

## Endocrinopathies associated with Ascites syndrome in Poultry of Kashmir Valley

Dr. Syed Bisma Ayub Kashani, Mohammad Maqbool Darzi, Dr. Nida Handoo, Dr. Ajaz Quadir

<sup>1</sup>Phd Scholar, Veterinary Pathology, FVSc & AH, SKUAST K, Shuhama, 190006.

<sup>2</sup>Professor and Head Division of Vety Pathology, FVSc & AH, SKUAST- K, Shuhama, 190006.

<sup>3</sup>Phd Scholar, Veterinary Surgery and Radiology, FVSc & AH, SKUAST K, Shuhama, 190006.

<sup>4</sup>Phd Scholar, Veterinary Anatomy, FVSc & AH, SKUAST K, Shuhama, 190006.

### ABSTRACT

The present work was undertaken with the objective of exploring the status of some endocrine glands like thyroid glands, adrenal glands and pancreas in Ascites syndrome of poultry in Kashmir valley. The birds (broilers as well as layers) belonged to various poultry farms in Srinagar and Ganderbal districts of Kashmir valley (Jammu and Kashmir, India). A total of 82 disease outbreaks were recorded over a period of one year (from June 2015- July 2016), out of which 16 outbreaks of Ascites were observed in broilers while as no outbreak of Ascites was noted in layers. Diagnosis was confirmed after conducting thorough post mortem examination. The disease was diagnosed grossly. On post mortem examination, the adrenal glands, thyroid glands and pancreas were properly collected from the carcasses of the ascites affected birds and preserved in 10% neutral buffered formalin for fixation. The tissue samples were processed for routine paraffin embedding technique and 5 micron thin tissue sections were stained with Haris Haematoxylin and Eosin. From the present study it could be concluded that any morphological or functional aberrations in the endocrine glands will definitely, therefore, affect the functional capacities of the target organs as well as the glands itself. Endocrinopathies co-existed with Ascites syndrome in broilers, and were mostly of hemodynamic derangements. Hence investigations into endocrine gland disorders in poultry needs attention for increasing the productivity and profitability from this enterprise.

**Key words:** Ascites, Adrenal glands, Histopathology, Pancreas, Thyroid glands.

### 1. INTRODUCTION

The endocrine system of birds consists of the hypothalamic-hypophyseal complex, the gonads, pancreatic islet cells, Adrenal glands, Thyroid glands, Parathyroid glands, Ultimobranchial glands and the endocrine cells of the gut. These organs release special compounds called hormones into the blood stream, which in turn, target particular

