9 weeks of age were 94.6 g and 2.9%, and 88.1 g and 3.8%, respectively. Variation among birds was extremely high. Positive correlations were observed between body weight and abdominal fat weight and between abdominal fat weight and fatty liver score. It was concluded that in broilers excessive deposition of fat tended to accompany the fatty liver (Akiba *et al.*, 1986). Cahaner *et al.* (1995) reported that abdominal fat decreased with increased protein intake in birds. Increasing the fat in ration may reduce protein intake resulting in to lower abdominal fat. Increasing dietary energy from 12.12 to 13.79 MJ ME/kg exhibited increase in relative abdominal fat of chickens in a linear manner (Yalcin *et al.*, 1998). Abdominal fat deposition also increased linearly with increasing the number of days in which the birds were fed tallow-enriched diet (Sanz *et al.*, 2000). However, no effect of dietary ME levels (3000, 3075, 3150, 3225 and 3300 Kcal ME/kg diet) was observed on abdominal fat of broilers from 22 to 42 days of age maintained under thermoneutral environment ($23.2 \pm 0.74^\circ\text{C}$) (Oliveira *et al.*, 2000). Abdominal fat was significantly lower in animals fed sunflower and linseed oils than in those fed tallow or olive oil ($P < 0.001$). In females, it increased with level of fat inclusion, only when fed tallow or olive oil, whereas it remained constant in birds fed sunflower or linseed oil. Polyunsaturated fatty acids reduced abdominal fat deposition than saturated or monounsaturated fatty acids (Crespo and Esteve-Garcia, 2001). Du and Ahn (2002) found no difference in the weight of abdominal fat among

the broilers fed on 0, 0.25, 0.5, or 1% conjugated linoleic acid. However, when the dietary conjugated linoleic acid was increased to 2 or 3%, the total body fat contents were reduced. Whole body fat content decreased from 14.2% in the control to 11.9 and 22.2% for 2 and 3% conjugated linoleic acid.

2.7 Meat Analysis

Muscle from broilers exhibited an increased sensitivity to acute heat stress exposure with age. Alterations in ante-mortem blood acid/base status and muscle membrane integrity induced by acute heat stress were associated with adverse effects upon breast meat quality. Exposure of broiler chickens to acute heat stress may cause alterations in muscle metabolism and membrane integrity, which results into undesirable meat characteristics (Sandercock *et al.*, 2001).

Birds of varied inherent growth rate and tendencies toward protein and fat deposition respond differently to dietary protein level under heat stress (Cahaner *et al.*, 1995). Santoso (2002) conducted a study to evaluate the effect of early feed restriction on meat composition in unsexed broiler chickens. In experiment 1, three hundred and fifty one-day-old broiler chicks were divided into 7 groups. Each treatment group was represented by five replicates of ten broilers each. One group was fed *ad libitum* as the control group and the other six groups were fed 25% *ad libitum* (25% multiplied by amount of feed intake of *ad libitum* chicks at the previous day) for 4 or 6 days, 50% *ad libitum* for 4 or 6 days, and 75% *ad libitum* for 4 or 6 days. In experiment 2, five hundred broiler chicks were divided into 10 groups. Each treatment group was represented by five replicates of ten broilers each. One group was fed *ad libitum* as the control group. Three initial ages at which broilers were restricted (2, 4 or 6 days of age) and three type of feed restriction (physical restriction, meal feeding and diet dilution) (3×3) were examined. Experimental results showed that broilers fed 25% *ad libitum* for 4-6

days tended to reduce leg meat fat, whereas breast meat fat was significantly higher in restricted birds.

Latour *et al.* (1994) studied the effects of dietary fat on the carcass composition; in experiment one, broiler chicks were fed nonisocaloric diets with either 0, 3 or 7% added lard, liver weight was suppressed by the inclusion of lard in the diet. The composition of liver was also changed and became higher in protein and lower in fat. Chicks given 7% added lard had lower liver fat throughout the trial than chicks fed no added lard. Additionally, these chicks had a higher body protein content. However, body fat only increased in chicks fed 7% added lard. Oliveira *et al.* (2000) reported that a level of 3232 Kcal ME, corresponding to the calculated energy: protein ratio of 16.49, gave the best performance and protein deposition in the carcass of broilers maintained under a thermoneutral environment. Whereas Yalcin *et al.* (1998) found no effect of dietary energy on the nutrient composition of breast meat when heterozygous naked neck chickens were reared under natural spring (average 21.2°C) and summer temperatures (average 27.1°C). Du and Ahn (2002) reported that after cooking, the breast meat from 2 or 3% dietary conjugated linoleic acid treatment was harder and drier (moisture content changed), and the color was a little darker than those of controls. They also reported that these changes could be caused by the decreased unsaturated fatty acid content in meat after conjugated linoleic acid feeding, which increased the melting point of the fat. Above review reflects that addition of fat and altering energy levels in diet may alter the chemical composition of meat.

2.8. Haematological Parameters

2.8.1. pH

Heat exposure resulted into increased blood pH in broilers birds (Branton *et al.*, 1986.) Teeter *et al.* (1985) observed that blood pH was higher in heat-stressed (32°C) panting birds (7.40) than either non-panting (7.28) or birds reared at

24°C (7.28). Acute heat stress (32 to 41°C over 20 minutes) further elevated blood pH to 7.52. Chronically heat-stressed broiler chicks had intermittent respiratory alkalosis during panting whereas with acute heat stress, chicks panted continuously and had alkalosis. Significantly elevated blood pH values in broilers were reported by Ching and Ching (1992), when they were kept under 38°C ambient temperature. Ait-Boulahsen *et al.* (1992) and Furlan *et al.* (1999) also found that heat stress was associated with increase in blood pH. Whereas Deyhim and Teeter (1991) reported no effect of high ambient temperature on the blood pH of broilers when subjected to either a thermoneutral (TN; 24°C) or cycling temperature (24 to 35°C) at 5 weeks post hatch.

