IMPLICATIONS OF VARYING ELECTROLYTES (SODIUM, POTASSIUM AND CHLORIDE) AND THEIR BALANCE ON GROWTH PERFORMANCE AND PHYSIOLOGICAL RESPONSES OF BROILERS

BY

Mirza Muhammad Haroon Mushtaq
B.Sc. (Hons.) Animal Husbandry
M.Sc. (Hons.) Animal Nutrition

2007-VA-559

A THESIS SUBMITTED IN THE PARTIAL FULFILLMENT OF THE REQUIREMENT FOR THE DEGREE

OF

DOCTOR OF PHILOSOPHY

IN

ANIMAL NUTRITION

DEPARTMENT OF FOOD AND NUTRITION
FACULTY OF ANIMAL PRODUCTION & TECHNOLOGY
UNIVERSITY OF VETERINARY AND ANIMAL SCIENCES, LAHORE-PAKISTAN

2010

To

The Controller Examinations,
University of Veterinary and Animal Sciences,
Lahore.

We the supervisory committee, certify that the contents and form of the thesis, submitted by **Mr. Mirza Muhammad Haroon Mushtaq, Registration No. 2007-VA-559** have been found satisfactory and recommend that it should be processed for the evaluation by the External Examiners for the award of degree.

Supervisor: _____
Prof. Dr. Talat Naseer Pasha

Member: _____
Dr. Saima

Member: _____
Prof. Dr. Muhammad Akram

In the name of almighty "**ALLAH**" who is **RAHMAN**

and **RAHEEM**

Oh Lord,

Make me

An Instrument of Your **Peace**

Where, there is **Hatred**

Let me Show **Love**

Where, there is Injury, **Pardon**

Where, there is Doubt, **Faith**

Where, there is Despair**, Hope**

Where, there is Darkness, **Light**

And where, there is Sadness, **Enjoy**

DEDICATED TO

HOLY PROPHET (PBUH)

The Greatest Social Reformer

To,

My beloved

MOTHER

My affectionate

FATHER

Who taught me

The first word to speak,

The first alphabet to write,

&

First step to take.

To,

Those who live in my mind,

In my heart,

Throughout the whole span of my life,

And are **nearest, dearest** and **deepest**, to me.

ACKNOWLEDGMENTS

All acclamations and appreciation are for ***ALMIGHTY ALLAH***, the Omnipotent, the omnipresent, the Merciful, the Beneficent, who presented me in a Muslim community and also bestowed and blessed me with such a lucid intelligence as I could endeavor my services towards this manuscript and who gave me an opportunity to add a drop in the wide ocean of knowledge.

This research work sounds incomplete without acknowledging the worthy guidance and cooperation extended by my supervisor **Prof. Dr. Talat Naseer Pasha**, Dean, Faculty of Animal Production and Technology, University of Veterinary & Animal Sciences, Lahore. His encouragement, support, guidance, valuable suggestion, boundless forbearance, indefatigable help with anything, anywhere, anytime, consummate advice and thought-provoking instructions in piloting this research venture and keen supervision made this work possible to reach its present effective culmination.

My greatest thanks go to my respected member of the supervisory committee, **Dr. Saima**, Assistant Professor, Department of Food and Nutrition, University of Veterinary & Animals Sciences Lahore and **Prof. Dr. Muhammad Akram**, Chairman, Department of Poultry Production, University of Veterinary & Animal Sciences Lahore, who are always a source of inspiration and continuous encouragement especially during hard and desperate phases of this research. Special thanks for them would always be due.

I would like to express my thanks to **Mr. Shahid Mahmood**, Lecturer in Department of Poultry Production, and **Mr. Umar Farooq**, Ex-Lecturer in Department of Poultry Production (Now PhD Fellow, University of Western Australia, Australia) for their cooperation, and guidance during the period of my academic studies at this prestigious institution

I especially thanks to my dear friend **Khalid Javed Iqbal**, who is always a source of continuous encouragement especially during hard and desperate phases of this research.

My acknowledgement is incomplete without thanking to my family for their everlasting love and support. They always acted as a light house for me in the dark oceans of life path. No words can really express the feelings that I have for my small family. The name of these people will always be in front of my eyes, as I will look on the cover of my thesis, even though my name may be printed on it.

*(May **Almighty ALLAH** bless them all)*

(Ameen)

Mirza Muhammad Haroon Mushtaq

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INTRODUCTION

Electrolytes are the substances dissolved into positive and negative particles in solution (acidic or basic) and known to maintain acid base balance and osmotic pressure within the body fluids of birds. Sodium (**Na^+**) is the principal cation of extracellular fluid and is known to involve in the regulation of extracellular fluid volume, basal metabolism acid base balance, membrane potential of cells etc (Elzbieta *et al*., 1979; Ruiz-Lopez *et al*., 1993; Nobakht, 2005). Concentration of Na in extracellular fluid is maintained through Na/K-ATPase pump which expels Na from the cell (Leeson and Summers, 2001). Dietary Na (**dNa**) and chloride (**dCl**) are inexpensive in terms of meeting requirements as Pakistan has huge reserves of sodium and chloride (**NaCl**) which are playing crucial role in body physiological responses and litter moisture contents. NRC (1994) recommended high levels of Na in prestarter phase of broiler's life, keeping in view the low level of feed intake and excreta during this period and, later on, confirmed by Maiorka *et al.,* (2004). Potassium (**K^+**) is found abundantly in animal body especially in muscles and nerve cells (Leeson and Summers, 2001). Actually these cations and anions are closely linked with the conditions of alkalosis and acidosis in birds. Most of the feed ingredients especially vegetable protein sources are rich in potassium. The dietary K (**dK**) has been studied extensively with other nutrients like Na, Cl, protein, amino acids (i.e., lysine, methionine, arginine etc.) and energy (Leach *et al.*, 1953; Stutz *et al.*, 1971; Scott and Austic, 1978; Reece *et al.*, 1984; Ribeiro *et al.*, 2008; Roussan *et al.*, 2008). It has been reported that K-rich diets favorably affect acid-base balance owing to precursors of bicarbonate, which neutralize acidogenic conditions in the biological fluids (Sebastian *et al.*, 1994, 2002). In recent research work, high levels of dK were used (Smith and Teeter, 1992; Mushtaq *et al*., 2005; 2007; Ahmad *et al*., 209) than the recommendations of NRC (1994) i.e. 0.30% and were indicated as because of larger muscle development and difference in environmental factors. Water intake and litter moisture are directly linked with the amount of dNa (Mongin, 1980) while dK and dCl did not play significant role *per se* in this regard (Vogt, 1971). It is anticipated that addition of cations (Na^+/K^+) could be used to compensate depressed growth in chickens caused by high levels of anions (Cl^-/HCO_3^-) and vice versa (Leach *et al.*, 1960; Nesheim *et al.*, 1964). Mongin (1980) considered the effect and interrelationship of Na^+, K^+ and Cl^- into an equation, called dietary electrolyte balance (**DEB** = Na+K-Cl, mEq/kg diet), and various researchers (Hulan *et al.*, 1985; NRC, 1994; Teeter and Belay., 1995; Murakami *et al.*, 2000; 2001; Rondon *et al.*, 2001; Borges *et al.*, 2003 ; 2003a; 2004 ; 2004a; Mushtaq *et al.*, 2005; 2007; Ahmad *et al.*, 2005; 2006; 2008)

recommended this balance for broilers of different ages, ranged from 150 to 350 mEq/kg with various combinations of Na^+, K^+ and Cl^-. These electrolytes and their interactions had been studied with reference to environment especially temperature and relative humidity. Under heat stress condition, the demand for Na and K are reduced while Cl demand increased and resulted in low blood electrolyte balance (Belay and Teeter, 1993). Brody (1999) recommended a range of pH from 7.35 to 7.45 for better growth efficiency in broilers. High DEB value (>300 mEq/kg) is made responsible for high litter moisture contents (Borges et al., 2003a; 2004a). However, Rondon *et al.* (2000) and Borges *et al.* (2004a) reported different DEB values for it was changed with different ions. Similarly, Mushtaq *et al.* (2005; 2007) reported that different ions acted differently on a similar DEB of 250 mEq/kg. Further to that most of the previous studies were conducted with special reference to heat stress in broiler chicken.

Addition of various salts is anticipated to change the bird's osmotic balance and this change is mainly influenced by disturbance in contributing electrolytes and leads to change in water consumption and excretion (Smith and Teeter, 1988; Borges *et al.* 2004; 2004a). Na-bicarbonate (**$NaHCO_3$**) is considered as the first-rated supplemental salt, being a source of Na^+ and HCO_3^- when compared with other ionic salts (Teeter *et al.* 1985; Gorman and Balnave, 1994; Johnson and Karunajeewa, 1985; Hooge, 2003; Mushtaq *et al.*, 2005; Ahmad *et al.* 2005; 2006). For example, Na-sulphate (**Na_2SO_4**) is known to induce blood acidosis with great severity when compared with other sulphate sources like potassium sulphate (**K_2SO_4**) or copper sulphate hence acidic properties of sulphates is directly linked with the supplemental salt (Ruiz-Lopez and Austic, 1993; Ahmad *et al.* 2006). Potassium carbonate (**K_2CO_3**) is considered as a good source of dK in broiler chicken (Hooge, 2003) especially under heat stress condition as it provides. Contrarily; better growth performance, water intake and high litter moisture were noted in potassium bicarbonate supplemented diet compared to K_2CO_3 or K_2SO_4 at a constant DEB of 250 mEq/kg under heat stress conditions (Ahmad *et al.*, 2005). Dietary Cl in calcium chloride (**$CaCl_2$**) is considered to be exchanged with bicarbonate ion in the lower digestive tract and make the whole environment acidic with the excretion of calcium as calcium carbonate (Mongin, 1981). Ahmad *et al.* (2005) indicated better growth performance and lower pH as a result of ammonium chloride (**NH_4Cl**) supplementation in heat-distressed broilers.

As modern poultry industry is shifting towards environment control systems so the nutrient requirements of today's broilers are changing with respect to varying growing potential, hence it is imperative to reconsider the requirements of dietary electrolytes and electrolyte salts with the concept of DEB for broilers fed under phase feeding system. The present study was, therefore, envisaged to evaluate the effect of supplementation of dietary electrolytes with

the applicability of the DEB using different salts on growth performance, carcass characteristics, body physiological responses, litter condition and serum mineral chemistry of the modern broiler strain reared under phase feeding program. In this way, we may be able to define best level of each electrolyte and salt source for each feeding phase of broiler's life.

Chapter 2

REVIEW OF LITERATURE

Electrolytes

Electrolytes are defined as the compounds that dissolve into positive (cation) and negative (anion) particles in solution and have the inherent ability to conduct an electrical current. The three elements that are predominant in satisfying the electrolyte definition in the poultry are sodium (**Na**), potassium (**K**) and chloride (**Cl**). All classes of poultry have definite requirements for these elements in the correct amounts/ ratio for acid-base homeostasis of body physiological system. Mongin (1981) studied the basic role of cation-anion balance for chickens and concluded that the electrolyte equilibrium could be described in a combination including these electrolytes. Actually these cations and anions are closely linked with the conditions of alkalosis and acidosis, respectively, of birds. The net alkalosis and acidosis could be measured by analyzing these ions in diet and then in the faeces. It is speculated that other than cations and anions, a third expression is also known to play crucial role in the acidosis and this expression is defined as endogenous acid production (**H^+ endo**). This endogenous acid production is usually related with the metabolism of dietary protein and amino acids (Mongin, 1981). All essential nutrients other than these electrolytes such as the amino acids, vitamins, minerals and oxygen must be continually supplied to maintain the well-being of poultry birds, plus obtain better growth and foster production efficiency. Likewise, sodium, potassium and chloride, which fall in the category of strong electrolytes, must be continually supplied to procure the desired production goals with all classes of poultry.

Sodium is the major cation in extracellular fluid and is closely associated with chloride and bicarbonate in managing acid-base balance (Ruiz-Lopez *et al.,* 1993), water intake (Mongin, 1980) and basal metabolism (Elzbieta *et al.,* 1979). Sodium works for the regulation of osmotic pressure and protection against excessive loss of body fluid. The permeability of cells and the sustentation of normal muscle irritability are additional functions of sodium (Nobakht, 2005). Sodium is also obligatory for absorption of amino acids and sugars in the small intestine, therefore utilization of digested protein and carbohydrates is diminished with an insufficiency of this cation. Likewise, sodium and chloride, both, aid in nutrient passage and waste removal in cell nourishment and maintenance. While sodium is found deficient in the diet, it will lead towards acid-base imbalance, reduced cardiac output, growth reduction, increased feed conversion, gonadal dormancy, bone-softening, corneal keratinization, and

decreased cellular volume and changes in cellular function. Diets deficient in sodium also lead towards adrenal malfunction and this impaired function results in increased uric acid levels which may cause shock and ultimately death in chicks (Leeson and Summers, 2001).

Meeting the requirements of the chicken for sodium and chloride is relatively easy in terms of price but body physiology is quite sensitive for both these ions *per se*. Both these electrolytes are cheaper especially in Pakistan where huge reserves of sodium and chloride (NaCl) are found. Chicken maintains its blood and tissue sodium homeostasis by excreting excess sodium and conserving short supply in diet, being hypotonic in nature (Leeson and Summers, 2001) However, the impact of these elements on litter moisture content places great emphasis on minimizing excessive levels in meeting the requirements of the chicken (Borges *et al.*, 2004; 2004a; Mushtaq *et al.*, 2005; 2007). Litter dampness is related with the amount of sodium and chloride and increased litter moisture put great emphasis on minimizing excessive levels in the diet. Excess water consumption contributes to increases in litter moisture and could be affected by the sodium concentration in the feed. Therefore, nutritionists in general are very cautious on the use of high levels of sodium in feeds. However, this argument may be of little application for a pre-starter feed, considering the small amount of feed intake and excreta produced in this period and high requirements during initial days of broiler's life (NRC, 1994). It is demonstrated that dehydration is a common problem in commercial poultry operations because large span of time generally passes from the time the chick hatches until its delivery to the broiler house. Evaluation of sodium requirements in this period should be considered carefully as higher levels of sodium than those usually accepted for initial stages of age would benefit the chick by inducing greater water intake. National Research Council (NRC; 1994) recommended 0.20 and 0.15% sodium and chloride level for the 1-3 and 4-6, respectively, weeks aged broiler. However, various researchers studied in different pattern, considering electrolyte, other nutrients, ambient conditions etc. Hence, it is anticipated that with the improving genetics and housing conditions, the requirement for the sodium and chloride should also be re-evaluated to fulfill the body requirement of modern-day broilers.

Potassium is the third most verdant element of the animal body (McDowell, 1992) and main intracellular cation as well. Blood, muscle and nerve cells are rich places of potassium reserves in the body. It participates in the processes that are essential to the body homeostasis such as acid-base equilibrium, osmotic pressure regulation, development of membrane potentials of cells (nerve transmission), and glucose and amino acid absorption and transport (Rinehart *et al.,* 1968; Reece, 1984; Leeson and Summers, 2001). It is conspicuous in the activation of a number of enzymes that play role in muscle activity, most eminently cardiac muscles. Potassium is extremely important for optimal nitrogen retention owing to a consistent ratio between nitrogen and potassium in fresh tissues (Leeson and Summers, 2001).

The reported requirement for potassium in relationship to nitrogen is 5mEq/g of amino acid nitrogen for optimal retention in poultry. Many poultry diets are marginal in potassium especially if animal protein comprises a large percentage of the diet protein. Additionally, the potassium availability from natural feedstuffs may be as low as 73 percent, leading to borderline conditions in certain rations during times of stress or for maximum processing yield of breast meat.

Potassium does not work alone *per se* therefore a correct balance with other electrolytes (Na and Cl) is necessary for higher level of animal performance, nutrient utilization and bone development (NRC, 1994). Hooge and Cummings (1995) stated that NRC (1994) recommended potassium dietary requirements for growing poultry (0.30%) were considerably lower than the levels typically present in commercial diets (up to 1.%; Table 2.1). However considerable rise in dietary potassium are now considered due to larger growth potential and muscle development. Difference in K requirements were indicated when environmental temperature, water intake, or stress conditions are increased (Smith and Teeter, 1992; Mushtaq *et al.*, 2005; 2007; Ahmad *et al.* 2009).

NRC (1994) recommended 0.30% potassium level for the whole life cycle of broiler, however, it is anticipated that with the improving body weight and better feed utilization, the requirement for the potassium would be increased from 0.30 to about 1.20% to fulfill the body requirement of modern-day broilers (Table 2.1). So, a big gap exists between NRC (1994) recommendations and various research work done in recent past. Rostagno *et al.* (2000) suggested that 0.50, 0.47 and 0.45% K were adequate for DEB in the periods from 1 to 21, 22 to 42 and 43 to 49 d, respectively. Other researchers estimated 0.82% (DEB=242 mEq/kg) as the potassium requirement from 7 up to 21 d, keeping levels of sodium from 0.15-0.17%, in a practical-type diet in broilers (Hurwitz *et al.,* 1973 cited by Hooge and Cummings, 1995). Thus, this information suggested that the K requirement for maximum body weight gain could be higher than the 0.30% level recommended by NRC (1994). It is still unclear how K supplementation improves BW, but it has been suggested that diets with high levels of lysine-HCl and arginine-HCl might need K supplementation as acetate or carbonate to minimize the antagonism between these ions on metabolism (O'Dell and Savage, 1957, O'Dell *et al.,* 1962; Nesheim *et al.,* 1964; Savage, 1972). Potassium salts were also shown to influence lysine catabolism, resulting on lower lysine:arginine ratio (Scott and Austic, 1978). The protein synthesis and BWG reduced tissue lysine concentration. This might be due to reduced arginase activity in the kidneys and lower urease activity in the intestines (microbial source). The arginine catabolism would then be reduced and this amino acid would become more available for protein synthesis (Stutz *et al.,* 1972). Young (1995, a personal communication with Hooge and Cummings, 1995) stated that K levels in some

ingredients were found to be considerably lower than expected compared with standard tabulated values, which in turn alter K levels and ultimately performance of the birds.

NRC (1994) recommended 0.20 and 0.15% level of chloride for starter (1-3 week) and finisher(4-6 week) phase of broiler age, while in recent past various workers recommended various values of chloride according to the age, environmental conditions (temperature and relative humidity) and dietary composition (protein composition particularly; Table 2.1). Miller and Soares (1972) observed better growth rate with decreasing Cl contents (from 0.54 to 0.23%). Later on, Mongin and Sauveur (1974) similarly reported a steep decline of weight gain in a semi-purified diets with increased level of Cl (i.e., 0.35%, 1.01% and 1.41%) when Na and K were kept 0.22% and 0.39%, respectively. Somewhat less steep decline was observed when Na and K were kept 0.40% and 0.68% and was almost prevented by 0.57% Na and 0.97% K. It means dietary cations should be balanced with dietary anions in order to keep the acid base balance in a normal range. A higher level of Cl was noticed to reduce weight gain by 16 to 22% (Ruiz-Lopez *et al.,* 1993). Borges *et al.* (2004a) found better performance when manipulated electrolytes were Na and Cl. He observed reduced performance in birds with high Cl (0.73%) and birds showed blood acidosis. This acidogenic condition would increase with increased supplementation of Cl that is evident from increased blood Cl contents (Ahmad *et al.* 2005). It is therefore demonstrated a wide range of Cl requirements in broilers i.e. 0.12-0.91% (Edwards, 1984; NRC, 1994; Rondon, 1999; Borges *et al.*, 2004) keeping in view other electrolytes in optimum range.

Table 2.1. Electrolytes and dietary electrolyte balance discussed in published data of various researchers in peer-reviewed journals

Reference	*Year*	*Age (Wk)*	*Na (%)*	*K (%)*	*Cl (%)*	*DEB (mEq/kg)*	*Journal*
Mushtaq *et al.*	2005	0-4	0.20, 0.25, 0.30	0.80 to 1.19	0.30, 0.40, 0.50	250	**PS**
Mushtaq *et al.*	2007	4-6	0.20, 0.25, 0.30	0.80 to 1.19	0.30, 0.40, 0.50	250	**JAPR**
Ahmad *et al.*	Basal 2005 Test	1-4	0.20	0.71	0.30	185	**AFST**
		4-6	0.15	0.67	0.23	174	
		1-4	0.20, 0.35	0.71, 0.97, 0.98	0.78, 0.31, 0.30	50 250	
		4-6	0.15, 0.33	0.67, 0.97, 0.98	0.67, 0.23, 0.24	50 250	
Ahmad *et al.*	2006	1-4	0.35	0.71	0.30	250	**BPS**
		4-6	0.33	0.67	0.23		
Ahmad *et al.*	2009	1-6	-	-	-	0, 50, 150, 250, 350	**JAPAN**
Rondon et al	2001	1-3 (Exp-1)	0.10, 0.15, 0..20, 0.25, 0.30, 0.35	0.95	0.20	232, 254, 286, 299, 321, 344	**PS**
		1-3 (Exp-2)	0.20	0.95	0.10, 0.15, 0..20, 0.25, 0.30, 0.35	305, 290, 276, 262, 248, 234	
Murakami et al	2001	3-6 (Exp-1)	0.10, 0.15, 0..20, 0.25, 0.30, 0.35	0.93	0.15	240, 263, 285, 308, 330, 353	**PS**
		3-6 (Exp-2)	0.15	0.96	0.10, 0.15, 0.20, 0.25, 0.30, 0.35	285, 271, 256, 243, 228, 214	
Murakami et al	Basal 1997a	1-3	0.02, 0.40	0.94, 0.93, 0.97	0.02, 0.40	-	**JAPR**

	Test		0.10, 0.15, 0.20, 0.25, 0.30, 0.35		0.10, 0.15, 0.20, 0.25, 0.30, 0.35		
Vieira et al	2003	1-7d	0.12, 0.24, 0.36 0.48	1.14	0.65, 0.84, 1.02, 1.20	240	**JAPR**
			0.12, 0.24, 0.36 0.48	1.23	0.45, 0.63, 0.80, 0.97	160	
Oleviera et al	2005	8-21d 22-42d 43-53d	0.22 0.19 0.19	0.30, 0.44, 0.58, 0.72, 0.84, 1.00	0.30 0.28 0.28	88 84 82	**BJPS**
Borges et al	2004	21-42d	0.15, 0.25, 0.35, 0.45	0.68, 0.68, 0.68, 0.85	0.70, 0.50, 0.30, 0.26	40 140 240 340	**IJPS**
			0.15, 0.20, 0.25, 0.30	0.68, 0.87 0.97, 1.06	0.70, 0.59, 0.40, 0.21	40 140 240 340	

PS – Poultry Science; JAPR – Journal of Applied Poultry Research; AFST – Animal Feed Science and Technology; BPS – British Poultry Science ; JAPAN – Journal of Animal Physiology and Animal Nutrition; BJPS – Brazilian Journal of Poultry Science; IJPS – International Journal of Poultry Science

Understanding the Ideology of Electrolyte Balance Hypothesis

Dietary electrolyte balance plays a vital role in broiler performance and is known to alter acid base balance. Metabolic pathways cannot function with maximum efficiency without proper dietary electrolyte so its role is very important to maintain better physiological status of bird. Sodium and chloride along with potassium in proper concentration and balance are indispensable for a number of physiological and biochemical processes. Some authors have developed equations to explain the relationship between cations and anions and the acid-base balance. Melliere and Forbes (1966) described this interrelationship previously in the following equation with some other cations and anions:

Relative level = mEq cation = Ca + Mg + Na + K

mEq anion = PO_4 + Cl + SO_4

However, the ions that are considered essential for the maintenance of production performance of today's broiler type chicken are: Na^+, K^+ and Cl^-. In addition to the minimum amount the birds need in their feed to meet their nutritional requirements, the proportion among these ions is vital to maintain the acid-base homeostasis and to get the best productive performance from the broiler chickens (Mongin, 1981). In order to maintain the acid-base balance, birds have to regulate acid intake and excretion; and there are differences in the intake and excretion of anions and cations in the diet. Mongin (1981) studied the basic role of cation-anion balance for chickens and concluded that the electrolyte equilibrium could be described by a formula including three main electrolytes. The abbreviated formula resulted in the sum of the positive ions (Na and K) minus the sum of the negative ions (Cl) known as dietary electrolyte balance (**DEB**). Percentage of different electrolytes could be changed into its standard units (mEq/kg) by multiply them with different factors which were derived by dividing 10,000 with the respective electrolyte elemental weight.

$$DEB\ (mEq/kg) = \%\ Na \times 435 + \%\ K \times 256 - \%\ Cl \times 282$$

It was described many reasons of including these ions in the equation in which acid-base balance and bioavailability of these ions are key factors to be considered.However, acids produced in the metabolism (endogenous H^+) of proteins particularly also contribute to the acid–base balance. The following equation can be described where birds are in a constant acid–base balance, with either acid or base excess or deficiency:

$$(Anions - Cations)_{Ingested} + H^+_{endogenous} = (Anions - Cations)_{excreted}$$

According to Mongin (1981), the electrolytic (Na + K – Cl) intake is equal to the difference of excreted cations and anions $\{_{excreted}\ (Anions - Cations)\}$ plus production of endogenous acid ($_{endogenous}\ H^+$) and excess bases (BEecf).

$$(Anions - Cations)_{Ingested} = (Anions - Cations)_{excreted} + H^+ + BEecf_{endogenous}$$

Or it can be stated as,

$$(Na^+ + K^+ - Cl^-)_{Ingested} = (Anions - Cations)_{excreted} + H^+ + BEecf_{endogenous}$$

In these equations, it is assumed that only the strong ions (Na^+, K^+ and Cl^-) have an impact on the acid-base balance, without considering the way of ingestion (dietary or through water). Na^+ and K^+ supplementation increases pH and blood HCO_3^-, while Cl^- addition decreases these parameters (Hurwitz *et al.,* 1973). Gorman and Balnave (1994) concluded that the weight gain is associated with the source (Na-carbonate), and the amount of this source was significantly different in diets with the same electrolyte balance. In relation to the elements that are not considered in the summarized equation by Mongin (1981) it is speculated that bivalent cations are not as rapidly absorbed as monovalent cations; magnesium is commonly supplied in feeds; phosphate is hard to assay owing to difference in sources; calcium absorption rate is controlled by the endocrine system; sulphate is present in small amounts, being related to methionine catabolism prevention. So various issues created hurdles to consider other minerals into this equation (i.e. DEB = Na+K-Cl).

The interrelationship between mineral ions and other nutrients, such as between Na^+ and Cl^- and the arginine: lysine ratio is of great importance especially with respect to ambient temperature (Brake *et al.,* 1998). In broilers (3 to 7 weeks) kept under heat stress condition, the ratio between arginine:lysine was standardized at 1.34 when Na^+ and Cl^- were added according to NRC (1994) recommendations. However, when Na^+ and Cl^- contents in the diet were increased, the arginine:lysine ratio was below 1.05, thus showing that under heat stress, the adjustment of the electrolyte balance for maximum bird performance can be related to the composition of amino acids in the diet (Brake *et al.,* 1998).

In diets formulated with the electrolyte balance concept, varying levels of each electrolyte (Na^+, K^+ and Cl^-) affect the DEB value in a different way. DEB (mEq/kg) can be altered by simply changing one, two or three electrolytes with varying effect on production response.

