EFFECT OF IPIL IPIL AND BEAN LEAVES SUPPLEMENTATION ON FEED CONVERSION, GROWTH PERFORMANCE AND EGG PRODUCTION OF CHICKEN

A THESIS

BY

F.M. BABRA HAMLIN
Registration No.: 1405121
Session: 2014-2015
Semester: January-June, 2015

MASTER OF SCIENCE (M S)
IN
PHARMACOLOGY

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

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Submitted to the Department of Physiology and Pharmacology
Hajee Mohammad Danesh Science and Technology University, Dinajpur, Bangladesh

In Partial fulfillment of the requirements
For the degree of

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IN
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Approved as to style and contents by

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(Dr. Rakibul Islam)	**(Dr. Md. Mahmudul Hasan)**
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DEPARTMENT OF PHYSIOLOGY AND PHARMACOLOGY
HAJEE MOHAMMAD DANESH SCIENCE AND TECHNOLOGY UNIVERSITY, DINAJPUR-5200

JUNE, 2016

DEDICATED
TO MY
BELOVED PARENTS

ACKNOWLEDGEMENT

The author is ever grateful to his creator Almighty Allah for his blessings to enable him to carry out this research work and complete this thesis.

The author would like to express heartfelt gratitude to his honorable Supervisor, ***Dr. Rakibul Islam****, Associate Professor and Chairman, Department of Physiology and Pharmacology, Hajee Mohammad Danesh Science and Technology University, Dinajpur for his supervision, scholastic guidance, innovative suggestions, constructive criticism, helpful comment, inspiration and timely instructions throughout the entire period of the research.*

The author expresses deep indebtedness to his Co-supervisor, ***Dr. Md. Mahmudul Hasan****, Assistant Professor, Department of Physiology and Pharmacology, Hajee Mohammad Danesh Science and Technology University, Dinajpur for his scholastic guidance, untiring assistance and advice in preparing the thesis.*

The author owes arrears of gratitude to ***Dr. Fahima Binthe Aziz****, Associate Professor and* ***Dr. Md. Bazlar Rashid,*** *Associate Professor, Department of Physiology & Pharmacology, Hajee Mohammad Danesh Science and Technology University, Dinajpur for their helpful advice and cooperation in providing facilities to conduct the experiment.*

The author humbly desires to express profound gratitude and thanks to all his reverend teachers of the Faculty of Veterinary and Animal Science, Hajee Mohammad Danesh Science and Technology University, Dinajpur for their kind help, cooperation, encouragement and valuable suggestions.

With due pleasure the author wishes to acknowledge the healthy working relationship of the staff of the Department of Physiology & Pharmacology, Hajee Mohammad Danesh Science and Technology University, Dinajpur.

The author deeply expresses cordial thanks to Ministry of Science and Technology for funding to the experimental work.

Finally, the author is very much grateful to his beloved parents, brother and sister for their sacrifice, inspiration, encouragement, endless love and continuous blessing for educating himself up to the postgraduate level.

The Author

ABSTRACT

This study was conducted to determine the effect of Ipil Ipil and Bean leaves supplementation on feed conversion, growth performance and egg production of chicken. A total of 28 "Hisex Brown layer" of 75 weeks old were purchased and assigned into four groups A, B, C and D. Group A was considered as control, fed only with commercial layer ration. Group B, C and D were supplemented with 2 gm grinded Ipil Ipil leaves, 2 gm grinded bean leaves and 1 gm grinded Ipil Ipil leaves plus 1 gm grinded Bean leaves per kg feed respectively. Observations were recorded for live body weight, feed consumption, feed conversion, growth performance and egg production of chicken. Increased egg production rate was observed in Ipil Ipil supplemented groups (group B). Body weights were increased significantly ($p<0.05$) in all treated groups in respect to the control and the highest weight was recorded in combined treatment of Ipil Ipil and bean leaves supplemented group (Group D). No significant ($p>0.05$) differences were observed among the groups for PCV and TEC values. Hb content increased and ESR decreased significantly ($p<0.05$) in the group (Group D) of combined treatment (Ipil Ipil and bean). In treatment groups there were significantly increased egg production, but in case of control group no incensement of egg production level observed. There was no significant pathological change in any internal organs of the layer of treated groups. Best result was found in the group D. The study revealed that the combined supplementation of Ipil Ipil and bean leaves gave better result than other groups in respect to body weight gain, feed consumption, feed conversion, growth performance, egg production, hemato-biochemical parameters and profitability without making any health hazard to the layers.

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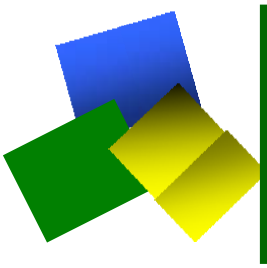
LIST OF ABBREVIATIONS

B. wt.	:	Body weight
Conc.	:	Concentration
Cu. mm	:	Cubic millimeter
d.w.	:	Drinking water
ESR	:	Erythrocyte Sedimentation Rate
et al.	:	Associates
Fig.	:	Figure
gm	:	Gram
Hb	:	Hemoglobin
i.e.	:	That is
J.	:	Journal
Kg	:	Kilogram
Lit	:	Liter
Ltd.	:	Limited
mg	:	Milligram
mm^3	:	cubic millimeter
No.	:	Number
PBS	:	Phosphate Buffer Solution
PCV	:	Packed Cell Volume
PM	:	Population Mean
SE	:	Standard Error
SM	:	Sample Mean
TEC	:	Total Erythrocyte Count
Vol.	:	Volume
µg	:	Microgram
%	:	Percent
&	:	And
@	:	At the rate of
<	:	Less than
>	:	Greater than
0^0C	:	Degree centigrade
FSH	:	Follicular Stimulating Hormone
LH	:	Luteinizing Hormone
PRL	:	Prolactin
ng	:	Nanogram
±	:	Plus minus
GDP	:	Gross Domestic Product



CHAPTER I

INTRODUCTION

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INTRODUCTION

Bangladesh is a highly populated country and growth of population is increasing very fast in comparison to its land size. As a result huge pressure is created on people's basic needs. The demand of protein of this booming population is a great threat for us. There are so many sources of protein but it is impossible to fulfill the demand without chicken meat and eggs. Chicken meat is popular to all of us and there is no religious restriction to consume.

Now a day's our unemployed young generation is coming in this business for long return of value and profit. Pharmaceutical companies take this advantage. They are convincing farmers to use synthetic phytase as a growth promoter and egg increaser for chicken. As a result each and every chicken is becoming a depot of antibiotics and other inorganic substances. When these chickens are consumed by human these antibiotic and other inorganic residue enters into human body and causing serious human health hazards with drug resistance (Islam, 2014).

So, scientists are again concentrating on the use of our ancient medicinal system to find beneficial herbs and plants which can be safely used to increase the production. Such plants, Ipil Ipil (*Leucaena leucocephala*) and Bean (*Lablab purpureus*) are common plant which can be used as an alternative source of phytase.

A phytase (myo-inositol hexakisphosphatephos phohydrolase) is any type of phosphatase enzyme that catalyzes the hydrolysis of phytic acid (myo-inositol hexakisphosphate) which is an indigestible, organic form of phosphorus that is found in grains and oil seeds and releases a usable form of inorganic phosphorus. While phytases have been found to occur in animals, plants, fungi and bacteria, phytases have been most commonly detected and characterized from fungi.

Phytic acid and its metabolites have several importance in seeds and grains, mostly phytic acid functions as a phosphorus store, as an energy store, as a source of cations and as a source of myo-inositol (a cell wall precursor). Phytic acid is the principal storage form of phosphorus in plant seeds and the major source of phosphorus in the grain based diets used in intensive livestock operations. The organic phosphate found in phytic acid

is largely unavailable to the animals that consume it but the inorganic phosphate that phytases release can be easily absorbed. Ruminant animals can use phytic acid as a source of phosphorus because the bacteria that inhabit in their gut are well characterized producers of many type of phytases. However, monogastric animals do not carry bacteria that produce phytase, thus these animals cannot use phytic acid as a major source of phosphorus and it is excreted in the feces.

Phytic acid and its metabolites have several other important roles in Eukaryotic physiological processes. As such phytic acid and its metabolites have been implicated in DNA repair, clathrin-coated vesicular recycling & control of neurotransmission and cell proliferation (Conway *et al*., 2007). The exact roles of phytases in the regulation of phytic acid and its metabolites in the physiological processes described above are still largely unknown and the subject of more research.

Phytase is used as animal feed supplement often in poultry and swine to enhance the nutritive value of plant material by liberation of inorganic phosphate from phytic acid (myo-inositol hexakisphosphate).

Ravindran *et al.* (1994) stated Ipil Ipil as a promising source of phytate phosphorus. George *et al.* (2006) extracted phytase (EC 3.1.3.8.) of Sanilac Navy Beans with 2% $CaCl_2$ and purified by ammonium sulfate fractionation and DEAE cellulose chromatography. Araújo *et al.* (2008) researched on phosphatase and phytase activities in nodules of common bean genotypes at different levels of phosphorus supply.

Kouas *et al.* (2009) studied changes in growth, symbiotic nitrogen fixation (SNF), acid phosphatase (ACP), and phytase activities to phosphorus availability of common bean. A single phytase was revealed for the nodules of both lines and was significantly enhanced by P deficiency. Three ACP types were found in roots and leaves, showing increasing activity under phosphorus deficiency

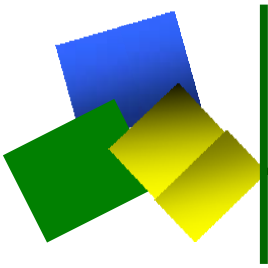
Lazali *et al.* (2014) worked on Localization of phytase transcripts in germinating seeds of the common bean (*Phaseolus vulgaris* L.) The work provides the first time evidence of tissue specific expression of a phytase gene in the parenchyma of germinating seeds whereas it was localized in the inner and middle cortex of nodules.

Phytate is the major form of phosphorus found in cereal grains, beans and oilseed meals feed to poultry birds (Ravindran *et al.,* 1995). Approximately 61–70%

phosphorus found in poultry diet ingredients is in the form of phytate phosphorus. But the monogastric animals like poultry birds are unable to utilize this phytate phosphorus, as they lack endogenous phytase, which necessitates in the addition of inorganic feed containing phosphates to poultry diets in order to meet the phosphorus requirements of poultry (Yu *et al.,* 2004). It results in relatively large amounts of phosphorus in the manure that contribute to environmental pollution (Guo *et al.,* 2009). Exogenous phytase of microbial origin can be used as an alternative that can help to reduce phosphorus excretion in poultry (Yu *et al.,* 2004). The beneficial effect of exogenous phytase in poultry ration has been supposed to be due to the direct hydrolytic effects on phytate and the subsequent improvement in the availability of minerals, amino acids, and energy (Selle and Ravindran, 2007). It has also been suggested that phytase in poultry diets improves gut health as indicated by reduced secretions from the gastrointestinal tract (GIT) which consequently improves the efficiency of utilization of energy (Oduguwa *et al.,* 2007; Pirgozliev *et al.,* 2008). The main goal of current study therefore is to determine the effect of Ipil ipil and Bean leaves supplementation as a source of phytase on the growth performance, feed efficiency, protein/amino acid digestibility, energy utilization, mineral retention, and bone growth and egg production of Chicken. By realizing all sorts of problem we are planning to rear Chicken by using herbal medication like Ipil Ipil and bean leaves extract instead of any Synthetic agent, to avoid human health hazards as well as economic Chicken production in Bangladesh.

Therefore, the present study was designed with the following objectives:

1. To assess the performance index: egg production, growth and body weight gain of chicken by providing Ipil Ipil and Bean leaves supplementation.
2. To evaluate the effects of Ipil Ipil and Bean leaves supplementation on blood parameters and gut biosis of the chicken.
3. To evaluate the hazardous effect of that supplementation on chicken



CHAPTER II

REVIEW OF LITERATURE

CHAPTER II

REVIEW OF LITERATURE

This chapter presents the review of relevant literatures, which consist of the effects of Ipil ipil and Bean leaves supplementation on feed conversion, growth performance and egg production of chicken. Many researchers have been conducted researches in these topics. But in Bangladesh, limited research work has been performed.

2.1 Ipil Ipil (*Leucaena leucocephala*)

Leucaena leucocephala is a small, fast-growing mimosoid tree native to southern Mexico and northern Central America (Belize and Guate mala), but is now naturalized throughout the tropics. Common names include white leadtree, jumbay, river tamarind, Subabul and white popinac (Hughes *et al.*, 1998). During the 1970s and 1980s, it was promoted as a "miracle tree" for its multiple uses. It has also been described as a "conflict tree" in that it is both promoted for forage production and spreads like a weed in some places (Gutteridge *et al*., 1998). The legume is promoted in several countries of Southeast Asia (at least Burma, Cambodia, Laos and Thailand), most importantly as a source of quality animal feed, but also for residual use for firewood or charcoal production.

2.1.1 The chemical constituents of *L. leucocephala* leaves

Chemical constituents	Amount Present
Crude Protein (%)	25.9
Carbohydrate (%)	40
Tannin (%)	4
Mimosin (%)	7.19
Total ash (%)	11
Total N (%)	4.2
Crude protein (%)	25.9
Calcium (%)	2.36
Phosphorus (%)	.23
b -carotene (mg/kg)	536
Gross energy (kJ/g)	20.1
Tannin (mg/g)	10.15

Meena Devi *et al.* (2013)

2.2 Bean

Bean is a common name for large seeds of several genera of the flowering plant family Fabaceae (also known as Leguminosae) which are used for human or animal food.

2.2.1 Types

Currently, the world gene banks hold about 40,000 bean varieties, although only a fraction are mass produced for regular consumption.

