

**EFFECT OF DIETARY CRUDE PROTEIN ON AMMONIA  
EMISSION, BLOOD PROFILE AND PRODUCTION  
PERFORMANCE OF BROILER**

**A Thesis**

**By**

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**MASTER OF SCIENCE IN ANIMAL NUTRITION  
DEPARTMENT OF ANIMAL NUTRITION, GENETICS  
AND BREEDING**

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### ***CERTIFICATE***

*This is to certify that the thesis entitled “**EFFECT OF DIETARY CRUDE PROTEIN ON AMMONIA EMISSION, BLOOD PROFILE AND PRODUCTION PERFORMANCE OF BROILER**” submitted to the Department of Animal Nutrition, Genetics and Breeding, Faculty of Animal Science & Veterinary Medicine, Sher-e-Bangla Agricultural University, Dhaka-1207, as partial fulfillment for the requirements of the degree of Master of Science (MS) in Animal Nutrition, embodies the result of a piece of bona fide research work carried out by **MD. TIPU SULTAN**, Registration No.: 14-06209, Semester: **JANUARY-JUNE/2021** under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma. I further certify that any help or source of information, received during the course of this investigation has been duly acknowledged.*

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*Dedicated  
To  
My Beloved Parents*

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## LIST OF ACRONYMS AND ABBREVIATION

ABBREVIATION		FULL MEANING
ANOVA	=	Analysis of variance
Avg.	=	Average
BWG	=	Body weight gain
DP	=	Dressing percentage
e.g.	=	Exempli gratia meaning for example
<i>et al.</i>	=	And others/associates
FC	=	Feed consumption
FCR	=	Feed conversion ratio
Gm	=	Gram
i.e.	=	That is
L	=	Liter
M.S.	=	Master of Science
No.	=	Number
SAU	=	Sher-e-Bangla Agricultural University
SE	=	Standard Error
SPSS	=	Statistical package for social sciences
Viz.	=	Such as
°C	=	Degree Celsius
/	=	Per
%	=	Percentage
±	=	Plus-minus
:	=	Ratio

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## **EFFECT OF DIETARY CRUDE PROTEIN ON AMMONIA EMISSION, BLOOD PROFILE AND PRODUCTION PERFORMANCE OF BROILER**

### **ABSTRACT**

The demand for poultry meat and their products in Bangladesh has grown significantly. Poultry sector of our country needs to increase to meet the growing domestic demand. However, atmospheric ammonia inhibits broiler performance. Therefore, a study was planned to investigate the effect of dietary crude protein on ammonia emission, blood profile and production performance of broiler. A total of 135 Day-Old Lohmann broiler chicks were reared in Sher-e-Bangla Agricultural University Poultry Farm, Dhaka-1207. Chicks were divided randomly into 3 experimental groups of 3 replications R<sub>1</sub>, R<sub>2</sub> and R<sub>3</sub> where each replication contains 15 birds. These three treatment groups were designated as T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub>. T<sub>1</sub> was high CP group containing 23% in starter phase and 22 % in grower phase. T<sub>2</sub> was medium CP group (21% in starter & 20 % in grower) and T<sub>3</sub> was low CP group (19 % in starter & 18 % in grower). Result demonstrated that the average ammonia level in T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> were insignificant (P>0.05) at the end of 1<sup>st</sup> week, however it varied significantly (P<0.05) at the end of 2<sup>nd</sup>, 3<sup>rd</sup> and 4<sup>th</sup> week. Here, T<sub>1</sub> indicates the highest ammonia emissions (ppm) at the end of 2<sup>nd</sup> (6.23<sup>a</sup>±0.03), 3<sup>rd</sup> (8.80<sup>a</sup>±0.05) and 4<sup>th</sup> (11.63<sup>a</sup>±0.17) week and T<sub>3</sub> showed the lowest ammonia emissions at the end of 2<sup>nd</sup> (3.46<sup>a</sup>±0.03), 3<sup>rd</sup> (5.30<sup>a</sup>±0.15) and 4<sup>th</sup> (6.53<sup>a</sup>±0.14) week. At the end of 4<sup>th</sup> week significantly (P<0.05) higher emissions of ammonia was found in T<sub>1</sub> (11.63<sup>a</sup>±0.17) and lower was found in T<sub>3</sub> (6.53<sup>c</sup>±0.14). BWG (Body Weight Gain) and BW (Body Weight) at the end of 4<sup>th</sup> week were significant (P<0.05) in different group. However, better BWG(g), BW(g) and FCR were found in T<sub>1</sub> (BWG 1885.34<sup>a</sup>±33.75, BW 1927.34<sup>a</sup>±33.75, FCR 1.35±0.02). Dressing percentage was significantly (P<0.05) higher in T<sub>1</sub> (70.81<sup>a</sup>±0.76) and lower in T<sub>3</sub> (66.52<sup>a</sup>±0.38). The weight of breast, wing, back and drumstick in T<sub>1</sub> was significantly higher (P<0.05) than T<sub>3</sub>. Survivability of the chicken was insignificantly higher (P>0.05) in T<sub>2</sub> and T<sub>3</sub> than T<sub>1</sub>. In case of benefit cost ratio analysis was insignificantly (P>0.05) higher in T<sub>1</sub> (1.35±0.02) and T<sub>2</sub> (1.35±0.01) than T<sub>3</sub> (1.33±0.02). The immune parameter (Hemoglobin, WBC, RBC) was significantly higher (P<0.05) in T<sub>2</sub>. The results of this experiment recommend that using the low CP concentrations may reduce performance therefore may not be advisable. On the other hand, high CP % can produce high NH<sub>3</sub> gas production which is detrimental. It should be keep in equilibrium state between high dietary CP supplementation and NH<sub>3</sub> gas production. Indeed, as subsequent further research is needed to identify the most accurate findings.

# CHAPTER I

## INTRODUCTION

### 1.1 General Background

Poultry farming has emerged as one of the fastest growing agribusiness industries in the world, even in Bangladesh. Research on meat production globally indicates poultry as the fastest growing livestock sector especially in developing countries. It is of utmost significance for the livestock industry to benefit from inexpensive, highly efficient, and safe diets. Ammonia ( $\text{NH}_3$ ) is generated in poultry houses by microbial degradation of poultry waste and can reach levels that limit poultry performance (Huff *et al.*, 1984). Ammonia volatilization has many sources including nitrogen components such as uric acid, urea, ammonia/ammonium and undigested protein. Important factors in this process include the dry matter and acidity of excreta and temperature in the house (Leenstra and Pit, 1990; Nahm, 2003). It is possible, at least in theory, to lower the rate and equilibrium ammonia gas concentration by manipulating either the source or the process of ammonia production.

A numbers of researches have studied different strategies to control ammonia including adequate ventilation and careful litter management (Reece *et al.*, 1979; Hartung and Phillips, 1994), and the addition of chemicals to the litter such as formaldehyde (Veloso *et al.*, 1974), zeolites (Nakaue *et al.*, 1981). All these tactics may gain widespread acceptance, but they impose an additional cost upon producers. Reducing nitrogen emission is the most efficient and the cheapest method of controlling ammonia production. To achieve this target, the nitrogen content of diet must be decreased. Attempts to reduce crude protein (CP) content of broiler diets have been successful up to a point, but most researchers agree that reduction in CP has noxious effects on performance. Inclusion of low crude protein content in poultry diets can be considered as one of the effective strategies to reduce ammonia emission from poultry wastes, environmental impact, and diet costs (Belloir *et al.*, 2017 and Barekatain *et al.*, 2019). However, such diets can result in negative consequences, including poor growth performance and carcass yield of broilers (Bregendahl *et al.*, 2002; Liu *et al.*, 2017; Allameh and Toghyani, 2019).

The negative effect of a low protein diet on chickens' growth performance might be attributed to the effects of protein deficiency on intestinal morphological changes (Yu *et al.*, 2019). Therefore, nutritionists have developed strategies to lessen the negative effects of the low CP diets on broiler chickens' growth performance (Kermani *et al.*, 2017; Goodarzi Boroojeni *et al.*, 2018 and Sharifuzzaman *et al.*, 2020). Essentially, dietary requirements for protein are requirements for amino acids. Therefore, dietary formulations based on the amino acid requirements of birds rather than CP can minimize N excretion by simply reducing total dietary N intake (NRC, 1994). Adequate intake of dietary amino acids is required by birds to achieve optimum performance measured in terms of growth, feed conversion ratio (FCR) and carcass quality (Bryden & Li, 2004). Dietary supplementation of amino acids can be used to reduce dietary CP content and N excretion. Waldroup *et al.* (1976) reported that minimizing excess dietary amino acids resulted in improved performance of broiler chicks. Corzo *et al.* (2004) also demonstrated improvements in nitrogen utilization in terms of plasma uric acid, dietary nitrogen intake, excretion and retention without deleterious consequences on live performance of chicken when low crude protein diet was fed to broilers.

## **1.2 State of the problems**

The NH<sub>3</sub> concentration in broiler house is a major concern of this modern poultry industry. Excess NH<sub>3</sub> in broiler house is frequently claimed for growth retardation, ascites, conjunctivitis and poor feed utilization. Therefore, farmers are trying to mitigate the NH<sub>3</sub> burden in broiler house by using some chemical compounds available in the market.

Ammonia emissions in poultry houses are mainly due to high protein formulated chicken diets. The chickens have no storage mechanisms for amino acids consumed beyond the requirement for protein synthesis, so the excess amino acids are deaminated and the derived nitrogen is excreted in the urine mainly as uric acid (80%), ammonia (10%) and urea (5%). High levels of ammonia in a poultry house have negative impact on poultry growth particularly at an early age. So it is very important to maintain ammonia level on broiler farming. Feed conversion and weight gains in poultry are also affected by high levels of ammonia. Not only does indoor ammonia pollution affect the chickens but also pose a risk to the health of the farmer in these facilities. Ammonia

gas has a characteristic pungent odor. High ammonia levels in broiler houses can reduce bird performance and increase susceptibility to disease and increase subsequent mortality.

Thus we can easily understand the harmful effect of ammonia to broiler, farmers & environment. The current study is to supply somewhat at a level that will generate a certain accepted concentration of NH<sub>3</sub> related with growth performance and economic aspects in broiler farming.

### **1.3 Justification of the study**

Reducing dietary crude protein (CP) in broiler diets has been advantageous in decreasing dietary cost, nitrogen excretion (Hernandez *et al.*, 2012), ammonia emissions (Ferguson *et al.*, 1998), and the incidence of pododermatitis (Nagaraj *et al.*, 2007). However, previous research reported that reduction of dietary CP content beyond 2.0% points may result in sub-optimum body weight (BW) gain and feed conversion ratio (FCR) of broilers (Waldroup *et al.*, 2005; Dean *et al.*, 2006; Namroud *et al.*, 2008; Hernandez *et al.*, 2012). Strategies to mitigate poor growth performance of broilers include potassium supplementation (Han *et al.*, 1992), the inclusion of Glu or Asp as a source of nitrogen (Aletor *et al.*, 2000), and increasing dietary energy (Hussein *et al.*, 2001). However, these approaches have produced inconsistent results (Han *et al.*, 1992; Aletor *et al.*, 2000; Hussein *et al.*, 2001). Adverse growth responses of broilers fed reduced-CP diets may be attributed to sub-optimum essential amino acid (AA) concentrations (Aftab *et al.*, 2006). When reducing dietary CP content in gradient increments, less limiting AA concentrations beyond Methionine, Lysine, and Threonine may be lower than diets with a higher CP content. Waldroup *et al.* (2005) observed a 3.1% increase in FCR when lowering dietary CP content from 22 to 19% in 28-day-old broilers. If we implement this study successfully we hope every people in our country will able to meet the requirement of broiler meat by increasing production.



## **1.4 Objectives**

From the above consideration, the present study was under taken to determine the effect of different level of dietary crude protein with the following specific objectives:

1. To assess the ammonia concentration level in broiler house;
2. To find out blood profile of broiler for immunity checking;
3. To evaluate the growth performance of broiler;
4. To estimate the economic impact in broiler rearing.

## CHAPTER II

### REVIEW OF LITERATURE

It is very important to review the past research works which are related to the proposed study before conducting any type of survey or experiment. The literatures reviewed here have been limited to those which are considered pertinent and related to the objectives of the present study. Ammonia is considered the most harmful gas in broiler chicken housing. It can cause environmental problems, which is detrimental to the health and performance of birds. However, some of the important and informative works and research findings related to this study have been reviewed in this chapter under the following headings.

#### **2.1 Present status of poultry farming in Bangladesh**

According to DLS (2020-21) there are 3041.06 lakh chicken, 3658.52 lakh poultry 84.40 Lakh Metric Ton meat production 2057.64 Crore Egg. Contribution of Livestock in Gross Domestic Product (GDP) (Constant Prices) 1.44% GDP growth rate of Livestock (Constant Prices) 3.80 % GDP volume (Current prices) (Million Taka) 50301 Share of Livestock in Agricultural GDP (Current prices) 13.10% Employment (Directly) 20% Employment (Partly) 50%.

WPSA-BB (2017) Within four decades, investment in poultry sector has spiked from 179 million USD to 3.573 billion USD. Around 2.5 million direct and another 3.5 million indirect jobs were created, where women account for 40 per cent of the total workforce. Bangladesh Economic Review reported that there are about 75,902 poultry farms up to February 2021 in the country (BER, 2021). Daily Star (2013) reported that in two years since 2011, nearly 25,000 farms were closed mainly due to the outbreak of the diseases.

