

**EFFECTS OF AZOLLA ON GROWTH, CARCASS AND  
HEMATOLOGICAL CHARACTERISTICS IN JAPANESE QUAIL**

**A Thesis**

**By**

**MD. RASEL PARVEZ**

**REGISTRATION NO. 1605192**

**SESSION: 2016-2017**

**SEMESTER: JANUARY–JUNE, 2018**

**MASTER OF SCIENCE (MS)**

**IN**

**PHARMACOLOGY**



**DEPARTMENT OF PHYSIOLOGY AND PHARMACOLOGY  
HAJEE MOHAMMAD DANESH SCIENCE AND TECHNOLOGY  
UNIVERSITY, DINAJPUR-5200**

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*Submitted to the*

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**June, 2018**

*Dedicated*  
*To*  
*My Beloved Parents*

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## ABSTRACT

This study was conducted at the Department of Physiology and Pharmacology, Hajee Mohammad Danesh Science and Technology University (HSTU), Dinajpur to investigate the effects of Azolla on growth, carcass characteristics and hematologic parameters of Japanese quail. A total of 40, fourteen days old quails were assigned into four groups T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub>. Group T<sub>0</sub> was considered as control, fed only with commercial quail ration. Groups T<sub>1</sub>, T<sub>2</sub>, and T<sub>3</sub> commercial quail ration were supplemented with 3%, 5% and 7% Azolla respectively. Live body weight, body weight gain, feed consumption, feed conversion ratio and carcass characteristics were observed and hematologic parameters were determined at the age of 21 and 42 days of quail. Live body weights were increased significantly ( $p < 0.01$ ) in all treated groups in compare to the control and the highest was found in the group T<sub>2</sub> supplement with 5% Azolla. Feed consumption were more or less similar among the control and treatment groups. Feed conversion ratio were increased significantly ( $p < 0.01$ ) in all treated groups than the control. Dressing percentage, breast and thigh weights were higher among the treatment groups compared to the control and differed significantly ( $p < 0.01$ ). Liver and gizzard weights were non significantly increased in treatment groups. No significant ( $p > 0.05$ ) differences were observed among the groups for Total Erythrocyte Count, Packed Cell Volume, Hemoglobin concentration and Erythrocyte Sedimentation Rate values. But Total Erythrocyte Count, Packed Cell Volume and Hemoglobin values were determined slightly higher in treatment groups compared to the control. Whereas the Erythrocyte Sedimentation Rate values were slightly higher in the control group. The present study reveals that the supplementation of 5% Azolla in the quail ration may be used to improve the performance in quail.

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## LIST OF ABBREVIATIONS

Conc.	:	Concentration
Cu mm	:	Cubic millimeter
CP	:	Crude Protein
CF	:	Crude Fiver
ESR	:	Erythrocyte Sedimentation Rate
FCR	:	Feed Conversion Ratio
<i>et al.</i>	:	Associates
g	:	Gram
GDP	:	Gross Domestic Product
Hb	:	Hemoglobin
i.e.	:	That is
J.	:	Journal
Kg	:	Kilogram
Mg	:	Milligram
ml	:	Milliliter
mm <sup>3</sup>	:	cubic millimeter
No.	:	Number
PCV	:	Packed Cell Volume
RBC	:	Red Blood Cell
SE	:	Standard Error
SM	:	Sample Mean
TEC	:	Total Erythrocyte Count
Vol.	:	Volume
µg	:	Microgram
%	:	Percent
&	:	And
@	:	At the rate of
<	:	Less than
>	:	Greater than
±	:	Plus minus

# CHAPTER I

## INTRODUCTION

Bangladesh is densely populated country and growth of population is increasing very fast in comparison to its land size, which increases the pressure on people's basic need. The economy of Bangladesh significantly depends on Agriculture. Our agriculture primarily depends on Livestock. Livestock provides new raw material for industry, serves a social security for the rural peoples, and provides security against crop failure or damage during draught or cyclone. The contribution of the livestock sub-sector to GDP at constant prices was 1.66% in the fiscal year 2015-16 (BER, 2017). The poultry meat alone contributes a substantial 37% of the total meat production in Bangladesh (Begum *et al.* 2011). The annual Avg. a deficit of chicken egg is 6939 million numbers and the annual average deficit of meat is 3.81 million metric ton (Andrew, 2003). With the rapid increase in total population, the demand for poultry products has been increasing the growing demand for poultry products, the development of poultry industry is very important. In this respect, commercial quail production seems to be one of the possible alternate sources of protein foods. On commercial basis the quail farming has many advantages in comparison to any other small animal for home food production (Akram *et al.* 2008).

Quail is the smallest and the latest domesticated poultry species. There are about 131 species and 17 to 18 varieties of wild quail found all over the world, of which Japanese, Bobwhite, King and Stable quail are most important. Japanese quail, a small-domesticated avian species, has assumed importance worldwide as bird and is commercially exploited for meat and egg production, (Wilson *et al.* 1961). Japanese quails are the natural inhabitant of Japan. Quails are reared in Japan from the time immemorial. The scientific name of Japanese quail is *Coturnix coturnix japonica* under the class aves and family Phasianoidea (Hashanuzzaman, 2013)

.The popularity of quail husbandry is increasing all over the world. In Bangladesh quail was introduced for the first time in 1988 (Das, 2004). Quail farming for egg and meat is quite popular in, Asia and some European countries. Only Bobwhite quail and Japanese quail have been domesticated for commercial purposes and in Bangladesh, these two are commercially available. Quail farming is very profitable like other farming ventures, such as chicken, turkey or duck farming business. Almost all types of weather conditions are suitable for starting

quail farming business. Meat and eggs of quail are very tasty and nutritious. Quail eggs are very nutritious than other poultry eggs. Because quail eggs contain comparatively more protein, phosphorus, iron, vitamin A, B<sub>1</sub> and B<sub>2</sub>. Besides, scientists developed many quail lines e.g. white egg shell line, meat line etc. Japanese quail, a recently introduced economic avian species is ideally suited for meat and egg under intensive management due to their low maintenance cost, early sexual maturity, higher exponential growth, higher heat tolerance, fitness for higher density rearing, higher disease resistance and higher egg production than that of other poultry species. Short generation interval and quick business returns and the requirement of low investment attracting people to rear them. It appears that quail rearing may be important to the chicken when chicken survived in hostile climates and also for havoc like avian influenza and salmonellosis. The climate and natural condition of Bangladesh are very suitable for quail rearing. Quail can be reared in this country throughout the year with a good performance in meat and egg production. It has a shorter life cycle and its production requires less capital and land (Vali *et al.* 2005).

Being an agricultural country the government of Bangladesh has shifted policy emphasis on poultry rearing. Quail supplies meats, eggs, and extra income also. The quail farming has the unique advantage of tapping the growing market demand for poultry products as a supplement of chicken and duck farming (Sultana *et al.* 2007). Nowadays a large number of quail farms have been established in Bangladesh to supply quail meats in hotels, shops, and household consumption as its demand is increasing day by day. Japanese quail is the smallest avian species farmed for meat production (Vali, 2008). The meat from broiler quail is very delicate and tasty. It is considered as a superior item in different restaurant and homes. The broiler quail attains 140-150 gm body weight within 5 weeks of age and yields 72.5 % carcass for consumption (Das, 2004). Success in poultry farming depends on scientific breeding, feeding, management and disease control of the flocks. There is a relationship of Japanese quails (heavy body weight) line to dietary energy levels and graded essential amino acid levels on growth performance and immune competence (Kaur *et al.* 2008).

The profitability in quail farming is possible by better management due to the above reasons. Reports on quail growth and body composition are numerous. The better growth performance and meat quality of broiler quail (Japanese quail) are supported by the findings of (Kaur *et al.* 2008 and Vali, 2008).

Today a large number of educated people are looking for self-employment. The time has now come for creating alternative employment opportunities for these educated people. The self-employment scheme is one probable answer and quail farming seems to be a promising enterprise in this direction. It is hoped that quail farming will be recognized as popular poultry sector one day in our country. Because of the climate and natural condition of Bangladesh is also very suitable for quail rearing. Quail can be reared in this country throughout the year and shows a good performance in meat and egg production. It has a shorter life cycle and its production requires less capital and land. Quail may be a source of income in addition to chicken and ducks for its immense potentiality for meat and egg production (Paul and Sarker, 1992).

Escalating prices of poultry feed which alone accounts of 65 to 70% of the total production cost is detrimental to the growth of poultry sector. Now a days the prices of crucial feedstuff have almost doubled. This is threatening the poultry industry to run on a razor-blade and requires immediate attention. Hence, several attempts are being constantly made by the poultry producers and Nutritionists to seek the alternative feed resources for economic consequence. FAO program focuses on increasing the feed base production systems to locally available feed resources in developing countries (Sansoucy, 1993).