Na 0.25% + K 0.80% - Cl 0.22% = 252 mEq/kg DEB
Na 0.25% + K 0.72% - Cl 0.15% = 251 mEq/kg DEB
Na 0.20% + K 0.85% - Cl 0.20% = 249 mEq/kg DEB
Na 0.25% + K 0.85% - Cl 0.25% = 256 mEq/kg DEB
Na 0.40% + K 0.50% - Cl 0.20% = 250 mEq/kg DEB

It means that the DEB value of 250mEq can be attained by varying the one or other values of each electrolyte. However, level of each electrolyte is also important as the level of DEB according to the physiology of the bird. Ingredient composition plays the pivotal role in the formulation e.g. by increasing the level of soybean level we can increase the level of potassium and chloride level by increasing fish meal in the formulation of poultry rations.

Electrolytes and Dietary Salts

Potassium chloride (**KCl**), hydrochloric acid (**HCl**), sodium chloride (**NaCl**), ammonium chloride (**NH_4Cl**), calcium chloride (**$CaCl_2$**) and sodium hydroxide (**NaOH**) are considered as strong electrolytes while sodium bicarbonate (**$NaHCO_3$**) and acetic acid (**CH_3COOH**) as weak electrolytes (Mongin, 1981; Hooge, 2003). The addition of various salts in the diet or water causes change in the birds' physiological performance and acid-base balance. The addition of salts to drinking water has been considered for the short-term treatment of heat stress in recent studies (Smith and Teeter, 1992; Beker and Teeter, 1994; Gorman and Balnave, 1994; Benton *et al.*, 1998; Roussan *et al.*, 2008). Dietary NH_4Cl at a concentration of 6.3 g/L in the drinking water caused a marked increase in blood acidosis for 42 to 52 day-old broilers (Branton *et al.,* 1986). Teeter and Smith (1986) obtained maximum weight gain in broilers at 35°C with 2 g/kg NH_4Cl in drinking water. Up to 3 g/kg dietary NH_4Cl may improve the growth rate of heat stressed broilers though it was not clear if this beneficial effect was either via electrolyte balance/blood pH or simply via the indirect effect of stimulating water intake (Leeson and Summers, 2001). Dietary $NaHCO_3$ added at 3.2 g/L had no effect on blood pH, whereas a short-term infusion of 2 g/L into the crop did induce alkalosis (Bottje and Harrison, 1985). It was observed by Owen *et al.* (1994) that dietary $NaHCO_3$ incorporated into the diet at the level of 1% caused a reduction in ascites incidence, ascribed as being due to its alkaline nature and the possible induction of alkalosis. In the same work, the addition of NH_4Cl at 1% level caused numerical increase in ascites incidence, and this was presumed to be due to its acidifying nature and the possible induction of acidosis. Sodium from $NaHCO_3$ at 1.45 g/L was harmful, both reducing body weight and increasing ascites mortality (Julian *et al.,* 1986), whereas a very high level of 7.50 g/L was found toxic and manifested syndrome characteristics mainly in 2-wk-old birds (Mirsalimi *et al.*, 1993). Under heat stress condition, Balnave and Gorman (1993) suggested $NaHCO_3$ at the level of 5.60 g/L in the water and did not find any reduction in body weight. The addition of Na has a specific ascites-inducing potential, that's why in this study $KHCO_3$was used rather $NaHCO_3$. Ahmad *et al.*, 2006 observed that non-chloride sodium salts ($NaHCO_3$, Na_2SO_4, and Na_2CO_3) showed better production performance and reduced mortality than the control diet containing sodium salt (NaCl). Sodium salts also enhanced water intake as well as water to feed intake ratio. This effect was more pronounced in broilers fed $NaHCO_3$ supplement (with NaCl in the basal diets). A lower mortality rate was noted with $NaHCO_3$ (15%), Na_2CO_3 (14%) and Na_2SO_4 (15%) supplements than with NaCl (33%) treatment. It is speculated that effect of Cl may be related to its supplemental salt. NH_4Cl added to provide total Cl at 0.15, 0.25, and 0.35% which increased water consumption from 102 to 105 to 114 mL/day (d 21-28), fecal moisture from 72 to 74 to 77% (14-28d) and weight gain of 735, 741, and 741 g, respectively,

in broiler chicks (Zisman, 1986). Moreover, adding chloride as NH_4Cl improved weight gain and decreased blood pH. While the effects of using combination of NH_4Cl and $NaHCO_3$ were synergistic in terms of increasing weight gain and slightly reducing alkalosis (Teeter and Smith, 1986). Bottje and Harrison (1985) suggested that treating heat stressed broilers with NH_4Cl could potentially be deleterious to the bicarbonate buffer system, as any metabolic acidosis associated with NH_4Cl catabolism might accentuate HCO_3^- loss due to increased respiratory rate. However, it needs care related to bicarbonate buffering system in immature birds like broilers rather than layers. In another study using non-conventional sodium salt sources; growth rates, feed efficiency and mortality were not affected by sodium when supplied in the form of sodium fluoride and sodium silicate; however, bone characteristics were improved by sodium fluoride but to some extent with sodium silicate (Merkley and Miller, 1983).

Dietary Electrolytes and Growth Performance

An improvement in production performance of broiler chickens was observed with variation in the levels of Na and Cl along with K in the feed keeping in view the variation in DEB. It is demonstrated that Na has a very important role in feed intake just after hatching, and also in secretion and activity of some digestive enzymes (Sklan and Noy, 2000). Na and K are involved in the process of absorption of some nutrients, such as glucose (Larbier and Leclercq, 1992). Cardiac output and blood pressure fall, hematocrit increases, elasticity of subcutaneous tissues decreases, and adrenal function is impaired in case of Na deficiency (Leeson and Summers, 2001). This deficiency would result in increased blood uric acid levels, which can result in shock and ultimately death.

When ratio between dietary cations and anions was kept around 1:1 containing 200 mEq/kg DEB, an improved growth pattern was seen in broilers (Hurwitz *et al.,* 1973). Johnson and Karunajeewa (1985) examined that DEB lower than 180 mEq/kg and higher than 300 mEq/kg decreased the weight of the chickens when assessed at day 42. Hulan *et al.* (1985) determined the effect of feeds containing electrolytes in different ratios and varying level of calcium, and found that the worst and the best weight gain were achieved when the DEB was 174 and 215 mEq/kg with 1.38 and 0.95% calcium, respectively. Leeson and Summers (2001) considered 250 mEq/kg DEB as an appropriate level for adequate poultry production potential, inspired from the work of Mongin (1981). According to Murakami *et al.* (2000), a vast range of DEB (150 and 350 mEq/kg) is recommended in commercial broiler diets for maximum bird performance. On the other hand, Na and Cl requirements in corn-soy diets were 0.28 and 0.25% (first 3 weeks) and 0.15% and 0.23% (last 3 weeks), respectively, for maximum performance of broiler chickens, and the best DEB was between 245 and 315 mEq/kg (first 3 wks.) and 249 and 261 mEq/kg from d 21 to 42 (Murakami *et al.,* 2001; Rondon *et al.,* 2001).

Borges *et al.* (2003a) evaluated that 240 mEq/kg DEB gave best weight gain and feed efficiency in Brazilian summer conditions and later on (Borges *et al.*, 2004) recommended 202-235 mEq/kg under normal ambient temperature, showing higher DEB requirements in heat stress than normal ambient conditions.

A positive quadratic effect of increasing Na levels (from 0.10 to 0.46%) in pre–starter phase was observed by Maiorka *et al.* (2004) on weight gain, feed to gain ratio, and feed and water consumption. He also suggested optimal Na level of 0.45% for water consumption, 0.40% for feed intake and weight gain and 0.38% for feed to gain ratio. Level of dietary Na needed for optimum feed conversion was supposed to be lower than needed for maximum body weight and water intake (Watkin *et al.,* 2005). Previously, Ross (1979) reported that the Na requirement of broiler chicks fed a wheat-soybean diet was greater than the 0.15% suggested by NRC (1971) when the water Na was 0.30%. Difference in requirements could be related with the production potential difference of strains of modern broilers. Watkins *et al.* (2005) suggested the estimated requirement of Na for body weight was 0.17% and suggested in between 0.14 and 0.19% Na^+ when the drinking water contained no added salt. A need for 0.20 to 0.25% Na for maximum performance during first 21 days period (Murakami *et al.,* 1997a, b), while, 0.25% Na and 0.30% Cl is recommended for greater feed intake for finishing broilers (from day 28 to 42; Rondon *et al.,* 2001). Oliveira *et al.* (2005) divided whole experiment into three phases viz. 8-21, 22-42 and 43-53d and recommended 0.63, 0.71 and 0.80% K, respectively, for weight gain and a bit higher for feed intake. Previously, lower levels viz. 0.50, 0.47 and 0.45% K were suggested by Rostagno *et al.* (2000) for 1-21, 22-42 and 43-49d, respectively.

Dietary Electrolytes, Heat Stress and Growth Performance

In the recent past, broiler production performance was evaluated in terms of varying electrolyte level and DEB with respect to environment. Ambient temperature is considered as the most important reason behind shuffling the level of electrolyte or DEB, besides considering the well-being of the bird (acid–base balance and internal homeostasis). Sodium is slightly more potent through the water than an equivalent intake through the feed (Ross, 1979), but nutritionists prefer the certainty of inclusion in feed rather than depending on sometimes mistaken dosing by flock owners through the water. $NaHCO_3$ has a solubility of about 11% in water at 37.8^{o}C (100 F).

A reduction in body weight (23%) and feed intake (15%) of broilers reared under high temperature (min, 27°C and max. 38°C) was reported by Yalcin *et al.* (1997) when compared with counterparts reared under normal temperatures. A significant reduction in feed consumption (7-14%) was observed in birds kept in 26.7°C compared with those kept at 21.1°C during finisher phase (4-6 week; Suk and Washburn, 1995). Reduction in feed intake

was 3.6% per degree increase between 22^0 and 32°C, which could be related to the age and genotype of the birds (Zuprizal *et al.,* 1993). Henken *et al.* (1983) observed that feed intake and growth rate were significantly reduced by 15.9 and 12.3% at 35°C, respectively, and 14.9 and 12.5% at cyclic temperatures of 30-40°C, respectively, compared to birds kept at 25°C. Cahaner and Leenstra (1992), Washburn *et al.* (1992) and Yunis and Cahaner (1999) were of the view that the effect of heat stress was more pronounced in fast growing broilers than those in slow-growing broilers. Improved blood parametre, litter condition and reduced abdominal fat were seen by Mushtaq *et al.* (2007) suggested best diets with 0.20-0.25% Na and 0.30% Cl at constant DEB (250 mEq/kg). Earlier, Mushtaq *et al.* (2005) recommended 0.25% Na and 0.30% Cl for better performance under high cyclic temperature. Recently, Ahmad *et al.* (2009) suggested 150-250 mEq/kg DEB for well-being of heat distressed broilers.

Dietary Electrolytes, Salts and Growth Performance

The requirement of dietary electrolytes (Na^+, K^+ and Cl^-) decreases with increasing age (NRC, 1994) however, the requirements of these electrolytes and their respective dietary salts tend to be different for different production and physiological parameters under different climatic conditions particularly ambient temperature and relative humidity. Addition to this, nature and purity of salt is also equally important in finding out the efficacy of a particular electrolyte.

Based on much better broiler performance during heat stress with $NaHCO_3$, a CO_2 generating buffer, than with Na_2CO_3 at the same dietary sodium and DEB levels, it has been suggested by Gorman and Balnave (1994) that heat stress may induce a requirement for the bicarbonate ion (HCO_3^-) due to condition of metabolic acidosis. Nutritionists can mediate during hot summer to provide $NaHCO_3$ and/or KCl through the feed or water, so that the detrimental effects of heat stress can be minimized in many cases. Preventive measures can be taken by formulating feed with high enough DEB using $NaHCO_3$ to prevent excessive mortality of heavy-weight broilers and to stimulate growth during the hot summer season. Various researchers (Mongin, 1981; Hooge, 2003; Ahmad *et al.*, 2005) recommended $NaHCO_3$ as salt of choice in hot summer conditions as because of having alkaline nature. Benton *et al.* (1998) stated that $NaHCO_3$ is incorporated into broiler diets at up to 0.5% and is recognized as saving 2 d growth to market weight during summer in Australia. They indicated that heat-stressed broilers respond better to supplements of $NaHCO_3$ than $KHCO_3$, Na_2CO_3, or K_2CO_3. Nagwa and Maghraby (1995) were of the view that broiler chicks (Arbor Acres) at environmental temperature 32.5°C kept on diets containing $NaHCO_3$ or NaCl to provide Na^+ 0.2%, 0.3% or 0.45% showed increased feed intake and improved feed conversion efficiency as compared to $NaHCO_3$ at all levels kept in pen system. Moreover, higher NaCl levels improved feed intake compared with lower doses. Bláha*et al.* (2000) reported that broiler

diets (Hybrid Cobb) containing 0.5% NaCl with 35 mg vitamin C showed better feed conversion than the diet without added NaCl or vitamin C. Feed efficiency was improved with a dietary Na^+/Cl^- ratio of unity in NaCl based diets while in a electrolyte mixture containing 56.3% NaCl and 43.7% $NaHCO_3$ based diet while the best ratio was 1.26 in 8 week old broilers (Hurwitz*et al.,* 1974). Similarly feed intake was found unaffected either by NH_4Cl (0.63%) or $NaHCO_3$ (0.32%) supplementation in drinking water on 8 hour severe heat exposure of 42-52 day-old broilers (Branton *et al.,* 1986).

When comparing different salts of sodium with potassium salts, it was reported that dietary supplements of $NaHCO_3$ gave significantly better feed conversion than supplements of $KHCO_3$ on equimolar bicarbonate concentration at 31ºC (Hayat *et al.,* 1999). Earlier, $KHCO_3$ was also observed to decrease feed efficiency (Teeter and Smith, 1986). Smith and Teeter (1987b) studied the impact of KCl supplementation on feed intake and feed efficiency of broilers exposed to chronic heat and cycling temperature stress. A dietary level of 1.5-2.0% K^+ in the form of KCl was reported to improve feed efficiency during chronic heat stress. KCl supplementation increased feed intake and weight gain of birds offered cold water, however, warm drinking water KCl supplementation had no beneficial effect on feed intake or weight gain when observing effect of drinking water temperature and KCl on production performance of heat-stressed broilers (Beker and Teeter, 1994). Deyhim and Teeter (1994) reported no effect of isomolar KCl or NaCl (0.067 mol/litre) supplementation in drinking water on birds kept under thermo-neutral or cyclic heat stress. Balnave and Gorman (1993) reported improvement in feed intake at high temperatures by diet or drinking water fortified with $NaHCO_3$. It is observed that about one-fourth of the broiler companies in America use either $NaHCO_3$ or $NaHCO_3 \bullet Na_2CO_3 \bullet 2H_2O$ (sodium sesquicarbonate) in the summer months (Donohue, 2001).

Dietary Electrolytes, Water Consumption and Litter Quality

Water consumption increases linearly with the increasing electrolytes in the diet (Mongin, 1980; Borges *et al.,* 1999). Mongin (1980) suggested a vital relationship between excessive sodium intake and litter dampness. Potassium can also be considered while role of chloride is considered inconsequential in this regard (Vogt, 1971), however, Borges *et al.* (1999) suggested that diets with high levels of Cl^- and/or K did not stimulate water intake during the first seven days of age.

It has been shown that the litter wetness and crustiness is the most prevalent form of footpad dermatitis, a form of dermatitis, in chicken and turkeys that is caused by a combination of moisture and chemical irritants present in the litter (Martland, 1984; 1985; Greene *et al.,* 1985; Ekstrand *et al.,* 1998). This condition is sometimes termed as 'contact dermatitis' and may also affect breasts (Greene *et al.,* 1985) and hocks (Martrenchar *et al.,* 2002). This

footpad dermatitis type seems to be related, mainly, to litter moisture and excreta stick to the skin (Abbott*et al.,* 1969; Jensen *et al.,* 1970; Harms *et al.,* 1977). Jensen *et al.* (1970) reported that increased incidence of foot dermatitis was seen when turkey pouts were fed diets with increased amounts of soybean meal. Borges *et al.* (1999) showed that diets containing 0.30 and 0.45% Na caused litter moisture similar to those obtained with the use of commercial diets at this age. Eichner *et al.* (2007) concluded that increased litter moisture and incidence and severity of footpad dermatitis was attributable to the diet formulation which based on all vegetable sources exclusively on corn and soybean meal when compared with diets containing poultry by-product meal or corn gluten meal in the starter and grower diets, respectively. This severity in high soybean diets is sometimes linked with high potassium contents. He further suggested that litter moisture and the occurrence and severity of footpad dermatitis increased as broilers aged to 40 d.

Dietary formulation and ingredient changes, particularly related to electrolytes, leading to higher water intake by birds are expected to increase excreta and litter moisture. Diets with increased Na and K result in an increased water intake and litter moisture, whereas an increase in Cl^- does not have the same effect (Murakami *et al.,* 2000; 2001; Rondon *et al.,* 2001). Hurwitz *et al.* (1973) indicated the moisture of the droppings have a positive reliance with total dietary Naand the Na^+ to Cl^- ratio. A negative correlation (r = -0.72) was found between water consumption and mortality by Branton *et al.* (1986).

The consequence of a higher water intake promoted by diets with high electrolytic balance leads to higher litter humidity and leg health issues. However, Maiorka *et al.* (2004) did not find any effect of the electrolytic balance (100, 150, 200, 250 e 300 mEq) on the humidity of the excreta up to 7-days of age when DEB was changed with dietary Na. No significant effects of DEB on litter moisture content was found by many workers when it is supplied through drinking water (Karunajeewa *et al.,* 1986; Borges *et al.,* 1999a; Salvador*et al.,* 1999); while many others (Champaign, 1994; Balnave and Gorman, 1993; Smith and Teeter, 1989; Nagwa and Maghraby, 1995) reported that it's all because of DEB. Castelló and Pontes (1992) reported lower water intake and higher moisture contents with low DEB diets i.e., 170-174 mEq/Kg than with 336-340 mEq/Kg. it is speculated that this increased water consumption in response to increased lead to more retention of water in the carcass leading to increase in dressing percentage. Karunajeewa and Barr (1988) observed the higher litter moisture content on the diets with an electrolyte balance of 205 mEq/kg than on the diets with either 125 or 165 mEq/kg.

Dietary Electrolytes, Heat Stress, Water Consumption and Litter Quality

A strong relationship exists between dietary electrolytes and water consumption especially in hot environment. Increased water intake could be used to reduce increased internal body

temperature especially under heat stress conditions. It is established by various researchers that increasing dietary Nahas a linear effect on water consumption (Murakami *et al.,* 2000; 2001; Rondon *et al.,* 2001) in heat distressed broilers (Mushtaq *et al.,* 2005). However, these reported results were specific to the initial days of birds. Moreover, Mushtaq *et al.* (2005) described that birds with high initial weights excreted less moisture through droppings. This demonstrated the ability of heavier birds to retain more water in the body, which led to improved litter condition (Mushtaq *et al.*, 2007). Vena *et al.,* (1990) reported that besides increased water consumption, Na^+ increases faeces moisture and Na^+ urinary excretion.

It is speculated that litter moisture is greatly linked with the adequate proportion between Na^+ and Cl^- (Freeman, 1983). A relationship between Na and Cl was established in heat-distressed broilers recently by Mushtaq *et al.* (2007) who observed that water consumption was significantly greater on 0.20% Na^+ when compared with higher levels of Na^+ (0.25 or 0.30%) without the effect of dietary Cl^- in high ambient temperature at a constant DEB level i.e. 250 mEq/kg. Maximum water consumption was noted with dietary Na × Cl of 0.20 × 0.30 and minimum water consumption with dietary Na × Cl of 0.25 × 0.50. It means that Cl intake is not linked with water consumption. Deyhim and Teeter (1994) reported that drinking water supplemented with either isomolar (0.067 mol/L) NaCl or KCl substantiated increased water consumption in heat distressed broilers. Branton *et al.* (1986) reported no difference in water intake with NH_4Cl (0.63%) and $NaHCO_3$ (0.32%) through drinking water supplementation during periods of heat stress. While water intake was increased by about 20% in birds given water containing $NaHCO_3$ (0.63%) was severely limited by NH_4Cl (0.31%). It was still unclear that beneficial effects of any salt (NH_4Cl) are either because of maintaining electrolyte balance and blood pH or by increasing water intake (Leeson and Summers, 2001; Ahmad *et al.,* 2009).

It is anticipated that with the increasing DEB, water consumption increases linearly that ultimately leads towards litter problems. Lowest water consumption was seen at lowest DEB i.e. 0 mEq/kg, while highest at highest level of DEB i.e. 350 mEq/kg in hot summer conditions (Ahmad *et al.,* 2009). This problem is more pronounced in last days of life. Borges *et al.* (2004) also observed higher water intake at highest level of DEB (240 mEq/kg) when compared with 0 mEq/kg.

Dietary Electrolytes, Salts, Water Consumption and Litter Quality

It was postulated that using different salts in the feed/ water determines the amount of water intake by poultry (Mongin, 1980). It has generally been accepted that addition of various salts to the diet and/or drinking water alters the bird's osmotic balance, increases water consumption and excretion to maintain water balance in the body (Borges *et al.,* 2004a). Various salts like KCl, $NaHCO_3$and NaCl significantly increase water intake in growing and

finishing stages (Borges, 1997). However, $NaHCO_3$ is considered as good source of Na^+ and HCO_3^- ions in heat stress condition (Hooge, 2003). The significance of electrolytes and their respective salts is vital when environmental norms, temperature and relative humidity, were considered for water consumption and dampness of bedding material. Various attributes are responsible for litter quality like housing and management (type of litter, temperature, humidity, ventilation, density, heating system, drinking system) and other disease factors (Coccidiosis, *E-Coli*, *Campylobacter*; Francesch and Brufau, 2004). As DEB increases (from 50 to 340 mEq/kg), this amplifies the litter moisture contents and bacterial contamination accordingly; and reduces the quality of litter in mild (Borges *et al.,* 2004a) and high (Ahmad *et al.*, 2009) temperature conditions. Moisture contents of litter may be altered by excreta composition, which can affect water retention and limit the evaporative water losses (Francesch and Brufau, 2004). The watery excreta is directly proportional with the electrolytes present in the diet that are the regulatory factors of body fluids. Borges *et al.* (2003a) described highest litter moisture (21%) during first week of age with 360 mEq/kg DEB ($NaHCO_3$ +$KHCO_3$, High Na + K diet); while go up to 55% litter moisture during the 6^{th} week of age. Similar findings of highest litter moisture were reconfirmed later on by Borges *et al.* (2004a) in the diet containing 340 mEq/kg DEB.

Dietary Electrolytes and Acid-Base Balance

The acid-base balance (ABB), blood pH and growth rate in broilers as influenced by the cation/anion ratio was earlier reviewed by Hurwitz *et al.* (1973). He demonstrated that ABB is influenced by a range of internal and external factors; among these, diet composition and environmental conditions are more prominent. These conditions affect regulation of pH in blood and tissues. Acids are known to be the final metabolites removed from the body by either kidneys or lungs; however, if these metabolites are kept building up in the body, they can alter the ABB from its normal status (Ruiz–Lopez and Austic, 1993). It is determined that concentration and chemical properties of the acids and bases dissolved in blood plasma are major determinants of its pH. Three major classes of acids and bases can be identified: 1) carbonic acid; 2) metabolizable acids (acetic and lactic); and 3) non-metabolizable inorganic acids and bases.

It is established earlier that the balance among strong electrolytes (Na^+, K^+ and Cl^-) mainly determines the ABB in body fluids (Stewart, 1978). Blood cations (Na^+ and K^+) are complemented inversely through shuffling of various anions (Cl^- and HCO_3^-) concentrations, so as Cl^- goes up, HCO_3^- goes down and vice versa. Supplementing the diet with extra chloride can increase blood Cl^- (acidosis) or, on the other hand, adding HCO_3^- to the diet can decrease blood Cl^- (alkalosis) within certain physiological limits. Addition of HCO_3^- to the

diet may lower the level of "effective Cl^{-}" of the diet by eliminating some Cl^{-} from the bird through the kidneys. Benton *et al.* (1998) stated that an increase in plasma Cl^{-} decreases H^{+} excretion and HCO_3^{-} reabsorption by the kidneys whereas an increase in plasma Na^{+} increases H^{+} excretion and HCO_3^{-} reabsorption and both situations affect blood pH differently. An increase in plasma K^{+} decreases HCO_3^{-} reabsorption from the kidneys. So, electrolyte relationships are identified in plasma as: 1) K^{+} content follows H^{+} (K^{+} inversely related to pH), 2) HCO_3^{-} content follows Na^{+} content, and 3) HCO_3^{-} content follows inversely Cl^{-}content. Berne and Levy (1993) described every 0.1 unit decrease in pH (H^{+} increase) causes plasma K^{+} to increase from 0.2 to 1.7 mEq/L.

Higher levels of dietary chloride are known to depress blood pH and blood bicarbonate concentration (Ruiz-Lopez and Austic, 1993) and to increase the occurrence of tibial dyschondroplasia (**TD**) and cartilage abnormalities (Nelson *et al.,* 1981) possibly attributable to competitive assimilation with Ca^{2+} in small intestine (Melliere and Forbes, 1966). Birds suffering from alkalosis cause blood concentration of the electrolytes (Na^{+}, K^{+}) to decrease. A decrease in Na:Cl ratio leads towards reduction in alkalosis with an 8% improvement in bird performance when 0.5 and 1.0% $CaCl_2$ was added (Teeter *et al.,* 1985). Also, respiratory alkalosis causes a reduced contest between H^{+} and K^{+} for urinary excretion thus increasing K^{+} loss in the urine. Excess K^{+} ions compete with buffer anions in the renal tubule fluid, preventing H^{+} removal, which in turns reabsorbed, and the result may be acidosis. H^{+} excretion and HCO_3^{-} reabsorption by the kidneys is reduced by increasing Cl^{-} ions. This might contribute to blood acidification which seems to be an appropriate response to alkalosis *per se*.

It is anticipated that bird performance is directly related with blood pH which indicates birds' normal physiology and biochemical status. Broiler growth was found greatest when blood pH was 7.28, with a decrease in growth being found when pH values were greater than 7.30 or lower than 7.20. Teeter *et al.,* (1985) have shown that pH values greater than 7.25 had depressed growth rate and feed efficiency during panting conditions. Actually this narrow range of pH determines the activity of enzymes which is depicted in terms of good health status and production performance (Lehninger, 1970). Rising blood pH can be adjusted by a reduction in the respiratory rate of birds; however this increase can occur with acclimatization temperatures, and can be improved by maintaining more water consumption (Belay and Teeter, 1993). However, electrolyte loss without any change in body water content reduces the osmolality of these fluids.

With the increasing DEB the blood pH augments linearly as at 0 DEB the blood pH is acidic while at 350 DEB it was resulted in alkalosis in hot summer conditions (Ahmad *et al.,* 2009). DEB is considered crucial in controlling blood pH and ABB which in turns control enzymatic

efficacy and ultimately bird performance (Patience, 1990; Butcher and Miles, 1994; Hooge, 1998). DEB for maximum growth varied from 226 to 260 mEq/kg; however if the response is totally due to changes in pH or to other electrolyte or metabolic effects is still to be defined. Changes in the ABB and imbalances in DEB cause loss of appetite with reduction in production performance; and ultimately lead to increased mortality rates (Mongin, 1981). As K^+ is involved in many metabolic and regulatory processes like ABB, arginine–lysine antagonism, nervous conduction, muscle contraction, synthesis of tissue proteins, safeguarding intracellular homeostasis, enzymatic reactions, and osmotic balance, so a consequence of involvement in many procedures, changes in K^+ homeostasis can affect cell function and in turn production efficiency of the birds. K^+ excretionis influenced by ABB, DEB and hormonal factors (aldosterone, anti–diuretic hormone and deoxycorticosterone). K^+ excretion rate by urine is variable, being connected to serum Na^+ concentrations and the bird's hydration status where losses can be caused by an increase in water consumption, since the osmotic gradient favours the movement of intracellular fluid water to the urine, which may carry K^+. The increase in K^+ intake results in greater urinary loss, as the bird having little capacity of conserving body K^+.