Raha (2013) reported that 6 Grand Parent farms which supply 80% of the total demand for parent stock and rest 20% are imported. In the country 82 parent stock farms are operating and of producing 55-60 lakh DOC of broiler and 5 lakh Layer DOC per week.

Daily star (2017) reported that there are 300 billion takas has been invested in the poultry industry. There is an estimated 150,000 poultry farms in Bangladesh.

Daily star (2017) also reported that the farms annually produce 570 million ton of meat and 7.34 billion eggs. Hossain *et al.* (2019) reported that total 198 feed mills are registered from DLS in Bangladesh up to 27 September 2018, which are involved in producing commercial poultry and cattle feed. Among 198 feed mills 96 are actively producing and marketing poultry and cattle feed. Based on internal estimates, current demand for poultry feed has been estimated to be 5.08 million MT/year (Fuad, 2017). WPSA-BB (2017) currently reported that 6 grandparent farms producing about (60 - 70) Thousand parent stocks per week and 140 parent stock farms producing 10.1 Million Day Old Chicks (DOC) per week. About 100,000 commercial farms produce over 15,000 MT of broiler meat and 2.4 Million eggs per week.

News today (2018) reported that there are 16 grandparent farms are producing about 6070 thousand parent stocks per week and 206 parent stock farms are producing 13 million Day Old Chicks (DOC) per week. About 100,000 (registered farms no 80,802) commercial farms produce over 15,000 MT of broiler meat and 2.4 million eggs per week. Annual per capita consumption of eggs in the country is 94 against the minimum requirement of 104 eggs. Per capita broiler meat consumption is 3.74 kg and the share of broiler meat out of total meat consumption is 54 percent. It is expected that per capita poultry meat consumption to be reached 8.42 kg in 2021 and contribution of poultry meat could increase to 78 percent. Besides commercial poultry, the growth of dairy industry has not been achieved a remarkable stage. DLS (2021) reported that there are 19 grandparent stock, 218 parent stock, 19801 layer farms and 56561 broiler farms in Bangladesh. Raha (2013) reported that based on poultry industry a number of industries are developed both in inputs sector and outputs sector along with a number of service providing organizations where at least 60 lakh people are involved.

## **2.2 Effect of ammonia on performance of broiler**

Ammonia has adverse effect particularly to the nasal cavity and eyes of affected chickens due to the gas's alkalinity and corrosiveness. When in contact with nasal moisture,  $\text{NH}_3$  reacts with the moisture and forms a corrosive basic solution. The

aqueous ammonium solution formed corrodes the respiratory lining of the affect chicken consequently weakening immunity in the respiratory system making the animal susceptible to bacterial infections especially E. coli. The corrosion of the respiratory lining ranges from cilia paralysis to complete loss of cilia of the epithelial cells. Cilia are hair like structures that are found on the trachea lining. Cilia clusters form the mucociliary apparatus that is mainly responsible for cleaning up the upper respiratory system of the chickens. The mucociliary apparatus removes dust particles by trapping and preventing them from reaching the lower respiratory system (air sacs). This happens by the use of propulsive action of the cilia where the dust particles are entrapped by mucus and propelled out of the respiratory system (Aziz and Barnes, 2010). When cilia are corroded or paralyzed by high NH<sub>3</sub> concentrations, mucus on the mucosal surface of the trachea cannot be cleared and thus entrapped bacteria on dust particles may reach the lungs and air sacs and cause infections (Aziz and Barnes, 2010). The most common bacterial diseases in chickens are pneumonia, septicemia and airsacculitis. In addition to respiratory diseases, high NH<sub>3</sub> concentrations have an effect on chicken eyes. Atmospheric NH<sub>3</sub> at high concentrations cause conjunctivitis (inflammation of conjunctiva) and damages the cornea of the eyes (Aziz and Barnes, 2010). The cornea is affected due to corneal erosion caused by NH<sub>3</sub> corrosion. High levels of ammonia in a poultry house have negative impact on poultry growth particularly at an early age. (Moore *et al.*, 2008), found that NH<sub>3</sub> levels as low as 20 ppm compromise the immune system of chickens, making them more susceptible to diseases and damage to respiratory system. Feed conversion and weight gains in poultry are also affected by high level of ammonia.

Miles *et al.* (2004), conducted a test for atmospheric ammonia is detrimental to the performance of modern commercial broiler. Quantified effects are based on older genetic stock with a BW of 2,000 g at 7 wk. In contrast, modern genetic stock reaches 3,200 g at 7 wk. of age. To assess the impact on present day broilers, 2 trials were conducted exposing male broilers to graded levels (0, 25, 50, and 75 ppm) of aerial ammonia from 0 to 4 wk. of age. Final BW was significantly depressed by 6 and 9% for the 50 and 75 ppm concentrations of ammonia as compared with 0 ppm. Also, mortality was significantly greater at the 75 ppm ammonia concentration, 13.9% compared with 5.8% for the 0 ppm treatment.

An experiment was conducted Nemat *et al.* (2011), to examine the effect of three levels of crude protein (CP) and three levels of Ca and available P on performance of broilers from hatching until 21 days of age. Results of the experiment showed that CP content had no significant effect on feed and water intake. However, body weight gain (BWG) significantly reduced and FCR increased ( $P < 0.05$ ) by 15% change in CP content of diet. Fifteen percent increase in mineral content of diets had no significant effect on feed intake. However, increase in Ca and Av. P significantly increased BWG and resulted in an improved feed conversion ratio (FCR) and increased water intake. There was an interactive effect of CP by Ca and Av. P levels on feed intake, BWG and FCR. A change in CP or mineral content of the diets had no significant effect on blood parameters except for potassium concentration.

### **2.3 Method for measuring ammonia emission from poultry house**

Reeves *et al.* (2002), test the ability of various available quick tests to determine ammonia concentration of poultry litters. A total of 136 samples was collected from brood chambers of poultry houses. Samples were equally divided between surface samples (top 25 mm) and core samples. Samples were frozen until analysis but received no further processing. Samples were analyzed for ammonia by auto analyzer (standard) and several quick tests (conductivity, Quantofix N-Volumeter, and Reflectquant). In addition, samples were analyzed by near-infrared spectroscopy by scanning samples using a large-sample transport device on a FOSSNIRS systems model 6500 (64 co-added scans from 400 to 2,498 nm). Results showed that, although ammonia could be determined with reasonable accuracy by near-infrared spectroscopy using data in the 1,100 to 2,498 nm spectral range, none of the quick tests, including near-infrared, worked as well as previously found with dairy manures. The best results were found using the Quantofix or Reflectoquant and conductivity worked only with the core samples. It is believed that interferences due to the presence of uric acid (spectroscopy, Quantofix, and Reflectoquant) and sodium bisulfate used to treat the litter (conductivity) were the cause of the decreased accuracies as compared to results achieved previously with dairy manures.

Ammonia emissions were measured by Moore *et al.* (1996). The small plastic containers were equipped with air inflows and outflows. Ammonia-free air was

continually passed through each chamber for 42 d per experiment, and any NH<sub>3</sub> volatilized from the litter was trapped in 2 consecutive traps containing 80 mL of boric acid solution that was titrated daily for NH<sub>3</sub> content with 0.1 N of HCl. Ammonia free air was generated by passing compressed through 2 consecutive 3-L acid traps containing 1 M HCl. After passing through the acid traps, the air was passed through 2 consecutive water traps, to remove any acid vapors. During experiment 2, the 2 water traps dried out for several days during the middle of the study, which may have resulted in acid fumes from the acid traps interacting with the litter.

#### **2.4 Effect of dietary protein on growth performance of broiler**

Several studies reported the inferior effects of low dietary protein on broiler growth performance (Hernandez *et al.*, 2012; Folorunso *et al.*, 2014 and Liu *et al.*, 2017). In the same vein, Allameh and Toghyani (2019) concluded that the weight of broiler chickens significantly decreased when the dietary CP reduced from 20.4% to 17.9%. Moreover, they concluded that feeding 85% of CP requirements had negative effects on the daily weight gain of broilers. Recently, Hilliar *et al.* (2020) and Macelline *et al.* (2020) concluded that birds were fed low protein diets had a low BWG, compared to chicks that were fed high protein diets. Reducing dietary CP below a minimum level even with maintained EAA level, depressed BW, FCR and feed intake. Other authors have reported similar results, confirming that reduction in CP during the grower period (10–28 days of age) could possibly reduce BW and feed intake (Ferguson *et al.*, 1998; Si *et al.*, 2004; Waldroup *et al.*, 2005; Yamazaki *et al.*, 2006).

The production of ammonia originated from poultry manure negatively impacts animal health and well-being as well as source of greenhouse gas (Ritz *et al.* 2004; Steinfield *et al.* 2006). Numerous approaches have been used in feed formulation for reducing N excretion (Namroud *et al.* 2008). For instance, low protein diet positively reduced N excretion when used in broiler diets (Hernandez *et al.* 2012). In quails, the dietary supplementation of crude protein (CP) ranging from 20 to 22% had significantly reduced N excretion without growth retardation (Omidwura *et al.* 2016). In contrast, reduced CP diet had minimal effect on the growth performance and feed efficiency of growing-meat birds (Soares *et al.* 2003). The difference might be due to the environment, nutrition, and genetic factors. Jacob *et al.* (1994) showed that the level of

N excreted in poultry waste can be reduced by up to 21% if dietary CP content is lowered by 2.5% and the diet is supplemented with synthetic amino acids.

The study by Kumari *et al.* (2016), was conducted to evaluate the effect of various levels of protein, in the diets of WLH layers, on production performance and on nitrogen (N) and phosphorus (P) levels in the excreta. Results demonstrated that egg production, egg weight and feed conversion ratio (FCR) were not influenced by the level of protein in diet. Body weight was increased with the increase of protein levels in diet. Gain over feed cost compared to control (17% CP) was recorded as \$1 and \$ 0.67 in low (13.38%) and medium (15.58%) protein groups, respectively. The percentage of N was increased and percentage of phosphorus was decreased significantly.

The study by Saleh *et al.* (2021), to evaluate low protein diets with amino acid supplement on growth, biochemical markers and muscle amino acids profile in broilers under high ambient temperature. Control fed optimal protein and optimal amino acids which contains 23% and 21% crude protein (CP) with 65% methionine + cysteine/lysine (Met + Cys/Lys) and 55% threonine/lysine (Thr/Lys), LPOA (low protein and optimal amino acids) which contains 21% and 19% CP with 65% Met + Cys/Lys and 55% Thr/Lys and 3. LPHA (low protein and high amino acids) which contains 21 and 19% CP with 74% Met + Cys/Lys and 67% Thr/Lys. Birds fed LPOA diets significantly highest body weight, while those fed LPHA recorded significantly the lowest body weight (BW). Dressing percentages not revealed significantly affected by reducing dietary protein levels, while the blood plasma total protein, albumin, and globulin were not significant differences due to dietary low protein. Chicks fed LPHA diets recorded the highest liver content of malonaldehyde. It concluded that feeding the Cobb 500 broilers on low protein diets with the same amino acid levels had no adverse effect on growth, carcass markers, and liver function, however increased amino acids levels to low protein diets may led negative impacts for the broiler performance under high ambient temperature.

The study by Nikoletta *et al.* (2021), was to investigate the effects of feeding low protein (LP) diets on the performance parameters and excreta composition of broiler chickens. The protein reduction in the grower and finisher phases were 1.8% and 2% respectively.

Beside the measurements of production traits, on day 24 and 40 representative fresh excreta samples were collected, their dry matter, total N,  $\text{NH}_4^+$ , N and uric Acid-N contents determined, and the ratio of urinary and fecal N calculated. Dietary treatments failed to cause significant differences in the feed intake, growth rate, and feed conversion ratio of animals. LP diets decreased the total nitrogen and uric acid contents of excreta significantly. The age of birds had also significant effect, resulting more reduction in the grower phase compared with the finisher. The ratio of urinary N was higher at day 40 compared with the age of day 24. The urinary N content of broiler chicken's excreta is lower than can be found in the literature, which should be considered in the ammonia inventory calculations.

The research by Latshaw *et al.* (2011), to determine the extent to which the CP content of laying diets can be reduced, based on performance criteria and to determine how ash: nitrogen ratios of manure, eggs, and hens are affected by dietary protein changes. Egg production averaged approximately 90%, and daily protein intake caused no effects on egg production or grams of egg per hen per day. Feed intake was higher for hens fed 13 g of protein than for hens in the other 2 treatments ( $P < 0.01$ ). Average feed intake for the experiment was approximately 95 g/d. Composition of the eggs was not affected by protein intake.

This study by Emous *et al.* (2019), determined the effects of different dietary crude protein (CP) levels on ammonia ( $\text{NH}_3$ ) emission, litter and manure composition, nitrogen (N) losses, and water intake in broiler breeders. Dietary protein level did not affect water intake and dry matter (DM) content of the litter or manure. Compared to birds fed the CP1 diets, the litter and manure samples of broiler breeders fed the CP2 had 8% lower Total-N and 13% lower Ammonia-N content resulting in a 9% lower ammonia concentration, 9% lower ammonia emission, and 11% lower total N losses. In conclusion of the study showed that reducing CP level in the diet of broiler breeders reduces ammonia emission and total N-losses from litter and manure.