Recently there is an increased emphasis in the use of aquatic plants in poultry rations because the protein and other nutrient content in them are comparatively rich than that of leguminous plants. Among the aquatic plants floating fern, the water fern *Azolla* (*Azolla pinnata*) is an unconventional feed ingredient. *Azolla* is a free floating fresh water fern belonging to the family *Azollaceae* and order *Pteridophyta*. There are six species of *Azolla*. It is commonly found in tropics and sub-tropics. It grows naturally in stagnant water of drains, canals, ponds, rivers, haors-baors, marshy lands. *Anabaena-Azollae*, living in the cavity of *Azolla* leaf, can fix high amount of atmospheric dinitrogen due to presence of symbiotic algae in the leaves (Becking, 1979). *Azolla* is rich in protein, total protein and other constituents in *Azolla* are minerals, chlorophyll, carotinoids, amino acids, vitamins etc. It is also a potential source of nitrogen and is a potential feed ingredient for livestock (Lumpkin, 1984 and Pannerker, 1988). It contains almost all essential amino acids, minerals such as iron, calcium, magnesium, potassium, phosphorus, manganese etc. apart from appreciable quantities of vitamin A precursor beta carotene and vitamin B<sub>12</sub>. It is capable of assimilating algae in its leaves. It is also found to contain probiotics and biopolymers (Pillai *et al.* 2005).

The water fern *Azolla*, grows in association with the blue-green algae, *Anabaena Azollae*, is considered to be the most promising because of the easiness of cultivation, high productivity and good nutritive value (Singh and Subudhi, 1978). *Azolla* is one of the plant resources with high biomass and protein production. *Azolla pinnata* was used as feedstuff in broiler chicken (Querubin *et al.* 1986 and Parthasarathy *et al.* 2002) and laying hens (Khatun, 1996). Poultry being the monogastric, lacks fiber degrading enzymes and hence the use of exogenous enzymes facilitates the degradation of these polymers, thereby, making available nutrients which would otherwise get excreted (Saxena *et al.* 2006).

Considering the above facts, experiments were designed on growth, carcass and hematological characteristics in Japanese quail.

- To know the live body weight, feed consumption and feed conversion ratio.
- To evaluate the dressing percentage, breast, thigh, liver, gizzard and heart weight.
- To determine the values of Total Erythrocyte Count, Packed Cell Volume, Hemoglobin Concentration and Erythrocyte Sedimentation Rate.



## CHAPTER II

### REVIEW OF LITERATURE

The chapter represents the review of relevant literatures, consisting of the effect of Azolla on growth, carcass characteristics and hematologic parameters of quail. Many researchers have been conducted researches in these topics in chicken. But limited research work has been performed in case of quail.

#### **2.1 Azolla**

Azolla (mosquito fern, duckweed fern, fairy mass and water fern) is a floating fern in shallow scale like leaves, with their roots hanging in the water. They are extremely reduced in form and water. It floats on the surface of water by means of numerous, small, closely overlapping specialized, looking nothing like conventional fern but more resembling duckweed or some mosses. Azolla form a symbiotic relationship with the blue green algae, *Anabaena Azollae* which fixes atmospheric nitrogen and convert to plant nitrogen. This had led to the plant being dubbed a “super plant”, as it can readily colonize areas of fresh water, and grow at great speed doubling its biomass every two to three days. Each leaf is bilobed, the lower, achlorophyllous lobe ensuring floatation and the upper, chlorophyllous one developing a cavity that remains in contact with the external environment through structurally sophisticated pore. The cyanobacterium *Anabaena Azollae* occurs as filaments located on the plant stem apexes and inside the leaf cavities (Vanhove and Lejeune, 2002).

#### **2.2 Distribution and Availability of Azolla**

Azolla has been used for centuries in Asia as green manure, as a fertilizer for rice fields and supplement in diets for pigs, poultry and fish (Moore, 1969 and Lumpkin, 1984). Azolla is widely distributed and is found in ponds, ditches and channels containing stagnant water with a temperature range of 15- 35 °C (Singh and Subudhi, 1978). Azolla can be used as food, mosquito inhibitor, green manure, herbicide, water saver, water purifier, nitrogen fertilizer saver, drugs and for reclaiming saline soils (Vanhove and Lejeune, 1996). Fresh Azolla is used in the preparation of compost. Since the fern has an excellent carbon nitrogen ratio, it decomposes rapidly and accelerates the decomposition of other organic residues inside the compost pit and used as a bio-fertilizer in coffee plantations (Anand and Geeta, 2007).

## **2.3 Chemical Composition and Nutritive Value of Azolla**

### **2.3.1 Crude Protein**

On proximate analysis of Azolla constituent on dry matter basis the crude protein were 24 to 34, 26.60, 25.78, 24.06, 21.4, 21.4,  $21.0 \pm 0.39$ , 21.37 and 28.24 percent, reported by Singh and Subudhi (1978), Sreemanannaryan *et al.* (1993), Basak *et al.* (2002), Chatterjee *et al.* (2013), Alalade and Iyayi (2006), Cherryl *et al.* (2014), Oladapo *et al.* (2007), Giridhar *et al.* (2012), Parashuramulu and Nagalakshmi (2013) and Indria and Ravi (2014), respectively.

### **2.3.2 Dry Matter**

On proximate analysis of Azolla constituent on dry matter basis the dry matter content of Azolla was 4.23 percent (Indria and Ravi 2014). The composition of Azolla (%) on dry matter basis the dry matter was  $4.99 \pm 0.16$  percent (Giridhar *et al.* 2012). whereas Cherryl *et al.* (2014) analyzed sun dried azolla sample for proximate principles. The dry matter (DM) content of sun dried azolla meal was 89.73 percent.

### **2.3.3 Total Ash**

On proximate analysis of Azolla constituent on dry matter basis the total ash were 20.7, 10.5, 11.3, 15.76, 19.47, 16.2, 24.26, 16.2, 14.80 percent reported by Samanta and Tamang.(1995), Singh and Subudhi. (1978), Sreemanannaryan *et al.* (1993), Basak *et al.* (2002), Chatterjee *et al.* (2013), Alalade and Iyayi (2006), Cherryl *et al.* (2014), Oladapo *et al.* (2007) and Indria and Ravi (2014), respectively.

### **2.3.4 Crude Fiber**

On proximate analysis of Azolla constituent on dry matter basis the crude fiber were 9.1, 11.3, 13.44, 12.7, 14.7, 12.7, 12.5, 22.25 percent, reported by Singh and Subudhi (1978), Sreemanannaryan *et al.* (1993), Chatterjee *et al.* (2013), Alalade and Iyayi (2006), Cherryl *et al.* (2014), Oladapo *et al.* (2007), Parashuramulu and Nagalakshmi (2013) and Indria and Ravi (2014), respectively.

**Table 1: Chemical Composition of Azolla on Dry Matter Basis**

<b>Constitute</b>	<b>Alalad and Iyayi (2006)</b>	<b>Lukiwati <i>et al.</i> (2008)</b>	<b>Prasanna <i>et al.</i> (2011)</b>	<b>Chatterjee <i>et al.</i> (2013)</b>	<b>Kavya, K. (2014)</b>
<b>Dry matter</b>	-	-	94.50	-	4.371
<b>Organic matter</b>	-	-	-	80.53	82.16
<b>Total ash</b>	16.2	28.7	16.21	19.47	17.84
<b>Crude protein</b>	21.4	23.7	23.44	24.06	21.66
<b>Crude fiber</b>	12.7	15.0	12.37	13.444	15.15
<b>Ether extract</b>	2.7	2.93	2.93	3.27	4.41
<b>NFE</b>	47	31.1	-	37.71	40.94
<b>Calcium</b>	1.16	2.07	01.28	0.8-4.99	1.64
<b>Phosphorus</b>	1.29	0.77	1.16	0.3-1.3	0.34
<b>Potassium</b>	1.25	-	-	-	2.71
<b>Copper</b>	16.74	17.66	-	-	9.1
<b>Manganese</b>	174.42	-	-	-	2418
<b>Zinc</b>	87.59	71.8	-	-	325
<b>Iron</b>	755.73	-	-	-	1569
<b>Cobalt</b>	-	-	-	-	8.11
<b>Chromium</b>	-	-	-	-	5.06
<b>Boron</b>	-	-	-	-	31
<b>Nickel</b>	-	-	-	-	5.33
<b>Lead</b>	-	-	-	-	8.1
<b>Cadmium</b>	-	-	-	-	1.2

**Table 2: Amino Acid Composition of Azolla**

<b>Amino acid(AA)</b>	<b>Alalade and Iyayi (2006)</b>		<b>Kavya (2014)</b>	
	AA % DM	AA% in CP	AA % DM	AA% in CP
<b>Lysine</b>	0.98	4.58	1.231	4.940
<b>Methionine</b>	0.34	1.59	0.431	1.657
<b>Cystine</b>	0.18	0.84	0.194	0.778
<b>Threonine</b>	0.87	4.07	1.164	4.671
<b>Arginine</b>	1.15	5.37	1.414	5.674
<b>Isoleucine</b>	0.93	4.35	1.160	4.655
<b>Leucine</b>	1.65	7.71	2.072	8.315
<b>Phenylalanine</b>	1.01	4.72	1.377	5.526
<b>Glycine</b>	1.00	4.60	1.341	5.318
<b>Serine</b>	0.90	4.21	1.124	4.510
<b>Valine</b>	1.18	5.51	1.445	5.799
<b>Alanine</b>	-	-	1.539	6.176
<b>Histidine</b>	-	-	0.488	1.956
<b>Proline</b>	-	-	1.032	4.141
<b>Aspartic Acid</b>	-	-	2.303	9.242
<b>Glutamic Acid</b>	-	-	2.740	10.995
<b>Total(WithoutNH3)</b>	-	-	21.037	84.418
<b>Ammonia</b>	-	-	0.509	2.043
<b>Total</b>	-	-	21.546	86.461

## 2.4 Cell Wall Constituents of Azolla

The NDF, ADF, hemicellulose, cellulose and lignin content of SDA ranged from 36.88 to 67.80%, 25.24 to 51.96%, 13.30 to 16.65%, 9.46 to 15.61 % and 8.08 to 28.24%, respectively.