Some bean types include:

- *Vicia*
  - *Vicia faba* (broad bean or fava bean)

Vicia faba or broad beans, known in the US as fava beans

- *Phaseolus*
  - *Phaseolus acutifolius* (tepary bean)
  - *Phaseolus coccineus* (runner bean)
  - *Phaseolus lunatus* (lima bean)
  - *Phaseolus vulgaris* (common bean; includes the pinto bean, kidney bean, black bean, Appaloosa bean as well as green beans, and many others)
  - *Phaseolus polyanthus* (a.k.a. *P. dumosus*, recognized as a separate species in 1995)
- *Vigna*
  - *Vigna aconitifolia* (moth bean)
  - *Vigna angularis* (adzuki bean)
  - *Vigna mungo* (urad bean)
  - *Vigna radiata* (mung bean)
  - *Vigna subterranea* (Bambara bean or ground-bean)
  - *Vigna umbellata* (ricebean)
  - *Vigna unguiculata* (cowpea; also includes the black-eyed pea, yardlong bean and others)
- *Cicer*
  - *Cicer arietinum* (chickpea or garbanzo bean)

- *Pisum*
  - *Pisum sativum* (pea)
- *Lathyrus*
  - *Lathyrus sativus* (Indian pea)
  - *Lathyrus tuberosus* (tuberous pea)
- *Lens*
  - *Lens culinaris* (lentil)
- *Lablab*
  - *Lablab purpureus* (Hyacinth bean/Country bean)
- *Glycine*
  - *Glycine max* (soybean)
- *Psophocarpus*
  - *Psophocarpus tetragonolobus* (winged bean)
- *Cajanus*
  - *Cajanus cajan* (pigeon pea)
- *Mucuna*
  - *Mucuna pruriens* (velvet bean)
- *Cyamopsis*
  - *Cyamopsis tetragonoloba* or (guar)
- *Canavalia*
  - *Canavalia ensiformis* (jack bean)
  - *Canavalia gladiata* (sword bean)
- *Macrotyloma*
  - *Macrotyloma uniflorum* (horse gram)
- *Lupinus* (lupin)
  - *Lupinus mutabilis* (tarwi)
  - *Lupinus albus* (lupini bean)
- *Arachis*
  - *Arachis hypogaea* (peanut)

2.2.2 Chemical Composition

Legumes are good sources of protein, minerals, vitamins dietary fiber and complex carbohydrates (FAO, 1982).

Proteins:

Legume has a high protein content ranging from 17 to 25% on dry weigh basis. Protein content of the edible protein of legume seeds is double than cereal and slightly lesser than meat fish and eggs (Walt and Merrill, 1963). Deka and Sakar (1990) reported the crude protein ranged from 22-31% of Hyacinth bean. However, (Schaaffhausen *et al.*, 1963) reported that the crude protein of Hyacinth bean ranged from 25-28%.

Amino acid:

Hyacinth bean is a good source of the amino acid, lysine and as such complement the general low lysine content of maize (corn) Daka and Sarkar (1990). General deficiency of lysine in most cereals e.g. sorghum and corn, thus consequence of their low content of albumins and globulin's (FAO, 1981).

Lablab Purpureus is especially low in the sulpher amino acids and tryptophan (Duke, 1981).

Carbohydrates:

Legumes have a larger sugar content which, contain 1-2%. Sucrose is a major sugar and unlike cereal, legumes contain appreciable quantities of oligosaccharides. These are indigestible by mammalian enzymes (FAO, 1982). The seed in addition to large proportion of protein (26%) and starch (48%) it contains various other carbohydrates as dietary fiber or unavailable carbohydrates (FAO, 1981) , Ripped seed Hyacinth bean contain (per 100 g) 61.49%. The total carbohydrates, 54.02 to 63.3% (Deka and Sakar, 1990).

Dietary fiber:

Dietary fiber is the term defined by Trowel, (1972) as the skeletal that remains of plant cell that are resistant to hydrolysis by the enzymes of man. It includes cellulose hemicelluloses (water soluble including pectin) and lignin. Wide range of unusual storage dietary fibers is commonly associated with legume (FAO, 1981). Deka and Sarkar (1990) reported the crude fiber of Hyacinth bean range from 7.6-9.6%. Moreover, Elhardallo and Eltinay (1985) reported 7.7% for crude fiber Hyacinth bean.

Phytic acid:

The other Anti Nutritional Factors which was found in Hyacinth bean is phytic acid. It's well known that high levels of phytic can reduce the availability in plant products (Cheryan *et al.,* 1983). *The important role of phytic acid nutrition was due to its ability to form insoluble* compounds with different mineral elements including calcium, phosphorus, iron, magnesium, zinc, rendering them unavailable to the animal and resultant phytates being excreted in feaces (Vohra *et al.,* 1965). Deka and Sarkar (1990) suggested that the level of phytic acid 1000 to 1350 (mg /100g).

2.3 Ipil ipil as animal and poultry feed supplement

Khan *et al.* (2009) experimented on one hundred twenty, two weeks old ISA Vedette broiler. The chicks were randomly allotted to four dietary treatments to investigate the effect of replacing fishmeal with two unconventional sources of leaf meals. The chicks were distributed into dietary treatments A (control-12% CP from fish meal), B (5% CP of fish meal replaced by *Leucaena*), C (5% CP of fish meal replaced by *Sesbania*) and D (2.5% CP of fish meal by *Leucaena* + 2.5% CP of fish meal by *Sesbania*) having three replications in each treatment group having 10 chicks in each replication. They found that *Leucaena* and *Sesbania* could be well utilized in broiler ration with no deleterious effects. Replacement of CP at 5% level by *Sesbania* leaf meal was slightly superior to that of *Leucaena* or a mixture of *Sesbania* and *Leucaena* in respect of broiler performance. Profit per kg live weight gain and per kg dressing yield at 5% level of CP replaced by *Sesbania* were only slightly superior over 12% CP from fish meal, 5% level of CP from *Leucaena* or a mixture of two leaf meals respectively. The findings also revealed that *Leucaena* and *Sesbania* leaf meals might replace individually or mixture of two leaf meals at 5% level of CP of fish meal for broiler production without affecting performance.

Oliveira *et al.* (2014) evaluated the effects of inclusion of *leucaena* leaf hay (LLH) on the performance and nutrient digestibility of diets for laying hens during the growth phase (14-18 weeks). Ninety pullets (Rhode Island Red and New Hampshire) were distributed in a completely randomized design with three treatments (0%, 5% and 10% inclusion of LLH) and five replicates, with six birds. The inclusion of LLH did not statistically influence nitrogen digestibility, apparent metabolizable energy and apparent

metabolizable energy corrected for nitrogen of diet. However, this inclusion significantly affected coefficients of dry matter (CDDM) and gross energy, resulting in lower CDDM and gross energy with inclusion of 10%. Whereas the use of nutrients by chicks fed on diets with the inclusion of LLH allowed the same amount of metabolizable energy, inclusion of up to 10% of LLH diet during the growth phase (14-19 weeks) of laying hens (Rhode Island Red and New Hampshire) may be recommended.

Zanu *et al.* (2011) conducted a study to assess the response of Cobb broiler chicks to diets containing varying levels (0%, 5%, 10% and 15%) of Leucaena leaf meal (LLM). They fed experimental starter diets (14-28 d) and finisher diets (28-56 d). Final weight, growth rate and feed conversion ratio significantly ($P<0.05$) declined as the level of LLM in the diets increased. Dressed and carcass weights also reduced significantly ($P<0.05$) with increasing level of LLM in the diets. All organ characteristics except liver kidney were significantly ($P<0.05$) affected by dietary treatments. Haematological variables were also not affected ($P<0.05$). The total cholesterol and Low Density Lipoprotein of serum decreased ($P<0.05$) when LLM was included to the diets. Feed cost reduced when LLM was incorporated in the diets, but the net revenue declined as LLM in diet increased. In this study inclusion of LLM in diets for broiler chickens did not affect their health status, but rather depressed their growth.

Hussain *et al.* (2007) investigated the effect of including Leucaena *(Leucaena leucocephala)* leaf meal (LLM) in broiler diets employing LLM at 0, 50, 100, 150 and 200 g/kg diet. Experiments were also conducted to investigate whether reduced food intake of diets containing LLM is responsible for the growth depression. LLM appears to be a good source of protein and calcium. Food intake was not significantly affected ($P>0.05$) with dietary LLM inclusion up to 200 g/kg. Inclusion of LLM up to 150 g/kg diet did not influence significantly ($P>0.05$) the performance of broilers in terms of body weight gain and food efficiency. However, inclusion of LLM at 200 g/kg diet had an adverse effect on weight gain and food efficiency, when compared to other dietary treatments. The profitability over food costs was low with the 200 g LLM/kg diet compared to those of other diets. It may be inferred from these experiments that LLM can safely be included in broiler diets up to 150 g/kg. Studies using force-feeding and restricted feeding techniques indicated that reduced food intake was not responsible for the growth depression in diets containing higher amounts of LLM.

Mutayoba *et al.* (2003) studied The effect of dietary energy (high and low) and Leucena leaf meal (LLM) levels (at 0 - 20%) on the performance of growing layers using 24 pullets in a digestibility trial and 288 birds aged 12 weeks in a growth study. The crude protein and crude fibre of LLM were 20.6 and 18.9%, respectively, whereas NFE was only 34.6%. LLM contained about 16.5 MJ/kg DM Gross energy and 1.9% mimosine in DM. Increasing LLM led to a reduction in most of the parameters measured with the exception of gizzard weights, large intestine length and age at first egg. Plasma glucose concentrations were not affected by treatment, while total plasma proteins fluctuated inconsistently. This study showed that LLM at 5% had no adverse effect on performance of growing layers but higher inclusion levels affected performance regardless of the dietary energy level**.**

Atawodi *et al.* (2004) supplemented Leucaena leucocephala (leucaena). At 20% supplementation level, the feed efficiency was highest in the group on standard diet and lowest in the group on sun dried leucaena supplement. These results suggest that at 20% supplementation level, leucaena causes reduced growth rate, and that ensiling (with or without ruminal fluid) only has marginal effect on the toxicity of mimosine found in Ipil ipil.

Abou-Elezz *et al.* (2004) conducted two experiments, aimed at determining the effect of the dietary inclusion of either Leucaena leucocephala (LLM) or Moringa oleifera (MOLM) leaf meals on Rhode Island Red (RIR) hens' egg production and quality. In the first experiment, thirty six RIR hens, at 36 weeks of age, were randomly divided into four groups each of nine birds and were allocated in individual cages. The four groups corresponded to four dietary treatments containing 0 (control), 5, 10, and 15 % of LLM, respectively. Simultaneously, the second experiment was carried out following the same design but using MOLM instead of LLM. The egg production and quality traits were monitored for five weeks, preceded by one week of adaptation. The results showed a quadratic effect on the egg laying rate (57.10, 57.46, 53.25, and 47.46 %), egg mass (g/hen/d) and feed conversion due to the LLM treatments (0, 5, 10, and 15 %, respectively). The MOLM treatments decreased linearly the egg laying rate (60.00, 59.72, 56.13, and 51.87 %) and the egg mass, and had a quadratic effect on the feed intake (111.15, 111.93, 107.08, and 100.47g/hen/d) when including 0, 5, 10, and 15 % of MOLM, respectively. The yolk color increased linearly by the rise in both the MOLM and the LLM levels. Other results were obtained in the albumen and yolk proportions

(%) and in the yolk coefficient, while no adverse effects were found on the other egg quality traits due to the LLM or MOLM treatments. The MOLM or the LLM could be acceptable as sustainable feed resource up to 10 % in laying hen diets.

Atawodi *et al.* (2007) Protein source is a limiting factor in poultry feed production in the tropics. Therefore, the suitability of leaves of *Leucaena leucocephala* as a protein rich multipurpose leguminous plant as feed supplement in laying hens was evaluated at 50, 100 and 200 g/kg (5, 10 and 20%) supplementation levels. *Leucaena* supplementation significantly decreased weekly average daily egg lay ($P < 0.01$) and progressively reduced cumulative weekly average daily egg lay to 88.2, 68.7 and 53.4% for 5, 10 and 20% supplementation levels, respectively. There was an inverse relationship between level of *L. leucocephala* supplementation and weekly average daily egg lay ($r = - 0.99$) which highly correlated with the crude fiber content of the diets ($r = 0.94$). Size and specific gravity of eggs were not significantly affected ($P >0.05$) by the different levels of *leucaena* supplementation. These results suggest that *L. leucocephala* leaves may only be useful as feed supplement in egg laying hens at low levels of supplementation.

Ayssiwede *et al.* (2010) used *Leuceana leucocephala* leaves meal as a protein ingredient source for indigenous Senegal chickens diets.The groups were corresponded to four dietary treatments (LL0, LL7, LL14 and LL21) containing respectively 0, 7, 14 and 21% of *Leuceana* leaves meal. Results showed that the *Leuceana* leaves were relatively rich in protein (24.9% DM), ether extract (6.4% DM), crude fiber (14.2% DM) and Neutral detergent fiber (22.4% DM). It contained respectively 43.1% and 11.4% DM of nitrogen free extract and ash, particularly calcium (1.8%) and potassium (1.1% DM) and 2573.8 kcal/kg DM of metabolizable energy. The results of the trial showed that the inclusion of *L. leucocephala* leaves meal in the diet at 21% level, has no significant adverse effect on feed intake, average daily weight gain, feed conversion ratio and nutrients utilization (except ether extract) of adult indigenous Senegal chickens. It has significantly ($p<0.05$) improved the crude protein and metabolizable energy utilization in birds fed the 7% level inclusion diet (LL7).

Adeparusi *et al.* (2005) supplemented *Leucaena leucocephala* and *Gliricidia sepium* leaf protein (LP) as either total or partial replacement for groundnut cake in the control diet. All diets were formulated at 30% crude protein. Fish (average weight 8.4g) were randomly assigned in triplicates of 30 fish per dietary treatment and fed at 5% body

weight daily for 8 weeks. Toasted Bam-nut supplemented with leaf protein gave the best crude protein digestibility followed by the fish fed a mixture Bam-nut and groundnut cake.