An experiment was conducted by Kriseldi *et al.* (2006), to determine the effects of feeding reduced crude protein (CP) diets to male broilers while maintaining adequate essential amino acid (AA) concentrations on growth performance, nitrogen excretion, and plasma uric acid (UA) concentration during the starter period. In trial 1, 11 dietary



treatments were fed from 1 to 18 d of age containing 1.20% digestible Lys. Diet 1 (23.2% CP) was formulated with DL-Met, L-Lys, and L-Thr to contain 1.70 total Gly + Ser to digestible Lys ratio whereas diets 2 (23.4% CP) to 11 were formulated with additional Gly to contain 1.90 total Gly + Ser to digestible Lys ratio. Free AA were added sequentially in the order of limitation (L-Val, L-Ile, L-Arg, L-Trp, L-His, L-Phe, and L-Leu) from diets 3 to 10 to decrease CP content from 22.6 to 18.8%, respectively. In diet 11, L-Gln was added to increase the CP content to 23.4%. Feed conversion of broilers fed diet 2 was lower ( $P < 0.05$ ) than those consuming diets 6 to 11 from 1 to 17 d of age. Nitrogen excretion (mg/b/d) decreased ( $P < 0.001$ ) by 14.1% when broilers were fed diet 4 compared with birds fed diet 2 from 15 to 16 d of age. Broilers fed diet 4 had lower ( $P = 0.011$ ) plasma UA concentration than birds fed diet 2 at 18 d of age. In trial 2, 8 dietary treatments containing 1.25% digestible Lys and 1.70 total Gly + Ser to digestible Lys ratio were fed from 1 to 21 d of age. Diet 1 (24.0% CP) was supplemented with DL-Met, L-Lys, and L-Thr. Free AA (L-Val, Gly, L-Ile, L-Arg, L-Trp, L-His, and L-Phe) were sequentially supplemented in the order of limitation to decrease CP content in diets 2 to 8 from 23.8 to 20.3%. Broilers fed diet 1 had higher ( $P < 0.05$ ) body weight gain and lower ( $P < 0.05$ ) feed conversion when compared with diet 7 or 8. Plasma UA concentration of broiler provided diets 4 to 8 was lower compared with diet 1 at 21 d of age. Placing a minimum on dietary CP percentage may not be necessary when proper AA ratios are implemented in diet.

A study was conducted by Namroud *et al.* (2008), in a completely randomized design to evaluate the performance, excreta characteristics, and some blood nitrogen metabolite concentrations of 28-d-old male broilers fed 4 experimental diets in which CP was decreased in a stepwise manner from 23 to 17%. The other 4 diets were formulated to have 19 and 17% CP, in which 2 of them contained an additional 10% of particular essential amino acids (EAA) and 2 were supplemented with Gly and Glu. Digestible quantities of all EAA were almost equal in the diets, and total amount of each EAA was maintained at or above NRC requirements. Decreasing dietary CP below 19% depressed performance and appetite and increased fat deposition in the whole body and abdominal cavity significantly. Adding the Gly and Glu mixtures to low-CP diets improved performance and decreased fat deposition. Uric acid, moisture, and acidity of excreta were decreased by reduction of dietary CP; excretory ammonia level was

increased in 17% CP diets. Blood ammonia level was increased and plasma uric acid was decreased with reduction of CP to 17%. Supplementing Gly and Glu increased plasma and excretory uric acid level in spite of decreasing blood ammonia concentration. The amino static hypothesis cannot explain the sharp reduction in appetite in this experiment, because alteration of dietary CP had no significant influence on most plasma free amino acid levels. Therefore, reduction of CP to 19% not only does not impair performance but also decrease nitrogen, ammonia, and pH of excreta that may improve upon litter and air quality. Adding large amounts of crystalline EAA to diets with low intact CP increased blood and excretory ammonia concentration, which due to its negative effects on tissue metabolism may be the main cause of retarded growth and appetite in decreased CP diets below 19%.

The experiment was conducted by Boontiam *et al.* (2019), to evaluate the effect of dietary crude protein (CP) levels and stocking density on growth performance, nutrient retention, blood metabolites, and carcass weight of growing-meat quails under high ambient temperature. 3 levels of CP (20, 22, and 24%) were in the experiment. 20% CP level significantly increased CP digestibility and decreased uric acid concentration compared to 24% CP.

A study was conducted by Namroud *et al.* (2009), in a completely randomized design to evaluate the effects of providing almost all important essential amino acids (EAA) in low-crude protein (CP) diets equal to that of higher CP diets in broiler chickens. Also the effects of additional mixture of glycine (Gly) and glutamic acid (Glu) or supplementation of excess EAA to low-CP diets on the live performance and excreta characteristics including pH, moisture, nitrogen, uric acid and ammonia concentration were measured to ascertain the optimum CP concentration for the maximum performance and reduced excreta ammonia concentration. Male broiler chickens growing from 10 to 28 days of age were fed eight experimental diets. Reducing dietary CP below 19% negatively affected performance. Adding the Gly and Glu mixtures to 17% CP diets improved live performance. Reducing CP to 19% with a normal amino acids status declined N, ammonia, uric acid, moisture and pH of excreta significantly. These findings suggest that diminishing dietary CP from 23% to 19% while maintaining adequate EAA levels during 10–28 days of age results in not only a significant decline

in N emission, but also a probable reduction in the NH<sub>3</sub> volatilization because of reduction in pH and moisture. Contrary to expectations, reduction of dietary CP below the minimum level (19%) resulted in more ammonia. All these factors may improve on litter and air quality within the housing facility and reduce the ventilation rate required to emit the elevated ammonia gas concentrations.

The study by Malomo *et al.* (2013), assessed the effects of dietary crude protein on performance and nitrogen economy of broilers. Chicks were allotted to 22, 20, 18 and 16% crude protein corn-soy diets in a completely randomized design for forty-two days. Several essential amino acids were observed to be deficient compared to recommendations for broiler chicks as the dietary crude protein level reduced. Feed intake, weight gain, feed to gain ratio, fecal nitrogen, nitrogen retention, anthropogenic potential and dressing percentage were influenced ( $P < 0.05$ ) by the dietary treatments. However, mortality was not significantly different ( $P > 0.05$ ). Feed intake, weight gain, nitrogen intake and output were significantly ( $P < 0.05$ ) depressed as the CP level reduced. However, the best ( $P < 0.05$ ) nitrogen retention and percentage fecal nitrogen was recorded for broilers fed 20% crude protein diet. Serum total protein, albumin, uric acid, creatinine and glucose were affected ( $P < 0.05$ ) across treatments. It was concluded that there is a limit to which dietary crude protein of broilers could be reduced without any detrimental effects on the performance and nitrogen economy of the birds, even when the requirements for methionine and lysine has been met, as several other amino acids could be limiting. Consequently, to achieve significant improvement in nitrogen economy and reduction in amount of fecal nitrogen, 20% crude protein diets could be fed to broilers. However, there may be need to further manipulate the amino acid profile of the diet so as to improve its performance to be at par with higher crude protein diets.

An experiment was conducted by Ferguson *et al.* (1998), to determine the effect of diets with reduced CP and supplemental amino acids on broiler performance, N excretion, litter characteristics, and equilibrium NH<sub>3</sub> gas concentration. Results suggest that reducing CP (and lysine) below 241 g/kg (13.7 g/kg lysine) in the diets fed during the first 3 wks. may slightly increase feed: gain and therefore may not be advisable. During the period 22 to 43 d of age there were no significant differences in weight gain and BW at 6 wks. of age when reducing CP from 215 g/kg (11.5 g/kg lysine) to 196 g/kg

(11.3 g/kg lysine), but feed intake and feed: gain ratio increased. However, reducing CP did cause equilibrium NH<sub>3</sub> gas concentration and litter N to decline by 31 and 16.5%, respectively. Both of these advantages will improve air quality within the housing facility and possibly reduce heating costs during winter associated with higher ventilation rates required to reduce elevated NH<sub>3</sub> gas concentrations.

An experiment was conducted by Ferguson *et al.* (1998), to determine whether broiler litter concentration of N and P and equilibrium NH<sub>3</sub> gas concentration can be reduced by reducing dietary CP and P levels and supplementing with amino acids and phytase, respectively, without adversely affecting bird performance. Equilibrium NH<sub>3</sub> gas concentration above the litter was measured. The experiment was divided into a starter period (1 to 21 d) and grower period (22 to 42 d), each having two different CP and P levels in a 2 x 2 factorial arrangements. The CP treatments consisted of a control with a mean CP of 204 and 202 g/kg for starter and grower periods, respectively, and a low CP diet with means of 188 and 183 g/kg, respectively, but with similar amino acid levels as the control. Reducing starter diet CP by 16 g/kg reduced weight gain by 3.5% and, hence, body weight at 21 d of age, but did not affect feed intake or feed efficiency. Reducing P did not affect feed intake and weight gain, but improved feed efficiency by 2.0%. Responses in feed intake and efficiency to CP depended on the level of dietary P. For the grower period there were no significant differences in feed intake, weight gain, and feed efficiency, nor in body weight at 42 d of age, after correcting for 21-d body weight, between CP and P treatments. There were significant (P<0.001) reductions in litter N and P concentrations, but not equilibrium NH<sub>3</sub> gas concentration, moisture content, or pH, for low CP and P diets.

Shlomo YAHAV *et al.* (2004) found the effects of various atmospheric ammonia concentrations at high ambient temperature (Ta = 32 °C) and relative humidity (rh = 60– 65%) on the performance and thermoregulation of male broiler chicken. Body weight declined significantly and proportionally with increasing ammonia concentration in the air. The decline in body weight coincided with a similar pattern in feed intake. Feed efficiency did not differ significantly among treatments, but the broilers exposed to the lowest concentration of ammonia showed the highest feed efficiency. Arterial pH increased with increasing ammonia concentration and was

similar in the upper 2 concentrations of ammonia. A similar but opposite trend was found in the partial pressure of arterial CO<sub>2</sub>. It can be concluded that exposure to increased ammonia concentrations impairs broiler performance.

Lemme *et al.* (2019), In the light of recently revised German legislation on on-farm nitrogen (N) management, the impact of decreasing dietary N-load at balanced dietary amino acid supply on growth performance and N-excretions was examined. In addition to diets representing the German standard with 22.0, 20.6, 20.0, and 19.5% crude protein (CP) in starter (1–10 d), grower I (11–16 d), grower II (17–30 d), and finisher feed (31–40 d), a N-reduced German standard (21.0%, 20.0%, 19.6%, and 18.9% CP) was fed in treatment 2. Dietary CP was further reduced in treatment 3 (21.0%, 19.5%, 18.7%, and 18.0% CP) and treatment 4 (21.0%, 19.0%, 18.0%, and 17.0% CP). Growth performance, feed conversion (FCR), carcass quality, N-utilization and litter quantity (at termination) were examined. Final body weight was similar between treatments 1, 2, and 3 but slightly impaired in treatment 4 ( $P < 0.05$ ) while FCR was not affected. Carcass and breast meat yield did not differ significantly between treatments in male broilers. In female broilers, carcass yield was similar between treatments 1, 2, and 3 whereas it was lower in treatment 4 ( $P < 0.05$ ). Breast meat yield of female broilers did not differ between treatments ( $P > 0.05$ ). Dietary N-reduction resulted in gradually improved N-utilization ( $P < 0.05$ ) resulting in reduction of N-excretion ( $P < 0.05$ ). Moreover, quantity of litter at termination of the trial gradually decreased with dietary protein while dry matter content of litter increased and N-content decreased ( $P < 0.05$ ).

## **2.5 Effect of dietary protein on hematological and immunological parameters**

The results of the study related to the effect of the CP level on the blood parameters were in accordance with the findings of Swennen *et al.* (2005), Kamran *et al.* (2010), and Mohamed *et al.* (2012) concluding that the level of dietary protein did not alter glucose concentration.

In addition, Alam *et al.* (2004) and Mohamed *et al.* (2012) indicated that the dietary protein level had no significant effect on the hemoglobin concentration of broilers. In addition, Mohamed *et al.* (2012) stated that the dietary protein caused only the globin section of the hemoglobin to increase with no effect on the haem section. A low level of albumin is related to the glycation of plasma proteins and HbA1c (Bhonsle *et al.*,

2012). According to Honma *et al.* (2017), protein levels in chicken diets could modify blood albumin levels and low dietary protein could increase the glycated amino acids in chicken plasma. Hemoglobin A1c reflects non-enzymatic glycosylation and fructosamine examined for glycation, associated primarily with albumin (Anguizola *et al.*, 2013). The *in vivo* experiment conducted by Bhonsle *et al.* (2012) indicated that blood albumin could control the plasma protein glycation. Moreover, Tiwari *et al.* (2015) found an inverse correlation between plasma albumin and HbA1c. Reduction of dietary protein in broiler diets not only reduces n-emissions but is also accompanied by several further benefits.

Another study was conducted by Mamdouh Omar *et al.* (2020), to investigate the effect of dietary protein levels and citric acid on the growth performance, carcass yield, abdominal fat, chemical composition of meat, intestinal morphology, and blood parameters of broiler chickens. The results showed that chickens fed the diet containing 100% required Crude Protein (CP) supplemented with citric acid which could significantly improve body weight gain, feed conversion ratio, carcass yield, abdominal fat, fat content in meat, intestinal morphology, cecal microbial content, and blood parameters (Albumin, hemoglobin, fructosamine and cholesterol). Chickens fed the low CP diet supplemented with citric acid could compensate for the growth performance equivalent to those fed the optimal CP diet. Both required protein level and citric acid were significantly improved blood albumin and reduced hemoglobin and fructosamine, which could serve as indicators of the blood protein glycation. In conclusion, citric acid addition could alleviate the negative effect of feeding broiler chickens on low CP diets through its beneficial impact on intestinal morphology, cecal bacterial counts, blood cholesterol reduction, and glycated proteins.