Dietary Electrolytes, Heat Stress and Acid-Base Balance

The bicarbonate system is the most vital buffer system for maintaining normal blood pH being under the dual regulatory control of the lungs and kidneys. Blood pH, as determined by the bicarbonate buffering system, is represented by the Henderson-Hassel Balch equation, i.e., $pH = 6.1 + \log [HCO_3^-] / 0.03\ pCO_2$; where $pC0_2$ is the partial pressure of carbon dioxide (Berne and Levy, 1993). Normally, the ratio of HCO_3^- and $0.03\ pCO_2$ is 20:1, resulting in a blood pH of approximately 7.4. Being a privileged source of bicarbonate, $NaHCO_3$ could affect blood pH by providing Na^+ and HCO_3^- ions as well (Mongin, 1968). The concentration of bicarbonate is mainly under the control of the kidneys and, to a much lesser extent, the lungs. The kidneys control the concentration of bicarbonate by modifying its reabsorption from the renal tubules to maintain ABB. Increased ventilation decreases $pC0_2$ during heat stress, which increases the log term in the Henderson-Hassel Balch equation, thereby increasing the pH and potentially inducing respiratory alkalosis. Potassium, sodium, and chloride levels in the plasma are all affected by respiratory alkalosis. Of the three ions, changes in K^+ are the best understood. In some studies with poultry, plasma K^+ has been shown to increase during heat stress (Kohne and Jones, 1975) but most studies have found K^+ to decrease during heat stress (Deetz and Ringrose, 1976; Harper *et al.,* 1977). Huston (1978) found plasma K^+ to decrease within 6 h when birds were transferred from 8 to 30°C but observed no change in plasma K^+ levels when the reverse transfer was performed. The decrease in K^+ has been attributed to increased excretion (Ait-Boulahsen *et al.,* 1989; Berne

and Levy, 1993) and an increase in K^+ uptake by cells. The former appears to predominate in chronic heat stress and the latter during acute heat stress. Berne and Levy (1993) demonstrated that plasma K^+ increases 0.2 to 1.7 mEq/L for every 0.1 unit decrease in pH. Mushtaq *et al.* (2007) showed reduced serum K level by increasing dietary Na in heat-distressed broilers. Several studies in hot summer conditions have observed an increase in plasma Na^+ accompanied by hemodilution (Ait-Boulahsen *et al.,* 1989; Parker and Boone, 1971; Whithow *et al.,* 1994). This increase has been imputed to peripheral vasodilation and increased blood flow to vascular beds. In the above mentioned studies it was not possible to distinguish between the effect of respiratory alkalosis and effects caused by hyperthermia, but it is worth mentioning that an increase in plasma Na^+ increases H^+ excretion and HCO_3^- reabsorption, which might lead to the conclusion that increase in plasma Na^+ is merely the result of hemodilution due to increased water intake and peripheral vasodilation during hyperthermia and does not represent a physiological response to alkalosis.

Plasma Cl^- has been found to increase during heat stress in poultry (Ait-Boulahsen *et al.,* 1989; Elkinton *et al.,* 1955; Giebish *et al.,* 1955). This increase has been ascribed to a mechanism known as 'chloride shift' across erythrocyte membranes as blood CO_2 levels decrease. An increase in plasma Cl^- appears to decrease H^+ excretion and reduce HCO_3^- reabsorption by the kidneys. This would contribute towards lowering of blood pH and, thus, be an appropriate response to alkalosis.

Hurwitz *et al.* (1973) found the growth rate of the chick to be maximal when plasma pH was 7.28 and observed a decline in growth rate when pH values were above 7.30 or below 7.20. Teeter *et al.* (1985) indicated that blood pH values in excess of 7.25 decreased growth rate and feeding efficiency during periods of panting. Increased blood pH can be reduced by lowering the respiration rate of the bird. This increase occurs with temperature acclimation but during acute heat stress the respiration rate can be reduced and the efficiency of respiratory heat loss can be improved by maintaining a high intake of drinking water (Beley and Teeter, 1993). Branton *et al.* (1986) found mortality due to heat stress and is inversely related to water consumption.

Dietary Electrolytes, Salts and Acid-Base Balance

There is always a tendency to link acid-base balance with electrolytes and environment especially temperature. Earlier, Mongin (1968) suggested that $NaHCO_3$ is quite good during heat stress as it provides elements, in the form of Na^+ and HCO_3^-, for maintaining pH. Teeter *et al.* (1985) observed that broilers given 0.5% $NaHCO_3$ in feed had the same blood pH (7.4) as control birds (without $NaHCO_3$) in summer conditions, indicating $NaHCO_3$ did not cause further alkalosis in heat stress. However, during panting under acute heat stress, similar blood pH in diets with and without $NaHCO_3$ indicated that the alkalosis was not exacerbated and the

birds perform better. The addition of 0.5 and 1.0% $NaHCO_3$ to the feed of broilers subjected to temperatures (varying from 39 to 41°C and 34 to 36°C) improved feed intake, weight gain and feed: gain (Fischer da silva *et al.,* 1994). Gorman and Balnave (1994) indicated that heat-distressed broilers responded better to supplements of $NaHCO_3$ than $KHCO_3$, Na_2CO_3 or K_2CO_3. Then, Smith and Teeter (1993) proposed KCl as privileged compound in the feed and/ or drinking water as a way to minimize the consequences of high temperatures on production performance of broilers. Later on, supplementing KCl, NaCl and $NaHCO_3$ in the feed of broilers resulted in improved weight gain (Borges, 1997). Finally, Hooge (2003b) stated that it was better to 'prevent' heat stress with $NaHCO_3$ and higher DEB. He also found that KCl could also be used in this scenario as supplement to put back body K^+ lost by altered tissue cell permeability and to supply Cl^- for alkalosis. However, depending upon the situation both these compounds viz. $NaHCO_3$ and KCl could be used as remedy under heat stress conditions. Teeter and Belay (1996) observed that addition of NH_4Cl to bring down blood pH to normal values had no effect on the performance of birds.

These observations led to the conclusion that different electrolyte sources led to positive impact on the performance of heat-distressed broilers. However, variations among different studies also indicated that there was a need to define the proper electrolyte source, its amount and combination of different sources keeping in view the concept of an appropriate DEB for optimum broiler production performance under heat stress conditions.

Dietary Electrolytes, Protein Source and Acid-Base Balance

It is anticipated that the connection between the metabolism of minerals (especially electrolytes) and of amino acids can often be used to adjust ABB. The protein source used in the feed can affect the electrolyte and ABB, because certain sources, particularly from animal by–products, increase the production of organic acids and reduce Na^+ and K^+ contribution, increasing the relative amount of Cl^- (Portsmouth, 1984). Broiler feeds containing soybean meal, which has low Na contents and high K contents, showed a significant response in the development of broilers supplemented with 0.5 and 1.0% NaCl (March, 1984). It means these vegetable protein sources are also important in terms of their mineral contents and metabolic products.

Excretion of acid or conservation of bicarbonate ions in the body is a critical function of the kidney in terms of acid-base balance (Brosnan *et al.,* 1987). Walser (1986) discussed that ammonium ion (NH_4^+) excretion only reflects increased acid removal if it is accompanied by Cl^- ions or occur in exchange for Na^+ ions only when glutamate is oxidized. The relationship of renal, hepatic and skeletal metabolism of amino acids clearly indicates the role of acid-base balance can play in the utilization of amino acids. Besides utilization of all other amino acids, glutamine remains a primary ammoniagenic precursor (Lowry *et al.,* 1987). Oxidation of

protein generally is considered a net contributor of acid, although this all depends on its amino acid profile. Oxidation of neutral amino acids has no effect on acid-base balance, whereas oxidation of dicarboxylic amino acids tends to cause metabolic alkalosis (Walser, 1986). If amino acid is phosphorylated, it will cause metabolic acidosis (Brosnan and Brosnan, 1982; Halperin *et al.,* 1986). Mongin (1981) initiated this issue that how protein source is directly related with endogenous acid production; composition of the nitrogen products, organic phosphorus and other mineral composition of the protein sources. DEB is supposed to be varies according to the changing dietary CP. The CP × DEB interaction was reported to be changing but non-significantly by Adekunmisi and Robbins (1987). Austic (1980), and Smith and Teeter (1987a) also discussed influence of ABB, electrolytes and their salts on birds' performance in low protein diets. Martinez-Amezcua *et al.* (1998) observed improved performance by increasing dietary lysine, without any effect of DEB level.

Increasing the DEB from 200 mEq/kg to 350 mEq/kg improved weight gain fed high CP diets (28.6%) but depressed gain and feed consumption of chicks fed low CP diets (14.3%). A new approach in this scenario could be observed when chicken growth fed low-CP diets reduces as DEB is altered by Na and K additions. A consistent interaction between K levels (0.90, 1.00 and 1.10%) and methionine sources was found for feed intake while deteriorated FCR was seen in highest level of K (Ribeiro *et al.,* 2008). Earlier, Borges *et al.* (2002) suggested that CP levels (21 and 23.5%) did not influence broiler performance. A positive response for 2-hydroxy-4 methylthio butanoic acid when compared with DL-Methionine was found at all levels of dietary Na (0.15, 0.20 and 0.25%) for weight gain and feed conversion ratio at 48 d of age (Ribeiro *et al.*, 2008).

Dietary Electrolytes and Blood Parametres

The blood system is particularly sensitive to changes in acid–base balance or other physiological chaos, ultimately lead to an important indicator of performance responses in chickens. Quantitative and morphological changes in blood cells are coupled with a number of factors such as haematocrit (**Hk**) value, leukocytes (**WBC**), erythrocytes (**RBC**) and haemoglobin (**Hb**) contents. Demand for Na^+ and K^+ contents dropped off (Borges, 1997) while Cl^- concentration increased (Belay and Teeter, 1993) as temperature rose and resulted in low blood electrolyte balance. There is about a 2.75:1 ratio of HCO_3^- concentration in plasma compared to intracellular fluid. Phosphate, HCO_3^- and Cl^- in conjunction with proteins including Hb, are most closely associated with the pH of the bloodstream, which is normally maintained within a range of about **7.35-7.45** (Brody, 1999). Ruiz-Lopez and Austic (1993) reported the depression in blood pH and blood HCO_3^- due to high dietary Cl^-. Previously, Walicka *et al.* (1979) also observed lower values of pH and HCO_3^- in NaCl deficient diets. Increasing dietary Cl^- from 0.30 to 0.40% decreased serum Na^+ and K^+ while

serum K^+ was found to positively correlated (r = 0.30) with increased water consumption (Ruiz-Lopez and Austic, 1993).

Dietary Electrolytes, Heat Stress and Blood Parametres

Heat stress is supposed to augment Hk, Hb, heterophil and heterophil:lymphocyte while reduce erythrocytes, lymphocytes and serum Na^+ and K^+. Supplementing water with KCl has been reported to regulate the level of erythrocytes and haemoglobin of broiler chicks stressed out by heat (Borges *et al.,* 1999a). Increased heterophil/lymphocyte ratio and decreased serum levels of K^+, Na^+ and Cl^- were observed by Salvador *et al.* (1999) as a consequence of high ambient temperature while bicarbonate in the feed and drinking water did not affect the heterophil/lymphocyte ratio, serum levels of K^+, Na^+, and Cl^-. Aengwanich (2007) was investigating the influence of high environment temperature on electrolyte status of Thai Indigenous, Thai Indigenous Crossbred and Broiler Chickens and found that time, breeding and environmental temperature had an influence on the electrolyte status of three strains and also suggested that Thai Indigenous Chicken preserved the electrolyte in their body better than Thai Indigenous Crossbred and broiler chickens, respectively.

Reduced blood Hk (Vo *et al.,* 1978; Zhou *et al.,* 1999) and Hb concentrations (Vecerck *et al.,* 2002; Bedanova *et al.,* 2003) while increased blood glucose (Borges, 1997; 2001) were observed in heat–distressed broilers in electrolyte studies. Mushtaq *et al.* (2007) observed a significant main and interaction effects of dietary Na and Cl on blood pH, serum Na^+ and K^+ under subtropical summer conditions on d 42. He observed that the increasing dietary Na from 0.20 to 0.30% increased serum HCO_3^- in linear fashion. The serum Na^+ unexpectedly decreased when dietary Na increased from 0.20 to 0.25%, and he has associated this change with the incremental increases of 0.10% in dietary K. The effects of different levels of dietary Na or Cl were more pronounced on blood parameters, which supported the view that DEB might be used as a good norm to predict or adjust acid–base status of the blood (Murakami *et al.,* 2001). In initial days of life, dietary Na or Cl did not affect acid-base status and serum Na^+ and K^+ (Mushtaq *et al.*, 2005). Maximum blood pH (7.39) at 0.40% Cl^- was seen and interactive effect of dietary cation–anion (Na × Cl) was found highly significant on blood pH. Birds fed the diet containing 0.25% Na^+ and 0.30% Cl^- showed minimum blood pH (acidosis). The blood pH increased with increased serum HCO_3^-, and as a result, the birds made some physiological adjustments to keep the blood pH near optimal (7.35; Mushtaq *et al.*, 2007). The physiological responses included increase in faecal moisture and urinary excretion and declined kidney glomerular filtration rate (Vena *et al.,* 1990; Pesti *et al.,* 1999; Rondon *et al.,* 2001).

Researchers have generally reported a reduction in plasma levels of K^+ and Na^+ (Deyhim *et al.,* 1990; Belay and Teeter, 1993; Ait-Boulahsen *et al.,* 1995; Borges, 1997) due to heat

stress, probably as a result of hemodilution following increased water consumption; however they assume that difference in the period of heat stress at the time of sampling may indicate no change in plasma K^+ and Na^+ levels. Kolb (1984) observed that blood glucose concentration is directly responsive to an increase in glucocorticoids for the period of stress. Higher concentration of dietary Na should be balanced with increasing Cl and reducing K contents so as to maintain plasma H^+. So, dietary manipulations will result in the adjustment of ABB. This was earlier postulated as cation-anion balance by Mongin and Sauveur (1974).

Dietary Electrolytes, Salts and Blood Parametres

It is contemplated that salts of various sources might have different effect on bird physiology and blood characteristics, depending upon whether they are included in the feed or water. Supplementing water with KCl has been reported to regulate the level of erythrocytes and haemoglobin of heat-stressed broiler chicks (Borges *et al.,* 1999a). He further observed that supplementation with 0.3 or 1.0% NH_4Cl in diets significantly decreased blood pH in heat-distressed broilers. Ahmad *et al.,* (2008) supplemented 0.6% KCl in drinking water and observed reduced panting-phase blood pH up to 7.31 and suggested enhanced physiological adaptation with this level of KCl supplementation due to more favourable pH under severe heat stress conditions (35 to 38°C). Supplementation of KCl (1.5%) and $NaHCO_3$ increased serum K^+ and HCO_3^- (Naseem *et al.*, 2005). Ahmad *et al.* (2005) compared different sources of electrolytes and noted that the adding NH_4Cl decreased blood pH while high pH values (7.33 and 7.34) with better performance were noticed in birds fed $NaHCO_3$ and $KHCO_3$ supplements in heat-distressed broilers. Increased HCO_3^- ion requirement was suggested during panting by Gorman and Balnave (1994). Source of salt is very important in order to determine the acidic properties of a particular anion like Cl^- and sulphates (SO_4^-). Acidic property of K_2SO_4 and Na_2SO_4 are 58 and 84%, respectively, mainly owing to the difference in source. This acidic property of source may not be apparent in heat stress condition as already high pH is compensated with anions (SO_4^-) in heat-distressed birds (Ruiz-Lopez and Austic, 1993; Ahmad *et al.*, 2006).

Dietary Electrolytes and Carcass Characteristics

Carcass responses are generally related with the protein and digestible amino acids concentration of the diet. The most limiting amino acids in poultry (methionine, lysine, and threonine) have become increasingly available for dietary supplementation at economical prices due to technological advancements. This has given poultry nutritionists the opportunity to formulate low crude protein diets which more closely meet the birds' amino acid needs while minimizing amino acid excesses. Al-Batshan and Hussein (1999) evaluated the effect of ambient temperature, energy and protein contents of the diet on carcass composition of

broilers and reported that hot cycling temperature decreased ($P<0.05$) breast meat, carcass weight, increased carcass yield, drum stick and thigh meat possibly due to less absorption of amino acids but did not affect abdominal fat. He suggested that increasing dietary energy did not alleviate the depressing effect of heat stress while increasing dietary protein up to 22% improved the performance of broilers irrespective of rearing temperature. He further added that energy contents of the diet should be increased or some other arrangements should be made to avoid the deleterious effects of heat stress on carcass composition. As crude protein is decreased, potassium is supposed to decrease that caused reduction in DEB which is known to adjust ABB. Carcass and dressing percentage increased while abdominal fat pad is generally not influenced on the high-energy diet. Increasing dietary amino acids linearly improved live weight and feed utilization and reduced abdominal fat pad weight (Waldroup *et al.,* 1990).

Along with all other production parametres, carcass characteristics were significantly affected by different electrolytic treatments and environmental (temperature and humidity) conditions. A significant effect of temperature treatment was detected for abdominal fat pad weight, where differences in fat pad weight were found among water treatments (5% KCl or 5% $NaHCO_3$) under normal ambient condition but not under cyclic environmental conditions (Whiting *et al.,* 1991a). Borges *et al.* (1999), and Johnson and Karunajeewa (1985) observed no effect on carcass yield and retail cuts in different DEB treatments. Karunajeewa *et al.* (1986) observed that higher concentration of inorganic phosphorus reduced dressing percentage and increased abdominal fat pad weight while similar effect of different DEB levels was observed on abdominal fat pad weights. Neither the dietary concentration of phosphorus nor the DEB was found not to effect ash content of tibia. Later on, Champaign (1994) reported no effect of electrolytes on carcass characteristics. Sharma and Gangwar (1987) reported that high temperature (32°C) decreased the concentration of Na^+ and K^+ in breast and thigh muscles of broilers from 4 to 8 weeks old. He further observed that breast muscles had shown lower Na^+ and higher K^+ concentrations than thigh muscles. Pourreza and Edriss (1992) reared straight-run broilers at 20 or 30°C and observed that high temperature decreased slaughter, carcass and abdominal fat weight and increased dressing percentage. Males raised under a normal temperature had higher starved, plucked and carcass weights than females reared under the same temperature. Somewhat comparable results were reported by Smith and Teeter (1987b) who subjected broilers to heat stress and observed that addition of KCl to the drinking water partially alleviated growth rate depression but did not affect carcass dry matter and fat by supplementing KCl in drinking water.

Dietary Electrolytes, Salts and Carcass Characteristics

Electrolytes contributed by different salts have variable effect on carcass characteristics of broilers. The effects of different levels of dietary Na or Cl^-, however, were found more pronounced on carcass parametres under heat stress condition (Mushtaq *et al.*, 2007; Murakami *et al.,* 2001). Whiting *et al.* (1991b) conducted two trials to investigate the effect of $NaHCO_3$ and KCl drinking water supplementation on post-mortem carcass and meat quality characteristics under thermoneutral and cyclic heat-stress climatic conditions. He reported that water chilled carcass weight changed water- and oven-cooked breast filled yields while similar responses in breast meat tenderness were found at the end of the experiment. Later on, the cyclic heat-stressed broiler carcass gained more weight than the thermoneutral (control) broiler carcass during ice-water chilling. Oven-cooked fillet yield was affected by environmental regimen; fillets from cyclic heat-stressed broilers had lower yields than thermoneutral (control) broiler fillets. Salts supplemented in drinking water showed a significant source of variation for breast meat tenderness. Broilers given 0.5% $NaHCO_3$ from 5 to 8 week of age had higher shear resistance values than broilers given a combination of 0.5% $NaHCO_3$ and 0.5% KCl or tap water-fed controls. Breast meat shear values were negatively correlated with 8 week broiler body weight and 5 to 8 week gain. On the other hand, Smith (1994) found similar effect of electrolytes on carcass characteristics when provided by KCl and NaCl in the drinking water to broilers grown at elevated temperatures. Similarly, no effect of supplemental salts of Na ($NaHCO_3$) and K (KCl) was observed in water on carcass yield and intestinal contents within 24 hours of processing (Gomes *et al.*, 2008).

Hooge *et al.* (1999) tested $NaHCO_3$ at levels of 0 to 0.4% inclusion in broiler chicken diets in pen trials conducted across several seasons and natural exposure to coccidia. Dietary $NaHCO_3$ levels of 0.2 to 0.4% yielded significant improvements in livability, carcass yield and breast yield, and occasionally, abdominal fat pad. Concentration of minerals (Na^+ and K^+) of the pectoral muscle and in the liver increased with increasing amounts of NaCl in the diet (Decus *et al.,* 1974) while lower fat contents of the thigh muscle were found in birds kept on water supplemented with NaCl, $NaHCO_3$ or vitamins when compared with birds reared on tap water. Bláha *et al.,* 2000 suggested that electrolyte caused a reduction in fat contents of broiler meat. Higher fat contents and lower cholesterol contents was observed in thigh muscles of 42 d Ross cockerels, supplemented in water with 20 mg ascorbic acid, 0.5% $NaHCO_3$ and 0.63% NH_4Cl and other receiving 35 mg ascorbic acid, and vitamin complex when compared with the un-supplemented group (Bláha and Kang, 1997).

Dietary Electrolytes and Coccidiostats

Wet litter problem is sometimes associated with the use of certain coccidiostats that would highlight the interaction of electrolyte and coccidiostats. Some coccidiostats compliment the

addition of electrolytes while others interfere with the efficacy of electrolytes. Non-ionophoric anticoccidials are not commonly used in broiler production (Leeson and Summers, 2001). However ionophoric anticoccidials influence transmembrane flow of monovalent ions viz. Na or K, and increase water intake in order to maintain osmotic balance. Monensin is considered as best ionophoric anticoccidials who works with the transmembrane flow of Na (Leeson and Summers, 2001)

Salinomycin has been comprehensively tested in combination with $NaHCO_3$ or sesquicarbonate and was found to be potentiated by them (Hooge, 2003). Monensin has also been evaluated in combination with these sodium supplements, with the same beneficial results as salinomycin. Limited testing of lasalocid with $NaHCO_3$ has shown enhanced effectiveness as well. $KHCO_3$ has been found, in combination with either monensin or salinomycin, to be only partially as effective as $NaHCO_3$ (Hooge *et al.,* 2000a). Ouart *et al.* (1995) tested different coccidiostats and found elevated levels of water consumption (particularly lasalocid) throughout the whole experimental period in different seasons. Leeson and Summers (1991) recommended that dietary Na level should be reduced to counter the effect of ionophores particularly lasalocid. To sustain production performance, chloride level should be reconsidered under the above prescribed conditions. Jensen (1982) reported that adding extra sodium (0.1%) from NaCl was found ineffective while finding ways to offset the growth depressing effects of high monensin.

Saylor and Fleet (1984) evaluated the effectiveness of various dietary levels of $NaHCO_3$ and $KHCO_3$ for reaching designated levels of K^+ with no coccidiostat, monensin, or lasalocid on 4-week body weights of broiler chicks without a coccidial challenge in battery brooders. Karunajeewa and Barr (1988) assessed the performance of male broiler chickens with different DEB (125, 165 and 205 mEq/kg) electrolyte balances, 2 sources of added K^+ (carbonate or sulphate) and 2 anticoccidial agents (monensin or lasalocid at 90 mg/kg) from 1 to 42 days. Similar effects were observed during starter phase while in the finisher phase, chickens fed the diets containing lasalocid utilized feed less efficiently than those given the diets containing monensin. Hooge *et al.* (1999) suggested that $NaHCO_3$ appear to complement ionophores and the optimal range of dietary $NaHCO_3$ for production improvements was 0.2% to 03% with an ionophore coccidiostat, either monensin or salinomycin, during all phases of growth and across all seasons when broilers were grown on built-up litter. On the other hand, $Na_2SO_4.10H_2O$ can potentially interact with ionophores (Soares, 1974).

It is anticipated that influence of Na sources along with various coccidiostats needs to be compared in different diets. Pesti (1998) observed no significant differences in male broiler performance on litter with seduramicin in corn-soy or corn-soy-meat diets, and showed no significant performance differences among three electrolyte balances (DEB 231, 275, or 362

mEq/kg), adjusted with a variety of electrolyte salts, including $NaHCO_3$ plus $KHCO_3$. Hurst *et al.* (1974) found that broiler chicks in battery brooders performed better with 0.30% dietary NaCl than with 0.075 to 0.225% NaCl, and performed equally well with 0.30% as with 0.375% NaCl, when monensin was included in the feed up to the age of 4 weeks. Nam *et al.* (1979) conducted litter and battery brooder trials with broiler chicks fed corn soy diets and found 0.15% sodium to be adequate with either monensin or lasalocid compared to 0.10, 0.20, or 0.35% Na. Gard *et al.* (1980) conducted a series of seven litter pen trials involving 218,200 birds (210 pens) with monensin at 100 or 121 ppm and dietary Na at 0.11, 0.17 to 0.19, and 0.24 to 0.27% using NaCl as the supplement. Broilers receiving 0.17-0.19% Na had heavier weights than those fed 0.11% or 0.24-0.27% Na. Increasing monensin from 100 to 121 mg/kg did not appear to increase the requirement for Na from NaCl. Edwards (1985) found significant improvements in 3-wk body weights and feed efficiencies of broiler chicks with or without monensin with increasing dietary NaCl levels from 0.11 to 0.22, 0.34, and 0.45%.

Saylor and Fleet (1984) recommended, without a coccidiostat, increasing Na from 0.15% to 0.30 and 0.45% was without effect at 1.0% K. With monensin, 0.15 and 0.45% Na treatments were equivalent at low or high K levels (0.5 or 1.0%) and 0.3% Na not tested. With lasalocid, lower K diets performed better, and increasing dietary Na to 0.30 and 0.45% improved growth over the 0.15% Na level with both dietary K levels. In another test, the best 4-wk body weights were reached with 0.42% Na, 0.55% K, and 0.27% Cl and either monensin or lasalocid. Fox *et al.* (1987) investigated 1.00% dietary $NaHCO_3$ for broiler chicks with or without coccidial inoculation in battery brooders and found that the supplement improved weight gain and feed efficiency. Weight gain was improved more in coccidiosis-infected chicks than in uninfected birds. No changes in the coccidiosis induced liver copper increase or the duodenal pH decrease were observed with high dietary $NaHCO_3$. Merrill (1993) found that 0.2% level of $NaHCO_3$ use in broiler feeds in Western Europe was the most common level, in conjunction with 0.1% added NaCl, and the main purpose was to lower dietary Cl and reduce wet litter problems. Diets typically contained not more than 25% soybean meal and included fish meals, the latter having about 0.60% each of Na and Cl. In diets without fish meal, it was typical to add up to 0.4% $NaHCO_3$.