systems or organs. Endocrine glands are ductless glands. They secrete their secretions into the surrounding blood capillaries, unlike exocrine glands, which secrete their products into a duct. When these hormones get out of balance, the bird's body cannot function properly and hence performance will suffer and in some cases an imbalance can even lead to death [1]. Thyroid gland is one of the most important glands of the endocrine system. Its hormones triiodothyronine (T3) and thyroxin (T4) are involved in wide range of metabolic activities influencing the growth and development of organisms. The thyroid hormones are primarily involved in energy production by increasing the metabolic rate. This increase in energy production is to the greatest extent manifested as heat production. In homeothermic animals and birds the thyroids are responsible for the maintenance of constant body temperature [2]. Thyroid hormones are involved in controlling metabolic rate, and the concentration of circulating T3 is positively correlated with oxygen consumption in broilers [3,4]. Thyroid growth has an essentially constant proportion to body growth in galliform birds during both embryonic lives [5] and post hatch growth. Since the production of broilers in the poultry industry lasts only 42 days, one would expect that thyroid hormones should play a vital role during this process. Adrenal glands are important endocrine glands. They help to maintain homeostasis as well as play important roles in all types of stress response [6,7]. The microanatomy of the avian adrenal gland differs that of mammals, in that, a distinct cortex and medulla is absent in avian adrenal gland [8]. It shows two morphologically and physiologically different endocrine regions, the inter renal tissue which synthesizes hormones such as cortisol, cortisone, testosterone and small amounts of estradiol during embryonic period and chromaffin cells producing epinephrine and norepinephrine. The inter renal chromaffin tissue ratio will aid in understanding the functional significance of the chicken adrenal gland. These hormones perform the vital functions of an organism such as metabolism, homeostasis and stress. Corticosterone is essential for survival in times of stress and regulates intermediary metabolism and hemodynamic functions and it also balances the production and action of biologically active substances produced during stress (catecholamines, prostaglandins) and young chickens respond quickly to stress stimuli. Epinephrine (catecholamine) when produced increases heart rate, contracts blood vessels and dilates air passages and participates in the "flight or fright" response [9]. Pancreas in poultry consists of a pale, elongated gland, situated in the interduodenal area and formed by the ascending and descending duodenal loops [10,11]. It is associated with the digestive system and is an exocrine as well as endocrine gland. The pancreatic islets are responsible for the control of blood sugar concentration and consists of isolated groups of pale staining islet cells called islets of Langerhans [12]. Ascites syndrome (accumulation of fluid in the peritoneal cavity) appears in fast-growing chickens, mostly during the winter, and is a considerable cause of mortality [13]. The pathogenesis of ascites is similar to that of high-altitude disease, characterized by an imbalance between oxygen supply and oxygen need, which causes hypoxemia. Hypoxemia initiates a cascade of events, including increased cardiac output, increased pulmonary blood pressure (hypertension), enlargement of the right ventricle, and cardiopulmonary dysfunction that results in ascites and death [14,15]. Increased susceptibility of broilers to ascites has been linked

with intensive growth[16]. Ambient temperature ( $T_a$ ) and dietary energy level are two factors that are related to the rate of metabolic activity and, hence, to the amount of oxygen required by the animal [17, 18, 19]. A higher metabolic rate is associated with increased secretion of the hormone thyroxine ( $T_4$ ), which is deiodinated to triiodothyronine ( $T_3$ ) in the periphery, mainly in the liver and kidneys. Triiodothyronine is the main metabolic stimulating hormone. Plasma  $T_3$  is associated with temperature regulation and is an important growth promoter in chickens [20, 21, 22] it may thus be involved in modification of growth rate in response to environmental temperature. The circulating concentration of  $T_3$  is increased at low temperatures [23,24] and is positively correlated to feed intake [25,26,27,]. Thus a better understanding of the pathomorphological alterations of various endocrine glands in Ascites syndrome of poultry will help in evaluation of their status. Moreover, the literature with regard to endocrine pathology in poultry is scantily documented. No study has been undertaken on the pathological aspect of endocrine glands in Kashmir valley. The present paper describes the endocrinopathies associated with Ascites syndrome in Poultry of Kashmir valley.

## **2. MATERIALS AND METHODS**

### **2.1. Study Material and Sampling Area**

The poultry birds ( broilers and layers) died of various diseases during the period from July 2015 to June 2016, and referred for postmortem examination in the Division of Veterinary Pathology, Faculty of Veterinary Sciences and Animal Husbandry, Sher-e-Kashmir University of Agricultural Sciences and Technology Kashmir, Shuhama, Ganderbal formed the material for the study. These were brought by the private poultry farmers from Srinagar and Ganderbal Districts and their adjoining areas. Dead birds were also collected from Government poultry farm located at Hariparbat, Srinagar.

### **2.2. Investigation approach**

The investigation approach was principally based on

**2.3. Disease Diagnosis:** The clinical history of dead birds was recorded from the owners of the poultry farms. Dead birds were then subjected to thorough post mortem examination. External examination and internal examination of birds was performed. Diagnosis of the disease was done in the Division of Veterinary Pathology.

### **2.4. Pathoanatomical Studies**

#### **2.4.1. Gross pathology**

Gross pathological changes suggestive of Ascites syndrome were recorded in the dead birds and the gross pathology of the endocrine glands under study i.e adrenal glands, thyroid glands and pancreas were recorded.