## **2.6 Research gap and scope of present investigation**

From the above review of the literature, it is clear that a few of works had been done on using different dietary crude protein to measure ammonia production and growth performance. To the author's knowledge, few papers had published about the effect of dietary crude protein on ammonia emission, blood parameters and growth performance of broiler.

Therefore, On the basis of previous research, the current study has been undertaken to investigate the effect of crude protein on NH<sub>3</sub> gas emission with production performance, blood parameters, carcass quality and economical utility in broiler rearing. The author also expect that Bangladeshi farmers will be benefited by the application of proper dietary crude protein in the farm at proper concentration level.

## CHAPTER III

### MATERIALS AND METHODS

#### 3.1 Statement of the experiment

The research work was conducted at Sher-E-Bangla Agricultural University, Poultry Farm, Dhaka, with 135-day-old chick for a period of 28 days from 17<sup>th</sup> June to 15<sup>th</sup> July, 2021 for a period of 4 weeks using different dietary crude protein. The experiment was performed by applying different concentration levels of protein to assess the ammonia emission, blood profile and production performance of broiler chickens.

#### 3.2 Collection of experimental broiler

A total of 135-day old chicks of “Lohman Meat (Indian River)” strain having  $42\pm 0.2$ g average body weight were collected from Kazi farm limited hatchery, taraganj, Savar, Dhaka.

#### 3.3 Experimental materials

The collected chicks were carried to the university poultry farm. They were kept in electric brooders equally for 7 days by maintaining standard brooding protocol. During brooding time only basal diet was given no additional protein was used as treatment. After seven days, 135 chicks were randomly selected from brooders and distributed for dietary treatments of different concentration of protein. For proper handling and data collection, the chicks of each treatment group were divided into three replications and in each replication of dietary treatment, there were 15 birds. After 28 days of nursing and feeding, data were collected for the following parameters: feed intake, live weight, body weight gain, feed conversion ratio, carcass characteristics, total blood count, profit per bird and benefit-cost ratio.



### 3.4 Experimental treatments

The market available kazi starter and grower ready feed was used as medium CP. Maize and soybean meal was mixed properly with commercial dietary feed to gain high CP and low CP. The experimental treatments were followings:

T<sub>1</sub>: Diet with high CP %

T<sub>2</sub>: Diet with medium CP % (Control)

T<sub>3</sub>: Diet with low CP %

**Table 3.1. Experimental layout**

Distribution of treatments and birds			No. of birds
T <sub>2</sub> R <sub>3</sub> (15)	T <sub>3</sub> R <sub>2</sub> (15)	T <sub>1</sub> R <sub>1</sub> (15)	45
T <sub>1</sub> R <sub>2</sub> (15)	T <sub>2</sub> R <sub>1</sub> (15)	T <sub>3</sub> R <sub>3</sub> (15)	45
T <sub>3</sub> R <sub>1</sub> (15)	T <sub>1</sub> R <sub>3</sub> (15)	T <sub>2</sub> R <sub>2</sub> (15)	45
<b>Total birds</b>			<b>135</b>

### 3.5 Collection of experimental feed

For the research of effect of different level of dietary protein supplements on ammonia emission, growth performance and immune parameter of Broiler Chicken. Grower and starter ready feed was collected from birulia road, savar. Maize and soybean meal was collected from nama bazar, savar.

### 3.6 Collection of Ammonia test kit

For assessment of ammonia emission in broiler house and to differentiate the effect of the treatment groups, ammonia test kit was collected from Newzealand during research of my supervisor and its details are given at 3.6.1. It was not easy to collect the test kit because of its unavailability in the market of our country.

#### 3.6.1 Ammonia test kit description

To assess ammonia level in broiler house, the commercially available ammonia test kit was Micro Essential Hydrion ammonia meter tester paper. The paper was packaged as a 15-foot roll in a pocket sized plastic dispenser with a polypropylene case. It comes

complete with a specially calibrated color chart. Per each: 10/ Carton. Micro Essential Labs #: AM-40. The commercially available ammonia test kit that was used in this study described below:

**Table 3.2. Description of ammonia test kit**

<b>Parameters</b>	<b>Feature</b>
Brand Name	Micro essential
Weight	120 gram
Model Number	MIC-AM-40
Part Number	AM-40
NH <sub>3</sub> range	0 To 100ppm

### **3.6.2 Ammonia assessment procedure**

Ammonia present in the air was detected using test strips of ammonia test kit. For that reason, 1 inch strip of paper was tear off and wet it with 1 or 2 drops of distilled water. Excess water was shaken in the paper and exposed for 15 seconds in air being tested. Then it compared with the color chart of the test kit. If color change matched, then it was recorded as data.

### **3.7 Preparation of broiler house**

The broiler shed was an open sided natural house. It was a tin shed house with concrete floor. The experimental room was properly cleaned and washed by using tap water. All the equipment of the broiler house was cleaned and disinfected. There was 1ft. side wall around the shed with no ceiling. The floor was above 1ft. from the ground and the top of the roof was above 15ft. from the floor. The house was disinfected by detergent before starting the experiment. After proper drying, the house was divided into pens as per lay-out of the experiment by polythene sheet so that air cannot pass one pen to another. The height of pens was 5 ft. Before placement of chicks the house was fumigated by formalin and potassium permanganate @ 500 ml formalin and 250 g potassium permanganate (i.e. 2:1) for 35 m<sup>3</sup> experimental area. Rice husk was used as a litter material to keep free the floor from moisture.

### 3.8 Experimental diets

Starter and grower commercial Kazi broiler feed were purchased from the local market. Starter diet was enriched with minimum 4 times daily by following Lohmann Meat (Indian River) Manual and *ad libitum* drinking water 2 times daily. Detail composition of feed are presented in table 3, 4 & 5,6 & 7.

**Table 3.3. Name and minimum percentage of ingredients present in starter ration**

Name of ingredients in Starter ration	Percentage Present
Protein	21.0
Fat	6.0
Fiber	5.0
Ash	8.0
Lysine	1.20
Methionine	0.49
Cysteine	0.40
Tryptophan	0.19

(Source: Kazi starter feed 50 kg packet)

**Table 3.4. Name and minimum percentage of ingredients present in grower ration**

Name of ingredients in Grower ration	Percentage Present
Protein	20.0
Fat	6.0
Fiber	5.0
Ash	8.0
Lysine	1.10
Methionine	0.47
Cysteine	0.39
Tryptophan	0.18
Threonine	0.75
Arginine	1.18

(Source: Kazi grower feed 50 kg packet)

**Table 3.5. Ration formulation chart**

Treatment	Status	1 <sup>st</sup> week	2 <sup>nd</sup> Week	3 <sup>rd</sup> and 4 <sup>th</sup> Week
T <sub>1</sub>	High CP	Basal diet(Starter)	Starter ready feed- 90%	Grower ready feed- 75%
			½ kg maize for 10 kg feed formulation	½ kg maize for 10 kg feed formulation
T <sub>2</sub>	Medium CP	Basal diet(Starter)	Starter ready feed	Grower ready feed
T <sub>3</sub>	Low CP	Basal diet(Starter)	Grower ready feed with slight crushing	Grower ready feed - 70 % Maize-25% Soymeal-5%

**Table 3.6. Nutrient composition of the ingredients used to formulate experimental diets**

Ingredients	DM (%)	ME (K Cal/kg)	CP (%)	CF (%)	Ca (%)	P (%)	Lys (%)	Meth (%)	Tryp (%)
Soybean meal	90	2710	44.50	7.5	0.26	0.23	2.57	0.76	0.57
Maize	89.5	3309	9.2	2.4	0.25	0.40	0.18	0.15	0.09

**Table 3.7. Proximate composition of feed (CP%)**

Treatment	Starter	Grower
T <sub>1</sub> (Prepared)	23	22
T <sub>2</sub> (Readymade feed)	21	20
T <sub>3</sub> (Prepared)	19	18

### **3.9 Care of day old chicks**

Just after arrival of day old chicks to the poultry house the initial weight of the chicks were recorded by a digital electronic balance, vaccination was done and distributed them under the hover for brooding. The chicks were supplied glucose water with Vitamin-C to drink for the first 3 hours to overcome dehydration and transportation stress. Subsequently small feed particles were supplied on the newspapers to start feeding for the first 24 hours.

#### **3.9.1 Brooding of baby chicks**

Electric brooder was used to brood chicks. Partitioning brooding was done due to different experimental treatment. Each brooder had one hover and a round chick guard to protect chicks and four partitioning chambers. Thereafter healthy baby chicks were randomly distributed to the pen according to the design of the experiment. The recommended brooding temperature was 35-21<sup>0</sup> C from 1<sup>st</sup> to 4<sup>th</sup> weeks of age. Sometimes day temperature was 31-35<sup>0</sup> C. So, at that time there was no need of extra heat to brood the baby chicks, but at night a 100-watt bulb was used in each pen to rise up low temperature according to heat requirement of brooding schedule. In case of high temperature cross ventilation was allowed by folding wall polythene and electric fans were used to reduce heat. After one week a 200-watt electric bulb was hanged in every pen up to the market age of birds. Moreover, at that time the wall polythene sheet spread over the net-wire to protect the broiler chicks from cold and wind.

#### **3.9.2 Room temperature and relative humidity**

Daily room temperature (°C) and humidity were recorded with a thermometer and a wet and dry bulb thermometer respectively. Daily of room temperature and percent relative humidity for the experimental period were recorded and presented. Average of room temperature and percent relative humidity for the experimental period was recorded and presented in Table 8.

**Table 3.8. Average temperature and humidity**

Week	Date	Temperature (°C)		Humidity (%)	
		Average Maximum	Average minimum	Average Maximum	Average minimum
1 <sup>st</sup>	17.06.21- 24.06.21	34.01	29.21	93.87	63
2 <sup>nd</sup>	25.06.21- 01.07.21	33.37	28.07	96.28	69.57
3 <sup>rd</sup>	02.07.21-08.07.21	31.27	27.44	97.85	71.85
4 <sup>th</sup>	09.07.21-15.07.21	33.04	28.31	95.85	59.85

### 3.9.3 Feeding and drinking

Crumble feed was used as starter (0-2 wks.) and pellet feed for grower (3-4 wks.) ration. *Ad libitum* feeding was allowed for rapid growth of broiler chicks up to the end of the four weeks. Fresh clean drinking water was also supplied *Ad libitum*. Feeds were supplied 3 times: morning, noon and night. Water was supplied two times daily: morning and evening. Left over feeds and water were recorded to calculate actual intake. Digital electronic balance and measuring plastic cylinder was used to take record of feed and water. Weekly feed consumption (gm)/bird were calculated to find out weekly and total consumption of feed. All feeders and drinkers were washed and sun-dried before starting the trial. One plastic made round feeder and one drinker were kept in the experimental pen. Feeder and drinker size were changed according to the age of the birds. Feeders were washed at the end of the week and drinkers once daily.

### 3.9.4 Lighting

At night there was provision of light in the broiler house to stimulate feed intake and rapid body growth. A 200watt incandescent bulb lights (1000 lumen) were provided to ensure 24 hours' light for first 2 weeks. Thereafter 23 hours' light and one-hour dark were scheduled up to marketable age. At night one-hour dark was provided in two times by half an hour.

### 3.9.5 Ventilation

The broiler shed was south faced and open-sided. Due to wire-net cross ventilation was easy to remove polluted gases from the farm. Besides, on the basis of necessity ventilation was regulated by folding polythene screen. The open space around the farm were favorable for cross ventilation.

### 3.9.6 Biosecurity measures

Biosecurity is a set of management practices that reduce the potential for introduction and spread of diseases causing organisms. To keep disease away from the broiler, farm the following vaccination, medication and sanitation program was undertaken. All groups of broiler chicks were supplied Vitamin B-Complex, Vitamin-A, D, E, K and Vitamin-C, Ca and Vitamin-D enriched medicine and electrolytes.

### 3.9.7 Vaccination

The vaccines were collected from medicine shop (Ceva Company) and applied to the experimental birds according to the vaccination schedule. One ampoule vaccine was diluted with distilled water according to the recommendation of the manufacturer. The cool chain of vaccine was maintained strictly up to vaccination. The vaccination schedule of broiler is shown in Table 9.

**Table 3.9. Vaccination schedule**

Age	Name of Disease	Name of Vaccine	Route of vaccination
Day 0	Infectious Bronchitis + Newcastle Disease (IB+ND)	CEVAC BIL	One drop in eye
Day 9	Gumboro (IBD)	CEVAC IBDL	Drinking water
Day 17	Gumboro (IBD)	CEVAC IBDL	Drinking water

### 3.9.8 Medication

Vitamin-B complex, vitamin-A, D3, and E were used against deficiency diseases. Electromin and Vitamin-C also used to save the birds from heat stress. The medication program is presented in the Table 3.10.

**Table 3.10. Medication program**

Medicine	Composition	Dose	Period
Liva -Vit	Vitamin B-complex	2-5ml/1L water	3-5 days (all groups)
Renasol AD3E (Vet)	Vitamin A, D & E	1 ml/5L water	3 -5 days (all groups)
Electromin powder	Electrolytes	1g/2L water	4 -5 days (all groups)
Cavit-P	Calcium, Phosphorus Vitamin-C, B12 Premix	1g/5L water	4 -5 days (all groups)
Hiprachok	Vitamin and amino acid	10 ml/100 bird	3-5 days (all groups)

### 3.9.9 Sanitation

Proper hygienic measures were maintained throughout the experimental period. Cleaning and washing of broiler shed and its premises were under a routine sanitation work. Flies and insects were controlled by spraying Phenol and Lysol to the surroundings of the broiler shed. The attendants used farm dress and shoe. There was a provision of Foot Bath at the entry gate of the broiler shed to prevent any probable contamination of diseases. Strict sanitary measures were followed during the experimental period.

