EFFECT OF SUPPLEMENTATION OF YEAST AS NUCLEOTIDE SOURCE ON THE PERFORMANCE OF JAPANESE QUAIL

Thesis

Submitted to the

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By

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B.V. Sc. & A. H.

IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE DEGREE OF

Master of Veterinary Science
(Poultry Science)

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ACKNOWLEDGEMENT

A, as its first of letters, every speech maintains; The “Primal Deity” is first through all the world’s domains.

Kural: 1.1

Behind every big achievement lies the help, guidance and support of many people. Acknowledgement is the simplest way to show our utmost gratitude towards them. So, here I take this great opportunity to give my sincere thanks to each and every person who helped me in one or the other way, from the beginning of my research work to up to this stage of completion of my thesis.

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CERTIFICATE

This is to certify that the thesis entitled “Effect of supplementation of yeast as nucleotide source on the performance of Japanese quail” submitted in partial fulfilment of the requirements for the degree of Master of Veterinary Science with major in Poultry Science of the College of Post-Graduate Studies, G.B. Pant University of Agriculture & Technology, Pantnagar, is a record of bona-fide research carried out by Mr. S. Prakash, Id. No. 42782 under my supervision and no part of the thesis has been submitted for any other degree or diploma.

The assistance and help received during the course of this investigation have been acknowledged.

Pantnagar  
June, 2013

(Jyoti Palod)  
Chairperson  
Advisory Committee
CERTIFICATE

We, the undersigned, members of the Advisory Committee of Mr. S. Prakash, Id. No. 42782 a candidate for the degree of Master of Veterinary Science with major in Poultry Science, agree that the thesis entitled “Effect of supplementation of yeast as nucleotide source on the performance of Japanese quail” may be submitted in partial fulfilment of the requirements for the degree.

(Jyoti Palod)
Chairperson
Advisory Committee

(R.K. Sharma)
Member

(Balwinder Singh)
Member

(Shive Kumar)
Member

(S. K. Singh)
Member
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ABBREVIATIONS USED

@ : At the rate of
%
% : Per cent
< : Less than
> : Greater than
µl : Microliter
/ : per
A.O.A.C. : Association of Official Analytical Chemists
ad lib : Ad libitum or free choice
AMP : Adenosine mono phosphate
B.P. : Boiling Point
ºC : Degree centigrade
CMP : Cytidine mono phosphate
CP : Crude protein
CRD : Completely randomized design
dl : Deciliter
DM : Dry matter
DNCB : Dinitro-chloro benzene
DTH : Delayed type hypersensitivity
EE : Ether extract
ºE : Degree east
FCR : Feed Conversion Ratio
g : Gram
GMP : Guanosine mono Phosphate
Hb : Haemoglobin
HDL : High Density Lipoprotein
Hrs : Hour
i.e. : id est, that is
Ig A : Immunoglobulin A
Ig G : Immunoglobulin G
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<tr>
<td>IMP</td>
<td>Inosine mono phosphate</td>
</tr>
<tr>
<td>IU</td>
<td>International Unit</td>
</tr>
<tr>
<td>Kg</td>
<td>Kilogram</td>
</tr>
<tr>
<td>LDL</td>
<td>Low Density Lipoprotein</td>
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<tr>
<td>m</td>
<td>Meter</td>
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<tr>
<td>M</td>
<td>Molar</td>
</tr>
<tr>
<td>MCH</td>
<td>Mean Corpuscular Haemoglobin</td>
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<tr>
<td>MCHC</td>
<td>Mean Corpuscular Haemoglobin Concentration</td>
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<tr>
<td>MCV</td>
<td>Mean Corpuscular Volume</td>
</tr>
<tr>
<td>mg</td>
<td>Milligram</td>
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<tr>
<td>Min</td>
<td>Minute</td>
</tr>
<tr>
<td>ml</td>
<td>Milliliter</td>
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<tr>
<td>mm</td>
<td>Millimeter</td>
</tr>
<tr>
<td>N</td>
<td>Normal</td>
</tr>
<tr>
<td>NK</td>
<td>Natural killer cell</td>
</tr>
<tr>
<td>Nm</td>
<td>Nano meter</td>
</tr>
<tr>
<td>OD</td>
<td>Optical Density</td>
</tr>
<tr>
<td>PCV</td>
<td>Packed cell volume</td>
</tr>
<tr>
<td>ppb</td>
<td>Parts per billion</td>
</tr>
<tr>
<td>RBC</td>
<td>Red blood cells</td>
</tr>
<tr>
<td>rpm</td>
<td>Revolutions per minute</td>
</tr>
<tr>
<td>SE</td>
<td>Standard error</td>
</tr>
<tr>
<td>SGOT</td>
<td>Serum Glutamate Oxaloacetate Transaminase</td>
</tr>
<tr>
<td>SGPT</td>
<td>Serum Glutamate Pyruvate Transaminase</td>
</tr>
<tr>
<td>EC</td>
<td>Total erythrocyte count</td>
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<td>TLC</td>
<td>Total leukocyte count</td>
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<td>TSP</td>
<td>Total serum protein</td>
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<td>UMP</td>
<td>Uridine mono phosphate</td>
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<tr>
<td>Viz</td>
<td>Videlicet</td>
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<tr>
<td>WBC</td>
<td>White blood cells</td>
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<td>Wt</td>
<td>Weight</td>
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<td>V/V</td>
<td>Volume by volume</td>
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Introduction
India is an agriculture based country as about 70 per cent population is dependent on farming and allied activities. Among the different sub-sectors of agriculture, livestock sector is gaining importance and within the livestock sector poultry farming occupies a premium position in India. Poultry products such as egg and meat are preferred over other kinds of animal products all over the world. There is no discouragement for the poultry products in any of the religions which is the reason for rapid growth of the poultry sector. The recent estimates indicate that poultry contributes around 25 per cent of world’s meat supply. In India, there is a great scope for poultry farming because of the high internal demand for poultry products. Poultry, which was considered as a backyard venture in the early 60’s has now been transformed into an organized, scientific and vibrant venture. Presently poultry sector contributes nearly 0.7 per cent of the national GDP and about 10 per cent of the livestock GDP. The growth of poultry sector has been over 5.58 per cent in egg and 10 per cent in poultry meat production which is very high as compared to 1.5 to 2 per cent growth in agriculture.

Now a days, poultry has made tremendous strides particularly in the private sector, with the result that India is now self-sufficient with regard to requirements of high quality breeding stock, modern poultry equipments, availability of medicines, vaccines and technically qualified skilled manpower. This industry has recorded magnificent growth with good prospects for a successful future. The present chicken population of our country is 866 millions and contributing 4.5 per cent of total world’s population (FAOSTAT, 2012). With an annual production of 63.02 billion eggs and 2.6 million tonnes of poultry meat, India is now the world’s third largest egg producer and fourth largest producer of broiler meat (Watt Executive Guide, 2012). The egg and broiler production is contributing Rs.15123 million and Rs. 30293 million to the gross national product (GNP) respectively and employing around 5 million people directly and indirectly. Per capita consumption of meat and eggs are 2.15 kg and 53 eggs respectively, which are much less than the ICMR
recommendation of 180 eggs and 11 kg of poultry meat (Basic Animal Husbandry Statistics, 2012). Therefore, there is wide scope of poultry production in India.

In last few decades, different species of poultry viz. Japanese quail, guinea fowl, turkey etc. have gained popularity for meat consumption among consumers. Among these Japanese quail by virtue of their short generation interval, lesser feed and space requirement and their physiological similarity with large avian species are being widely used as a model poultry bird for breeding experiments (Anthony et al., 1996). In the past few decades, meat and eggs of Japanese quail have become more popular in European and Middle East countries because of their small size, taste and low fat content.

Japanese quails were first domesticated in Japan for meat and egg purpose in 1595. After the World War II, domestication of Japanese quail took place globally. In India, Japanese quails were first imported by CARI from California in the year 1974 for diversification of poultry farming from chicken and duck. Mainly two species of quails are found in India; the black breasted quail (Coturnix coromendelica) found in jungle and the brown coloured Japanese quail (Coturnix coturnix japonica) which is being used for the commercial quail production. There are about 45 varieties of quails among which Japanese quail is the largest species which weigh up to 200 g and lays around 280 eggs, whereas Indian quail weighs only 100 g and lays about 100 eggs a year.

Poultry farmers face heavy economic problems due to the increasing susceptibility of birds to various diseases and stressors (associated with high productivity) along with the ban of antibiotics and restricted use of medication. Under these circumstances, effective control of diseases is most important as per the proverb ‘Prevention is better than cure’ for better economic results. Vaccination becomes unavoidable in the poultry farming because of the increased incidence of immunosuppressive diseases. However, bird’s immune system should be enhanced in order to increase natural resistance to diseases and to improve efficacy of vaccines. Genetics, environment and stressors usually cannot be influenced by farmers. Therefore, the feed must be regarded as a target to optimize both production and immune performance of the birds. Balanced feeds with essential and semi essential nutrients during different stages of growth or production are a prerequisite of economical performance.
Feed is an important and critical input for the poultry industry as it accounts for 60 to 70 per cent of production cost. Various feed additives or growth promoters have been developed to improve growth rate, feed efficiency, product quality, reduce mortality and to reduce the production cost. These feed additives are mostly non-nutrient feed additives (Singh and Panda, 1992). In order to prevent performance losses in poultry production, nucleotides are emerging as one of the potential feed additives.

Nucleotides are low molecular weight biological compounds, act as the building blocks of the cell and are required to make new RNA and DNA. They consist of a nitrogen base (either purine or a pyrimidine) linked to a pentose sugar with at least one phosphate group attached. Nucleic acids are the polymer of nucleotides and when the sugar is ribose, the result is ribonucleic acid (RNA) or, if deoxyribose, deoxyribonucleic acid (DNA). They are fundamental to the physiological and biochemical functions of the body including encoding and deciphering genetic information, mediating energy metabolism and cell signalling as well as serving as components of coenzymes, allosteric effectors and cellular agonists (Carver and Walker, 1995; Cosgrove, 1998). For many years nucleotides have been considered as non essential nutrients as neither biochemical malfunctions nor classical deficiency signs develop in human or animal models. However, this opinion was later changed due to research studies which suggest that dietary nucleotide deficiency may impair liver, heart, intestine and immune functions (Grimble and Westwood, 2000). Dietary nucleotides are of main significance in the growth and development of tissues with rapid turnover like intestinal mucosa, red blood cells, white blood cells, bone marrow cells and some brain cells because of their inability to produce sufficient amount of nucleotides, hence nucleotides are described as ‘conditionally essential’.

The physiological demand of nucleotides for all organisms is met by two biosynthetic pathways i.e. de novo synthesis and salvage pathway. The nucleotide synthesis from small precursors is by means of de novo pathway, whereas the salvage pathway uses nucleosides and bases resulting from the breakdown of nucleotides to re-synthesize nucleic acids. In order to maintain a balance, each organism must produce millions of new cells every second. Although biosynthetic pathways produce nucleotides internally, they are inefficient to supply huge demand of additional
nucleotides required for cell proliferation during the times of extraordinary stress (such as growth, reproduction, environmental change or challenge, combat disease and recovery from injury). As a result the affected or compromised birds reduce their performance or slow down development. The increased demand in combination with relatively slow supply of nucleotides by the bird itself may result in the need for extra nucleotides added directly to the poultry diet. Therefore, external supply of both purine and pyrimidine nucleotides can save the high energetic cost of their synthesis by internal mechanisms.

Nucleotides are present in most ingredients of plant and animal origin. Through intestinal absorption they become available to the animal from the diet. Nucleotides reach the blood stream after the enzymatic metabolism in the intestine. Compared to the de novo synthesis, reassembly of the nucleotides requires less energy in the intestinal cells. Therefore, external supply of the nucleotides would be more beneficial. The feed ingredients such as fish meal, legumes, yeast extracts and unicellular organisms like yeasts and bacteria are rich in RNA or DNA. However, among the ingredients, there is a huge difference in the nucleotide content, proportion and availability. Digestibility of the ingredients is the main factor which affects the availability of nucleotides. It is possible to increase the nutritional availability over 95 per cent by means of purifying nucleotides and make them easily absorbed through intestinal wall.

Research in human as well as certain animal and aquatic species revealed that dietary supplementation of nucleotides shows various beneficial effects like increased resistance to bacterial and viral infections, acceleration of antibody production, increase in number of neutrophils and macrophages, reversal of malnutrition and starvation-induced immunosuppression, increase in plasma HDL cholesterol, decrease in plasma LDL cholesterol, faster recovery of the liver after injury, positive effects on the intestines, intestinal repair after diarrhoea and stress relief (Wissman, 2006).

In contrast, limited information is available in the literature about the need for nucleotides and their role on the production performance, development of the immune system and the intestinal tissue in poultry. For Japanese quails such information is lacking. Therefore, the present investigation was undertaken to study
the effects of dietary nucleotide supplementation in Japanese quails with the following objectives:

1. To study the effect of dietary nucleotide supplementation on growth performance in Japanese quails
2. To study the effect of dietary nucleotide supplementation on nutrient utilization in Japanese quails
3. To study the effect of dietary nucleotide supplementation on carcass traits and meat composition in Japanese quails
4. To study the effect of dietary nucleotide supplementation on hemato-biochemical parameters in Japanese quails
5. To study the effect of dietary nucleotide supplementation on immune status in Japanese quails
6. To study the effect of dietary nucleotide supplementation on intestinal morphology in Japanese quails
Review of Literature
In the last few decades there has been considerable progress in poultry industry due to improved genetics, nutrition, metabolism and disease control. The ban of antibiotics and restricted use of medication have aggravated the economic problems on farms along with the increasing susceptibility of birds to various diseases and stressors. Effective control of diseases and improvement in the performance of birds is possible only by enhancing the bird’s immune system. Along with environmental factors, external stressors and the genetic makeup of the birds, various nutrients also play an important role in triggering performance. Among these, balanced diet is the easiest way to optimize performance of birds. Nutritionists and feed producers know very well about the feasibility to assemble well balanced diets including all essential as well as minor nutrients. Nevertheless there is always the possibility of lacking certain nutrients in a given diet. Mostly these missing nutrients are not classified as being essential for performance. However, under specific conditions some of these non-classified nutrients may switch in their importance quite drastically. Therefore these nutrients are called conditionally essential nutrients or semi essential nutrients. Now days, nucleotide supplements are used as feed additives because of its significant role in improvement of performance.

Nucleotides are the monomers of the nucleic acids (DNA or RNA) i.e. group of nucleotides linked together to form nucleic acids. Nucleotides are normal components of the diet and the body provides mechanisms for their absorption and incorporation into tissues (Sanchez and Gil, 2002). Nucleotides are either themselves or in combination with molecules are involved in almost all activities of the cell and the body like creating cells, replacing cells, developing immune cells, sperm cells and supporting female reproductive tract.

Nucleotides are ubiquitous molecules with considerable structural diversity. They are composed of a nitrogenous base linked to a pentose sugar to which at least one phosphate group is attached. The pentose sugar may be a ribose for a ribonucleic acid (RNA) or a deoxyribonucleic acid (DNA). The nitrogenous base can be a purine or a pyrimidine. Pyrimidine base are composed of six membered rings and comprise uridine, cytosine and thymine. Purine bases have an additional five membered ring.
and comprise adenine, guanine, and hypoxanthine. The phosphate group may be in a mono, di or tri phosphate form, and is commonly esterified to the C-5’ hydroxyl group of the pentose sugar.

When the phosphate group is absent, the compound is known as a nucleoside. A chain of nucleotides attached together via a phosphodiester linkage at the 3’ and 5’ position of neighbouring ribose units are called polynucleotides or nucleic acids. Nucleic acids conjugated to protein are called nucleoproteins. The concentration of ribonucleotides is relatively constant in all cells, while the concentration of deoxyribonucleotides varies with the stage of the cell cycle (Barness, 1994).

**Sources of nucleotides**

Protein rich diets are good sources of nucleotides especially Inosine mono phosphate (Carver and Walker, 1995). Generally feed ingredients containing cellular elements are potential dietary source of nucleotides in the form of nucleoproteins. Organ meats, poultry and sea food are good source of nucleoprotein (Barness, 1994). Single cell proteins, bakers and brewer’s yeast, and yeast extract are ingredients that have a relatively high concentration of nucleotides (Maloney, 1998 and Tibbets, 2002). Most commonly used feed ingredients contain relatively low amounts of nucleotides. The nucleotide content of some feed ingredients is given in Table 2.1.

**Table 2.1: Nucleotide concentrations in some feed ingredients on as is basis** (Mateo et al., 2004).

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>5’CMP</th>
<th>5’AMP</th>
<th>5’GMP</th>
<th>5’UMP</th>
<th>5’IMP</th>
</tr>
</thead>
<tbody>
<tr>
<td>Barley</td>
<td>0.002</td>
<td>0.001</td>
<td>0.001</td>
<td>0.000</td>
<td>0.001</td>
</tr>
<tr>
<td>Casein</td>
<td>0.001</td>
<td>0.000</td>
<td>0.000</td>
<td>0.000</td>
<td>0.000</td>
</tr>
<tr>
<td>Corn</td>
<td>0.003</td>
<td>0.002</td>
<td>0.003</td>
<td>0.000</td>
<td>0.001</td>
</tr>
<tr>
<td>Fish meal</td>
<td>0.026</td>
<td>0.011</td>
<td>0.002</td>
<td>0.001</td>
<td>0.035</td>
</tr>
<tr>
<td>Naked oats</td>
<td>0.003</td>
<td>0.003</td>
<td>0.003</td>
<td>0.001</td>
<td>0.001</td>
</tr>
<tr>
<td>Plasma</td>
<td>0.002</td>
<td>0.002</td>
<td>0.002</td>
<td>0.000</td>
<td>0.001</td>
</tr>
<tr>
<td>Whey, dried</td>
<td>0.270</td>
<td>0.019</td>
<td>0.000</td>
<td>0.001</td>
<td>0.004</td>
</tr>
<tr>
<td>Protein plasma, spray dried</td>
<td>0.016</td>
<td>0.008</td>
<td>0.003</td>
<td>0.009</td>
<td>0.002</td>
</tr>
<tr>
<td>Red blood cells, spray dried</td>
<td>0.000</td>
<td>0.044</td>
<td>0.003</td>
<td>0.002</td>
<td>0.006</td>
</tr>
<tr>
<td>Soya bean meal, 44%</td>
<td>0.016</td>
<td>0.008</td>
<td>0.003</td>
<td>0.009</td>
<td>0.002</td>
</tr>
</tbody>
</table>
Digestion, absorption and metabolism of nucleotides

Enzymatic hydrolization of dietary nucleoprotein, nucleic acids, and nucleotides are needed prior to absorption because only nucleosides, bases and small amount of nucleotides are absorbed. Endonucleases, phosphodiesterases and nucleoside phosphorylases of small intestine are the major enzymes involved in this process. These enzymes originate from the brush border epithelium (Morley et al., 1987), pancreatic juice (Weickman et al., 1981), and bile (Holdsworth and Coleman, 1975).

The duodenum has the greatest absorptive capacity (Bronk and Hastewell, 1987). Under physiological conditions, nucleotides have a limited capacity to pass through the microvillous membrane of the enterocytes (Sanderson and Youping, 1994). This may be due to the absence of a nucleotide transport system. Nucleotides also have a high negatively charged phosphate group that hinders absorption. Therefore, the nucleoside form is the major vehicle for the entry of purines and pyrimidines into the enterocytes. Nucleosides transport into the enterocytes occurs by facilitated diffusion and by specific Na+ dependent carrier mediated mechanisms (Bronk and Hastewell, 1987). This is a relatively efficient process and more than 90% of dietary nucleosides and bases are absorbed into the enterocyte (Salati et al., 1984). From the enterocyte, partial metabolic products of dietary and endogenous nucleotides and nucleosides enter the hepatic portal vein. These molecules are carried to the hepatocytes for further metabolism. From the liver, partial metabolic products of dietary and endogenous nucleotides and nucleosides are released into the circulation and enter the muscle tissues. If the products are not reutilized for nucleotides production or not absorbed in a specific tissue, the purine and pyrimidine bases are catabolized into uric acid and B-alanine or B – aminoisobutyrate (Carver and Walker, 1995; Thorell et al., 1996). In avians and primates, uric acid is excreted in the urine, but in mammals other than primates, uric acid is further catabolized into allantoin via the enzyme uricase. Allantoin is then excreted via the urine. The products of pyrimidine catabolism are β-alanine and β- aminoisobutyrate. They are further metabolized into NH3, CO2 and Acetyl CoA.
Synthesis of nucleotides

Nucleotides can be synthesized in the cytosol of hepatocytes by de novo synthesis provided the required precursors are available. The purine IMP is synthesized from α-D-ribose-5-phosphate via a process involving 11 reactions. Glutamine is the N-donor in the synthesis of IMP. Both AMP and GMP are subsequently formed from IMP via adenylosuccinate and xanthosine monophosphate, respectively (Rodwell, 2000). The precursor for pyrimidine synthesis are carbamoyl phosphate and aspartate. The pyrimidine UMP is formed in a process involving 6 reactions. A dephosphorylation of UMP yields UDP which is subsequently turned into CMP and TMP, respectively (Rodwell, 2000). The denovo synthesis of both purine and pyrimidine nucleotides is a metabolically costly process requiring a significant amount of energy in the form of ATP. In addition, both reactions require glutamine.