### 2.4.2. Histopathology

Representative samples of adrenal glands, thyroid glands and pancreas were collected from dead birds that died of Ascites syndrome, subsequently preserved and fixed in 10% buffered formalin for fixation. The tissue samples were processed for routine paraffin embedding technique employing alcohol and acetone as dehydrating agent, benzene as clearing agent and paraffin wax of melting point 60°C. The sections of 5µm thickness were cut and stained with Haris' Haematoxylin and Eosin for routine examination [28].

## 2.5. RESULTS AND DISCUSSION

### 2.5.1. Gross pathology

The affected broilers revealed pot bellied appearance due to abdominal distension (Fig 4) with accumulation of straw yellow coloured fluid in different compartments of peritoneal cavities. Lungs showed varying degrees of congestion and oedema. Thyroid glands, adrenals and pancreas were congested and oedematous.

### 2.5.2. Histopathology

#### 2.5.2.1. Thyroid gland

Thyroid follicles were lined essentially with squamous type of epithelium and filled with colloid. Occasionally follicular epithelium was cuboidal to columnar. The histological structure of thyroid glands reveals primarily the capsule enclosing the follicles that contained colloid which forms the parenchyma of the gland. The thyroid follicles are filled with a homogenous colloidal mass. The variability in the type of follicular epithelium like cuboidal, columnar or squamous along with the colour intensity (light pink to pink) of colloid were attributed to different functional status of the gland in the present study. Cuboidal to columnar cell lined follicles were considered active follicles while squamous cell lined follicles with no vacuolation in the colloid were treated as inactive follicles. [29] reported that in the follicles which are lined with simple cuboidal or low columnar epithelium the colloid is predominantly basophilic, while those follicles lined by simple squamous epithelium have acidophilic colloid. [30] reported that the freshly secreted colloid in human thyroid assumed a blue colour and later, on aging it became pink. [31] recorded that the thyroid colloid was acidophilic when dense and basophilic when dilute. The colloid stains basophilic in active follicles and contains vacuoles, whereas it stains eosinophilic for inactive follicles with no vacuoles or rare ones. Follicles were lined by single layer of epithelial cells and type of epithelium depends upon their functional status. Therefore, based on the type of epithelium and nature of colloid, the follicles were categorized as active follicles and inactive follicles. The active follicles were lined by simple cuboidal epithelium, while the inactive follicles were lined by simple squamous epithelium in fowl and quail [ 32,33,34]. The quantity of

the colloid varied according to the activity of the thyroid gland. In the inactive follicles it was more and thick due to accumulation of large amount of colloid without being utilized, whereas in active follicles it was lesser and thinner due to regular production and consumption. Physiological significance ascribed to this multiple staining reaction appeared to be due to difference in the concentration of protein that depended upon the direction and rate of penetration of the fixatives into the tissue. Occasionally focal mononuclear cell infiltration was noted in the parenchyma of the gland (Fig 5). The glands revealed marked vascular congestion (Fig 6). The host response to the infectious disease alters metabolism [35]. The systemic stress response to infection contributes to the expression of metabolic disorders [36] and the immune response to infectious agents has a profound effect on general metabolic processes [37]. The monokines, acute phase proteins and other mediators released by macrophages, lymphocytes and other leucocytes increase body temperature which might have further resulted in haemodynamic changes leading to significant increase in blood flow and decreased vascular resistance. Oedema was also observed ( Fig 7) might be a part of generalized vascular derangement seen in the disease condition.

#### **2.5.2.2. Adrenal gland**

Adrenal glands revealed congestion, which is attributed to the stress factor induced by the Ascites syndrome. [38] reported marked hyperemia in adrenal glands of broilers exposed to the long term sound stress. Severe oedema was also observed which was attributed by stress and pulmonary hypertension in ascites, and mononuclear cell infiltration was also evident ( Figs. 8,9,10) which might be avian stress reaction [39,40].