### 3.10 Recorded parameters

Daily temperature, humidity, light intensity & ammonia level was calculated. Weekly live weight, weekly feed consumption and death of chicks to calculate mortality



percent were taken during the study. FCR was calculated from final live weight and total feed consumption per bird in each replication. After slaughter carcass weight and gizzard, liver, spleen and heart were measured from each broiler chicken. Dressing yield was calculated for each replication to find out dressing percentage.

### **3.11 Data collection**

#### **3.11.1 Ammonia emission**

During the study, the data of ammonia emission from broiler litter was collected daily at several times from each replication of all treatment groups and untreated also. The average of the daily recorded ammonia emission was calculated.

#### **3.11.2 Live weight**

The initial day-old live weight and weekly live weight of each replication was kept to get final live weight record per bird.

#### **3.11.2 Dressing yield**

Dressing yield of bird was obtained from live weight subtracting blood, feathers, head, shank and inedible viscera.

#### **3.11.3 Feed consumption**

Daily feed consumption record of each replication was kept to get weekly and total feed consumption record per bird.

#### **3.11.4 Survivability of chicks**

Daily death record for each replication was counted up to 28 days of age to calculate mortality if occurred that indicated the survivability of the bird.

### **3.11.5 Dressing procedures of broiler**

Three birds were picked up at random from each replicate at the 28th day of age and sacrificed to estimate dressing percent of broiler chicken. All birds to be slaughtered were weighed and fasted by halal method or overnight (12 hours) but drinking water was provided *ad-libitum* during fasting to facilitate proper bleeding. All the live birds were weighed again prior to slaughter. Birds were slaughtered by severing jugular vein, carotid artery and the trachea by a single incision with a sharp knife and allowed to complete bleed out at least for 2 minutes. Outer skin was removed by sharp scissor and hand. Then the carcasses were washed manually to remove loose singed feathers and other foreign materials from the surface of the carcass. Heart and liver were removed from the remaining viscera by cutting them loose and then the gall bladder was removed from the liver. Cutting it loose in front of the proventriculus and then cutting with both incoming and outgoing tracts removed the gizzard. Giblet were collected after removing the gall bladder. All the carcasses were washed with cold water inside and out to remove traces blood, loosely attached tissue or any foreign materials. Then the eviscerated weight of carcasses was recorded. Thereafter the weight of carcass cuts such as breast, thigh (both), drumstick (both), back, neck, wing (both), heart, liver and gizzard was taken. Dressing yield was found by subtracting blood, feathers, head, shank, liver, heart and digestive system from live weight. Liver, heart, gizzard and neck were considered as giblet. Percent of breast, thigh, drumstick, back, wing, giblet and abdominal fat were found as DP.

### **3.11.6 Immune parameter**

At the end of the experiment blood sample was collected randomly from each replication of every treatment. 2mL blood was collected from wing vein with syringe in a vacutainer. Vacutainer contains EDTA solution which prevent blood coagulants. Few hours after collection the blood sample was tested by Auto Blood Analyzer in dhanmondi diagnostic and consultation center, Dhaka.

### 3.12 Calculations

Each data was collected by the following formulae:

#### 3.12.1 Live weight gain

The average body weight gain of each replication was calculated by deducting initial body weight from the final body weight of the birds.

$$\text{Body weight gain(g)} = \text{Final weight(g)} - \text{Initial weight (g)}$$

#### 3.12.2 Feed intake

Feed intake was calculated as the total feed consumption in a replication divided by number of birds in each replication.

$$\text{Feed intake (g/bird)} = \frac{\text{Feed intake in a replication}}{\text{No. of birds in a replication}}$$

#### 3.12.3 Feed conversion ratio (FCR)

Feed conversion ratio (FCR) was calculated as the total feed consumption divided by weight gain in each replication.

$$\text{FCR} = \frac{\text{Feed intake (kg)}}{\text{Weight gain (kg)}}$$

#### 3.12.4 Dressing percentage

Dressing yield was found by subtracting blood, feathers, head, shank and digestive system from live weight. Liver, heart, gizzard and neck were considered as giblet.

Dressing percentage of bird was calculated by the following formula-

$$DP = \frac{\text{Dressing yield (g)}}{\text{Live weight(g)}} \times 100$$

Dressing yield = Breast, thigh, drumstick, back, wing, giblet, abdominal fat weight.

### **3.13 Statistical analysis**

Total data were compiled, tabulated and analyzed in accordance with the objectives of the study. Excel Program was practiced for preliminary data calculation. The collected data was subjected to statistical analysis by applying one-way ANOVA using Statistical Package for Social Sciences (SPSS version 16.0, 2008). Differences between means were tested using Duncan's multiple comparison test, LSD and significance was set at  $P < 0.05$ .

**Some photographic view of chick management and experimental procedure**



**Plate 1. Washing of floor by detergent**



**Plate 2. Washing of feeder and drinker**



**Plate 3. Brooding of chicks**



**Plate 4. Preparation of broiler shed**



**Plate 5. Monitoring of research activities by the supervisor**





**Plate 6. Vaccination of chick**



**Plate 7. Giving starter feed of chick**



**Plate 8. Preparation of feed of different protein concentration**



**Plate 9. Different equipment for data collection**



**Plate 10. Data collection of ammonia emission, light intensity**



**Plate 11. Collection of weight**



**Plate 12. Blood collection**



**Plate 13. Weight of slaughtered broiler**



**Plate 14. Weight of gizzard**



**Plate 15. Weight of liver**



**Plate 16. Weight of heart**



**Plate 17. Weight of Back**



**Plate 18. CP test by kjeldahl method**



## CHAPTER IV

### RESULTS AND DISCUSSION

#### 4.1 Ammonia gas emissions

The rate of NH<sub>3</sub> emissions against 28 days of rearing period of birds in different concentration level of CP had shown in Table 4.1. Results demonstrated that the rate of NH<sub>3</sub> emissions was low where low dietary CP was supplied.

However, the rate of NH<sub>3</sub> emission from broiler litter was reduced when concentration level of CP was low. ANOVA analysis revealed that the average NH<sub>3</sub> level were insignificant ( $P>0.05$ ) in 1<sup>st</sup> week but it varied statistically significant ( $P<0.05$ ) at 2<sup>nd</sup>, 3<sup>rd</sup> and 4<sup>th</sup> week of rearing period. The rate of NH<sub>3</sub> generation was higher gradually after 7 days of rearing period. The highest level of NH<sub>3</sub> (ppm) was found in high CP treated group T<sub>1</sub> ( $11.63\pm 0.30$  ppm) followed by T<sub>2</sub> ( $10.36\pm 0.45$ ) and T<sub>3</sub> ( $6.53\pm 0.25$ ) respectively at 28 days of rearing period.

This result agrees with Hernández *et al.* 2012. For instance, low protein diet positively reduced N excretion when used in broiler diets. Jacob *et al.* (1994) showed that the level of N excreted in poultry waste can be reduced by up to 21% if dietary CP content is lowered by 2.5% and the diet is supplemented with synthetic amino acids. The study by Emous *et al.* 2019 also agree with this result that reducing CP level in the diet of broiler breeders reduces ammonia emission and total N-losses from litter and manure.

#### 4.2 Production performances of broiler

Production performances of broiler chicken was evaluated by body weight gain, feed consumption, feed Conversion Ratio (FCR), dressing percentage (DP), carcass weight. The parameters research data analysis is given and discussed below.

##### 4.2.1 Final Live Weight

Data submitted in Table 4.2 expressed that the effect of treatments on final live weight (gram per bird) was significant ( $P<0.05$ ). The relative final live weight (g) of broiler chickens in the different groups T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> were  $1927.34\pm 33.57$ ,  $1835.80\pm 15.83$  and

1779.20.00±18.91 respectively. The highest result was found in T<sub>1</sub> (1927.34g) and lowest result was found in T<sub>3</sub> (1779.20g) group and that was statistically significant (P<0.05). Results also expressed that the body weights also different among the treatment groups having statistical significance (P<0.05). The higher body weight in T<sub>1</sub> group might be due to the positive effect of high dietary protein.

This results are in agreement with those obtained by reducing dietary CP below a minimum level even with maintained EAA level, depressed BW, FCR and feed intake. Other authors have reported similar results, confirming that reduction in CP during the grower period (10–28 days of age) could possibly reduce BW and feed intake (Ferguson *et al.*, 1998; Si *et al.*, 2004; Waldroup *et al.*, 2005; Yamazaki *et al.*, 2006).

#### **4.2.2 Weekly body weight gain**

Data presented in Table 4.3 showed that the effect of treatments on total body weight gain (gram per broiler chicken) was not significant (P>0.05) in 2<sup>nd</sup> and 3<sup>rd</sup> week but significant (P<0.05) in 1<sup>st</sup> and 4<sup>th</sup> week. Somehow there are difference in total body weight gain among treatments. The relative total body weight gain (g) of broiler chickens in the dietary group T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> were 1885.34±33.75, 1793.98±15.92 and 1737.20±18.91 respectively. The highest result was found in T<sub>1</sub> (1885.34±33.75) and lowest result was in T<sub>3</sub> (1737.20±18.91) group. At the 2<sup>nd</sup>, 3<sup>rd</sup> and 4<sup>th</sup> week there were higher body weight gain value recorded in T<sub>1</sub> group than T<sub>2</sub> and T<sub>3</sub> group.

This results are in agreement with those obtained by Hernandez *et al.*, 2012; Folorunso *et al.*, 2014 and Liu *et al.*, 2017. Allameh and Toghyani (2019) concluded that the weight of broiler chickens significantly decreased when the dietary CP reduced from 20.4% to 17.9%. Moreover, they concluded that feeding 85% of CP requirements had negative effects on the daily weight gain of broilers. Hilliar *et al.* (2020) and Macelline *et al.* (2020) concluded that birds were fed low protein diets had a low BWG, compared to chicks that were fed high protein diets.

#### **4.2.3 Feed Consumption**

All the treatment groups (Table 4.2) showed significant (P<0.05) differences in FC of broiler chicken. T<sub>1</sub> group consumed higher amount of feed (2560.57g±47.32) and T<sub>3</sub>

group consumed relatively lower amount of feed ( $2317.30\pm 24.70$ ). T<sub>1</sub> group contain high CP feed and relatively less amount of energy. To fulfill the energy requirement bird can be fed high amount of feed than T<sub>2</sub> and T<sub>3</sub> group.

This results are in disagreement with the experiment conducted by Nemat *et al.* (2011), to examine the effect of three levels of crude protein (CP) and three levels of Ca and available P on performance of broilers from hatching until 21 days of age. Results of the experiment showed that CP content had no significant effect on feed and water intake.

#### **4.2.4. Weekly feed consumption**

Data regarding presented in Table 4.4 showed that the feed consumption (g) of broiler chicks at the end of 1<sup>st</sup>, 2<sup>nd</sup> and 3<sup>rd</sup> week in different groups were no significant ( $P>0.05$ ) effect. The feed consumption (g) of broiler chicks at the end of 4<sup>th</sup> week in different groups was significant ( $P<0.05$ ) effects. Somehow feed consumption increased with high CP supplement. The mean feed consumption (g) of broiler chicks at the end of 4<sup>th</sup> week in dietary group T<sub>1</sub>, T<sub>2</sub>, and T<sub>3</sub> are  $1068.82\pm 39.60$ ,  $899.74\pm 60.29$  and  $837.34\pm 69.27$  gm respectively. The higher feed consumption was in T<sub>1</sub> and comparatively lower in T<sub>3</sub>.

Reducing dietary CP below a minimum level (in this experiment, 19%), even with maintained EAA level, depressed BW, FCR and feed intake. Other authors have reported similar results, confirming that reduction in CP during the grower period (10–28 days of age) could possibly reduce BW and feed intake (Ferguson *et al.*, 1998; Si *et al.*, 2004; Waldroup *et al.*, 2005; Yamazaki *et al.*, 2006).

#### **4.2.5. Feed conversion ratio**

Data presented in Table 4.2 showed that feed conversion ratio were not significant ( $P>0.05$ ) in any group. Feed supplemented with high CP at T<sub>1</sub>, feed conversion ratio (FCR) was higher in T<sub>1</sub> group ( $1.35\pm 0.02$ ) and comparatively same in T<sub>2</sub> group ( $1.32\pm 0.02$ ) and T<sub>3</sub> ( $1.33\pm 0.01$ ) respectively.

This results are in agreement with the findings of Mamdouh Omar *et al.* (2020), conducted the test in which chickens fed the diet containing 100% required Crude

Protein (CP) supplemented with citric acid which could significantly improve body weight gain, feed conversion ratio. Chickens fed the low CP diet supplemented with citric acid could compensate for the growth performance equivalent to those fed the optimal CP diet.

#### **4.2.6. Weekly feed conversion ratio**

The mean weekly FCR of broiler chicks in different groups were presented in Table 4.5. The FCR of 2<sup>nd</sup>, 3<sup>rd</sup> and 4<sup>th</sup> weeks were insignificant ( $P>0.05$ ) among the treated group. In 4<sup>th</sup> week FCR of T<sub>1</sub> group is somewhat higher ( $1.61\pm 0.07$ ) than other groups T<sub>2</sub> ( $1.52\pm 0.05$ ) and T<sub>3</sub> ( $1.58\pm 0.03$ ).