Safwat *et al*. (2015) evaluated the nutrient digestibility of growing rabbits fed diets with different levels of either *Leucaena leucocephala* (LLM) or *Moringa oleifera* (MOLM) leaf meals and also to compare total collection and TiO_2 marker methods for estimating digestibility. A total of 30 California growing rabbits (1.81±0.19 kg live weight on average) were randomly distributed into five experimental groups of six rabbits each and were housed in individual cages. The groups were control, 30% LLM, 40% LLM, 30% MOLM, and 40% MOLM. All groups received pelleted diets for two weeks; diets also contained 4 g/kg titanium dioxide as dietary marker. The results showed that there were no difference ($p>0.05$) in feed, dry matter (DM), organic matter (OM), crude protein (CP), digestible energy, and crude fiber (CF) intake between the control group and the other experimental groups. The apparent digestibility values of DM, OM, CP, CF, acid detergent fiber, and gross energy were the highest for control group ($p = 0.001$), meanwhile MOLM diets had generally higher nutrient digestibility coefficients than LLM diets. Increasing the inclusion level of leaf meal in the diet from 30% to 40% improved the digestibility of CF from 45.02% to 51.69% for LLM and from 48.11% to 55.89% for MOLM.

Eichie *et al.* (2015) carried out a study to determine the effect of replacing soybean meal (SBM) with *Leucaena leucocephala* leaf meal (LLM) in the diet of broiler chickens. One hundred and fifty one-day-old broiler chicks were allotted to five treatments. The feeding trial lasted for 8 weeks. The treatments were Diet 1 (control, without LLM replacement) and Diets 2, 3, 4, and 5 where soybean meal (SBM) was replaced by 25%, 50%, 75%, and 100% of LLM, respectively from 0-56 days of rearing. Substitution of SBM with LLM significantly ($P < 0.05$) influenced the performance of broilers from day 0-28, but not at 29-56 days. Body weight gain of 907 g was obtained in birds on Diet 3 (50% LLM), while the least value of 553 g was obtained for Diet 5 with 100% LLM. The diet containing 50% LLM had better FCR (2.66) than the other treatments. Substitution of SBM with LLM did not significantly influence the haematological parameters except Erythrocyte Sedimentation Rate (ESR) concentration, which was significantly ($P < 0.05$) affected. Results of the study showed that LLM could be used to supplement SBM up to

50% in the diets of broiler chickens to improve performance and without adverse effect on the birds.

2.4 Bean leaves as animal and poultry feed supplement:

Assessment of lablab *(Lablab purpureus*) leaf meal as feed ingredient and yolk colouring agent in the diet of layers was applied by A. A. Odunsi (2003). Feeding trial was conducted to determine the performance nutrient digestibility and egg quality of layers fed 0, 50,100 and 150 kg leaf meal of *Lablab purpureus*. Feeding Lablab at 100 and 150 g/kg significantly reduced feed intake and egg production while egg weight, feed conversion efficiency and body weight changes were not affected by dietary treatments. Apparent nutrient digestibility of dry matter and crude protein decreased with Lablab inclusion while ether extract was not significantly influenced. Internal and external egg quality values were comparable amongst dietary groups except for yolk color, which was affected higher in layers fed Lablab compared to those without. Boiling had no effect on the proportion of egg components but boiling affected a percentage reduction of 62, 56 and 52 in the egg yolk colour of 50, 100 and 150 g/kg lablab fed layers, respectively. The persistence of the color change after withdrawal of lablab ranged from 5 days 50 g/kg to 15 days 150 g/kg, based on egg quality. Lack of mortality and similar biological efficiency, it may be possible to include *Lablab purpureus* in layer diets up to 100 and 150g /Kg in situation of acute scarcity and or high cost of grain and concentrates.

Islam *et al.* (2002) worked in Evaluation of toxic effects of *Lablab purpureus* seed meal as a dietary protein supplement in broiler feed. Group (A) was fed ration with ground raw country bean 250 mg/kg diet, group (B) with ration containing ground fried country bean 250 mg/kg diet supplemented with methionine and lysine (1g/kg) diet and group (C) control diet. Weight, feed intake and water consumption and presenting signs were recorded daily for 36 days. Groups A and B showed toxic signs such as anorexia, depression, drowsiness, incoordination, recumbency and ruffled feathers. The control group had the highest feed intake followed group B and then group A. reduction in body weight were recorded on the 7^{th}, 14^{th}, 21^{st}, 28^{th}, and 36^{th} day after feeding with raw and fried bean seed supplemented meal, due to the low nutritive value and the anti-nutritive effects of proteinase inhibitors, tannins, phytic acid, lectins. There were increases in weight of intestinal, pancreas and gizzard of groups A and B due to the present of lectins and trypsin inhibitors. Raw and fried

bean seed meals were toxic when supplemented at 12.5% in diet of ground broilers. Sarwatt *et al.* (1991) worked on effects of substituting *Dolichos bean* meal with soya bean meal on the performance of broiler chicken, Two hundred Cobb broiler chicken one day-old were randomly allocated to five rations containing levels of *Dolichos beans (Lablab purpureus*) meal at 0, 5, 15, 20 and 25 percent and *soya bean (Glycine max*) meal at 25, 20, 10, 5 and 0 percent. Feed intake, feed utilization efficiency, growth and mortality rates were determined from 2 to 8 weeks at which time the birds were slaughtered and dressing percentages and organ weights were determined. As the level of dolichos bean meal increased there was a decrease in crude protein and an increase in crude fiber in the diets, but the changes were not affected. Weight gain was highest (28.6 g/day) for the ration containing 25 percent soya bean meal and lowest (26.6 g/day) for the diet containing 25 percent *Dolichos* bean meal. Feed intake was highest for the ration containing 25 percent *Dolichos bean* meal but there was no difference between the treatments. Although the mortality rate was highest (16%) in the diet containing 25 percent *Dolichos bean* meal, the beans were well accepted by the birds, and the protein appeared to be well utilized, with a feed gain ratio of 3.02. This value was only slightly lower than that recorded for the diet containing 25 percent soya bean meal.

Robinson and Singh (2001) worked in the effect of Hyacinth bean, *chick pea* and *Mung bean* on layer performance. The material tested in this trial were *Lablab bean* (100, 200 and 400 g/kg), chick pea (200 and 300 g/kg) and Mung bean (300 and 450 g/kg) there were also mash and pellet control treatments without grain legumes and all diets were formulated to similar nutrient specifications. The start of trial at 17-18 weeks of age, parameters measured were egg number, egg mass, egg weight, feed intake, and body weight. Treatments mean performance results over the experimental period that Mung bean (450 g/kg) and *Lablab* at all three concentrations were associated with significantly adverse effects when compared to the control diet. The diet containing 400 g/kg Hyacinth bean resulted in lower egg number, egg mass output and feed intake than any of the other treatments ($P<0.001$). Average egg weight of birds given the 400 g/kg Lablab diet was lower than that of birds given 100 g/kg Hyacinth bean diet. Birds given *chick* pea or Hyacinth bean at the highest concentration lost most weight.

Omole *et al.* (2007) used *Stylosanthes guianensis* and *Lablab purpureus* as sole feed for growing rabbit. Thirty-six cross-bred growing rabbits of mean weight 515 ± 2.3g were

used for the study. The animals were randomly allotted to 3 different treatments. The animals in T1 were fed *S. guanensis* only, while animals in T2 and T3 were fed solely on *L. purpureus* and sunflower leaf (control), respectively. Feed intake and weight gain were measured on daily and weekly basis respectively. The results showed that rabbits fed *S. guanensis* and *L. purpureus* compared favourably with those fed sunflower leaf in terms of feed intake, weight gain and feed conversion ratio. The results also revealed that the nutrients digestibility (dry matter, crude protein and crude fibre) were also better in rabbit fed *S. guanensis* and *L. purpureus*. The dressing percent, lung weight, heart and kidney weight were not affected by the dietary treatment.

Babikr (2000) studied the effect of decorticated Hyacinth bean on the performance of broiler chicks and some blood parameters. One hundred and twenty eight one day old unsexed broiler chicks (Hubbard) were used, chicks were divided into four groups with four replicates of 8 birds / replicate, the birds were fed on 4 experimental diets, with varying levels of Hyacinth bean (0, 5, 10, and 15%). The results of the experiment indicated that dietary treatments had significant ($P<0.01$) effect on feed intake, weight gain and feed conversion ratio (FCR). Inclusion of Hyacinth bean in broiler diets resulted in reduction in feed intake and weight gain. Moreover, the treatments had no effect on dressing percentage. Rickets occurred in 12.5% the birds fed 10 and 15% Hyacinth bean Supplementation of Hyacinth bean in broiler diets had no significant ($P>0.05$) effect on serum calcium and total serum protein. However, the treatment had significantly decrease ($P<0.01$) serum phosphorus and bone ash. The results of recovery period indicated that birds fed previously Hyacinth bean had improved in feed intake, weight gain and feed conversion ratio (FCR).

2.5 Review of literature on effect of phytase on egg production

Lucky *et al.* (2014) worked on effect of dietary exogenous phytase on laying performance of chicken at older ages. A total of 48 Shaver-579 chicken layers aged between 85 to 94 weeks were reared in individual cages and given a basal diet amounting to 115g feed/bird/day. The basal diet fortified with 0.05, 0.10 or 0.15% RenaPhytase-400 constituted of 3 experimental diets to see the effects of exogenous phytase on egg production and egg quality. Results indicated that increasing level of exogenous phytase in diet almost linearly ($p<0.05$) increased egg production and feed conversion but did not affect egg quality. Providing phytase in the diet at 0.05, 0.10 and 0.15% increased egg

production by 11.86, 22.2 and 24.58%, respectively. It was shown that highest egg production was found at 0.15% phytase levels in diet. They concluded that egg production of aged hen is increased by adding exogenous phytase in the diet.

Abeyrathna *et al.* (2014) the objective of this study was to determine the maximum inclusion levels of dietary RB with or without exogenous phytase for laying Japanese quail. In a completely randomized design with a 3 x 2 factorial arrangement, 108 quails in 36 cages received six experimental diets ad libitum from 8 to 15 week. Experimental factors were three dietary RB inclusion levels (20, 30 and 40%) and two phytase levels (0 and 1000 FTU/kg). The level of dietary RB, phytase supplementation and their interaction had no significant effects on live weight or feed intake. The total egg production of the quail fed 40% RB was significantly lower than that of quail fed 20 and 30% RB. Egg laying rate of the quail fed 40% RB was significantly lower than those of the quail fed 20 or 30% RB from 6th week onwards. By eighth week, 30% RB resulted in significantly lower egg laying rate compared to the quail fed 20% RB. Feed conversion ratio (FCR) of the quail fed 40% RB was significantly higher than those of 20 or 30 % RB fed. Adverse effects of phytate in 30 or 40% RB on egg number, egg mass and FCR were not mitigated by the supplemental phytase. They concluded that inclusion of more than 20% RB in the diets of laying Japanese quail reduces the production performances.

Ademola *et al.* (2013) investigated the effects of microbial phytase and native wheat bran phytase on laying performance, egg quality and shell phosphorus of hens fed two forms of diets. Five experimental diets were formulated for the study. Control and basal diets contained similar levels of nutrients. However, basal diet (T1) containing 15% wheat bran (WB) had lower available phosphorus (AVP). Diet forms (mash and pelleted) and microbial phytase supplementation (0 and 900 phytase unit (FYT) were arranged to examine their interaction effects. The 0 FYT microbial phytase represented the native wheat bran phytase activity in the mash diet only. T1 and T2 were mash and pelleted unsupplemented diets respectively. Diets in T3 and T4 were microbial phytase supplemented in mash and pelleted forms respectively. Laying hens fed unsupplemented mash basal diet (T1) had the highest hen day production (HDP) ($P<0.024$), and the best feed conversion ($P<0.012$). However, those fed mash supplemented diet (T3) had the lowest HDP and worst feed conversion. Microbial phytase supplementation to mash diet (T3) resulted in lowest egg mass of 45.35 gram daily ($P<0.025$). Pelleting the

unsupplemented diet (T2) yielded poorer feed conversion than those fed unsuplemented mash diet (T1). Hens fed pelleted supplemented diet (T4) had slightly reduced HDP and significantly lower egg mass when compared to the control group. These hens had significantly highest yolk index (P<0.036) and egg shell with the most concentrated phosphorus content (P<0.002). It is concluded that native wheat bran phytase in mash diet containing 15% WB was effective for improved laying performance.

Ahmadi *et al.* (2012) added of nonphytate P (NPP) in diets for laying hens without negatively affecting their productivity and heath is crucial for sustainable egg production. A meta-analytical approach using a full quadratic model was applied to quantify relationships between dietary NPP and phytase levels and performance of laying hens. Except for the quadratic effect of dietary phytase on FCR were significant (P < 0.05). There was a relatively strong relationship between observed and predicted and FCR. Analyses of the model revealed that corn soybean meal based diets containing 0.22% of NPP without supplemental phytase resulted in high EP, EM, and feed efficiency in layers In the presence of phytase in feed.

Mohebbifar *et al.* (2011) investigated effects of phytase supplementation of low phosphorous diets included graded levels of rice bran on productive performance of laying hens and egg quality characteristics. They experimented on Lohmann LSL-Lite hens after production peak were randomly divided in 48 cages and experimental diets including three levels of rice bran with or without phytase and two levels of dietary nonphytate phosphorus were fed to hens with 4 replicates per diet during 7-week trial period. The results indicated that dietary inclusion of rice bran decreased. Phytase supplementation increased egg production. Dietary inclusion of rice bran increased feed intake and feed conversion ratio comparing with control diet. Phytase did not affect on feed intake; however improved feed conversion ratio. Dietary non-phytate phosphorus level did not affect on feed intake, egg production and feed conversion ratio. From the results of this study, it can be concluded that rice bran can be included in laying hens' diets up to 7.5% with no adverse effects on performance. Decreasing non-phytate phosphorus level of Lohmann LSL-Lite hens' diet up to 0.029% would be beneficial way to minimize environmental pollution and decrease dietary phosphorous expenses with no adverse effect on productive performance and egg quality characteristics.