#### **2.5.2.3. Pancreas**

Pancreas showed vascular congestion ( Fig 11) , which is in accordance with the findings of [41] however no significant change was seen in the endocrine part of the gland.

### **2.6. CONCLUSION**

From the present study it could be concluded that endocrinopathies co-existed with Ascites syndrome in broilers, and were mostly of hemodynamic derangements. Endocrinology has gained importance both in humans as well as in animals as their derangement is directly going to affect the productive capacities of animal. Poultry is not an exception. Endocrinopathies in birds do occur though however avian practitioners seem to remain mostly unaware to endocrinology. Knowledge about the endocrinopathies in birds is going to gain momentum as working know how of avian endocrinology is explored more and more, and appropriate clinical diagnostic tests can be used to document endocrine abnormalities.

### **2.7. Acknowledgements**

The authors are highly thankful to the entire staff of the Division of Veterinary Pathology for their timely support during the research work.

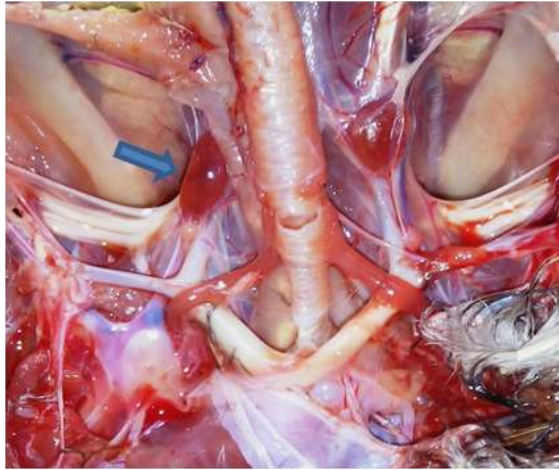


Fig 1. Pair of normal thyroid glands (arrow) on each side of the trachea at the level of clavicle in poultry.

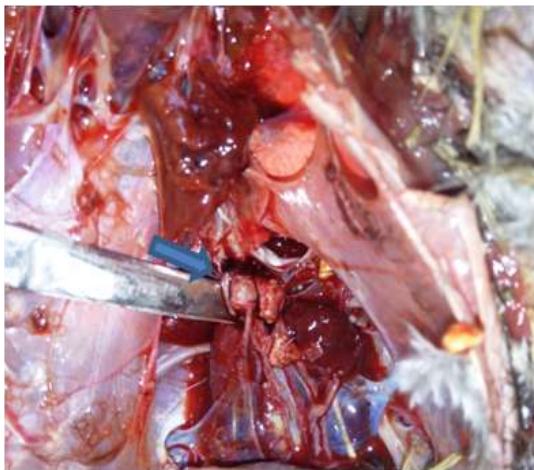


Fig 2. Pair of normal adrenal glands (arrow) located on each side of the median line just anterior to the bifurcation of the caudal vena cava.



Fig 3. Normal pancreas located in the inter-duodenal area and formed by the ascending and descending loop.

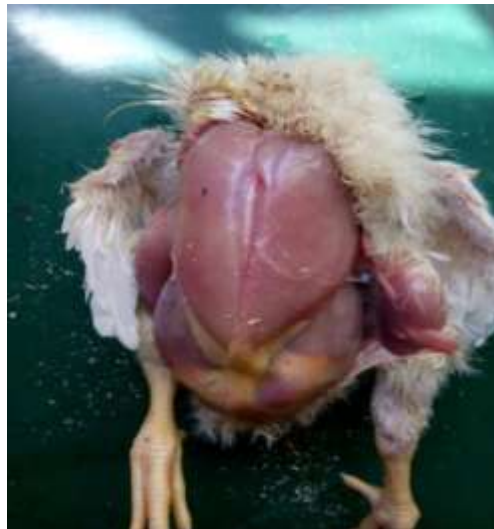


Fig 4. A broiler chicken affected with ascites showing distended abdomen due to accumulated ascitic fluid.