Feed restriction and/or use of carbohydrate-free diets increases survival time under conditions of acute heat stress; both these mechanisms are thought to induce a metabolic acidosis that helps to maintain the pH in situations of respiratory alkalosis (Leeson, 1986). Heat stress in *ad libitum*-fed birds was found to be associated with an increase in blood pH and a decrease in pCO₂, and in restricted birds with a decrease in blood pH and an increase in pCO₂ (Hocking *et al.*, 1994). Maintenance of blood pH through feeding management is a key factor in birds survival during heat stress (Nillipour and Melo, 1999).

2.8.2. Haemoglobin

High environmental temperature adversely affects hemoglobin contents of blood. Whereas the other factors, which influence the concentration of hemoglobin in blood are age, sex, season, acclimatization and nutrition etc. Most of the studies revealed that high environmental temperature decreases haemoglobin level in chickens (Deaton *et al.*, 1969a; Zimmerman *et al.*, 1973, 1975) and male turkeys (Parker and Boone, 1971). Deaton *et al.* (1969b) observed that haemoglobin contents in broilers reared at 7.2°C were higher than those kept at 40.6°C. The chickens kept from 1 to 10 weeks of age at ambient temperature of 23.9°C had significantly (P<0.05) higher haemoglobin level than those reared at 37.8°C (Sabry *et al.*, 1978). Huston *et al.* (1962) stated that

oxygen consumption in fowl decreased with increase in ambient temperature, and this may be associated with a depressed haemotopoietic activity as a result of lowered basal metabolic rate (BMR). Chickens show seasonal variation in their hemoglobin levels. During cold weather, haemoglobin content of the hens has been reported to increase (Winter, 1935). Yaqoob (1966) observed that haemoglobin level in hens increased at 37.8°C specially in Desi (local) breed, however, it fell sharply at 43.3°C.

Deaton *et al.* (1969b) observed the haemoglobin level of broilers reared at 32.2°C for the first 24 hours and then established over a 6 hour period at 32.0°C, 23.9°C and 7.2°C, with dew points of 10°, 10° and 0°C respectively, in 3 chambers for 55 days. Results did not show any effect on haemoglobin level at the end of first week. Beginning with the second week and continuing throughout the 8th week, both sexes reared at 7.2°C temperature had significantly higher rate of haemoglobin. It was therefore, concluded that winter level of haemoglobin was higher than summer. With the change of environmental temperature, a definite change in blood system occurs in chicken. Haemoglobin concentration was found to be dropped in male broiler chickens as ambient temperature rose from 10 to 30°C or 15 to 35°C (Yahav, *et al.*, 1997). Vo and Boone (1975) reared male and female broilers from 2 to 8 weeks of age at 21.7, 29.4 and 37.8°C. The average haemoglobin levels in the three groups were 13.25, 13.22 and 10.67 gm/100 ml of blood, respectively. Factors that affect erythropoiesis and red cell number also affect haemoglobin level (Sturkie and Griminger, 1976). Furlan *et al.* (1999) studied the effect of acute heat stress (35°C/4 hour) on haematological and gasometric parameters from five commercial broiler chicken strains (Arbor Acres, Cobb, Hubbard-Petterson, Isa and Ross) at different ages (4 and 7 weeks of age). The haematological parameters (erythrocyte number, haematocrit and haemoglobin) were not affected by chicken strain. Whilst, age of the birds significantly affected haemoglobin content with higher values at 7 weeks of age.

Shlomo (2000) reported that homeotherms acquire thermal tolerance under potentially deleterious thermal stresses by acclimation and thermal conditioning. In both strategies alterations in heat production and/or heat loss occur in response to changes in the environment. Environmental conditions are commonly considered to be a combination of ambient temperature and relative humidity (rh), therefore, the effects of these two parameters on the performance and thermoregulation of broiler chickens and turkeys were studied, using acclimation or thermal conditioning strategies. Acclimation of broilers and turkeys to a wide range of constant environmental temperatures suggested the range of 18-20°C, as the optimal one for maximal performance. However, in practice fowls are exposed to diurnal temperature cycling, therefore it was suggested to expose them to ranges of ambient temperatures (Tas) of 15°C and 30°C. Relative humidity played a major role in performance of chickens and turkeys exposed to Tas 28°C, and >30°C, respectively. The preferred rh for raising chickens has been found to be 60-65%, whereas in turkeys it was age dependent, i.e., up to 8 weeks the preferred rh was 40-45% and thereafter 70-75%. Acclimation to altered environmental conditions resulted in changes in the blood system, to accommodate to changing energy needs (changes in haematocrit/haemoglobin concentrations), on one hand, and, on the other hand, changes to accommodate heat dissipation (increase in plasma volume, alterations in the blood acid-base balance, and heat loss by radiation). Thermal conditioning (exposing chicks at the age of 5 days to 36°C, 70-80% rh for 24 h) of broiler chickens, resulted in thermotolerance improvement during exposure to heat stress (35°C and 20-30% rh for 6 h) at marketing age, and this coincided with significantly improved performance. However, while the improved thermotolerance was significantly higher than that of the control, it was far inferior to that of chickens acclimated to similar conditions. Both strategies may be useful, depending on the environmental conditions that the birds has to face during the growth period, and on the economic restraints.