Mika *et al.* (2011) conducted a study to evaluate the effects of dietary supplementation of phytase B (product of the *Aspergillus nigerphy* B gene expressed in *Trichoderma reesei*) on feed intake, laying performance, eggshell quality, and on phosphorus and calcium balance in laying hens. Seventy-two, 40 weeks old Hy-Line Brown hens were fed for 14 weeks the following four phosphorus-deficient (0.12% nonphytate phosphorus, NPP), maize-soybean meal-based diets: (1) calcium-deficient (2.8% Ca) control diet; (2) diet 1 + phytase B at the activity of 2.5 acid phosphatase units (AcPU/kg); (3) control diet (3.8% Ca); (4) diet 3 + phytase B at the activity of 2.5 AcPU/ kg. Each dietary treatment was fed to 18 cages of hens, 1 hen/cage kept in individual cages. Hens fed the NPP and Ca deficient diets consumed more feed ($P < 0.01$) and excreted less calcium ($P < 0.01$) than those receiving P-deficient diets with the standard calcium level. There were no effects of calcium level on feed utilization, egg mass, egg weight, and eggshell breaking strength. Egg production, although numerically higher in hens fed low Ca diets with no enzyme added, failed to be significantly different due to the low number of hens investigated and therefore the measurement should be considered as preliminary and supplementary. Phytase B increased mean egg weight by about 7% in layers fed the NPP and Ca deficient diet (Ca × phytase B interaction, $P<0.05$), increased shell breaking strength, particularly at the standard calcium level, significantly enhanced amounts of calcium retained by layers and amounts of phosphorus retained by hens fed the Ca-deficient diets. Additionally, phytase B improved Ca retention at both dietary Ca levels and phosphorus retention in hens fed the Ca-deficient diets. Results of the study indicate that the efficacy of phytase B in NPP-deficient diets is strongly influenced by the dietary calcium level and the enzyme may modulate egg weight, eggshell quality, phosphorus and calcium retention in laying hens fed low-NPP, maize-soybean meal-based diets.

Mohammed *et al.* (2010) a total number of three hundred and fifteen 22-weeks old, commercial Hy-line White-36 hens were randomly assigned into five groups, each group contains nine replicate (seven birds per each) to study the effect of phytase supplementation to diets containing rice bran on performance and egg quality of laying hens. The first group was fed basal diet without phytase supplementation (control) and the 2, 3, 4 and 5 groups, were fed on basal diet supplemented with phytase at levels 0.1, 0.15, 0.20 and 0.25 % respectively. The present results indicated that phytase supplementation significantly ($p<0.05$) or ($p<0.01$) increased henday production, accumulative eggs number, egg mass and also improved significantly ($p<0.05$) feed

conversion ratio. While, egg weight was significantly decreased. However, feed consumption and egg quality measurements expressed as shell thickness, yolk percentage, albumen percentage did not affected by phytase enzyme supplementation. On the other hand, the overall mean of shell percentage was significantly ($p<0.05$) increased. In conclusion, It is concluded that the best level of phytase supplementation was (2 kg phytse / ton feed) in diets contains rice bran for laying hens performance and egg shell percentage.

Jubarah *et al.* (2010) phosphorus is an indispensable mineral, crucial to growth and development both structurally and metabolically but phytate (a major storage form of phosphorus in seeds of plants) decreases mineral bioavailability and nutrient digestibility. Subsequently, in determining the influence of phytate with/without phytase or citrate on laying performance, 5 soybean-maize based diets of similar nutrient density were formulated: a control diet with 0.21% phytate-P and 0.36 % nonphytate-P but no added phytate, and a second similar diet except that the inorganic phosphorus source was replaced by added phytate-P. The third, fourth and fifth diets were similar to the second diet but they contained 0.3% phytase, 1.5% citrate, 0.3% phytase plus 1.5% citrate, respectively. Each treatment was randomly assigned to 35-week-old laying hens. Hens fed the phytase diet showed a pronounced increase in egg production ($P<0.05$), followed by the citrate treatment, while the phytate-P based diet depressed egg production compared to the rest of the treatments. Though the differences in eggshell thickness were insignificant ($P>0.05$), egg weights were affected ($P<0.05$) by the various dietary treatments; the phytase plus citrate diet yielded the highest egg weight improvement. Carbonic anhydrase in the shell gland mucosal extracts depicted significant variation in its specific activity ($P<0.05$), with no observed differences between the control and the phytate-P, phytase or citrate treatments, but phytase combined with citrate had significantly the highest activity of carbonic anhydrase. Serum zinc and yolk iron were significantly improved ($P<0.05$) by phytase and/or citrate treatments, but serum copper or yolk total lipid and mineral concentrations were not affected by the dietary treatments ($P>0.05$). Both phytase and phytase plus citrate treatments yielded high serum and yolk total cholesterol, followed by the citrate and phytate-P, and the least for the control diet. Supplementary phytase and/or citrate seem to increase nutrients availability. Whether citrate increases susceptibility of phytate to phytase by removing cations bound to phytate or creating digesta pH conducive for phytate hydrolysis is yet to be determined.

Lim *et al.* (2003) An experiment employing a factorial arrangement of two levels (3.0 and 4.0%) of Ca, two levels (0.15 and 0.25%) of nonphytate phosphorus (NPP), and two levels (0 and 300 U/kg diet) of microbial phytase was carried out with 960 ISA-brown layers from 21 to 41 wk of age. There was a significant interaction between NPP level and phytase for egg production. High NPP level and phytase supplementation increased egg production only in the second 10 weeks period (31 to 41 wk). High NPP and low Ca increased feed intake, and a significant interaction between levels of NPP and Ca was observed in the first 10 wk. High NPP improved feed efficiency only in the second 10-wk period. Low NPP improved egg specific gravity and eggshell thickness but decreased Haugh units in the first 10 weeks period; high NPP decreased the percentage of broken and soft-shell eggs in the second period. Low Ca decreased egg specific gravity, eggshell strength, and eggshell thickness in both periods and increased Haugh units in the second 10-wk period. Phytase supplementation decreased the percentage of broken and soft-shell eggs. High NPP increased fiber availability but decreased Ca availability. High Ca decreased Ca availability, whereas phytase increased availability of dry matter, fiber, and P. High NPP increased retention of P and Fe but also increased excretion of P. High Ca decreased retention of Zn and Fe. Phytase supplementation increased P retention, resulting in decrease of P excretion. In conclusion, supplementation of microbial phytase at a level of 300 U per kg diet of laying hens can improve egg production, decrease broken and soft egg production rate, and P excretion. The effects of phytase supplementation are significantly modified by the level of Ca and NPP

Jalal *et al.* (2000) Hens were fed corn-soybean meal diets containing nonphytate phosphorus (NPP). Phytases A and B were added at 0.25, 0.15, and 0.10% at 250 to 300 units of phytase (FTU)/kg feed in a 3 × 3 factorial; 0.35% was a control diet. Phytase supplementation had a significant effect on several production parameters: feed intake, feed conversion, and egg mass. Results showed nonsignificant effects ($P < 0.06$) on feed intake when hens were supplemented with phytase A or B and consumed more feed compared to the basal diet at 0.10% NPP. The feed conversion of birds fed 0.10% NPP without phytase was the least efficient compared to the other nine treatments ($P < 0.05$). Egg mass was significantly greater for hens supplemented with phytases A and B than for hens fed the basal diet at low (0.10%) NPP ($P < 0.05$). There were no significant differences in egg production, egg weight, specific gravity, Haugh units, wet shell, or dry yolk percentages. Phytase supplementation improved Ca and P digestibilities to varying

degrees. Supplementation of phytase in normal, corn-soybean meal diets improved feed intake, feed conversion, and egg mass and elicited a response in shell quality and egg components at the low (0.10%) NPP.

2.2 Review of literature on effects of phytase on Feed conversion and growth performance

Saima *et al.* (2014) A 28 days' trial was conducted to evaluate efficacy of microbial phytase in diets for Japanese quails. For this purpose, 900 experimental birds were divided into six groups, each group containing 150 chicks and further sub-divided into10 replicates. Diet A (positive control) was formulated according to NRC (1994) requirements set for the Japanese quail (CP 24% and ME 2900 Kcal/Kg). Diet B differed from diet A in Ca (Calcium) and P (Phosphorus) i.e. 0.15% Ca and 0.20% P less to Diet A, respectively. Four different levels of phytase enzyme (250, 500, 750, 1000 FTU/kg of feed) were added to diet B to formulate diets C, D, E and F treatments respectively. Results revealed that body weight gain, feed consumption, FCR, keel /shank length, dressing percentage of birds in groups consuming 750 and 1000 FTU/kg phytase were significantly higher ($P<0.05$) than those of B, C and D. The growth performance of group E and F was comparable with those of group A (+ve control). Maximum leg weakness, swollen joints and crippled legs were observed in group B (39.30%) followed by C, D (21.33%, and 16.0%). Keeping in view, performance and mortality rate, it is recommended that microbial phytase may be used with greater confidence in Japanese quail ration.

Narasimha *et al.* (2013) conducted a trial to evaluate pure NSP enzyme combination derived from in vitro studies and commercially available phytase to corn-soybean meal based low energy diets singly and combination of both. The experiment was conducted by using completely randomized design on one hundred and fifty layer birds (40 weeks) of uniform body weight and production with five treatments, six replicates and five hens in each replicate for three laying periods with twenty eight days in each laying period. The performance was measured in terms of egg production, feed intake, weight changes, feed efficiency, egg quality, nutrient retention, and gut health. Egg production improved ($P<0.05$) with supplementation of phytase alone or in combination of phytase and NSP (non-starch polysaccharides) enzymes. No effect of supplementing NSP enzymes, phytase alone or in combination was observed on feed intake, FCR, egg quality traits and

retentions of DM, OM and NFE. Significantly (P<0.05) higher retentions of CP, CF, EE, GE and phosphorus was observed with supplementation of NSP enzymes and similar trend was observed with both NSP and phytase to BD except for phosphorus indicating no associative effect of phytase and NSP enzymes on above nutrient retentions. Intestinal pH, viscosity and E. coli count significantly (P<0.05) reduced with supplementation of NSP enzymes and no further improvement was observed on these variables with supplementation of phytase with NSP enzymes. Gut histology revealed broad and disrupted villi with little goblet cell activity. No significant (P<0.05) effect on feed cost due to addition of phytase and/or NSP enzymes to BD was observed. The cost of feed to produce dozen eggs was comparable among SD, BD and BD supplemented with NSP enzymes and phytase.

Gao *et al.* (2013) compared the efficacy of a novel transgenic corn-derived phytase (TCDP) and two other commercial microbial phytases (PA and PB) in the long-term feeding study of laying hens. The treatments consisted of a positive control (PC) diet adequate in phosphorus (P); a negative control diet low in P (0.10% NPP. Eight diets were fed to Hy-line hens (n = 576) from 50 to 66 weeks of age. They found that with a reduction in dietary P in the NC diet, egg production, egg mass, feed intake, final BW, BW gain, eggshell thickness, and eggshell strength of laying hens decreased (P<0.05). In addition, the number of soft-shelled, cracked and broken eggs increased (P<0.05) in the NC group. The addition of TCDP, PA or PB significantly increased laying production and egg quality (P<0.05), and performed similarly in hens fed the PC diet. Hens fed each source of phytase had greater apparent ileal P digestibility, tibia ash, and bone breaking strength than hens fed the NC diet (P<0.05). Results from this study indicate that the addition of TCDP to a P-deficient diet improves laying performance, egg quality, ileal P utilization, and bone mineralization, and TCDP is as efficacious as two commercial microbial phytases when P-deficient diets for laying hens were supplemented with it.

Tang *et al.* (2012) The experiment was conducted to assess the effects of a novel thermostable phytase in male broiler chicks (Ross 308) fed available P (AP)-deficient diets on growth performance and bone mineralization. The treatments consisted of 8 experimental diets: 1 positive control diet containing an adequate level of AP, 1 negative control diet deficient in AP, and 6 diets with the same level of AP as in the negative control but supplemented with different levels phytase. The addition of phytase significantly improved (P < 0.05) BW gain, feed intake, FCR, toe ash, tibia ash, and tibia

P of broilers compared with those fed the negative control diet. No significant differences (P > 0.05) were found in FCR and bone mineralization among the broilers fed different levels of phytase and those fed the positive control diet. In conclusion, normal growth performance and bone mineralization were maintained in broilers fed AP-deficient diets supplemented with thermostable phytase.

Abou-Elezz *et al.* (2011) this study consisted of two experiments, aimed at determining the effect of the dietary inclusion of either *Leucaena leucocephala* (LLM) or *Moringa oleifera* (MOLM) leaf meals on Rhode Island Red (RIR) hens' egg production and quality. In the first experiment, thirty six RIR hens, at 36 weeks of age, were randomly divided into four groups each of nine birds and were allocated in individual cages. The four groups corresponded to four dietary treatments containing 0 (control), 5, 10, and 15% of LLM, respectively. Simultaneously, the second experiment was carried out following the same design but using MOLM instead of LLM. The egg production and quality traits were monitored for five weeks, preceded by one week of adaptation. The results showed a quadratic effect on the egg laying rate (57.10, 57.46, 53.25, and 47.46%), egg mass (g/hen/d) and feed conversion due to the LLM treatments (0, 5, 10, and 15%, respectively). The MOLM treatments decreased linearly the egg laying rate (60.00, 59.72, 56.13, and 51.87%) and the egg mass, and had a quadratic effect on the feed intake (111.15, 111.93, 107.08, and 100.47g/hen/d) when including 0, 5, 10, and 15% of MOLM, respectively. The yolk color increased linearly by the rise in both the MOLM and the LLM levels. Other results were obtained in the albumen and yolk proportions (%) and in the yolk coefficient, while no adverse effects were found on the other egg quality traits due to the LLM or MOLM treatments. The MOLM or the LLM could be acceptable as sustainable feed resource up to 10 % in laying hen diets.