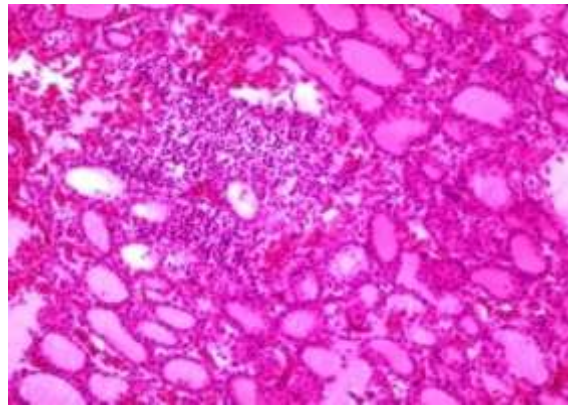


Fig 5. Section of thyroid gland of a broiler chicken affected with Ascites syndrome showing focal mononuclear cell infiltration in the parenchyma of the gland. H& E. Original magnification-400 X.

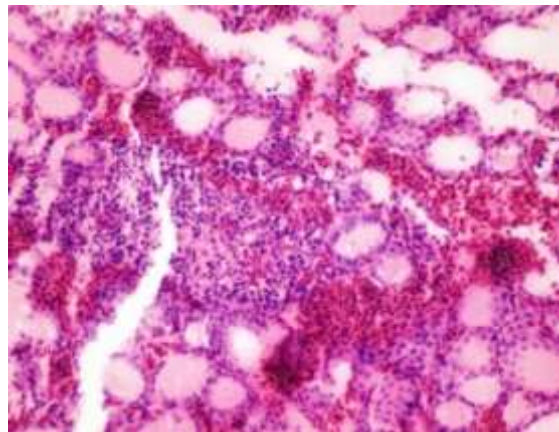


Fig 6. Section of thyroid gland of a broiler chicken affected with Ascites syndrome showing congestion and focal mononuclear cell infiltration. H&E. Original magnification-400X.



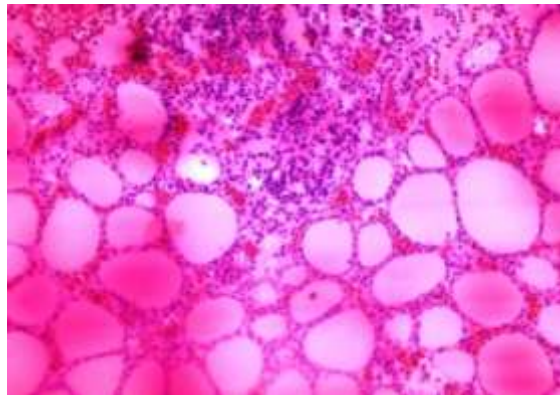


Fig 7. Section of thyroid gland of a broiler chicken affected with Ascites syndrome revealing, congestion, oedema and infiltration of mononuclear cells. H&E. Original magnification-400X.

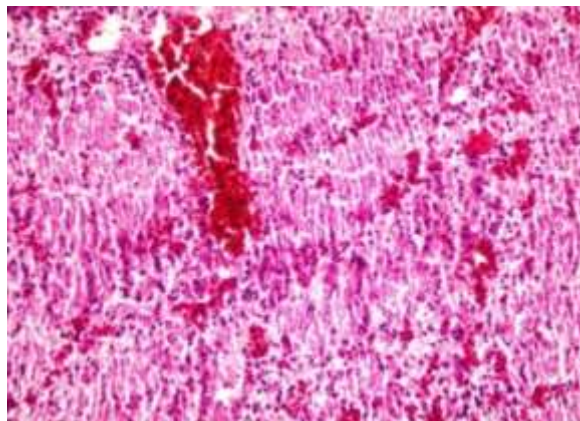


Fig 8. Section of adrenal gland of a broiler chicken affected with Ascites syndrome revealing severe congestion. H&E . Original magnification-400X.