## 2.5 Effect of Azolla on the Growth of Poultry

**Basak *et al.* (2002)** conducted an experiment was conducted with 120 seven days old Vencobb commercial broiler chicks and continued up to 42 days of age to determine the feasibility of Azolla (*Azolla pinnata*) as a feed ingredient in broiler ration. The broilers are randomly allotted to four dietary treatments; T<sub>1</sub> (control diet without Azolla meal), T<sub>2</sub> (diet with 5% Azolla meal), T<sub>3</sub> (diet with 10% Azolla meal) and T<sub>4</sub> (diet with 15% Azolla meal) diets replacing sesame meal by Azolla meal. Live weight, production number and protein efficiency were ( $p < 0.01$ ) significantly improved at the level off 5% Azolla meal in broiler ration. Feed conversion ratio and energy efficiency were significantly ( $p < 0.01$ ) improved in diet with 5% Azolla meal and control diet. It can be conducted that use of Azolla meal up to 5% in the broiler ration found to improve performance and may be used in broiler diet as a safe level. Azolla meal had no deleterious effect on the palatability of broiler diets. In another study the body weight of Rhode Island Red chicks on day 49 of feeding were 3445, 3532, 3590, 4081 and 3272 g for the group fed control, control with Spirodela, Lemna and Azolla mixture, or Azolla, Spirodela or Spirogyra diets, respectively (Shahjahan *et al.* 1992).

**Bhattacharyya *et al.* (2016)** conducted that one hundred and twenty, one week old, Cobb 400 broiler chickens were randomly distributed into three dietary treatments having four replicates each with ten birds. The birds of the control group (T<sub>1</sub>) were fed a basal diet (23.16% CP 1-3 weeks & 19.68% CP 3-6 weeks) while the other two groups were offered the treatment diets (T<sub>2</sub>& T<sub>3</sub>) replacing 4.50% or 5.50%, of the dry matter of the basal diet with dry *Azolla pinnata* powder on dry matter basis, respectively. Feeding Azolla meal did not significantly ( $p > 0.05$ ) affect the weekly body weight gain of the birds during the experimental period. Thus, it may be inferred that replacement of basal diet with dry *Azolla pinnata* meal on dry matter basis did not adversely affect the growth. In another study the partial replacement of SBM at 2.5 and 5 per cent levels by Azolla meal has no adverse effect on weekly body weights of broilers. However, in third, fourth and fifth weeks, the body weights were higher than that of control ( Dhumal *et al.* 2009).

**Rout et al. (2017)** studied the 150 day-old broiler chicks of either sex were distributed randomly in five dietary treatments with 3 replicates each, C<sub>1</sub>: basal diet, T<sub>1</sub>: basal diet + 10% dietary protein from Azolla, T<sub>2</sub>: basal diet + 10% dietary protein from Azolla + enzyme, T<sub>3</sub>: basal diet+ 15% dietary protein from Azolla, T<sub>4</sub>: basal diet + 15% dietary protein from *Azolla* + enzyme. The experiment was carried out for 92 days. Each of the Azolla fed group, compared to control, showed significantly ( $P \leq 0.05$ ) higher body weight. T<sub>4</sub> showed higher gains than T<sub>1</sub> till 4 th week whereas no significant ( $P \leq 0.05$ ) difference was recorded between the Azolla fed groups from 5 th week onwards. Azolla fed group registered significantly ( $P \leq 0.05$ ) higher consumptions than the control. However between the two non-enzymes groups T<sub>1</sub> showed higher ( $P \leq 0.05$ ) values than T<sub>3</sub>. Enzyme supplementation resulted in a lower consumption in 10% Azolla group while it led to a higher consumption in 15% Azolla group, and that between the two enzyme groups, group T<sub>4</sub> showed higher values ( $P \leq 0.05$ ) than group T<sub>2</sub>. Throughout the experiment feed efficiency of Azolla fed group showed higher than the control. It was concluded that Azolla could be included in the broiler ration up to 15% level which resulted in higher body weight gain and feed efficiency.

**Sujatha et al. (2013)** Forty-week old, 72 chicks were divided into two groups of 36 birds for the study. The control group was given commercial feed (basal diet) at the rate of 120 g per chick per day, while the experimental group was given raw Azolla, at the rate of 200 g per chick per day in separate feeder, in addition to 120 g of basal diet, from 45-60 weeks. The final body weight of the birds ( $1560.0 \pm 26.8$  g), and gain in body weight/ day ( $2.77 \pm 1.78$  g) were higher, and the feed conversion ratio ( $36.10 \pm 1.19$  g) was better in Azolla supplemented group than the birds in control group, during the experimental period ( $p < 0.05$ ).

**Parthasarathy et al. (2002)** reported no significant difference in body weight gain of broilers on basal and 5 per cent Azolla diets whereas higher levels of inclusion resulted in significant ( $p < 0.01$ ) reduction in body weight gain (1752, 1676, 1650 g for 10, 15 and 20 per cent Azolla based diets, respectively) thereby concluded that the Azolla could safely be included at 5 per cent level, replacing 2.6 per cent wheat bran and 2.4 per cent fish meal without any adverse effect. In another study conducted that the cumulative body weight gain of broilers at sixth week of age in groups fed 0, 1.5, 3.0 and 4.5 percent Azolla incorporated diets were statistically similar at 2123, 2123, 2066 and 2040g, respectively indicating that inclusion of Azolla up to 4.5 per cent in rations did not have any influence on body weight in broiler chicken (Balaji et al. 2009).

**Becerra et al. (1995)** studied the performance of duck by incorporating 15, 30, 45 and 60 percent Azolla microphylla in boiled soya bean diet supplying 15.2, 17.9, 24.4 and 30.3 percent of the total protein. The daily weight gain was found to decrease with increasing levels of Azolla when this exceeded 15 per cent of the dietary protein. Feed intake and FCR deteriorated with increasing level of Azolla ( $p < 0.01$ ) and Gavina (1993) conducted that feeding of duck supplemented with fresh Azolla with 60 percent and 40 percent as the percentage of commercial feed in their diet. The results revealed that there was a significant difference in the average gain in the final weight of the duck, concluding that commercial feed could be reduced considerably in the ducks diet and thus encouraging Azolla as feed supplement.

## **2.6 Effect of Sun Dried Azolla on the Performance of other Livestock**

**Querubin et al. (1986)** conducted a trial to evaluate the feeding value of Azolla (*A. microphylla*) silage in growing pigs by randomly allotting into three treatments. Pigs in treatment 1 were fed the control diet while those in treatment 2 and 3 were fed diets with Azolla silage A (70% Azolla + 25% corn) and Azolla silage B (70% Azolla + 20% corn + 5% molasses), respectively. Both silages were fed in combination with a concentrate mixture at the rate of 20% of the diet. The pigs were fed their respective diets for 35 days. Average weight gain, feed intake and feed efficiency of the pigs fed diets with Azolla silage did not differ significantly from those fed the control diet and Gavina (1993) reported no significant difference in the average final weight, feed consumption and feed conversion efficiency of pigs fed diets containing Azolla up to 40 percent and Duran (1994) reported that the aquatic plant *Azolla filiculoides* can replace up to 20 per cent of the soya bean protein with no deterioration in performance of growing and finishing pigs.

**Sreemannaryana et al. (1993)** An experiment was conducted in New Zealand and Russian Grey Giant rabbits by dividing into 4 groups. Group 1 rabbits were fed a commercial pellet feed *ad libitum* while fresh Azolla at 10, 15 and 20% level (w/w) replacing pellet feed on dry matter basis was offered to rabbits in groups II, III and IV, respectively. Replacement of pelleted feed by fresh Azolla at varying levels did not affect the weight gains recorded at weekly intervals. Inclusion of Azolla up to 20% level did not affect the palatability.

**Indira et al. (2009)** conducted a trial with two groups of male buffalo calves. Control group was fed on hybrid Napier, paddy straw and concentrate mixture as per their requirements. In the second group, 50% of GNC nitrogen of control diet was replaced by Azolla nitrogen. The average daily gain, average dry matter intake and average feed conversion efficiency for two groups was 240 and 294 g, 3.21 and 2.91 kg/100 kg body weight, 13.24 and 10.07%, respectively. Higher average daily gain was recorded in experimental group. Feed conversion efficiency was significantly ( $p < 0.01$ ) superior in experimental diet.