Yan *et al.* (2009) experimented on 68 weeks old Hy-Line brown laying in a 6-week feeding trial to compare the efficacy of phytases Optiphos (OPT) and Natuphos (NAT), which were isolated from Escherichia coli and Aspergillusniger, respectively. Feed intake, egg production, egg quality, apparent nutrient digestibility and serum P and Ca concentration were evaluated to compare the effect of the two phytases. Feed intake and eggshell thickness were not affected by the treatments. Superior effects ($p<0.05$) of OPT were only observed in egg production and egg weight compared with NAT. Characteristics such as eggshell breaking strength, apparent digestibility of N, Ca and P and serum P concentration were equally increased with the supplementation of both

phytases ($p<0.05$), where no significant difference was observed in those characteristics between PC and phytase supplementation at 500 FTU/kg. Equally effective improvements ($p<0.05$) were also observed in egg production and DM digestibility, where no improvements were observed ($p<0.05$) between the PC group and the groups with phytase supplementation at 500 FTU/kg. Equal increases in the serum Ca level were observed when the groups with phytase supplementation were compared to the PC group. Overall, the results of this study suggest that NAT and OPT are equally effective at liberating phytate-bound complexes when included in 0.2% available phosphorus diets for 68 weeks laying hens; either source of phytase can be fed to commercial 68 weeks laying hens at 500 FTU/kg to correct the negative effects associated with a 0.2% available phosphorus diet. In conclusion, either source of phytase can be fed to commercial first cycle laying hens at 500 FTU/kg to effectively replace inorganic phosphorus when economically justified.

Liu *et al.* (2007) the effects of phytases on the performance of layers and the ileal nutrient digestibility of corn, soybean, and by-product meal-based diets were assessed with 320 Hy-Line brown layers from 23 to 28 weeks of age. Layers were grouped randomly into 5 treatments, with 8 replicates per treatment and 8 layers per replicate. The 5 diets consisted of a positive control diet with adequate Ca (3.30%), total P (0.50%), and nonphytate P (NPP; 0.28%), and a negative control diet and 3 phytases (phytase A derived from Aspergillusniger, and phytases B and C derived from Escherichia coli) supplemented at 300 phytase units/kg of feed, respectively. Phytase supplementation in the negative control diet improved the digestibility of P and Ca by 11.08 and 9.81% ($P < 0.05$), respectively, whereas it improved the digestibility of amino acids by 2 to 8% ($P<0.05$). Supplementing phytases in the negative control diet improved the rate of lay, egg mass and eggshell quality to the level of birds fed the positive control diet. These results suggest that supplementing phytases can improve the digestibility not only of Ca and P but also of amino acids in layers. The use of phytase reduces phosphorus excretion in poultry manure by allowing the birds to utilize more of the phytate phosphorus. Phytate phosphorus has the ability to complex with cations such as calcium, magnesium, zinc, copper and nitrogen and certain gastrointestinal proteases thus reducing the availability of these cations and of amino acids. The use of phytase may free these cations and proteases bound in phytate phosphorus complexes and improve many production parameters and body structure characteristics in broilers and

laying hens, such as body weight, bone ash content, feed consumption, egg weight and eggshell quality.

Francesch *et al.* (2005) performed a 24 weeks trial to evaluate the efficacy of an experimental phytase on performance, egg quality, tibia ash content and phosphorus excretion in laying hens fed on either maize or barley based diet. At the end of the trial, an ileal absorption assay was conducted in order to determine the influence of phytase supplementation on the apparent absorption of calcium and total phosphorus. Each experimental diet was formulated either as a positive control containing 3.2 g/kg Non-Phytate Phosphorus (NPP), with the addition of dicalcium phosphate or as a low P one without DCP addition. Both low phosphorus diets (containing 1.3 or 1.1 g/kg NPP) were supplemented with microbial phytase at 0, 150, 300 and 450 U/kg. Low dietary NPP (below 1.3 g/kg) was not able to support optimum performance of hens during the laying cycle (from 22 to 46 weeks of age), either in maize or barley diets. Rate of lay, daily egg mass output, feed consumption, tibia ash percentage and weight gain were reduced in hens fed low NPP diets. The adverse effects of a low Phosphorus diet were more severe in hens on a maize diet than in those on a barley diet. Low dietary NPP reduced egg production, weight gain, feed consumption and tibia ash content and microbial phytase supplementation improved these parameters. Hens given low NPP diets supplemented with phytase performed as well as the hens on positive control diets containing 3.2 g/kg of NPP. A 49% reduction of excreta Phosphorus content was achieved by feeding hens on low NPP diets supplemented with phytase, without compromising performance. Phytase addition to low NPP diets increased total phosphorus absorption at the ileal level from 0.25 to 0.51 in the maize diet and from 0.34 to 0.58 in the barley diet. Phosphorus absorption increased linearly with increasing levels of dietary phytase

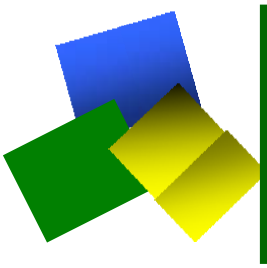
Musapuor *et al.* (2005) conducted an experiment to study the effects of different levels of phytase, vitamin D3, calcium and available phosphorus on phytate phosphorus utilization in laying hens. Dietary phytase caused a significant ($P<0.05$) increase in feed consumption, feed conversion ratio, tibia ash weight, tibia ash percentage, tibia phosphorus plasma phosphorus and phosphorus digestibility. However, dietary phytase caused a significant ($P<0.05$) decrease in plasma alkaline phosphatase activity and excreta phosphorus percentage. Also phytase had no beneficial effect on egg shell quality traits. Interaction between phytase and calcium on tibia phosphorus, plasma calcium and excreta phosphorus were significant ($P<0.05$). Interaction between phytase and available

phosphorus on tibia phosphorus was significant (P<0.05). Overall, they concluded that in low phosphorus diet which food consumption is low, phytase would increase food consumption as well as retention of phosphorus in bones. Also, the lower excreta of phosphorus by using phytase could decrease pollution.

Casartelli *et al.* (2004) conducted an experiment to evaluate the effects of the enzyme phytase in diets formulated with different phosphorus sources on performance, eggshell quality and excretion of commercial laying hens. Two hundred and eighty-eight commercial Hyssex Brown laying hens were evaluated during two production phases, which included eight twenty-eight-day cycles, using a completely randomized design in a 3x2 factorial with six replicates of eight birds per treatment. Three phosphorus sources (calcium and sodium phosphate, micro-granulated dicalcium phosphate and triple super phosphate) and two phytase levels (0 or 1000 FTU/kg diet) were tested in the composition of the diets. After the post-peak period, triple super phosphate decreased bird performance and eggshell quality. It was possible to reduce the levels of phosphorus supplementation when phytase was added to the diet. Besides, phytase supplementation reduced phosphorus, calcium and nitrogen excretions but affected mean egg weight at production peak.

Berry *et al.* (1980) Forty eight laying hens were used in a 6 x 8 randomized block design to compare the efficiency of Leucaena leaf meal and grass meal as sources of yolk pigments. Leaf meals were added to a low pigment diet (LP) to supply 10 or 20 mg dihydroxyxanthophyll (DHX) / kg. Leucaena from Malawi was added to supply both 10mg (L10) and 20mg DHX/kg (L20) that from Bangkok was added to supply 10mg DHX/kg (B10) and the grass to supply 20mg DHX/kg. A fifth diet (L20c) was the same as the L20 diet except that coconut oil replaced the groundnut oil in the basal diet. There were no significant differences between treatments with respect to egg production, egg yield, mean daily food intake and live weight change during the 28-day experimental period. After 7 days, significant differences (P<0.001) in Roche fan score (RFS) and Beta- carotene equivalent (BCE) values were evident between the 3 DHX levels (0, 10 and 20 DHX/kg). Further changes during the second week were followed by stabilization in yolk colour during the last two weeks. No differences were detected within DHX level for visual yolk colour measurement (RFS) except that on day 28 the RFS for the L20c diet was detected as significantly greater (P <0.05) than the other two 20mg DHX treatments (L20 and G20). BCE measurements gave significant differences (P<0.001)

within the 20mg DHX treatments (L20 vs. L20c +G20) from day 7 onwards. In addition on day 28 the BCE values for diets L20c and G20 were detected as significantly different ($P<0.001$). The absence of any deleterious effects on egg production in this short term study suggests that at inclusion rates of 10-25 g/kg leucaena is an effective yolk pigmenter.



CHAPTER III

MATERIALS AND METHODS

CHAPTER III

MATERIALS AND METHODS

The experiment was conducted in the Department of Physiology & Pharmacology, Hajee Mohammad Danesh Science and Technology University, Dinajpur.

Study Area: Karnai, Basherhat, Dinajpur

Study Duration: Six weeks (From 01 June, 2015 to 14 July, 2015)

3.1 Experimental Layout

28 chickens of 81 weeks old were randomly selected and equally divided into 4 groups (A, B, C and D).

Chickens of Group 'A': 7 chickens were kept and are considered as control, fed only with commercial layer ration.

Chickens of Group 'B': 7 chickens were supplemented with formulation of 2 gm grinded Ipil Ipil leaves per kg feed.

Chickens of Group 'C': 7 chickens were supplemented with formulation of 2 gm grinded Bean leave per kg feed.

Chickens of Group 'D': 7 chickens were supplemented with formulation of 1 gm grinded Ipil Ipil leaves plus 1 gm grinded Bean leaves per kg feed.

All the 28 chickens of control and treated groups were closely observed for 6 weeks to complete the research work following steps were followed.

LAYOUT OF THE EXPERIMENT

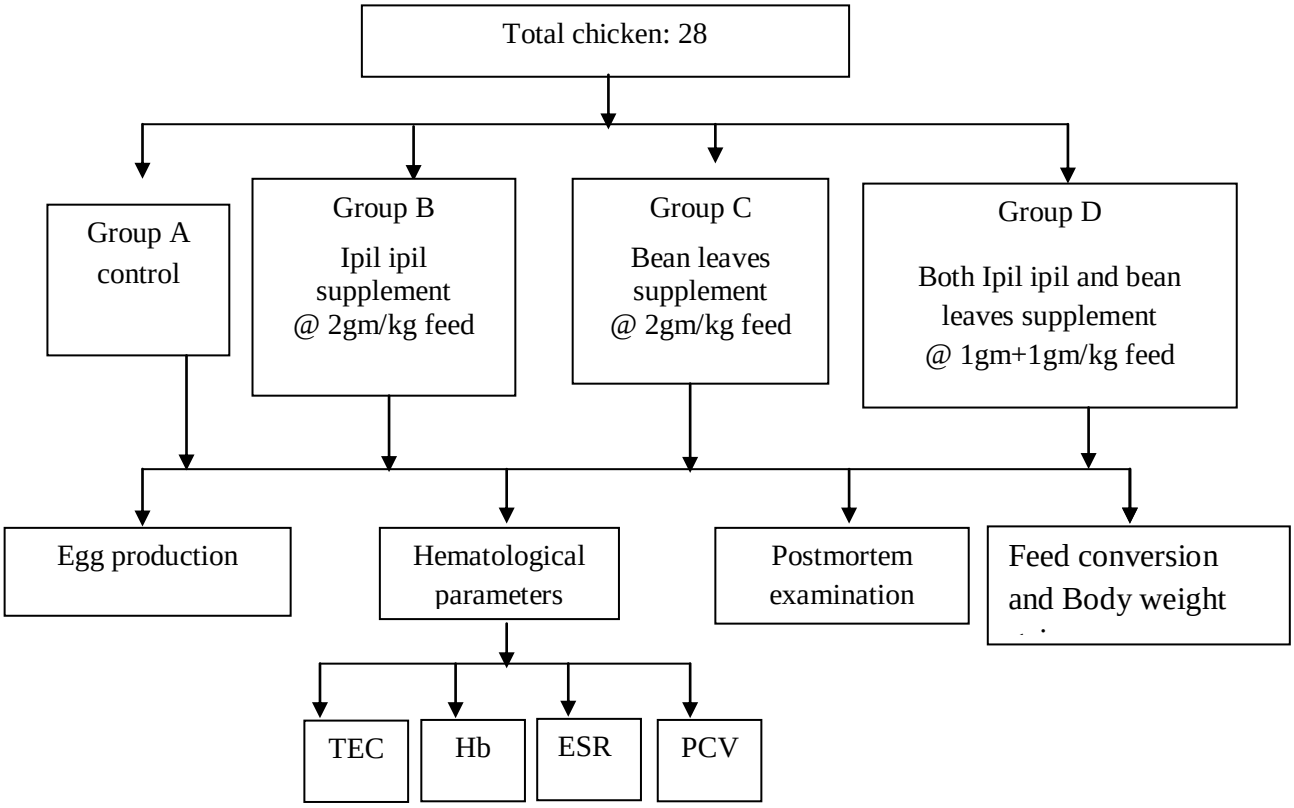


Fig. 1: Layout of the experimental design (each group consisting of seven layer)

3.2 Collection and management of chickens

At 6 weeks of age, Hisex brown layer chickens were collected from local market. The body weights of assigned chickens were taken with digital electronic balance and the results were recorded. The finally selected 28 chickens were housed under normal husbandry condition and reared in layer cage. All the birds were fed with commercial crumbled feed at the rate of 125 gm per bird daily with fresh water *adlibitum.*

Plate 1: Hisex Brown layer in experimental shed

3.3 Measurement of body weight

The body weight of each layer was measured with the help of digital balance.

3.4 Collection of Ipil Ipil and bean leaves

Ipil Ipil and bean leaves were collected from the research field of HSTU

3.5 Feed preparation and supply

The Ipil Ipil and bean leaves were dried and grinded. The grinded leaves were added with commercial layer ration and served to different groups of chickens.

3.6 Clinical examination

Egg production, pause days, body weight, egg quality and hematgological parameters were recorded.

3.7 Hematological test

Blood samples were collected from wing vein of layers of both control and treated groups to study the effect of experiment at blood serum level and the following parameters were observed:

(a) Total Erythrocyte Count (TEC)
(b) Hemoglobin estimation (Hb)
(c) Packed Cell Volume (PCV)
(d) Erythrocyte Sedimentation Rate (ESR)

3.8 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 mark 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 mark. 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 (10X) objective. 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 down 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 objective (45X). 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/µl of blood.

3.9 Determination of Hemoglobin concentration (Hb)

The N/10 hydrochloric acid 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 cmm. 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 hydrochloric acid (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 gm%. The above procedure was matched by the Hellige hemometer method as described by Lamberg and Rothstein (1977).