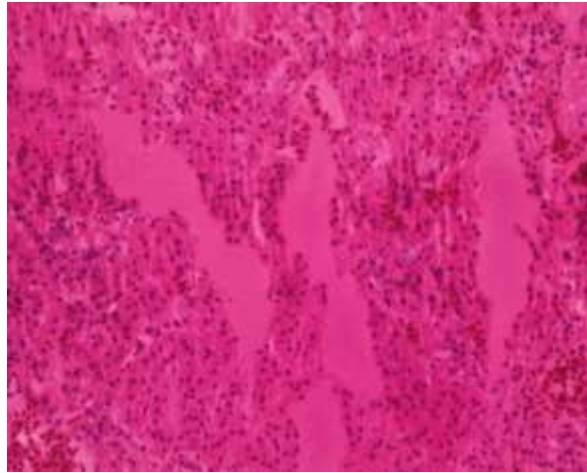


Fig 9. Section of adrenal gland of a broiler chicken affected with Ascites syndrome showing severe oedema. H&E. Original magnification-400X.

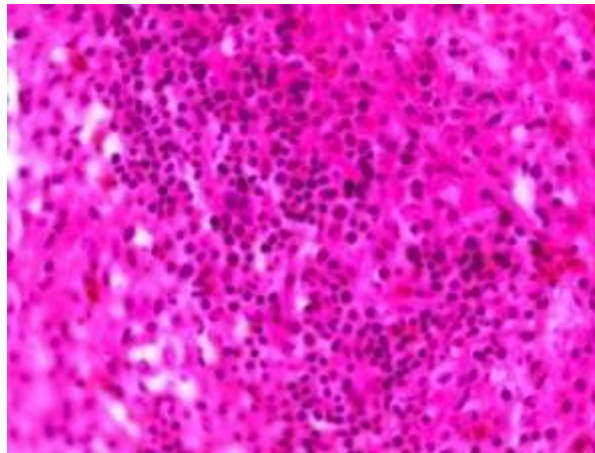
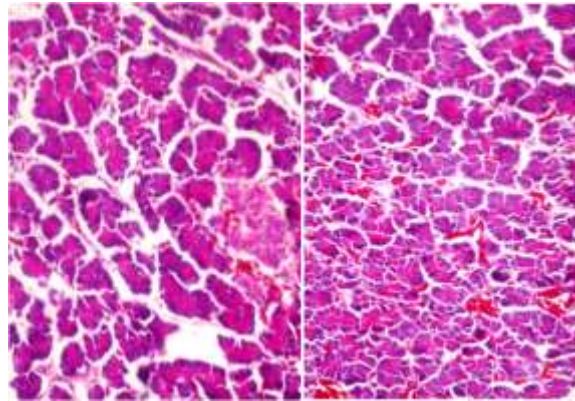


Fig 10. Section of adrenal gland of a broiler chicken affected with Ascites syndrome revealing infiltration of mononuclear cells. H&E. Original magnification-400X.



**Fig 11. Section of pancreas of a broiler chicken affected with Ascites syndrome showing vascular congestion. H&E . Original magnification-400X.**