**Wadhvani et al. (2010)** A trial was carried out in Marwari, Patanwadi and Merino x Patanwadi weaner lambs (12.40 to 14.68 kg) with three dietary treatments viz. T1: TMR-I Conventional, T2 : TMR-II supplemented non-conventional group I (Azolla- 10%) and T3 : TMR-III supplemented non-conventional group II (Azolla- 20%). The treatment groups did not differ from each other in their final body weights. The body weight gain and body measurements were significantly ( $p < 0.05$ ) influenced by the treatments. In another study indicated that the sun-dried azolla can be incorporated up to 20 percent of concentrate mixture of goat kids without any deleterious effects on the performance, digestibility of various nutrients, carcass characteristics, haematological and biochemical parameters (Tamang and Samanta, 1993)

**Reddy et al. (2010)** conducted a trial in adult Nellore Sheep reared under intensive and semi intensive systems of management by feeding four iso- nitrogenous diets formulated using Azolla (*Azolla pinnata*) as low protein supplement and Sheanut cake (*Vitellaria paradoxa*) as energy supplement. Results indicated significant ( $p < 0.05$ ) higher dry matter intake (kg) per day for experimental concentrate mixtures T2 (concentrate diet with replacement of 30 parts of GNC with Azolla) and T3 (concentrate diet replacement of 16 parts of Rice bran with Sheanut cake) compared to T4 (concentrate diet containing replacement of 30 parts of GNC with Azolla and 16 parts of Rice bran with Sheanut cake) in both the system of management. The dry matter intake/100 kg body weight was significantly ( $p < 0.01$ ) higher in semi intensive system than intensive system.

## **2.7 Effect of Azolla on Nutrient Digestibilities**

**Alcantra and Querubin (1985)** indicated that nutrient digestibility's were not affected in broilers up to 15% level of inclusion of Azolla in the ration. They also indicated that broilers



can readily digest crude fibre in Azolla but not the crude fibre in rice bran. In another study carried out with two species of Azolla in layers, it is reported that the digestibility of CP, CF, EE and NFE were not significantly influenced by the Azolla species. Inclusion of increasing level of Azolla meal in the diet did not significantly affect the digestibility of CP, CF and NFE (Bastian, 1987).

**Tamang and Samanta (1993)** conducted a digestion trial in goats and reported that sun dried Azolla can be incorporated up to 20 per cent of concentrate mixture without any affect on the digestibility of nutrients. In another study carried out with the digestibility (%) of OM, CF, EE and NFE was similar while that of DM and CP decreased significantly ( $p < 0.05$ ) in weaner lambs fed diets containing 0, 10 and 20% Azolla. However, there was no significant difference between 10 and 20% level of inclusion of Azolla. (Wadhvani *et al.* 2010-) and Indira *et al.* (2009) reported significantly ( $p < 0.01$ ) higher digestibility coefficient of DM, CP, CF, EE, NDF, ADF and NFE in cattle fed experimental diet where 50% GNC nitrogen is replaced by Azolla nitrogen compared to cattle fed control diet.

**Domínguez *et al.* (1996)** studied ideal and in vitro digestibility of three floating aquatic macrophytes (water hyacinth, duckweed and Azolla) in pigs and reported a sharp increase in N content with increasing levels of Azolla meal in the diet. No evident treatment effect on ideal digestibility of nutrients and energy was found although ideal digestibility of ash and organic matter seemed to decrease ( $p < 0.10$ ) in treatments where Azolla meal was included.

## **2.8 Carcass Characteristics**

**Samanta and Dey (1991)** showed that dressing yield in Japanese quail fed on rations containing 27 percent crude protein and 2700 kcal of ME /kg was 75.5 percent at seventh week of age. They also reported an eviscerated yield of 71.3 percent and giblet yield of 4.19 percent. Dhariwal *et al.* (2004) reported 73.65 percent dressed yield, 61.90 percent eviscerated yield and 8.37 percent giblet yield in non-selected control lines of Japanese quail at six week of age. Whereas Pakhira and Samanta (2005) observed dressing percentage of 69.88 and giblet yield of 8.29 percent at sixth week in Japanese quails.

**Basak *et al.* (2002)** conducted an experiment was conducted with 120 seven days old Vencobb commercial broiler chicks and continued up to 42 days of age to determine the

feasibility of *Azolla* (*Azolla pinnata*) as a feed ingredient in broiler ration. The broilers are randomly allotted to four dietary treatments; T<sub>1</sub> (control diet without *Azolla* meal), T<sub>2</sub> (diet with 5% *Azolla* meal), T<sub>3</sub> (diet with 10% *Azolla* meal) and T<sub>4</sub> (diet with 15% *Azolla* meal) diets replacing sesame meal by *Azolla* meal. Dressing percentage was significantly ( $p < 0.01$ ) increased on diet with 5% *Azolla* meal. Giblet percentage on dietary treatment T<sub>4</sub> significantly ( $p < 0.05$ ) increased than other treatments.

**Shrivastav *et al.* (1995)** reported eviscerated yield of 62.93 per cent and dressed yield of 90.46 per cent and giblet yield of 6.04 per cent in Japanese quail at five weeks of age whereas **Mandal *et al.* (1996)** reported dressed yield of 68.75 per cent in Japanese quail fed rations containing 26 per cent crude protein and 2600 kcal of ME/ kg and **Kumararaj *et al.* (1997)** reported a ready to cook yield of 73.20 per cent at six weeks of age in Japanese quail fed with ration having 26 per cent crude protein and 2700 kcal of ME / kg. He also reported a giblet yield of 8.16 per cent and blood and feather loss as 1.60 and 5.20 per cent, respectively.

**Bhattacharyya *et al.* (2016)** conducted that one hundred and twenty, one week old, Cobb 400 broiler chickens were randomly distributed into three dietary treatments having four replicates each with ten birds. The birds of the control group (T<sub>1</sub>) were fed a basal diet (23.16% CP 1-3 weeks & 19.68% CP 3-6 weeks) while the other two groups were offered the treatment diets (T<sub>2</sub>& T<sub>3</sub>) replacing 4.50% or 5.50%, of the dry matter of the basal diet with dry *Azolla pinnata* powder on dry matter basis, respectively. Dressing percentage was significantly higher ( $p < 0.05$ ) in T<sub>2</sub> compared to the other two treatment groups compared to control group. However, there was no significant difference among the treatment groups in the other carcass traits.

**Balaji *et al.* (2009)** reported that the per cent dressed yield, eviscerated yield and ready-to-cook yield in broilers were not influenced by dietary supplementation of dried *Azolla*. The per cent giblet yield of birds fed with 4.5% *Azolla* was significantly higher ( $p < 0.05$ ) than control and other treatments. Whereas **Dhumal *et al.* (2009)** reported that the carcass yield obtained from different groups of broilers fed on diets containing 0, 2.5 and 5% *Azolla* were 75.73, 75.89 and 74.95%, respectively and concluded that water fern *Azolla* could replace SBM in broiler ration up to 5% level without any adverse effect on carcass quality of broiler birds.

## 2.9 Hematologic Parameters

**Alagbe (2017)** studied that one hundred and sixty (160) Arbo-acre day old chick of mixed sex were divided into four groups with four replicates, each of ten birds in a completely randomized design. Treatment 1 was fed basal diet without Azolla –Moringa leaf mixture, treatment 2, 3 and 4 were fed basal diets supplemented with Azolla -Moringa leaf mixture at levels 5, 10 and 15% respectively. No significant influence ( $P>0.05$ ) were observed for some of the hematological parameters PCV, Hb and RBC at 42 days of the experimental period.

**Alalade *et al.* (2007)** investigated in the treatments of 0, 5, 10 and 15 percent, of Azolla in growing pullets and in packed cell volume (PCV) reported as 27.7, 28.3, 27.3 and 26.3 per cent, respectively. Whereas, in red blood cell (RBC) it was denoted that 2.01, 2.94, 2.33 and  $1.98 \times 10^6 /\text{mm}^3$ , respectively. Haemoglobin (Hb) content of blood were 8.47, 8.53, 7.80 and 7.20 g/dl respectively.

## CHAPTER III

### MATERIALS AND METHODS

The experiment was conducted at the poultry research unit under the department of Physiology and Pharmacology, Hajee Mohammad Danesh Science & Technology University, Dinajpur. The duration of experiment was 28 days.

#### 3.1 Collection and Management of Quails

At 14 days of age Japanese quails were collected from Abdul Hakim quail hatchery and farm Gaibandha. The body weights of assigned quails were taken with digital balance and data were recorded. The finally 40 quails were housed under normal husbandry condition and divided into 4 groups T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub>, and T<sub>3</sub> and reared in cage system. Here Group T<sub>0</sub> were kept control. Group T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub> were fed 3%, 5% and 7% Azolla supplementation respectively. All of them were fed with commercial crumbled plus mesh feed and fresh water ad-libitum.