3.10 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 mark of the right sided scale. 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).

$$\text{PCV\%} = \frac{\text{(Height of the red cell (cm)}}{\text{(Height of total blood (cm)}} \text{ X } 100$$

3.11 Determination of Erythrocyte Sedimentation Rate (ESR)

The fresh anticoagulated blood was taken into the Wintrobe hematocrit tube by using special loading pipette exactly up to 0 mark of the left sided scale. 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.

3.12 Egg production record

Egg production was recorded for each layer at the same time each day during laying period. The incidence of broken eggs and soft-shelled eggs were identified and recorded. The number of eggs laid on successive days by a particular hen determined the length of each sequence and the number of pauses in each hen's oviposition determined the number of sequences. For each layer the length of laying sequence was determined on the day the last egg of the current clutch was laid.

3.13 Observation of internal and external egg quality

Egg qualities were measured from those eggs laid by layers of different treatment group. Measured egg qualities were egg weight, shell dry weight, fresh albumin weight, fresh yolk weight, egg shell thickness, height of the thick albumin, height of the yolk, width of the yolk, width of the egg and diameter of the egg albumin. For quality determination egg weight was recorded by an electric weighing balance. The length of egg was measured by a slide calipers. The width was also estimated by slide calipers. The eggs were then carefully broken down on a glass plate (40 x 20cm) to determine the internal egg qualities.

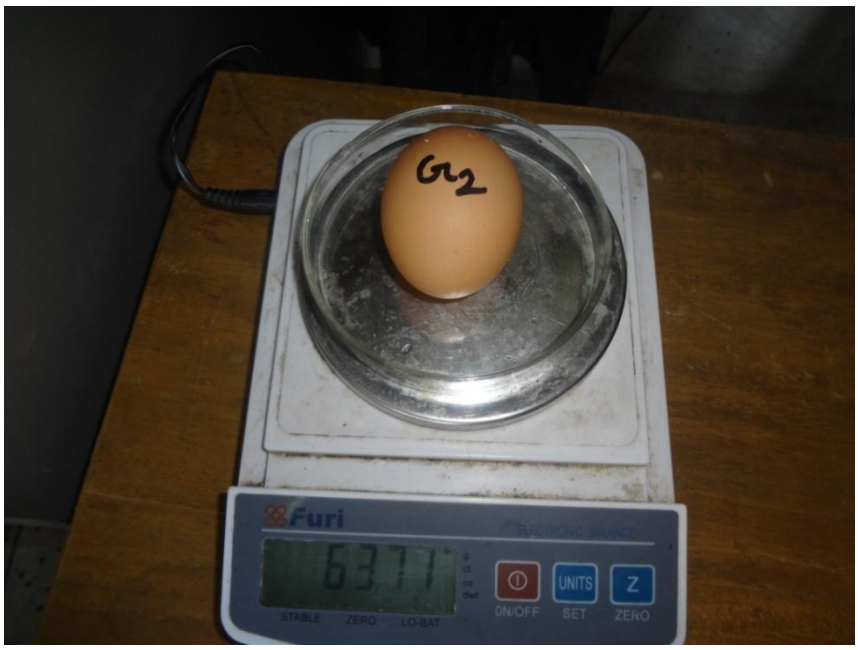


Plate 2: Measurement of egg weight

Plate 3: Measurement of egg length

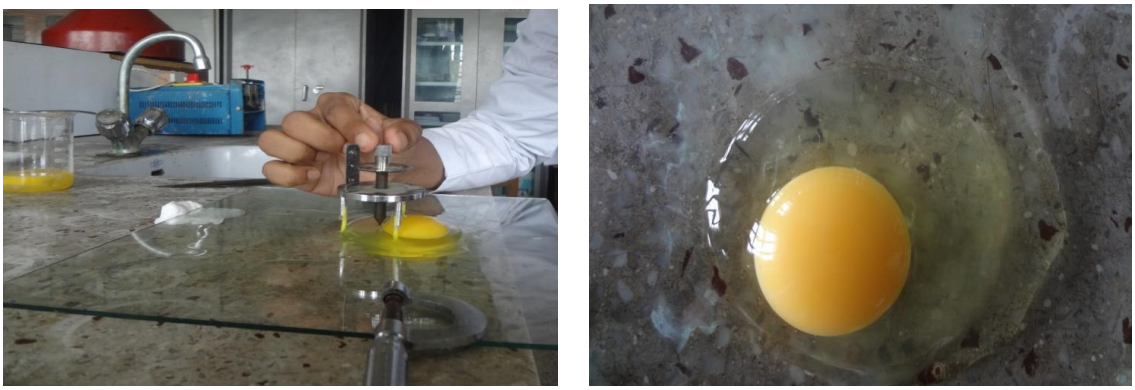


Plate 4: Observation of internal and external egg quality

3.13.1 Weight of different egg component

The method outlined by Chowdhuri (1988) was followed for partitioning different egg components. At first, egg was broken on glass plate. Then the yolk was separated carefully from albumin with the help of a spatula and transferred to a previously weighed petridish by a spatula and weighed. Precautions were taken at all stages to avoid rupture of yolk.

Plate 5: Measurement of fresh shell weight **Plate 6: Measurement of fresh yolk weight**

The shell of the broken eggs were rinsed and washed thoroughly in tap water keeping the membranes intake. The washed shells with membrane were immersed in a beaker of water for removal of the shell membranes. The shell and shell membranes were oven dried separately at 105 cover night keeping them in a glass petridish. On the following day, oven dried shell and shell membranes were taken. Finally the following calculations were made for different components suggested by Chowdhuri (1988).

1. Fresh yolk weight: {(weight of yolk + weight of petridish) - weight of petridish}.

2. Fresh albumin weight: {(Weight of wet albumin + weight of petridish)-weight of petridish}.

3.14 Shell thickness

After removing of shell membrane, shell thickness (mm) was measured by screw gauge.

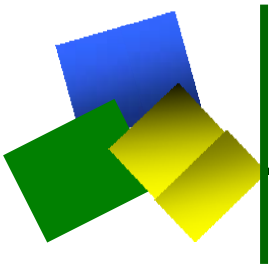
Plate 7: Measurement of shell thickness

3.15 Postmortem examination for side effect

Two layers from every group were slaughtered to see if there any pathological changes present on the period of experiment. There was no significant pathological change in any internal organs of the layers of treated groups. In case of treated birds increase number follicles in ovary was observed than in control.

3.16 Statistical analyses

Data were analyzed by analysis of variance using Completely Randomized Design with factorial arrangement of time and treatments (Steel and Torrie, 1986). All analyses were performed by MSTATC and SPSS program.



CHAPTER IV

RESULTS AND DISCUSSION

CHAPTER IV

RESULTS AND DISCUSSION

This study investigated the effect of Ipil Ipil and Bean leaves extract supplementation on feed conversion, growth performance and egg production of chicken.

This experiment was conducted in the Department of Physiology and Pharmacology, Faculty of Veterinary and Animal Science. The results of these studies are discussed under following headings.

4.1 Recording of egg production

Egg production of different groups of hens is presented in Table 1. Layers fed with Ipil Ipil, Bean leaves and combined treatment supplementation showed an increased egg production, reduced the number of laying pauses between sequences than control group (Normal commercial feed).

Table 1: Effect of Ipil Ipil and bean leaves supplementation on egg production of chicken during experimental period

Group (n=7)	**82 weeks (Mean ± SE)**	**83 weeks (Mean ± SE)**	**84 weeks (Mean ± SE)**	**85 weeks (Mean ± SE)**	**86 weeks (Mean ± SE)**	**87 weeks (Mean ± SE)**	**Total**	**Average**	**Percentage**
A (Control)	$29^{a} \pm 1.15$	$32^{a} \pm 1.73$	$31^{a} \pm 2.08$	$31^{b} \pm 2.31$	$29^{b} \pm 2.31$	$28^{b} \pm 1.73$	180	30	61.22%
B (Ipil Ipil)	$34^{a} \pm 1.73$	$36^{a} \pm 2.65$	$35^{a} \pm 2.30$	$37^{ab} \pm 1.73$	$38^{a} \pm 1.74$	$37^{a} \pm 2.89$	217	36.16	73.81%
C (Bean leaves)	$35^{a} \pm 1.74$	$35^{a} \pm 1.15$	$36^{a} \pm 1.53$	$36^{ab} \pm 1.73$	$35^{ab} \pm 2.30$	$36^{ab} \pm 2.31$	213	35.5	72.44%
D (Both)	$35^{a} \pm 2.65$	$37^{a} \pm 1.73$	$38^{a} \pm 2.31$	$39^{a} \pm 2.30$	$40^{a} \pm 1.73$	$38^{a} \pm 2.88$	227	37.83	77.21%

Note: Values followed by same superscripts in the same column are not statistically significant ($p>0.05$), different superscripts indicate that difference is significant ($P<0.05$).

In this and other tables, A = control (Normal ration), B=Supplied with Ipil Ipil, C = Supplied with Bean leaves D= Supplied with both preparation.

Table 1 revealed that,

On 82th week the layers of A, B, C and D showed significant ($P<0.05$) changes in egg production. Group B, C and D showed maximum egg production.

On 83th and 84th week there was no significant difference among groups in terms of egg production.

On 85^{th} 86^{th} and 87^{th} week the egg production rate steadily progressed and group D showed best performance among four groups. The egg production difference was statistically significant ($P<0.01$).

The layers of group D fed with both Ipil Ipil and bean leaves preparation statistically significant ($P<0.01$) and more efficient among the other groups.

Table 1 also revealed that,

In Group A total egg production during experimental period 180 and total pause day 114.

In Group B total egg production during experimental period 217 and total pause day 77.

In Group C total egg production during experimental period 213 and total pause day 81.

In Group D total egg production during experimental period 227 and total pause day 67.

The layers of Group D showed increasement in egg production, reduced the number of laying pauses between sequences among the other groups and it is significant at 1% ($p<0.01$) level. The current study showed similarities with Mutayoba *et al.* (2003) at lower rate of supplementation.

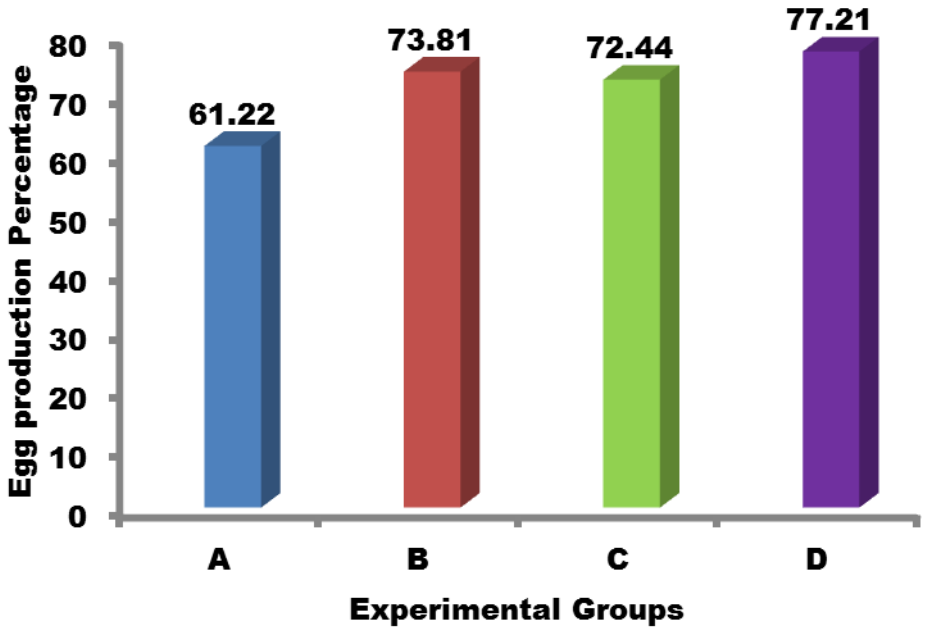


Fig. 2: Graphical presentation of egg production percentage in different groups

In this graph the total egg production of four experimental groups is shown. Group A showed less egg production (61.22%) than other three groups. Group B and C showed significant increase in egg production and quality. On the other hand Group D showed maximum egg production performance (77.21).