Garcia *et al.* (1992) observed that food-deprived males exposed to 40°C for 4 h had a significant increase in haemoglobin and haematocrit and a decrease in leukocyte count than those subjected normal rearing conditions. Walton *et al.* (1999) conducted three experiments with 405 broilers using hypobaric chambers and control pens, fed diets containing flax oil (25 or 50 g/kg feed) or control diets with equivalent amounts of animal/vegetable (A/V) blend oil for 4 weeks. The effect of these diets on haematological variables and the extent of right ventricular hypertrophy (RVH) leading to ascites were determined. Feeding the 50 g flax oil/kg diet under hypobaric conditions reduced the haematocrit and haemoglobin content.

2.8.3. Packed Cell Volume

Environmental temperature, age, season, feed, diurnal effect, and fasting are related to packed cell volume in chicken. Huston (1965) observed that haematocrit in birds reared at 30°C was significantly less than those reared at 8 and 19°C, however, non-significant difference existed between haematocrit at 8 and 19°C. Fowls kept in a cold environment showed much higher haematocrit values, than those kept at room temperature. The effect of environmental temperature (8°C and 30°C) on haematocrit levels in the summer and winter months was studied by Moye *et al.* (1969). Haematocrit levels at weekly interval were determined upto 63 days of age. Low temperature (8°C) caused an increase in the haematocrit, while the higher temperature (30°C) caused a decline in it. High environmental temperatures depressed packed cell volume in chickens Kubena *et al.*, 1972b; Zimmerman *et al.*, 1973; Zimmerman *et al.*, 1975; Andrade *et al.*, 1976).

Response of environmental temperature on the haemodynamic changes in male broiler chickens was studied during exposure to constant temperature of 10 to 35°C or diurnal temperature cycles of 10:30°C and 15:35°C, and during acute heat or cold by Yahav *et al.* (1997). Haematocrit percentages recorded at 8 weeks of age for chickens exposed to constant or diurnal cyclic

ambient temperature in the range of 10 to 30°C in trial 1 were 34.5, 31.1, 25.6, 30.9 and 30.8 at 10°, 20° and 30°C, and at 10° phase and 30°C phase, respectively. These values for the chickens in trial 2, exposed to constant or diurnal cyclic ambient temperatures in the range of 15 to 35°C were 35.2, 32.4, 27.5, 30.9 and 28.7 % at 15°, 25° and 35°C, constant and at 15 and 35°C phase, respectively. Acclimation to low temperature resulted in an increase in packed cell volume, recorded at 20°C. Authors concluded that changes in haematocrit levels were parallel to changes in heat production and were probably aimed at modulation of the supply of oxygen to accommodate the changing needs. Acclimation to altered environmental conditions resulted changes in haematocrit to accommodate to changing energy needs (Shlomo, 2000).

Heat stress caused significant reduction in packed cell volume and the decreased haematocrits appeared due to decreased cell count (Sahota *et al.*, 1993). Deyhim and Teeter (1991) reported that ambient temperature (38°C) for 0, 3, 17, 29 and 43 h declined haematocrit of broilers, from 3 to 43 h. Acute heat stress was associated with a decrease in packed cell volume (Furlan *et al.* 1999). Food-deprived males exposed to 40°C had significant increases in haematocrit (Garcia *et al.* 1992). Al-Rawashdeh *et al.* (2000) reported that feed-restricted birds had slightly higher PCV, than birds fed *ad libitum*. Packed cell volume (PCV) was lower for four week-old birds and tended to increase up to 6 weeks of age. Whereas Gonzalez *et al.* (2000b) reported that haematocrit (39.7 vs. 37.1%) were lower only on day 20 with feed restriction. Haematocrits increased across time in birds fed 3% fat in starter diets through 10 day of age (Peebles *et al.* 1997).

2.8.4. Erythrocytes

Acute heat stress has been found to be associated with a decrease in red blood cell number whereas erythrocyte number was not affected by chicken strain (Furlan *et al.* 1999). Maxwell *et al.* (1992a) reported that erythrocytes and

thrombocytes were longer and thinner after heat stress, possibly due to dehydration, these changes seen in the cells after heat stress may affect their functions in raising an immune response. Increased erythrocyte numbers and mean cell volume resulted into higher packed cell in chickens in cold than at hot environments (Huston, 1960; Washburn and Huston, 1968; Moya *et al.*, 1969; Soliman and Huston, 1974).

2.8.5. Total Leukocyte Count

Heat stress caused an increase in corticosterone level, and decrease in total leucocytic count (Ben Nathan *et al.*, 1976), which can be used as indicators for measurement of stress response. Environmental temperature had a significant effect on White Blood Cell (WBC) count. Broilers those were not provided cooling during thermal stress exhibited the highest WBC count (Datta *et al.*, 1984). Differential leucocyte responses to various degrees of food restriction in broilers, turkeys and ducks were carried out by Maxwell *et al.* (1992b) at higher environmental temperature. Temperature of 20 to 24°C during week 1 to 5 and decreasing to 13 to 16°C at 9 weeks of age were maintained. The duckling were bled at 1, 2, 9 and 12 weeks of age. In broilers blood samples were taken at 12, 22, 33, and 42 days of age. Whereas, in turkey blood sampling was done at 3, 5, 6, 7, 8, 9 and 14 weeks of age, respectively. Feed restricted broilers showed increase in heterophil and basophil number with decrease in lymphocytes. The heterophil/lymphocyte ratio also increased and no difference between different strains of broiler was noted. In case restricted feeding of ducks heterophils were significantly raised in 2 week old ducks. *Ad libitum* feeding also raised heterophils at 21 weeks of age. After food restriction turkey responded with significant heterophil/lymphocyte ratios following two degrees of restricted feeding. It was concluded that in poultry a heterophilia may be the response to mild to moderate stress but a basophilia may result after severely stressed birds. The authors describes that it is widely accepted that various stressors of environment reduce the efficiency of animal production. A temperature high enough to cause increased body temperature changed circulating leucocyte components in broilers (Altan *et al.*, 2000).