A level of 0.25% dietary $NaHCO_3$ without monensin significantly improved 42-day body weight and feed:gain and decreased mortality (Hooge *et al.,* 2000). Turkey diets with monensin (99 mg/kg) significantly improved body weight, feed:gain and mortality compared to the zero monensin diets. The combined monensin and $NaHCO_3$ diets had the best body weight, feed:gain and mortality compared to monensin alone or $NaHCO_3$ alone treatments. Hooge *et al.* (2000) reported that dietary $NaHCO_3$ (0.20%) or $KHCO_3$ (0.14%; more expensive) were equally effective in a used litter, coccidial-inoculation broiler pen trial at

improving 45-day body weight, feed conversion ratio, or mortality with lasalocid or monensin. In another trial on used litter and with coccidial challenge, dietary $KHCO_3$ was only zero to 40%, depending on parameter, as effective as $NaHCO_3$ at improving performance with salinomycin up to 46 day. Coccidiosis causes wet litter problems which can be prevented by using higher levels of the ionophore within the legal range or by addition of $NaHCO_3$ to the diets along with the ionophore.

Chapter 3

MATERIALS AND METHODS

Housing and Management

A total of 1440 (experiment-1) and 1656 (experiment-2 and -3) day-old straight-run Hubbard broiler chicks (Hubbard × Hubbard) were allocated to eight (8) dietary treatments replicated four (4) times in such a way that each replicate had 40 (experiment-1) and 46 (experiment-2 and -3) birds. A floor space of 0.09 m^2 was provided to each bird. Each replicate pen was equipped with separate overhead, transparent and volume-graduated 20L water bottle linked with nipple drinker line. Water bottles were cleaned and filled with fresh water after measuring the water consumption on daily basis. One flat bottom round feeder was provided for each experimental pen. Birds were housed in environment control systems where variations in temperature and relative humidity were recorded and maintained according to the production manual. Continuous light was provided 24 hours for the first 3 days and then 23L:1D light pattern was adopted for the rest of the experimental period. A 7.5 cm deep fresh saw dust was used as litter material over a concrete floor. For the first 3 days, house temperature was maintained at 32°C and thereafter reduced by 0.5°C per day until 24°C was attained at d 19.

Birds were vaccinated against Newcastle Disease (**ND**) plus Infectious Bronchitis viruses at 4 d, Infectious Bursal Disease virus at 8 d and again at 14 d; Hydropericardium Syndrome virus at 18 d and ND-Lasota strain at 22 d following the locally designed vaccination schedule.

Dietary Plan

Experiment 1: A basal diet having dNa, K and Cl in amounts of 0.08, 0.71 and 0.20%, respectively with dietary electrolyte balance (DEB) value of 160 mEq/kg (Table 3.1 and 3.2) was formulated on least-cost basis. Four levels of dNa (i.e., (0.17, 0.26, 0.35 and 0.44%; Table 3.2) were supplemented to the basal diet with either commercially available feed-grade sodium bicarbonate (**$NaHCO_3$**; crystalline fine powder containing 27.4% Na) or sodium sulphate (**Na_2SO_4;** in granular crystalline form, containing 32.4% Na) which corresponded to DEB values of 200, 240, 280 and 320 mEq/kg, respectively.

Experiment 2: A basal diet was formulated having dK, Na and Cl as 0.70, 0.15 and 0.30%, respectively with a DEB value of 160 mEq/kg (Table 4.1 and 4.2). Four levels of dK (i.e., 0.86, 1.02, 1.18 and 1.34%) were supplemented to the basal mash diet with either commercially available feed-grade potassium carbonate (**K_2CO_3**; in granular powder form,

containing 56.7% K) or potassium sulphate (**K_2SO_4**; in powder form, containing 44.9% K) which corresponded to DEB values of 200, 240, 280 and 320 mEq/kg, respectively.

Experiment 3: A basal diet having dCl, Na and K as 0.17, 0.30 and 0.92%, respectively with a DEB value of 320 (Table 5.1 and 5.2) was formulated on least-cost basis. Four levels of dCl (0.31, 0.45, 0.59 and 0.73%) were supplemented to the basal diet with either commercially available feed-grade calcium chloride (**$CaCl_2$**; in powder form, containing 63.9% Cl) and ammonium chloride (**NH_4Cl**; in granular form, containing 66.3% Cl) which corresponded to DEB values of 280, 240, 200, and 160 mEq/kg, respectively.

For this purpose, a large batch of basal diet was prepared for each phase and then experimental diets were prepared according to research plan using this basal diet. The experimental period was divided into four phases i.e., pre-starter (1 to 10 d), starter (11 to 20 d), grower (21 to 33 d) and finisher (34 to 42 d) which met or exceeded the nutrient specifications recommended by the Hubbard management guide.

All the ingredients were assayed for their proximate composition (AOAC, 2005) prior to diet formulation and actual values were used in the formulation. The Na and K contents of each diet were analyzed by flame photometer (AOAC, 2005) and Cl by titration with $AgNO_3$ (Lacroix *et al.*, 1970). The K, Na and Cl contents of the final diets were again verified prior to start of the experiment by the above mentioned methods. The ME of each ingredient was calculated by the appropriate regression equation suggested by NRC (1994). Amino acid composition of each ingredient was calculated using AminoDat™ 3.0 Platinum (Degussa AG, Germany) based on the DM and CP contents of each ingredient. The amino acid composition of each diet met or exceeded the ideal amino acid ratio suggested by Han and Baker (1994). The experiment lasted for 42 d of age, offering mash diets throughout the experimental period.

Data Collection

Live performance: Data on feed intake (**FI**), BW gain (**BWG**) and feed-to-gain ratio (**FG**) were recorded for each phase. Feed was withheld for 6 hrs before weighing the birds at the end of each phase to ensure the emptying of the digestive tract of the bird. Water intake (**DWI**) was recorded on daily basis and a ratio between DWI and FI (**DWI:FI**) was also calculated for each phase. Mortality was recorded on daily basis and dead bird was weighed prior to removal to correct FG.

Litter moisture: Litter was collected at the end of each phase to determine its moisture (**LM)**. For this purpose, about 500g litter sample was randomly collected from different locations in each replicate pen. Each sample was homogenized and a representative sample of 100 g was taken and oven dried at 105°C for 24 hrs (AOAC, 2005) to determine moisture contents.

Water analysis: Water characteristics were also recorded twice (morning and noon) daily to check pH by pH metre (LT-Lutron pH-207 Taiwan), dissolved oxygen by DO metre (**DO;** YSI 55 Incorporated, Yellow Springs, Ohio, 4387, USA). Moreover, temperature, electrical conductivity (**EC**), total dissolved solids (**TDS**) and salinity were recorded by Combo metre (H M Digital, Inc. CA 90230). These observations were recorded randomly from different replicates.

Carcass and physiological responses: At the end of 42 d, two birds were randomly selected from each replicate for carcass and blood responses. Blood was collected from wing vein in EDTA-coated vacutainer for immediate pH monitoring. The same birds were killed by severing neck vein and blood samples were collected. Blood serum was separated by centrifugation of blood samples at 2000 × g for 15 minutes (Hayat *et al.*, 1999; Ahmad *et al.*, 2005) and was analyzed for glucose and mineral (Na, K, Cl, Ca, P, Mg, HCO_3) contents. The same birds were further used for evaluation of carcass characteristics. Carcass responses were evaluated in terms of dressing, breast, thigh, abdominal fat, gizzard, proventriculus, heart, liver, kidney, spleen, pancreas, bursa, gall bladder, intestine and lungs weights, and for shank and intestine lengths and were presented as percent of dressed weight.

Statistical Analyses

The experiment was executed under completely randomized design with factorial arrangement using four (4) levels of each electrolyte from two (2) sources of salt. Pen mean was an experimental unit. The data obtained at the end of the experiment were subjected to ANOVA technique using General Linear Model (**GLM**) of Minitab 15.1 (Minitab Inc., State College PA) where all the linear, quadratic and cubic terms were used in the model. When quadratic and cubic responses were found significant ($P < 0.05$), regression equations were used to estimate requirements of each electrolyte (95% of maximum or minimum response). The level of significance was 0.05 unless or otherwise stated.

Chapter 4

RESULTS AND DISCUSSION

Experiment 1

Water was evaluated on daily basis for its quality parametres like temperature, pH, EC, TDS, DO and salinity (Table 4.3). At the start of the experiment, water was also analyzed to contain sodium absorption ratio (25.6) and residual sodium carbonate (9.02). It is anticipated that the concentration of various minerals (cations plus anions) and values of above mentioned parametres in the drinking water could maneuver the electrolyte concentration of ingesta thus their concentration in the water were evaluated in the present study and by previous researchers (Teeter *et al.*, 1985; Borges *et al.*, 2003a;b). The results showed that the electrolytic concentration in the water was too low to have any impact on the growth performance. Water pH values (7.17–7.49) were found within the range (6.0–8.5) considered as optimal for broiler performance (Socha *et al.*, 2002; Borges *et al.*, 2003a; b). Previous reports (Good, 1985; Grizzle *et al.*, 1996) showed retarded growth up to a pH level of 6.3. Water TDS levels ranged from 1000-3000 ppm and these levels were considered satisfactory for broilers (Chiba, 2009) and analyzed values (1060-1284) in the present study were observed not to disturb experimental theme.

BWG ($P \leq 0.04$; Table 4.4) and FI ($P \leq 0.001$; Table 4.5) were significantly affected during d 1-10 by amount and salt of dNa (quadratic and linear term for BWG and FI, respectively, for amount and salt of dNa). When BWG (BWG = 353.3 - 2356 Na + 8343 Na^2 - 9511 Na^3 using $NaHCO_3$ and BWG = 214.6 - 721 Na + 2473 Na^2 - 2521 Na^3 using Na_2SO_4); and FI (FI = 293.3 - 487.3 Na + 1008 Na^2 - 697 Na^3 using $NaHCO_3$ and FI = 480.4 - 2895 Na + 10499 Na^2 - 11702 Na^3 using Na_2SO_4) were optimized for dNa, maximum responses of these parameters were achieved at lowest level (0.17%) of dNa ($NaHCO_3$) and higher level of dNa (Na_2SO_4). It means higher levels of sulphates in Na_2SO_4 supplemented diets induced appetite in birds that would result in higher weight gain. This result could also be attributed towards the better absorption of sulphates ($\mathbf{SO_4^-}$) i.e. sodium dependant active process (Ahearn and Murer, 1984; Florin *et al.*, 1991; Langridge-Smith *et al.*, 1983) and also known to involve in cysteine sparing effects that would result in better growth efficiency in initial days of birds in Na_2SO_4 supplemented diets. With increasing amounts of dNa, BWG ($P \leq 0.05$; Table 4.4) and FG ($P \leq 0.04$; Table 4.6) were declined during d 11-20. Cubic responses of dNa were found significant on BWG for d 21-33 ($P \leq 0.04$) and d 34-42 ($P \leq 0.009$). Upon optimization with regression equations, 0.17% (Y = 1166 - 3134 dNa+ 9423 dNa^2 - 8930 dNa^3) and 0.24% (Y = - 935.8 + 16138 dNa - 52913 dNa^2 + 54434 dNa^3) dNa were observed for maximum BWG

during d 21-33 and d 34-42, respectively. Improved FG was found at the level of dNa i.e. 0.37%, supplemented with Na_2SO_4 when compared with $NaHCO_3$ which is optimized at 0.20% dNa during d 21-33 ($P \leq 0.02$). It means birds can tolerate high levels of dNa when Na_2SO_4 was added as source of Na when compared with $NaHCO_3$. Previously Mushtaq *et al.*, (2007) has suggested dietary requirements of 0.20 to 0.25% dNa for the finisher phase (d 29-42) by adding $NaHCO_3$ and NaCl. The difference between results might be due to high ambient temperature (32-40°C), varying levels of Cl^- (0.30, 0.40 and 0.50%) or extended period of rearing (d 29-42). We have designed phase feeding experiment to determine phase requirement of amount and sources of dNa but as some phases show non-significant results so their requirements could not be determined *per se* except BWG.

Significant ($P \leq 0.04$) linear rise in DWI was observed during d 21-33 ($P \leq 0.007$), d 34-42 ($P \leq 0.01$) and d 1-42 ($P \leq 0.04$) by increasing dNa levels from 0.17 to 0.44% (Table 4.7). It is obvious from previous studies that increasing dNa levels enhances the water consumption linearly (Julian *et al.*, 1992; Murakami *et al.*, 2000; Rondon *et al.*, 2001; Mushtaq *et al.* 2005). Actually supplementation of electrolytes (Na, K, Cl) by using various salts could modify the body fluid and osmotic balance that increases thirst and leads to enhance water consumption (Smith and Teeter, 1988; Borges *et al.*, 2004; 2004a). Birds showed similar responses in water intake with increasing dNa in initial stages while drank more water in last stages of life ($P > 0.05$). However this water was not excreted through faeces as evident from similar litter condition in all dietary treatments in later stages of life and was retained in the body. It means heavier birds retain more water with increasing dNa. DWI:FI was found significant during d 1-10 ($P \leq 0.04$) and d 34-42 ($P \leq 0.03$) by replacing salts (Table 4.8). Numerically, increasing level of dNa by adding $NaHCO_3$ salt showed increased DWI:FI up to d 20 while same pattern was observed in Na_2SO_4 supplemental diets in the rest of the phases ($P > 0.05$; Table 3.8). Higher water intake limits the feed consumption, excretes nutrients in faeces thus reduces weight gain in $NaHCO_3$ (d 1-10) and Na_2SO_4 (d 34-42) supplemented diets. Ahmad *et al.*, (2005) observed pronounced effects on DWI:FI during overall period when comparing $NaHCO_3$ with other salt sources. The difference in results might be owing to the difference in high cyclic ambient temperature (29-33°C) and fixation of DEB (250 mEq/kg).

LM ($P \leq 0.01$; Table 4.9; linear term) and mortality ($P \leq 0.04$; Table 4.10; cubic term) were affected by amount and source of dNa during d 1-10. Linear decrease in LM was observed by using $NaHCO_3$ while linear increase in LM in case of Na_2SO_4. Similar situation was observed in BWG and FI in initial days that did not affect feed efficiency *per se*. After d 10, birds showed similar pattern of LM throughout the experimental period. It seems birds excreted less owing to reduced water intake by increasing dNa in initial days ($P > 0.05$). It is anticipated

that birds can retain more water in muscle tissues and excrete less quantity of water with high levels of dNa. This observation is supported from higher breast and thigh meat yield ($P<0.05$) with increasing dNa. In contrast, Mushtaq *et al.* (2007) found reduced breast meat and dressing percentage. These results could be differentiated as environmental (heat stress) condition and rearing age. During heat stress it is assumed that most nutrients were consumed for maintenance of physiological system (Mushtaq *et al.*, 2007). When mortality was optimized against levels of dNa by supplementing Na_2SO_4, lowest level of mortality (i.e. 0.37%) was observed at 0.37% dNa while highest mortality (i.e. 2.52%) in the counterpart (0.37% dNa) by using $NaHCO_3$. It indicates that salt source is more important in this scenario. Mortality results were found similar in Na_2SO_4 and $NaHCO_3$ supplemental salt groups by Ahmad *et al.* (2005). It was demonstrated that the birds that drank more water show less mortality under heat stress.

At 13th d, birds were found lame, and both amount and source were responsible for it (cubic term between amount and source; $P\leq0.05$; Table 4.11). By using polynomial regression equation, lowest level of lameness i.e. 3.50 and 0.10%, were found in 0.44 and 0.33% dNa in $NaHCO_3$ and Na_2SO_4 supplemental salts, respectively. It is observed that excess dNa could be involved in excessive excretion of phosphorus so causes its deficiency (Hooge, 2003) and leads towards calcium phosphorus imbalance and creates leg abnormality.

Significant quadratic rise in pH ($P\leq0.01$) and decrease in DP ($P\leq0.05$) were observed by amount and source of salt of dNa (Table 4.11). The diets containing high dNa showed increased pH i.e. 7.37 (mild alkalosis). The value of pH greater than 7.30 is known to decrease production performance in broilers as evident from decline in weight gain (however non-significant) when data was pooled for d 1-42. This decline in weight gain might be the impact of acid base imbalance (Hurwitz *et al.* 1973). Mushtaq *et al.* (2007) also observed quadratic increase in pH by dNa (0.20 to 0.30%) during d 29-42. Actually for proper functioning of intermediary metabolism and cellular activity, body physiological system requires optimum pH which in turn controls enzymatic activity and ultimately leads towards better growth performance (Lehninger, 1970). Mongin (1981) was of the view that when DEB is other than 250 mEq/kg of diet, either acidosis or alkalosis develops which may lead to retarded growth performance. This is also evident from present study that highest level of dNa (0.44%; DEB = 320 mEq/kg) showed reduced growth (1864g) as compared to 0.26% dNa or DEB = 240 mEq/kg (1959g; $P>0.05$). Reduced DP in birds fed $NaHCO_3$ supplemented diets was also observed by Ahmad *et al.* (2005; 2006) who found better performance however, when compared this salt with ionic salts. This reduced DP might be due to the heat distressed condition offered in the experiment of Ahmad *et al.* (2005; 2006). In heat stress condition, bicarbonate ions were utilized to correct the stress condition hence more nutrients were

converted for valuable meat production. Breast (P≤0.001) and thigh (P≤0.001) meat yield increased with dNa (Table 4.11). Increased breast and thigh meat yield might be the result of increased tissue water accumulation in high dNa diets as evident from high water intake (P≤0.04). These findings were not in line the results of Mushtaq *et al.*, (2007) who observed reduced breast and leg meat by increasing dNa from 0.20 to 0.30%. This could be attributed towards heat stress condition provided in the experiment so more nutrients were consumed to maintain acid base balance and not converted to meat. It could be concluded from present study that by increasing dNa under normal physiological condition might improve the basal metabolism and energy is mainly utilized for meat production and less is wasted as abdominal fat.

Increasing levels of dNa from 0.17 to 0.44% increased gizzard weight (P≤0.002) while kidney weight was increased by amount and salt (P≤0.006; Table 4.12; linear interaction). Bicarbonate (HCO_3^-) buffer system mainly determines the blood acid-base balance and functions under the regulatory control of kidneys (Hooge, 2003) that's why kidney size was increased in $NaHCO_3$ supplemented diets. Pancreas (P≤0.05), gall bladder (P≤0.002) weights, and shank length (P≤0.05; cubic term), while bursa (P≤0.006) and lungs (P≤0.05; quadratic term) weights were affected by amount and source of dNa (Table 4.13).

Serum K (P≤0.001), Cl (P≤0.001), Mg (≤0.001) and HCO_3 (P≤0.001) were affected by supplementation of dNa (cubic term remain significant for amount and source of Na), however quadratic increase in serum Na (P≤0.001) was observed with increasing dNa (Table 4.14). This significant increase in serum Na caused more water intake and ultimately increased litter dampness as evident from the current study. Mushtaq *et al.* (2005) had suggested to maintain a balance between cations and anions in order to keep the acid-base homeostasis. This balance could be maintained by providing proper DEB so that mild alkalosis caused by increased supplementation of dNa could be compensated by dietary supplementation of anions. Increased serum anionic concentration might be to compensate increased cations in the blood. It is anticipated that serum Na and K are directly related to each other in blood and leads towards osmoregulation of body fluids (Mushtaq *et al.*, 2005). Effect of dNa is different on different serum mineral concentrations, so considering only the individual electrolyte is not sufficient to understand the whole phenomenon of DEB.

It was concluded that birds showed better growth performance and reduced mortality against high levels of dietary sodium in Na_2SO_4 than $NaHCO_3$ supplemented diets, however it is difficult to define amount of dietary sodium for each phase against each parametre. Significant rise in breast and thigh meat yield while reduced dressing percentage were observed with increasing dietary sodium.

Table 4.1. Ingredient composition of the basal diets fed four levels of sodium with two sources of sodium salts at different stages of growth in broilers[1]

Ingredients (%)	Pre-starter (1 – 10 d)	Starter (11 – 20 d)	Grower (21 – 33 d)	Finisher (34 – 42 d)
Corn	46.93	47.54	67.46	67.94
Broken rice	13.46	16.16	0.07	–
Soybean meal	27.63	29.11	26.93	24.47
Canola Meal	6.39	1.24	–	–
Oil	1.62	2.21	1.88	3.94
DCP	2.16	2.02	1.89	1.60
Limestone	1.04	0.94	1.05	1.10
L-Lysine HCl	0.23	0.22	0.20	–
L-Lysine sulphate	–	–	–	0.34
NaCl	0.15	0.16	0.16	0.16
KCl	0.04	0.03	0.05	0.14
DL-methionine	0.20	0.20	0.17	0.17
L-threonine	0.05	0.07	0.03	0.04
Premix[2]	0.10	0.10	0.10	0.10

[1]All the diets were supplemented with 4 levels of either $NaHCO_3$ (0.33, 0.66, 0.99, or 1.32%) or Na_2SO_4 (0.28, 0.56, 0.84 or 1.12%) to make final Na concentrations of 0.17, 0.26, 0.35 or 0.44%, respectively. The basal diet has 0.08% Na in it.

[2]Provides per kg of finished diet: vitamin A, 12 mg; vitamin D_3, 7 mg; vitamin E, 100 mg; vitamin K_3 (50% as MNB), 3 mg; vitamin B_1 (98%), 3 mg; vitamin B_2 (800,000 mg), 12 mg; vitamin B_3 (niacin; 99%), 600 mg; vitamin B_6 (98%), 4 mg; vitamin B_9 (folic acid; 95%), 2 mg; vitamin B_{12} (0.10%), 20 mg; Biotin (0.10%), 5 mg; Ca-Pentothenate (98%), 12 mg; cholin (70% as choline sodium), 1 g; MnO (60%), 169 mg; $FeSO_4$ (21%), 200 mg; $ZnSO_4$ (36%), 150 mg; $CuSO_4$ (25%), 40 mg; Se (sodium selenite 0.40%), 100 mg; KI (68%), 2 mg; Salinomycin, 60 mg; Zinc bacitracin (as Albac 10%), 50 mg

Table 4.2. Nutrient composition of the basal diets for different phases of birds fed four levels of sodium with two sources of sodium salts[1]

Nutrients (%)	Pre-starter (1 – 10 d)	Starter (11 – 20 d)	Grower (21 – 33 d)	Finisher (34 – 42 d)
ME (kcal/kg)	2900	3000	3000	3147
Crude Protein	21.00	20.00	19.00	18.00
Crude fiber	4.14	3.72	3.37	3.21
Calcium	1.00	0.90	0.90	0.85
Available Phos.	0.45	0.42	0.40	0.35
Sodium	0.08	0.08	0.08	0.08
Potassium	0.71	0.71	0.71	0.71
Chloride	0.20	0.20	0.20	0.20
DEB (mEq/kg)	160	160	160	160
Dig Lys	1.10	1.05	0.97	0.93
Dig Met/ Dig Lys	0.45	0.45	0.44	0.44
Dig Met + Dig Cys/Dig Lys	0.72	0.71	0.72	0.72
Dig Thr/Dig Lys	0.66	0.67	0.66	0.66
Dig Try/Dig Lys	0.18	0.18	0.18	0.17
Dig Arg/Dig Lys	1.12	1.14	1.12	1.09

[1] $NaHCO_3$ and Na_2SO_4

Table 4.3. Drinking water properties during the experimental period

Phase	Item[1]	Salinity (g/kg)	TDS[2] (mg/kg)	EC[3] (millisemen/cm)	Temperature (°C)	pH	DO[4] (mg/L)
Phase 1	Max	1.30	1284	1.39	29.7	7.49	5.40
	Min	1.20	1198	1.27	26.2	7.31	3.90
	Average	1.25	1251	1.32	27.3	7.40	4.65
Phase 2	Max	1.20	1111	1.21	26.4	7.35	5.07
	Min	1.20	1060	1.06	25.8	7.17	3.70
	Average	1.20	1088	1.12	26.0	7.27	4.39
Phase 3	Max	1.30	1187	1.24	25.2	7.40	5.60
	Min	1.20	1108	1.08	24.3	7.21	3.70
	Average	1.22	1144	1.15	24.9	7.33	4.65
Phase 4	Max	1.20	1194	1.21	24.8	7.33	5.07
	Min	1.20	1110	1.07	24.0	7.23	3.70
	Average	1.20	1141	1.11	24.2	7.29	4.39

[1]Max, Min and Average values were taken twice daily so one value is equal to days in phase × 2
[2]TDS - Total Dissolved Solids; [3]EC - Electrical Conductivity; [4]DO - Dissolved Oxygen

Table 4.4. Effect of dietary sodium and sodium salts on body weight gain of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			g		
Basal (0.08%)	152	337	827	600	1916
SEM	*2.4*	*13.8*	*9.0*	*24.1*	*31.3*
Dietary Na (%)					
0.17	149	336	862	546	1893
0.26	144	344	831	640	1959
0.35	150	328	840	564	1883
0.44	141	314	851	558	1864
SEM	*4.2*	*13.5*	*19.8*	*26.4*	*30.5*
Salts					
$NaHCO_3$	137	317	838	591	1884
Na_2SO_4	155	344	854	563	1916
SEM	*3.0*	*9.5*	*14.0*	*18.7*	*21.6*
Na × Salts					
0.17 × $NaHCO_3$	147	338	864	592	1941
0.26 × $NaHCO_3$	138	324	849	630	1941
0.35 × $NaHCO_3$	143	295	807	584	1829
0.44 × $NaHCO_3$	122	311	833	558	1824
0.17 × Na_2SO_4	151	334	859	500	1844
0.26 × Na_2SO_4	150	364	813	650	1977
0.35 × Na_2SO_4	157	361	874	545	1936
0.44 × Na_2SO_4	161	318	868	558	1905
SEM	*6.0*	*19.0*	*28.0*	*37.3*	*43.1*
ANOVA			***Probability***		
Na	-	0.05	-	-	-
Na_L	-	-	-	-	-
Na_Q	-	-	-	-	-
Na_C	-	-	0.04	0.009	-
Salt	0.009	-	-	-	-
Salt × Na	0.04	-	-	-	-
Salt × Na_L	-	-	-	-	-
Salt × Na_Q	0.04	-	-	-	-
Salt × Na_C	-	-	-	-	-

Na_L, Na_Q & Na_C are linear, quadratic and cubic terms for Na, respectively.

Table 4.5. Effect of dietary sodium and sodium salts on feed intake of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			g		
Basal (0.08%)	236	632	1775	1093	3736
SEM	*5.2*	*5.3*	*11.6*	*39.1*	*99.3*
Dietary Na (%)					
0.17	235	643	1649	1036	3564
0.26	227	663	1654	1089	3634
0.35	234	652	1652	1067	3605
0.44	228	681	1612	1101	3622
SEM	*3.3*	*15.9*	*49.2*	*24.0*	*45.9*
Salts					
$NaHCO_3$	222	653	1640	1089	3604
Na_2SO_4	240	667	1644	1058	3608
SEM	*2.3*	*11.2*	*34.8*	*17.0*	*32.4*
Na × Salts					
0.17 × $NaHCO_3$	236	635	1644	1071	3586
0.26 × $NaHCO_3$	222	658	1652	1099	3631
0.35 × $NaHCO_3$	216	650	1725	1057	3649
0.44 × $NaHCO_3$	215	668	1539	1129	3551
0.17 × Na_2SO_4	234	651	1654	1002	3542
0.26 × Na_2SO_4	232	669	1657	1080	3637
0.35 × Na_2SO_4	251	653	1578	1078	3560
0.44 × Na_2SO_4	242	694	1685	1073	3694
SEM	*4.6*	*22.4*	*69.5*	*33.9*	*64.9*
ANOVA			***Probability***		
Na	-	-	-	-	-
Na_L	-	-	-	-	-
Na_Q	-	-	-	-	-
Na_C	-	-	-	-	-
Salt	≤0.001	-	-	-	-
Salt × Na	0.002	-	-	-	-
Salt × Na_L	0.001	-	-	-	-
Salt × Na_Q	-	-	-	-	-
Salt × Na_C	-	-	-	-	-

Na_L, Na_Q & Na_C are linear, quadratic and cubic terms for Na, respectively.