Salvage pathway, synthesize the nucleotide from the nucleoside and a phosphate group. The nucleosides used in the salvage pathway may originate from dietary source because most dietary nucleotides are changed to nucleosides prior to phosphoribosylation of purines and pyrimidines formed during the catabolism of nucleotides. This pathway may spare energy and allow cells that are incapable of de novo synthesis (i.e. leukocytes, erythrocytes, bone marrow cells, intestinal mucosal cells and lymphocytes) to maintain their nucleotide pools (Sanderson and Youping, 1994). As all cells of body require large numbers of nucleotides and de novo and salvage both mechanisms may not always supply enough nucleotides. When birds are unable to produce adequate nucleotides, they must get them from feeds. Though some of the feeds are rich in nucleotides but most of them have quite low nucleotides as compared to their need. Relatively high concentrations of nucleotides are found in intestines, bacterial and yeast cultures, none of which are usually consumed. Therefore, looking to the need of nucleotides in body for various functions exogenous source of nucleotides such as dietary supplement could optimize various body functions.

Roles of nucleotides

Nucleotides are the building blocks of nucleic acids (DNA and RNA). However, they also have physiological roles in the body such as source of energy
(i.e. ATP and GTP), cofactors in oxidation and reduction (i.e. FAD, NAD\(^+\), and NADP\(^+\)), physiological regulator (i.e. cAMP and cGMP), carry activated intermediate (i.e. UDP-glucose, CMP-sialic acid and CDP-choline) and acyl group (i.e. CoA). In addition, nucleotides have been shown to influence the development of the immune system, enhance stress and disease resistance, performance and reproductive efficiency, increases the microflora of the intestinal tract and better development of intestine. They also reduce the mycotoxin problems.

The literature pertaining to the effects of dietary nucleotides have been reviewed as under.

(A) Effect on growth performance

Zhang et al. (2005) conducted an experiment to investigate the effects of *Saccharomyces cerevisiae* (SC) cell components on the growth performance, meat quality, and ileal mucosa development by feeding Whole yeast (WY), SC extract (YE), and SC cell wall (CW) added at 0.5, 0.3 and 0.3% levels respectively to the broilers. From the results they concluded that dietary yeast components i.e. WY or CW supplementation improved growth performance. Both YE and CW had oxidation-reducing effects and CW improved ileal villus development.

Teles et al. (2006) conducted a 10 week experiment on sea bream juveniles to evaluate the effect of nucleic acids on performance of growth and ureagenesis. The fishes were separated into 5 groups of 3 replicates with 50 juveniles each. The control group was provided with diet having fish meal as protein source. In the treatment groups, part of the fish meal was replaced with two levels of nucleic acids and brewer’s yeast. From the results, they found that the inclusion of nucleotide (RNA/brewer’s yeast) had significant effect on the growth and feed intake. The plasma urea level was higher in groups supplemented with RNA that confirms that inclusion of nucleotide has significant effect on ammonia and urea excretion.

Garcia et al. (2007) conducted an experiment to determine the efficacy of a nucleotide preparation in broiler chickens by feeding nucleotide preparation @ 0, 500, 1000 mg/ kg feed. They found that the performance of the birds fed with 500 mg nucleotide/ kg feed on 21\(^{st}\) day showed significantly improved body weight by 1 % and FCR by 1.9 %, while at 1000 mg/ kg level there was no significant effect. There
were no significant effects between 21 and 42 days but body weight and FCR were numerically better in the treatments containing nucleotides.

Puig et al. (2007) performed an experiment to study the impact of nucleotide supplements on weaned piglets under correct health conditions. The productive performance of the piglets fed with nucleotide supplements from weaning (21 days) to 56 days were compared with piglets fed with standard commercial diets on days 21, 28, 35 and 56. During the Prestarter stage (21 to 35 days) the piglets supplemented with nucleotides revealed greater growth than the control group. During the starter stage (35 to 56 days), improvements were observed in both body weight and feed conversion ratio in the group supplemented with nucleotides.

Chumpawadee et al. (2009) conducted a study to investigate the effect of dietary inclusion of cassava yeast (*Saccharomyces cerevisiae*) on growth performance and carcass percentage in Japanese quails. The results showed that feed intake, feed conversion ratio, average daily gain and carcass percentage were not significantly different among treatments supplemented with *Saccharomyces cerevisiae* @ 0, 10^6, 10^7 and 10^8 organisms/kg feed.

Shareef and Dabbagh (2009) performed an experiment of 21 days to study the effect of yeast supplementation on the performance of broilers. The birds were separated into five treatments (T1 to T5) and fed with diets having 0, 0.5, 1.0, 1.5, 2.0 per cent baker yeast respectively. The results of the experiment indicated that the body weight gain, feed intake and feed conversion were significantly higher in the treatments T3, T4 and T5.

Ezema and Eze (2010) performed an experiment to study the effect of probiotic (*Saccharomyces cerevisiae*) supplementation on the growth performance and haematological parameters of rabbits supplemented with bioactive yeast @ 0.08, 0.12, 0.16 and 0 g yeast/kg diet. The results of the experiment showed that yeast supplementation at the level of 0.12 g/kg of diet had significant effect on the health status and growth rate of rabbits.

Singhal et al. (2010) conducted a study to reveal the effects of nucleotide supplementation on weight gain and head growth in formula-fed infants. The results of the study revealed that the infants fed with nucleotide-supplemented formula had
greater occipitofrontal head circumference at ages 8, 16, and 20 weeks than infants fed control formula even after adjustment for potential confounding factors. Weight at 8 weeks and the increase in both occipitofrontal head circumference and weight from birth to 8 weeks were also greater in infants fed nucleotide-supplemented formula than in those fed control formula.

Król (2011) conducted a study to determine effect of calf milk replacer feed additives i.e. mannanooligosaccharides, inulin and yeast nucleotides on rumen microflora, level of serum immunoglobulin and calf health condition. The results of the study indicated that the supplemented milk replacers had better effect on health condition, final body weight, daily body weight gain, concentrate intake and feed conversion ratio of calves. Feed additives, especially yeast nucleotides had beneficial influence on faecal scores (less watery and well formed). Additives applied in calf milk replacers did not clearly affect morphological, blood and most of the serum biochemical parameters. However, they increased blood glucose level and decreased cholesterol and urea levels. The higher level of gamma-globulin as well as better passive immunity transfer were shown in calves receiving yeast nucleotides in amount 4 g/day/head in milk replacer. Concentration of total volatile fatty acids in rumen, especially acetate and propionate were also higher in calves receiving milk replacer prebiotic feed additives, especially mannanooligosaccharides and yeast nucleotide that was confirmed by higher body weight gains.

Hosseini (2011a) conducted a trial by feeding the yeast extract to Ross 308 chicks @ 0.05, 0.1, 0.15, 0.2 per cent levels from 1 to 49 days. The results showed that yeast (Saccharomyces cerevisiae) extract had significant effect on growth performance and carcass traits at a level of 0.2 per cent.

Bruno et al. (2012) conducted an experiment on broilers by supplementing the feed with six different levels of nucleotide (0; 100; 200; 300; 400 and 500 g/ton of ration) and evaluated their performance during the initial and growth phase. From the results of the experiment they observed that the nucleotide supplementation in the feed had directly proportional effect on the performance i.e., feed conversion ratio and body weight of broilers during the initial and growth phase.

Hassan et al. (2012) performed an experiment to evaluate the effect of yeast (Saccharomyces cerevisiae) supplementation on growth performance and colonization...
of Salmonella enteriditis in intestinal tract of 240 unsexed Japanese quails under four treatments. T₁ and T₂ served as non infected and infected controls, while T₃ and T₄ served as non infected and infected groups with yeast supplementation of 3 g / kg basal diet. The results of the study showed that the birds of the non infected group with yeast supplementation revealed better growth performance with increased erythrogram and leukogram compared to the control. In infected group birds, yeast supplementation reduced the caecal colonization and restored the drastic reduction of Hb and PCV.

Bael and Roxas (2013) conducted an experiment to study the effect of yeast based nutritional enhancers (YNE) i.e. mannoproteins, betaglucans and nucleotides on growth performance of weaning piglets. A total of eighty piglets were randomly allocated in to four treatments with four replicates each. The treatment groups were supplied with feeds as follows: basal diet (BD), BD + 0.05% YNE, BD + 0.15% YNE and BD + 0.30% YNE. From the results they found that addition of YNE @ 0.30% significantly increased the feed intake and average daily weight gain. However, feed efficiency ratio was not significantly affected by dietary YNE supplementation.

(B) Effect on haemato-biochemical parameters

Choudhury et al. (2005) observed the effects of dietary ribonucleic acid or chitin supplementation on haematological parameters and resistance to Aeromonas hydrophila by conducting a feeding trial of 60 days in Labeo rohita juveniles under 6 groups. The groups were fed with diets containing different concentrations of either ribonucleic acid or chitin except the control group, viz., control, T₁ (0.1% ribonucleic acid), T₂ (0.2% ribonucleic acid), T₃ (0.4% ribonucleic acid), T₄ (25mg chitin/kg) and T₅ (50 mg chitin/kg). Based on the observations they found that the T₃ group was significantly different from the others with higher total leukocyte count, total protein, globulin and lower A/G ratio and better survival percentage after challenging with Aeromonas hydrophila. However, there was no significant difference in the growth performance, haemoglobin content and total erythrocyte count between the control and treatment groups.

Paryad and Mahmoudi (2008) performed an experiment to study the effect of feeding different levels (0%, 0.5, 1.5 and 2%) of yeast Saccharomyces cerevisiae (SC)
on growth performance, blood constituents and carcass characteristics of Ross broiler chicks. The results showed that chick’s ration containing 1.5 and 2% yeast reduced plasma cholesterol and triglycerides concentration and increased plasma HDL level. Chicks fed 1.5% *S. cerevisiae* had higher total plasma protein, albumin and globulin concentration and also had higher body weight gain (BWG), feed intake (FI) and better feed conversion ratio (FCR).

Shareef and Dabbagh (2009) performed an experiment of 21 days to study the effect of yeast supplementation on the performance of broilers. The birds were separated into five treatments (T₁ to T₅) and fed with diets having 0, 0.5, 1.0, 1.5, 2.0 per cent baker yeast respectively. The results of the experiment indicated that the total serum protein and glucose were higher in yeast supplemented groups. The serum triglycerides were significantly reduced in the treatments supplemented with yeast. However, no effect was observed in the serum calcium, uric acid, ALT and AST activities of the different treatments compared with the control.

Hosseini (2011c) conducted an experiment by feeding yeast extract to the Ross 308 broiler chicks @ 0.5, 0.1, 0.15, 0.2 per cent levels from 1 to 49 days and studied their effects on blood parameters. The results showed that 0.2% *Saccharomyces cerevisiae* supplementation had significant effect on haematological parameters viz. haemoglobin, total red blood cell, white blood cell count, mean corpuscular haemoglobin, mean corpuscular volume and mean corpuscular haemoglobin concentration.

Mansour *et al.* (2011) conducted a study to evaluate the effect of dietary supplementation of yeast culture @ 0, 1, 1.25 and 1.5 g of YC/kg diet on growth performance and hematological parameters of broilers for a period of 42 days. The results of this study indicated that the addition of yeast culture in broiler diet improved body weight gain and feed efficiency and decreased H:L ratio, especially at 1.25 g YC/kg diet level. However, no significant differences were found among treatment groups for haemoglobin, total red blood cells, haematocrit, mean corpuscular volume and mean corpuscular haemoglobin.

Wu *et al.* (2011) conducted a study to investigate the differences in serum biochemistry between breast-fed and formula-fed infants of 4 and 8 weeks age. The results of this study revealed that there were no significant differences between the
two groups in terms of growth or anthropometric measurements. Serum cholesterol, triglycerides (TG), alanine aminotransferase (ALT), aspartate aminotransferase (AST), gamma glutamyl transferase (GGT), total bilirubin and direct bilirubin levels were significantly higher in the breast-fed group compared with those measured in the formula-fed group at both 4 and 8 weeks of age. Serum blood urea nitrogen (BUN) and inorganic phosphate (IP) levels were significantly lower in the breast-fed group compared with the formula-fed group at 4 and 8 weeks of age. They suggested that the beneficial effects in breast fed infants may be due to the considerable amount of nucleotides in the breast milk.

Aluwong et al. (2012) conducted a study on broiler chickens to investigate the effect of supplemental probiotic yeast (*Saccharomyces cerevisiae*) preparation under four groups (Control-C, E₁ 0.5%, E₂ 1.5% and E₃ 2.0%) on performance indices and serum biochemistry. From the results they observed that the broilers of E₃ group had significantly higher body weight gain compared to control. However, the activity of AST in broilers of treatment group was insignificantly increased while ALT was significantly reduced compared to the control. The glucose and cholesterol concentration were significantly reduced in the E₂ and E₁ group respectively compared to control.

Recently, Abdelrahman (2013) conducted a feeding trial on 400 male broilers (Ross) to investigate the effect of using dry fat and 2 levels of yeast culture (*Saccharomyces cerevisiae*) on performance, blood glucose, cholesterol, calcium, phosphorus, cobalt, copper, magnesium, manganese and zinc levels. The results indicated that the addition of yeast culture (3 kg/t) to diets containing dry fat improved broiler growth performance and positively affected the carcass characteristics by reducing the abdominal fat and blood cholesterol levels. The serum cholesterol levels were significantly (P < 0.05) reduced by adding the yeast culture.

Mohebbi et al. (2013) conducted an eight week feeding experiment to study the changes on lipid contents of rainbow trout fingerlings by supplementing different levels of dietary nucleotides (0, 0.5, 1, 1.5 and 2 g/kg of feed). The results of experiment revealed reduced levels of serum triglycerides and low density lipoprotein (LDL) cholesterol while increased levels of serum high density lipoprotein (HDL)
cholesterol in trout received diets containing 1.5 and 2 g nucleotides/ kg compared to the control group.

(C) Effect on immunological status

Giancamillo et al. (2003) performed an experiment on piglets by feeding glutamine and nucleotides for a period of 28 days and studied their effects on growth performance and gut associated lymphoid tissues. They found that supplementation of glutamine and nucleotides have no significant effect on the growth performance of the piglets. However, increase in villus height, crypt depth, decrease in villus:crypt ratio and fast restoration of mucosal thinning that occurs at weaning were observed in groups supplemented with glutamine and nucleotides. The percentage of macrophages and intraepithelial lymphocytes were also higher in piglets of treatment groups compared to the control.

Choudhury et al. (2005) observed the effects of dietary ribonucleic acid or chitin on haematological parameters and resistance to Aeromonas hydrophila by conducting a feeding trial of 60 days in Labeo rohita juveniles under 6 groups. The groups were fed with diets containing different concentrations of either ribonucleic acid or chitin except the control group, viz., control, T_1 (0.1% ribonucleic acid), T_2 (0.2% ribonucleic acid), T_3 (0.4% ribonucleic acid), T_4 (25mg chitin/kg) and T_5 (50 mg chitin/kg). Based on the observations they found that the T_3 group was significantly different from the others with higher total leukocyte count, total protein, globulin and lower A/G ratio and better survival percentage after challenging with Aeromonas hydrophila. However, there was no significant difference in the growth performance, haemoglobin content and total erythrocyte count between the control and treatment groups.

Deng et al. (2005) conducted an experiment to study the carry–over effects of dietary yeast RNA as a source of nucleotides on the immune system in Leghorn–type chickens for a period of 12 weeks by supplementing 0, 5 (low RNA, LR) or 10 (high RNA, HR) g yeast RNA/kg of feed during the initial 4 weeks. From the results they concluded that the addition of yeast RNA as a source of nucleotides to a commercial diet had minimal effects on humoral and cell–mediated immune responses in growing
Leghorn–type chickens. Supplementation stimulated the development of spleen. However, this effect did not persist into the later stages of the chickens’ life.

Gheisari and Kholeghipour (2006) conducted an experiment to evaluate the effects of four levels (0, 0.1, 0.2 and 0.3%) of two forms (powdery and granular) of live yeast i.e. *Saccharomyces cerevisiae* supplementation over a period of 49 days on performance, humoral immune responses and blood parameters of broilers. The results of the experiment suggested that both granular and powdery forms of live yeast *S. cerevisiae* had not a growth stimulating effect in broiler chicks. However as compared to granular type, addition of 0.1 or 0.2% powdery form of live yeast *S.cerevisiae* to diet improved humoral immune response, decreased serum lipids and suppressed abdominal fat accumulation in broiler chickens.

Hawkes et al. (2006) conducted an experiment to study the growth and immune functions of infants provided with nucleotide based formula diet. The results of the experiment indicated that the growth of the infants at different ages was not significantly affected by nucleotide supplementation. However, infants supplemented with nucleotides @ 33.5 mg/l of formula food showed mild improvement in the immune response.

Puig et al. (2007) performed an experiment under field conditions to determine the impact of nucleotide supplement on productive performance of piglets weaned at 21 days of age. They were divided into 3 groups and fed with three experimental diets (commercial control diet, diet with 750 and 1000 ppm of nucleotides respectively) for a period of 2 weeks. Feed consumption, growth, incidence of diarrhoea and mortality were monitored on a weekly basis. There was a significant reduction in the incidence of diarrhoea in the animals supplemented with 750 ppm (3.13%) and 1,000 ppm of nucleotides (1.53%) compared to the control group (15.6%), as well as a numeric reduction in the mortality rate.

Hosseini (2011b) conducted an experiment to determine the effects of yeast (*Saccharomyces cerevisiae*) extract on visceral and immune organs of broilers. In this trial they fed the yeast extract to the Ross 308 chicks @ 0.05, 0.1, 0.15, 0.2 per cent levels from 1 to 49 days. The results showed that *Saccharomyces cerevisiae* had significant effect on weight, length and per cent of viscera (proventriculus, small
intestine, colon, cecum, lungs) and immune (spleen, gall bladder, bursa of fabricius) organs.

Andrino et al. (2012) conducted an experiment on pacific white shrimp (*Litopenaeus vannamei*) by feeding nucleotide formula @ 0%, 0.2%, 0.4% and 0.6% of feed and studied their effect on growth, immune response and survivability against white spot syndrome virus (WSSV). From the results they found that dietary supplementation of nucleotide @ 0.2% improved the growth, feed utilisation, protein utilisation and also accelerated immune response against WSSV infection.

Fathi et al. (2012) conducted a study for a period of 42 days to evaluate the effect of supplemental yeast culture (YC) @ 0, 1, 1.25, 1.5 g /kg diet on carcass characteristics and humoral immune response of broilers. The results revealed that the broilers fed yeast cultures had statistically increased body weight gain only at 5-6 weeks of age. Also the treatment broilers showed low mortality and high percentage of major and minor breast muscles to the control. A significant increase in IgM titer at 7 days post-injection with sheep red blood cells was noticed in broilers fed diet containing YC more than 1g/kg. Similarly, broilers fed a diet containing 1.25g/kg YC exhibited a higher level of antibody titer against Newcastle disease virus at the age of 42 days compared with the other groups.

Sauer et al. (2012) studied the effect of nucleotide supplementation on performance, humoral immunity and gut morphology of piglets weaned at 20 days of age. The experiment was conducted on 5 litters each of 7 piglets. One piglet/litter was slaughtered at zero day as baseline and the remaining 30 were divided into two groups (control and treatment group supplemented with nucleotide). From the observations of the study, they found that nucleotides don’t have significant effect on daily body weight gain, final body weight and gut morphology. However, the average daily feed intake and concentration of Ig A were significantly higher in group supplemented with nucleotides. Based on this, they concluded that dietary nucleotide in weaning piglets improved the humoral immunity.