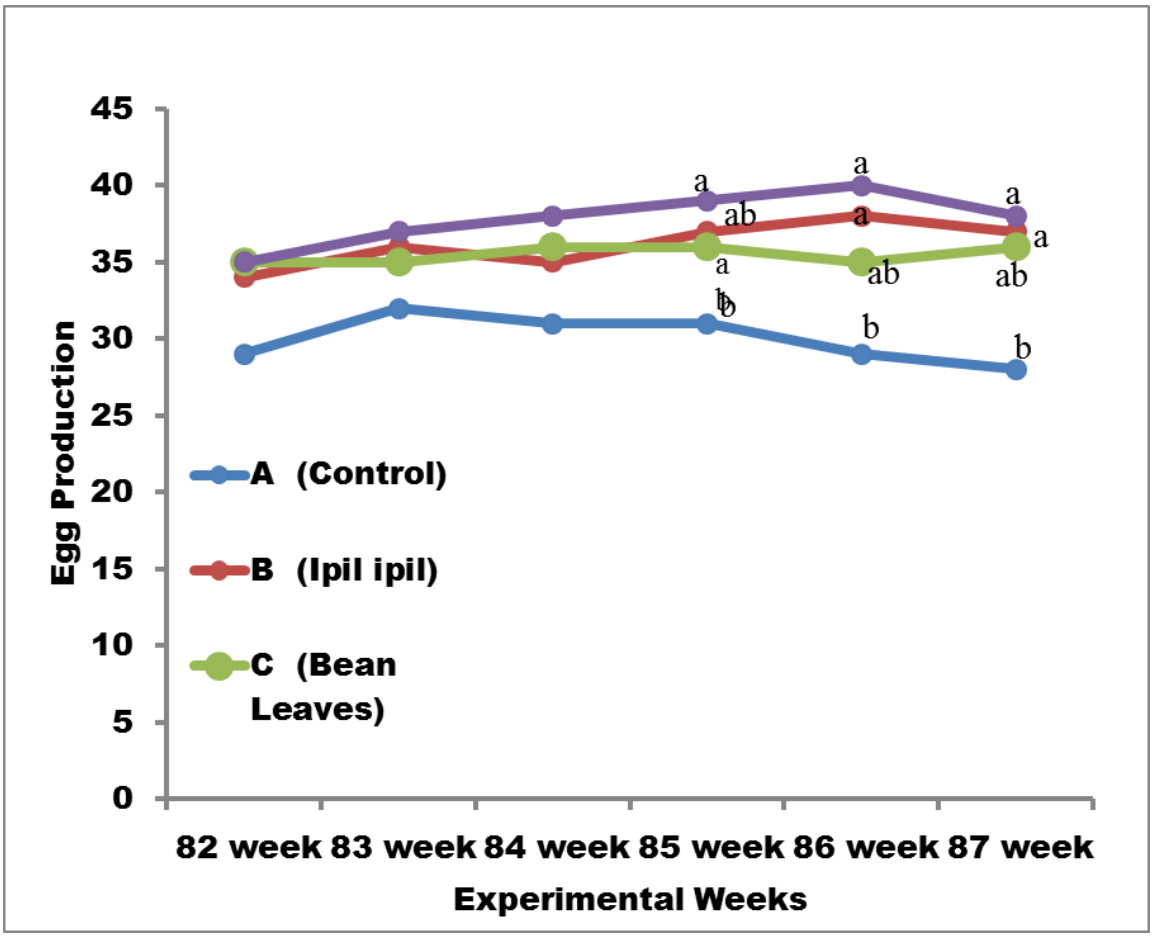


Fig. 3: Graphical representation of progress of egg production

Here is a line graph of egg production performance of the layers. Group A showed gradual decrease in egg production week by week. On the other hand other three groups showed steady progress. Group D showed best performance in egg production. The egg production performance of Group B and C was better than group A which is in agreement with the findings of the present study.

4.2 Recording of Feed Intake

Table 2: Feed consumption

Feed consumption/bird/day (gm)							
Group	82 weeks	83 weeks	84 weeks	85 weeks	86 weeks	87 weeks	Average
A	117	116	118	120	124	123	119.67
B	117	119	122	125	123	125	121.8
C	116	118	120	120	125	127	121
D	119	120	123	121	124	130	122.83

The table shows the daily feed intake of four groups of birds. The birds of group A took slightly more amount of feed than other three groups.

4.3 Recording of Body weight

Table 3: Effect of Ipil Ipil and bean leaves supplementation on body weight of chicken during experimental period

Group (n=7)	82 weeks	83 weeks	84 weeks	85 weeks	86 weeks	87 weeks
A (Control)	$1772.85^{a} \pm 2.64$	$1772.14^{a} \pm 1.84$	$1768.57^{b} \pm 2.60$	$1765.00^{b} \pm 2.67$	$1760.00^{b} \pm 2.88$	$1757.14^{b} \pm 2.85$
B (Ipil Ipil)	$1770.71^{a} \pm 2.54$	$1769.28^{a} \pm 2.77$	$1773.57^{ab} \pm 1.43$	$1774.28^{a} \pm 2.54$	$1779.29^{a} \pm 1.70$	$1779.29^{a} \pm 2.02$
C (Bean)	$1772.14^{a} \pm 2.85$	$1765.71^{a} \pm 2.97$	$1770.00^{ab} \pm 2.67$	$1772.14^{a} \pm 1.84$	$1775.71^{a} \pm 2.97$	$1780.00^{a} \pm 3.08$
D (Both)	$1766.43^{a} \pm 3.03$	$1770.71^{a} \pm 2.97$	$1775.71^{a} \pm 2.02$	$1778.57^{a} \pm 2.60$	$1781.43^{a} \pm 2.60$	$1786.43^{a} \pm 2.82$

Note: Values followed by same superscripts in the same column are not statistically significant ($p>0.05$), different superscripts indicate that difference is significant ($P<0.05$).

Table 3. revealed that,

The weekly body weight among group A, B, C and D didn't differ significantly. After 85^{th} weeks significant changes took place. In 86^{th} and 87^{th} week's body weight gain showed significant increase at the .005 level. Most development was found in Group D, group C and D showed improvement in body weight gain.

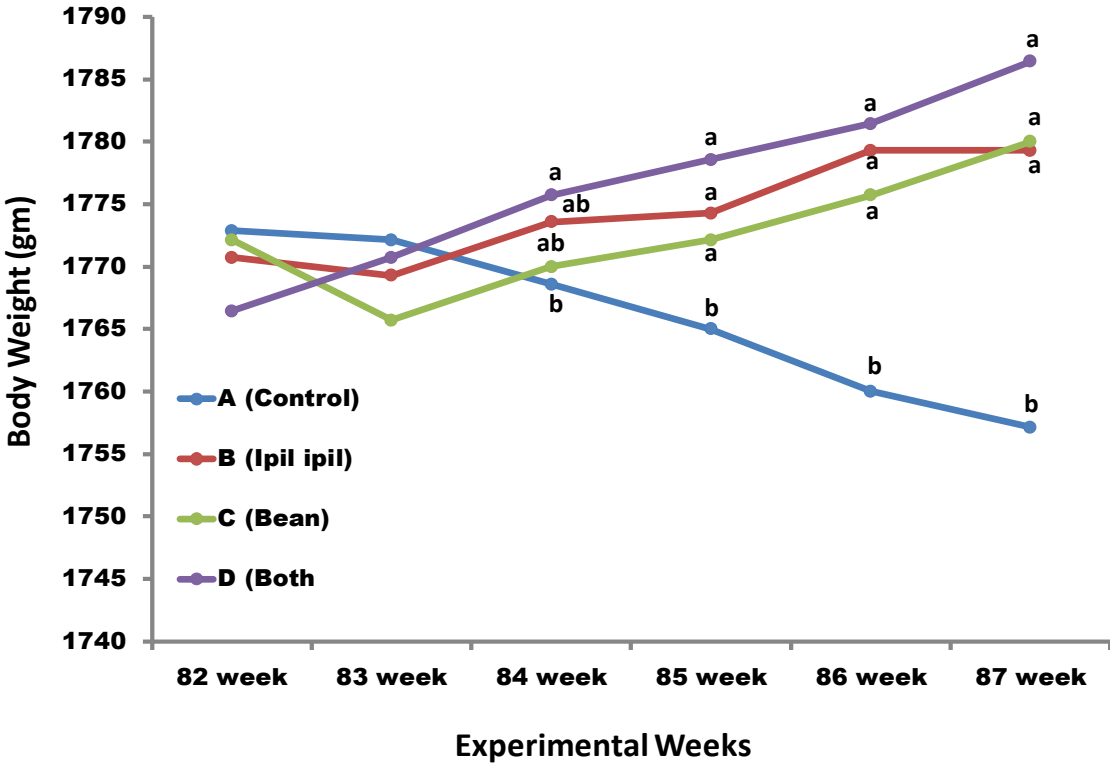


Fig. 4: Line graph of body weight gain during experimental weeks

Here is a graphical representation of progress of the 4 groups through 6 weeks. The layers of group D fed with Ipil Ipil and bean leaves supplement showed statistically significant ($P<0.05$) body weight gain and more efficient among the other groups which is in agreement with the findings of the present study. The result expresses similarities with Eichie *et al.* (2015)

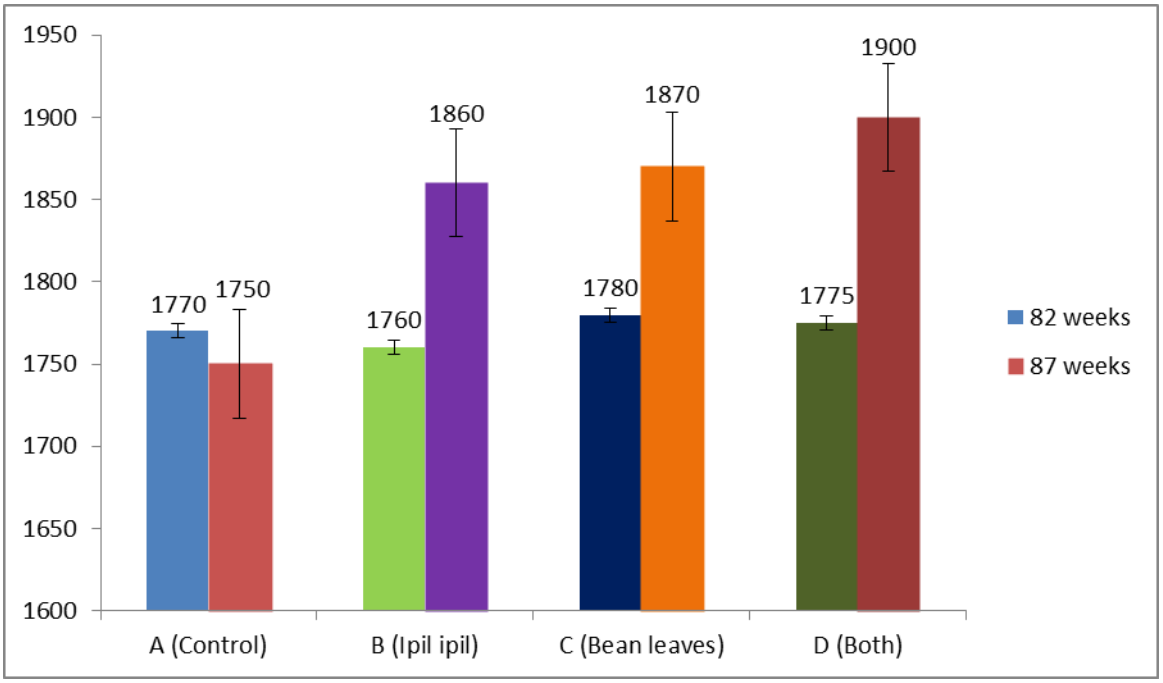


Fig. 5: Body weight gain during experimental weeks in four groups

Body weight of different groups of hens is presented in Table 2. Layers feed with Ipil Ipil (Group B), Bean leaves (Group C) and combined treatment supplementation (Group D), showed an increased body weight, reduced the number of laying pauses between sequences than control group (Normal commercial feed).

4.4 Feed conversion

Feed Conversion Ratio (FCR)

FCR represents the proportion of food that is converted into meat. FCR can be calculated over a set period, e.g. monthly, quarterly, annually or on a room, house or herd basis.

$$\text{FCR} = \frac{\text{Feed intake}}{\text{Average daily gain}}$$

Average Daily Gain (ADG)

The ADG can be calculated by following formulae.

$$\text{AGD} = \frac{\text{Finish weight} - \text{start weight}}{\text{Age (days)}}$$

Feed conversion for egg production on different diets varied significantly (P<O.O1). Change in Feed Conversion in relation to phytase supplemented diet recorded coincide

with some previous findings (Fuller, 1997; Abdul Rahini *et al.,* 1996; Haddadin *et al.,* 1996; Nahashon *et al.,* l996a; Tortuero *et al.,* 1995).

4.5 Overall performance

The overall performance of laying pullets was up to the level of breeder's performance standard. During experimental period the temperature ranged from a minimum of 27 to a maximum of 31°C.

4.6 Cost benefit analysis

Cost of per kg feed was forty (40) taka.

Table 4: Data showing economics of egg production kept under different treatment groups during experimental period (6 week)

Description	A	B	C	D
Cost /layer(Taka)	300	300	300	300
Total cost of hens(Taka)/group	2100	2100	2100	2100
Feed cost /day(Taka)	4.79	4.87	4.84	4.91
Total feed cost (Taka)/group	1408	1431	1423	1444
Total medicine cost (Taka)	18	18	18	18
Miscellaneous cost (Taka)	20.00	20.00	20.00	20.00
Total cost for electricity	10	10	10	10
Sale price/ hen (Taka)	300	315	315	320
Total sale price of hens (Taka)	2150	2205	2205	2240
Sale price/egg (Taka)	8.00	8.00	8.00	8.00
Total sale price of egg (Taka)	1440	1736	1704	1816
Total income (Taka)	3590	3941	3909	4056
Net profit (Taka)	38	362	338	464

Table 4. Revealed that,

The average rearing costs of experimental hens are kept under different treatment groups viz. A, B, C and D was 3552 TK, 3579 TK, 3571TK, 3592 TK respectively. Miscellaneous cost summed up 20Tk in case of A, B, D and C. The average income in group A, B, C and D was 3590 TK, 3941 TK, 3909 TK, 4056 TK respectively. The hen

was sold at the rate of 150 TK/kg and egg was sold 8TK/egg. The net profit the respective group was found to be taka 38, 362, 338, 464 respectively. Supplementation with Ipil Ipil and bean leaves extract was found to be more profitable than control group.

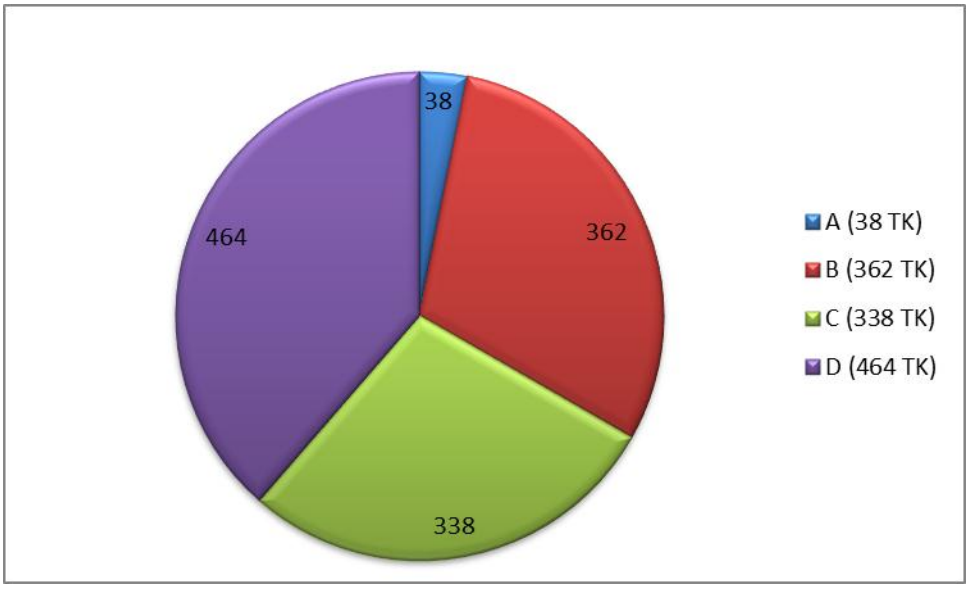


Fig. 6: Economics of Production

4.7 Hematological parameter:

The results of various blood parameters are depicted in table 5.