Table 4.6. Effect of dietary sodium and sodium salts on feed:gain of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			g/g		
Basal (0.08%)	1.56	1.88	2.15	1.83	1.88
SEM	*0.052*	*0.089*	*0.137*	*0.084*	*0.056*
Dietary Na (%)					
0.17	1.58	1.92	1.92	2.06	1.95
0.26	1.59	1.94	2.00	1.71	1.80
0.35	1.57	2.01	1.97	1.90	1.89
0.44	1.64	2.22	1.90	1.98	1.95
SEM	*0.046*	*0.098*	*0.058*	*0.166*	*0.076*
Salts					
$NaHCO_3$	1.64	2.10	1.97	1.85	1.88
Na_2SO_4	1.55	1.95	1.93	1.98	1.91
SEM	*0.032*	*0.069*	*0.041*	*0.117*	*0.053*
Na × Salts					
0.17 × $NaHCO_3$	1.61	1.88	1.91	1.81	1.82
0.26 × $NaHCO_3$	1.62	2.05	1.96	1.76	1.83
0.35 × $NaHCO_3$	1.54	2.22	2.14	1.81	1.91
0.44 × $NaHCO_3$	1.78	2.25	1.86	2.02	1.98
0.17 × Na_2SO_4	1.55	1.96	1.93	2.32	2.08
0.26 × Na_2SO_4	1.55	1.84	2.04	1.66	1.76
0.35 × Na_2SO_4	1.61	1.81	1.81	1.99	1.88
0.44 × Na_2SO_4	1.51	2.20	1.94	1.93	1.91
SEM	*0.065*	*0.138*	*0.082*	*0.234*	*0.107*
ANOVA			***Probability***		
Na	-	-	-	-	-
Na_L	-	0.04	-	-	-
Na_Q	-	-	-	-	-
Na_C	-	-	-	-	-
Salt	-	-	-	-	-
Salt × Na	-	-	0.05	-	-
Salt × Na_L	-	-	-	-	-
Salt × Na_Q	-	-	-	-	-
Salt × Na_C	-	-	0.02	-	-

Na_L, Na_Q & Na_C are linear, quadratic and cubic terms for Na, respectively.

Table 4.7. Effect of dietary sodium and sodium salts on water intake of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			mL		
Basal (0.08%)	647	1721	3559	2393	8320
SEM	*26.5*	*70.2*	*95.3*	*79.5*	*110.0*
Dietary Na (%)					
0.17	685	1777	3411	2422	8294
0.26	683	1817	3740	2657	8898
0.35	666	1751	3730	2561	8709
0.44	650	1722	3835	2733	8940
SEM	*26.1*	*54.7*	*96.2*	*68.7*	*176.2*
Salts					
$NaHCO_3$	674	1766	3589	2544	8574
Na_2SO_4	668	1767	3769	2642	8846
SEM	*18.5*	*38.7*	*68.0*	*48.6*	*124. 6*
Na × Salts					
0.17 × $NaHCO_3$	720	1833	3340	2349	8241
0.26 × $NaHCO_3$	695	1740	3651	2596	8683
0.35 × $NaHCO_3$	670	1796	3631	2472	8570
0.44 × $NaHCO_3$	611	1696	3734	2760	8801
0.17 × Na_2SO_4	650	1721	3482	2494	8347
0.26 × Na_2SO_4	671	1894	3829	2718	9112
0.35 × Na_2SO_4	662	1706	3830	2649	8848
0.44 × Na_2SO_4	689	1747	3936	2706	9078
SEM	*36.9*	*77.4*	*136.0*	*97.1*	*249.1*
ANOVA			***Probability***		
Na	-	-	0.02	0.02	-
Na_L	-	-	0.007	0.01	0.04
Na_Q	-	-	-	-	-
Na_C	-	-	-	-	-
Salt	-	-	-	-	-
Salt × Na	-	-	-	-	-
Salt × Na_L	-	-	-	-	-
Salt × Na_Q	-	-	-	-	-
Salt × Na_C	-	-	-	-	-

Na_L, Na_Q & Na_C are linear, quadratic and cubic terms for Na, respectively.

Table 4.8. Effect of dietary sodium and sodium salts on water intake-to-feed intake ratio of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			mL/g		
Basal (0.08%)	2.74	2.72	2.03	2.19	2.23
SEM	*0.086*	*0.131*	*0.131*	*0.064*	*0.060*
Dietary Na (%)					
0.17	2.91	2.76	2.09	2.36	2.33
0.26	3.01	2.74	2.28	2.44	2.45
0.35	2.87	2.71	2.28	2.40	2.42
0.44	2.84	2.53	2.40	2.48	2.47
SEM	*0.110*	*0.091*	*0.108*	*0.072*	*0.062*
Salts					
$NaHCO_3$	3.03	2.72	2.20	2.34	2.38
Na_2SO_4	2.79	2.66	2.32	2.51	2.46
SEM	*0.078*	*0.064*	*0.077*	*0.051*	*0.044*
Na × Salts					
0.17 × $NaHCO_3$	3.05	2.89	2.03	2.19	2.30
0.26 × $NaHCO_3$	3.13	2.64	2.22	2.36	2.39
0.35 × $NaHCO_3$	3.10	2.78	2.13	2.34	2.35
0.44 × $NaHCO_3$	2.84	2.55	2.43	2.44	2.48
0.17 × Na_2SO_4	2.78	2.64	2.15	2.52	2.36
0.26 × Na_2SO_4	2.90	2.84	2.34	2.52	2.51
0.35 × Na_2SO_4	2.63	2.63	2.43	2.46	2.49
0.44 × Na_2SO_4	2.84	2.51	2.36	2.53	2.46
SEM	*0.156*	*0.128*	*0.153*	*0.102*	*0.087*
ANOVA			*Probability*		
Na	-	-	-	-	-
Na_L	-	-	-	-	-
Na_Q	-	-	-	-	-
Na_C	-	-	-	-	-
Salt	0.04	-	-	0.03	-
Salt × Na	-	-	-	-	-
Salt × Na_L	-	-	-	-	-
Salt × Na_Q	-	-	-	-	-
Salt × Na_C	-	-	-	-	-

Na_L, Na_Q & Na_C are linear, quadratic and cubic terms for Na, respectively.

Table 4.9. Effect of dietary sodium and sodium salts on litter moisture of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			%		
Basal (0.08%)	14.8	36.9	38.9	34.2	31.2
SEM	*1.25*	*2.12*	*2.22*	*2.74*	*1.20*
Dietary Na (%)					
0.17	14.6	38.3	47.8	35.0	33.9
0.26	14.1	32.3	41.9	40.4	32.2
0.35	14.8	34.8	42.7	35.6	32.0
0.44	15.5	38.0	47.6	37.1	34.6
SEM	*0.67*	*2.68*	*3.94*	*2.86*	*1.45*
Salts					
$NaHCO_3$	14.8	37.0	45.6	35.5	33.2
Na_2SO_4	14.7	34.6	44.4	38.5	33.1
SEM	*0.48*	*1.90*	*2.78*	*2.02*	*1.02*
Na × Salts					
0.17 × $NaHCO_3$	15.8	42.4	44.7	33.7	34.1
0.26 × $NaHCO_3$	15.0	32.7	43.4	35.9	31.8
0.35 × $NaHCO_3$	13.8	35.9	48.2	32.0	32.5
0.44 × $NaHCO_3$	14.6	37.2	46.1	40.5	34.6
0.17 × Na_2SO_4	13.3	34.2	50.9	36.3	33.7
0.26 × Na_2SO_4	13.3	31.9	40.4	45.0	32.6
0.35 × Na_2SO_4	15.7	33.6	37.3	39.1	31.4
0.44 × Na_2SO_4	16.4	38.9	49.1	33.7	34.5
SEM	*0.95*	*3.79*	*5.57*	*4.04*	*2.05*
ANOVA			***Probability***		
Na	-	-	-	-	-
Na_L	-	-	-	-	-
Na_Q	-	-	-	-	-
Na_C	-	-	-	-	-
Salt	-	-	-	-	-
Salt × Na	-	-	-	-	-
Salt × Na_L	0.01	-	-	-	-
Salt × Na_Q	-	-	-	-	-
Salt × Na_C	-	-	-	-	-

Na_L, Na_Q & Na_C are linear, quadratic and cubic terms for Na, respectively.

Table 4.10. Effect of dietary sodium and sodium salts on mortality of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			%		
Basal (0.08%)	4.0	1.7	0.0	0.0	5.7
SEM	*3.26*	*1.09*	*0.00*	*0.00*	*4.20*
Dietary Na (%)					
0.17	2.3	0.6	0.6	0.6	4.0
0.26	1.4	0.3	1.1	0.0	2.8
0.35	1.4	0.6	1.1	0.0	3.1
0.44	1.4	0.0	0.6	0.3	2.3
SEM	*0.87*	*0.36*	*0.52*	*0.22*	*1.28*
Salts					
$NaHCO_3$	1.1	0.0	0.9	0.0	2.0
Na_2SO_4	2.1	0.7	0.9	0.4	4.1
SEM	*0.62*	*0.25*	*0.37*	*0.15*	*0.91*
Na × Salts					
0.17 × $NaHCO_3$	2.3	0.0	0.6	0.0	2.8
0.26 × $NaHCO_3$	0.0	0.0	1.1	0.0	1.1
0.35 × $NaHCO_3$	2.3	0.0	1.7	0.0	4.0
0.44 × $NaHCO_3$	0.0	0.0	0.0	0.0	0.0
0.17 × Na_2SO_4	2.3	1.1	0.6	1.1	5.1
0.26 × Na_2SO_4	2.8	0.6	1.1	0.0	4.6
0.35 × Na_2SO_4	0.6	1.1	0.6	0.0	2.3
0.44 × Na_2SO_4	2.8	0.0	1.1	0.6	4.6
SEM	*1.23*	*0.51*	*0.73*	*0.31*	*1.82*
ANOVA			***Probability***		
Na	-	-	-	-	-
Na_L	-	-	-	-	-
Na_Q	-	-	-	-	-
Na_C	-	-	-	-	-
Salt	-	-	-	-	-
Salt × Na	-	-	-	-	-
Salt × Na_L	-	-	-	-	-
Salt × Na_Q	-	-	-	-	-
Salt × Na_C	0.04	-	-	-	-

Na_L, Na_Q & Na_C are linear, quadratic and cubic terms for Na, respectively.

Table 4.11. Effect of dietary sodium and sodium salts on blood and carcass responses of broilers at the end of the experiment

Item	Blood pH	Dressing weight[1]	Breast weight[2]	Thigh weight[2]	Intestinal weight[3]	Intestinal length[4]	Lame Birds[5]
Basal (0.08%)	7.27	53.89	32.22	46.16	58.0	1.9	
SEM	*0.005*	*0.580*	*0.686*	*1.150*	*1.53*	*0.05*	
Dietary Na (%)							
0.17	7.30	56.12	31.85	45.26	58.2	1.9	6.2
0.26	7.33	54.18	32.98	46.28	54.3	1.8	3.1
0.35	7.35	52.94	33.79	47.34	54.9	1.9	3.8
0.44	7.37	52.07	34.31	48.30	56.1	1.7	4.1
SEM	*0.002*	*0.423*	*0.347*	*0.627*	*1.15*	*0.07*	*1.25*
Salts							
$NaHCO_3$	7.35	53.95	33.16	46.28	56.3	1.8	5.1
Na_2SO_4	7.33	53.70	33.30	47.31	55.4	1.9	3.6
SEM	*0.002*	*0.299*	*0.246*	*0.443*	*0.81*	*0.05*	*0.88*
Na × Salts							
0.17 × $NaHCO_3$	7.31	55.68	32.11	45.29	58.8	1.9	5.0
0.26 × $NaHCO_3$	7.34	54.85	32.56	45.05	56.2	1.8	4.5
0.35 × $NaHCO_3$	7.36	53.38	33.57	46.79	58.7	2.1	7.2
0.44 × $NaHCO_3$	7.38	51.90	34.40	48.01	51.6	1.7	3.5
0.17 × Na_2SO_4	7.30	56.56	31.60	45.24	57.7	2.0	7.5
0.26 × Na_2SO_4	7.32	53.50	33.40	47.51	52.4	1.8	1.75
0.35 × Na_2SO_4	7.34	52.51	34.01	47.90	51.1	1.8	0.25
0.44 × Na_2SO_4	7.36	52.24	34.22	48.58	60.7	1.8	4.75
SEM	*0.003*	*0.599*	*0.492*	*0.886*	*1.62*	*0.10*	*1.76*
ANOVA		———	———	***Probability***	———	———	
Na	≤0.001	≤0.001	≤0.001	0.008	-	-	-
Na_L	≤0.001	≤0.001	≤0.001	≤0.001	-	-	-
Na_Q	≤0.001	-	-	-	0.03	-	-
Na_C	-	-	-	-	-	-	-
Salt	≤0.001	-	-	-	-	-	-
Salt × Na	0.03	-	-	-	≤0.001	-	0.05
Salt × Na_L	-	-	-	-	0.01	-	-
Salt × Na_Q	0.01	≤0.05	-	-	≤0.001	-	-
Salt × Na_C	-	-	-	-	0.04	-	-

[1]% of live weight (without visceral organs)
[2]% of dressed weight
[3]measured in gram, [4]centimeters and [5]percentage, respectively
Na_L, Na_Q & Na_C are linear, quadratic and cubic terms for Na, respectively.

Table 4.12. Effect of dietary sodium and sodium salts on body organ weights of broilers at the end of the experiment

Item	Gizzard[1]	Proventriculus[1]	Heart[1]	Liver[1]	Kidney[1]
		% of dressed weight			
Basal (0.08%)	2.77	0.60	0.56	2.55	0.41
SEM	*0.207*	*0.064*	*0.036*	*0.398*	*0.051*
Dietary Na (%)					
0.17	2.41	0.61	0.52	2.61	0.31
0.26	2.57	0.63	0.59	2.89	0.36
0.35	2.98	0.63	0.66	2.90	0.37
0.44	2.99	0.64	0.59	2.74	0.37
SEM	*0.15*	*0.04*	*0.04*	*0.14*	*0.02*
Salts					
$NaHCO_3$	2.85	0.64	0.62	2.88	0.42
Na_2SO_4	2.62	0.61	0.56	2.68	0.28
SEM	*0.10*	*0.03*	*0.03*	*0.10*	*0.02*
Na × Salts					
0.17 × $NaHCO_3$	2.72	0.61	0.56	2.90	0.42
0.26 × $NaHCO_3$	2.65	0.63	0.65	3.08	0.47
0.35 × $NaHCO_3$	2.98	0.73	0.69	2.76	0.41
0.44 × $NaHCO_3$	3.05	0.61	0.59	2.80	0.39
0.17 × Na_2SO_4	2.11	0.61	0.47	2.32	0.21
0.26 × Na_2SO_4	2.48	0.63	0.53	2.70	0.24
0.35 × Na_2SO_4	2.98	0.53	0.63	3.04	0.33
0.44 × Na_2SO_4	2.93	0.66	0.60	2.68	0.35
SEM	*0.21*	*0.06*	*0.06*	*0.20*	*0.04*
ANOVA		***Probability***			
Na	0.01	-	-	-	-
Na_L	0.002	-	-	-	-
Na_Q	-	-	-	-	-
Na_C	-	-	-	-	-
Salt	-	-	-	-	≤0.001
Salt × Na	-	-	-	-	0.03
Salt × Na_L	-	-	-	-	0.006
Salt × Na_Q	-	-	-	-	-
Salt × Na_C	-	-	-	-	-

[1]% of live weight (with visceral organs)

Na_L, Na_Q & Na_C are linear, quadratic and cubic terms for Na, respectively.

Table 4.13. Effect of dietary sodium and sodium salts on body organ weights of broilers at the end of the experiment

Item	Spleen[2]	Pancreas[2]	Bursa[2]	Gall bladder[2]	Lungs[2]	Abdominal fat[1]	Shank length[3]
			% of dressed weight				
Basal (0.08%)	0.04	0.36	0.25	0.11	0.49	2.96	6.24
SEM	*0.017*	*0.021*	*0.03*	*0.01*	*0.03*	*0.43*	*0.18*
Dietary Na (%)							
0.17	0.13	0.29	0.23	0.08	0.56	3.01	6.06
0.26	0.08	0.31	0.15	0.08	0.58	3.12	6.26
0.35	0.07	0.29	0.21	0.09	0.60	2.44	5.95
0.44	0.08	0.31	0.25	0.12	0.48	2.61	6.44
SEM	*0.02*	*0.02*	*0.02*	*0.01*	*0.03*	*0.24*	*0.16*
Salts							
$NaHCO_3$	0.09	0.30	0.21	0.07	0.58	2.74	6.12
Na_2SO_4	0.09	0.30	0.22	0.11	0.54	2.86	6.23
SEM	*0.01*	*0.01*	*0.01*	*0.01*	*0.02*	*0.17*	*0.12*
Na × Salts							
0.17 × $NaHCO_3$	0.11	0.28	0.16	0.07	0.64	2.96	6.24
0.26 × $NaHCO_3$	0.10	0.34	0.18	0.07	0.61	2.93	5.85
0.35 × $NaHCO_3$	0.08	0.27	0.22	0.04	0.54	1.93	6.01
0.44 × $NaHCO_3$	0.07	0.30	0.26	0.11	0.51	3.12	6.38
0.17 × Na_2SO_4	0.15	0.29	0.29	0.10	0.49	3.06	5.89
0.26 × Na_2SO_4	0.16	0.28	0.13	0.09	0.56	3.31	6.66
0.35 × Na_2SO_4	0.06	0.31	0.20	0.13	0.64	2.94	5.89
0.44 × Na_2SO_4	0.08	0.33	0.24	0.12	0.46	2.11	6.50
SEM	*0.02*	*0.02*	*0.02*	*0.01*	*0.04*	*0.35*	*0.23*
ANOVA				***Probability***			
Na	-	-	0.001	0.01	0.05	-	-
Na_L	0.02	-	-	0.005	-	-	-
Na_Q	-	-	0.01	-	0.03	-	-
Na_C	-	-	0.03	-	-	-	-
Salt	-	-	-	≤0.001	-	-	-
Salt × Na	-	-	0.001	0.003	0.04	0.04	-
Salt × Na_L	-	-	0.008	-	-	-	-
Salt × Na_Q	-	-	0.006	0.03	0.05	0.02	-
Salt × Na_C	-	0.05	-	0.002	-	-	0.05

[1]% of live weight (without visceral organs); [2]% of dressed weight;
[3]shank length was measured from hock joint to tarsometatarsus joint in centrimetre;
Na_L, Na_Q & Na_C are linear, quadratic and cubic terms for Na, respectively

Table 4.14. Effect of dietary sodium and sodium salts on serum mineral chemistry of broilers at the end of the experiment

Item	Na	K	Cl	Ca	P	Mg	HCO_3
		mmol/L					
Basal (0.08%)	130	3.04	101	2.36	2.79	1.05	29.7
SEM	*0.3*	*0.005*	*0.5*	*0.047*	*0.084*	*0.093*	*0.09*
Dietary Na (%)							
0.17	131	3.04	102	2.52	2.86	1.06	30.8
0.26	132	3.05	102	2.56	2.86	1.15	31.1
0.35	135	3.07	104	2.68	2.92	1.18	30.0
0.44	137	3.04	101	2.71	2.79	1.14	30.6
SEM	*0.2*	*0.004*	*0.3*	*0.056*	*0.037*	*0.018*	*0.14*
Salts							
$NaHCO_3$	134	3.06	102	2.50	2.86	1.14	31.4
Na_2SO_4	132	3.05	102	2.73	2.86	1.12	30.8
SEM	*0.1*	*0.003*	*0.2*	*0.039*	*0.026*	*0.038*	*0.05*
Na × Salts							
0.17 × $NaHCO_3$	132	3.04	103	2.45	2.88	1.11	31.1
0.26 × $NaHCO_3$	134	3.06	102	2.40	2.86	1.07	31.4
0.35 × $NaHCO_3$	137	3.09	104	2.56	2.95	1.28	29.7
0.44 × $NaHCO_3$	138	3.02	100	2.60	2.74	1.11	30.7
0.17 × Na_2SO_4	130	3.04	101	2.58	2.84	1.01	30.5
0.26 × Na_2SO_4	131	3.04	102	2.71	2.86	1.22	30.7
0.35 × Na_2SO_4	133	3.05	103	2.79	2.89	1.09	30.2
0.44 × Na_2SO_4	135	3.06	102	2.82	2.84	1.18	30.4
SEM	*0.3*	*0.005*	*0.4*	*0.079*	*0.052*	*0.025*	*0.205*
ANOVA				***Probability***			
Na	<0.001	-	-	-	-	-	-
Na_L	<0.001	-	-	0.007	-	<0.001	0.007
Na_Q	0.002	<0.001	<0.001	-	-	<0.001	-
Na_C	-	<0.001	<0.001	-	-	-	<0.001
Salt	-	0.03	-	<0.001	-	-	0.08
Salt × Na	-	-	-	-	-	-	-
Salt × Na_L	-	0.01	<0.001	-	-	-	-
Salt × Na_Q	<0.001	<0.001	-	-	-	-	-
Salt × Na_c	-	<0.001	<0.001			<0.001	<0.001

Na_L, Na_Q & Na_C are linear, quadratic and cubic terms for Na, respectively

Experiment 2

Water was analyzed daily for its EC, TDS, pH, salinity, DO and temperature (Table 4.17). As the presence of ions in the drinking water may maneuver the DEB of ingesta thus their concentration in the water were analyzed. The results showed that the electrolytic concentration in the water were too low to affect the diet structure. Water pH values (6.96–7.24) were within the normal range of 6.0–8.5 (Socha *et al.*, 2002; Borges *et al.*, 2003a, 2003b). The analyzed values of TDS of water (960 to 1194 mg/kg) were in safe range of 1000 to 3000 mg/kg as reported by Chiba (2009).

Significant cubic responses of dK during 34 to 42 d on BWG ($P \leq 0.03$) and FG ($P \leq 0.05$) were noted in present study. When optimized, maximum BWG and improved FG were projected at 1.20% of dK (DEB of 288 mEq/kg), associated with the regression equation ($Y = 17102 - 48262\ \%\ dK + 45876\ \%\ dK^2 - 14294\ \%\ dK^3$; R^2 0.72) and ($Y = -85.15 + 253.0\ \%\ dK - 239.0\ \%\ dK^2 + 74.00\ \%\ dK^3$; R^2 0.80), respectively. This finding is in accordance with Johnson and Karunajeewa (1985) who reported DEB values of less than 180 and more than 300 mEq/kg could decrease BWG. However, Mongin (1981) and Borges *et al.* (2004a) recommended DEB at 250 and between 202 to 235 mEq/kg, the difference in results may be due to the manipulated electrolytes which was Na (Mongin, 1981) and Na+K (Borges *et al.*, 2004a). At optimized value of dK in present study for BWG and FG showed highest DWI which may be a reason of increased BWG as a result of more water deposition in muscles. It was noted that BWG (Table 4.18) and FG (Table 4.20) were not directly affected by the levels of dK when it was increased from 1.02% to 1.18 in present study. The BWG was improved by 20% at 1.18% (DEB of 283 mEq/kg) compared to 0.86% (DEB of 201) and reduced by 24.3% at 1.34% (DEB of 320 mEq/kg) compared to 1.18%. No effect of salt source was noted on FI, BWG or FG in present study. The present study showed that that birds' performance is more sensitive to the levels of dK than sources especially in last phases of production. It has been concluded in many studies (Muntwyler *et al.*, 1953; Leach *et al.*, 1959; Sullivan, 1963; Chavez *et al.*, 1979; Austic, 1983; Hooge and Cummings, 1995) that optimum dK supply is necessary for increased protein metabolism and similar response in BWG up to the level of 1.18% dK was also noted in present study. Oliveira *et al.* (2005) established relationship between dK and mass (protein) synthesis and reported that dK requirements are high in early phases of production cycle with observed muscle K contents of 110 mEq/kg at 12 d compared to that of 30 mEq/kg on hatching. It was also suggested that requirements of dK coincided with the growth pattern of the birds. However, the optimized value of dk i.e., 1.20% (DEB of 288 mEq/kg) for BWG during 34 to 42 d in the present study was quite high than that estimated previously (Oliveira *et al.*, 2005). If the same concept hold true, the difference in values may be due to reason that dK is playing role in protein metabolism, and mass

development is still continue to increase. The findings of the present study are also in agreement with Boulahsen *et al.* (1995) and Ahmad *et al.* (2008) who observed increasing BWG with increasing level of dK in drinking water when supplemented as KCl.

A significant response on FI ($P \leq 0.05$) was noted in present study where FI was increased by 7.4% in K_2SO_4 than that of K_2CO_3 diets in prestarter phase (1 to 10 d; Table 4.19). However weight gain and feed efficiency ($P > 0.05$) was not improved by this significant rise in FI. Neither dK levels nor sources significantly affected feed consumption in other phases of the experiment. The interaction between salt sources and dK levels showed no response on other performance parameters during the present experiment.

The interaction effects between salt sources and dk were found significant on daily water intake (DWI; Table 4.21) during 10 to 20 d ($P \leq 0.002$) and 1 to 42 d ($P \leq 0.04$) whereas DWI-to-FI ratio (Table 4.8) was observed significant ($P \leq 0.001$) during 10 to 20 d. When dK was optimized for DWI for 10 to 20 d and 1 to 42 d, minimum DWI was observed at 0.86% (DEB of 200 mEq/kg; K_2SO_4) or 1.34% (DEB of 320 mEq/kg; K_2CO_3). This showed that birds can retain more water in body in K_2SO_4 supplemented diets which is also evident from reduced litter dampness at high levels of dK in such diets (Table 4.23). Previous studies (Gorman and Balnave, 1994; Hooge, 2003; Ahmad *et al.*, 2005) also highlighted the same issue and reported the palatability matter of K_2CO_3 when over-supplemented. However, the interaction effect mainly changed DWI and DWI-to-FI ratio only. The DWI was reduced by 6% during 34 to 42 d ($P \leq 0.04$) for K_2CO_3 as compared to those with K_2SO_4. It is also speculated that adding various salts in different diets regulate the body's osmotic balance and associate water consumption and excretion to sustain body fluids (Borges *et al.*, 2004).

Litter moisture (**LM**) was significantly ($P \leq 0.03$) affected by dK × source during 1 to 42 d (Table 4.23). Lowest percentage (i.e., 40.23%) of LM was optimized at highest dK level (DEB of 320 mEq/kg) when used with K_2SO_4 whereas it was found lowest (i.e., 39.93%) at 0.98% dK level (DEB of 232 mEq/kg) when used with K_2CO_3 (quadratic effect of dK levels × salt sources). It means birds can sustain higher amounts of K_2SO_4 which is absorbed readily (>80%) as evident from isotopic tracer studies in man (Bauer, 1976; Florin *et al.*, 1991). Poultry litter wetness could be affected by dietary nutrients (mainly electrolytes), health of gastrointestinal tract, age of the birds, regional production difference and various environmental factors (Patterson *et al.*, 1998). The high intracellular concentration of K^+ has a characteristic impact on thirst, and body fluids are passively distributed corresponding to osmotic activity which is due to osmotically active potassium (Edelman *et al.*, 1958). As a result, birds having K_2CO_3-supplemented diets will tend to consume greater amounts of water in order to satisfy the internal toxic conditions of body fluids created by the increased K_2CO_3 intake (Gorman and Balnave, 1994; Hooge, 2003; Ahmad *et al.*, 2005). Replacing K_2SO_4 with

K_2CO_3 results in reduced ($P \leq 0.02$; 42%) mortality during the 1 to 42 d (Table 4.24). Ahmad *et al.* (2005) also noted that birds having more water intake showed less mortality and improved survivability in heat-distressed broilers. In heat stress condition, increased water intake acts as heat sink from the body and thus lowering deep body temperature so reduced mortality was observed. It may be possible that increased sulfur contents in K_2SO_4 diets reduced the pH of the birds so that in high stress of production this caused comparatively high mortality. However, the levels of dK did not affect the mortality.