Shankar et al. (2012) conducted a trial to evaluate the effect of nucleotides @ 0, 1.5, 2.25 and 3.0 g/kg diet on growth, survival, immunity and resistance to white muscle disease and *Aeromonas hydrophila* infection in freshwater prawn (*Macrobrachium rosenbergii*). At the end of the 60 days feeding trial, growth was
recorded and non-specific immune parameters, such as, prophenol oxidase activity, superoxide anion production, total haemocyte count and total serum protein were studied in haemolymph samples. They found that total haemocyte count, prophenol oxidase activity, superoxide anion production and the relative per cent survival of prawn after the challenge test against white muscle disease were significantly higher in prawns fed nucleotide-based diets. From the results, they concluded that the dietary level of 1.5 g nucleotide/kg improved the growth, survival and enhanced the immune response in *Macrobrachium rosenbergii*.

Superchi *et al.* (2012) investigated the effect of dietary nucleotide supplementation on growth and immune response during 9 – 55 days age (pre weaning 9 – 21 days, post weaning 22 – 55 days) piglets. A total of 108 piglets were divided into two groups provided with diets having 0% and 0.1% of yeast extract nucleotides. Significant increase in body weight was noticed in piglets at 21, 35 and 55 days of age. However, the feed intake was not different between groups. The lymphocyte subpopulation and peripheral mononuclear concentration at 21 and 35 days of age were higher in piglets of supplemented groups. From the results, they concluded that nucleotide supplementation enhanced growth performance and immune response even under the weaning stress.

**(D) Effect on carcass characteristics**

Pelícia *et al.* (2010) conducted a study on 600, Ross 308 male broilers for evaluating the effect of nucleotides on the performance and carcass yield of broilers fed diets with no antibiotic growth promoters (AGP), anticoccidials or animal feedstuffs. Under the conditions of this experiment, diets supplemented with nucleotides did not influence broiler performance or carcass yield at 42 days of age.

Chiofalo *et al.* (2011) conducted a study to evaluate the effects of administration of nucleotides (0.1%) in broiler diet on the lipid composition of the *Pectoralis major* muscle after slaughtering at 52nd day of the trial. The results indicated that the nucleotides influenced lipid content significantly in the muscle, probably in relation to the physiological effect of nucleotides to stimulate the alfa-lipoprotein synthesis during the neonatal period. Also the results suggested that there
was no induction of PUFA accumulation in *Pectoralis major* muscle owing to dietary supplementation with nucleotides.

**(E) Effect on intestinal villus growth**

Adjei *et al.* (1996) conducted an experiment on mice which were divided into eight groups and fed with 20% casein diet (control), protein free diet, protein free diet supplemented with 3 M cytidine, uridine, thymidine, inosine, guanosine monophosphate, and nucleoside-nucleotide mixture respectively for four weeks. At the end of 4 weeks, each mouse was injected with lipopolysaccharide (50 micrograms/500 microliters) intraperitonially and the incidence of bacterial translocation and the ileal histology were noted after 48 hours. It was found that the death rate and the bacterial translocation to the mesenteric lymph node were lower and the villous height, crypt depth and the wall thickness of the intestine were higher in the nucleoside-nucleotide mixture groups. Based on the results they suggested that dietary nucleotide and nucleosides improve the survivability and intestinal repair after damage.

Domeneghini *et al.* (2004) performed an experiment to study the effect of dietary supplementation of glutamine and nucleotide on intestinal mucosa of 16 weaned piglets. Piglets were separated into 4 groups and supplemented with 4 different diets for 28 days i.e. control (C), C + 0.05% glutamine, C + 0.05% nucleotides and C + 0.05% glutamine + 0.05% nucleotides. The results obtained during the histological examination of distal ileum at the end of experiment revealed increase in villi (V) height, crypt (C) depth, decrease in V:C ratio and mitotic mucosal cell. Similarly, greater percentage of mucosal macrophages were noted in treatment groups. However, there was no significant difference in growth performance among groups.

Puig *et al.* (2007) designed an experiment to determine the effect of dietary nucleotide supplementation on development of intestinal mucosa using 24 piglets of 21 days of age from 6 litters. The piglets were randomly distributed in 4 groups i.e. one group of 6 piglets remained in lactation, whereas the other 3 groups were weaned. The weaned groups were fed with a commercial Prestarter diet (control group), diet supplemented with 1,000 and 2000 ppm of nucleotide. They found that the nucleotide
supplement brought about a significant reduction (P<0.001) in the atrophy of the villosities (characteristic of weaning) in the distal jejunum samples collected seven days after weaning.

Chumpawadee et al. (2008) conducted a study to investigate the effect of dietary inclusion of cassava yeast on growth performance, ileum morphology and carcass percentage in Japanese quails. The results showed that feed conversion ratio, average daily gain and carcass percentage were not significantly different among treatments supplemented with *Saccharomyces cerevisiae* @ 0, $10^6$, $10^7$ and $10^8$ organisms/kg feed. Significant differences were observed in feed intake during finisher period, tight percentage and villus height. The results of the experiment showed that dietary inclusion of cassava yeast to Japanese quail had minimal influence on their performance.

Moore et al. (2011) conducted an experiment on ninety individually housed castrated pigs to study the effect of yeast protein concentrate or its major active components viz. nucleotide, inositol and glutamate on pig performance, indices of gut structure and circulating measures of immune function. The results of the experiment indicated that daily weight gain and feed intake were not affected by dietary supplementation. However, the pigs of yeast protein concentrate supplemented group had showed a low feed conversion ratio and improved duodenal villous height compared to the other groups.

Jung and Batal (2012) conducted two experiments to determine the effects of dietary nucleotide supplementation (Nupro) on broiler performance, physical and morphological development of the gastrointestinal tract under normal and stressed conditions. No significant differences were noticed from 0 to 16 days by supplementing the diets with up to 0.5 per cent Torula yeast RNA under normal conditions. However under high stocking density, broilers fed on the diets supplemented with 2 and 6 per cent Nupro from 0 to 14 d of age had better feed conversion (feed:gain) ratios over the entire experiment (0 to 32 d of age), even though the diets were supplemented with Nupro from 0 to 14 d of age. The broilers fed on the diets supplemented with 0.25 per cent Torula yeast RNA and 2 per cent Nupro had higher villus height and improved villus height-to-crypt depth ratio as compared with birds fed on the control or 6 per cent Nupro diet at 14 d of age.
Materials and Methods
Chapter 3 MATERIALS AND METHODS

The present investigation was carried out to study the effect of dietary supplementation of different levels of nucleotide source (yeast) in Japanese quails. The study was conducted as two experiments during October (2012) – February (2013) at Instructional Poultry Farm (IPF), College of Veterinary and Animal Sciences, Govind Ballabh Pant University of Agriculture and Technology, Pantnagar (U.S. Nagar), Uttarakhand. Different levels of nucleotide supplement through feed were provided to Japanese quails of different groups. The details of experimental and analytical procedures carried out to study the effect of various levels of nucleotide supplement on growth performance, nutrient retention, carcass traits, meat composition, haematological, certain serum biochemical and health status related parameters, intestinal morphology and immunological status in Japanese quail (*Coturnix coturnix japonica*) are described in this chapter.

3.1 EXPERIMENT I

The experiment I was conducted during October - November, 2012 in order to find out the approximate level of the nucleotide supplement. Based on the results of the experiment I, the levels of supplement for the experiment II were fixed.

3.1.1 Experimental birds

For experiment I, day-old broiler red plumaged Japanese quail chicks (*Coturnix coturnix japonica*), belonging to the same hatch, were reared at Instructional Poultry Farm, Pantnagar. After three days of brooding (Fig. 3.1, 3.2), chicks were used for actual experiment. On day 4th, all the chicks were individually weighed and 120 of them were randomly allocated into 4 different treatment groups with three replicates of 10 Japanese quail chicks in each. Over and underweight chicks were discarded.

3.1.2 Housing and management

The chicks were housed in deep litter system (Fig. 3.3, 3.4). All the housing and managerial conditions were similar for different treatment groups in the
experiment. They were given 18 hour light. Control as well as supplemented diets and fresh drinking water were provided \textit{ad libitum} to the experimental birds during the entire experimental period on 42 days.

**3.1.3 Design of experiment**

The experiment was carried out in a completely randomized design (CRD) in which nucleotide supplement at different levels were provided. After three days of brooding, 120 chicks were divided randomly into four treatment groups consisting of 30 chicks each and each group was subdivided into three replicates of 10 chicks each.

**3.1.3.1 Feed supplement and feeding schedule**

Standard feed for starter and finisher period was provided to the quails. The composition of starter feed provided for 1-2 weeks and finisher feed provided for 3-6 weeks period to meet the requirement of all the essential nutrients for broiler Japanese quails during experiment I is given in Table 3.1.

**Table 3.1: Nutrient composition (on dry matter basis) of basal ration used during Experiment I**

<table>
<thead>
<tr>
<th>Nutrient (%)</th>
<th>Starter feed</th>
<th>Finisher feed</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moisture</td>
<td>9.75</td>
<td>9.70</td>
</tr>
<tr>
<td>Crude protein</td>
<td>23.11</td>
<td>21.69</td>
</tr>
<tr>
<td>Crude fibre</td>
<td>5.82</td>
<td>5.96</td>
</tr>
<tr>
<td>Ether extract</td>
<td>4.09</td>
<td>4.26</td>
</tr>
<tr>
<td>Total ash</td>
<td>6.54</td>
<td>7.00</td>
</tr>
<tr>
<td>Acid insoluble ash</td>
<td>1.94</td>
<td>1.97</td>
</tr>
</tbody>
</table>

Commercial nucleotide supplement (Nucleoplus) from yeast \textit{Saccharomyces cerevisiae} was bought from A.A. Biotech Pvt. Ltd., Chennai. It was stored in air tight container as per the precautions advised by the manufacturer. Specifications (as given by manufacturer) and proximate analysis of the nucleotide supplement are presented in Table 3.2 and Table 3.3, respectively.
Table 3.2: Specification of the nucleotide supplement

<table>
<thead>
<tr>
<th>Content</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moisture</td>
<td>12 % max.</td>
</tr>
<tr>
<td>Crude Protein</td>
<td>50 % min.</td>
</tr>
<tr>
<td>Nucleotide content</td>
<td>6 % min.</td>
</tr>
<tr>
<td>Minerals</td>
<td>9 % max.</td>
</tr>
</tbody>
</table>

Table 3.3: Proximate analysis of nucleotide supplement

<table>
<thead>
<tr>
<th>Content</th>
<th>Value (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dry matter</td>
<td>88.18</td>
</tr>
<tr>
<td>Crude Protein</td>
<td>52.42</td>
</tr>
<tr>
<td>Ether Extract</td>
<td>2.33</td>
</tr>
<tr>
<td>Ash</td>
<td>8.76</td>
</tr>
</tbody>
</table>

3.1.3.2: Experimental treatments

There were four treatments employed in the experiment. The first treatment was considered control (T₁) in which no supplement was added to basal feed, while in T₂, T₃ and T₄ groups nucleotide supplement was provided through feed at different concentrations i.e. 0.5, 1.0 and 1.5 per cent respectively.

3.1.4 Nutrient analysis

The proximate analysis viz. moisture, crude protein, crude fibre, ether extract and total ash of feed, feed supplement, excreta and meat were determined as per the methods of AOAC (2003).

3.1.5 Experimental schedule

Treatment supplementation was continued for six weeks to determine the effect of nucleotide supplement. Body weight and feed intake were recorded at weekly intervals under two periods i.e. I and II week (Starter period), III - VI week (Finisher period). Growth parameters viz. body weight gain, average feed intake, FCR and performance index were calculated for starter, finisher and overall period.
Delayed type of hypersensitivity test was conducted on the 21st day of experiment by randomly selecting six Japanese quails from each group. The sensitized Japanese quails were challenged two weeks later and assessment of reaction was done 24 and 48 hours post challenge.

A metabolic trial of 7 days duration was conducted between 36th to 42nd days of the experiment to estimate the nutrient utilization. Four birds i.e. two male and two female from each replicate were used for metabolic trial.

At the end of experiment on 42nd day, carcass traits as well as meat composition studies were done. Blood was also collected for haemato-biochemical parameters and humoral immune response.

3.1.6 Growth performance

The data on growth parameters of broiler Japanese quails during the six weeks of experimental period as affected by dietary nucleotide supplementation were collected and growth parameters were studied under following heads:

i. Feed intake

ii. Body weight gain

iii. Feed conversion ratio

iv. Performance index

These growth parameters were studied as follows:

3.1.6.1 Feed intake

Feed consumption of the experimental birds under each treatment group was calculated on replicate basis at weekly interval for six weeks (4th – 45th days of age). For this purpose daily feed offered to different groups was recorded. At the end of week, feed residue and spillage were collected, weighed and their amount was deducted from total feed offered to calculate the net feed intake.

3.1.6.2 Body weight gain

All the chicks were weighed individually (Fig. 3.5, 3.6) from each replicate group at weekly interval (I - VI weeks) to get week wise body weight. The average value of body weight gain under each treatment group was then calculated.
3.1.6.3 Feed conversion ratio (FCR)

Feed conversion ratio for each week was calculated from feed intake and body weight gain of that period as under:

\[
\text{Feed conversion ratio} = \frac{\text{Feed consumed (g)}}{\text{Weight gain (g)}}
\]

3.1.6.4 Performance index (PI)

The performance index of broilers during different periods of growth was calculated by using the formula given by Bird (1955).

\[
\text{Performance index} = \frac{\text{Body weight gain (g)}}{\text{FCR}}
\]

3.1.7 Nutrient retention

A metabolic trial of 7 days duration was conducted between 36th to 42nd days of the experiment to estimate the nutrient utilization. Four birds i.e. two males and two females from each replicate were housed in metabolic cages (Fig 3.7). The experimental feed and water were provided ad libitum. Birds were given 4 days adaptation period followed by 3 days collection period. During the adaptation period, excess amount of weighed feed was offered to the birds at a fixed time in morning and the residue left was weighed next day morning at the same time. During the collection period, 70 per cent of the feed consumed in the adaptation period per day was calculated and offered to the birds at the same time every day. Simultaneously, galvanized tin trays layered with polythene sheets were placed for collection of excreta. Excreta were pooled, weighed and dried in a hot air oven for dry matter estimation and thereafter stored for further analysis. Representative samples of dried excreta were drawn for chemical analysis. For nitrogen estimation, fresh samples of excreta were preserved in 5 per cent sulphuric acid (v/v). The pooled samples of feed and excreta were analyzed to determine nutrient balances.

\[
\text{Nutrient utilization (\%)} = \frac{\text{Nutrient intake in feed} - \text{Nutrient loss in faeces}}{\text{Nutrient intake in feed}} \times 100
\]
3.1.8 Carcass characteristics

At the end of experiment on 45th day of age, two quails from each replicate (6 quails per treatment) were randomly selected and slaughtered for carcass trait study. Prior to slaughter the Japanese quails were off fed for 12 hours. For dressing and processing of the experimental quails, different steps followed were as under:

1. Slaughtering: The Japanese quails were weighed alive just prior to slaughter. They were killed by cutting the carotid artery and jugular vein by single clean cut with a sharp knife and left for bleeding.

2. Bleeding: For complete bleeding one minute was allowed without any struggling and then the carcass was again weighed and the blood loss was recorded.

3. Scalding: The bled carcass was dipped in hot water for one and half minutes. The temperature of water was kept 58°C.

4. Defeathering: The feathers were removed manually, after the removal of pin feather, the carcass was again weighed to record the feather loss.

5. Dressing: The Japanese quails were dressed by removing the head by cutting between the first cervical and occipital bone. The neck at the base where it joins the body was cut off, the blood adhering to it was removed. The shanks were cut off from the hock joint and the head and shanks were discarded.

6. Evisceration: It was done by making a slit from the tip of breast bone up to the area around the cloaca. The visceral organs were removed by supporting the bird with one hand through the incised abdomen. Liver was removed carefully. Gall bladder was removed gently without rupture. Gizzard and heart were also removed carefully. The internal layer of gizzard lining was removed retaining its muscular portion. The pericardium of heart was removed.

7. Washing: After evisceration, thorough washing and cleaning was done with running tap water.

8. Draining: The carcasses were kept hanging on the special racks for 5 – 10 minutes.

Various parameters viz. dressed yield without or with giblet, organ weights i.e. liver, heart and gizzard weights, cut-up parts i.e. drumsticks and thighs, wings, neck,
back and breast and processing losses as blood loss, feather loss, head and shank losses as per cent of live weight were then calculated.

\[
\text{Dressed yield (\%)} = \frac{\text{Live weight} - (\text{Weight loss as blood, feathers, head, shank and viscera})}{\text{Live weight}}
\]

After weighment, around 20 g of meat sample from both breast and thigh muscles were collected for the analysis of meat composition.

### 3.1.9 Intestinal morphology

After evisceration the intestine of the birds were carefully separated and the length of the intestine from duodenum to end of the cloaca was measured using a measuring tape to study the effect of nucleotide supplementation on the intestinal gross morphology. A sample of two cm from proximal jejunum was collected and preserved in 10 per cent formalin to study the histological changes in the cross section of intestine.

Two cross sections (4-5 µm) of 10 per cent formalin-preserved and processed segments from each jejunum sample were then prepared using microtome for staining with haematoxylin and eosin using standard paraffin embedding procedures (Uni et al., 1995). A total of three intact well-oriented villi were selected in two replicates from each jejunum cross section (six measurements for each jejunum sample, with thirty six measurements per treatment). Villus height was measured from the tip of the villus to the bottom of the villus, and crypt depth was measured from the villus bottom to the crypt base. Villus height and crypt depth ratio was also calculated.

### 3.1.10 Analysis of feed, excreta and meat samples

The proximate analysis viz. moisture, crude protein, crude fat, crude fibre, NFE and total ash of feed and dry matter, crude protein and crude fat content of meat and excreta were determined as per the methods of AOAC (2003).