**Figure 1: Japanese Quail in Experimental Cage**

## Layout of the Experiment

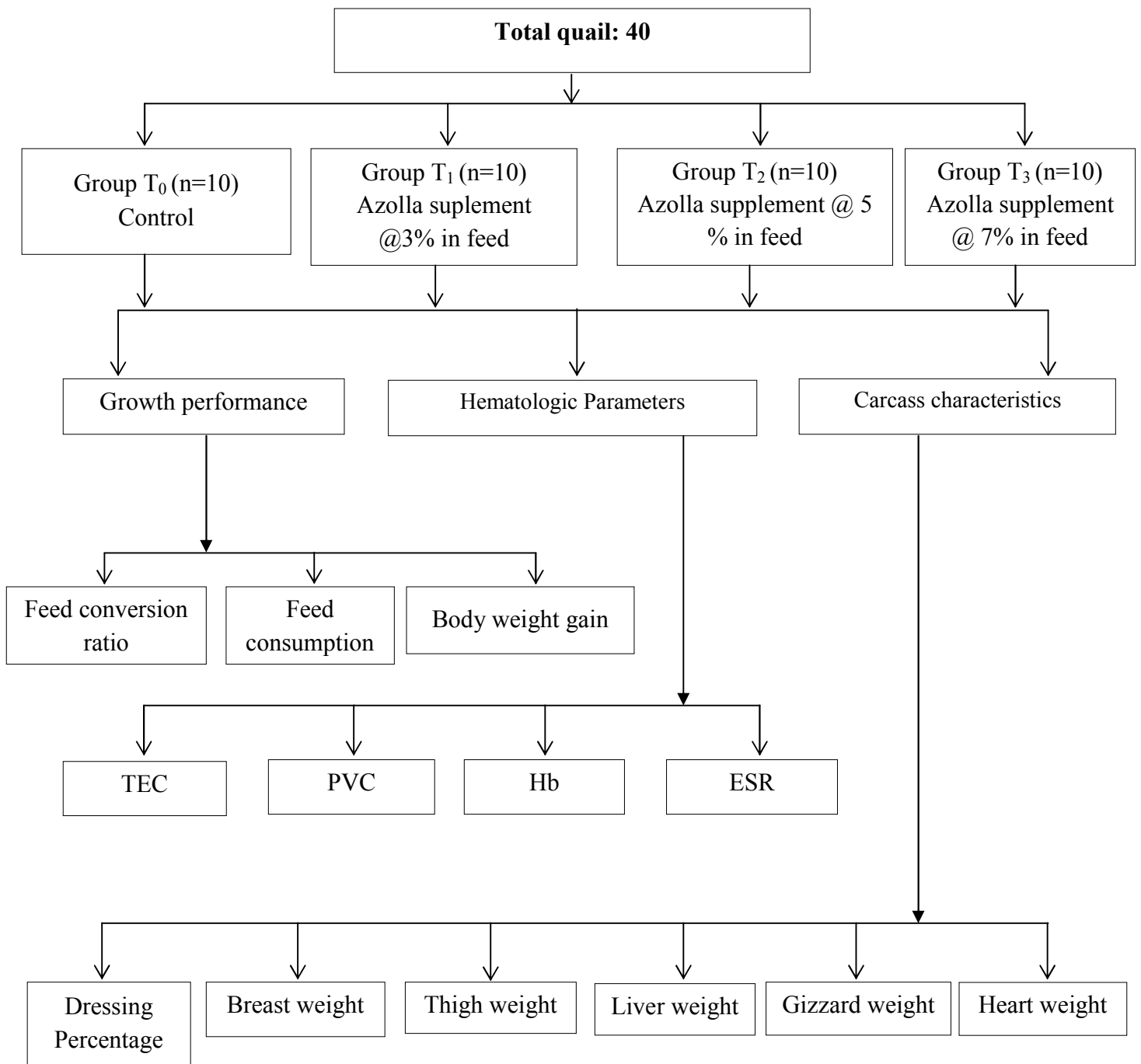


Figure 2: Layout of the Experiment

### 3.2 Collection of Azolla

Azolla were collected from few ponds located near the Baserhat, Dinajpur.



Figure 3: Collection of Azolla

### 3.3 Experimental Diets

Azolla were dried in the sun and grinded. The grinded Azolla were added with commercial quail ration and served to different groups.



Figure 4: Grinded Azolla



Figure 5: Experimental Diets

### 3.4 Growth Performance Parameters

Several parameters were studied to observe the effect of Azolla in growth performance of quail:

#### 3.4.1 Recording of Live Body Weight Gain

The body weights of birds were recorded at 7 days interval and the body weight gains were arrived at for each week. The body weight gain was also calculated on a cumulative basis. Accordingly, the body weight gains in different dietary groups were compared.



Figure 6: Measurement of Body Weight

### 3.4.2 Estimation of Feed Consumption

The daily feed offered to all individual groups was accurately recorded and at the end of each week and the residual amount of feed was weight and subtracted from the known weight of feed at the beginning of week. The product was divided by the total number of bird.

### 3.4.3 Calculation of Feed Conversion Ratio

The feed conversion ratio (FCR) expressed as the ratio of amount of feed consumed to the body weight gained under each group of bird.

$$\text{FCR} = \frac{\text{Amount of Feed Consumed}}{\text{Live Body Weight Gain}}$$

### 3.5 Carcass Characteristics

At the end of the experiment ( day 42), three birds from each group, were separated and starved for 12 hours however with a provision of plenty of water. Then immediately after recording their live body weights (pre-slaughter bird weight), the birds were slaughtered and the carcasses were subjected for the study of following parameters:

#### 3.5.1 Dressing Percentage

The slaughtered birds were defeathered, denecked and eviscerated along with two legs beneath the hock joint to observe the effect of various experimental diets on the dressing

percentage. The dressing percentage was calculated as the percent of the carcass weight obtained after removing the feathers, neck, legs and internal viscera, to its live body weight.

### 3.5.2 Organometry

From the sacrificed birds, different organs liver, gizzard and heart and the breast meat portion as well as thigh were carefully separated and weighed to observe the effect of different dietary treatments on growth and development of certain organs.

$$\text{Relative weight (g)} = \frac{\text{Organ Weight (g)}}{\text{Live Weight of Bird (g)}} \times 100$$



Figure 7: Postmortem Examination



Figure 8: Carcass of quail



Figure 9: Different organs of quail



### 3.6 Hematological Parameters:

Blood samples were collected from wing vein of quail of among the control and treated groups at 21<sup>th</sup> and 42<sup>th</sup> days to study the effect of the Azolla on Total Erythrocyte Count (TEC), Packed Cell Volume (PCV), Hemoglobin Concentrations (Hb) and Erythrocyte Sedimentation Rate (ESR) of quail.



Figure 10: Collection of Blood.

#### 3.6.1 Determination of Total Erythrocyte Count (TEC)

Total erythrocyte count was done following the method described by Lamberg and Rothstein (1977). Well-mixed blood sample was drawn with red blood cell diluting pipette exactly up to 0.5 marks of the pipette. Outside of the tip of the pipette was wiped with cotton. Then the pipette was immediately filled with the red cell diluting fluid (Hayem's solution) up to 101 marks. The free end of the pipette was wrapped around with the rubber tube stretching to both the ends and held with thumb and middle finger. The content of the pipette was mixed thoroughly by shaking with 8-knot motion for 3-5 minutes. Then the counting chamber was placed with special cover glass under microscope using low power (10 x) objectives. After discarding 2 or 3 drops of fluid from the pipette, a small drop was placed to the edge of the cover glass on the counting chamber as the entire area under the cover glass was filled by the fluid. One-minute time was spared to allow the cells to settle on the chamber under the cover glass. Taking 5 larger squares (4 in the 4 corners and the central one) of the central large square, the cells were counted from all the 80 small squares (16 x 5) under high power objectives (45 x). After completion of counting, the total number of RBC was calculated as number of cells counted x 10, 000 and the result was expressed in million/ $\mu$ l of blood.

### **3.6.2 Determination of Packed Cell Volume (PCV)**

The citrated well mixed blood sample was drawn into special loading pipette (Wintrobe pipette). The tip of the pipette was inserted up to the bottom of a clean, dry Wintrobe hematocrit tube. Then the Wintrobe tube was filled from the bottom by pressing the rubber bulb of the pipette. As blood came out, the pipette was slowly withdrawn but pressure was continued on the rubber bulb of the pipette so as to exclude air bubbles. The tip of the pipette was tried to keep under the rising column of blood to avoid foaming and the tube was filled exactly to the 10 cm mark. Then the Wintrobe hematocrit tube was placed in the centrifuge machine and was centrifuged for 30 minutes at 3000 rpm. Then the hematocrit or PCV was recorded by reading the graduation mark; the percent volume occupied by the hematocrit was calculated by using the following formula as described by Lamberg and Rothstein (1977).

$$\text{PCV}\% = \frac{\text{Height of red cell volume in cm}}{\text{Height of total blood in cm}} \times 100$$

### **3.6.3 Determination of Hemoglobin Concentrations (Hb)**

The N/10 hydrochloric acid (HCl) was taken in a graduated tube up to 2 marks with the help of a dropper. Well-homogenized blood sample was then drawn into the Sahli pipette up to 20 cm. mark. The tip of the pipette was wiped with sterile cotton and the blood of the pipette was immediately transferred into the graduated tube containing hydrochloric acid. This blood and acid were thoroughly mixed by stirring with a glass stirrer. There was a formation of acid hematin mixture in the tube by hemolysing red blood cells by the action of HCl. The tube containing acid hematin mixture was kept standing in the comparator for 5 minutes. After that distilled water was added drop by drop. The solution was mixed well with a glass stirrer until the color of the mixture resembled to the standard color of the comparator. The result was read in daylight by observing the height of the liquid in the tube considering the lower meniscus of the liquid column. The result was then expressed in g %. The above procedure was matched by the Hellige hemometer method as described by Lamberg and Rothstein (1977).