The present finding was in agreement with Lemme *et al.* (2019) who reported that addition of CP to broiler chicken diets does not affect FCR significantly.

#### **4.2.7 Dressing percentage (DP)**

Data presented in Figure 4.1 showed that the dressing percentage at all group was significant ( $P<0.05$ ). carcass percentage compared with T<sub>1</sub> ( $70.81\pm 0.76$ ) and the other treatment group T<sub>2</sub> ( $67.10\pm 0.70$ ) and T<sub>3</sub> ( $66.52\pm 0.38$ ) respectively. Experiment, evaluation of dressing percentage on slaughtered representative birds revealed that T<sub>1</sub> group had significantly higher dressed percentage followed by T<sub>2</sub> and T<sub>3</sub> groups.

These results are in agreement with the study by Saleh *et al.* (2021), Dressing percentages revealed significantly affected by reducing dietary protein levels, while the blood plasma total protein, albumin, and globulin were not significant differences due to dietary low protein.

#### **4.2.8. Survivability**

The survivability rate showed on Table 4.2. Survivability rate was statistically higher for the T<sub>2</sub> and T<sub>3</sub> treated group ( $100\pm 0.00$ ) than T<sub>1</sub> group ( $97.67\pm 2.23$ ) but no significant ( $P>0.05$ ) difference among them. The overall survivability (0-4 weeks) during the experimental period was almost higher in all the treatment group. The variation in mortality among the group might be due to the seasonal influence of summer. The

another possible cause of less survivability might be due to more ammonia gas emission in T<sub>1</sub>.

The mortality observed in the present study agreed with the report of Malomo *et al.* (2013), assessed the effects of dietary crude protein on performance and nitrogen economy of broilers. The result showed that mortality was not significantly different ( $P>0.05$ ).

#### **4.2.9 Carcass weight**

Data presented in Table 4.6 showed that the carcass weight in T<sub>1</sub> groups are better than other two group T<sub>2</sub> and T<sub>3</sub> group. The results revealed that the treatments had significant effects ( $P<0.05$ ) in dressed wings, breast, back, thigh, drumstick. However, in treatment T<sub>1</sub> group the carcass weight is better than other treatment groups.

The present findings were in agreement with previous findings by Malomo *et al.* (2013), who reported that higher carcass and breast meat yield in broilers' carcass that high protein containing feed.

#### **4.3 Economic impact**

The cost of different treatment groups and control group presented in Table 4.7. Production cost included feed cost and common costs (litter cost, vaccine, medicine, electricity etc.) for both the treated groups and untreated group. Total expenditure per bird was higher high cp treated groups T<sub>1</sub> ( $199.05\pm 2.39$ ) and lower in low CP group T<sub>3</sub> ( $186.83\pm 1.22$ ). Feed cost was comparatively higher in high CP group. Profit per bird (PPB) and Benefit cost ratio (BCR) also presented in Table 10, demonstrated the economic impact of the treatment groups compared with the untreated group. Return was calculated after selling the live birds per kg weight and profit was computed by subtracting the expenditure. Profit per bird (TK.) was higher in treatment groups but not significant T<sub>1</sub> ( $70.73\pm 4.19$ ), T<sub>2</sub> ( $67.27\pm 2.99$ ) and T<sub>3</sub> ( $62.18\pm 3.83$ ). Among the treatment groups T<sub>1</sub> performed better than others. BCR was also statistically higher ( $P<0.05$ ) in treatment groups T<sub>1</sub> ( $1.35\pm 0.02$ ),) compared with the T<sub>3</sub> ( $1.32\pm 0.02$ ) (Table 4.7).

This results are in agreement with those of previous researchers (The study by Kumari *et al.* (2016)) reported the application of high dietary protein on economic analysis revealed that it could be cost-effective management practice to improve shed environment and in turn performance of broiler chicks.

#### **4.4 Immune Parameter**

The immune parameter Hemoglobin, WBC, RBC, Lymphocyte, Monocyte, Neutrophil, Eosinophil, PCV and MCV was counted and the data has presented in Table 4.8. The Hemoglobin, WBC, RBC, Platelet, Neutrophil, Lymphocyte, Monocyte, PCV and MCV was statistically insignificant ( $P>0.05$ ) among different treatment. The highest Hemoglobin ( $12.55\pm 1.72$ ), WBC ( $18000\pm 818.53$ ), RBC ( $5.13\pm .4$ ), Monocyte ( $2.00\pm 1.00$ ), Lymphocyte ( $17\pm 3.60$ ) found in T<sub>2</sub>. Highest platelet was in T<sub>3</sub> ( $465000\pm 0127573.5$ ) and neutrophil ( $83.00\pm 1.73$ ) found in T<sub>3</sub>. The lowest WBC ( $14200\pm 2351.5$ ), RBC ( $3.79\pm .43$ ) and Hemoglobin ( $8.66\pm 1.02$ ) found in T<sub>3</sub>.

This results are agreed with the findings of Swennen *et al.* (2005), Kamran *et al.* (2010), and Mohamed *et al.* (2012). They concluded that the level of dietary protein did not alter glucose concentration. In addition, Alam *et al.* (2004) and Mohamed *et al.* (2012) indicated that the dietary protein level had no significant effect on the hemoglobin concentration of broilers. In addition, Mohamed *et al.* (2012) stated that the dietary protein caused only the globin section of the hemoglobin to increase with no effect on the haem section. (Moore *et al.*, 2008), found that NH<sub>3</sub> levels as low as 20 ppm compromise the immune system of chickens, making them more susceptible to diseases and damage to respiratory system. Feed conversion and weight gains in poultry are also affected by high level of ammonia.

**Table 4.1. Weekly ammonia gas emission**

<b>Treatment</b>	<b>1<sup>st</sup> week</b>	<b>2<sup>nd</sup> week</b>	<b>3<sup>rd</sup> week</b>	<b>4<sup>th</sup> week</b>
<b>T<sub>1</sub></b>	3.2±00	6.23 <sup>a</sup> ±0.03	8.80 <sup>a</sup> ±0.05	11.63 <sup>a</sup> ±0.17
<b>T<sub>2</sub></b>	3.2±00	5.80 <sup>b</sup> ±0.05	8.30 <sup>b</sup> ±0.11	10.36 <sup>b</sup> ±0.26
<b>T<sub>3</sub></b>	3.2±00	3.46 <sup>c</sup> ±0.03	5.30 <sup>c</sup> ±0.15	6.53 <sup>c</sup> ±0.14
<b>Mean±SE</b>	<b>3.2±00</b>	<b>5.16±0.43</b>	<b>7.46±.54</b>	<b>9.51±0.77</b>
<b>Significance</b>	<b>NS</b>	<b>*</b>	<b>*</b>	<b>*</b>

Here, T<sub>1</sub> = (High CP feed), T<sub>2</sub> = (Medium CP feed), T<sub>3</sub> = (Low CP feed); Values are Mean±SE (n=9) one-way ANOVA (SPSS, Duncan method).

- ✓ Mean with different superscripts are significantly different (P<0.05)
- ✓ SE= Standard Error
- ✓ Mean within same superscripts don't differ (P>0.05) significantly
- ✓ NS=Non-Significant (P>0.05)
- ✓ \* =Significant (P<0.05)

**Table 4.2. Production performance**

<b>Treatment</b>	<b>Final Live Weight (g/bird)</b>	<b>Avg. BWG (g/bird)</b>	<b>Total FC (g/bird)</b>	<b>Final FCR</b>	<b>Survivability rate %</b>
T <sub>1</sub>	1927.34 <sup>a</sup> ±33.75	1885.34 <sup>a</sup> ±33.75	2560.57 <sup>a</sup> ±47.32	1.35±0.02	97.76±2.23
T <sub>2</sub>	1835.80 <sup>b</sup> ±15.83	1793.98 <sup>b</sup> ±15.92	2374.71 <sup>b</sup> ±22.43	1.32±0.02	100.00±0.00
T <sub>3</sub>	1779.20 <sup>b</sup> ±18.91	1737.20 <sup>b</sup> ±18.91	2317.30 <sup>b</sup> ±24.70	1.33±0.01	100.00±0.00
<b>Mean± SE</b>	<b>1847.44</b> <b>±24.72</b>	<b>1805.50</b> <b>±24.72</b>	<b>2417.53±</b> <b>40.33</b>	<b>1.33±0.01</b>	<b>99.25±0.74</b>
<b>Significance</b>	<b>*</b>	<b>*</b>	<b>*</b>	<b>NS</b>	<b>NS</b>

Here, T<sub>1</sub> = (High CP feed), T<sub>2</sub> = (Medium CP feed), T<sub>3</sub> = (Low CP feed); Values are Mean±SE (n=9) one-way ANOVA (SPSS, Duncan method).

- ✓ Mean with different superscripts are significantly different (P<0.05)
- ✓ SE= Standard Error
- ✓ Mean within same superscripts don't differ (P>0.05) significantly
- ✓ NS=Non-Significant (P>0.05)
- ✓ \* =Significant (P<0.05)



**Table 4.3. Weekly body weight gain**

Treatments	Weekly Body Weight Gain				Total BWG
	1 <sup>st</sup> week	2 <sup>nd</sup> week	3 <sup>rd</sup> week	4 <sup>th</sup> week	
T <sub>1</sub>	220.56 <sup>a</sup> ±2.27	392.15±4.90	617.02±22.85	659.60 <sup>a</sup> ±17.2	1885.34 <sup>a</sup> ±33.75
T <sub>2</sub>	217.27 <sup>a</sup> ±1.36	384.71±7.67	594.46±9.24	597.53 <sup>b</sup> ±2.76	1793.98 <sup>b</sup> ±15.92
T <sub>3</sub>	207.20 <sup>b</sup> ±1.92	378.97±7.48	579.74±12.88	571.28 <sup>b</sup> ±2.44	1737.20 <sup>b</sup> ±18.91
<b>Mean±SE</b>	<b>215.01±2.22</b>	<b>385.28±3.90</b>	<b>597.07±9.68</b>	<b>609.47±14.04</b>	<b>1805.50</b> <b>±24.7</b>
<b>Significance</b>	*	NS	NS	*	*

Here, T<sub>1</sub> = (High CP feed), T<sub>2</sub> = (Medium CP feed), T<sub>3</sub> = (Low CP feed); Values are Mean±SE (n=9) one-way ANOVA (SPSS, Duncan method).

- ✓ Mean with different superscripts are significantly different (P<0.05)
- ✓ SE= Standard Error
- ✓ Mean within same superscripts don't differ (P>0.05) significantly
- ✓ NS=Non-Significant (P>0.05)
- ✓ \* =Significant (P<0.05)

**Table 4.4. Weekly Feed Consumption**

Treatment	Weekly Feed Consumption				Total Feed Consumption
	1 <sup>st</sup> Week	2 <sup>nd</sup> week	3 <sup>rd</sup> week	4 <sup>th</sup> week	
T <sub>1</sub>	226±00	456.27±4.41	810.14±11.99	1068.82 <sup>a</sup> ±39.60	2560.57 <sup>a</sup> ±47.32
T <sub>2</sub>	226±00	450.83±8.35	764.92±7.54	899.74 <sup>ab</sup> ±60.29	2374.71 <sup>b</sup> ±22.43
T <sub>3</sub>	226±00	432.81±10.27	753.45±26.01	837.34 <sup>b</sup> ±69.27	2317.30 <sup>b</sup> ±24.70
<b>Mean±SE</b>	<b>226±00</b>	<b>446.64±5.36</b>	<b>776.17±12.16</b>	<b>935.30±45.04</b>	<b>2417.53±40.33</b>
<b>Significance</b>	<b>NS</b>	<b>NS</b>	<b>NS</b>	<b>*</b>	<b>*</b>

Here, T<sub>1</sub> = (High CP feed), T<sub>2</sub> = (Medium CP feed), T<sub>3</sub> = (Low CP feed); Values are Mean±SE (n=9) one-way ANOVA (SPSS, Duncan method).

- ✓ Mean with different superscripts are significantly different (P<0.05)
- ✓ SE= Standard Error
- ✓ Mean within same superscripts don't differ (P>0.05) significantly
- ✓ NS=Non-Significant (P>0.05)
- ✓ \* =Significant (P<0.05)

**Table 4.5. Weekly Feed Conversion Ratio**

Treatments	Weekly FCR				Total FCR
	1 <sup>st</sup> week	2 <sup>nd</sup> week	3 <sup>rd</sup> week	4 <sup>th</sup> week	
T <sub>1</sub>	1.06±0.03	1.16±0.02	1.31±0.03	1.61±0.07	1.35±0.02
T <sub>2</sub>	1.02±0.01	1.16±0.008	1.28±0.01	1.52±0.05	1.32±0.02
T <sub>3</sub>	1.05±0.01	1.11±0.03	1.30±0.06	1.58±0.03	1.33±0.01
<b>Mean±SE</b>	<b>1.04±0.01</b>	<b>1.14±0.01</b>	<b>1.30±0.02</b>	<b>1.57±0.03</b>	<b>1.33±0.01</b>
<b>Significance</b>	<b>NS</b>	<b>NS</b>	<b>NS</b>	<b>NS</b>	<b>NS</b>

Here, T<sub>1</sub> = (High CP feed), T<sub>2</sub> = (Medium CP feed), T<sub>3</sub> = (Low CP feed); Values are Mean±SE (n=9) one-way ANOVA (SPSS, Duncan method).