Maxwell *et al.* (1992a) found that number of electron dense granules in small lymphocytes were reduced due to heat stress. Eosinophils showed no significant differences in cytoplasmic mitochondria, granules, vacuoles or lobules, but all values were slightly higher after heat stress. Salvador *et al.* (1999) observed increased heterophil/lymphocyte ratio as a consequence of high ambient temperature. Exposure of broilers to acute heat stress ($39\pm 1^{\circ}\text{C}$ for 2 h at 44 d of age) decreased monocyte and lymphocyte proportions whereas the proportion of eosinophil and the haematocrit values were not affected (Altan *et al.*, 2000). Feed restricted birds had slightly higher heterophils, lymphocytes and monocytes than birds fed *ad libitum* (Al-Rawashdeh *et al.* 2000). Hocking *et al.* (1994) found that feed restriction was associated with lower plasma triglyceride concentrations, lower number of heterophils and monocytes and a lower ratio of heterophils to lymphocytes. Above studies indicate that heat stress and feed restriction effects differential leucocyte count hence study of this parameter may be a wise step to understand the effect of treatments under study.

2.9. Blood Biochemistry

2.9.1. Glucose

Environmental temperature and feeding management are the factors which may influence the change in blood glucose level in birds. Broiler kept at 43°C in chambers for 2 hr showed a concurrent decline in plasma level of glucose (Edens, 1978). Yang *et al.* (1992) observed high blood sugar levels (223.6 and 221.7 mg/100ml) in broiler exposed to 23 and 28°C , respectively. Sahota *et al.* (1994) reported that blood glucose of birds increased with increase in the ambient temperature. At 17.5°C blood glucose was 196 and 179 mg in 5 weeks old Lyallpur Silver Black and White Leghorn chicks, respectively, which increased to 206 and 195 mg at 27.5°C when they were 12 weeks old. Blood glucose level changed with change in environmental temperature and advancing age in both breeds. It was also observed that high environmental temperature of 39°C increased blood glucose levels for subsequent 6 weeks at 5.88 and 8.28 percent in Lyallpur Silver Black and White Leghorn breeds, respectively. The authors concluded that the elevation of

blood glucose in experimental birds under heat stress was due to hydrolysis of glucagon as a result of increased body temperature. Significant increase in blood sugar was observed in broilers subjected to 35°C for 6 h/day from days 32 to 49 of age (Deyhim *et al.*, 1995). Lin *et al.* (2000b) also found that plasma glucose level was increased by heat exposure (35°C, for 2 h) and this effect was aggravated by increase in exposure time (24 h).

Hazelwood and Lorzen (1959) studied the effects of prolonged fasting in adult chickens, and after observing the usual decrease in blood glucose, 24 to 36 hours after the starvation period commenced, found that a progressive rise in blood glucose occurred, reaching a maximum (22 percent above fed levels) six days later concurrently, liver glucagon, depleted early during fasting, was partially replaced by prolongation of the starvation period. Simultaneous studies of nitrogen metabolism indicate that increased gluconeogenesis appears likely not only to be responsible for the blood glucose increase but also to play a role in the restoration of liver glucagon after the initial glycogenolysis, induced by fasting. Pierce and Fanguy (1971) noted a reduction in the blood glucose during the first 72 hours under food deprivation stress but slightly elevated due to 144 hours post fasting. In another study the data on blood sugar indicated that insulin secretion was inhibited at high temperatures and that the use of interrupted feeding (2h F-2h NF) and watering decreased this effect (Suprunov *et al.* 1995). Whereas Zulkifli *et al* (2000) reported that serum concentrations of glucose were elevated by the heat challenge, but were not affected by the feeding regimes; (1) *Ad libitum* feeding (ALF); (2) 40% feed restriction at 4, 5, and 6 d of age (F40); (3) 60% feed restriction at 4, 5 and 6 d of age (F60); and (4) 80% feed restriction at 4, 5 and 6 d of age (F80). The intent of the present work was to observe the behaviour of birds regarding glucose variation as a result of feeding regimens and addition of fat.

2.9.2. Cholesterol

Cholesterol concentration in birds may be influenced by sex, age, ration, hyperthermia and fasting. Hevia and Vinsek (1979) stated that starvation increased blood cholesterol level in chickens. Fasting caused mobilization of fat through gluconeogenesis which ultimately increased blood cholesterol level. Dickson (1975)

observed that thyroid deficiency resulted in marked increase of serum cholesterol level. Blood total lipids, triglycerides, and cholesterol at 4 and 7 weeks of age were not affected by the type of dietary fat (Rotter *et al.* 1987). Zulkifli *et al.* (1999) reported high serum cholesterol concentrations in birds in response to heat treatment ($36\pm 1^{\circ}\text{C}$). However, the effect of feeding management on cholesterol concentration in birds under stressful environment needs to be addressed.

2.9.3. Protein

Broilers kept for 4 weeks at $18-22^{\circ}\text{C}$ exhibited more serum total protein than those kept at $33-35^{\circ}\text{C}$ (Ward and Peterson, 1973). Ching and Ching (1992) determined the plasma protein content of Taiwan country chickens and broilers kept at 38°C for 0, 3, 17, 29 and 43 hours. The plasma protein content of these birds decreased at 3 hours and remained at lower level until 43 hour, however, no difference was observed between the two strains. Pardue *et al.* (1985) found that heat stress considerably decreased the total protein in blood. Yang *et al.* (1992) found higher total protein content in the serum of broilers exposed to 12°C group than those kept under 23 and 28°C . Decrease in protein content of the heat stressed birds was also observed by Geraert *et al.* (1996).