Significantly high blood pH and dressing weight (as percent of live-weight) was noted by increasing levels of dK from 0.86 to 1.34% (Table 4.25). Diets containing high levels of dK tended to create a condition of mild alkalosis that may be the reason behind poor BWG at the highest level of 1.34% dK (DEB of 320 mEq/kg) when compared to that of 1.18% (DEB of 283 mEq/kg). The results of the present study showed that acid-base balance of body fluids of the birds are maintained at maximum at 1.20% dK levels (DEB of 288 mEq/kg) to sustain higher productivity. Significant responses on carcass characteristics to increasing dK were noted in the present study (Table 4.25-4.26). The increased carcass percentage attributes with the increasing dK may be correlated to high water intake in such diets which in turn deposited in the muscles and responded in high dressing characteristics in the present study. Reduced blood pH ($P \leq 0.001$) and dressing weights ($P \leq 0.04$) were observed by shifting salt source from K_2CO_3 to K_2SO_4 (Table 4.25). Replacing K_2CO_3 to K_2SO_4 may add more sulphate ions to the diets that lead to metabolic acidosis which resulted in reduced blood pH. However, during high growth rate these sulphates as sulfur amino acids become a part of body tissues (Shaw, 1989) that's why numerically high growth rate was seen in K_2SO_4 supplemented diets in present study (Table 4.18). Lehninger (1970) was of the view that enzymatic activity always requires optimum pH in the body physiological system for proper functioning of intermediary metabolism and cellular activity. When compared 1.34% dK levels of both salts (DEB of 320 mEq/kg), lower blood pH (i.e., 7.35) was noted in K_2SO_4 than K_2CO_3 supplemented diets which show acidic properties of sulphates (Ruiz-Lopez and Austic, 1993). Ahmad *et al.* (2008) also reported increased level of blood pH at higher level of DEB (350 mEq/kg). Increased gizzard and pancreatic weights may have increased digestive capacity and hence responded in improved BWG in present study. Optimized value of dK in case of K_2CO_3 supplemented salt for maximum pancreas and spleen weight was 1.20% (DEB of 288 mEq/kg) which in turn depicted in increased BWG. Blood glucose value was not affected either by dK, source or their interaction. As pancreas plays an important role in insulin secretion which maintains glucose level and controls Na/K-ATPase pump which in turn controls the high intracellular concentration of the potassium (Helderman *et al.*, 1983). However role of spleen in bird's body is related to erythrocyte production and immune

system (antibodies production and vaccine efficacy), and maximum weight at 1.20% dK (DEB of 288 mEq/kg) might have resulted in better immune system which leads to better growth efficiency as evident from higher BWG in present study. Reduction in bursa weight with increasing dK was more pronounced in K_2CO_3 as compared to K_2SO_4. This decrease may reduce immunity level of birds which is evident from higher mortality percentage in K_2CO_3 supplemented diets. Reduced abdominal fat in the present study (Table 4.27) was observed at lowest level of K_2CO_3 or highest level of K_2SO_4. It might be possible that more sulphates in the form of sulfur amino acids are available by using K_2SO_4 for tissue mass. By increasing dK may improve the basal metabolism so dietary nutrients become part of useful meat production rather as abdominal fat.

Serum Na ($P \leq 0.001$), Cl ($P \leq 0.03$), Mg ($P \leq 0.03$) and HCO_3 ($P \leq 0.001$) were affected by supplementation of dK (cubic term remain significant for amount and source of K) however quadratic increase in serum K ($P \leq 0.02$) was observed with increasing dK (Table 4.28). This significant increase in serum K caused more water intake and ultimately increased litter dampness as evident from current study. Mushtaq *et al.* (2005) suggested to maintain a balance between cations and anions in order to keep the acid base homeostasis. This balance could be maintained by providing proper DEB so that mild alkalosis caused by increased supplementation of dK could be compensated by dietary supplementation of anions. Increased serum anionic concentration might compensate increased cations in the blood. Serum Na and K are directly related to each other in blood and leads towards osmoregulation of body fluids (Mushtaq *et al.*, 2005). Linear effect of dK was found on serum Ca ($P \leq 0.01$; source and amount interaction) and P ($P \leq 0.03$; amount). Effect of dK is different on different serum mineral concentrations, so considering only the individual electrolyte is not sufficient to understand the whole story of DEB.

From the present study, it can be concluded that dK requirements for optimum performance are high when studied independently in modern strains of broilers reared for maximum performance in thermo-neutral zone. As no other ions were changed in this study, the response to dK may actually be response to DEB and a level of 1.20% K (288 mEq/kg) responded in the last phase of life-cycle and optimum performance of the birds was noted. As the information on dK requirements for optimum growth in thermo-neutral zone is scarce, further studies are recommended to investigate if there is some interaction between different cations at similar DEB or if all the cations respond similarly at same DEB. The results of the present study were variable for different phases, and level of 1.20% dK was found to affect on growth parametres in finisher phase while interaction effects was found effective for water intake and litter moisture. It is also suggested to verify the requirements by changing other electrolytes (Na and Cl) with other salt sources at constant DEB in low nutrient density diets.

To be very concise, high levels of dK remained important for last stages of broiler production. K_2CO_3 increased survivability and dressing responses however both, level and salts play an important role for water intake, litter condition and body characteristics.

Table 4.15. Ingredient composition of the basal diets for different phases of birds fed 4 levels of potassium with two sources of potassium salts[1]

Ingredients (%)	Pre-starter (1 – 10 d)	Starter (11 – 20 d)	Grower (21 – 33 d)	Finisher (34 – 42 d)
Corn	–	25.00	11.65	40.00
Broken rice	56.00	35.00	52.50	22.00
Soybean meal	14.80	13.60	21.90	14.60
Canola Meal	20.60	17.50	6.90	13.00
Fish meal	2.00	2.25	–	2.50
Oil	2.00	2.00	3.00	4.30
L-Lysine HCl	0.28	0.28	0.20	0.18
L-Lysine sulphate	0.28	0.28	–	–
DL-methionine	0.26	0.26	0.23	0.18
L-threonine	0.32	0.32	0.24	0.16
MCP	1.42	1.38	1.20	1.12
Limestone	1.27	1.27	1.35	1.18
NaCl	0.28	0.28	0.30	0.32
Premix[2]	0.50	0.50	0.50	0.50

[1]All the diets were supplemented with 4 levels of either K_2CO_3 (0.14, 0.28, 0.42 or 0.56%) or K_2SO_4 (0.18, 0.36, 0.54 or 0.72%) to make final K concentrations of 0.86, 1.02, 1.18 or 1.34%, respectively. These K values corresponded to DEB values of 200, 240, 280 and 320 mEq/kg, respectively.
The basal diet has 0.70% K in it corresponded to DEB of 160 mEq/kg.
[2]Provides per kg of finished diet: vitamin A, 12 mg; vitamin D_3, 7 mg; vitamin E, 100 mg; vitamin K_3 (50% as MNB), 3 mg; vitamin B_1 (98%), 3 mg; vitamin B_2 (800,000 mg), 12 mg; vitamin B_3 (niacin; 99%), 600 mg; vitamin B_6 (98%), 4 mg; vitamin B_9 (folic acid; 95%), 2 mg; vitamin B_{12} (0.10%), 20 mg; Biotin (0.10%), 5 mg; Ca-Pentothenate (98%), 12 mg; cholin (70% as choline chloride), 1 g; MnO (60%), 169 mg; $FeSO_4$ (21%), 200 mg; $ZnSO_4$ (36%), 150 mg; $CuSO_4$ (25%), 40 mg; Se (sodium selenite 0.40%), 100 mg; KI (68%), 2 mg; Salinomycin, 60 mg; Zinc bacitracin (as Albac 10%), 50 mg.

Table 4.16. Nutrient composition of the basal diets for different phases of birds fed 4 levels of potassium with two sources of potassium salts[1]

Nutrients (%)	Pre-starter (1 – 10 d)	Starter (11 – 20 d)	Grower (21 – 33 d)	Finisher (34 – 42 d)
ME (kcal/kg)	2,901	2,974	3,082	3,147
Crude Protein	21.01	19.57	19.00	17.89
Crude fiber	5.23	4.70	4.13	4.17
Calcium	1.00	0.94	0.90	0.88
Available Phos.	0.45	0.44	0.42	0.40
Sodium	0.15	0.15	0.15	0.15
Potassium	0.70	0.86	0.70	0.70
Chloride	0.30	0.30	0.30	0.30
DEB (mEq/kg)	160	160	160	160
Dig Lys	1.22	1.05	0.97	0.90
Dig Met/ Dig Lys	0.54	0.46	0.44	0.42
Dig Met + Dig Cys/Dig Lys	0.81	0.72	0.72	0.65
Dig Thr/Dig Lys	0.83	0. 70	0.66	0.61
Dig Try/Dig Lys	0.17	0.15	0.18	0.13
Dig Arg/Dig Lys	1.23	1.11	1.12	1.04

[1] K_2SO_4 and K_2CO_3

Table 4.17. Drinking water properties during the experimental period

Phase	Item[1]	Salinity (g/kg)	TDS[2] (mg/kg)	EC[3] (millisemen/cm)	Temperature (°C)	pH	DO[4] (mg/L)
Phase 1	Max	0.6	1184	1.39	33.2	7.27	4.65
	Min	0.4	1161	1.27	29.8	6.96	2.91
	Average	0.5	1171	1.32	31.7	7.11	3.84
Phase 2	Max	0.5	1011	1.41	29.5	7.24	4.50
	Min	0.3	960	1.06	26.0	6.99	3.91
	Average	0.4	984	1.22	27.5	7.13	4.22
Phase 3	Max	0.6	1187	1.24	26.0	7.24	4.59
	Min	0.5	1008	1.08	25.0	6.98	3.49
	Average	0.5	1044	1.15	25.4	7.15	4.06
Phase 4	Max	0.5	1194	1.21	25.0	7.18	4.45
	Min	0.4	1010	1.07	24.4	7.01	3.29
	Average	0.4	1111	1.11	24.7	7.12	3.98

[1]Max, Min and Average values were taken twice daily so one value is equal to days in phase × 2
[2]TDS - Total Dissolved Solids; [3]EC - Electrical Conductivity; [4]DO - Dissolved Oxygen

Table 4.18. Effect of dietary potassium and potassium salts on body weight gain of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			grams		
Basal (0.70%)	201	343	707	401	1,652
SEM	*2.9*	*8.2*	*39.5*	*29.6*	*67.4*
Dietary K (%)					
0.86	196	333	743	434	1,706
1.02	201	332	730	434	1,698
1.18	196	329	740	544	1,809
1.34	191	330	716	412	1,648
SEM	*2.8*	*8.5*	*27.1*	*33.0*	*50.0*
Salts					
K_2CO_3	194	336	716	448	1,693
K_2SO_4	198	326	749	464	1,737
SEM	*2.0*	*6.0*	*19.2*	*23.3*	*35.4*
K × Salts					
0.86 × K_2CO_3	193	335	718	419	1,664
1.02 × K_2CO_3	200	333	695	402	1,630
1.18 × K_2CO_3	194	340	738	540	1,812
1.34 × K_2CO_3	190	334	713	432	1,668
0.86 × K_2SO_4	200	331	768	450	1,748
1.02 × K_2SO_4	202	331	766	467	1,767
1.18 × K_2SO_4	198	317	743	548	1,807
1.34 × K_2SO_4	191	326	718	392	1,628
SEM	*4.0*	*12.1*	*38.3*	*46.7*	*70.7*
ANOVA			***Probability***		
K	-	-	-	-	-
K_L	-	-	-	-	-
K_Q	-	-	-	-	-
K_C	-	-	-	0.03	-
Salt	-	-	-	-	-
Salt × K_L	-	-	-	-	-
Salt × K_Q	-	-	-	-	-
Salt × K_C	-	-	-	-	-

K_L, K_Q & K_C are linear, quadratic and cubic terms for K, respectively.

Table 4.19. Effect of dietary potassium and potassium salts on feed intake of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			grams		
Basal (0.70%)	274	328	1,613	1,025	3,241
SEM	*10.5*	*6.1*	*34.2*	*65.0*	*76.3*
Dietary K (%)					
0.86	287	322	1,640	1,096	3,344
1.02	286	341	1,740	1,114	3,480
1.18	283	328	1,700	1,117	3,427
1.34	285	332	1,663	1,041	3,322
SEM	*10.5*	*10.6*	*56.4*	*55.3*	*89.4*
Salts					
K_2CO_3	274	337	1,638	1,070	3,319
K_2SO_4	296	325	1,734	1,114	3,468
SEM	*7.4*	*7.5*	*39.9*	*39.1*	*63.2*
K × Salts					
0.86 × K_2CO_3	279	340	1,539	1,040	3,199
1.02 × K_2CO_3	272	340	1,671	1,065	3,347
1.18 × K_2CO_3	282	333	1,743	1,127	3,484
1.34 × K_2CO_3	266	334	1,599	1,047	3,245
0.86 × K_2SO_4	294	303	1,741	1,152	3,490
1.02 × K_2SO_4	301	341	1,808	1,163	3,613
1.18 × K_2SO_4	285	323	1,656	1,107	3,371
1.34 × K_2SO_4	305	331	1,727	1,035	3,398
SEM	*14.8*	*14.9*	*79.8*	*78.2*	*126.5*
ANOVA			***Probability***		
K	-	-	-	-	-
K_L	-	-	-	-	-
K_Q	-	-	-	-	-
K_C	-	-	-	-	-
Salt	0.05	-	-	-	-
Salt × K_L	-	-	-	-	-
Salt × K_Q	-	-	-	-	-
Salt × K_C	-	-	-	-	-

K_L, K_Q & K_C are linear, quadratic and cubic terms for K, respectively.

Table 4.20. Effect of dietary potassium and potassium salts on feed:gain of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			grams/grams		
Basal (0.70%)	1.37	0.96	2.30	2.87	1.87
SEM	*0.05*	*0.04*	*0.14*	*0.22*	*0.09*
Dietary K (%)					
0.86	1.47	0.97	2.24	2.72	1.85
1.02	1.43	1.04	2.40	2.77	1.91
1.18	1.45	1.00	2.40	2.18	1.76
1.34	1.51	1.00	2.36	2.77	1.91
SEM	*0.062*	*0.032*	*0.139*	*0.197*	*0.070*
Salts					
K_2CO_3	1.42	1.00	2.34	2.58	1.84
K_2SO_4	1.51	1.00	2.36	2.64	1.88
SEM	*0.044*	*0.023*	*0.099*	*0.139*	*0.049*
K × Salts					
0.86 × K_2CO_3	1.46	1.02	2.17	2.76	1.85
1.02 × K_2CO_3	1.37	1.02	2.42	2.79	1.90
1.18 × K_2CO_3	1.46	0.98	2.51	2.23	1.79
1.34 × K_2CO_3	1.40	1.00	2.25	2.54	1.80
0.86 × K_2SO_4	1.48	0.92	2.31	2.68	1.85
1.02 × K_2SO_4	1.49	1.05	2.37	2.75	1.92
1.18 × K_2SO_4	1.44	1.02	2.29	2.13	1.72
1.34 × K_2SO_4	1.63	1.01	2.47	2.99	2.02
SEM	*0.088*	*0.046*	*0.197*	*0.278*	*0.099*
ANOVA			***Probability***		
K	-	-	-	-	-
K_L	-	-	-	-	-
K_Q	-	-	-	-	-
K_C	-	-	-	0.05	-
Salt	-	-	-	-	-
Salt × K_L	-	-	-	-	-
Salt × K_Q	-	-	-	-	-
Salt × K_C	-	-	-	-	-

K_L, K_Q & K_C are linear, quadratic and cubic terms for K, respectively.

Table 4.21. Effect of dietary potassium and potassium salts on water intake of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			mL		
Basal (0.70%)	926	1,160	3,186	2,590	7,863
SEM	*39.0*	*72.4*	*74.5*	*43.5*	*139.0*
Dietary K (%)					
0.86	927	1,180	3,439	2,793	8,339
1.02	950	1,217	3,482	2,904	8,552
1.18	947	1,241	3,610	3,111	8,909
1.34	979	1,204	3,589	2,978	8,750
SEM	*45.4*	*49.7*	*78.1*	*140.1*	*231.4*
Salts					
K_2CO_3	918	1,203	3,455	2,798	8,374
K_2SO_4	983	1,218	3,605	3,095	8,901
SEM	*32.1*	*35.1*	*55.2*	*99.0*	*163.6*
K × Salts					
0.86 × K_2CO_3	876	1,081	3,277	2,527	7,761
1.02 × K_2CO_3	959	1,174	3,407	2,745	8,284
1.18 × K_2CO_3	912	1,197	3,526	2,860	8,495
1.34 × K_2CO_3	925	1,361	3,610	3,058	8,954
0.86 × K_2SO_4	977	1,280	3,601	3,059	8,917
1.02 × K_2SO_4	941	1,261	3,556	3,062	8,820
1.18 × K_2SO_4	982	1,284	3,694	3,362	9,322
1.34 × K_2SO_4	1,033	1,046	3,568	2,899	8,546
SEM	*64.2*	*70.3*	*110.4*	*198.1*	*327.2*
ANOVA			***Probability***		
K	-	-	-	-	-
K_L	-	-	-	-	-
K_Q	-	-	-	-	-
K_C	-	-	-	-	-
Salt	-	-	-	0.04	0.03
Salt × K_L	-	0.002	-	-	0.04
Salt × K_Q	-	-	-	-	-
Salt × K_C	-	-	-	-	-

K_L, K_Q & K_C are linear, quadratic and cubic terms for K, respectively.

Table 4.22. Effect of dietary potassium and potassium salts on water intake-to-feed intake ratio of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			mL/grams		
Basal (0.70%)	3.38	3.55	1.98	2.56	2.43
SEM	*0.05*	*0.28*	*0.04*	*0.16*	*0.01*
Dietary K (%)					
0.86	3.24	3.74	2.10	2.58	2.50
1.02	3.33	3.57	2.01	2.66	2.47
1.18	3.37	3.80	2.15	2.82	2.61
1.34	3.49	3.64	2.17	2.90	2.65
SEM	*0.197*	*0.172*	*0.076*	*0.185*	*0.094*
Salts					
K_2CO_3	3.38	3.59	2.13	2.65	2.54
K_2SO_4	3.34	3.78	2.09	2.83	2.58
SEM	*0.140*	*0.122*	*0.054*	*0.131*	*0.066*
K × Salts					
0.86 × K_2CO_3	3.16	3.24	2.14	2.50	2.45
1.02 × K_2CO_3	3.52	3.45	2.05	2.62	2.48
1.18 × K_2CO_3	3.27	3.60	2.07	2.57	2.45
1.34 × K_2CO_3	3.55	4.09	2.27	2.92	2.76
0.86 × K_2SO_4	3.33	4.23	2.07	2.67	2.55
1.02 × K_2SO_4	3.15	3.70	1.97	2.71	2.45
1.18 × K_2SO_4	3.46	4.00	2.23	3.08	2.77
1.34 × K_2SO_4	3.43	3.19	2.08	2.88	2.54
SEM	*0.279*	*0.242*	*0.108*	*0.262*	*0.133*
ANOVA			***Probability***		
K	-	-	-	-	-
K_L	-	-	-	-	-
K_Q	-	-	-	-	-
K_C	-	-	-	-	
Salt	-	-	-	-	
Salt × K_L	-	0.001	-	-	
Salt × K_Q	-	-	-	-	
Salt × K_C	-	-	-	-	

K_L, K_Q & K_C are linear, quadratic and cubic terms for K, respectively.

Table 4.23. Effect of dietary potassium and potassium salts on litter moisture of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			%		
Basal (0.70%)	24.2	71.3	29.6	42.1	41.8
SEM	*3.98*	*1.01*	*4.29*	*1.97*	*1.28*
Dietary K (%)					
0.86	21.6	73.1	30.1	41.7	41.6
1.02	22.4	71.1	34.2	42.0	42.4
1.18	25.2	72.5	32.3	41.6	43.0
1.34	24.6	73.8	34.8	44.4	44.4
SEM	*2.49*	*1.69*	*3.09*	*2.80*	*1.64*
Salts					
K_2CO_3	24.6	71.2	34.9	42.8	43.4
K_2SO_4	22.3	74.0	30.9	42.0	42.3
SEM	*1.76*	*1.19*	*2.19*	*1.98*	*1.60*
K × Salts					
0.86 × K_2CO_3	25.2	71.4	26.5	44.2	41.8
1.02 × K_2CO_3	19.0	68.8	34.3	36.4	39.6
1.18 × K_2CO_3	25.8	68.3	37.3	41.4	43.2
1.34 × K_2CO_3	28.3	76.3	41.7	49.2	48.9
0.86 × K_2SO_4	18.0	74.7	33.7	39.1	41.4
1.02 × K_2SO_4	25.8	73.5	34.2	47.6	45.3
1.18 × K_2SO_4	24.6	76.7	27.9	41.8	42.7
1.34 × K_2SO_4	20.8	71.3	28.0	39.6	39.9
SEM	*3.52*	*2.38*	*4.37*	*3.97*	*2.32*
ANOVA			***Probability***		
K	-	-	-	-	-
K_L	-	-	-	-	-
K_Q	-	-	-	-	-
K_C	-	-	-	-	-
Salt	-	-	-	-	-
Salt × K_L	-	-	0.02	-	0.04
Salt × K_Q	0.05	0.04	-	0.03	0.03
Salt × K_C	-	-	-	-	-

K_L, K_Q & K_C are linear, quadratic and cubic terms for K, respectively.

Table 4.24. Effect of dietary potassium and potassium salts on mortality of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			%		
Basal (0.70%)	0.0	0.5	0.0	2.7	3.3
SEM	*0.00*	*0.54*	*0.00*	*1.63*	*1.40*
Dietary K (%)					
0.86	0.8	0.3	1.1	2.2	4.4
1.02	1.4	1.4	0.3	2.2	5.2
1.18	0.8	0.3	1.4	1.6	4.1
1.34	1.6	0.0	0.8	1.6	4.1
SEM	*0.82*	*0.36*	*0.50*	*0.84*	*1.18*
Salts					
K_2CO_3	0.7	0.3	0.8	1.5	3.3
K_2SO_4	0.6	0.7	1.0	2.3	5.6
SEM	*0.58*	*0.25*	*0.36*	*0.59*	*0.84*
K × Salts					
0.86 × K_2CO_3	0.0	0.0	1.1	2.7	3.8
1.02 × K_2CO_3	1.6	1.1	0.0	1.1	3.8
1.18 × K_2CO_3	1.1	0.0	1.6	1.6	4.4
1.34 × K_2CO_3	0.0	0.0	0.5	0.5	1.1
0.86 × K_2SO_4	1.6	0.5	1.1	1.6	4.9
1.02 × K_2SO_4	1.1	1.6	0.5	3.3	6.5
1.18 × K_2SO_4	0.5	0.5	1.1	1.6	3.8
1.34 × K_2SO_4	3.3	0.0	1.1	2.7	7.1
SEM	*1.16*	*0.51*	*0.71*	*1.18*	*1.67*
ANOVA			***Probability***		
K	-	-	-	-	-
K_L	-	-	-	-	-
K_Q	-	-	-	-	-
K_C	-	-	-	-	-
Salt	-	-	-	-	0.02
Salt × K_L	-	-	-	-	-
Salt × K_Q	-	-	-	-	-
Salt × K_C	-	-	-	-	-

K_L, K_Q & K_C are linear, quadratic and cubic terms for K, respectively.

Table 4.25. Effect of dietary potassium and potassium salts on blood and carcass responses of broilers at the end of the experiment

Item	Blood pH	Blood glucose[0]	Dressing weight[1]	Breast weight[2]	Thigh weight[2]	Intestinal weight[3]	Intestinal length[4]
Basal (0.70%)	7.25	214.0	55.92	41.71	45.34	49.6	2.0
SEM	*0.041*	*15.70*	*1.591*	*1.022*	*1.553*	*4.39*	*0.10*
Dietary K (%)							
0.86	7.27	210.6	57.36	38.57	44.38	46.7	1.7
1.02	7.31	190.5	60.23	38.86	41.34	55.6	1.7
1.18	7.34	202.1	58.63	37.21	42.67	54.8	1.8
1.34	7.36	218.8	64.35	36.94	40.11	51.2	1.7
SEM	*0.004*	*12.67*	*1.428*	*1.324*	*1.312*	*3.31*	*0.07*
Salts							
K_2CO_3	7.33	202.2	61.61	36.70	41.26	49.5	1.7
K_2SO_4	7.31	208.8	58.67	39.08	42.99	54.6	1.7
SEM	*0.003*	*8.96*	*1.010*	*0.936*	*0.927*	*2.34*	*0.05*
K × Salts							
0.86 × K_2CO_3	7.28	211.9	61.04	36.25	42.75	45.6	1.9
1.02 × K_2CO_3	7.32	178.5	60.77	37.41	40.44	55.0	1.7
1.18 × K_2CO_3	7.35	215.8	58.53	37.93	42.64	48.8	1.7
1.34 × K_2CO_3	7.38	202.5	66.12	35.22	39.22	48.6	1.7
0.86 × K_2SO_4	7.26	209.2	53.68	40.89	46.02	47.7	1.6
1.02 × K_2SO_4	7.30	202.5	59.69	40.32	42.24	56.2	1.7
1.18 × K_2SO_4	7.33	188.5	58.73	36.48	42.70	60.8	1.8
1.34 × K_2SO_4	7.35	235.0	62.58	38.66	41.00	54.0	1.8
SEM	*0.005*	*17.91*	*2.019*	*1.872*	*1.855*	*4.68*	*0.10*
ANOVA		———	***Probability***	———			
K	≤0.001	-	0.004	-	-	-	-
K_L	0.01	-	-	-	-	-	-
K_Q	0.004	-	-	-	-	-	-
K_C	-	-	-	-	-	-	-
Salt	≤0.001	-	0.04	-	-	-	-
Salt × K_L	≤0.001	-	-	-	-	-	0.03
Salt × K_Q	-	-	-	-	-	-	-
Salt × K_C	-	-	-	-	-	-	-

[0] mg/dL; [1]% of live weigh (without visceral organs)
[2]%t of dressed weight; [3]measured in gram, and [4]centimeters, respectively
K_L, K_Q & K_C are linear, quadratic and cubic terms for K, respectively.