- ✓ Mean with different superscripts are significantly different (P<0.05)
- ✓ SE= Standard Error
- ✓ Mean within same superscripts don't differ (P>0.05) significantly
- ✓ NS=Non-Significant (P>0.05)

**Table 4.6. Carcass characteristics**

<b>Treatment</b>	<b>Breast</b>	<b>Thigh</b>	<b>Wing</b>	<b>Back</b>	<b>Drumstick</b>
T <sub>1</sub>	564 <sup>a</sup> ±7.54	193.33±4.97	89.66 <sup>a</sup> ±3.17	228 <sup>a</sup> ±9.23	184.33 <sup>a</sup> ±4.40
T <sub>2</sub>	527 <sup>b</sup> ±4.35	180±6.08	86.33 <sup>ab</sup> ±3.48	207.66 <sup>ab</sup> ±3.38	170 <sup>ab</sup> ±6.08
T <sub>3</sub>	518 <sup>b</sup> ±5.50	174.33±8.35	77 <sup>b</sup> ±2.88	199 <sup>b</sup> ±2.64	157 <sup>b</sup> ±4.16
<b>Mean±SE</b>	<b>536.33±7.64</b>	<b>182±4.34</b>	<b>84.33±2.47</b>	<b>211.55±5.20</b>	<b>170.44±4.66</b>
<b>Significance</b>	<b>*</b>	<b>NS</b>	<b>*</b>	<b>*</b>	<b>*</b>

Here, T<sub>1</sub> = (High CP feed), T<sub>2</sub> = (Medium CP feed), T<sub>3</sub> = (Low CP feed); Values are Mean±SE (n=9) one-way ANOVA (SPSS, Duncan method).

- ✓ Mean with different superscripts are significantly different (P<0.05)
- ✓ SE= Standard Error
- ✓ Mean within same superscripts don't differ (P>0.05) significantly
- ✓ NS=Non-Significant (P>0.05)
- ✓ \* =Significant (P<0.05)

**Table 4.7. Economic Impact**

<b>Treatment</b>	<b>Total Expenditure</b>	<b>Receipt Per Bird</b>	<b>Profit Per Bird</b>	<b>Benefit Cost Ratio</b>
T <sub>1</sub>	199.05 <sup>a</sup> ±2.39	269.78 <sup>a</sup> ±4.70	70.73±4.19	1.35±0.02
T <sub>2</sub>	189.71 <sup>b</sup> ±1.13	256.99 <sup>b</sup> ±2.21	67.27±2.99	1.35±0.01
T <sub>3</sub>	186.83 <sup>b</sup> ±1.22	249.01 <sup>b</sup> ±2.61	62.18±3.83	1.33±0.02
<b>Mean± SE</b>	<b>191.86±2.02</b>	<b>258.59±3.45</b>	<b>66.72±2.23</b>	<b>1.34±0.01</b>
<b>Significance</b>	*	*	NS	NS

Here, T<sub>1</sub> = (High CP feed), T<sub>2</sub> = (Medium CP feed), T<sub>3</sub> = (Low CP feed); Values are Mean±SE (n=9) one-way ANOVA (SPSS, Duncan method).

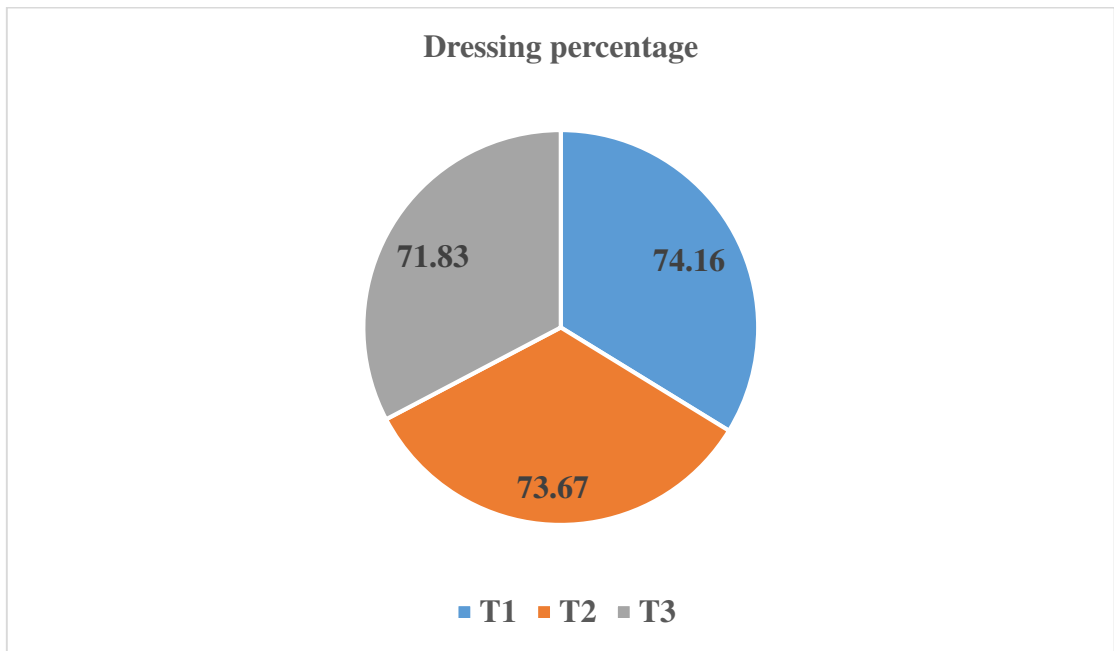
- ✓ Mean with different superscripts are significantly different (P<0.05)
- ✓ SE= Standard Error
- ✓ Mean within same superscripts don't differ (P>0.05) significantly
- ✓ NS=Non-Significant (P>0.05)
- ✓ \* =Significant (P<0.05)

**Table 4.8. Immune Parameter**

<b>Treatment</b>	<b>Hemoglobin</b>	<b>WBC</b>	<b>RBC</b>	<b>Platelet</b>	<b>Neutrophil</b>	<b>Lymphocyte</b>	<b>Monocyte</b>	<b>Eosinophil</b>	<b>PCV</b>
T <sub>1</sub>	9.61 <sup>a</sup> ±1.54	15000 <sup>a</sup> ±1059.87	4.64 <sup>a</sup> ±0.50	385000±43301.27	77.33 <sup>a</sup> ±1.20	17a±1.73	1.33±0.33	4.33±0.88	30.58±5.78
T <sub>2</sub>	12.55 <sup>ab</sup> ±0.99	18000 <sup>ab</sup> ±472.58	5.13 <sup>ab</sup> ±0.23	423333.33±68879.92	76.33 <sup>a</sup> ±2.60	17a±2.08	2±0.57	4.66±0.33	38.87±2.92
T <sub>3</sub>	8.66 <sup>b</sup> ±0.58	14200 <sup>b</sup> ±1357.69	3.79 <sup>b</sup> ±0.25	465000±73654.59	83 <sup>b</sup> ±1.00	11b±00	1.66±0.66	4.33±1.45	26.57±1.75
<b>Mean±SE</b>	<b>10.27±0.80</b>	<b>15733.33±774.77</b>	<b>4.52±0.26</b>	<b>424444.44±33721.12</b>	<b>78.88±1.35</b>	<b>15±1.26</b>	<b>1.66±0.28</b>	<b>4.44±0.50</b>	<b>32.01±2.65</b>
<b>Significance</b>	*	*	*	NS	*	*	NS	NS	NS

Here, T<sub>1</sub> = (High CP feed), T<sub>2</sub> = (Medium CP feed), T<sub>3</sub> = (Low CP feed); Values are Mean±SE (n=9) one-way ANOVA (SPSS, Duncan method).

- ✓ Mean with different superscripts are significantly different (P<0.05)
- ✓ SE= Standard Error
- ✓ Mean within same superscripts don't differ (P>0.05) significantly
- ✓ NS=Non-Significant (P>0.05)
- ✓ \* =Significant (P<0.05)



**Figure 4.1. Effects of dietary crude protein on dressing % of broiler**

## CHAPTER V

### CONCLUSION AND RECOMMENDATION

The present study was conducted at Sher-e-Bangla Agricultural University (SAU), Dhaka Poultry Farm for a period of four weeks using different level of dietary protein concentration in feed. The specific objectives of this study was under taken to determine the effect of different level of dietary crude protein to assess ammonia gas emission & production performance of broiler. A total of 135 day-old Lohman meat broiler chicks were purchased from Kazi hatchery, Savar, Dhaka. The experimental broilers were allocated randomly to three treatment groups with three replications having 15 broilers per replication. The experiment lasted for 4 weeks and the treatment of various groups consisted of group T<sub>1</sub> (high CP feed), group T<sub>2</sub> (medium CP feed), group T<sub>3</sub> (low CP feed). The parameters evaluated in this study were NH<sub>3</sub> emission from broiler litter, immune parameter, the bird's performance like body weight, feed consumption, FCR, survivability, carcass characteristics and economic impact on broiler rearing that includes production cost, profit per bird (PPB) and benefit cost ratio (BCR).

Result demonstrated that the average ammonia level in T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> were insignificant ( $P>0.05$ ) in 1<sup>st</sup> week ( $3.2\pm 0.00$ ) but it varied statistically significant ( $P<0.05$ ) at 2<sup>nd</sup> ( $5.16\pm 0.43$ ), 3<sup>rd</sup> ( $7.46\pm 0.54$ ) and 4<sup>th</sup> ( $9.51\pm 0.77$ ) week of rearing period. High CP treated group T<sub>1</sub> showed higher amount of NH<sub>3</sub> gas emission in broiler house than T<sub>2</sub> and T<sub>3</sub>. A statistically significant difference ( $P<0.05$ ) was noted on feed consumption ratio, NH<sub>3</sub> gas emission, carcass weight and dressing percentage value of the birds. Feed consumption of 4<sup>th</sup> week ( $1068.82\pm 39.60$ ) significantly ( $P<0.05$ ) higher in T<sub>1</sub>. There was no significant ( $P>0.05$ ) difference in weekly feed consumption among T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub>. The mean body weight gain (g) at 1<sup>st</sup> ( $215.01\pm 2.22$ ), 2<sup>nd</sup> ( $385.28\pm 3.90$ ) and 4<sup>th</sup> ( $609.47\pm 14.04$ ) week of T<sub>1</sub> were significantly higher ( $P>0.05$ ) than T<sub>2</sub> and T<sub>3</sub>. The better Feed conversion ratio (FCR) was significantly ( $P<0.05$ ) observed in high CP group T<sub>1</sub> than T<sub>2</sub> and T<sub>3</sub> group. At the last two weeks (3<sup>rd</sup> and 4<sup>th</sup> week) T<sub>1</sub> showed significantly ( $P<0.05$ ) higher FCR value ( $1.31\pm 0.03$  and  $1.61\pm 0.07$ ) than T<sub>3</sub> group ( $1.30\pm 0.06$  and  $1.58\pm 0.03$ ). Carcass percentage significantly ( $P<0.05$ ) higher in T<sub>1</sub> and T<sub>2</sub> than T<sub>3</sub> group and T<sub>1</sub> showed a greater carcass percentage value. Breast meat



percentage and edible portion of birds was significantly ( $P<0.05$ ) higher in  $T_1$  ( $564\pm7.54$ ) than  $T_3$  ( $518\pm5.50$ ). Drumstick percentage found higher in  $T_1$  ( $184.33\pm4.40$ ) and lower in  $T_3$  ( $157\pm4.16$ ) group but was insignificant ( $P>0.05$ ). Total expenditure per bird was significantly higher ( $P<0.05$ ) in  $T_1$  ( $199.05\pm2.39$ ) than  $T_3$  ( $186.83\pm1.22$ ). Feed cost was comparatively higher in  $T_1$  that was statistically insignificant ( $P>0.05$ ). Profit per bird was insignificantly higher ( $P>0.05$ ) in  $T_1$  ( $70.73\pm4.19$ ) than  $T_3$  ( $62.18\pm3.83$ ). BCR was also statistically higher ( $P<0.05$ ) in  $T_1$  ( $1.35\pm0.02$ ) compared with  $T_3$  ( $1.33\pm0.02$ ). Among the treatment groups  $T_1$  performed better than others.

Analyzing the above research findings, this study suggested that the high CP (23% in starter phase and 21% in grower phase) was for better production performance, to improve carcass quality and more economic benefit in broiler rearing. Though high CP feed increase somewhat production but it is not always true because high CP feed takes high cost and produce more ammonia gas emission. It was concluded that It should be keep in equilibrium state between high dietary CP supplementation and  $NH_3$  gas production. Indeed, as subsequent further research is needed to identify the most accurate findings.