During the study of hemodynamic changes of fowl under high environmental temperature one of the important constituent; plasma protein was studied by Yahav *et al.* (1997). Three trials were conducted using Cobb male broiler chickens raised for 4 weeks in battery brooders at 26°C . In trial 1, the birds acclimated during the 5th week of age to constant environmental temperatures of 10, 20 and 30°C or to diurnal temperatures of low (10°C) and high (30°C) upto 8 weeks of age. In trial 2, the cyclic environmental temperatures were 15 and 35°C and constant temperatures were 15, 25 and 35°C , whereas in trial 3, non-acclimatized 8 week old chickens were exposed to higher temperature of 35°C . In trial 1, the total protein percentages were 4.80, 4.34, 4.19, 4.17 and 4.28 at 10,

20°, 30°, 10° and 30°C phase, respectively. Whereas the values at 15°, 25°, 35°, 15° and 35°C phase were 4.09, 4.26, 4.44, 3.66 and 4.31 percent, respectively. Plasma protein concentration increased significantly only during the 35°C phase of the 15:35°C cycle but decreased at higher temperature.

2.9.4. Albumin and Globulin

Heat distress (from days 32 to 49 subjected to 35°C for 6 h/day) reduced ($P<0.05$) serum total protein and albumin (Deyhim *et al.*, 1995) in broilers. Serum total protein and albumin also decreased ($P<0.05$) with trace mineral and/or vitamin supplementation. Yang *et al.* (1992) reported that serum globulin content of broilers decreased with increase in the environmental temperature. Liu *et al.* (2000) exposed the broilers to high temperature (42°C) for 3 h and found lower serum total protein and albumin values than those kept under thermoneutral zone (control).

Ehinger (1977) reported that diet did not affect plasma albumin of broilers fattened from 1 to 46 days on 1 of 4 diets containing metabolizable energy 2800 or 3250 Kcal/kg with protein 25.0 or 20.0%. There was no correlation between plasma protein and total carcass fat. Whereas Zavarey (1984) found that nutritional factors affected mainly serum total proteins, the level being higher on high protein diet. Plasma protein concentrations were increased in birds fed 7% fat added diet through 10 d of age (Peebles *et al.*, 1997).

Little information regarding the response of birds with respect to the serum albumin and globulin fractions under various feeding managements during high environmental temperature and humidity led to the inclusion of the parameter in the scheme of this study.

2.9.5. Urea

Yang *et al.* (1992) reared 5 groups of broilers at 12, 18, 23, 28 and 32°C for 4 weeks to investigate the uric acid concentration. Serum uric acid contents of broilers were significantly affected by environmental temperature and sex with the minimum levels at 23-28°C. The concentration of uric acid was significantly higher

in the 12°C treated group than those kept under 23°C and 28°C. Ward and Peterson (1973) reported that broilers exposed for 4 hours to 33-35 °C had higher levels of plasma uric acid (6.7 against 5.5 mg/dL) than birds kept at 18-22 °C. Whereas Deyhim *et al.* (1995) found that heat distress (from days 32 to 49 subjected to 35°C for 6 h/day) reduced uric acid. Lin *et al.* (2000b) conducted four experiments, involving 400 broilers, to evaluate the effect of temperature and humidity on biochemical indices of Arbor Acres broilers at different ages (3-7 weeks). They reported that temperature had significant effect on the uric acid levels ($P < 0.01$) under one of his four experiments, whereas in another experiment they found that high temperature (32°C) or high humidity (85%) had no significant effect on uric acid concentration. A controversy in the findings was the initiative to add this parameter in the study.

2.10 Liver Enzymes

Arad and Marder (1984) studied the SGPT in normally dehydrated fowls. Stepwise increase of heat stress from 35 to 40°C for 2-3 hours and water deprivation for 48 hours increased SGPT level in blood serum. The results supported that fowls can regulate body temperature by acclimatization and enzymic changes during heat exposure and dehydration. Likewise, the objective of this study was to observe the response of broiler with reference to enzymatic changes under different feed managemental programs during summer. Amubode and Fetuga (1984) observed the effect of dietary methionine, protein, and caloric density on the glutamic-oxalacetate transaminase (GOT) and glutamic-pyruvate transaminase (GPT). They found no effect of protein and methionine levels on GOT either in the plasma or liver. Plasma GPT tended to increase ($P < 0.01$) between 0.28% and 0.44% methionine levels in experiment I and between 0.26% and 0.50% in second experiment. Whereas GPT activity in the liver was similar in both the experiments. The interaction between methionine + cystine and protein was significant in their effect on the GPT activity. This enzyme decreased in the plasma with increasing methionine + cystine level,

while it increased with increasing protein level. The GPT activity in the liver was found negatively correlated ($r = -0.52$) with the energy level.

2.11. Micro and Macro Minerals

Environmental temperature and composition of diet may affect the mineral content of plasma and muscle. Ward and Peterson (1973) reported that broilers exposed to 33-35°C for 4 hours had higher chloride contents (114 against 111 meq/litre) than broilers maintained at 18-22°C. Edens (1978) found that acute heating (43°C) in chambers, for periods up to 2 hr in duration resulted in decline in plasma levels of, sodium/potassium ratio, total calcium, and inorganic phosphate in broilers. The enhanced respiration rate during heat stress is critical for body temperature maintenance. However, the increased alveolar ventilation rate, necessary for evaporative cooling, also results in carbon dioxide loss and acid-base perturbations (Teeter *et al.*, 1985). Specific consequences of the acid-base perturbations have indeed been speculative. Teeter *et al.* (1985) reported that manipulating sodium : chloride ratios by addition of calcium chloride increased body weight gain 8% and slightly reduced severity of alkalosis. The results indicate that blood alkalosis limits growth rate of broiler chicks reared under chronic heat stress and that the respiratory alkalosis and weight gain depressions can partially be alleviated by diet.