#### 3.1.10.1 Determination of dry matter

Representative sample was taken in the pre- weighted petri dish and kept in hot air oven at 100°C for 24 hrs. Weight of the petri dish was again taken after 24 hours. Dry matter was calculated as follows:
Dry matter (%) = \( \frac{b}{a} \times 100 \)

where, 
\( a = \) Fresh weight of sample (g) 
\( b = \) Weight of sample after drying (g) 

Moisture (%) = 100 – Dry matter (%)

3.1.10.2 Determination of nitrogen and crude protein

The protein content was determined by Kjeldahl method. For the purpose, 2 g of representative sample was taken in a digestion flask followed by addition of 3 g of digestion mixture (\( \text{K}_2\text{SO}_4:\text{CuSO}_4 \) in 9:1 ratio) and 20 ml of conc. sulphuric acid. The contents were then digested till a blue/green transparent liquid was obtained. After cooling the volume of digested mixture was made 100 ml with distilled water. A 20 ml aliquot of digested mixture was distilled with excess of 40 per cent NaOH solution and liberated ammonia was collected in 20 ml of 2 per cent boric acid solution containing 2 to 3 drops of mixed indicator (10 ml of 0.1 per cent bromocresol green + 2 ml of 0.1 per cent methyl red indicator in 95 per cent alcohol). The entrapped ammonia was titrated against 0.1N HCl. A reagent blank was similarly digested and distilled. Nitrogen content in sample was calculated as follows:

\[
\text{Nitrogen} \% = \frac{\text{Sample Titre} - \text{Blank Titre} \times \text{Normality of HCl} \times 14 \times \text{Volume made up}}{\text{Aliquot of digest taken} \times \text{Weight of sample taken}}
\]

Crude protein % was then calculated as under

\[
\text{Crude protein} (\%) = \text{Nitrogen} (\%) \times 6.25
\]

3.1.10.3 Determination of ether extract

For estimation of ether extract Soxhlet method was used. In this method 2 g of dried and grinded sample was transferred to a thimble and weight of the empty oil flask was noted. Thimble was placed in Soxhlet’s apparatus for 8 hours in straight direction. Petroleum ether (B.P. 40-60°C) was used as solvent which was subsequently evaporated. After 8 hours the thimble was taken out and oil flask was placed in hot air oven for evaporation of ether, it was then removed from hot air oven, cooled in a desiccator and weighed. The calculation of ether extract (%) was done as under.
Ether extract (%) = \frac{b}{a} \times 100

where,

a = weight of sample
b = (weight of oil flask after extraction) – (weight of oil flask before extraction)

3.1.10.4 Determination of crude fibre

The dry sample after de-fatting was transferred from thimble to spoutless beaker of one litre capacity and in beaker; 200 ml of 1.25 per cent H₂SO₄ was added. It was refluxed for 30 minutes on hot plate after the boiling started and thereafter, filtered through muslin cloth. The residue was washed 5-6 times with hot water until it became acid free. The residual material on the muslin cloth was again transferred to the respective beaker and in beaker 200 ml of 1.25 per cent sodium hydroxide (NaOH) solution was added. It was again refluxed for 30 min. after the boiling started and thereafter filtered through muslin cloth and washed with hot water for 5-6 times until it became free from alkali. Thereafter, total residue was transferred in a clean, dry silica crucible and dried in hot air oven at 100ºC for 24 hrs. Then, it was cooled in desiccator and weighed. The residue was then ignited in muffle furnace at 600ºC for 2 hrs. After 12 hrs silica crucible containing ash was removed from the furnace and transferred into desiccator, cooled and weighed again. Weight loss during ignition was recorded as the weight of crude fibre:

\[
\text{Crude fibre (\%) on DM basis} = \frac{b-c}{a} \times 100
\]

Where,

a = weight of sample on DM basis (g)

b = weight of silica crucible before ignition (g)

c = weight of silica crucible containing residue after ignition (g)

3.1.10.5 Determination of total ash

Five g of ground sample was taken in previously weighed silica crucible. The crucible along with sample was kept on a heater and burnt till no more smoke was
given off by the charred mass of sample. Thereafter, the silica crucible containing charred mass of sample was transferred into muffle furnace with the help of metal tong and ignited at 600°C for 2 hrs. After 12 hrs, the crucible containing ash was removed from the furnace and then transferred into desiccator, cooled and weighed. Total ash was calculated as follows:

\[
\text{Total ash (\% on DM basis)} = \frac{a-b}{c} \times 100
\]

where,

- \(a\) = weight of silica crucible with ash (g)
- \(b\) = weight of empty silica crucible (g)
- \(c\) = weight of sample taken for ashing on dry matter basis (g)

3.1.10.6 Estimation of acid insoluble ash

For the estimation of acid insoluble ash 20 ml dilute HCl (1:1) was added in the crucible containing ash. The crucible was covered with watch glass and the sample was digested for 20 minutes on water bath. After digestion the watch glass was removed and rinsed with water into the contents of the crucible. The digested material was then filtered through Whatman filter paper No. 1 into a volumetric flask, the residue in the crucible was washed and transferred repeatedly with small volumes of 5% HCl solution in volumetric flask. The filter paper along with the residue was transferred to the same crucible, dried in hot air oven and then ignited in muffle furnace at 600°C for 2 hours. After few hours crucible was removed from furnace, cooled in a desiccator and its weight was recorded. The increase in weight of crucible gave the weight of acid insoluble ash.

\[
\text{Acid insoluble ash (\%)} = \frac{b}{a} \times 100
\]

where,

- \(a\) = original weight of sample (g)
- \(b\) = weight of insoluble ash (g)
3.1.11 Blood sample collection

Blood samples were collected at the end of the experiment. On last day of experiment blood was collected from two birds of each replicate aseptically through the jugular vein in sterilized disposable syringe (24 gauge needle). Out of the whole blood collected, part of blood was transferred to the vials containing ethylene diamene tetra acetate (EDTA) and used for estimation of haemoglobin (Hb), packed cell volume (PCV), red blood cells (RBC) and white blood cells (WBC) counts. While, remaining blood was transferred in a clean test tube for serum separation to study the biochemical parameters.

3.1.12 Haematological parameters

Following haematological parameters were determined.

3.1.12.1 Total erythrocyte count

Total erythrocytes count (TEC) was done by using Neubauer’s counting chamber as described by Natt and Herrick (1952). The blood sample was then diluted with Vallarino’s solution (Iodine 0.3 g, Potassium iodide 0.4 g, Sodium citrate 2.0 g and Distilled water 100 ml). Well mixed blood sample was sucked up to 0.5 mark in RBC diluting pipette, excess blood was wiped out from the stem of the pipette. Diluting fluid was sucked up to 101 marks slowly and carefully to get 1:200 dilution of blood. The pipette was rolled in between the palms in horizontal position to mix blood and diluting fluid. One secondary square of haemocytometer was focused under high power of microscope and a cover slip was placed over the counting area. First 2-3 drops of diluted blood were discarded and then one drop of diluted blood was gently placed between cover slip and haemocytometer slide so that fluid spread under the cover slip by capillary action. The cells were allowed to settle down for five minutes. The total number of cells in four corners and one central secondary square (i.e. total 80 tertiary squares) were counted. Total erythrocyte numbers ($10^6/\mu l$) were calculated as follows:

\[
\text{Total RBCs in 5 secondary squares} = n
\]

\[
\text{Volume of diluted blood in 5 secondary squares} = (1/250) \times 5 = 1/50 \mu l
\]
Since n is the no. of RBCs present in 1/50 µl of diluted blood. Therefore 1 µl of diluted blood had 50n x 200 RBCs (200 dilution factor). Therefore, TEC of sample was 10000 n/µl of blood.

3.1.12.2 Total leukocyte count

Total Leukocyte Count (TLC) was performed with Neubauer’s counting chamber (Jain, 1986) using Thomas diluting fluid (2 ml glacial acetic acid, 1 ml 1 per cent Gentian violet in 100 ml of distilled water). For this purpose, well mixed blood samples were sucked up to 0.5 mark in WBC diluting pipette, excess blood was wiped out from the stem of the pipette. Diluting fluid was sucked up to 11 mark to provide 1:20 dilution of blood. The pipette was rolled in between the palms in horizontal position to mix blood and diluting fluid. One primary square (1 mm²) of haemocytometer was focused under low power of microscope and a cover slip was placed over the counting area. First 2-3 drops of diluted blood were discarded and then one drop of diluted blood was gently placed between cover slip and haemocytometer slide so that fluid spread under the cover slip by capillary action. The cells were allowed to settle down for five minutes. The total numbers of WBC were counted in all four primary squares (i.e. total 64 secondary squares). The number of total leukocytes was expressed in thousands per microlitre (10³/µl) and calculated as follows.

Total WBCs in 4 primary square = n
Volume of 4 primary squares = (1/10) x 4 =0.4 µl

Since 0.4 µl of diluted blood contained n cells, so 1 µl of diluted blood had n/0.4=2.5 n and 1 µl of whole blood had 2.5n x 20 = 50n WBC (20 dilution factor). Therefore, TLC of sample was 50 n/µl of blood.

3.1.12.3 Packed cell volume

Packed cell volume (PCV) was estimated using micro haematocrit method as described by Sharma and Singh (2000). Fresh anticoagulant added blood was drawn into micro capillaries and sealed with wax at one end. Capillaries were centrifuged at 10,000 rpm for 3 minutes. Packed cell volume was directly read by using Citro Cap Microhaematocrit tube reader and expressed in per cent.
3.12.4 Haemoglobin

Haemoglobin (Hb) concentration was estimated spectrophotometrically at 540 nm by cyanomethemoglobin method, using Drabkin’s solution (Fudge, 2000) of Span Diagnostics. The concentration of Haemoglobin was expressed in g/dl.

3.12.5 Mean corpuscular volume (MCV)

Mean corpuscular volume was calculated by using following formula and expressed in femtoliter (fl).

\[
\text{MCV} = \frac{\text{Packed cell volume (％)}}{\text{Total erythrocyte count (10⁶/µl)}} \times 10
\]

3.12.6 Mean corpuscular haemoglobin (MCH)

Mean corpuscular haemoglobin was calculated by using following formula and expressed in pg.

\[
\text{MCH} = \frac{\text{Haemoglobin (g/dl)}}{\text{Total erythrocyte count (10⁶/µl)}} \times 10
\]

3.12.7 Mean corpuscular haemoglobin concentration (MCHC)

Mean corpuscular haemoglobin concentration was calculated by using following formula and expressed in per cent

\[
\text{MCHC} = \frac{\text{Haemoglobin (g/dl)}}{\text{PCV (％)}} \times 100
\]

3.12.8 Differential leukocyte count

Six blood smears, from each treatment group (two/replicate), were prepared. The differential counting of white blood cells were done on blood smears stained with Giemsa stain via enumeration of 100 cells (Coles, 1986) and the heterophil/lymphocyte ratio was also calculated.

3.13 Serum biochemical parameters

Serum samples obtained from the blood were used for certain biochemical parameters, protein profile, some metabolites and enzymes estimation. Glucose and
enzymes estimations were done within 24 hours of collection while for other estimations serum was stored in refrigerator till the analysis. Following biochemical parameters were determined using the blood samples.

3.1.13.1 Serum glucose

Estimation of glucose was done spectrophotometrically by enzymatic GOD-POD method with the help of Span Diagnostic Kit at 505 nm wavelength (Sacks, 1998). Concentration of serum glucose was expressed in mg/dl.

3.1.13.2 Serum lipid profile

a) Serum total cholesterol

Total cholesterol concentration in serum was estimated spectrophotometrically using Span Diagnostic Kit with enzymatic CHOD-PAP method at 505 nm wavelength (Tietz, 1998) and expressed in mg/dl.

b) Serum HDL cholesterol

HDL cholesterol concentration was estimated spectrophotometrically at 560 nm by phosphotungstate method (Tietz, 1998) using Span Diagnostic Kit and expressed in mg/dl.

c) Serum LDL cholesterol

The serum LDL cholesterol was expressed in mg/dl and calculated using Friedwald’s equation as follows:

\[ \text{LDL cholesterol} = \text{Total cholesterol} - \left( \frac{\text{Triglycerides}}{5} \right) - \text{HDL cholesterol} \]

d) Serum triglycerides

The concentration of serum triglycerides was estimated at 505 nm spectrophotometrically using Span Diagnostic Kit by GPO – PAP, end point assay (Stein and Mayer, 1995). The concentration was expressed in mg/dl.

3.1.13.3 Health status related parameters

Following health status related parameters were determined using the serum obtained from blood samples.
3.1.13.3.1 Serum creatinine

The creatinine concentration in the serum was estimated at 520 nm spectrophotometrically by Alkaline picrate method (Toro and Ackermann, 1975) using Span Diagnostic kit and expressed in mg/dl.

3.1.13.3.2 Serum uric acid

Serum uric acid was estimated by enzymatic method using Span Diagnostic Kit at 660 nm wave length (Tietz, 1998). The values were expressed in mg/dl.

3.1.13.3.3 Serum protein profile

Following parameters were estimated and calculated.

a) Serum total protein

Total protein concentration in serum was estimated by Biuret method with the help of Span Diagnostic Kit at 540 nm wavelength (Johnson et al., 1999) and expressed in g/dl.

b) Serum albumin

Albumin concentration in serum was estimated by bromocresol green, end point assay method with the help of Span Diagnostic Kit at 630 nm wavelengths (Johnson et al., 1999) and expressed in g/dl.

c) Serum globulin

The albumin content was deducted from total protein to calculate globulin and expressed in g/dl (Johnson et al., 1999).

d) Albumin-globulin ratio

The albumin-globulin ratio was calculated by using following formula :

\[
\text{Albumin- globulin ratio} = \frac{\text{Serum albumin (g/dl)}}{\text{Serum globulin (g/dl)}}
\]

3.1.13.3.4 Serum enzyme profile

Following Serum enzymes were estimated using the serum obtained from the blood samples.
a) Serum glutamate oxaloacetate transaminase (SGOT)

SGOT concentration in serum was estimated by DNPH colorimetric method with the help of Span Diagnostic Kit at 505 nm wavelength (Tietz, 1998). Concentration of SGOT was expressed in IU/L.

b) Serum glutamate pyruvate transaminase (SGPT)

SGPT concentration in serum was estimated by DNPH colorimetric method with the help of Span Diagnostic Kit at 505 nm wavelength (Tietz, 1998). Concentration of SGPT was expressed in IU/L.

3.1.14 Immunological studies

The cell mediated immune response of the birds was evaluated using delayed type hypersensitivity and humoral response (serum total immunoglobulin) was estimated by zinc sulphate turbidity test.

These immunological parameters were determined as follows:

3.1.14.1 Cell mediated immunity

The cell mediated immune response (CMIR) was studied by delayed type of hypersensitivity (DTH) reaction to 2, 4 dinitro-chloro benzene (DNCB), as per method adopted by Tiwary and Goel (1985) with slight modification.

3.1.14.1.1 Sensitization of cockrels to DNCB

On 21st day of experiment, six birds from each group were selected for the test. The relatively featherless elliptical area of approximately 20 to 30 square mm was marked on both right and left lateral side of abdomen for epi-cutaneous application of DNCB (Fig. 3.8). The left side served as test area while right side area was control during the challenge dose. The marked area on left side was cleaned with alcohol and then 0.25 ml of DNCB (10 mg/ml) in a vehicle consisting of acetone and olive oil (4:1) mixture was applied as a sensitizing dose.

3.1.14.1.2 Challenge

The sensitized birds were challenged two weeks later by applying 0.25 ml of DNCB (1 mg/ml) to marked area on left side. On the right side which was marked as control, only vehicle was applied.
3.1.14.1.3 Assessment of reaction:

Skin thickness was measured by Vernier’s caliper at zero (prior to challenge), 24 hours and 48 hours post challenge to assess the reaction (Fig. 3.9). Increase in mean skin thickness (MST) of birds was recorded as difference in thickness of skin at 24 hours and 48 hours against zero hour thickness. Each measurement was repeated thrice on a constant area and overall MST for each group was calculated and recorded.

3.1.14.2 Humoral immunity

Total serum immunoglobulins were estimated by using zinc sulphate turbidity test (McEvan et al., 1969; Mondesire, 2004). For performing this test, 0.1 ml serum and 6 ml of distilled water was used as control while 0.1 ml serum with 6 ml zinc sulphate solution (250 mg of zinc sulphate dissolved in 1 litre of distilled water) was taken as a test. These were shaken well and then kept at room temperature for 60 min. The optical density (O.D) was read at 545 nm by spectrophotometer. Total immunoglobulins were calculated as follows:

Zinc sulphate turbidity (ZST units) = Reading of test – Reading of control × 10

Total immunoglobulins (g dL⁻¹) = 0.04 + 0.98 ZST units.

3.2 EXPERIMENT II

The experiment II was conducted during Jan – Feb 2013. In the experiment I, T₂ group Japanese quails supplemented with 0.5 per cent nucleotide supplement showed better results compared to other groups. On the basis of the experiment I results, the levels of the nucleotide supplement for the second experiment were fixed as 0, 0.25, 0.5 and 0.75 per cent in feed, for the treatment groups T₁, T₂, T₃ and T₄ respectively.

In the experiment II, the performance of Japanese quails in terms of growth and nutrient utilization (metabolic trial) were studied similar to the experiment I using 120, three day old chicks of red plumaged Japanese quails (*Coturnix coturnix japonica*) which were individually weighed and randomly allocated into 4 different treatment groups with three replicates of 10 Japanese quails in each.
The composition of standard starter feed provided for I – II weeks and finisher feed provided for III – VI weeks period to meet the requirement of all the essential nutrients for broiler Japanese quails during experiment II is given in Table 3.4.

Table 3.4: Nutrient composition (on dry matter basis) of basal ration used during Experiment II

<table>
<thead>
<tr>
<th>Nutrient (%)</th>
<th>Starter feed</th>
<th>Finisher feed</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moisture</td>
<td>9.70</td>
<td>9.6</td>
</tr>
<tr>
<td>Crude protein</td>
<td>22.78</td>
<td>20.93</td>
</tr>
<tr>
<td>Crude fibre</td>
<td>5.97</td>
<td>6.03</td>
</tr>
<tr>
<td>Ether extract</td>
<td>4.52</td>
<td>4.86</td>
</tr>
<tr>
<td>Total ash</td>
<td>6.32</td>
<td>6.62</td>
</tr>
<tr>
<td>Acid insoluble ash</td>
<td>1.82</td>
<td>1.90</td>
</tr>
</tbody>
</table>

3.3 Statistical analysis

All the data obtained in the experiment were finally analyzed using analysis of variance (ANOVA) and the critical difference (CD) was calculated to determine any significant difference among the treatment means (Snedecor and Cochran, 1994) and by using STPR 3 software system.
Fig. 3.1 and 3.2: Brooding of Japanese quails under canopy

Fig. 3.3 and 3.4: Rearing of Japanese quails in deep litter system

Fig. 3.5 and 3.6: Weighing of Japanese quails
Materials and Methods

Fig. 3.7: Rearing of Japanese quails in metabolic cages

Fig. 3.8: Epi cutaneous application of DNCB for delayed type of hypersensitivity test in Japanese quails

Fig. 3.9: Measuring skin thickness in DNCB sensitized Japanese quails
Results and Discussion
Chapter 4

RESULTS AND DISCUSSION

The present investigation was conducted to study the effect of supplementation of yeast as nucleotide source on the growth performance, nutrient utilization, carcass traits, carcass composition, haematological parameters, certain serum biochemical and health status related parameters, intestinal morphology and immunological response of Japanese quails. The study was conducted in two experiments, each for a period of 6 weeks. For this purpose in both of the experiments, a total of one twenty Japanese quail chicks of three days age were divided randomly into four groups (T₁, T₂, T₃ and T₄), each group having three replicates of 10 chicks. These corresponded to control and three treatment groups. In the experiment I, the diets were prepared by addition of commercial yeast extract enriched with nucleotides in different concentrations (group T₁- control, T₂-0.5 per cent, T₃-1.0 per cent and T₄-1.5 per cent). Based on the performance of the birds in the experiment I, the levels for experiment II were fixed as 0, 0.25, 0.5 and 0.75 per cent for groups T₁, T₂, T₃ and T₄ respectively. In both of the experiments, growth parameters were studied at weekly interval in two different periods of age viz. Starter (I – II week) and Finisher (III – VI week). On 36th day of both the experiments, four birds (two male and two female) from each replicate were placed for metabolic trial to determine the effect of nucleotide supplementation on nutrient utilization of Japanese quails. In the experiment I, at the end on 42nd day, two birds from each replicate (6 birds / treatment) were randomly sacrificed for the study of carcass yield, processing losses, yield of cut up parts and organ weights. Meat samples from breast and thigh muscles were taken to analyze the composition as per the methods of AOAC (2003). Length of the intestine was measured after evisceration and an intestine sample of 2 cm from proximal jejunum was collected for morphological studies of intestine. Blood was also collected for haematobiochemical parameters and humoral immune response. Delayed type of hypersensitivity was studied from 21st day to 35th day of the experiment. Results obtained in both of the experiments were statistically analyzed and are presented and discussed in this chapter.

Results and Discussion ..............
4.1 Growth performance

The growth performance of experimental chicks of Japanese quails (*Coturnix coturnix Japonica*) with respect to feed intake, body weight gain, feed conversion ratio and performance index was calculated at weekly interval, starter period, finisher period as well as overall basis.

4.1.1 Feed intake (Experiment I)

The average feed consumption of Japanese quails in experiment I obtained at weekly interval as well as overall basis is presented in Table 4.1 and Fig. 4.1.

During first week of the experiment, Japanese quail chicks of control group consumed minimum (21.90 ± 0.98 g) feed which was statistically (P>0.05) similar to the feed intake of T₂ and T₄ groups. In the second week, the quail chicks of the T₂ and T₃ groups consumed significantly (P<0.05) less feed compared to the control (T₁) group. During starter period, the feed consumption of the chicks were significantly (P<0.01) lower in the groups T₂ and T₃ compared to the T₁ and T₄ groups. In the third, fourth and fifth week, the feed intake of the supplemented group birds were significantly (P<0.05) lower than the control. In the sixth week, feed intake in the control group of Japanese quails was minimum (169.16 ± 4.02 g) and significantly (P<0.05) lower than the T₂ and T₃ groups. Between III - VI week of feeding trial (finisher period), Japanese quails of T₂ and T₄ groups showed significant (P<0.01) reduction in feed intake compared to the control, whereas for overall period, all the supplemented groups showed significant (P<0.01) reduction in the feed intake compared to the control. Among all the treatments, the feed intake of T₁ group quails was highest (716.58 ± 14.15 g) and lowest in T₂ (650.55 ±7.31 g). However, during all the periods, T₂ group fed with 0.5 per cent nucleotide supplement/kg of feed (i.e. around 400 mg of nucleotide/kg of feed) showed significant (P<0.01) reduction in feed intake compared to the control and other supplemented groups.