## REFERENCES

- [1]. O David, Norris, and A.C James, *Vertebrate Endocrinology (Overview Of Chemical Bioregulation in vertebrates)*. Academic Press Science 2013).
- [2]. E Danforth, and A. Burger, The role of thyroid hormones in the control of energy expenditure, *Clinics in endocrinology and metabolism*, 13(3), 1984, 581-595.
- [3]. S Bobek, M. Jastrzebski, and M. Pietras, Age related changes in oxygen consumption and plasma thyroid hormone concentration in the young chicken, *General and Comparative Endocrinology*, 31, 1977, 169–174.
- [4]. J Gabarrou, C. Duchump, J. Williams, and P. Geraert, A role of thyroid hormones in the regulation of diet induced thermogenesis in birds, *British Journal of Nutrition*, 78, 1997, 963–973.
- [5]. F McNabb, and R. McNabb, Thyroid development in precocial and altricial avian embryos. *The Auk*, 94, 1981, 736-742.
- [6]. B Freeman, Stress and the domestic fowl: Physiological fact or fancy? *World's Poultry Journal*, 4, 1985, 45-51.
- [7]. D Randall, W. Burggren, and K. French, *Glands and Hormones*. (Animal Physiology: Mechanisms and Adaptations, Freeman and Company, New York. 2002)
- [8]. A Ghosh, S. Carmichael, and M. Mukherje, 2001. Avian adrenal medulla: cytomorphology and function, *Acta Biologica Szegediensis*, 45, 2001, 1-11.
- [9]. R Kumar, S. Kumar, M. Ali, A. Kumar, A. Nath, K. Lawrence, and J. Singh, Impact of stress on histology and biochemical parameters of liver and kidney of Mice. *Innovative Journal of Medical and Health Science*, 2(4), 2012, 63-66.
- [10]. P. D. Sturkie, *avian physiology* ( Springer, Verlag: New York , 1986).

- [11]. T Fitzgerald, *The coturnix quail*( anatomy and histology. Iowa State University Press, Iowa. 1969)
- [12]. E Aughey, and F.L Frye, *Comparative veterinary histology with clinical correlates.*( Manson, London. 2001).
- [13]. M Maxwell, I. Alexander, J. Robertson, M. Mitchell, and C. Corquodale, Identification of tissue hypoxia in the livers of ascitic and hypoxia induced broilers using trypan blue. *British Poultry Science*, 36, 1995, 791–798.
- [14]. R Julian, *ascites in poultry*( *Avian Pathology* 1993).
- [15]. M Maxwell, and G. Robertson, UK survey of broiler ascites and sudden death syndromes in 1993, *British Poultry Science*, 39, 1998, 203–215.
- [16]. R Owen, R. Wideman, R. Leach, B. Cowen, P. Dunn, and B. Ford, Physiologic and electrocardiographic changes occurring in broilers reared simulated high altitude. *Avian Diseases*, 39, 1995, 108–115.
- [17]. E Kuhn, E. Decuyper, and P. Rudas, Hormonal and environmental interactions on thyroid function in the chick embryo and post-hatching chickens, *Journal of Experimental Zoology*, 232, 1984b, 653–658.
- [18]. G Jones, Energy and nitrogen metabolism and oxygen use by broilers susceptible to ascites and grown at three environmental temperatures, *British Poultry Science*, 35, 1994, 97–105.
- [19a]. N Buys, C. Scheele, W. Kwakernaak, and E. Decuyper, Performance and physiological variables in broiler chicken lines differing in susceptibility to the ascites syndrome: 2. Effect of ambient temperature on partial efficiencies of protein and fat retention and plasma hormone concentrations, *British Poultry Science*, 40, 1999a, 140–144.
- [19b]. N Buys, C. Scheele, C. Kwakernaak, and E. Decuyper, Susceptibility to the ascites syndrome: 1. Changes in blood gases as a function of ambient temperature, *British Poultry Science*, 40, 1999b 135–139.
- [20]. L Carew, K. Evarts, and F. Alster, 1998. Growth, feed intake, and plasma thyroid hormone levels in chicks fed dietary excesses of essential amino acids, *Poultry Science*, 77, 1988, 295–298.
- [21]. E Gonzales, J. Buyse, M. Sartori, and E. Decuyper, Metabolic disturbances in male broilers of different strains. 2. Relationship between the thyroid and somatotrophic axes with growth and mortality. *Poultry Science*, 78, 1999, 516–521.
- [22]. S Yahav, Relative humidity at moderate ambient temperatures: its effect on male broiler chickens and turkeys, *British Poultry Science*, 41, 2000, 94–100.
- [23]. S Yahav, S. Goldfeld, I. Plavnik, and S. Hurwitz, Effects of diurnally cycling versus constant temperatures on chickens growth and food intake, *British Poultry Science*, 37, 1996, 43–54.
- [24]. H Klandorf, and S. Harvey, Food intake regulation of circulating thyroid hormones in domestic fowl, *General and Comparative Endocrinology*, 60, 1985, 162–170.