## CHAPTER VI

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## CHAPTER VII

### APPENDICES

#### Appendix I. Average temperature and humidity

Week	Date	Temperature (°C)		Humidity (%)	
		Average Maximum	Average minimum	Average Maximum	Average minimum
1 <sup>st</sup>	17.06.21- 24.06.21	34.01	29.21	93.87	63
2 <sup>nd</sup>	25.06.21- 01.07.21	33.37	28.07	96.28	69.57
3 <sup>rd</sup>	02.07.21-08.07.21	31.27	27.44	97.85	71.85
4 <sup>th</sup>	09.07.21-15.07.21	33.04	28.31	95.85	59.85

**Appendix II. Ammonia content (ppm) in litter using different concentration levels of dietary crude protein in different rearing period**

<b>Treatment</b>	<b>Replication</b>	<b>1<sup>st</sup> week</b>	<b>2<sup>nd</sup> week</b>	<b>3<sup>rd</sup> week</b>	<b>4<sup>th</sup> week</b>
<b>T<sub>1</sub></b>	<b>R<sub>1</sub></b>	3.2	6.3	8.8	11.7
	<b>R<sub>2</sub></b>	3.2	6.2	8.7	11.9
	<b>R<sub>3</sub></b>	3.2	6.2	8.9	11.3
<b>T<sub>2</sub></b>	<b>R<sub>1</sub></b>	3.2	5.9	8.5	9.9
	<b>R<sub>2</sub></b>	3.2	5.7	8.1	10.4
	<b>R<sub>3</sub></b>	3.2	5.8	8.3	10.8
<b>T<sub>3</sub></b>	<b>R<sub>1</sub></b>	3.2	3.4	5.2	6.8
	<b>R<sub>2</sub></b>	3.2	3.5	5.6	6.3
	<b>R<sub>3</sub></b>	3.2	3.5	5.1	6.5

**Appendix III. Feed consumption (g/bird) of 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup> and 4<sup>th</sup> week under different treatment groups**

<b>Treatment</b>	<b>Replication</b>	<b>1<sup>st</sup> Week FC</b>	<b>2<sup>nd</sup> Week FC</b>	<b>3<sup>rd</sup> Week FC</b>	<b>4<sup>th</sup> Week FC</b>	<b>Total FC</b>
<b>T<sub>1</sub></b>	<b>R<sub>1</sub></b>	226.00	465.00	791.90	1041.23	2524.13
	<b>R<sub>2</sub></b>	226.00	450.76	832.76	1146.92	2656.44
	<b>R<sub>3</sub></b>	226.00	453.07	805.76	1018.33	2503.16
<b>T<sub>2</sub></b>	<b>R<sub>1</sub></b>	226.00	443.46	780.00	880.38	2329.84
	<b>R<sub>2</sub></b>	226.00	441.53	757.69	971.92	2397.14
	<b>R<sub>3</sub></b>	226.00	467.50	757.08	947.30	2397.88
<b>T<sub>3</sub></b>	<b>R<sub>1</sub></b>	226.00	448.46	702.30	905.38	2282.14
	<b>R<sub>2</sub></b>	226.00	413.46	787.30	878.07	2304.83
	<b>R<sub>3</sub></b>	226.00	436.53	770.76	931.66	2364.95

**Appendix IV. Body weight gain (BWG) (g/bird) of 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup> and 4<sup>th</sup> week under different treatments**

<b>Treatment</b>	<b>Replication</b>	<b>1<sup>st</sup> Week</b>	<b>2<sup>nd</sup> Week</b>	<b>3<sup>rd</sup> Week</b>	<b>4<sup>th</sup> Week</b>	<b>Total</b>
<b>T<sub>1</sub></b>	<b>R<sub>1</sub></b>	224.92	383.08	580.00	630.00	1818.00
	<b>R<sub>2</sub></b>	217.23	396.92	658.75	659.23	1923.13
	<b>R<sub>3</sub></b>	219.53	393.46	612.31	689.59	1914.89
<b>T<sub>2</sub></b>	<b>R<sub>1</sub></b>	214.92	383.08	610.00	603.07	1811.07
	<b>R<sub>2</sub></b>	217.23	372.31	578.01	594.62	1762.17
	<b>R<sub>3</sub></b>	219.66	398.75	595.38	594.92	1808.71
<b>T<sub>3</sub></b>	<b>R<sub>1</sub></b>	209.53	393.85	595.09	568.46	1766.93
	<b>R<sub>2</sub></b>	203.38	373.08	590.00	576.15	1742.61
	<b>R<sub>3</sub></b>	208.69	370	554.15	569.23	1702.07

**Appendix V. Effects of dietary protein on production performances of broiler**

<b>Treatment</b>	<b>Replication</b>	<b>Average Live Weight (G/Bird)</b>	<b>Total Feed Consumption (G/Bird)</b>	<b>Total Body Weight Gain (G/Bird)</b>	<b>Final FCR</b>	<b>Survivability</b>
<b>T<sub>1</sub></b>	<b>R<sub>1</sub></b>	1860.00	2524.13	1818.00	1.38	100
	<b>R<sub>2</sub></b>	1965.13	2656.44	1923.13	1.38	100
	<b>R<sub>3</sub></b>	1956.89	2503.16	1914.89	1.31	100
<b>T<sub>2</sub></b>	<b>R<sub>1</sub></b>	1853.07	2329.84	1811.07	1.29	100
	<b>R<sub>2</sub></b>	1804.17	2397.14	1762.17	1.36	100
	<b>R<sub>3</sub></b>	1850.17	2397.88	1808.17	1.33	100
<b>T<sub>3</sub></b>	<b>R<sub>1</sub></b>	1808.93	2282.14	1766.93	1.29	100
	<b>R<sub>2</sub></b>	1784.61	2304.83	1742.61	1.32	100
	<b>R<sub>3</sub></b>	1744.07	2364.95	1702.07	1.38	92

**Appendix VI. Live weight, eviscerated weight and dressing percentage of broiler under different treatments**

<b>Treatment</b>	<b>Replication</b>	<b>Average Live weight (g)</b>	<b>Eviscerated weight (g)</b>	<b>Dressing (%)</b>
<b>T<sub>1</sub></b>	<b>R<sub>1</sub></b>	1860.00	1290	69.35
	<b>R<sub>2</sub></b>	1965.13	1414	71.95
	<b>R<sub>3</sub></b>	1956.89	1392	71.13
<b>T<sub>2</sub></b>	<b>R<sub>1</sub></b>	1853.07	1267	68.37
	<b>R<sub>2</sub></b>	1804.17	1209	67.01
	<b>R<sub>3</sub></b>	1850.17	1220	65.93
<b>T<sub>3</sub></b>	<b>R<sub>1</sub></b>	1808.93	1217	67.29
	<b>R<sub>2</sub></b>	1784.61	1179	66.06
	<b>R<sub>3</sub></b>	1744.07	1155	66.22



**Appendix VII. Weight (g) of liver, heart, neck, gizzard, giblet and intestine, of broiler under different treatment groups**

<b>Treatment</b>	<b>Replication</b>	<b>Liver</b>	<b>Heart</b>	<b>Neck</b>	<b>Gizzard</b>	<b>Giblet</b>	<b>Intestine</b>
<b>T<sub>1</sub></b>	<b>R<sub>1</sub></b>	42	9	42	42	133	85
	<b>R<sub>2</sub></b>	49	11	45	59	147	119
	<b>R<sub>3</sub></b>	47	10	44	52	146	127
<b>T<sub>2</sub></b>	<b>R<sub>1</sub></b>	40	10	42	38	137	102
	<b>R<sub>2</sub></b>	33	10	41	50	125	114
	<b>R<sub>3</sub></b>	36	9	40	41	146	84
<b>T<sub>3</sub></b>	<b>R<sub>1</sub></b>	40	12	42	23	141	81
	<b>R<sub>2</sub></b>	34	11	40	25	128	111
	<b>R<sub>3</sub></b>	33	10	39	31	107	83

**Appendix VIII. Weight (g) of carcass cut of broiler under different treatment groups**

<b>Treatment</b>	<b>Replication</b>	<b>Breast</b>	<b>Thigh</b>	<b>Wing</b>	<b>Back</b>	<b>Drumstick</b>
<b>T<sub>1</sub></b>	<b>R<sub>1</sub></b>	558	184	87	212	186
	<b>R<sub>2</sub></b>	579	201	96	244	191
	<b>R<sub>3</sub></b>	555	195	86	228	176
<b>T<sub>2</sub></b>	<b>R<sub>1</sub></b>	520	190	80	201	180
	<b>R<sub>2</sub></b>	535	169	87	212	159
	<b>R<sub>3</sub></b>	526	181	92	210	171
<b>T<sub>3</sub></b>	<b>R<sub>1</sub></b>	513	191	72	198	165
	<b>R<sub>2</sub></b>	529	167	82	204	151
	<b>R<sub>3</sub></b>	512	165	77	195	155

**Appendix IX. Production cost of the birds at 28 days of rearing period.**

<b>Parameter</b>	<b>Amount (BDT)</b>
Day Old chick cost (120 no.)	3120
Feed cost (6bag+Maize+Soymeal))	16500
Litter cost	900
Medicine cost	500
Vaccine cost	1000
Others cost	3000
<b>Total</b>	<b>25020</b>

**Appendix X. Selling price of the birds under different treatment group**

<b>Treatment</b>	<b>Replication</b>	<b>NO. of bird</b>	<b>Average Live body weight kg</b>	<b>Selling price (BDT) @ 140 TK/ Kg live weight)</b>	<b>Total selling price (per replication)</b>
<b>T<sub>1</sub></b>	<b>R<sub>1</sub></b>	13	1.860	260.40	3385.20
	<b>R<sub>2</sub></b>	13	1.965	275.10	3576.30
	<b>R<sub>3</sub></b>	13	1.956	273.84	3559.92
<b>T<sub>2</sub></b>	<b>R<sub>1</sub></b>	13	1.853	259.42	3372.46
	<b>R<sub>2</sub></b>	13	1.804	252.56	3283.28
	<b>R<sub>3</sub></b>	12	1.850	259.00	3367.00
<b>T<sub>3</sub></b>	<b>R<sub>1</sub></b>	13	1.808	253.12	3290.56
	<b>R<sub>2</sub></b>	13	1.784	249.76	3246.88
	<b>R<sub>3</sub></b>	12	1.744	244.16	2929.92
<b>Total sell</b>					<b>30011.52</b>

**Appendix XI. Economic impact of treatments on broiler production**

<b>Treatment</b>	<b>Replication</b>	<b>Feed cost (BDT) Per Bird</b>	<b>Common Expenditure (BDT) Per Bird</b>	<b>Total Expenditure(BDT) Per Bird</b>
<b>T<sub>1</sub></b>	<b>R<sub>1</sub></b>	126.20	71	197.20
	<b>R<sub>2</sub></b>	132.80	71	203.80
	<b>R<sub>3</sub></b>	125.15	71	196.15
<b>T<sub>2</sub></b>	<b>R<sub>1</sub></b>	116.45	71	187.45
	<b>R<sub>2</sub></b>	119.85	71	190.85
	<b>R<sub>3</sub></b>	119.85	71	190.85
<b>T<sub>3</sub></b>	<b>R<sub>1</sub></b>	114.10	71	185.10
	<b>R<sub>2</sub></b>	115.20	71	186.20
	<b>R<sub>3</sub></b>	118.20	71	189.20

**Appendix XII. Net return of the birds under different treatment groups**

<b>Treatment</b>	<b>Replication</b>	<b>Receipt per bird @ 140 TK./ Kg LW)</b>	<b>Total Expenditure(BDT) Per Bird</b>	<b>Profit per bird (BDT)</b>	<b>Benefit Cost Ratio</b>
<b>T<sub>1</sub></b>	<b>R<sub>1</sub></b>	260.40	197.20	63.20	1.32
	<b>R<sub>2</sub></b>	275.10	203.80	71.30	1.35
	<b>R<sub>3</sub></b>	273.84	196.15	77.69	1.40
<b>T<sub>2</sub></b>	<b>R<sub>1</sub></b>	259.42	187.45	71.97	1.38
	<b>R<sub>2</sub></b>	252.56	190.85	61.71	1.32
	<b>R<sub>3</sub></b>	259.00	190.85	68.15	1.35
<b>T<sub>3</sub></b>	<b>R<sub>1</sub></b>	253.12	185.10	68.02	1.36
	<b>R<sub>2</sub></b>	249.76	186.20	63.56	1.34
	<b>R<sub>3</sub></b>	244.16	189.20	54.96	1.29

**Appendix XIII. Effects of dietary crude protein on immune parameters of broiler**

<b>Treatment</b>	<b>T<sub>1</sub></b>			<b>T<sub>2</sub></b>			<b>T<sub>3</sub></b>		
<b>Replication</b>	<b>R<sub>1</sub></b>	<b>R<sub>2</sub></b>	<b>R<sub>3</sub></b>	<b>R<sub>1</sub></b>	<b>R<sub>2</sub></b>	<b>R<sub>3</sub></b>	<b>R<sub>1</sub></b>	<b>R<sub>2</sub></b>	<b>R<sub>3</sub></b>
<b>Hemoglobin(g/dl)</b>	10.12	6.73	12	10.79	14.24	12.63	8.40	9.79	7.86
<b>WBC(/cumm)</b>	16300	15800	12900	18200	17100	18700	14100	16600	11900
<b>RBC(/cumm)</b>	4.98	3.65	5.31	4.67	5.41	5.32	3.99	4.09	3.29
<b>Platelet</b>	310000	460000	385000	520000	290000	460000	370000	415000	610000
<b>Neutrophils(%)</b>	78	75	79	81	76	72	84	81	84
<b>Lymphocytes(%)</b>	17	20	14	13	18	20	11	11	11
<b>Monocytes(%)</b>	01	02	01	01	02	03	03	01	01
<b>Eosinophil(%)</b>	04	03	06	05	04	05	02	07	04
<b>PCV(%)</b>	32.10	19.88	39.76	33.86	43.98	38.79	25.98	29.87	23.87
<b>MCV(fl)</b>	87.43	73.55	87.51	79.65	91.66	91.09	74.31	78.54	71.60
<b>MCH(pg)</b>	28.90	25.87	29.88	28.90	30.98	30.54	26.87	28.90	24.30
<b>MCHC(g/dl)</b>	29.76	25.81	30.72	32.19	33.09	33.21	30.51	32.19	28.51