Heat stress increased plasma Na^+ to Ca^{2+} ratio, heat exposure (24 increasing to 29° and 29° increasing to 37°C) decreased Ca^{2+} with only 40-50% of the decrease accounted for by the increase in blood pH. Changes in body temperature and Ca_2^+ followed a similar pattern. The magnitude of decrease in Ca^{2+} seemed to be negatively related to tolerance of chickens to heat stress. Thus maintaining Ca^{2+} improved heat tolerance (Ait-Boulahsen *et al.*, 1992). Heat stress increased urinary excretion for potassium, sodium, zinc and molybdenum and increased fecal excretion for calcium, manganese, selenium and copper. Mineral retention for magnesium and phosphorous was reduced by a combination of urinary and fecal excretion (Belay *et al.*, 1993). Heat stress decreased plasma concentration of Na^+

and K⁺ when broiler chickens were reared in thermoneutral (24°C) or cycling heat stressing environments (24-35-24°C) (Deyhim and Teeter 1994). Whereas heat distress (from days 32 to 49 subjected to 35°C for 6 h/day) did not significantly affect muscle and spleen trace mineral concentrations (Deyhim *et al.*, 1995).

Jamadar and Jalnapurkar (1995) observed that mean serum Fe concentration of chicks exposed to a high ambient temperature (40 ± 1°C) for 12 h of the day (07.00 to 19.00 h) was lower than those maintained at the prevailing room temperature (22 to 31°C). They suggested that possible release of corticosteroids during heat exposure might have caused low serum Fe concentration and distribution of Fe to the organs leading to higher organ Fe concentration. Salvador *et al.* (1999) reported decreased serum levels of K⁺, Na⁺, Cl⁻ as a consequence of high ambient temperature. Lin *et al.* (2000a) found no significant changes in plasma Ca concentration, by heat exposure (35°C) whereas temperature (10, 20, 30, 33°C) had significant effect on the levels of K and Cl. Chloride contents were increased at high temperature (33°C) (P<0.01), and K level was decreased by high (33°C) or low (10°C) temperature and increased at medium temperature (30°C) (P<0.01). The humidity (35, 85% RH) only had a significant effect on Cl concentration, which was decreased, by high humidity levels (P<0.01). Temperature and humidity had no significant effect on K concentration.

Trace minerals act as keys, which unlock the ability of the immune system to ward off invaders. Proper trace mineral supplementation will not eliminate disease, but it will allow the animal's immune system to respond with peak efficiency to minimize the risk of significant economic losses (Berger, 1996), hence it may be concluded that any phenomena causing a disturbance in mineral component of body may affect the immunity. Chicks inoculated with *S. gallinarum* had increased (P<0.01) survival when iron (100 ppm of diet or more) was added to a basal diet containing 200 ppm of iron. Anemia was found in the diseased chicks three days post-infection and continued through day nine as measured by decreased hemoglobin and hematocrits. However, birds receiving additional iron, up through

600 ppm, had less severe anemia and increased antibody titres. These and other data in broilers show that once the infection has occurred, increased supplemental iron enhances the immune system in destroying the invading organism.

Colnago *et al.* (1984) challenged male broiler chicks with *E. tenella* oocysts from 22 to 32 days of age in six experiments to determine if selenium would affect the ability of the birds to cope with coccidiosis. Graded selenium levels from 0.1 to 1.0 ppm or 100 ppm of vitamin E/kg were added to the diet from day one of age. Dietary supplementation of at least 0.25 ppm selenium or vitamin E reduced mortality ($P < 0.05$) and increased weight gains ($P < 0.05$). They observed that feeding 0.25 ppm selenium or more, increased leukocyte numbers in the blood after infection with coccidia and may explain the immune enhancement. Southern and Baker (1983) reported that adding 50 ppm zinc to broiler diets containing 40 ppm zinc increased ($P < 0.05$) gain and feed efficiency when infected with *E. acervulina*. Substantial evidence has been reported that adding zinc above the supposed requirement enhances disease resistance in chickens.

Stahl *et al.* (1984) reported that the immunocompetence of progeny chicks from hens was affected by dietary zinc. White Leghorn breeding hens were fed a corn-soy diet supplemented with 0, 10, 20, 40 or 150 ppm zinc. Progeny from unsupplemented hens had reduced titers to sheep red blood cells compared to those receiving the 10 and 20 ppm zinc treatments. However, excessive zinc (150 ppm) also depressed the immunocompetence of the progeny. Dietary levels of minerals can affect immunocompetence. While deficient levels of sodium and chloride decrease humoral immunity, levels of these nutrients which supported maximum growth also supported maximal humoral immunity. Low dietary zinc levels did not affect indicators of immunocompetence in the chick. Several indicators of immune responsiveness are depressed when chicks are vitamin E and/or selenium deficient (Latshaw, 1991).

2.12. Hormones

2.12.1. Triiodothyronine (T₃) and Thyroxine (T₄)

Decrease in thyroid size (Chiasson and Combest, 1979) and lowerd secretion with high environmental temperature have been well documented (Mueller and Amezua, 1959; Stahl and Turner, 1961; Huston *et al.*, 1962; Cogburn and Harrison, 1980) when young broilers were fed T₃ and T₄ and heat stressed. Pineal gland is associated with regulation of body temperature. Thyroid response in respect to triiodothyronine (T₃) of pinealectomized cockerels to different ambient temperatures was investigated by Cogburn and Harrison (1980). Pinealectomized, sham operated and controlled cockerels were randomly assigned to three ambient temperatures of 7, 23 and 37°C at 8 weeks of age and blood was taken from birds on 0, 4, 8, 12 and 16 days of the exposure period. Results showed that serum T₃ measured by radioimmunoassay technique was depressed significantly in all birds held at 37°C environment. The average values for the T₃ at 7° and 23°C environment were 410.2 ng/ml being higher than that noted for birds kept in the hot environment i.e. 261.3 ng/100 ml. Pinealectomized birds had a slightly higher average T₃ concentration of 370.6 ng/100 ml as compared to sham operated of 350.5 ng/100 ml. This difference was caused mainly by higher serum T₃ concentration in pinealectomized cockerels at 7° and 37°C but not at 23°C. It was concluded that serum T₃ was depressed in birds held in the hot environment.