4.1.1.2 Feed Intake (Experiment II)

The average feed consumption of Japanese quails in experiment II obtained at weekly interval as well as overall basis is presented in Table 4.2 and Fig. 4.2.
Table 4.1: Effect of nucleotide supplementation on average feed intake (g) of Japanese quails (Experiment I)

<table>
<thead>
<tr>
<th>Treatments</th>
<th>I*</th>
<th>II**</th>
<th>(I–II)**</th>
<th>III**</th>
<th>IV**</th>
<th>V**</th>
<th>VI*</th>
<th>(III - VI)**</th>
<th>(I–VI)** Overall</th>
</tr>
</thead>
<tbody>
<tr>
<td>T1</td>
<td>21.90(^b) ± 0.98</td>
<td>80.63(^a) ± 1.15</td>
<td>102.52(^a) ± 1.10</td>
<td>132.77(^a) ± 1.81</td>
<td>152.77(^a) ± 1.84</td>
<td>159.37(^a) ± 1.26</td>
<td>169.16(^b) ± 4.02</td>
<td>614.06(^a) ± 13.30</td>
<td>716.58(^a) ± 14.15</td>
</tr>
<tr>
<td>T2</td>
<td>23.18(^b) ± 0.32</td>
<td>65.21(^b) ± 3.20</td>
<td>88.38(^b) ± 3.52</td>
<td>103.75(^c) ± 1.04</td>
<td>129.59(^b) ± 5.07</td>
<td>136.84(^c) ± 0.43</td>
<td>192.00(^a) ± 6.45</td>
<td>562.17(^b) ± 9.17</td>
<td>650.55(^c) ± 7.31</td>
</tr>
<tr>
<td>T3</td>
<td>25.77(^a) ± 0.71</td>
<td>65.99(^b) ± 2.98</td>
<td>91.76(^b) ± 3.47</td>
<td>111.52(^b) ± 2.53</td>
<td>135.64(^b) ± 2.52</td>
<td>152.78(^b) ± 3.40</td>
<td>192.80(^a) ± 0.65</td>
<td>592.73(^a) ± 8.95</td>
<td>684.48(^b) ± 12.43</td>
</tr>
<tr>
<td>T4</td>
<td>24.06(^a) ± 0.49</td>
<td>80.63(^a) ± 1.15</td>
<td>104.68(^a) ± 1.47</td>
<td>104.17(^c) ± 2.08</td>
<td>132.29(^b) ± 1.15</td>
<td>140.00(^c) ± 1.25</td>
<td>185.38(^a) ± 6.99</td>
<td>561.83(^b) ± 6.50</td>
<td>666.51(^b) ± 6.15</td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01
Fig. 4.1: Effect of nucleotide supplementation on average feed intake (g) of Japanese quails (Experiment I)
Table 4.2: Effect of nucleotide supplementation on average feed intake (g) of Japanese quails (Experiment II)

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Weeks</th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>I*</td>
<td>II**</td>
<td>(I – II)**</td>
<td>III**</td>
<td>IV*</td>
<td>V**</td>
<td>(III - VI)*</td>
</tr>
<tr>
<td>T₁</td>
<td>30.25 ± 0.69</td>
<td>62.30 ± 0.08</td>
<td>92.55 ± 0.64</td>
<td>83.73 ± 0.29</td>
<td>144.63 ± 1.30</td>
<td>190.73 ± 0.44</td>
<td>626.91 ± 4.70</td>
</tr>
<tr>
<td>T₂</td>
<td>27.19 ± 0.67</td>
<td>57.40 ± 0.32</td>
<td>84.59 ± 0.36</td>
<td>75.90 ± 0.92</td>
<td>139.43 ± 0.21</td>
<td>195.16 ± 0.63</td>
<td>613.06 ± 2.48</td>
</tr>
<tr>
<td>T₃</td>
<td>27.38 ± 0.47</td>
<td>54.87 ± 0.48</td>
<td>82.24 ± 0.47</td>
<td>75.33 ± 0.48</td>
<td>142.40 ± 0.44</td>
<td>193.95 ± 0.35</td>
<td>626.09 ± 1.41</td>
</tr>
<tr>
<td>T₄</td>
<td>27.03 ± 0.60</td>
<td>53.13 ± 0.11</td>
<td>80.17 ± 0.64</td>
<td>78.47 ± 0.07</td>
<td>141.17 ± 0.79</td>
<td>196.05 ± 0.14</td>
<td>621.66 ± 0.58</td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01
Fig. 4.2: Effect of nucleotide supplementation on average feed intake (g) of Japanese quails (Experiment II)
During the first week of experiment, Japanese quail chicks of the supplemented groups consumed significantly (P<0.05) less feed compared to the control. Minimum feed intake (27.19 ± 0.67 g) was noted in chicks of T_4 group. In the second week, feed intake of the quails of all groups were significantly (P<0.01) different among themselves. The highest and lowest feed intake were recorded in quail chicks of T_1 (62.30 ± 0.08 g) and T_4 (53.13 ± 0.11 g) groups respectively. During the starter period, feed intake of the birds were significantly (P<0.01) reduced with increase in concentration of nucleotides in the feed. The highest and lowest feed intake were recorded in quail chicks of T_1 (92.55 ± 0.64 g) and T_4 (80.17 ± 0.64 g) groups respectively. During third week, the feed intake of all the treatment groups were significantly (P<0.01) lower than the control. Minimum (75.33 ± 0.48 g) feed intake was noted in quail chicks of T_3 groups. In the fourth week, Japanese quails of T_2 and T_4 groups showed significant (P<0.05) reduction in feed intake compared to the T_1 (control) group. However, the feed intake of T_1 during the fifth week was significantly lower and minimum (190.73 ± 0.44 g) than the other groups. In the sixth week, significantly (P<0.05) higher feed intake (214.41 ± 1.07 g) was noticed in the T_3 group compared to the rest of the treatment groups. All the other treatment groups had statistically similar feed intake with minimum (202.57 ± 1.43 g) in Japanese quails of T_2 group. During the finisher period, T_2 group Japanese quails consumed significantly (P<0.05) lower feed (613.06 ± 2.48 g) compared to other groups. For overall period, all the supplemented groups showed significant (P<0.01) reduction in the feed intake compared to the control. Feed intake of T_1 group quails was maximum (719.46 ± 5.27 g) and significantly (P<0.01) higher while that of T_2 group (697 ± 2.27 g) was minimum.

In both the feeding trials, the feed intakes of groups supplemented with nucleotides were lower than the control. These findings are in agreement with Garcia et al. (2007) who also noted significant reduction in the feed intake of broilers supplemented with 0.5 per cent nucleotide / kg of feed during the starter period.

In contrast to the results of present investigation, Domeneghini et al. (2004) found no significant difference in feed intake of piglets due to the supplementation of glutamine.
and nucleotides. Puig et al. (2007) and Pelicia et al. (2010) also noted no significant effect of nucleotide supplementation in piglets and broilers.

Reduced feed intake in nucleotide supplemented groups may be due to the additional nutrients supplied by them during growth.

**4.1.2.1 Body weight gain (Experiment I)**

Effect of supplementation of yeast extract enriched with nucleotides on body weight gain of Japanese quails for a period of six weeks in the experiment I is presented in Table 4.3 and Fig. 4.3.

During first week, the weight gain of Japanese quails of control group was minimum (13.97 ± 0.29 g) and significantly (P<0.01) lower than the supplemented groups. In the second week, body weight gain of Japanese quails of T2 group was maximum (30.71 ± 0.36 g) and significantly (P<0.01) higher as compared to the quails of other groups. All the other treatment groups had statistically similar body weight gains. The average body weight gain of Japanese quails during starter period was maximum (45.92 ± 0.13 g) and significantly (P<0.01) higher in T2 group quails while it was minimum (40.95 ± 0.43 g) and significantly lower in T1 group compared to the birds of other groups.

In the third week, the quails of control group (38.54 ± 0.19 g) showed significantly (P<0.01) higher body weight gain compared to nucleotide supplemented groups. During the fourth and sixth week, the body weight gains were significantly (P<0.01) higher in treatment groups compared to the control. Highest body weight gains were observed in quails of T2 group i.e., 32.33 ± 0.37 g and 21.13 ± 0.27 g respectively. During fifth week there was no significant difference in the weight gain of Japanese quails among control and supplemented groups. For finisher period, the average body weight gain of the T2 group was maximum (117.25 ± 0.07 g) and significantly (P<0.01) higher than the other groups. There was no significant difference in body weight gain between Japanese quails of T1 and T4 groups.
Table 4.3: Effect of nucleotide supplementation on body weight gain (g) of Japanese quails (Experiment I)

<table>
<thead>
<tr>
<th>Treatments</th>
<th>I**</th>
<th>II**</th>
<th>(I–II)**</th>
<th>III**</th>
<th>IV**</th>
<th>V</th>
<th>VI**</th>
<th>(III–VI)**</th>
<th>(I–VI)**</th>
</tr>
</thead>
<tbody>
<tr>
<td>T&lt;sub&gt;1&lt;/sub&gt;</td>
<td>13.97&lt;sup&gt;c&lt;/sup&gt; ± 0.29</td>
<td>26.99&lt;sup&gt;b&lt;/sup&gt; ± 0.14</td>
<td>40.95&lt;sup&gt;c&lt;/sup&gt; ± 0.43</td>
<td>38.54&lt;sup&gt;a&lt;/sup&gt; ± 0.19</td>
<td>29.77&lt;sup&gt;c&lt;/sup&gt; ± 0.15</td>
<td>27.18 ± 0.46</td>
<td>16.91&lt;sup&gt;b&lt;/sup&gt; ± 0.33</td>
<td>112.41&lt;sup&gt;c&lt;/sup&gt; ± 0.32</td>
<td>153.36&lt;sup&gt;d&lt;/sup&gt; ± 0.53</td>
</tr>
<tr>
<td>T&lt;sub&gt;2&lt;/sub&gt;</td>
<td>15.21&lt;sup&gt;b&lt;/sup&gt; ± 0.46</td>
<td>30.71&lt;sup&gt;a&lt;/sup&gt; ± 0.36</td>
<td>45.92&lt;sup&gt;a&lt;/sup&gt; ± 0.13</td>
<td>36.17&lt;sup&gt;b&lt;/sup&gt; ± 0.13</td>
<td>32.33&lt;sup&gt;a&lt;/sup&gt; ± 0.37</td>
<td>27.63 ± 0.14</td>
<td>21.13&lt;sup&gt;a&lt;/sup&gt; ± 0.27</td>
<td>117.25&lt;sup&gt;a&lt;/sup&gt; ± 0.07</td>
<td>163.17&lt;sup&gt;a&lt;/sup&gt; ± 0.18</td>
</tr>
<tr>
<td>T&lt;sub&gt;3&lt;/sub&gt;</td>
<td>16.63&lt;sup&gt;a&lt;/sup&gt; ± 0.18</td>
<td>27.30&lt;sup&gt;b&lt;/sup&gt; ± 0.05</td>
<td>43.93&lt;sup&gt;b&lt;/sup&gt; ± 0.20</td>
<td>36.68&lt;sup&gt;b&lt;/sup&gt; ± 0.71</td>
<td>30.92&lt;sup&gt;b&lt;/sup&gt; ± 0.43</td>
<td>27.47 ± 0.14</td>
<td>19.51&lt;sup&gt;a&lt;/sup&gt; ± 0.89</td>
<td>114.58&lt;sup&gt;b&lt;/sup&gt; ± 0.39</td>
<td>158.50&lt;sup&gt;b&lt;/sup&gt; ± 0.36</td>
</tr>
<tr>
<td>T&lt;sub&gt;4&lt;/sub&gt;</td>
<td>15.65&lt;sup&gt;ab&lt;/sup&gt; ± 0.45</td>
<td>28.04&lt;sup&gt;b&lt;/sup&gt; ± 0.52</td>
<td>43.69&lt;sup&gt;b&lt;/sup&gt; ± 0.22</td>
<td>34.00&lt;sup&gt;c&lt;/sup&gt; ± 0.64</td>
<td>31.63&lt;sup&gt;ab&lt;/sup&gt; ± 0.20</td>
<td>26.38 ± 1.05</td>
<td>19.67&lt;sup&gt;a&lt;/sup&gt; ± 0.61</td>
<td>111.67&lt;sup&gt;c&lt;/sup&gt; ± 0.18</td>
<td>155.36&lt;sup&gt;c&lt;/sup&gt; ± 0.09</td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01
Fig. 4.3: Effect of nucleotide supplementation on body weight gain (g) of Japanese quails (Experiment I)
Table 4.4: Effect of nucleotide supplementation on body weight gain (g) of Japanese quails (Experiment II)

<table>
<thead>
<tr>
<th>Treatments</th>
<th>I*</th>
<th>II**</th>
<th>(I–II)*</th>
<th>III**</th>
<th>IV**</th>
<th>V**</th>
<th>VI**</th>
<th>(III - VI)**</th>
<th>(I–VI)**</th>
</tr>
</thead>
<tbody>
<tr>
<td>T1</td>
<td>19.48± 0.11</td>
<td>28.10± 0.04</td>
<td>47.58± 0.14</td>
<td>35.60± 0.35</td>
<td>31.23± 0.05</td>
<td>24.70± 0.47</td>
<td>17.90± 0.20</td>
<td>109.43± 0.60</td>
<td>157.01± 0.49</td>
</tr>
<tr>
<td>T2</td>
<td>18.08± 0.28</td>
<td>29.20± 0.35</td>
<td>47.28± 0.16</td>
<td>35.58± 0.18</td>
<td>33.23± 0.23</td>
<td>25.43± 0.29</td>
<td>18.47± 0.31</td>
<td>112.72± 0.23</td>
<td>160.00± 0.08</td>
</tr>
<tr>
<td>T3</td>
<td>19.19± 0.03</td>
<td>29.85± 0.30</td>
<td>49.04± 0.33</td>
<td>37.77± 0.28</td>
<td>32.60± 0.21</td>
<td>24.85± 0.19</td>
<td>21.84± 0.04</td>
<td>117.06± 0.35</td>
<td>166.10± 0.57</td>
</tr>
<tr>
<td>T4</td>
<td>18.77± 0.32</td>
<td>28.28± 0.26</td>
<td>47.06± 0.49</td>
<td>37.78± 0.16</td>
<td>29.72± 0.23</td>
<td>27.87± 0.16</td>
<td>21.89± 0.02</td>
<td>117.26± 0.16</td>
<td>164.32± 0.41</td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01
Fig. 4.4: Effect of nucleotide supplementation on body weight gain (g) of Japanese quails (Experiment II)
The overall mean value of body weight gain for Japanese quails of all treatment groups were significantly (P<0.01) different among themselves. Overall body weight gain was maximum (163.17 ± 0.18 g) in Japanese quails of T₂ group and minimum (153.36 ± 0.53 g) in control group.

4.1.2.2 Body weight gain (Experiment II)

Effect of supplementation of yeast extract enriched with nucleotides on body weight gain of Japanese quails for a period of six weeks in the experiment II is depicted in Table 4.4 and Fig. 4.4.

During first week, the maximum (19.48 ± 0.11 g) weight gain was observed in the control group quail chicks which were statistically similar to the weight gain of T₃ and T₄ group chicks. In the second week, the weight gains of Japanese quail chicks of T₂ and T₃ groups were significantly (P<0.01) higher than the T₁ and T₄ groups. But, there was no significant difference between these two groups. During the starter period, the weight gain of T₃ group quails was significantly (P<0.05) higher than the quails of other groups. There was no significant difference in the weight gain of Japanese quails among other treatment groups.

In the third week, quails of T₃ and T₄ groups attained significantly (P<0.01) higher body weight gains compared to the T₁ and T₂ group quails. In the fourth week of experiment, body weight gain of all the groups were significantly (P<0.01) different with each other. The highest weight gain of 33.23 ± 0.23 g and lowest of 29.72 ± 0.23 g were observed in quails of T₂ and T₄ groups respectively. However in the fifth week, quails of T₄ group showed significant (P<0.01) increase in weight gain (27.87 ± 0.16 g) compared to the quails of other groups. During sixth week, the weight gain of T₃ and T₄ group quails were significantly higher compared to the weight gain of control and T₂ group Japanese quails with maximum (21.89 ± 0.02 g) in T₄ group.

During finisher period, significant (P<0.01) increase in weight gain was noted in Japanese quails of supplemented groups as compared to the T₁ (control) group.
Maximum (117.26 ± 0.16 g) weight gain was noted in quails of T₄ group which was statistically similar to that of T₃ group. Minimum (109.43 ± 0.60 g) and significantly (P<0.01) lower weight gain compared to the supplemented groups was noted in quails of control group. For overall period, the weight gains of quails of different treatment groups were significantly different amongst each other. Maximum weight gain (166.10 ±0.57 g) was noted in the T₃ group quails fed with 0.5 per cent of supplement, whereas minimum weight gain (157.01 ± 0.49 g) was observed in quails of control group.

Findings of the present investigation in both of the experiments regarding effect of nucleotide supplementation on weight gain revealed that maximum weight gain for overall period was noted in quails supplemented with 0.5 per cent nucleotide/kg of feed and weight gain was improved by all the levels of dietary nucleotide supplementation. These findings corroborated with those of Garcia et al. (2007) who found 1.6 per cent improvement in body weight gain of chicken supplemented 500 mg of nucleoforce / kg of feed. Shankar et al. (2012) reported same results in prawn. Jung and Batal (2012) found that inclusion of nucleotides increased weight gain of broilers under stress condition.

During the period of fast replication, nucleotides are required in larger amount. Therefore external supplementation of the nucleotide in the present investigation helped to meet the nucleotide requirement of fast growing Japanese quails. Probably, this may be the reason for the improved weight gain in Japanese quails of nucleotide supplemented groups.

4.1.3.1 Feed conversion ratio (Experiment I)

The data on feed conversion ratio (FCR) calculated as feed consumption per unit weight gain at weekly intervals as well as on over all basis for Japanese quails in the experiment I have been shown in Table 4.5 and Fig. 4.5.

In the first week, no significant difference in the feed conversion ratio was noted among different treatment groups. The average feed conversion ratios of Japanese quails of T₂ and T₃ groups in the second week were 2.12 ± 0.09 and 2.42 ± 0.11 respectively,
which were significantly (P<0.01) lower than the FCR of T₁ and T₄ groups. There was a significant difference in FCR of T₂ and T₃ groups during this period. During starter period, the FCR of Japanese quails of T₂ and T₃ groups were significantly (P<0.01) lower than the T₁ and T₄ groups. But, there was no significant difference in FCR between these two groups. Maximum FCR (2.50 ± 0.04) was noted in quails of control group. In third and fourth weeks, significantly (P<0.05) lower feed conversion values were observed in the groups supplemented with nucleotides compared to the control group. Minimum FCR was noted in the quails of T₂ group. During the fifth week, the FCR of T₂ and T₄ group quails were significantly (P<0.01) lower compared to the FCR of control group while FCR of T₃ group quails were statistically (P>0.05) similar to that of control group. Statistically (P>0.05) similar values for FCR were noted in all the groups during the sixth week with minimum value noted in T₂ group quails.

During the finisher period, FCR of supplemented group quails were significantly (P<0.01) lower than the control (T₁) group. FCR of T₂ and T₄ groups as well as T₃ and T₄ groups were statistically similar. Minimum FCR (4.79 ± 0.08) was noted in T₂ group quails, whereas maximum (5.46 ± 0.11) and significantly higher FCR was noted in Japanese quails of control group.

The overall mean values for feed conversion ratio of Japanese quails were minimum (3.99 ± 0.04) and significantly (P<0.01) lower in T₂ group and maximum (4.67 ± 0.06) and significantly (P<0.01) higher in T₁ group. The supplementation of nucleotide at all levels significantly (P<0.01) reduced FCR. Moreover, best results regarding FCR were also observed in the treatment group T₂ provided with 0.5 per cent nucleotide.