Table 4.26. Effect of dietary potassium and potassium salts on body organs weights of broilers at the end of the experiment

Item	Gizzard	Proventriculus	Heart	Liver	Kidney
		% of dressed weight			
Basal (0.70%)	2.20	0.46	0.56	3.30	0.53
SEM	*0.242*	*0.040*	*0.025*	*0.231*	*0.040*
Dietary K (%)					
0.86	2.23	0.53	0.51	3.10	0.59
1.02	1.84	0.41	0.51	3.14	0.53
1.18	1.95	0.48	0.53	2.99	0.63
1.34	1.76	0.53	0.51	2.71	0.77
SEM	*0.116*	*0.019*	*0.032*	*0.121*	*0.092*
Salts					
K_2CO_3	1.91	0.48	0.52	3.07	0.62
K_2SO_4	1.98	0.50	0.52	2.90	0.64
SEM	*0.082*	*0.014*	*0.023*	*0.086*	*0.065*
K × Salts					
0.86 × K_2CO_3	2.08	0.47	0.51	2.93	0.53
1.02 × K_2CO_3	1.93	0.44	0.52	3.37	0.48
1.18 × K_2CO_3	1.90	0.51	0.52	3.20	0.70
1.34 × K_2CO_3	1.75	0.49	0.53	2.76	0.77
0.86 × K_2SO_4	2.38	0.60	0.52	3.27	0.66
1.02 × K_2SO_4	1.75	0.39	0.51	2.91	0.58
1.18 × K_2SO_4	2.01	0.45	0.54	2.77	0.55
1.34 × K_2SO_4	1.78	0.57	0.49	2.66	0.77
SEM	*0.164*	*0.027*	*0.045*	*0.171*	*0.129*
ANOVA		***Probability***			
K	0.03	≤0.001	-	-	-
K_L	0.02	-	-	0.02	-
K_Q	-	≤0.001	-	-	-
K_C	-	0.03	-	-	-
Salt	-	-	-	-	-
Salt × K_L	-	-	-	-	-
Salt × K_Q	-	≤0.001	-	0.02	-
Salt × K_C	-	-	-	-	-

K_L, K_Q & K_C are linear, quadratic and cubic terms for K, respectively.

Table 4.27. Effect of dietary potassium and potassium salts on body organs weights of broilers at the end of the experiment

Item	Spleen	Pancreas	Bursa	Gall bladder	Lungs	Abdominal fat	Shank length[1]
			% of dressed weight				
Basal (0.70%)	0.08	0.23	0.13	0.08	0.64	2.91	7.58
SEM	*0.012*	*0.026*	*0.014*	*0.012*	*0.047*	*0.232*	*0.14*
Dietary K (%)							
0.86	0.10	0.26	0.12	0.09	0.65	3.28	7.55
1.02	0.06	0.31	0.08	0.09	0.59	3.20	7.04
1.18	0.11	0.28	0.08	0.09	0.66	3.38	7.36
1.34	0.11	0.26	0.07	0.08	0.59	2.81	7.42
SEM	*0.007*	*0.020*	*0.010*	*0.007*	*0.039*	*0.231*	*0.136*
Salts							
K_2CO_3	0.08	0.23	0.08	0.09	0.60	3.42	7.51
K_2SO_4	0.11	0.32	0.09	0.08	0.64	2.91	7.18
SEM	*0.005*	*0.014*	*0.007*	*0.005*	*0.028*	*0.163*	*0.096*
K × Salts							
0.86 × K_2CO_3	0.07	0.24	0.13	0.13	0.63	3.11	7.54
1.02 × K_2CO_3	0.05	0.22	0.08	0.10	0.61	3.46	7.26
1.18 × K_2CO_3	0.11	0.26	0.07	0.08	0.60	3.79	7.53
1.34 × K_2CO_3	0.09	0.21	0.05	0.06	0.58	3.34	7.71
0.86 × K_2SO_4	0.13	0.28	0.11	0.05	0.66	3.45	7.56
1.02 × K_2SO_4	0.07	0.40	0.08	0.07	0.57	2.94	6.82
1.18 × K_2SO_4	0.11	0.31	0.10	0.10	0.73	2.96	7.19
1.34 × K_2SO_4	0.13	0.31	0.09	0.09	0.59	2.29	7.13
SEM	*0.011*	*0.028*	*0.015*	*0.010*	*0.055*	*0.327*	*0.192*
ANOVA				***Probability***			
K	≤0.001	-	0.02	-	-	-	-
K_L	0.02	-	0.007	-	-	-	0.04
K_Q	0.006	-	-	-	-	-	-
K_C	≤0.001	-	-	-	-	-	-
Salt	≤0.001	-	≤0.001	0.04	-	0.03	0.02
Salt × K_L	-	≤0.001	0.04	≤0.001	-	0.04	-
Salt × K_Q	0.03	-	-	-	-	-	-
Salt × K_C	-	0.01	-	-	-	-	-

[1] measured in centimeter (cm)

K_L, K_Q & K_C are linear, quadratic and cubic terms for K, respectively.

Table 4.28. Effect of dietary potassium and potassium salts on serum mineral chemistry of broilers at the end of the experiment

Item	Na	K	Cl	Ca	P	Mg	HCO_3
		mmol/L					
Basal (0.70%)	129	2.94	101	2.78	3.03	1.20	29.7
SEM	*0.2*	*0.002*	*0.4*	*0.140*	*0.180*	*0.055*	*0.09*
Dietary K (%)							
0.86	131	2.97	102	2.41	3.06	1.10	30.4
1.02	132	3.01	102	2.46	3.19	1.12	31.0
1.18	133	3.06	102	2.33	2.97	1.14	30.5
1.34	137	3.12	101	2.04	2.88	1.15	32.4
SEM	*0.2*	*0.002*	*0.2*	*0.098*	*0.107*	*0.051*	*0.08*
Salts							
K_2CO_3	134	3.06	102	2.55	2.89	1.10	31.4
K_2SO_4	132	3.01	101	2.07	3.16	1.15	30.8
SEM	*0.1*	*0.002*	*0.2*	*0.070*	*0.075*	*0.036*	*0.05*
K × Salts							
0.86 × K_2CO_3	132	2.99	102	2.80	3.03	1.01	31.1
1.02 × K_2CO_3	132	3.03	102	2.84	3.09	1.20	31.4
1.18 × K_2CO_3	135	3.08	103	2.51	2.80	1.06	29.8
1.34 × K_2CO_3	138	3.16	100	2.03	2.64	1.11	33.2
0.86 × K_2SO_4	129	2.95	101	2.02	3.08	1.18	29.8
1.02 × K_2SO_4	132	2.99	101	2.07	3.29	1.04	30.5
1.18 × K_2SO_4	132	3.03	101	2.14	3.14	1.21	31.2
1.34 × K_2SO_4	135	3.09	101	2.05	3.13	1.18	31.7
SEM	*0.3*	*0.003*	*0.3*	*0.139*	*0.151*	*0.073*	*0.109*
ANOVA		***Probability***					
K	<0.001	<0.001	-	-	-	-	<0.001
K_L	<0.001	<0.001	-	0.006	-	-	<0.001
K_Q	<0.001	<0.001	0.01	-	-	-	<0.001
K_C	0.03	<0.04	0.04	-	-	-	0.04
Salt	<0.001	<0.001	0.007	<0.001	0.01	-	<0.001
Salt × K_L	-	<0.001	0.01	0.002	-	-	<0.001
Salt × K_Q	0.009	<0.02	-	-	-	-	<0.001
Salt × K_C	<0.001	-	0.03	-	-	0.03	<0.001

K_L, K_Q & K_C are linear, quadratic and cubic terms for K, respectively

Experiment 3

Water was analyzed daily for its EC, TDS, pH, salinity, DO and temperature (Table 4.31), keeping in view the concept that the concentration of various minerals (cations plus anions) and above said parametres in the drinking water could maneuver the DEB of ingesta thus their values in the water were evaluated (Teeter *et al.*, 1985; Borges *et al.*, 2003a;b). Electrolyte concentration was found negligible. Water pH values (7.17–7.49) were found within the range (6.0–8.5) considered as optimal for broiler performance (Socha *et al.*, 2002; Borges *et al.*, 2003a,b). Previous report of Grizzle *et al.*, (1996) also showed retarded growth up to pH level of 6.3. Water TDS level ranges from 1000-3000 ppm was considered satisfactory for broilers (Chiba, 2009) and analyzed values (1010-1234) were found within safe zone.

Significant cubic responses in BWG were observed during d 21-33 ($P \leq 0.04$) and d 34-42 ($P \leq 0.009$; Table 4.32). When optimized, maximum BWG i.e. 702 and 579 were observed at 0.60 (DEB = 197 mEq/kg) and 0.42% (DEB = 248 mEq/kg) dCl, associated with the regression equation for d 21-33 (BWG = 924.8 - 1770 Cl + 3983 Cl^2 - 2755 Cl^3) and d 34-42 (BWG = - 2106 + 16130 Cl - 31222 Cl^2 + 19138 Cl^3), respectively. Not major numerical, however statistically significant, difference exist in BWG (677 to 702 g) during grower phase. Effects of amount and source were found significant for BWG during d 1-10 (quadratic term for dCl levels and salt sources). When dCl was optimized for BWG for d 1-10, highest BWG was observed at 0.73% dCl (DEB = 160 mEq/kg) in cases of both salts. It means with the increasing age, requirements for dCl decreased and DEB increased as dCl was dropped from 0.73% (DEB = 160 mEq/kg; d 1-10) to 0.60% (DEB = 197 mEq/kg; d 21-33) and then to 0.42% (DEB = 248 mEq/kg; d 34-42). This drop was in accordance with the recommendations of NRC (1994) but numerical value is quite high in our experiment. This might be due to the fact that the levels recommended by NRC (1994) were determined using Vantress × White Plymouth Rock (Oliveira *et al.* 2005) which had lower potential for mass and skeletal development than the strains of today's broiler. Murakami *et al.* (1997) suggested that requirements of Na and Cl were increasing in recent research works. He also observed that higher levels of these minerals didn't appear to counter the growth effects in terms of body weight and feed efficiency. However, Borges *et al.* (2004b) found maximum BWG at 202 mEq/kg. This difference in result might be owing to the changing elements (Na and Cl) and the extended period of rearing (d 21-42). Cubic ($P \leq 0.006$) responses were observed for FI for the period of d 1-42 (Table 4.33), however, maximum intake was achieved at 0.60% dCl (DEB = 197 mEq/kg) when optimized according to the regression equation (FI = 3629 - 1485 Cl + 5128 Cl^2 - 4297 Cl^3). Murakami *et al.* (1997) suggested a direct effect of dCl on appetite and observed better FI by increasing Na and Cl. Higher values of dCl were observed for each phase than previously recommended. However it is anticipated that the negative effects of

higher dCl on growth parametres in present study may be compensated by higher levels of both Na and K. This cation-anion ratio has a strong impact on acid base balance in broilers and blood pH of 7.28 (Hurwitz *et al.* 1973) is found to be ideal for maximum growth efficiency which is also evident from present study. Although a lot of research has been conducted on individual electrolyte however decisive requirements are not fully dogged out yet. This might be due to reason that numerous different strategies had been applied in dietary plan and environmental conditions. However, birds consume more of 0.63% (DEB = 188 mEq/kg) and 0.61% (DEB = 194 mEq/kg) dCl diets when $CaCl_2$ was used in comparison to 0.38 (DEB = 259 mEq/kg) and 0.42% (DEB = 248 mEq/kg) dCl diets when NH_4Cl was used during d 21-33 ($P \leq 0.001$) and d 34-42 ($P \leq 0.04$), respectively (cubic terms for amount and source of dCl). It was found that intake of dCl is directly linked with supplemental salts in later part of life. It is also clear that birds supplemented with $CaCl_2$ diets convert feed into weight with great efficiency than in NH_4Cl supplemented diets in later part of life ($P>0.05$). However the concept of DEB (mEq/kg) might tackle phenomenon of intake in birds as with increasing DEB (mEq/kg), intake was also increased. This increase might be to adjust blood electrolyte and acid base balance. Levels, sources nor interaction effects were observed in prestarter and starter phases. Except for FG during prestarter phase ($P \leq 0.03$), there was no difference in response of chicks by replacing $CaCl_2$ with NH_4Cl (Table 4.34). However, marked effects (cubic responses) for amount of dCl were seen for FG for finisher ($P \leq 0.004$) and overall ($P \leq 0.007$) periods. Upon optimization, improved FG were observed at 0.38% (DEB = 259 mEq/kg) for finisher (2.15) and overall (1.92) periods, respectively. It means amount of dCl is not directly responsible for change in production efficiency of chicks. It has been recommended in previous studies that FG is strongly affected by dCl (Hurwitz *et al.* 1973; Nam and McGinnis, 1981; Murakami *et al.* 1997). It was evident that sources play pivotal role in production performance in initial days of life while after that birds become more sensitive to the amount of dCl. No other study has yet been carried out to evaluate dCl requirements during phase feeding program so this finding also leads towards a dire need for establishing biochemical processes involved and understanding the individual effect of each electrolyte that was not estimated previously in different phases of broiler life.

Significant response was observed in DWI during prestarter phase (quadratic term for amount and source of dCl; $P \leq 0.003$; Table 4.35). When dCl amount was increased from 0.31 to 0.59% (DEB = 280-200 mEq/kg), a 13% decrease in DWI was observed while same percentage was increased from 0.59 to 0.73% (DEB = 200-160 mEq/kg) in case of $CaCl_2$ salt. However, 14% decrease in DWI was observed when amount of dCl was increased from 0.31 to 0.45% (DEB = 280-240 mEq/kg) while 40% increase was observed when dCl was increased from 0.45 to 0.73% (DEB = 240-160 mEq/kg) in case of NH_4Cl supplemental salt.

This increase in water intake did not result to increase body weight so excess water is excreted to enhance litter dampness (quadratic term for amount and source of dCl) during d 1-10. No effect of dCl or salt source on DWI was observed in previous reports (Smith and Teeter, 1993; Mushtaq *et al.* 2005, 2007). The difference in results may be due to partitioning of experimental period into short phases and providing normal environmental conditions in the present study. Neither amount, neither source nor interaction responses on DWI were observed in rest of the phases. Ratio between DWI and FI (DWI:FI) was significantly affected by source and level interaction for d 1-10 ($P \leq 0.05$) and d 21-33 ($P \leq 0.05$; cubic term; Table 4.36). This ratio might be affected by the significant change in DWI and FI during these phases. Mushtaq *et al.* (2005, 2007) and Ahmad *et al.* (2005) observed no response of dCl on DWI:FI when comparing different levels, electrolytes or salts, respectively.

Litter moisture (LM) is quadratically affected by source, level and interaction effects for d 1-42 (Table 4.37). Upon optimization, least level of LM (26.93 and 32.52%) was observed at least level (i.e. 0.31%) of NH_4Cl and 0.35% dCl of $CaCl_2$, respectively ($P \leq 0.001$) for d 1-42. Highest level of water intake might show maximum litter moisture at highest level of dCl during 1-42d ($P > 0.05$). Linear increase in LM was observed by Mushtaq *et al.* (2005, 2007) by increasing dCl levels. Reason behind this linear increase might be due to the narrower range of dCl (0.30-0.50%) and also balancing this dCl with increasing Na level (0.20-0.30%) at constant DEB (250 mEq/kg). Murakami *et al.* (2001) observed no difference in LM with various levels of dCl and concluded that dCl should be balanced with Na in order to optimize kidney excretion (Freeman, 1983). It is anticipated that poultry litter wetness could be affected by dietary nutrients (mainly electrolytes), health of gastrointestinal tract (**GIT**), age of the birds, regional production difference and various environmental factors (Patterson *et al.*, 1998). Except that mortality in finisher phase (quadratic term of amount and source of dCl; $P \leq 0.03$), neither amount nor sources of dCl were found significant for mortality. Highest level of mortality, upon optimization, was observed in highest level of dCl (0.73% or DEB = 160 mEq/kg) in both salts during d 34-42. These results are in accordance with the findings of Ahmad *et al.* (2008) who observed high mortality in the diet having the lowest level of DEB (150 mEq/kg).

Amount and source of dCl was found effective for blood pH ($P \leq 0.05$), serum glucose ($P \leq 0.004$), DP ($P \leq 0.008$), intestinal weight ($P \leq 0.02$) and length ($P \leq 0.01$; Table 4.39). Blood pH decreased linearly with dCl (quadratic term for amount and source of dCl), however this gap is more extended in case of NH_4Cl (7.35-7.29) than $CaCl_2$ (7.29-7.27). Lower pH value in $CaCl_2$ supplemented diets might be indicator of reduced absorption of Ca in the distal part of intestine and more excretion as $CaCO_3$ as Cl is retained to reduce blood pH (Mongin, 1981). It means the role of dCl is more sensitive in case of $CaCl_2$ rather NH_4Cl supplemented diets in

order to maintain acid base balance. A 6% decrease in serum glucose was observed with dCl in $CaCl_2$ supplemented diets while in contrary to this, 14% increase in serum glucose was observed in NH_4Cl supplemented diets (linear term for amount and source of dCl). Ahmad *et al.* (2005) also found reduced blood glucose in $CaCl_2$ supplemented diets when compared with other ionic salts. Better glucose level overall depicts better health status. $CaCl_2$ supplemented diets prove to have negative effect on DP with increasing dCl while positive results were found in NH_4Cl supplemented diets (linear term for amount and source of dCl). Birds might be able to contain more water in muscles to increase DP as evident from higher water intake in NH_4Cl supplemented diets ($P>0.05$). Mushtaq *et al.* (2007) also observed reduced DP in birds of d 28-42 in the diets containing increasing 0.30-0.50% dCl. Quadratic responses of dCl were seen on intestinal length and weight by supplementing both salts. Breast and thigh meat yield was found higher when $CaCl_2$ diets were replaced with NH_4Cl. Ahmad *et al.* (2005) had also found higher breast meat yield in NH_4Cl supplemented group when comparing with other ionic salts.

Weight of proventriculus ($P\leq0.03$), heart ($P\leq0.005$), liver ($P\leq0.01$) and kidney ($P\leq0.03$) were affected by both, amount and salt of dCl (Table 4.40). However, quadratic and cubic responses were found significant in case of proventriculus and liver weights, respectively.

Cubic term effects were found in pancreas and gall bladder however linear interaction effects were found in abdominal fat (Table 4.41). Reduced abdominal fat was seen in $CaCl_2$ supplemented diets while abdominal fat was increased in NH_4Cl supplemented diets with increasing dCl. However, lowest abdominal fat was found in NH_4Cl supplemented diets by Ahmad *et al.* (2005), this difference in results may be due to fixation of DEB (250 mEq/kg) in the experiment of Ahmad *et al.* (2005). It means energy is better utilized in $CaCl_2$ diets which is evident because of the dressing percentage ($P\leq0.02$) and body weight gain ($P>0.05$) in $CaCl_2$ diets. Significant salt replacing effects ($CaCl_2$ diets with NH_4Cl) were observed in shank length that leads towards more skeletal development and ultimately shows potential of birds to sustain more mass production (Table 4.41).

Serum Cl was affected by supplementation of chloride salts ($P<0.001$) and dCl ($P<0.03$; quadratic term remained significant for amount and source of K). Mushtaq *et al.* (2005) suggested to maintain a balance between cations and anions in order to keep the acid base homeostasis. This balance could be maintained by providing proper DEB so that mild alkalosis caused by increased supplementation of dK could be compensated by dietary supplementation of anions. Increased serum anionic concentrations might be to compensate increased cations in the blood. Serum Na and K are directly related to each other in blood and leads towards osmoregulation of body fluids (Mushtaq *et al.*, 2005).

It is concluded from the present study that birds are more sensitive to amount and source of dCl in later part of their life while remain ineffective during initial phases of life. Increasing amounts of dCl and replacing $CaCl_2$ with NH_4Cl proved to be effective for carcass characteristics. Higher requirements of dietary chloride were found but did not appear to disturb growth performance *per se*.

Table 4.29. Ingredient composition of the basal diets for different phases of birds fed 4 levels of chloride with two sources of chloride salts[1]

Ingredients (%)	Pre-starter (1 – 10 d)	Starter (11 – 20 d)	Grower (21 – 33 d)	Finisher (34 – 42 d)
Corn	30.00	19.00	10.00	36.00
Broken rice	25.25	39.00	50.00	25.70
Soybean meal	26.50	32.00	30.00	28.00
Canola Meal	10.00	3.25	–	–
Oil (Grease)	2.85	2.00	3.00	5.75
L-Lysine sulphate	0.60	0.29	0.23	0.16
DL-methionine	0.33	0.29	0.25	0.23
L-threonine	0.37	0.33	0.30	0.24
MCP	1.70	1.75	1.62	1.52
Limestone	1.56	1.56	1.50	1.46
NaCl	0.21	0.19	0.18	0.20
K_2CO_3	0.11	0.17	0.28	0.26
Premix[2]	0.50	0.50	0.50	0.50

[1]All the diets were supplemented with 4 levels of either $CaCl_2$ (0.11, 0.22, 0.33 or 0.44%) or NH_4Cl (0.105, 0.21, 0.315 or 0.42%) to make final Cl concentrations of 0.31, 0.45, 0.59 or 0.73%, respectively. The basal diet has 0.17% Cl in it.

[2]Provides per kg of finished diet: vitamin A, 12 mg; vitamin D_3, 7 mg; vitamin E, 100 mg; vitamin K_3 (50% as MNB), 3 mg; vitamin B_1 (98%), 3 mg; vitamin B_2 (800,000 mg), 12 mg; vitamin B_3 (niacin; 99%), 600 mg; vitamin B_6 (98%), 4 mg; vitamin B_9 (folic acid; 95%), 2 mg; vitamin B_{12} (0.10%), 20 mg; Biotin (0.10%), 5 mg; Ca-Pentothenate (98%), 12 mg; cholin (70% as choline chloride), 1 g; MnO (60%), 169 mg; $FeSO_4$ (21%), 200 mg; $ZnSO_4$ (36%), 150 mg; $CuSO_4$ (25%), 40 mg; Se (sodium selenite 0.40%), 100 mg; KI (68%), 2 mg; Salinomycin, 60 mg; Zinc bacitracin (as Albac 10%), 50 mg

Table 4.30. Nutrient composition of the basal diets for different phases of birds fed 4 levels of chloride with two sources of chloride salts[1]

Nutrients (%)	Pre-starter (1 – 10 d)	Starter (11 – 20 d)	Grower (21 – 33 d)	Finisher (34 – 42 d)
ME (kcal/kg)	2,892	3,005	3,099	3,176
Crude Protein	20.97	20.06	19.08	18.27
Crude fiber	4.45	3.86	3.82	3.50
Calcium	1.00	0.95	0.90	0.87
Available Phos.	0.45	0.45	0.42	0.40
Sodium	0.30	0.30	0.30	0.30
Potassium	0.92	0.92	0.92	0.92
Chloride	0.17	0.17	0.17	0.17
DEB (mEq/kg)	317	319	320	319
Dig Lys	1.23	1.06	0.98	0.90
Dig Met/ Dig Lys	0.54	0.47	0.43	0.40
Dig Met + Dig Cys/Dig Lys	0.77	0.65	0.60	0.57
Dig Thr/Dig Lys	0.82	0. 71	0.66	0.60
Dig Try/Dig Lys	0.14	0.13	0.13	0.11
Dig Arg/Dig Lys	1.27	1.23	1.29	1.15

[1] $CaCl_2$ and NH_4Cl

Table 4.31. Drinking water properties during the experimental period

Phase	Item[1]	Salinity (g/kg)	TDS[2] (mg/kg)	EC[3] (millisemen/cm)	Temperature (°C)	pH	DO[4] (mg/L)
Phase 1	Max	1.0	1234	1.39	33.1	7.49	5.03
	Min	0.8	1180	1.27	30.0	7.31	3.41
	Average	0.9	1211	1.32	31.7	7.40	4.25
Phase 2	Max	0.9	1061	1.31	29.6	7.35	4.79
	Min	0.8	1010	1.06	26.1	7.17	3.81
	Average	0.8	1036	1.17	27.4	7.27	4.31
Phase 3	Max	1.0	1187	1.24	26.2	7.40	5.10
	Min	0.9	1058	1.08	25.4	7.21	3.60
	Average	0.9	1094	1.15	25.7	7.33	4.36
Phase 4	Max	0.9	1194	1.21	25.1	7.33	4.76
	Min	0.8	1060	1.07	24.4	7.23	3.50
	Average	0.8	1126	1.11	24.8	7.29	4.19

[1]Max, Min and Average values were taken twice daily so one value is equal to days in phase × 2
[2]TDS - Total Dissolved Solids; [3]EC - Electrical Conductivity; [4]DO - Dissolved Oxygen

Table 4.32. Effect of dietary chloride and chloride salts on body weight gain of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			g		
Basal (0.17%)	147	339	694	468	1649
SEM	*6.1*	*10.5*	*10.6*	*64.4*	*46.7*
Dietary Cl (%)					
0.31	135	340	677	464	1615
0.45	130	336	684	574	1724
0.59	131	329	701	473	1633
0.73	138	320	683	476	1618
SEM	*3.0*	*7.7*	*18.4*	*37.4*	*37.4*
Salts					
$CaCl_2$	138	337	690	488	1653
NH_4Cl	129	326	682	505	1642
SEM	*2.1*	*5.4*	*13.0*	*26.4*	*26.5*
Cl × Salts					
0.31 × $CaCl_2$	138	352	708	455	1653
0.45 × $CaCl_2$	140	340	666	608	1753
0.59 × $CaCl_2$	132	332	723	386	1574
0.73 × $CaCl_2$	142	324	664	502	1631
0.31 × NH_4Cl	132	329	645	473	1578
0.45 × NH_4Cl	121	332	702	540	1695
0.59 × NH_4Cl	129	326	679	559	1693
0.73 × NH_4Cl	135	317	703	449	1604
SEM	*4.3*	*10.9*	*26.0*	*52.9*	*53.0*
ANOVA			***Probability***		
Cl	-	0.05	-	-	-
Cl_L	-	-	-	-	-
Cl_Q	-	-	-	-	-
Cl_C	-	-	0.04	0.009	-
Salt	0.009	-	-	-	-
Salt × Cl	-	-	-	-	-
Salt × Cl_L	-	-	-	-	-
Salt × Cl_Q	0.04	-	-	-	-
Salt × Cl_C	-	-	-	-	-

Cl_L, Cl_Q & Cl_C are linear, quadratic and cubic terms for Cl, respectively.

Table 4.33. Effect of dietary chloride and chloride salts on feed intake of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			g		
Basal (0.17%)	224	502	1519	1238	3483
SEM	*12.6*	*55.7*	*46.1*	*80.8*	*163.0*
Dietary Cl (%)					
0.31	221	516	1678	1118	3534
0.45	211	506	1626	1265	3608
0.59	207	516	1717	1216	3656
0.73	230	504	1598	1274	3606
SEM	*8.1*	*26.2*	*36.9*	*39.3*	*59.0*
Salts					
$CaCl_2$	217	498	1663	1233	3612
NH_4Cl	218	523	1646	1203	3590
SEM	*5.7*	*18.5*	*26.1*	*27.8*	*41.7*
Cl × Salts					
0.31 × $CaCl_2$	212	512	1669	1150	3544
0.45 × $CaCl_2$	215	492	1546	1237	3490
0.59 × $CaCl_2$	203	489	1832	1283	3807
0.73 × $CaCl_2$	238	498	1608	1264	3607
0.31 × NH_4Cl	229	520	1687	1087	3523
0.45 × NH_4Cl	207	520	1706	1293	3726
0.59 × NH_4Cl	211	543	1602	1148	3504
0.73 × NH_4Cl	223	510	1588	1284	3605
SEM	*11.5*	*37.0*	*52.2*	*55.6*	*83.4*
ANOVA			***Probability***		
Cl	-	-	-	0.03	-
Cl_L	-	-	-	0.03	-
Cl_Q	-	-	-	-	-
Cl_C	-	-	-	-	0.006
Salt	-	-	-	-	-
Salt × Cl	-	-	-	-	-
Salt × Cl_L	-	-	-	-	-
Salt × Cl_Q	-	-	-	-	-
Salt × Cl_C	-	-	0.001	0.04	-

Cl_L, Cl_Q & Cl_C are linear, quadratic and cubic terms for Cl, respectively.