Rudas and Pethes (1984) reported that no change in T₃ concentration was found after exposure to 35°C for 1 hour. The involvement of thyroid gland in the response of young chickens to heat stress was examined by Bowen *et al.* (1984). The levels of thyroxine were observed in male broiler chickens by maintaining at 35°C at the first week and then decreasing 2.8°C weekly and thereafter with a constant temperature of 27°C. Birds were heat stressed at 3 or 4 weeks of age by keeping them in a forced draft heat chamber with a temperature maintained at 50±1°C for 6 hours and were injected with different levels of thyroxine i.e. 12, 18 or 24 hour before exposing them to heat stress. The experiment proved that the ability

of young chicken to withstand heat stress is decreased under hyperthyroidism and increased under hypothyroidism. Feeding T_3 caused reduction in serum T_4 concentration but T_3 dieting had no effect on T_3 concentration. In another study of El-Gendy *et al.* (1995) plasma T_3 level of the heat stressed broilers at 6 weeks of age was significantly lower than that of control group. Similar results were presented by Gu. *et al.* (1995) in hens.

Conditioning at an early age chicken exposed to heat stress ($36\pm 1^\circ\text{C}$, 70 to 80% RH) for 24 h at the age of 5 (group 1), or 5 and 7 days (group 2), resulted in haemodynamic changes (significant decrease in heart weight and haematocrit) and reduced plasma triiodothyronine (T_3) concentration. It was suggested that reduced T_3 and haemodynamic changes may be part of the mechanism associated with improved thermotolerance by early age temperature conditioning (Yahav *et al.*, 1996). Gu *et al.* (1999) reported that plasma T_3 level decreased significantly at 30°C , compared with the control group. Whereas humidity had no effect on plasma T_3 . Similarly Lin *et al.* (2000b) found that plasma T_3 level was declined by high temperature (33°C), and this phenomenon disappeared in birds under high temperature and high humidity. T_4 concentration in plasma was not affected by temperature, but increased by high or low humidity.

Barbosa and Compos (1992) founds no effect of dietary ME level on serum concentration of free triiodothyronine. Gonzales *et al.* (1998a) reported that during the period of feed restriction, plasma T_3 and IGF-I concentrations decreased whereas plasma T_4 and growth hormone (GH) increased compared with those of the age-matched *ad libitum* fed counterparts. Junqueira *et al.* (1999) observed no correlation between plasma thyroid hormone (T_4 and T_3) concentration and energy intake (2600, 2900, and 3200 Kcal ME/kg).

2.12.2. Corticosterone

Heat stress stimulated the release of corticosterone from adrenal glands in chickens (Ben Nathan *et al.*, 1976), turkey (El-Halwani *et al.*, 1973) and pigeons (Pilo *et al.*, 1985). El-Halawani *et al.* (1973) studied the response of 9-week old

male turkeys to acute and chronic cold and to warm environments compared with normal temperature. High temperature increased corticosterone level. Edens (1978) found that acute heating (43°C) in chambers for periods up to 2 hr in duration caused acute adrenal cortical insufficiency (AACI) to develop in young chicken. The development of AACI in heat stressed broiler cockerels was characterized by a sharp increase in levels of plasma corticosteroids, which was followed by a rapid decline. The decline of plasma corticosteroid levels was associated with significant reduction of adrenal cortical levels of corticosteroids. The development of AACI in heat stressed chickens may predispose them to irreversible heat prostration and death due to cardiovascular failure.

Seasonal changes in concentration of corticosterone have been determined in wild birds, in natural and outdoor environment. Gould and Siegel (1985) studied the effects of corticotropin and heat on serum corticosterone in 6-8 weeks old White Plymouth Rock chickens. Total corticosteroids were measured in chicks, injected adrenocorticotropin hormone (ACTH). Four experiments were conducted with different doses of ACTH at environmental temperatures of 31 and 46°C. A single heat episode consisting of three 30 minutes intervening periods at constant temperature caused significant increase in serum corticosteroids. The birds in which serum was sampled after 2 heat exposures consisting of 11 hours intervals, corticosterone levels were not increased significantly.

The effect of heat stress on the corticosteroid in chickens and quails was studied by Bowen and Washburn (1985). Groups of chickens that were handled on each of 4 days and non-handled control groups were bled on the 5th day with or without heating for 1 hour at 50°C. Corticosterone levels noted in non-heated handled and not handled birds on average were 6.1 and 11.2 ng/ml respectively, whereas the corticosterone levels in the heated handled and not handled birds were found to be 13.8 and 22.5 ng/ml, respectively. Handling decreased the corticosterone levels, while heating increased its levels but magnitude of the increase in heating was similar for both groups. It was concluded that increased resistance to heat stress in the handled birds was associated with the decreased pre-

heating corticosterone levels of the handled birds. Heating increased corticosterone about 2 fold in both the handled and non-handled groups, because the basal corticosterone level of the handled groups was lower, the same percentage increase resulted in lower levels in the heated groups.