4.1.3.2 Feed conversion ratio (Experiment II)

The data on feed conversion ratio (FCR) of Japanese quails calculated at weekly intervals as well as on over all basis in the experiment II have been summarized in Table 4.6 and Fig. 4.6.
Table 4.5: Effect of nucleotide supplementation on feed conversion ratio of Japanese quails (Experiment I)

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Weeks</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>I</td>
</tr>
<tr>
<td>T₁</td>
<td>1.57 ± 0.05</td>
</tr>
<tr>
<td>T₂</td>
<td>1.53 ± 0.05</td>
</tr>
<tr>
<td>T₃</td>
<td>1.55 ± 0.05</td>
</tr>
<tr>
<td>T₄</td>
<td>1.54 ± 0.06</td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01
Fig. 4.5: Effect of nucleotide supplementation on feed conversion ratio of Japanese quails (Experiment I)
Table 4.6: Effect of nucleotide supplementation on feed conversion ratio of Japanese quails (Experiment II)

<table>
<thead>
<tr>
<th>Treatments</th>
<th>I*</th>
<th>II**</th>
<th>(I–II)**</th>
<th>III**</th>
<th>IV**</th>
<th>V**</th>
<th>VI**</th>
<th>(III - VI)**</th>
<th>(I–VI)**</th>
</tr>
</thead>
<tbody>
<tr>
<td>T1</td>
<td>1.55^a ± 0.04</td>
<td>2.22^a ± 0.00</td>
<td>1.95^a ± 0.02</td>
<td>2.35^a ± 0.02</td>
<td>4.63^b ± 0.03</td>
<td>7.73^a ± 0.13</td>
<td>11.61^a ± 0.22</td>
<td>5.73^a ± 0.05</td>
<td>4.58^a ± 0.04</td>
</tr>
<tr>
<td>T2</td>
<td>1.50^ab ± 0.01</td>
<td>1.97^b ± 0.01</td>
<td>1.79^b ± 0.01</td>
<td>2.13^b ± 0.03</td>
<td>4.20^d ± 0.03</td>
<td>7.67^a ± 0.06</td>
<td>10.98^b ± 0.27</td>
<td>5.44^b ± 0.03</td>
<td>4.36^b ± 0.02</td>
</tr>
<tr>
<td>T3</td>
<td>1.43^b ± 0.02</td>
<td>1.84^c ± 0.03</td>
<td>1.68^c ± 0.01</td>
<td>1.99^c ± 0.02</td>
<td>4.37^c ± 0.03</td>
<td>7.81^a ± 0.06</td>
<td>9.82^c ± 0.04</td>
<td>5.35^bc ± 0.03</td>
<td>4.26^c ± 0.02</td>
</tr>
<tr>
<td>T4</td>
<td>1.44^b ± 0.02</td>
<td>1.88^c ± 0.02</td>
<td>1.70^c ± 0.02</td>
<td>2.08^b ± 0.01</td>
<td>4.75^a ± 0.02</td>
<td>7.04^b ± 0.04</td>
<td>9.41^c ± 0.02</td>
<td>5.30^c ± 0.01</td>
<td>4.27^c ± 0.01</td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01
Fig. 4.6: Effect of nucleotide supplementation on feed conversion ratio of Japanese quails (Experiment II)
During the first week, the minimum FCR (1.43 ± 0.02) was observed in the quail chicks of T₃ group which was statistically similar to the FCR of T₂ and T₄ group quails and significantly (P<0.05) lower than the control group. However, the FCR of T₁ and T₂ groups were statistically similar. The average FCR of Japanese quails were significantly (P<0.01) lower in the treatment groups compared to the control in the second week. Best FCR (1.84 ± 0.03) for second week was observed in T₃ group quails which was statistically similar to that of T₄ group. FCR was worst (2.22 ± 0.00) in quails of T₁ (control) group. During starter period, minimum (1.68 ± 0.01) FCR was noted in the T₃ group quails which was statistically similar to the FCR of T₄ group. Maximum (1.95 ± 0.02) and significantly (P<0.01) higher FCR was noted in the quails of T₁ group. In the third and fourth weeks, FCR of supplemented group quails were significantly (P<0.01) lower than the control except the FCR of T₄ group in the fourth week which was significantly higher than other groups. In the fifth week, quails of T₄ group had minimum (7.04 ± 0.04) and significantly (P<0.01) lower FCR than other treatment groups. Quails of T₁, T₂ and T₃ groups had statistically similar FCR. During sixth week, the FCR of control group quails was maximum (11.61 ± 0.22) and significantly (P<0.01) higher than the nucleotide supplemented groups. Among the supplemented groups, best result (9.41 ± 0.02) was observed in T₄ group quails.

During finisher period, FCR of nucleotide supplemented groups were significantly (P<0.01) lower than the control and best result was observed in the T₄ group (5.30 ± 0.01) quails. The overall average FCR of control group quails was maximum (4.58 ± 0.04) and significantly (P<0.01) higher compared to the supplemented groups. Statistically similar FCR results were noted between the T₃ and T₄ groups during this period. Minimum FCR (4.26 ± 0.02) was noted in quails of T₃ group.

In the present investigation the results on FCR revealed that during starter, finisher and overall period it was better in Japanese quails provided nucleotide in the feed. The increased surface area of the intestinal mucosa of the quails supplemented with nucleotide may be the reason for better utilization of nutrients and therefore lower feed to
gain ratio. Similar trend for FCR were recorded by Garcia et al. (2007) who found 1.9% difference in feed to gain ratio of broilers supplemented with 500 mg of nucleotide/kg of feed. The results of Jung and Batal (2012) also supported the beneficial effect of nucleotide supplementation in the birds under stress condition. However, in contrast to the findings of present experiment, Pelicia et al. (2010) found no significant difference in the FCR of broilers supplemented with nucleotides in the feed. Similarly, Domeneghini et al. (2004) in their experiment found no significant difference on feed conversion ratio of piglets supplemented with glutamine and nucleotides. The reason may be the difference in level of nucleotide or species difference.

4.1.4.1 Performance index (Experiment I)

The data on performance index of Japanese quails of different treatment groups at weekly intervals as well as on over all basis in the experiment I have been summarized in Table 4.7 and Fig. 4.7.

During the first week of the trial, there was no significant difference noted in the performance index among different treatment groups of Japanese quails. In the second week and starter period, the performance index of T_2 and T_3 group Japanese quails were significantly (P<0.01) higher than the control and T_4 groups. Maximum and significantly higher performance index were noted in quails of T_2 group whereas minimum and significantly lower performance index were noted in quails of control group.

The performance index of different groups were statistically (P>0.05) similar in the third and fifth week. During the fourth week, the performance index of quails supplemented with nucleotide were significantly (P<0.01) higher than the quails of control (5.83 ± 0.29) group. Maximum (8.08 ± 0.21) and significantly higher performance index was noted in the quails of T2 group. During the sixth week, performance index of T_2 group was maximum (2.33 ± 0.11) and significantly (P<0.05) higher than the control group. However, T_3 and T_4 group quails showed insignificantly better performance index compared to the control group. Performance index of all the nucleotide supplemented group Japanese quails were statistically similar.
Table 4.7: Effect of nucleotide supplementation on performance index of Japanese quails (Experiment I)

<table>
<thead>
<tr>
<th>Treatments</th>
<th>I</th>
<th>II**</th>
<th>(I–II)** Starter</th>
<th>III</th>
<th>IV**</th>
<th>V</th>
<th>VI*</th>
<th>(III - VI)** Finisher</th>
<th>(I–VI)** Overall</th>
</tr>
</thead>
<tbody>
<tr>
<td>T₁</td>
<td>8.93 ± 0.28</td>
<td>9.04c ± 0.22</td>
<td>16.37c ± 0.43</td>
<td>11.19 ± 0.05</td>
<td>5.83c ± 0.08</td>
<td>4.64 ± 0.19</td>
<td>1.69b ± 0.03</td>
<td>20.59c ± 0.25</td>
<td>32.85c ± 0.54</td>
</tr>
<tr>
<td>T₂</td>
<td>10.01 ± 0.66</td>
<td>14.52a ± 0.58</td>
<td>23.93a ± 0.91</td>
<td>12.61 ± 0.20</td>
<td>8.08a ± 0.21</td>
<td>5.58 ± 0.04</td>
<td>2.33a ± 0.11</td>
<td>24.47a ± 0.37</td>
<td>40.93a ± 0.37</td>
</tr>
<tr>
<td>T₃</td>
<td>10.75 ± 0.39</td>
<td>11.34b ± 0.51</td>
<td>21.10b ± 0.93</td>
<td>12.10 ± 0.67</td>
<td>7.08b ± 0.30</td>
<td>4.95 ± 0.16</td>
<td>1.98ab ± 0.18</td>
<td>22.17b ± 0.45</td>
<td>36.75b ± 0.84</td>
</tr>
<tr>
<td>T₄</td>
<td>10.21 ± 0.66</td>
<td>9.75c ± 0.22</td>
<td>18.24c ± 0.17</td>
<td>11.10 ± 0.28</td>
<td>7.56ab ± 0.14</td>
<td>4.98 ± 0.35</td>
<td>2.09ab ± 0.13</td>
<td>22.20b ± 0.22</td>
<td>36.22b ± 0.37</td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01
Fig. 4.7: Effect of nucleotide supplementation on performance index of Japanese quails (Experiment I)
Table 4.8: Effect of nucleotide supplementation on performance index of Japanese quails (Experiment II)

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Weeks</th>
<th>I*</th>
<th>II**</th>
<th>(I–II)**</th>
<th>III**</th>
<th>IV**</th>
<th>V**</th>
<th>VI**</th>
<th>(III - VI)**</th>
<th>(I–VI)**</th>
</tr>
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<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>T&lt;sub&gt;1&lt;/sub&gt;</td>
<td>Starter</td>
<td>12.56&lt;sup&gt;bc&lt;/sup&gt; ± 0.41</td>
<td>12.67&lt;sup&gt;c&lt;/sup&gt; ± 0.03</td>
<td>24.47&lt;sup&gt;c&lt;/sup&gt; ± 0.31</td>
<td>15.14&lt;sup&gt;c&lt;/sup&gt; ± 0.28</td>
<td>6.75&lt;sup&gt;c&lt;/sup&gt; ± 0.04</td>
<td>3.20&lt;sup&gt;b&lt;/sup&gt; ± 0.12</td>
<td>1.54&lt;sup&gt;c&lt;/sup&gt; ± 0.04</td>
<td>19.11&lt;sup&gt;c&lt;/sup&gt; ± 0.24</td>
<td>34.27&lt;sup&gt;c&lt;/sup&gt; ± 0.32</td>
</tr>
<tr>
<td>T&lt;sub&gt;2&lt;/sub&gt;</td>
<td></td>
<td>12.03&lt;sup&gt;c&lt;/sup&gt; ± 0.09</td>
<td>14.86&lt;sup&gt;b&lt;/sup&gt; ± 0.29</td>
<td>26.43&lt;sup&gt;b&lt;/sup&gt; ± 0.21</td>
<td>16.69&lt;sup&gt;b&lt;/sup&gt; ± 0.35</td>
<td>7.92&lt;sup&gt;a&lt;/sup&gt; ± 0.11</td>
<td>3.31&lt;sup&gt;b&lt;/sup&gt; ± 0.06</td>
<td>1.69&lt;sup&gt;b&lt;/sup&gt; ± 0.07</td>
<td>20.73&lt;sup&gt;b&lt;/sup&gt; ± 0.16</td>
<td>36.70&lt;sup&gt;b&lt;/sup&gt; ± 0.16</td>
</tr>
<tr>
<td>T&lt;sub&gt;3&lt;/sub&gt;</td>
<td></td>
<td>13.46&lt;sup&gt;a&lt;/sup&gt; ± 0.21</td>
<td>16.25&lt;sup&gt;a&lt;/sup&gt; ± 0.44</td>
<td>29.24&lt;sup&gt;a&lt;/sup&gt; ± 0.41</td>
<td>18.94&lt;sup&gt;a&lt;/sup&gt; ± 0.34</td>
<td>7.46&lt;sup&gt;b&lt;/sup&gt; ± 0.10</td>
<td>3.18&lt;sup&gt;b&lt;/sup&gt; ± 0.05</td>
<td>2.22&lt;sup&gt;a&lt;/sup&gt; ± 0.01</td>
<td>21.89&lt;sup&gt;a&lt;/sup&gt; ± 0.18</td>
<td>38.95&lt;sup&gt;a&lt;/sup&gt; ± 0.36</td>
</tr>
<tr>
<td>T&lt;sub&gt;4&lt;/sub&gt;</td>
<td></td>
<td>13.04&lt;sup&gt;abc&lt;/sup&gt; ± 0.26</td>
<td>15.06&lt;sup&gt;b&lt;/sup&gt; ± 0.31</td>
<td>27.63&lt;sup&gt;b&lt;/sup&gt; ± 0.55</td>
<td>18.19&lt;sup&gt;a&lt;/sup&gt; ± 0.17</td>
<td>6.26&lt;sup&gt;d&lt;/sup&gt; ± 0.07</td>
<td>3.96&lt;sup&gt;a&lt;/sup&gt; ± 0.04</td>
<td>2.33&lt;sup&gt;a&lt;/sup&gt; ± 0.01</td>
<td>22.12&lt;sup&gt;a&lt;/sup&gt; ± 0.06</td>
<td>38.47&lt;sup&gt;a&lt;/sup&gt; ± 0.15</td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01
Fig. 4.8: Effect of nucleotide supplementation on performance index of Japanese quails (Experiment II)

Results and Discussion
In the finisher phase, the performance index of Japanese quails supplemented with nucleotides was significantly (P<0.01) higher than the control group. Maximum (24.47 ± 0.37) and significantly (P<0.01) higher and minimum (20.59 ± 0.25) and significantly lower performance index were observed in the quails of T₂ and T₁ groups respectively. However, the performance index of the T₃ and T₄ group quails were statistically similar. For the overall duration of experiment, the Japanese quails supplemented with nucleotides showed significantly (P<0.01) better performance index compared to the control (32.85 ± 0.54). Maximum (40.93 ± 0.37) and significantly higher performance index was noted in the T₂ group Japanese quails. T₃ and T₄ group quails showed statistically (P>0.05) similar performance index.

4.1.4.2 Performance index (Experiment II)

The data on performance index of Japanese quails calculated at weekly intervals as well as on over all basis in different groups during the experiment II have been summarized in Table 4.8 and Fig. 4.8.

During the first week of the experiment, maximum (13.46 ± 0.21) and minimum (12.03 ± 0.09) performance index were noticed in the Japanese quails of T₃ and T₂ groups respectively. Performance index of T₃ and T₄ groups, T₁ and T₄ groups as well as T₁ and T₂ groups were statistically (P>0.05) similar. In the second week, performance index of treatment group quails were significantly (P<0.01) better than the control (12.67 ± 0.03). Maximum and significantly higher performance index was noted in the T₃ group (16.25 ± 0.44). During starter period, Japanese quails of supplemented groups showed significantly (P<0.01) better performance index compared to the control group. Significantly higher and maximum (29.24 ± 0.41) and significantly lower and minimum (24.47 ± 0.31) performance index were observed in the T₃ and T₁ group Japanese quails, respectively.

In the third week, the quails of supplemented groups showed better and significant results compared to the control. The performance indexes of the quails were higher in the third week compared to the other weeks during the entire experiment. During the fourth week, the performance index of treatment and control groups were significantly (P<0.01) different among each other. Maximum and significantly higher and minimum performance index were noted in T₂ (7.92 ± 0.11)
and T₄ (6.26 ± 0.07) groups, respectively. However, the performance index of T₄ group (3.96 ± 0.04) was significantly higher in the fifth week compared to the other groups which were statistically similar among each other. In the sixth week, the groups T₃ and T₄ revealed better and significantly (P<0.01) higher performance index compared to the T₂ and T₁ group quails. Minimum performance index (1.54 ± 0.04) was noted in quails of T₁ group.

In the finisher phase, the performance indexes of supplemented groups were significantly higher than the control (19.11 ± 0.24) group. Among the supplemented groups, Japanese quails of T₄ group showed best performance index (22.12 ± 0.06) whereas lower performance index was observed in T₂ group (20.73 ± 0.16). The overall performance during the entire period was significantly (P<0.01) higher in the Japanese quails provided nucleotides compared to the control group. Maximum (38.95 ± 0.36) and significantly higher performance index was observed in T₃ group quails which was statistically similar to that of T₄ group, whereas minimum (34.27 ± 0.32) performance index was observed in the T₁ group quails.

From the results of both experiments, it is clearly revealed that the groups fed with nucleotide supplement showed better and significantly higher performance index compared to the control. Best performance index for overall period was noted in Japanese quails of 0.5 per cent nucleotide supplemented group. Effect of nucleotide supplementation on performance index of Japanese quails and other poultry species has not been studied. Therefore, literature pertaining to this aspect is scanty, rather more experimentation on other aspects of growth have been documented in the literature. The predominant mechanism involved in the improved performance is the role of nucleotide in better intestinal growth which increases the nutrient absorption and is thus responsible for better performance.

4.2.1 Nutrient utilization (Experiment I)

Data pertaining to the average nutrient utilization of different supplemented and control group in the experiment I are presented in Table 4.9 and depicted in Fig 4.9.

A) Dry matter

The study on dry matter utilization (per cent) in Japanese quails by nucleotide supplementation revealed the following results. The Japanese quails of supplemented
groups absorbed significantly (P<0.01) higher amount of dry matter compared to the T1 group (control) quails, which showed lowest (64.59 ± 0.51 per cent) absorption during the experiment. Among the supplemented groups maximum (68.77 ± 0.37 per cent) utilization was noted in the T2 group quails and minimum (67.36 ± 0.46 per cent) in T4 group. Dry matter digestibility of Japanese quails in T3 group was statistically (P>0.05) similar to both T2 and T4 groups.

B) Ether extract

The ether extract utilization in Japanese quails showed significant (P<0.01) difference among treatment groups. Maximum (76.69 ± 1.49 per cent) ether extract utilization was noted in the Japanese quails of T2 group, which was statistically (P>0.05) similar to the utilization of T3 group. The Japanese quails of control group showed significantly lower and minimum (68.90 ± 0.34 per cent) utilization of ether extract among the treatments.

C) Crude protein

Japanese quails of groups supplemented with nucleotides had significantly (P<0.01) better crude protein (per cent) utilization than the T1 group (68.99 ± 1.43 per cent). Japanese quails of T2 group (71.86 ± 0.13 per cent) absorbed highest percentage of crude protein among all groups of the experiment. At the same time no significant difference (P>0.05) was observed among the Japanese quails of T3 and T4 groups.

D) Total carbohydrates

Study of total carbohydrate utilization in Japanese quails indicated that, there was a significant (P<0.05) impact of nucleotide supplementation. Maximum (43.65 ± 0.16 per cent) utilization of total carbohydrate was observed in Japanese quails of T2 group, while significantly (P<0.05) lower and minimum (41.23 ± 0.25 per cent) utilization was noted in T1 group. Utilization of total carbohydrates was statistically (P>0.05) similar among the nucleotide supplemented Japanese quails.

Results and Discussion ............