### **3.6.4 Determination of Erythrocyte Sedimentation Rate (ESR)**

The fresh anticoagulant blood was taken into the Wintrobe hematocrit tube by using special loading pipette exactly up to 0 marks. Excess blood above the mark was wiped away by

sterile cotton. The filled tube was placed vertically undisturbed on the wooden rack for one hour. After one hour the ESR was recorded from the top of the pipette. The result was expressed in mm/in 1st hour.

### **3.7 Statistical Analysis**

The data were analyzed by analysis of variance using complete randomized design with factorial arrangement of time and treatments (Steel and Torrie, 1986). All analyses were performed by SPSS Program Version 22.

## CHAPTER IV

### RESULTS AND DISCUSSION

The present study was undertaken with a view to study the effect of Azolla supplementation in growth, carcass characteristics and hematologic parameters of quails. After doing the analysis, the findings of the present investigation was presented and discussed in this chapter under the following headings

#### 4.1 Effect of Azolla Supplementation on Growth Performance

##### 4.1.1 Live Body Weight

The observations for live body weight (g) means of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> groups after six weeks of the experimental period were 116.20 ±1.43 g, 123.20 ±1.16 g, 128.20±1.46 g and 119.00 ±1.00 g respectively (Table 3) and graphically presented at figure 11. So quails of Group T<sub>2</sub> (supplemented with 5% Azolla) got the maximum weight (p<0.01) followed by Group T<sub>1</sub> (supplemented with 3% Azolla) and group T<sub>3</sub> (supplemented with 7% Azolla ) among all of the experimental groups and the control group T<sub>0</sub> (without supplementation of Azolla) got the lowest body weight. This may be attributed to the beneficial effect of Azolla, possibly due to better utilization of protein and other nutrients. The improvement in body weight is similar to the finding of Basak *et al.* (2002) who observed significant (p<0.01) improvement in live weight of broiler chicks than control diet when they were fed with 5 per cent Azolla meal replacing sesame meal, while lower live weights were recorded in 10 and 15 per cent Azolla incorporated diets. Similarly, Naghshi *et al.* (2014) reported chickens fed diets containing 5% Azolla powder significantly (p<0.01) improved daily weight gain compared to control. Further, Balaji *et al.* (2009) reported that dietary inclusion of dried Azolla up to 4.5% level did not have any adverse effect on production performance of broiler chicken.

##### 4.1.2 Feed Consumption

The average feed consumption of quails from 14 to 42 days of age in different groups were presented in( Table 3) In the present study average feed consumption were 438 g , 440 g , 440 g and 439 g among the experimental groups T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> respectively. The quails of treatment groups T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> consumed slightly higher than control group T<sub>0</sub>. Similarly higher results were obtained by Dhumal *et al.* (2009) who observed feed consumption for

groups treated with azolla meal was slightly higher than control (A-983.79, B-1045 and C-1009 g).

#### **4.1.3 Feed Conversion Ratio**

The feed conversion ratio of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> groups at 42 days of the experimental period were  $4.88 \pm 0.10g$ ,  $4.56 \pm 0.09g$ ,  $4.33 \pm 0.05$  and  $4.75 \pm 0.08g$  respectively (Table 3) and graphically presented at figure 12. So quails of Group T<sub>2</sub> (supplemented with 5% Azolla) got the maximum weight ( $p < 0.01$ ) followed by Group T<sub>1</sub> (supplemented with 3% Azolla) and group T<sub>3</sub> (supplemented with 7% Azolla) among all of the experimental groups and the control group T<sub>0</sub> (without supplementation of Azolla) got the lowest body weight. This finding was similar to the finding of Basak *et al.* (2002) feed conversion ratio and energy efficiency were significantly ( $p < 0.01$ ) improved in diet with 5% Azolla meal than control diet. Sujatha *et al.* (2013) also reported chickens fed diets containing 5% Azolla powder significantly ( $p < 0.05$ ) improved feed conversion ratio compared to control.

**Table 3: Live Body Weight of Experimental Quails at 7 Days Interval and Weight Gain, Feed Consumption and Feed Conversion Ratio of Quail from 14 to 42 Days of Age.**

Days	T <sub>0</sub>	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	P-value	Level of significance
Day 14 Mean ± SD (g)	26.40 <sup>a</sup> ±0.68	26.60 <sup>a</sup> ± 0.68	26.60 <sup>a</sup> ± 0.24	26.40 <sup>a</sup> ± 0.87	0.992	NS
Day 21 Mean ± SD (g)	45.60 <sup>a</sup> ± 1.03	46.80 <sup>a</sup> ± 0.58	47.60 <sup>a</sup> ± 0.75	45.80 <sup>a</sup> ± 0.49	0.236	NS
Day 28 Mean ± SD (g)	65.60 <sup>a</sup> ± 1.44	68.40 <sup>ab</sup> ± 1.21	71.80 <sup>b</sup> ± 1.46	66.20 <sup>a</sup> ± 1.28	0.021	*
Day 35 Mean ± SD (g)	91.80 <sup>a</sup> ± 1.39	96.20 <sup>b</sup> ± 0.86	101.80 <sup>c</sup> ± 1.28	93.00 <sup>ab</sup> ± 1.14	0.000	**
Day 42 mean ± SD (g)	116.20 <sup>a</sup> ± 1.43	123.20 <sup>b</sup> ± 1.16	128.20 <sup>c</sup> ± 1.46	119.00 <sup>a</sup> ± 1.00	0.000	**
Weight gain (g)	89.80 <sup>a</sup> ± 1.83	96.60 <sup>a</sup> ±1.81	101.60 <sup>a</sup> ±1.29	92.60 <sup>a</sup> ± 1.57	.001	**
Feed consumption (g)	438 <sup>a</sup>	440 <sup>a</sup>	440 <sup>a</sup>	439 <sup>a</sup>		
Feed conversion ratio (FCR) g feed consumed/g weight gain	4.88 <sup>a</sup> ± 0.10	4.56 <sup>ab</sup> ± 0.09	4.33 <sup>b</sup> ± 0.05	4.75 <sup>c</sup> ± 0.08	.001	**

Different letters in the same row indicate the significant difference

\*= Significant at 5% level of probability (p < 0.05)

\*\*= Significant at 1% level of probability (p < 0.01)

NS= Non-Significant (P > 0.05)

Here, T<sub>0</sub> indicates commercial diet without Azolla; T<sub>1</sub> indicates Commercial diet with 3% Azolla; T<sub>2</sub> indicates Commercial diet with 5% Azolla; T<sub>3</sub> indicates Commercial diet with 7% Azolla.