Heat stress under cyclic temperature (24 to 35°C) resulted in to increased plasma corticosterone by 53% (Deyhim and Teeter, 1991). Increased corticosterone due to heat stress was also reported (Yang *et al.*, 1992, Jamadar and Jalnapurkar, 1995) in broilers. Sokolowicz and Herbut (1998) conducted a study on broilers, allocated to 2 equal groups and reared for 49 days. Group 1 chickens were reared at 33°C from day 1, which was reduced to 18°C by day 49 (optimum temperature); group 2 chickens were reared at +10°C higher than the optimum temperature throughout the rearing period. Twenty chickens from each group were killed every week to day 49. Overall, adrenal gland weights were higher in group 2 than group 1 chickens, and similar trend for blood corticosterone concentrations and blood glucose levels were observed.

Freeman *et al.* (1981) reported that plasma corticosterone concentration after 1 week of restriction was 73% greater than in controls (non restricted group) which decreased progressively, falling within the normal range at 5 weeks. Similarly blood corticosterone concentration was found to be significantly ($P<0.01$) higher on skip-a-day (SD) feed restriction regimen from 4 to 15 weeks old than in controls. It was concluded that SD feed restriction caused a persistent increase in plasma corticosterone in broiler breeders (Mench, 1991). Latshaw (1991) observed that feed restriction caused higher plasma corticosterone levels, which were known to decrease the immune response, possibly through effects on cytokinase.

2.13. Immune Response

High environmental temperature adversely affects the immune response in birds. Suba-Rao and Glick (1977) observed the effect of various environmental temperature changes on the antibody production of chickens. Birds exposed to 32.2°C and above had significantly depressed agglutinin levels. Short term cold

exposure 2 or 4 times following the antigen injection enhanced the agglutinin and hemolysin response. Thirty minute cold exposure for 2 or 4 times significantly increased IgM antibody production and markedly reduced the IgG antibody.

Edens and Siegel (1974); Edens (1976) investigated the role of adrenal medulla of chickens in response to acute exposure to high environmental temperature. He demonstrated that during the course of heating blood, levels of epinephrine and calcium increased. Latshaw (1991) reported that immune response changed metabolism resulting into decreased growth thereby decreasing the need for amino acids during heat stress. Dietary levels of minerals can also affect immunocompetence. Which indicates that mineral loss due to any reason may lead to same situation. El-Gendy *et al.*, (1995) observed that antibody titre of the heat stressed broilers against Newcastle disease vaccine was lower than that of broilers kept under normal temperatures. The effect of heat stress on immune organs was studied by Zhang *et al.* (1998), who reported that weights of the spleen, bursa Fabricii and thymus and the ratio of their weights to body weight in the test group were significantly lower than those of the control group. Stannius follicles of bursa Fabricii in the test group were small and few. There were other abnormal features of the immune organs in the heat stressed group.

Spalatin and Hanson (1974) found that hens deprived of water and food significantly altered their reactivity to Newcastle disease. A lower antibody titer was found by Ben Nathen *et al.* (1976) due to feed and water withdrawal. Latshaw (1991) observed that feed restriction causes higher plasma corticosterone levels, which are known to decrease the immune response, possibly through effects on cytokines. Relationships among short-term fasting, heat stress and response to infectious bursal disease (IBD) vaccinations were studied in 144 broiler chicks by Zulkifli *et al.* (1997). Starting at 28 days of age, for 14 days, birds were fasted from 09.00 h to 17.00 h or fed *ad libitum*, were exposed to high ambient temperatures ($36\pm 2^{\circ}\text{C}$) from 12.00 h to 17.00 h or were untreated controls (minimum 25°C ; maximum, 34°C). Live IBD vaccine (Nobilif strain D78) was administered

intraocularly at 14 and 28 days of age. At 35 days of age, while feeding regimen had negligible effect on immune response, the heat treatment suppressed antibody titres. At 42 days of age, feed-restricted chicks had a higher antibody response to IBD vaccinations than those fed *ad libitum*.

During heat stress, birds supplemented with 500 ppm ascorbic acid developed increased IBV antibody titres. The serum corticosterone concentration was lowered in these birds. There was a positive correlation ($R^2=0.95$) between antibody titres and ascorbic acid concentration and a negative correlation ($r^2=.54$) between the serum corticosterone concentration and the antibody titres (Tuekam *et al.*, 1994). Friedman and Sklan (1995) observed that when chicks were fed four diets containing 12% added fat made up of different proportions of palm oil and soybean oil and immunized against bovine serum albumin at 14 to 16 d of age, antibody production developed more rapidly, reached a higher level, and was more persistent in the chicks fed lower levels of linoleic acid. It was concluded that dietary fatty acid composition could influence immune response in broilers.

Zouelfakar and Moubarak (1998) studied the effect of nutrition on the immune response of broilers; first group (high energy treatment) was fed on a starter diet (24% CP, 3300 Kcal/kg ME), then on a grower diet (21% CP, 3100 Kcal/kg ME) followed by a finisher diet (19% CP, 3100 Kcal/kg ME). Whereas the second group (conventional energy control) was fed on a pre-starter diet (24% CP, 2900 Kcal/kg ME) then on a starter diet (21% CP, 2800 Kcal/kg ME) and a grower diet (19% CP, 2800 Kcal/kg ME). The sustainability of maternal immunity against Newcastle disease and infectious bursal disease virus was not affected by dietary treatment. The non specific immune response to sheep red blood cells was significantly higher in birds fed on high energy ration; the stimulation index of lymphocyte transformation was also significantly increased compared with those of the conventional energy treatment.

Keeping in the view the above mentioned review of literature, it may be concluded that weight gain, feed consumption, feed conversion ratio, body

temperature, mineral composition of meet and immune response are the factor which are effected by environmental temperature. Whereas, use of appropriate feeding method may help to appease the environmental stress and thus may improve production performance of broilers during summer.