Table 5. Effect of Ipil Ipil and bean leaves supplementation on hematological parameters

Groups	TEC (million/mm^3) (Mean ± SEM)	Hb (gm/dl) (Mean ± SEM)	PCV (%) (Mean ± SEM)	ESR (in 1^{st} hr) (Mean ± SEM)
A	$3.04^a \pm 0.60$	$10.30^a \pm 0.68$	$30.55^a \pm 0.12$	$5.23^a \pm 0.34$
B	$2.99^a \pm 0.40$	$10.68^a \pm 0.67$	$30.37^a \pm 0.92$	$5.83^a \pm 0.11$
C	$2.99^a \pm 0.59$	$10.63^a \pm 0.64$	$30.30^a \pm 0.51$	$5.89^a \pm 0.20$
D	$2.93^a \pm 0.08$	$10.07^a \pm 0.68$	$30.40^a \pm 0.11$	$5.97^a \pm 0.53$

Note: Values followed by same superscript in the same column are not statistically significant ($p>0.05$), different superscripts indicate that difference is significant ($P<0.05$).

A. Total Erythrocyte Count (million/ mm^3):

Total erythrocyte count is presented in (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 A and lowest in Group D. Although these values show a little fluctuation they were not statistically significant ($p>0.05$).

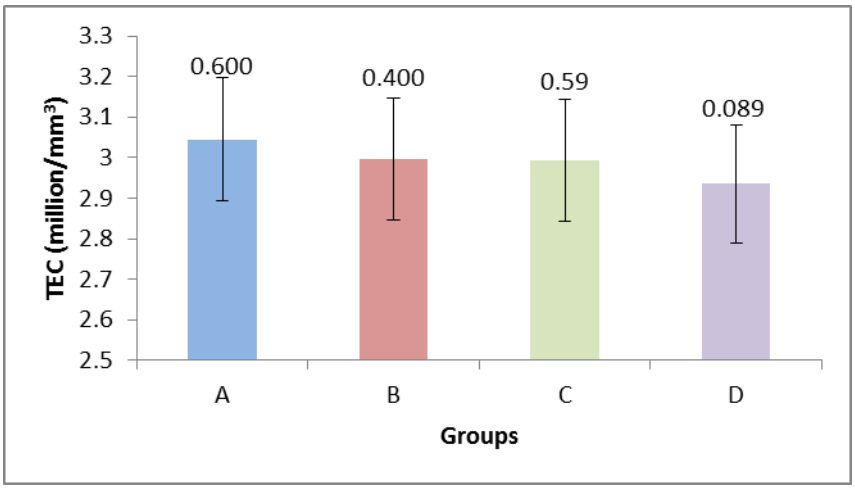


Fig. 7: TEC (million/mm^3) during experimental period

The superscript number above bar indicates standard error (SE)

B. Estimation of Hemoglobin (gm/dl)

Hemoglobin content is presented in (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. These values show a little fluctuation they were not statistically significant ($p>0.05$).

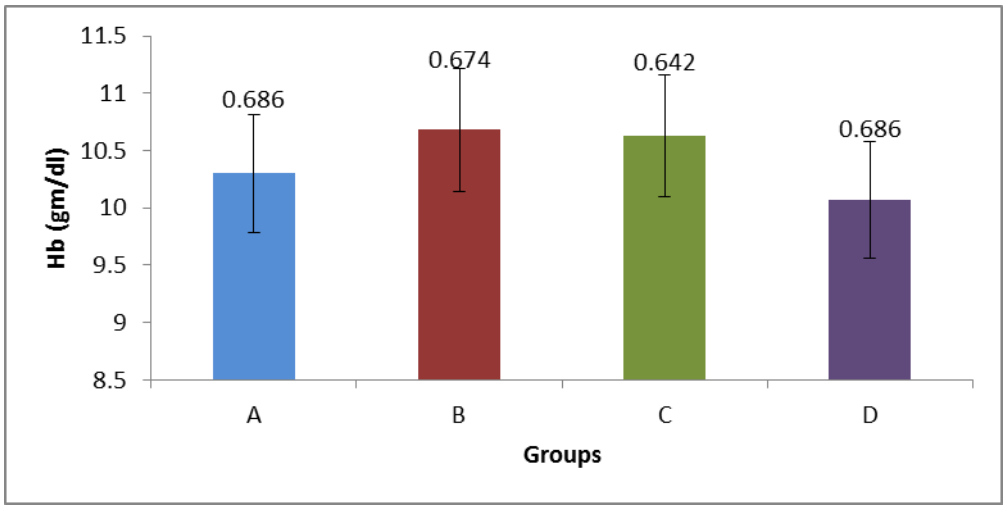


Fig. 8: Hb (gm/dl) during experimental period

The superscript number above bar indicates standard error (SE)

C. Packed Cell Volume (%)

Packed cell volume is presented in (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. These values show a little fluctuation they were not statistically significant ($p>0.05$).

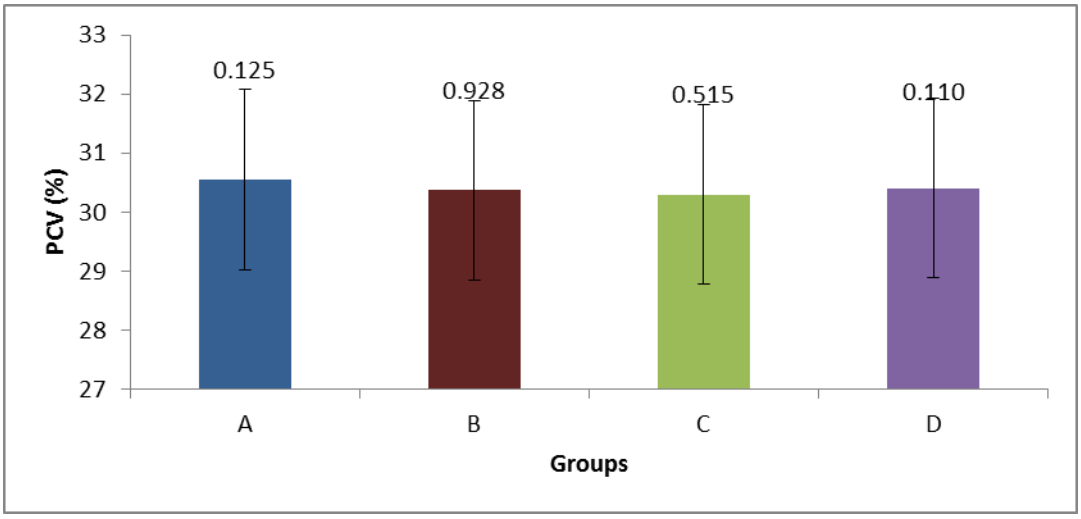


Fig. 9: PCV (%) during experimental period

The superscript number above bar indicates standard error (SE)

D. Erythrocyte Sedimentation Rate (mm/ in 1st hr):

Erythrocyte sedimentation rate content is presented in (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 Group D and lowest in Group A. Although these values show a little fluctuation they were not statistically significant ($p>0.05$).

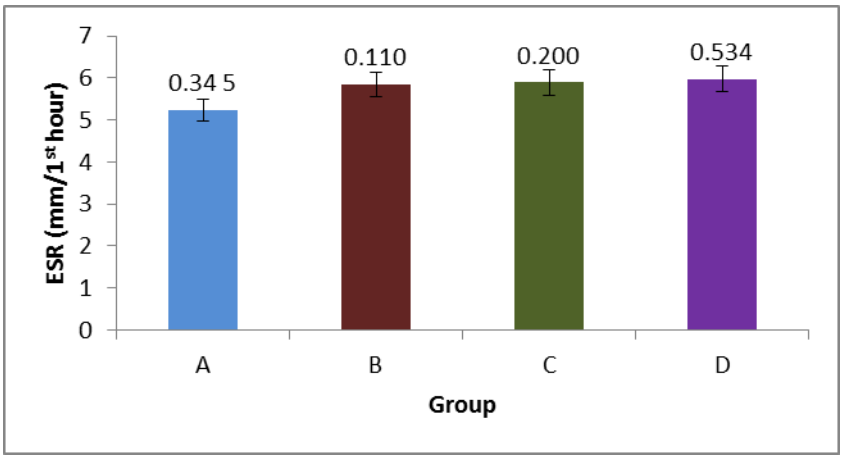
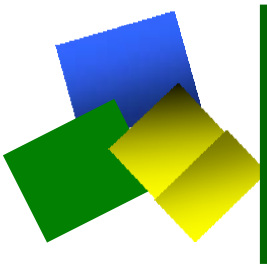


Fig. 10: ESR (mm/1st hour) during experimental period

The superscript number above bar indicates standard error (SE)



CHAPTER V

CONCLUSION

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CONCLUSION

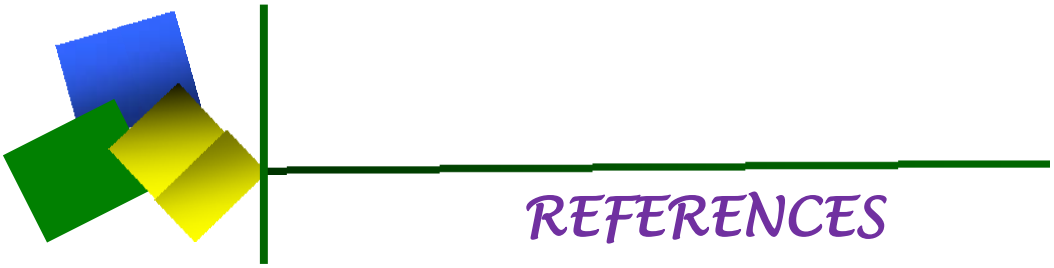
This research work was conducted to study the effects of Ipil Ipil and bean leaves extract supplementation on feed conversion, growth performance and egg production of chicken. A total of 28 "Hisex Brown layer" of 75 weeks old were collected from local market and reared throughout the entire period and it were usually 6 weeks. All the 28 chicken's randomly divided into 4 groups (n=7) to carry out this research work. Keeping one group as normal control group (A) and other three groups (B, C and D) as group subjected to treatment with Ipil Ipil and Bean leaves extract. The group of B supplemented with formulation of 2gm grinded Ipil Ipil leaves, C was supplemented with 2gm grinded Bean leaves and D was supplemented with 1gm grinded Ipil Ipil leaves plus 1 gm grinded Bean leaves per kg feed respectively. Observations were recorded for body weight, weight gain, feed consumption, feed conversion, growth performance and egg production of chicken during experimental period. The treatment group B,C,D recorded statistically significant ($p<0.01$) increase for egg production and decrease pause days than that of control group A. Net egg production was increased in Ipil Ipil and bean leaves extract supplemented group (28 ± 1.73*, 37 ± 2.89*, 36 ± 2.31*, 38 ± 2.88*) than control group is 28 ± 1.73 and net pause days was decreased than control group A. Body weights were increased significantly ($p<0.05$) in all treated groups in respect to the control and highest was recorded in combine Ipil Ipil and bean leaves supplemented groups (Group D). The average income in group B, C and D was 3941 TK, 3909 TK, 4056 TK respectively control group was 3590 TK. It is concluded that supplementation of 1gm grinded Ipil Ipil leaves plus 1gm grinded Bean leaves per kg feed (D) of treatment groups caused significant increase in egg production ($p<0.01$) and decrease in pause days as compared to that of other groups of hens. From this experiment we found that, between the control group and the treatment group of birds, the (D) group is more profitable than any other groups. No significant ($p>0.05$) differences were observed among the groups for PCV and TEC values. Hb content increased and ESR decreased significantly ($p<0.05$) with combine Ipil Ipil and bean supplemented group (Group D). That there exist a significant ($P<0.05$) difference among the mean values like Weight of the egg (gm), Diameter of the albumin (mm), Height of the yolk (mm), Fresh albumin weight (gm) corresponding to the different treatment. But no significant ($p>0.05$) difference among the mean values like Width of the egg (mm), Length of the egg (mm, Height of the thick albumin (mm), Width of the yolk (mm), Shell thickness (mm), Fresh yolk wt (gm), Shell dry weight

(gm) corresponding to the different treatment. From the present field and laboratory trial, it can be concluded that combined supplementation of 1 gm grinded Ipil Ipil leaves plus 1gm grinded Bean leaves per kg feed is highly beneficial for enhancing egg production without making any potential hazards of Hisex Brown layer and our formulations could be used as an egg enhancer and growth promoter for layer especially for older layer.

This is a preliminary work and the feed preparation technique is very simple. Farmers could adopt this technology without any specialized technical knowledge and medicinal ingredients. As a result by using grinded Ipil Ipil and Bean leaves with normal commercial ration small scale layer farmers would be able to sustain in their farming business and fulfill our protein demand. As well as a positive contribution in our national GDP of Bangladesh, these can helps in alleviating poverty through creating employment opportunity especially for rural population.

Recommendations

- Further research on supplementing Ipil ipil and bean leaves at varying levels should be considered to assess its effect on production performance.
- There is scope for further study to confirm the results that Ipil ipil and bean leaves supplementation can be incorporated in the diet of layers for better growth performance and to increase eggs production.
- Proper feeding of high quality feed should be done. Studies on feeding/feeds quality versus management systems are required.
- Disease, parasite control and management of chickens kept under different management systems should be taken into consideration.
- Other environmental factors such as sanitation, stress, feeding and water management practices and bio security measures should be taken into consideration.



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