Table 4.34. Effect of dietary chloride and chloride salts on feed:gain of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			g/g		
Basal (0.17%)	1.53	1.47	2.19	2.82	2.00
SEM	*0.137*	*0.133*	*0.077*	*0.486*	*0.165*
Dietary Cl (%)					
0.31	1.63	1.53	2.52	2.50	2.05
0.45	1.63	1.51	2.38	2.24	1.94
0.59	1.59	1.57	2.45	2.88	2.12
0.73	1.67	1.58	2.34	2.75	2.08
SEM	*0.048*	*0.085*	*0.080*	*0.224*	*0.062*
Salts					
$CaCl_2$	1.57	1.48	2.41	2.72	2.05
NH_4Cl	1.68	1.61	2.43	2.47	2.05
SEM	*0.034*	*0.060*	*0.056*	*0.158*	*0.044*
Cl × Salts					
0.31 × $CaCl_2$	1.53	1.47	2.36	2.70	2.01
0.45 × $CaCl_2$	1.54	1.45	2.33	2.04	1.84
0.59 × $CaCl_2$	1.54	1.47	2.54	3.50	2.26
0.73 × $CaCl_2$	1.68	1.54	2.43	2.64	2.07
0.31 × NH_4Cl	1.74	1.58	2.68	2.31	2.08
0.45 × NH_4Cl	1.71	1.58	2.43	2.44	2.04
0.59 × NH_4Cl	1.63	1.67	2.36	2.26	1.98
0.73 × NH_4Cl	1.65	1.61	2.26	2.87	2.10
SEM	*0.068*	*0.120*	*0.113*	*0.317*	*0.088*
ANOVA			***Probability***		
Cl	-	-	-	-	-
Cl_L	-	-	-	-	-
Cl_Q	-	-	-	-	-
Cl_C	-	-	-	0.004	0.007
Salt	0.03	-	-	-	-
Salt × Cl	-	-	0.02	-	-
Salt × Cl_L	-	-	0.02	-	-
Salt × Cl_Q	-	-	-	-	-
Salt × Cl_C	-	-	-	-	-

Cl_L, Cl_Q & Cl_C are linear, quadratic and cubic terms for Cl, respectively.

Table 4.35. Effect of dietary chloride and chloride salts on water intake of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			mL		
Basal (0.17%)	513	1189	3801	2582	8085
SEM	*20.5*	*52.8*	*288.0*	*47.9*	*385.0*
Dietary Cl (%)					
0.31	534	1108	3705	2478	7824
0.45	484	1138	3807	2420	7848
0.59	488	1091	3650	2479	7708
0.73	579	1101	3781	2587	8047
SEM	*22.8*	*38.3*	*71.6*	*83.2*	*158.9*
Salts					
$CaCl_2$	510	1107	3733	2499	7848
NH_4Cl	533	1111	3739	2482	7866
SEM	*16.1*	*27.1*	*50.6*	*58.8*	*112.4*
Cl × Salts					
0.31 × $CaCl_2$	535	1083	3808	2443	7870
0.45 × $CaCl_2$	508	1105	3742	2434	7789
0.59 × $CaCl_2$	466	1140	3633	2496	7735
0.73 × $CaCl_2$	528	1100	3748	2623	7998
0.31 × NH_4Cl	533	1132	3601	2512	7779
0.45 × NH_4Cl	460	1170	3873	2405	7907
0.59 × NH_4Cl	511	1042	3668	2462	7682
0.73 × NH_4Cl	630	1102	3814	2550	8096
SEM	*32.2*	*54.2*	*101.3*	*117.6*	*224.7*
ANOVA			***Probability***		
Cl	-	-	-	-	-
Cl_L	-	-	-	-	-
Cl_Q	-	-	-	-	-
Cl_C	-	-	-	-	-
Salt	-	-	-	-	-
Salt × Cl	-	-	-	-	-
Salt × Cl_L	-	-	-	-	-
Salt × Cl_Q	0.003	-	-	-	-
Salt × Cl_C	-	-	-	-	-

Cl_L, Cl_Q & Cl_C are linear, quadratic and cubic terms for Cl, respectively.

Table 4.36. Effect of dietary chloride and chloride salts on water intake-to-feed intake ratio of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			mL/g		
Basal (0.17%)	2.31	2.44	2.50	2.11	2.33
SEM	*0.136*	*0.231*	*0.170*	*0.125*	*0.119*
Dietary Cl (%)					
0.31	2.45	2.20	2.21	2.24	2.22
0.45	2.32	2.27	2.36	1.93	2.18
0.59	2.40	2.16	2.14	2.05	2.11
0.73	2.55	2.21	2.37	2.04	2.23
SEM	*0.167*	*0.127*	*0.065*	*0.094*	*0.058*
Salts					
$CaCl_2$	2.38	2.25	2.26	2.04	2.18
NH_4Cl	2.48	2.16	2.28	2.09	2.20
SEM	*0.118*	*0.090*	*0.046*	*0.067*	*0.041*
Cl × Salts					
0.31 × $CaCl_2$	2.57	2.13	2.28	2.14	2.22
0.45 × $CaCl_2$	2.41	2.26	2.45	1.98	2.24
0.59 × $CaCl_2$	2.30	2.40	1.99	1.95	2.03
0.73 × $CaCl_2$	2.24	2.23	2.33	2.09	2.22
0.31 × NH_4Cl	2.34	2.27	2.14	2.35	2.22
0.45 × NH_4Cl	2.23	2.28	2.27	1.88	2.13
0.59 × NH_4Cl	2.49	1.92	2.29	2.16	2.19
0.73 × NH_4Cl	2.85	2.19	2.41	1.99	2.25
SEM	*0.236*	*0.180*	*0.092*	*0.133*	*0.083*
ANOVA			***Probability***		
Cl	-	-	-	-	-
Cl_L	-	-	-	-	-
Cl_Q	-	-	-	-	-
Cl_C	-	-	0.009	-	-
Salt	-	-	-	-	-
Salt × Cl	0.05	-	-	-	-
Salt × Cl_L	-	-	-	-	-
Salt × Cl_Q	-	-	-	-	-
Salt × Cl_C	-	-	0.05	-	-

Cl_L, Cl_Q & Cl_C are linear, quadratic and cubic terms for Cl, respectively.

Table 4.37. Effect of dietary chloride and chloride salts on litter moisture of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			%		
Basal (0.17%)	15.20	31.91	39.25	31.81	29.54
SEM	*0.490*	*1.150*	*1.650*	*1.280*	*0.463*
Dietary Cl (%)					
0.31	15.85	37.87	33.57	32.28	29.89
0.45	14.35	35.37	44.30	40.98	33.75
0.59	15.23	36.55	56.87	40.97	34.91
0.73	13.26	36.07	55.12	35.04	34.87
SEM	*0.321*	*1.042*	*1.348*	*1.647*	*0.578*
Salts					
$CaCl_2$	14.75	41.28	49.79	36.47	35.57
NH_4Cl	14.60	31.65	40.15	38.16	31.14
SEM	*0.227*	*0.737*	*0.953*	*1.165*	*0.408*
Cl × Salts					
0.31 × $CaCl_2$	17.30	43.19	36.59	34.34	32.86
0.45 × $CaCl_2$	11.76	34.42	45.25	43.14	33.64
0.59 × $CaCl_2$	15.10	39.39	49.90	44.20	37.15
0.73 × $CaCl_2$	14.84	48.12	67.40	24.21	38.64
0.31 × NH_4Cl	14.40	32.56	30.55	30.22	26.93
0.45 × NH_4Cl	16.94	36.32	43.35	38.82	33.86
0.59 × NH_4Cl	15.36	33.71	43.84	37.73	32.66
0.73 × NH_4Cl	11.68	24.02	42.84	45.87	31.10
SEM	*0.454*	*1.473*	*1.907*	*2.330*	*0.817*
ANOVA			***Probability***		
Cl	0.04	-	≤0.001	-	≤0.001
Cl_L	≤0.001	-	≤0.001	-	≤0.001
Cl_Q	≤0.001	≤0.001	≤0.001	≤0.001	0.01
Cl_C	≤0.001	-	-	-	0.04
Salt	-	≤0.001	≤0.001	-	≤0.001
Salt × Cl	-	0.006	0.001	0.003	-
Salt × Cl_L	-	≤0.001	≤0.001	≤0.001	-
Salt × Cl_Q	0.002	-	0.04	0.009	≤0.001
Salt × Cl_C	-	-	-	-	-

Cl_L, Cl_Q & Cl_C are linear, quadratic and cubic terms for Cl, respectively.

Table 4.38. Effect of dietary chloride and chloride salts on mortality of broilers during various phases of the experiment

Item	1 – 10 d	11 – 20 d	21 – 33 d	34 – 42 d	1 – 42 d
			%		
Basal (0.17%)	8.7	1.1	0.0	0.5	10.3
SEM	*1.98*	*1.09*	*0.00*	*0.54*	*1.37*
Dietary Cl (%)					
0.31	5.2	1.1	0.5	0.0	6.8
0.45	7.3	1.9	0.8	0.0	10.0
0.59	6.2	1.4	0.3	0.0	7.9
0.73	6.2	2.4	0.0	1.1	9.8
SEM	*1.37*	*0.74*	*0.31*	*0.22*	*1.36*
Salts					
$CaCl_2$	7.5	1.4	0.3	0.3	9.4
NH_4Cl	5.0	2.0	0.5	0.3	7.9
SEM	*0.97*	*0.52*	*0.22*	*0.16*	*0.96*
Cl × Salts					
0.31 × $CaCl_2$	4.9	1.1	0.5	0.0	6.5
0.45 × $CaCl_2$	9.8	1.1	0.5	0.0	11.4
0.59 × $CaCl_2$	8.2	2.2	0.0	0.0	10.3
0.73 × $CaCl_2$	7.1	1.1	0.0	1.1	9.2
0.31 × NH_4Cl	5.4	1.1	0.5	0.0	7.1
0.45 × NH_4Cl	4.9	2.7	1.1	0.0	8.7
0.59 × NH_4Cl	4.4	0.5	0.5	0.0	5.4
0.73 × NH_4Cl	5.4	3.8	0.0	1.1	10.3
SEM	*1.93*	*1.05*	*0.44*	*0.31*	*1.92*
ANOVA			***Probability***		
Cl	-	-	-	0.004	-
Cl_L	-	-	-	0.003	-
Cl_Q	-	-	-	-	-
Cl_C	-	-	-	-	-
Salt	-	-	-	-	-
Salt × Cl	-	-	-	-	-
Salt × Cl_L	-	-	-	-	-
Salt × Cl_Q	-	-	-	0.03	-
Salt × Cl_C	-	-	-	-	-

Cl_L, Cl_Q & Cl_C are linear, quadratic and cubic terms for Cl, respectively.

Table 4.39. Effect of dietary chloride and chloride salts on blood and carcass responses of broilers at the end of the experiment

Item	Blood pH	Blood glucose[0]	Dressing weight[1]	Breast weight[2]	Thigh weight[2]	Intestinal weight[3]	Intestinal length[4]
Basal (0.17%)	7.35	183.3	55.70	38.60	45.02	64.87	178.4
SEM	*0.003*	*7.18*	*1.280*	*1.240*	*1.230*	*3.260*	*4.31*
Dietary Cl (%)							
0.31	7.32	171.9	55.96	39.55	44.83	61.13	177.2
0.45	7.30	177.7	55.07	39.13	45.52	57.82	181.0
0.59	7.29	175.5	55.74	39.36	45.09	61.01	181.1
0.73	7.28	178.3	56.24	40.38	44.49	60.05	167.7
SEM	*0.002*	*4.20*	*0.624*	*0.646*	*0.614*	*2.528*	*2.90*
Salts							
$CaCl_2$	7.28	168.3	56.53	38.66	44.26	63.75	177.5
NH_4Cl	7.32	183.4	54.97	40.55	45.71	56.25	176.0
SEM	*0.001*	*2.97*	*0.441*	*0.457*	*0.434*	*1.787*	*2.05*
Cl × Salts							
0.31 × $CaCl_2$	7.29	173.9	57.62	38.15	44.42	63.38	179.4
0.45 × $CaCl_2$	7.28	171.3	56.79	37.57	44.38	64.57	175.8
0.59 × $CaCl_2$	7.28	164.6	55.84	39.47	44.09	67.90	180.4
0.73 × $CaCl_2$	7.27	163.3	55.90	39.46	44.14	59.16	174.6
0.31 × NH_4Cl	7.35	169.8	54.30	40.94	45.24	58.88	175.1
0.45 × NH_4Cl	7.32	184.0	53.34	40.68	46.66	51.06	186.2
0.59 × NH_4Cl	7.30	186.3	55.65	39.26	46.09	54.12	181.8
0.73 × NH_4Cl	7.29	193.3	56.58	41.31	44.84	60.94	160.8
SEM	*0.002*	*5.94*	*0.882*	*0.913*	*0.869*	*3.575*	*4.10*
ANOVA		———	———	***Probability***	———	———	
Cl	≤0.001	-	-	-	-	-	0.005
Cl_L	≤0.001	-	-	-	-	-	0.03
Cl_Q	-	-	-	-	-	-	0.005
Cl_C	-	-	-	-	-	-	-
Salt	≤0.001	0.001	0.02	0.005	0.02	0.004	-
Salt × Cl	≤0.001	0.04	0.04	-	-	-	0.03
Salt × Cl_L	≤0.001	0.004	0.008	-	-	-	-
Salt × Cl_Q	0.05	-	-	-	-	0.02	0.01
Salt × Cl_C	-	-	-	-	-	-	-

[1]% of live weight (without visceral organs)
[2]% of dressed weight; [0] mg/dL; [4] gram; [5] centimetres
Cl_L, Cl_Q & Cl_C are linear, quadratic and cubic terms for Cl, respectively.

Table 4.40. Effect of dietary chloride and chloride salts on body organs weights of broilers at the end of the experiment

Item	Gizzard[1]	Proventriculus[1]	Heart[1]	Liver[1]	Kidney[1]
		% of dressed weight			
Basal (0.17%)	2.67	0.57	0.79	3.19	0.74
SEM	*0.100*	*0.028*	*0.034*	*0.094*	*0.037*
Dietary Cl (%)					
0.31	2.91	0.53	0.69	2.83	0.61
0.45	2.73	0.61	0.74	3.20	0.72
0.59	2.82	0.50	0.70	3.31	0.70
0.73	2.93	0.54	0.71	3.14	0.62
SEM	*0.092*	*0.024*	*0.026*	*0.094*	*0.031*
Salts					
$CaCl_2$	2.85	0.55	0.72	3.02	0.66
NH_4Cl	2.84	0.54	0.71	3.22	0.66
SEM	*0.065*	*0.017*	*0.018*	*0.067*	*0.022*
Cl × Salts					
0.31 × $CaCl_2$	2.99	0.48	0.73	2.44	0.56
0.45 × $CaCl_2$	2.64	0.63	0.79	3.24	0.72
0.59 × $CaCl_2$	2.80	0.55	0.69	3.15	0.70
0.73 × $CaCl_2$	2.99	0.55	0.66	3.27	0.68
0.31 × NH_4Cl	2.83	0.59	0.65	3.21	0.66
0.45 × NH_4Cl	2.82	0.59	0.69	3.17	0.73
0.59 × NH_4Cl	2.84	0.46	0.72	3.47	0.71
0.73 × NH_4Cl	2.88	0.52	0.76	3.01	0.57
SEM	*0.130*	*0.033*	*0.036*	*0.134*	*0.044*
ANOVA		***Probability***			
Cl	-	0.02	-	0.005	0.02
Cl_L	-	-	-	-	-
Cl_Q	-		-	0.005	0.003
Cl_C	-	0.003	-	-	-
Salt	-	-	-	0.05	-
Salt × Cl	-	0.03	0.02	0.001	-
Salt × Cl_L	-	0.04	0.005	0.002	0.03
Salt × Cl_Q	-	0.03	-	-	-
Salt × Cl_C	-	-	-	0.01	-

[1]% of live weight (with visceral organs)
Cl_L, Cl_Q & Cl_C are linear, quadratic and cubic terms for Cl, respectively.

Table 4.41. Effect of dietary chloride and chloride salts on body organs weights of broilers at the end of the experiment

Item	Spleen[2]	Pancreas[2]	Bursa[2]	Gall bladder[2]	Lungs[2]	Abdominal fat[1]	Shank length[3]
		% of dressed weight					
Basal (0.17%)	0.08	0.38	0.26	0.10	0.64	3.29	6.33
SEM	*0.005*	*0.024*	*0.015*	*0.011*	*0.040*	*0.281*	*0.255*
Dietary Cl (%)							
0.31	0.13	0.33	0.20	0.08	0.76	3.07	6.30
0.45	0.12	0.35	0.23	0.10	0.77	2.73	6.32
0.59	0.13	0.37	0.25	0.10	0.74	2.86	6.26
0.73	0.13	0.31	0.21	0.09	0.73	2.82	6.18
SEM	*0.010*	*0.026*	*0.016*	*0.007*	*0.042*	*0.189*	*0.078*
Salts							
$CaCl_2$	0.12	0.33	0.22	0.10	0.74	2.73	6.19
NH_4Cl	0.13	0.35	0.22	0.08	0.76	3.01	6.34
SEM	*0.007*	*0.018*	*0.011*	*0.005*	*0.029*	*0.133*	*0.055*
Cl × Salts							
0.31 × $CaCl_2$	0.12	0.30	0.18	0.10	0.70	3.14	6.18
0.45 × $CaCl_2$	0.12	0.35	0.23	0.13	0.82	2.87	6.11
0.59 × $CaCl_2$	0.13	0.31	0.26	0.11	0.73	2.62	6.28
0.73 × $CaCl_2$	0.13	0.36	0.22	0.09	0.69	2.30	6.19
0.31 × NH_4Cl	0.14	0.36	0.23	0.07	0.82	3.00	6.42
0.45 × NH_4Cl	0.12	0.35	0.23	0.06	0.72	2.58	6.54
0.59 × NH_4Cl	0.13	0.43	0.24	0.10	0.75	3.10	6.24
0.73 × NH_4Cl	0.12	0.27	0.20	0.09	0.76	3.35	6.16
SEM	*0.013*	*0.037*	*0.023*	*0.009*	*0.059*	*0.267*	*0.110*
ANOVA		*Probability*					
Cl	-	-	-	-	-	-	-
Cl_L	-	-	-	-	-	-	-
Cl_Q	-	-	0.04	0.05	-	-	-
Cl_C	-	-	-	-	-	-	-
Salt	-	-	-	≤0.001	-	-	0.05
Salt × Cl	-	0.05	-	0.009	-	0.03	-
Salt × Cl_L	-	-	-	-	-	0.01	-
Salt × Cl_Q	-	-	-	-	-	-	-
Salt × Cl_C	-	0.04	-	0.02	-	-	-

[1]% of live weight (without visceral organs);
[2]% of dressed weight;
[3]shank length was measured from hock joint to tarsometatarsus joint in centimetres;
Cl_L, Cl_Q & Cl_C are linear, quadratic and cubic terms for Cl, respectively

Table 4.42. Effect of dietary chloride and chloride salts on serum mineral chemistry of broilers at the end of the experiment

Item	Na	K	Cl	Ca	P	Mg	HCO_3
	———————	mmol/L	———————				
Basal (0.17%)	136	3.05	106	2.60	2.89	1.02	33.6
SEM	*0.5*	*0.005*	*0.2*	*0.091*	*0.083*	*0.025*	*0.09*
Dietary Cl (%)							
0.31	138	3.08	107	2.46	3.11	1.09	33.0
0.45	136	3.05	109	2.31	2.92	1.09	32.4
0.59	135	3.06	111	2.04	2.99	1.18	31.9
0.73	136	3.05	112	2.09	3.12	1.08	31.4
SEM	*0.4*	*0.006*	*0.2*	*0.101*	*0.116*	*0.048*	*0.07*
Salts							
$CaCl_2$	134	3.06	110	2.40	3.03	1.13	31.9
NH_4Cl	138	3.07	109	2.05	3.04	1.09	32.4
SEM	*0.1*	*0.004*	*0.2*	*0.071*	*0.082*	*0.034*	*0.05*
K × Salts							
0.31 × $CaCl_2$	138	3.08	108	2.90	3.09	1.20	32.7
0.45 × $CaCl_2$	133	3.05	109	2.50	2.88	1.01	32.2
0.59 × $CaCl_2$	132	3.05	111	2.09	2.89	1.27	31.6
0.73 × $CaCl_2$	135	3.05	112	2.12	3.26	1.04	31.1
0.31 × NH_4Cl	138	3.09	106	2.02	3.13	0.98	33.2
0.45 × NH_4Cl	139	3.06	108	2.12	2.97	1.16	32.6
0.59 × NH_4Cl	138	3.08	110	2.00	3.09	1.09	32.2
0.73 × NH_4Cl	138	3.05	112	2.06	2.97	1.12	31.7
SEM	*0.6*	*0.008*	*0.3*	*0.143*	*0.164*	*0.068*	*0.10*
ANOVA		———————	*Probability*	———————			
Cl	-	-	-	-	-	-	-
Cl_L	0.005	0.001	≤0.001	0.003	-	-	≤0.001
Cl_Q	0.001	-	0.04	-	-	-	-
Cl_C	-	0.03	-	-	-	-	-
Salt	≤0.001	-	0.001	0.001	-	-	≤0.001
Salt × Cl_L	0.001	-	-	0.003	-	-	-
Salt × Cl_Q	≤0.001	-	-	-	-	-	-
Salt × Cl_c	-	-	-	-	-	0.004	-

Cl_L, Cl_Q & Cl_C are linear, quadratic and cubic terms for Cl, respectively

Chapter 5

SUMMARY

A series of experiments were envisaged to evaluate the effect of supplementation of dietary electrolytes with applicability of dietary electrolyte balance by using different salts on growth and carcass responses, body physiological responses and litter condition of modern day broiler chickens under phase feeding system.

Day-old straight-run Hubbard broiler chicks were randomly allocated to eight dietary treatments replicated four times in such a way that a floor space of 0.09 m^2 was provided to each bird. Birds were housed in environmental control system. Continuous light was provided 24 hours for the first 3 days and thereafter a light pattern of 23L:1D was adopted for the entire experimental. In each experiment, a basal diet was formulated having lowest level of each electrolyte. In experiment 1, Na and DEB in the basal diet were maintained at 0.08% and 160 mEq/kg, respectively. This basal diet was then supplemented with sodium bicarbonate ($NaHCO_3$) and disodium sulphate (Na_2SO_4) to maintain four levels of Na^+ (0.17, 0.26, 0.35, and 0.44%) by fixing K and Cl with DEB 200, 240, 280 and 320 mEq/kg, respectively. In experiment 2, a basal diet was prepared to contain the lowest level of K and DEB i.e. 0.70% and 160 mEq/kg, respectively. This basal diet was supplemented with potassium sulphate (K_2SO_4) and potassium carbonate (K_2CO_4) by fixing Na and Cl. So, four levels of K (0.86, 1.02, 1.18, and 1.34%) were maintained in eight dietary treatments. In experiment 3, a basal diet was prepared to contain the lowest level of Cl and DEB i.e. 0.17% and 320 mEq/kg, respectively. This basal diet was supplemented with ammonium chloride (NH_4Cl) or calcium chloride ($CaCl_2$), so that, in each diet, we can have the increase of 40 mEq/kg DEB at 0.31, 0.45, 0.59 and 0.73% of Cl at DEB 280, 240, 200 and 160 mEq/kg, respectively, by fixing Na and K.

At the end of each phase (pre-starter, starter, grower and finisher); data of feed intake, weight gain, feed to gain ratio, mortality, water intake, water intake-to-feed intake ratio and litter quality were collected and evaluated. At the end of each experiment, two birds were slaughtered for their carcass and body physiological responses. Blood was also collected from these same birds for blood pH, glucose and serum mineral analyses. For statistical analyses, four (4) levels of electrolyte were used with two (2) sources of salt in a factorial arrangement of 4 × 2 under completely randomized design using GLM.

In experiment 1, highest weight gain and feed intake were found in birds consuming 0.17% ($NaHCO_3$) and 0.44% (Na_2SO_4) dNa, respectively during d 1-10. However during d 11-20, weight gain and feed:gain were reduced with same levels of dNa. Maximum weight gain was

found in diets containing 0.17 and 0.24% dNa during d 21-33 and 34-42, respectively. Improved FG was the result of diets containing 0.20% ($NaHCO_3$) and 0.37% (Na_2SO_4) dNa during d 21-33. Linear rise in water intake was observed in birds with increasing dNa during d 1-42. Minimum litter dampness was seen at 0.37% ($NaHCO_3$) and 0.21% (Na_2SO_4) during d 1-10. Minimum and maximum mortality were observed at 0.37% level of dNa in case of supplementation of $NaHCO_3$ and Na_2SO_4, respectively. Significantly increased pH and kidney weight while reduced dressing percentage were observed by amount and salt of dNa. Increased breast, thigh and gizzard weights were observed with increasing sodium. Weights of pancreas, gall bladder, bursa, and lungs, and shank length were affected by interaction of amount and salt of dNa.

In experiment 2, BWG ($P \leq 0.03$) and feed:gain ($P \leq 0.05$) was improved at 1.20% dK during 32 to 42 d of age. K_2SO_4 supplemented diets increased feed intake during 1 to 10 d ($P \leq 0.05$), water intake during 34 to 42 d ($P \leq 0.04$) and mortality during 1 to 42 d ($P \leq 0.02$). Water intake was increased linearly with increasing dK when supplemented by K_2CO_3 whereas this was decreased linearly with increasing dK with that of K_2SO_4 during 11 to 20 d ($P \leq 0.002$). The K_2SO_4 supplemented diets lowered the blood pH ($P \leq 0.001$), dressing ($P \leq 0.04$), abdominal fat ($P \leq 0.03$) weights and shank length ($P \leq 0.02$). A significant salt × dK effect was observed where low levels of dK with K_2CO_3 and high levels with K_2SO_4 exhibited lower litter moisture during all phases. Increasing concentration of serum cations was observed by increasing dK, by balancing of increasing serum HCO_3 with decreasing Cl at the end of the experiment.

In experiment 3, body weight gain and water consumption were optimized at 0.73%, and 0.73% ($CaCl_2$) and 0.45% (NH_4Cl), respectively, during d 1-10. During d 21-33, maximum weight gain and feed intake were observed at 0.42%, and 0.63% ($CaCl_2$) and 0.63% (NH_4Cl), respectively. Highest weight gain (0.60% dCl), feed intake (0.61% $CaCl_2$; 0.42% NH_4Cl) and mortality (0.73%) while improved feed:gain (FG; 0.38% dCl) were obtained by interaction effects of amount and source of dCl during d 34-42. FI (0.60%), feed:gain (0.38%) and litter moisture (0.31% NH_4Cl; 0.35 $CaCl_2$) was affected during 1-42d by amount of dCl. Increased blood pH, serum glucose and dressing percentage were found by dCl and replacing $CaCl_2$ with NH_4Cl. Improved breast meat, thigh meat and shank length while reduced abdominal fat were observed by replacing salts ($CaCl_2$ with NH_4Cl).

It is concluded that birds showed better growth performance and reduced mortality against high levels of dietary sodium in Na_2SO_4 than $NaHCO_3$ supplemented diets, while significant rise in pH, breast and thigh meat yield while reduced dressing percentage were observed with increasing dietary sodium. The importance of high concentration of dK for better weight gain and feed efficiency was depicted in later stages of production. K_2CO_3 increased survivability and dressing responses but both dK levels and salts played important role for water intake,

litter condition, carcass characteristics and serum mineral concentration. Birds were also suggested to be more sensitive to amount and source of dCl in later part of their life.

Chapter 6

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