## Body Weight of Experimental Quail

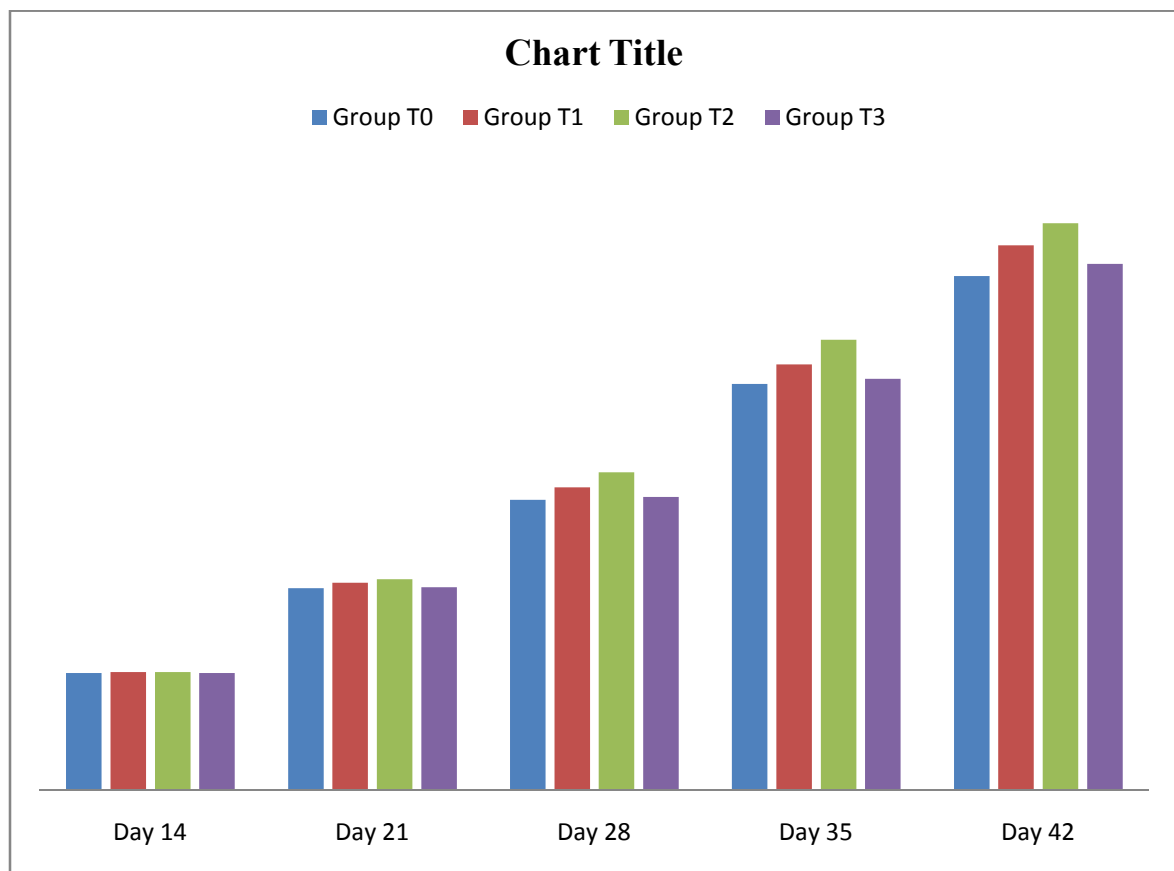


Figure 11: Body Weight of Experimental Quail

## Feed Conversion Ratio

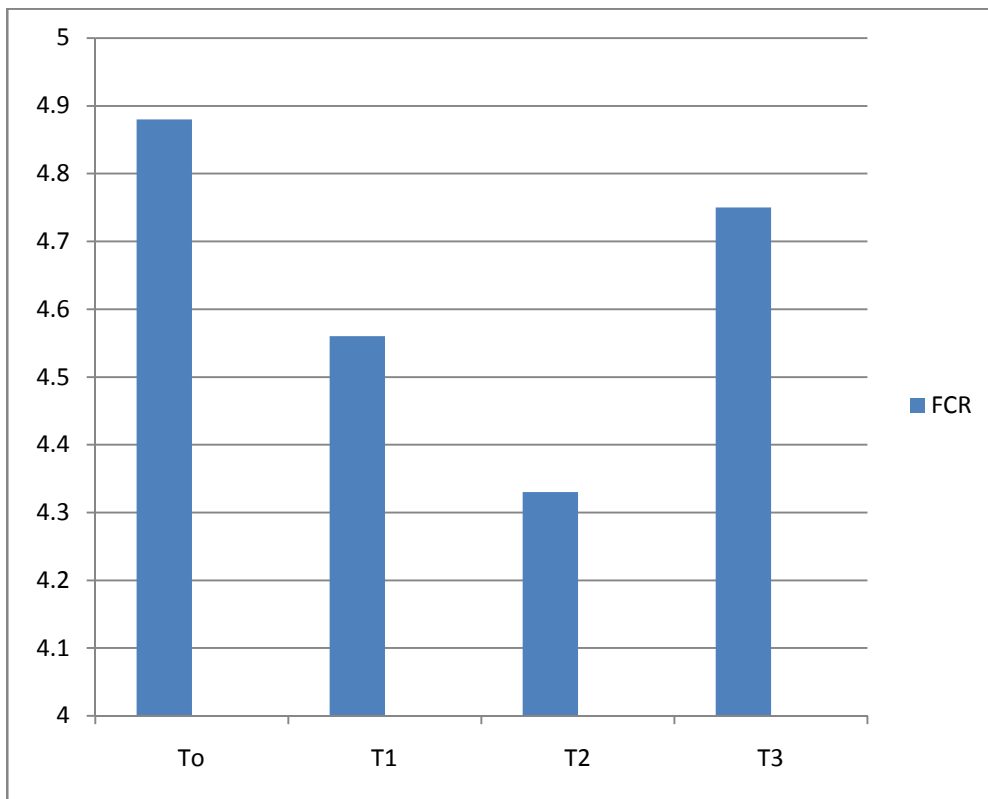


Figure 12: Feed Conversion Ratio



## **4.2 Carcass Characteristics**

### **4.2.1 Dressing Percentage**

The dressing percentage of  $T_0$ ,  $T_1$ ,  $T_2$  and  $T_3$  groups at 42 days age of the experimental quails were  $62.78 \pm 0.25g$ ,  $63.39 \pm 0.30g$ ,  $64.26 \pm 0.19g$ , and  $62.95 \pm 0.24g$  respectively (Table 4). So dressing percentage was significantly higher ( $p < 0.01$ ) in  $T_2$  compared to the other two treatment groups and control group got the lowest dressing percentage. Similarly Basak *et al.* (2002) and Parthasarathy *et al.* (2002) reported highest dressing percentage in the birds fed with 5 percent level of Azolla than control.

### **4.2.2 Breast Weight**

The breast weight (g) means of  $T_0$ ,  $T_1$ ,  $T_2$  and  $T_3$  groups at 42 days of the experimental period were  $19.60 \pm 0.45$ ,  $22.13 \pm 0.26$ ,  $25.36 \pm 0.94$  and  $20.89 \pm 0.49g$  respectively (Table 4). So, quails of group  $T_2$  (supplemented with 5% Azolla) got the highest breast weight ( $p < 0.01$ ) followed by group  $T_1$  (supplemented with 3% Azolla) and group  $T_3$  (supplemented with 7% Azolla) among all of the experimental groups and the control group  $T_0$  (without supplementation of Azolla) got the lowest body weight.

### **4.2.3 Thigh Weight**

The thigh weight (g) means of  $T_0$ ,  $T_1$ ,  $T_2$  and  $T_3$  groups at 42 days of the experimental period were  $6.23 \pm 0.28$  g,  $7.37 \pm 0.40$  g,  $8.28 \pm 0.37g$  and  $6.69 \pm 0.02$  g respectively (Table 4). So quails of group  $T_2$  (supplemented with 5% Azolla) got the maximum thigh weight ( $p < 0.01$ ) followed by group  $T_1$  (supplemented with 3% Azolla) and group  $T_3$  (supplemented with 7% Azolla) among all of the experimental groups and the control group  $T_0$  (without supplementation of Azolla) got the lowest thigh weight. Similarly, Naghshi *et al.* (2014) reported supplementation of 5% Azolla powder significantly increased ( $p < 0.05$ ) thigh weight.

### **4.2.4 Liver Weight**

The liver weight (g) means of  $T_0$ ,  $T_1$ ,  $T_2$  and  $T_3$  groups at 42 days of the experimental period were  $2.27 \pm 0.28$  g,  $2.76 \pm 0.23$  g,  $2.82 \pm 0.26$  g and  $2.51 \pm 0.01g$  respectively (Table 4). So, quails of group  $T_2$  (supplemented with 5% Azolla) got the maximum liver weight

followed by group T<sub>1</sub> (supplemented with 3% Azolla) and group T<sub>3</sub> (supplemented with 7% Azolla) among all of the experimental groups and the control group T<sub>0</sub> (without supplementation of Azolla) got the lowest liver weight. But not significant ( $p > 0.05$ ) difference among the groups. Similarly Naghshi *et al.* (2014) reported supplementation of 5% Azolla powder not significantly ( $p > 0.05$ ) differed liver weight.

#### **4.2.5 Gizzard Weight**

The gizzard weight (g) means of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> groups at 42 days of the experimental period were  $1.70 \pm 0.01$  g,  $1.64 \pm 0.02$  g,  $1.81 \pm 0.23$  g and  $1.67 \pm 0.00$  g respectively (Table 4). So, quails of group T<sub>2</sub> (supplemented with 5% Azolla) got the highest gizzard weight among all of the experimental groups. But no significant ( $p > 0.05$ ) difference among the groups. Similarly Naghshi *et al.* (2014) reported supplementation of 5% Azolla powder not significantly ( $p > 0.05$ ) differed gizzard weight.

#### **4.2.6 Heart Weight**

The heart weight (g) means of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> groups at 42 days of the experimental period were 0.86 g,  $0.82 \pm 0.01$  g,  $0.78 \pm 0.01$  g and 0.83 g respectively (Table 4). So, quails of control group T<sub>0</sub> got the highest heart weight and significantly ( $p < 0.01$ ) higher among the groups.