Results and Discussion ............
### Table 4.9: Effect of nucleotide supplementation on nutrient utilization (%) of Japanese quails (Experiment I)

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Parameters</th>
<th>Dry matter**</th>
<th>Ether extract**</th>
<th>Crude protein*</th>
<th>Total carbohydrates**</th>
</tr>
</thead>
<tbody>
<tr>
<td>T&lt;sub&gt;1&lt;/sub&gt;</td>
<td>64.59&lt;sup&gt;c&lt;/sup&gt; ± 0.51</td>
<td>68.90&lt;sup&gt;c&lt;/sup&gt; ± 0.34</td>
<td>68.99&lt;sup&gt;b&lt;/sup&gt; ± 1.43</td>
<td>41.23&lt;sup&gt;b&lt;/sup&gt; ± 0.25</td>
<td></td>
</tr>
<tr>
<td>T&lt;sub&gt;2&lt;/sub&gt;</td>
<td>68.77&lt;sup&gt;a&lt;/sup&gt; ± 0.37</td>
<td>76.69&lt;sup&gt;a&lt;/sup&gt; ± 1.49</td>
<td>71.86&lt;sup&gt;a&lt;/sup&gt; ± 0.13</td>
<td>43.65&lt;sup&gt;a&lt;/sup&gt; ± 0.16</td>
<td></td>
</tr>
<tr>
<td>T&lt;sub&gt;3&lt;/sub&gt;</td>
<td>68.05&lt;sup&gt;ab&lt;/sup&gt; ± 0.30</td>
<td>75.76&lt;sup&gt;a&lt;/sup&gt; ± 0.59</td>
<td>70.17&lt;sup&gt;b&lt;/sup&gt; ± 0.71</td>
<td>43.23&lt;sup&gt;a&lt;/sup&gt; ± 0.38</td>
<td></td>
</tr>
<tr>
<td>T&lt;sub&gt;4&lt;/sub&gt;</td>
<td>67.36&lt;sup&gt;b&lt;/sup&gt; ± 0.46</td>
<td>71.48&lt;sup&gt;b&lt;/sup&gt; ± 1.04</td>
<td>69.38&lt;sup&gt;b&lt;/sup&gt; ± 0.65</td>
<td>42.89&lt;sup&gt;a&lt;/sup&gt; ± 0.49</td>
<td></td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01

### Table 4.10: Effect of nucleotide supplementation on nutrient utilization (%) of Japanese quails (Experiment II)

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Parameters</th>
<th>Dry matter*</th>
<th>Ether extract</th>
<th>Crude protein**</th>
<th>Total carbohydrates*</th>
</tr>
</thead>
<tbody>
<tr>
<td>T&lt;sub&gt;1&lt;/sub&gt;</td>
<td>63.30&lt;sup&gt;b&lt;/sup&gt; ± 0.65</td>
<td>67.82 ± 0.53</td>
<td>68.24&lt;sup&gt;b&lt;/sup&gt; ± 0.95</td>
<td>40.87&lt;sup&gt;b&lt;/sup&gt; ± 0.28</td>
<td></td>
</tr>
<tr>
<td>T&lt;sub&gt;2&lt;/sub&gt;</td>
<td>64.84&lt;sup&gt;a&lt;/sup&gt; ± 0.74</td>
<td>68.01 ± 0.72</td>
<td>68.93&lt;sup&gt;b&lt;/sup&gt; ± 0.93</td>
<td>44.06&lt;sup&gt;a&lt;/sup&gt; ± 0.59</td>
<td></td>
</tr>
<tr>
<td>T&lt;sub&gt;3&lt;/sub&gt;</td>
<td>65.82&lt;sup&gt;a&lt;/sup&gt; ± 1.13</td>
<td>68.58 ± 0.80</td>
<td>70.61&lt;sup&gt;a&lt;/sup&gt; ± 0.59</td>
<td>42.98&lt;sup&gt;a&lt;/sup&gt; ± 0.60</td>
<td></td>
</tr>
<tr>
<td>T&lt;sub&gt;4&lt;/sub&gt;</td>
<td>66.18&lt;sup&gt;a&lt;/sup&gt; ± 0.60</td>
<td>68.88 ± 0.70</td>
<td>71.28&lt;sup&gt;a&lt;/sup&gt; ± 0.58</td>
<td>42.56&lt;sup&gt;ab&lt;/sup&gt; ± 0.67</td>
<td></td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01
Fig. 4.9: Effect of nucleotide supplementation on nutrient utilization of Japanese quails (Experiment I)

Fig. 4.10: Effect of nucleotide supplementation on nutrient utilization of Japanese quails (Experiment II)
4.2.2 Nutrient utilization (Experiment II)

Data pertaining to the average nutrient utilization of supplemented and control groups in the experiment II are presented in Table 4.10 and depicted in Fig. 4.10.

A) Dry matter

The results on nutrient utilization revealed a positive impact of nucleotide supplementation on dry matter utilization of Japanese quails. Japanese quails of T₁ group showed significantly (P<0.05) lower and minimum (63.30 ± 0.65 per cent) utilization of dry matter compared to the supplemented groups. Maximum (66.18 ± 0.60 per cent) dry matter utilization was noted in the Japanese quails of T₄ group which was statistically (P>0.05) similar to the other treatment groups supplemented with nucleotides.

B) Ether extract

The ether extract utilization in Japanese quails did not show any significant (P>0.05) impact of nucleotide supplementation. The highest mean (68.88 ± 0.70 per cent) ether extract utilization was recorded in Japanese quails of T₄ group.

C) Crude protein

Japanese quails of groups T₃ and T₄ had significantly (P<0.01) more crude protein utilization than the T₁ and T₂ group quails. The highest percentage (71.28 ± 0.58 per cent) of crude protein utilization was noted in the Japanese quails of T₄ group which was statistically (P>0.05) similar to the T₃ group. The Japanese quails of T₁ group utilised lowest (68.24 ± 0.95 per cent) percentage of crude protein among all treatment groups. At the same time no significant difference (P>0.05) in crude protein utilization was observed among the Japanese quails of T₁ and T₂ groups.

D) Total carbohydrates

The total carbohydrate utilization showed a positive impact of nucleotide supplementation. Maximum (44.06 ± 0.59 per cent) absorption was noted in the Japanese quails of T₂ group which was statistically (P>0.05) similar to other nucleotide supplemented groups. Significantly lower and minimum (40.87 ± 0.28 per cent) carbohydrate utilization was observed in the T₁ (control) group quails. However,
absorption of total carbohydrates of Japanese quails in T4 group was statistically similar to both control and the groups supplemented with nucleotides.

Effect of nucleotide supplementation on nutrient utilization of poultry and animals has not been studied. Therefore, literature pertaining to this aspect is scanty, rather more experimentation on other aspects have been documented in the literature. The increased surface area of the intestinal mucosa in the birds supplemented with nucleotides may be responsible for the better nutrient utilization in Japanese quails of supplemented groups (Hosseini, 2011a; Jung and Batal, 2012 and Puig et al, 2007).

4.3 Carcass traits

In Japanese quails carcass evaluation is of prime importance along with economic traits. The ultimate objective of Japanese quail broiler production is to obtain good quality edible meat, which has quality composition for human consumption. Carcass traits were studied in the first trial of present investigation to discern the effect of nucleotide enriched yeast supplementation on dressed yield (with or without giblet), cut-up parts, organ weights and processing losses of Japanese quail broilers, which were considered for the evaluation. Two birds from each replicate (6 birds/treatment) were randomly selected and slaughtered at the end of feeding trial i.e. on 42nd day of trial to study carcass traits. Carcass traits in the present study include dressed yield without or with giblet, cut up parts (thighs, drumstick, breast, back, neck and wings), organ weights (heart, liver and gizzard) and processing losses (blood, feather, head and shanks).

4.3.1 Dressed yield

The effects of nucleotide supplementation on dressed yield (with or without giblet) of Japanese quails have been presented in Table 4.11 and Fig. 4.11.

There was no significant difference in the dressed yield with giblet among the treatment groups i.e. the dressed yield with giblet was not significantly (P<0.05) affected by nucleotide supplementation. The dressed yield without giblet of control group quails (69.90 ± 1.12 per cent) was significantly (P<0.05) higher than the T2 and T4 group quails. However, that of T3 group (69.33 ± 0.20 per cent) was similar to the control group. Similar findings were reported by Pelicia et al. (2010) who found no
significant improvement in the dressed yield of broilers supplemented with nucleotides. Chumpawadee (2009), Fathi et al. (2012) and Abdelrahman (2013) also found no significant difference in the dressing percentage of broilers supplemented with yeast. However, Paryad and Mahmoudi (2008) found significant (P<0.05) increase in the dressing yield of broilers supplemented with 1.5 and 2.0 per cent of yeast culture in the feed. In contrast to better FCR and performance index carcass yield was not improved due to nucleotide supplementation which may be attributed to the high proportion of the intestine and visceral organs in nucleotide supplemented quails.

4.3.2 Cut-up parts

The effect of nucleotide supplementation on the cut-up parts viz. drumsticks, thighs, breast, back, wings and neck have been presented in Table 4.12 and Fig. 4.12.

Thigh weights of Japanese quails in T3 group (10.32 ± 0.18 per cent) were numerically (P>0.05) increased compared to the quails of control group (9.70 ± 0.27 per cent) and significantly (P<0.05) increased compared to the T2 and T4 groups. However, there was no significant difference in the percentage of thigh weight between the T1, T2 and T4 groups of Japanese quails.

The percentage of breast weight as per cent live weight was statistically similar among different treatment group quails with maximum breast weight (24.21 ± 0.57 per cent) noted in the quails of T3 group and minimum (22.73 ± 0.51 per cent) in quails of T2 group. Similarly, there was insignificant (P>0.05) difference in the percentage of drumstick and neck weights between the treatment groups. Numerically, maximum drumstick (6.29 ± 0.35 per cent) and neck (3.61 ± 0.13 per cent) weights were observed in quails of T3 group. There was significant (P<0.05) increase in wing weights of Japanese quails of T3 group compared to the control and the other treatment groups. There was no significant (P>0.05) difference in wing weights among the quails of control, T2 and T4 groups. The Japanese quails of T1 group showed significant increase in the back weight (22.73 ± 0.53 per cent) compared to the quails of T3 and T4 groups.
Table 4.11: Effect of nucleotide supplementation on carcass yield (% live weight) of Japanese quails

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Carcass yield</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Dressed yield with giblet</td>
<td>Dressed yield without giblet*</td>
<td></td>
</tr>
<tr>
<td>T&lt;sub&gt;1&lt;/sub&gt;</td>
<td>75.30 ± 0.82</td>
<td>69.90&lt;sup&gt;a&lt;/sup&gt; ± 1.12</td>
<td></td>
</tr>
<tr>
<td>T&lt;sub&gt;2&lt;/sub&gt;</td>
<td>73.38 ± 0.77</td>
<td>66.67&lt;sup&gt;b&lt;/sup&gt; ± 0.84</td>
<td></td>
</tr>
<tr>
<td>T&lt;sub&gt;3&lt;/sub&gt;</td>
<td>75.33 ± 0.08</td>
<td>69.33&lt;sup&gt;a&lt;/sup&gt; ± 0.20</td>
<td></td>
</tr>
<tr>
<td>T&lt;sub&gt;4&lt;/sub&gt;</td>
<td>73.28 ± 0.37</td>
<td>66.67&lt;sup&gt;b&lt;/sup&gt; ± 0.68</td>
<td></td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01

Table 4.12: Effect of nucleotide supplementation on cut up parts (% live weight) of Japanese quails

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Cut up parts</th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Thigh*</td>
<td>Breast</td>
<td>Drumstick</td>
<td>Back*</td>
<td>Neck</td>
</tr>
<tr>
<td>T&lt;sub&gt;1&lt;/sub&gt;</td>
<td>9.70&lt;sup&gt;ab&lt;/sup&gt; ± 0.27</td>
<td>23.31 ± 0.14</td>
<td>5.64 ± 0.21</td>
<td>22.79&lt;sup&gt;a&lt;/sup&gt; ± 0.53</td>
<td>3.01 ± 0.13</td>
</tr>
<tr>
<td>T&lt;sub&gt;2&lt;/sub&gt;</td>
<td>9.36&lt;sup&gt;b&lt;/sup&gt; ± 0.25</td>
<td>22.73 ± 0.51</td>
<td>6.09 ± 0.23</td>
<td>20.95&lt;sup&gt;ab&lt;/sup&gt; ± 0.61</td>
<td>3.09 ± 0.13</td>
</tr>
<tr>
<td>T&lt;sub&gt;3&lt;/sub&gt;</td>
<td>10.32&lt;sup&gt;a&lt;/sup&gt; ± 0.18</td>
<td>24.21 ± 0.57</td>
<td>6.29 ± 0.35</td>
<td>19.41&lt;sup&gt;b&lt;/sup&gt; ± 0.44</td>
<td>3.61 ± 0.13</td>
</tr>
<tr>
<td>T&lt;sub&gt;4&lt;/sub&gt;</td>
<td>9.31&lt;sup&gt;b&lt;/sup&gt; ± 0.08</td>
<td>23.82 ± 0.41</td>
<td>6.01 ± 0.15</td>
<td>19.35&lt;sup&gt;b&lt;/sup&gt; ± 0.69</td>
<td>3.43 ± 0.27</td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01
Fig. 4.11: Effect of nucleotide supplementation on carcass yield (% live weight) of Japanese quails

Fig. 4.12: Effect of nucleotide supplementation on cut up parts (% live weight) of Japanese quails
However, there was no significant difference between control and $T_2$ (20.95 ± 0.61 per cent) group. Minimum back weight (19.35 ± 0.69) was observed in the quails of $T_4$ group which was statistically similar to that of $T_3$ group.

The results of the present investigation revealed that the weight of various cut up parts were mostly comparable in nucleotide supplemented as well as control group which indicates that there was no effect of nucleotide supplementation on the weights of cut-up parts of Japanese quail. The results of the present study were supported by Pelicia et al. (2010) who found no significant improvement in the cut-up parts yield in broilers supplemented with nucleotides. In contrast, Paryad and Mahmoudi (2008) found significant ($P<0.05$) increase in the breast and leg muscles yield in broilers supplemented with 1.5 and 2.0 per cent of yeast. Fathi et al. (2012) also found significant increase in the breast muscles yield in broilers supplemented with yeast.

### 4.3.3 Organ weights

The effect of nucleotide supplementation on organ weights viz. heart, liver and gizzard weights of the Japanese quails have been shown in Table 4.13 and Fig. 4.13.

There was no significant ($P<0.05$) difference in the heart weights amongst the Japanese quails of different groups. However, significant increase in liver and gizzard weight were shown by the quails of groups fed with nucleotide supplement. Maximum and significantly (3.11 ± 0.05 per cent) higher liver weights were noticed in the birds of $T_2$ and $T_4$ groups while minimum liver weight (2.07 ± 0.18 per cent) was noticed in control group quails. Whereas, maximum gizzard weight (2.81 ± 0.03 per cent) was noticed in the quails of $T_3$ group, minimum and significantly ($P<0.05$) lower gizzard weight (2.52 ± 0.06 per cent) was noted for control group quails.

As regards liver weight, supplementation of nucleotide at the concentration of 0.5 and 1.5 per cent in the feed showed significant ($P<0.05$) impact on Japanese quails compared to 0.1 per cent concentration in feed and the control groups. Similarly, the gizzard weight was significantly ($P<0.05$) increased in Japanese quails of $T_2$ and $T_3$ groups supplemented with nucleotides compared to the birds of control and $T_4$ groups.

Thus there was significant ($P<0.05$) positive impact of nucleotide supplementation on gizzard and liver percentage. The results are in accordance with
Table 4.13: Effect of nucleotide supplementation on organ weight (% live weight) of Japanese quails

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Heart</th>
<th>Liver**</th>
<th>Gizzard**</th>
</tr>
</thead>
<tbody>
<tr>
<td>T1</td>
<td>0.81 ± 0.04</td>
<td>2.07\textsuperscript{b} ± 0.18</td>
<td>2.52\textsuperscript{b} ± 0.06</td>
</tr>
<tr>
<td>T2</td>
<td>0.80 ± 0.01</td>
<td>3.11\textsuperscript{a} ± 0.05</td>
<td>2.79\textsuperscript{a} ± 0.04</td>
</tr>
<tr>
<td>T3</td>
<td>0.82 ± 0.01</td>
<td>2.37\textsuperscript{b} ± 0.14</td>
<td>2.81\textsuperscript{a} ± 0.03</td>
</tr>
<tr>
<td>T4</td>
<td>0.78 ± 0.01</td>
<td>3.11\textsuperscript{a} ± 0.01</td>
<td>2.72\textsuperscript{a} ± 0.04</td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01

Fig. 4.13: Effect of nucleotide supplementation on organ weight (% live weight) of Japanese quails
Hosseini (2011b) who also found increase in gizzard and liver per cent in broilers supplemented with yeast. The reason for this increase may be the improvement in digestion and attraction of nutrients by nucleotides for which weight and size of digestive organs might increase.

In contrast, Chumpawadee et al. (2009) found that inclusion of cassava yeast had no significant influence on the liver and gizzard weights of broilers. Similarly, Shareef and Al-Dabbagh (2009) also found no significant difference in the organ weights of broilers supplemented with Saccharomyces cerevisiae yeast.

4.3.4 Processing losses

The effect of nucleotide supplementation at different levels on processing losses viz. blood loss, feather loss, head and shank of the Japanese quails have been shown in Table 4.14.

Table 4.14: Effect of nucleotide supplementation on processing losses (% live weight) of Japanese quails

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Blood loss</th>
<th>Feather loss</th>
<th>Head</th>
<th>Shank</th>
</tr>
</thead>
<tbody>
<tr>
<td>T\textsubscript{1}</td>
<td>3.90 ± 0.04</td>
<td>8.15 ± 0.12</td>
<td>5.51 ± 0.28</td>
<td>1.86 ± 0.05</td>
</tr>
<tr>
<td>T\textsubscript{2}</td>
<td>3.89 ± 0.05</td>
<td>8.19 ± 0.09</td>
<td>5.33 ± 0.11</td>
<td>1.82 ± 0.02</td>
</tr>
<tr>
<td>T\textsubscript{3}</td>
<td>3.85 ± 0.01</td>
<td>8.27 ± 0.15</td>
<td>5.26 ± 0.00</td>
<td>1.88 ± 0.04</td>
</tr>
<tr>
<td>T\textsubscript{4}</td>
<td>3.81 ± 0.05</td>
<td>8.36 ± 0.08</td>
<td>5.25 ± 0.12</td>
<td>1.79 ± 0.00</td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01

The overall mean value for blood loss were recorded as 3.90 ± 0.04, 3.89 ± 0.05, 3.85 ± 0.01 and 3.81 ± 0.05 per cent, for feather loss 8.15 ± 0.12, 8.19 ± 0.09, 8.27 ± 0.15 and 8.36 ± 0.08 per cent, for head loss 5.51 ± 0.28, 5.33 ± 0.11, 5.26 ± 0.00 and 5.25 ± 0.12 per cent and for shank loss 1.86 ± 0.05, 1.82 ± 0.02, 1.88 ± 0.04 and 1.79 ± 0.00 per cent from groups T\textsubscript{1}-T\textsubscript{4}, respectively.

The data of the present investigation indicated that the processing losses were not affected due to nucleotide supplementation. Effects of nucleotide on processing...
losses of Japanese quails or broilers have not been studied and the literature pertaining to this aspect is scanty. The above results regarding the processing losses revealed a numerical variation between the groups provided supplemental nucleotide. Therefore, it may be concluded that the supplementation of nucleotide did not materially affect the processing losses of Japanese quails.

4.4 Meat composition

The proximate composition with respect to moisture, crude protein, ether extract and total ash contents was estimated in thigh and breast muscle samples. The results of same are presented in Table 4.15 and Fig. 4.14 and Table 4.16 and Fig. 4.15 respectively.

4.4.1 Thigh muscles

The moisture, crude protein and total ash content of thigh revealed non-significant impact of nucleotide supplementation. Though, the crude protein and ash content of the thigh muscles of supplemented group Japanese quails were numerically higher than the quails of control group. As regards fat content, highest fat content (3.64 ± 0.11 per cent) was found in thigh muscles of Japanese quails of group T₁ which was significantly (P<0.01) decreased in supplemented groups with the level of nucleotides. Lowest fat content of thigh muscles (2.06 ± 0.06 per cent) was recorded in the Japanese quails of group T₄ which was statistically (P>0.05) similar to that of T₃ group.

4.4.2 Breast muscles

The moisture, crude protein and total ash content of breast revealed non-significant impact of nucleotide supplementation. Though, the crude protein and ash content of the breast muscles of supplemented group Japanese quails were numerically higher than the quails of control group. As regards fat content, highest fat content (3.29 ± 0.17 per cent) was found in breast muscles of Japanese quails of group T₁ which decreased significantly (P<0.05) in nucleotide supplemented groups. Lowest fat content of breast muscles (2.42 ± 0.12 per cent) was recorded in the Japanese quails of group T₃ which was statistically (P>0.05) similar to the breast fat content of T₃ and T₄ groups Japanese quails.
Table 4.15: Effect of nucleotide supplement on proximate composition (%) of thigh meat of Japanese quails

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Thigh meat composition</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Moisture</td>
</tr>
<tr>
<td><strong>T&lt;sub&gt;1&lt;/sub&gt;</strong></td>
<td>71.17 ± 0.36</td>
</tr>
<tr>
<td><strong>T&lt;sub&gt;2&lt;/sub&gt;</strong></td>
<td>71.76 ± 0.48</td>
</tr>
<tr>
<td><strong>T&lt;sub&gt;3&lt;/sub&gt;</strong></td>
<td>72.41 ± 0.23</td>
</tr>
<tr>
<td><strong>T&lt;sub&gt;4&lt;/sub&gt;</strong></td>
<td>71.16 ± 0.66</td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01

Table 4.16: Effect of nucleotide supplement on proximate composition (%) of breast meat of Japanese quails

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Breast meat composition</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Moisture</td>
</tr>
<tr>
<td><strong>T&lt;sub&gt;1&lt;/sub&gt;</strong></td>
<td>70.28 ± 0.51</td>
</tr>
<tr>
<td><strong>T&lt;sub&gt;2&lt;/sub&gt;</strong></td>
<td>71.29 ± 0.14</td>
</tr>
<tr>
<td><strong>T&lt;sub&gt;3&lt;/sub&gt;</strong></td>
<td>71.30 ± 0.03</td>
</tr>
<tr>
<td><strong>T&lt;sub&gt;4&lt;/sub&gt;</strong></td>
<td>71.32 ± 0.28</td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01
Fig. 4.14: Effect of nucleotide supplement on proximate composition (%) of thigh meat of Japanese quails

Fig. 4.15: Effect of nucleotide supplement on proximate composition (%) of breast meat of Japanese quails
From the results of the experiment, it is clear that the proximate composition with respect to moisture, crude protein and total ash contents of thigh and breast muscles were not affected due to nucleotide supplementation while fat content of thigh and breast muscles were significantly (P<0.05) decreased in the Japanese quails of nucleotide supplemented groups compared to control group. The results of the present experiment are supported by Paryad and Mahmoudi (2008) who found significant (P<0.05) reduction in the ether extract per cent in the breast muscles of broilers supplemented with 2.0 per cent yeast in the feed. In contrast, Chiofalo et al. (2011) found significant increase in the fat content of breast muscles in nucleotide supplemented broilers. However, they noticed no significant difference in the protein content of breast muscles of nucleotide supplemented broilers.