- [25]. S Yahav, S. Goldfeld, I. Plavnik, and S. Hurwitz, Physiological responses of chickens and turkeys to relative humidity during exposure to high ambient temperature, *Journal of Thermal Biology*, 20, 1995, 245–253.
- [26]. S Yahav, S. Goldfeld, I. Plavnik, and S. Hurwitz, Response of turkeys to relative humidity at high ambient temperature, *British Poultry Science*, 39, 1998, 340–345.
- [27]. S Yahav, A. Straschnow, I. Plavnik, and S. Hurwitz, S. Effects of diurnally cycling versus constant temperatures on chickens growth and food intake, *British Poultry Science*, 37, 1996 43–54.
- [28]. L Luna, Manual of Histologic staining methods of the armed forces, (Institute of Pathology NEWYORK, Mc Graw Hill Book Company, New York, 1968).
- [29]. K Balasundaram, and M. Mookkappan, Histomorphology of the thyroid gland in the Domestic Fowl (*Gallus domesticus*), *Journal of Veterinary and Animal Sciences*, 31, 2000, 28 - 31
- [30]. H Stein The volumes of colloid in follicles of normal human (Bantu) thyroid gland with a note on the staining reactions of the colloid, *American Journal of Anatomy*, 66, 1940, 197-200.
- [31]. Beresford, Endocrine system. Lecture notes on histology( Blackwell scientific publications, Asney Mead and Oxford 1983).
- [32]. R Hodges, *the histology of the fowl* (Academic Press, London, 1974).
- [33]. S Enura, I. Isogai, H. Tranloom, Ultrastructure of quail thyroid gland. *Japanase Poultry Science*, 14, 1977, 121-130.
- [34]. P Wight, and D. Shannon The morphology of the thyroid glands of quails and fowls maintained on the diet containing rape seed, *Avian Pathology*, 14, 1985, 383-399.
- [35]. G Colditz, Effects of the Immune system on metabolism: implications for production and disease resistance in livestock. *Livestock Production Science*, 75, 2002, 257-268.
- [36]. K Klasing Metabolic disease: exacerbation of metabolic disease by infectious disease. Program and abstracts. In: XIII Congress of the World Veterinary poultry Association Denver, American Association of Avian Pathologists, Athens, GA, 2003. 54-55.
- [37]. B Humphrey, and K. Klasing, Modulation of nutrient metabolism and homeostasis by the immune system, *World's Poultry Science*, 60, 2004, 90 - 100.
- [38]. D Zikic, G. Uscebrka, D. Gledic, M. Lazarevic, S. Stojanovic, and Z. Kanack, The influence of long term sound stress on histological structure of broiler's adrenal glands, *Journal of Biotechnology Animal Husbandry*, 27, 2011, 1613–1619.
- [39]. M. Lazarevic, D. Zikic, and G. Uscebrka, The influence of long term sound stress on the blood leukocyte count, heterophil/lymphocyte ratio and cutaneous basophil hypersensitive reaction to phytohemagglutinin in broiler chickens, *Acta Veterinaria -Beograd*, 50, 2000, 63-76.

- [40]. I Bedanova, P. Chloupek, P.Vosmerova, J. Chloupek, and V. Vecerek, Time course changes in selected biochemical stress indices in broilers exposed to short-term noise, *Acta veterinaria Brno*, 79, 2010, 35-40.
- [41]. A Tafti, and M. Karima, Morphological studies on natural ascites syndrome in broiler chickens, *Veterinarski arhiv*, 70, 2000, 239 - 250.