**Table 4: Dressing Percentage (%), Breast, Thigh, Liver, Gizzard and Heart Weight (g) of quail**

Variables	Control	Treatments			P Value	Level of significance
	T <sub>0</sub>	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>		
Dressing Percentage (%)	62.78 <sup>a</sup> ±0.25	63.39 <sup>a</sup> ±0.30	64.26 <sup>b</sup> ±0.19	62.95 <sup>a</sup> ± 0.24	0.012	*
Breast Weight(g)	19.60 <sup>a</sup> ±0.45	22.13 <sup>b</sup> ±0.26	25.36 <sup>c</sup> ±0.94	20.89 <sup>ab</sup> ±0.49	0.001	**
Thigh Weight(g)	6.23 <sup>a</sup> ± 0.28	7.37 <sup>b</sup> ± 0.40	8.28 <sup>c</sup> ± 0.37	6.69 <sup>ab</sup> ± 0.02	0.007	**
Liver Weight (g)	2.27 <sup>a</sup> ± 0.28	2.76 <sup>a</sup> ± 0.23	2.82 <sup>a</sup> ± 0.26	2.51 <sup>a</sup> ± 0.01	0.351	NS
Gizzard Weight(g)	1.70 <sup>a</sup> ± 0.01	1.64 <sup>a</sup> ± 0.02	1.81 <sup>a</sup> ± 0.23	1.67 <sup>a</sup> ± 0.00	0.756	NS
Heart Weight (g)	0.86 <sup>c</sup> ± 0.00	0.82 <sup>b</sup> ± 0.01	0.78 <sup>a</sup> ± 0.01	0.83 <sup>bc</sup> ± 0.00	0.001	**

Different letters in the same row indicate the significant difference

\*= Significant at 5% level of probability (p < 0.05)

\*\*= Significant at 1% level of probability (p < 0.01)

NS= Non-Significant (P > 0.05)

**Table 5: Hematological Parameters of Quails**

Days of post treatment	Treatment		Mean $\pm$ SE	Significance value
21 <sup>st</sup> day	TEC (million/mm <sup>3</sup> )	T <sub>0</sub>	2.20 <sup>a</sup> $\pm$ 0.15	NS
		T <sub>1</sub>	2.27 <sup>a</sup> $\pm$ 0.15	
		T <sub>2</sub>	2.20 <sup>a</sup> $\pm$ 0.06	
		T <sub>3</sub>	2.23 <sup>a</sup> $\pm$ 0.23	
	PCV %	T <sub>0</sub>	35.10 <sup>a</sup> $\pm$ 0.10	
		T <sub>1</sub>	35.13 <sup>a</sup> $\pm$ 0.59	
		T <sub>2</sub>	35.33 <sup>a</sup> $\pm$ 0.33	
		T <sub>3</sub>	35.03 <sup>a</sup> $\pm$ 1.73	
	Hb(gm/dl)	T <sub>0</sub>	8.07 <sup>a</sup> $\pm$ 0.58	
		T <sub>1</sub>	8.10 <sup>a</sup> $\pm$ 0.10	
		T <sub>2</sub>	8.07 <sup>a</sup> $\pm$ 0.41	
		T <sub>3</sub>	8.03 <sup>a</sup> $\pm$ 0.29	
	ESR (mm in 1 <sup>st</sup> hour)	T <sub>0</sub>	10.84 <sup>a</sup> $\pm$ 0.65	
		T <sub>1</sub>	10.70 <sup>a</sup> $\pm$ 0.30	
		T <sub>2</sub>	10.83 <sup>a</sup> $\pm$ 0.60	
		T <sub>3</sub>	10.67 <sup>a</sup> $\pm$ 0.67	
42 <sup>nd</sup> day	TEC(million/mm <sup>3</sup> )	T <sub>0</sub>	2.30 <sup>a</sup> $\pm$ 0.10	
		T <sub>1</sub>	2.43 <sup>a</sup> $\pm$ 0.25	
		T <sub>2</sub>	2.57 <sup>a</sup> $\pm$ 0.40	
		T <sub>3</sub>	2.40 <sup>a</sup> $\pm$ 0.26	
	PCV %	T <sub>0</sub>	35.40 <sup>a</sup> $\pm$ 0.31	
		T <sub>1</sub>	35.77 <sup>a</sup> $\pm$ 0.62	
		T <sub>2</sub>	35.87 <sup>a</sup> $\pm$ 0.13	
		T <sub>3</sub>	35.70 <sup>a</sup> $\pm$ 0.30	
	Hb (gm/dl)	T <sub>0</sub>	8.37 <sup>a</sup> $\pm$ 0.32	
		T <sub>1</sub>	8.43 <sup>a</sup> $\pm$ 0.09	
		T <sub>2</sub>	8.60 <sup>a</sup> $\pm$ 0.25	
		T <sub>3</sub>	8.07 <sup>a</sup> $\pm$ 0.12	
	ESR (mm in 1 <sup>st</sup> hour)	T <sub>0</sub>	9.70 <sup>a</sup> $\pm$ 0.35	
		T <sub>1</sub>	9.50 <sup>a</sup> $\pm$ 0.36	
		T <sub>2</sub>	9.40 <sup>a</sup> $\pm$ 0.31	
		T <sub>3</sub>	9.63 <sup>a</sup> $\pm$ 0.35	

Same letters in the same column indicate the no significant difference

\*= Significant at 5% level of probability (p < 0.05)

\*\*= Significant at 1% level of probability (p < 0.01)

NS= Non-Significant (P > 0.05)

### 4.3 Hematologic Parameters

#### 4.3.1 Total Erythrocyte Count (million/ mm<sup>3</sup>)

Total erythrocyte count of of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> groups at 21 days were 2.20 ± 0.15, 2.27 ± 0.15, 2.20 ± 0.06 and 2.23 ± 0.23 respectively (Table 5) and at 42 days were 2.30 ± 0.10, 2.43 ± 0.25, 2.57 ± 0.40 and 2.40 ± 0.26 respectively (Table 5). The values of TEC in all treated groups and control group were more or less similar and the values were within the normal range. The highest TEC was recorded in group T<sub>2</sub> and lowest in Group T<sub>0</sub> at 42 days of age. Although these values show a little fluctuation they were not statistically significant (p>0.05). Similarly Alagbe, (2017) and Alalade *et al.* (2007) reported that the Total Erythrocyte Count were not significantly (p>0.05) differed by the dietary inclusion of Azolla in birds but the highest TEC was recorded in treatment groups compared to control group.

#### 4.3.2 Packed Cell Volume (%)

Packed Cell Volume of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> groups at 21 days were 35.10 ± 0.10, 35.13 ± 0.59, 35.33 ± 0.33 and 35.03 ± 1.73 respectively (Table 5) and at 42 days were 35.40 ± 0.31, 35.77 ± 0.62, 35.87 ± 0.13 and 35.70 ± 0.30 respectively (Table 5). The values of PCV in all treated groups and control group were more or less similar and the values were within the normal range. The highest PCV was recorded in group T<sub>2</sub> followed by T<sub>1</sub>, T<sub>3</sub>, and control group T<sub>0</sub> was lowest at 42 days of age. Although these values show a little fluctuation they were not statistically significant (p>0.05). Similarly Alagbe (2017) and Alalade *et al.* (2007) reported that the Pack Cell Volume (PCV) values were not significantly (P>0.05) differed by the dietary inclusion of Azolla in birds but the highest PCV was recorded in treatment groups compared to control group.

#### 4.3.3 Hemoglobin (g/dl)

Hemoglobin count of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> groups at 21 days were 8.07 ± 0.58, 8.10 ± 0.10, 35.33 ± 0.33 and 8.03 ± 0.29 respectively (Table 5) and at 42 days were 8.37 ± 0.32, 8.43 ± 0.09, 8.60 ± 0.25 and 8.07 ± 0.12 respectively (Table 5). The values of Hb in all treated groups and control group were more or less similar and the values were within the normal range. The highest Hb was recorded in group T<sub>2</sub> than control group T<sub>0</sub> at 42 days of age. All the data were statistically not significant (p>0.05). Similarly Alagbe, (2017) and Alalade *et al.* (2007) reported that the Hb counts were not significantly (p>0.05) differed by the dietary

inclusion of Azolla in birds but the highest Hb was recorded in treatment groups compared to control group.

#### **4.3.4 Erythrocyte Sedimentation Rate (mm/1<sup>st</sup> hour)**

Erythrocyte sedimentation rate of T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> groups at 21 days were  $10.84 \pm 0.65$ ,  $10.70 \pm 0.30$ ,  $10.83 \pm 0.60$  and  $10.67 \pm 0.67$  respectively (Table 5) and at 42 days were  $9.70 \pm 0.35$ ,  $9.50 \pm 0.36$ ,  $9.40 \pm 0.31$  and  $9.63 \pm 0.35$  respectively (Table 5). The values of ESR in all treated groups and control group were more or less similar and the values were within the normal range. The highest ESR was recorded in control group T<sub>0</sub> and lowest in group T<sub>2</sub> at 42 days of age. Although these values show a little fluctuation they were not statistically significant ( $p > 0.05$ ).

## CHAPTER V

### CONCLUSION

The present study indicates that Azolla may be used as a growth promoter for Japanese quail up to 5% level. The highest body weight was obtained in T<sub>2</sub> followed by T<sub>0</sub> which differ significantly ( $p < 0.01$ ) from each other. The feed consumption was similar for all treatments all over the experimental period. Cumulative feed consumption for T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub>, and T<sub>3</sub> were 438g, 440g, 440g and 439g respectively. Feed conversion ratio (FCR) improved significantly for T<sub>2</sub> and T<sub>1</sub> which were  $4.33 \pm 0.05$  and  $4.56 \pm 0.09$  respectively. FCR for T<sub>3</sub> and T<sub>0</sub> were poorer i.e.  $4.75 \pm 0.08$  and  $4.88 \pm 0.10$ . The highest dressing percentage, breast, thigh liver and gizzard were observed for treatment groups than control group. However, further research using large number of birds with similar objectives is needed before giving final recommendation to use Azolla meal as a feed ingredient in quail ration.

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