Increased protein content of muscles may be due to increase in net protein retention in nucleotide supplemented groups of Japanese quails. Reduction of ether extract content of thigh and breast muscles in nucleotide supplemented groups is an important finding of present investigation for lean meat production.

4.5 Haematological parameters

In the present investigation effects of supplementation of nucleotide as yeast extract on certain haematological parameters were considered at 42\textsuperscript{nd} day of trial. The results obtained have been presented in Table 4.17 and Fig. 4.16.

4.5.1 Total erythrocyte count (TEC)

Supplementation of nucleotide in feed of Japanese quails produced a marked effect on the total erythrocyte count (×10\textsuperscript{6} / µl). The mean TEC values of the Japanese quails of supplemented groups were significantly (P<0.01) higher than the control group quails which showed minimum (2.68 ± 0.40 ×10\textsuperscript{6} / µl) TEC value at the end of the experiment. TEC values were statistically (P>0.05) similar among the treatment groups with maximum value noted in the Japanese quails of T\textsubscript{4} group.

There was a linear increase in the total red blood cells in the Japanese quails with increase in the concentration of nucleotide supplementation. Similar results were also reported by Shareef and Al-Dabbagh (2009) and Hosseini (2011c) in broilers supplemented with yeast (*Sacchromyces cerevisiae*). Similarly Krol (2011) noticed
significant increase of total RBC in calves supplemented with yeast nucleotides. Shankar (2012) also noted that dietary nucleotides significantly increased the total haemocyte count in fresh water prawns.

In contrast, Choudhury et al. (2005) could not found any significant difference in the total RBC of Rohu juveniles supplemented with RNA nucleotides. Gheisari and Kholeghipour (2006), Prayad and Mahmoudi (2008) and Mansour et al. (2011) had also not found any significant increase in TEC with yeast supplementation in broilers.

**4.5.2 Total leukocyte count (TLC)**

Total leukocyte count in the present investigation revealed a significant (P<0.05) impact of nucleotide supplementation with maximum value in treatment group T₄ (21.20 ± 0.12 ×10³/µl) which was statistically (P>0.05) similar to group T₃, while minimum value was noted in group T₁ (19.32 ± 0.12 ×10³/µl). TLC value in T₂ group quails was significantly (P<0.05) lower than the T₄ and T₃ groups but higher than the TLC value of control group.

The results are in agreement with the findings of Choudhury et al. (2005) who reported maximum increase in the total WBC of rohu juveniles supplemented with 0.4 per cent of RNA nucleotides. Similarly, Prayad and Mahmoudi (2008), Shareef and Al-Dabbagh (2009) and Hosseini (2011c) also reported significant increase in the total leukocytes with yeast supplementation in broilers.

However, Gheisari and Kholeghipour (2006) found that supplementation of yeast did not increase the total WBC in broilers. Reduction in total WBC was reported by Mansour et al. (2011) in broilers supplemented with yeast culture.

**4.5.3 Packed cell volume (PCV)**

Nucleotide supplementation in Japanese quails revealed a marked positive effect on the packed cell volume. PCV values of treatment groups were significantly (P<0.01) higher than the control. The maximum (36.06 ± 0.20 %) and minimum (32.31 ± 0.22 %) PCV values were observed in the T₄ and T₁ groups, respectively. However, there was no significant difference in the PCV values among the supplemented groups.
Table 4.17: Effect of nucleotide supplementation on haematological profile of Japanese quails

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Parameters</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>TEC** (10^6/µl)</td>
</tr>
<tr>
<td>T_1</td>
<td>2.68 ± 0.40</td>
</tr>
<tr>
<td>T_2</td>
<td>3.05 ± 0.21</td>
</tr>
<tr>
<td>T_3</td>
<td>3.15 ± 0.23</td>
</tr>
<tr>
<td>T_4</td>
<td>3.34 ± 0.29</td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01
Fig. 4.16: Effect of nucleotide supplementation on haematological profile of Japanese quails
The results of the experiment are in corroboration with Shareef and Al-Dabbagh (2009) who found best results of PCV with 2 per cent inclusion of yeast in broilers. Hosseini (2011c) also noted that dietary inclusion of yeast increased the PCV in broilers.

However, Gheisari and Kholeghipour (2006) and Mansour et al. (2011) did not find any significant effect of nucleotide supplementation on PCV in broilers. Similarly, Krol (2011) also found non-significant effect of nucleotide supplementation on PCV of calves.

4.5.4 Haemoglobin (Hb)

Haemoglobin (g/dl) in the present investigation revealed a significant (P<0.05) positive impact of nucleotide supplementation in Japanese quails. The maximum (12.92 ± 0.20 g/dl) concentration was observed in the Japanese quails of group T₄ while minimum (10.13 ± 0.59 g/dl) in T₁ group. The haemoglobin level among the supplemented groups were statistically (P>0.05) similar.

The results of the present study are in agreement with those of Gheisari and Kholeghipour (2006), Shareef and Al-Dabbagh (2009) and Hosseini (2011c) who found significant increase in haemoglobin due to supplementation of yeast in broilers. In contrast, Choudhury et al. (2005) observed no significant effect on haemoglobin in rohu juveniles supplemented with RNA nucleotides. Mansour (2011) found no significant effect of nucleotide supplementation on the haemoglobin level in broilers. Similarly, Krol (2011) did not find any significant difference in Hb of calves due to supplementation of yeast nucleotides.

4.5.5. Mean corpuscular volume (MCV)

The values of mean corpuscular volume (fl) in Japanese quails of different treatment groups on 42nd day of the experiment did not differ significantly. However, numerically higher (121.18 ± 7.14 fl) value of MCV was noted in control group.

The results are in contrast with the findings of Shareef and Al-Dabbagh (2009) and Hosseini (2011c) who found increased MCV in broilers supplemented with yeast.
4.5.6 Mean corpuscular haemoglobin (MCH)

Mean corpuscular haemoglobin in Japanese quails was not affected by nucleotide supplementation. Maximum (39.76 ± 0.36 pg) MCH value was noted in the Japanese quails of T₂ group, while minimum (38.01 ± 2.77 pg) was noted in T₁ group quails.

The results are in contrast with the findings of Shareef and Al-Dabbagh (2009) and Hosseini (2011c) who found increased MCH in broilers supplemented with yeast.

4.5.7 Mean corpuscular haemoglobin concentration (MCHC)

MCHC values were markedly affected by the nucleotide supplementation. Maximum value of MCHC (35.83 ± 0.90 %) was observed in the Japanese quails of T₄ group which was significantly (P<0.01) higher than control group. MCHC values were statistically (P>0.05) similar among the supplemented groups. The quails of control group showed significantly lower and minimum (31.33 ± 0.76 %) value of MCHC compared to the other treatment groups at the end of the experiment.

The present experiment indicates that the inclusion of nucleotides significantly increase the value of MCHC in Japanese quails. The results are in agreement with the observations of Shareef and Al-Dabbagh (2009) and Hosseini (2011c) who found increase in MCHC of broilers supplemented with yeast.

The increased villous surface area of intestine in the Japanese quails supplemented with nucleotides may maximize the absorption of iron ions and other trace minerals and increase in the availability in the blood which may simultaneously increase the haemoglobin and erythrocytes synthesis and other haematological parameters.

The haemoglobin content in the blood and oxygen consumption increases when birds are under stress. Under such conditions, there is an increase in release of immature RBCs from the haemopoietic organs, which in turn elevate haemoglobin concentration in blood. The increased haemoglobin content in the Japanese quails along with high level of MCH and MCHC clearly indicates that the circulating RBCs are mature. Hence, it clearly indicates that the nucleotide supplementation reduce stress condition in birds.
4.5.8 Differential leukocyte count

The data pertaining to the effect of nucleotide supplementation on differential leukocyte count (DLC) among different treatment groups of Japanese quails at 42\textsuperscript{nd} day of trial are presented in Table 4.18.

4.5.8.1 Heterophil count

Supplementation of nucleotide showed a non-significant (P>0.05) impact on heterophil percentage in Japanese quails. Maximum heterophil (23.00 ± 0.29) percentage was noted in Japanese quails of control group whereas, minimum (21.00 ± 0.29 per cent) heterophil percentage was noticed in Japanese quails of T\textsubscript{4} group.

4.5.8.2 Basophils and Eosinophils count

In the present investigation, there was no significant effect of nucleotide supplementation on basophil and eosinophil percentage of Japanese quails.

4.5.8.3 Monocyte count

Nucleotide supplementation had no significant impact on the percentage of monocyte in the Japanese quails. Maximum (2.83 ± 0.17 per cent) monocyte percentage was noticed in Japanese quails of T\textsubscript{3} group while minimum (2.33 ± 0.17) in quails of T\textsubscript{1} group.

4.5.8.4 Lymphocyte count

The percentage of lymphocytes was statistically (P>0.05) similar among quails of different treatment groups. However, lymphocyte percentage was numerically higher in Japanese quails of supplemented groups compared to the quails of control (69.17 ± 1.20 per cent) group.

4.5.8.5 Heterophil – Lymphocyte ratio (H/L ratio)

H/L ratio exhibited non–significantly (P>0.05) similar results in all the treatment groups. However, numerically maximum (0.33 ± 0.01) value was observed in T\textsubscript{1} group quails and minimum (0.30 ± 0.1) in quails of T\textsubscript{4} group.
Table 4.18: Effect of nucleotide supplementation on differential leukocyte count of Japanese quails

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Parameters</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Heterophils (%)</td>
</tr>
<tr>
<td><strong>T&lt;sub&gt;1&lt;/sub&gt;</strong></td>
<td>23.00 ± 0.29</td>
</tr>
<tr>
<td><strong>T&lt;sub&gt;2&lt;/sub&gt;</strong></td>
<td>21.83 ± 0.44</td>
</tr>
<tr>
<td><strong>T&lt;sub&gt;3&lt;/sub&gt;</strong></td>
<td>21.00 ± 0.76</td>
</tr>
<tr>
<td><strong>T&lt;sub&gt;4&lt;/sub&gt;</strong></td>
<td>21.00 ± 0.29</td>
</tr>
</tbody>
</table>

* P<0.05; ** P<0.01
The results of the present experiment are supported by Shareeff and Al-Dabbagh (2009) who found no significant effect on percentage of heterophils, basophils, eosinophils, lymphocytes and monocytes in broilers supplemented with yeast. However, Gheisari and Kholeghipour (2006) found significant reduction in the H/L ratio of broilers supplemented with 0.3 per cent of yeast. Similarly, Paryad and Mahmoudi (2008) found reduced H/L ratio values in broilers supplemented with 1.5 and 2.0 per cent of yeast.

4.6 Serum-biochemical parameters

In the present investigation effect of supplementation of nucleotides on certain serum-biochemical parameters viz. cholesterol, triglyceride, HDL cholesterol, LDL cholesterol and glucose were studied in Japanese quails at 42nd day of trial. The results of these are presented in Table 4.19 and Fig. 4.17.

4.6.1 Serum glucose

A significant effect of nucleotide supplementation was observed on serum glucose level of different groups. Maximum glucose level (121.16 ± 0.28 mg/dl) was found in Japanese quails of T2 group which was statistically similar to the glucose level of control group. It was found that the concentration of glucose in serum decreased proportionally when the concentration of nucleotide supplement increased in the feed. Minimum (115.88 ± 0.77 mg/dl) and significantly (P<0.01) lower level of glucose was found in quails of T4 group which were fed 1.5% of nucleotide supplement.

The results of the present experiment revealed that there was significant decrease in level of glucose at higher levels of nucleotide supplementation. Earlier studies also support the results in which the dietary supplementation of nucleotides reduced serum glucose levels in broilers and calves (Shareef and Al-Dabbagh, 2009 and Krol, 2011). Similarly, significant reduction in the serum glucose concentration was noted by Aluwong et al. (2012) in broilers supplemented with yeast. However, Abdelrahman (2013) could not find effect of yeast supplementation on serum glucose in broilers.
Reduction in the serum glucose level of supplemented groups clearly indicates that dietary inclusion of nucleotides has a role in minimizing the stress.

4.6.2 Serum lipid profile

The mean values of serum lipid profile showing the effect of nucleotide supplementation in Japanese quails are shown in Table 4.19 and Fig. 4.17.

4.6.2.1 Serum total cholesterol

Japanese quails of nucleotide supplemented groups at 1.0 and 1.5 per cent level had significantly (P<0.01) lower serum total cholesterol as compared to T1 and T2 groups. Maximum serum cholesterol level (238.19 ± 11.81 mg/dl) was found in quails of group T1 (control). Japanese quails of T2 group showed statistically (P>0.05) similar level of serum cholesterol to that of group T1. The cholesterol content in serum of group T3 was lowest (176.75 ± 3.84 mg/dl) which was insignificantly different from the serum cholesterol level of T4 group quails.

4.6.2.2 Serum triglycerides

Serum triglyceride level (mg/dl) was significantly (P<0.05) different among Japanese quails of supplemented groups and control. Maximum (105.29 ± 2.70 mg/dl) serum triglyceride concentration was found in the quails of T2 group whereas, in the control group quails the concentration of triglyceride was minimum (82.72 ± 0.34 mg/dl). The concentration of triglyceride in all the supplemented groups were significantly higher (P<0.01) than the triglyceride level of control group. However, they were statistically (P>0.05) similar among themselves.

4.6.2.3 Serum HDL-cholesterol

Serum HDL cholesterol showed a significant impact of nucleotide supplementation. Minimum serum HDL-cholesterol (17.93 ± 0.08 mg/dl) level was recorded in quails of T1 group which was significantly lower than the serum HDL cholesterol levels of Japanese quails of T2 and T3 groups. HDL-cholesterol concentration between Japanese quails of control and T4 groups were statistically similar. Maximum (59.87 ± 6.04 mg/dl) and significantly higher level of serum HDL-cholesterol was noted in quails of T3 group.
4.6.2.4 Serum LDL-cholesterol

There was significant effect of nucleotide supplementation on the serum LDL-cholesterol levels of Japanese quails. Minimum (97.74 ± 10.66 mg/dl) and significantly (P<0.01) lower level of serum LDL-cholesterol was found in Japanese quails of T₃ group, whereas maximum (203.72 ± 11.71 mg/dl) level of LDL-cholesterol was observed in Japanese quails of control (T₁) group which was statistically similar to the LDL-cholesterol levels of T₂ group quails. Serum LDL-cholesterol level between the quails of T₂ and T₄ groups were also statistically similar.

Findings of the present investigation regarding effect of nucleotide supplementation on serum lipid profile of Japanese quails corroborated with Gheisari and Kholeghipour (2006) who found significant decrease in total cholesterol and increase in HDL-cholesterol of broilers supplemented with yeast. However, they observed no significant effect on the triglyceride level due to nucleotide supplementation. Similarly, Prayad and Mahmoudi (2008) and Shareef and Al-Dabbagh (2009) observed significant reduction in serum total cholesterol and triglycerides whereas significant increase in HDL-cholesterol in broilers supplemented with yeast. Krol (2011) noted significant reduction in the serum cholesterol level of calves supplemented with yeast nucleotides. Recent studies conducted by Aluwong et al. (2012) and Abdelrahman (2013) also revealed reduction in the cholesterol level of broilers supplemented with yeast culture.

In contrast to the findings of present investigation Chung Wu et al. (2011) noted significant increase in blood cholesterol of breast fed infants (breast milk is rich in nucleotides).

4.6.3 Health status related parameters

In the present investigation effect of supplementation of nucleotides on some of the health status related parameters viz. enzymes i.e. serum glutamate oxaloacetate transaminase (SGOT), serum glutamate pyruvate transaminase (SGPT), serum metabolites i.e. serum uric acid, serum creatinine, protein profile i.e. total serum protein, serum albumin, serum globulin and albumin-globulin ratio of Japanese quails at 42nd day of trial were considered.
4.6.3.1 Serum protein profile

The values of the serum protein profile of Japanese quails supplemented with nucleotides at different concentration are furnished in Table 4.20 and Fig 4.18.

4.6.3.1 (a) Serum total protein

Nucleotide supplementation as yeast extract had marked effect on serum total protein levels. Maximum and significantly (5.85 ± 0.45 g/dl) higher serum total protein compared to the control was found in Japanese quails of group T2 which was statistically (P>0.01) similar to the serum total protein of group T3 (4.77 ± 0.22 g/dl). Japanese quails of group T4 showed statistically (P>0.01) similar serum total protein to that of both T1 and T3 group birds. Minimum and significantly lower (3.42 ± 0.32 g/dl) serum total protein was observed in the Japanese quails of control group and it was comparable to that of T4 group.

4.6.3.1 (b) Serum albumin

The concentration of serum albumin (g/dl) was significantly affected by the nucleotide supplement in Japanese quails. The groups T2 and T3 supplemented with nucleotide were significantly different in the concentration of serum albumin from the control group. The maximum concentration (1.33 ± 0.11 g/dl) of albumin was observed in the T3 group supplemented with 1.0% of nucleotide whereas, the concentration of serum albumin was minimum (0.95 ± 0.07 g/dl) in control group. The serum albumin concentration of T4 group was statistically similar (P>0.05) to control as well as other supplemented groups.

4.6.3.1 (c) Serum globulin

The supplementation of nucleotide had marked effect on serum globulin concentration of Japanese quails. The concentration of serum globulin was significantly (P<0.01) lower in the birds of control group (2.47 ± 0.14 g/dl) compared to the supplemented groups. Maximum (4.62 ± 0.24 g/dl) and significantly higher concentration of serum globulin as compared to control and T4 group was observed in the T2 group quails. The serum globulin concentration of T3 group quails (3.44 ± 0.31 g/dl) was statistically similar to serum globulin level of both T2 and T4 group quails while serum globulin level of T4 group was significantly different from both T1 and T2 groups.
## Table 4.19: Effect of nucleotide supplementation on serum glucose and lipid profile of Japanese quails

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Serum Glucose** (mg/dl)</th>
<th>Serum total cholesterol** (mg/dl)</th>
<th>Serum triglycerides** (mg/dl)</th>
<th>Serum HDL-cholesterol** (mg/dl)</th>
<th>Serum LDL-cholesterol** (mg/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>T&lt;sub&gt;1&lt;/sub&gt;</td>
<td>119.54&lt;sup&gt;a&lt;/sup&gt; ± 0.66</td>
<td>238.19&lt;sup&gt;a&lt;/sup&gt; ± 11.81</td>
<td>82.72&lt;sup&gt;b&lt;/sup&gt; ± 0.34</td>
<td>17.93&lt;sup&gt;c&lt;/sup&gt; ± 0.08</td>
<td>203.72&lt;sup&gt;a&lt;/sup&gt; ± 11.71</td>
</tr>
<tr>
<td>T&lt;sub&gt;2&lt;/sub&gt;</td>
<td>121.16&lt;sup&gt;a&lt;/sup&gt; ± 0.28</td>
<td>232.79&lt;sup&gt;a&lt;/sup&gt; ± 6.58</td>
<td>105.29&lt;sup&gt;a&lt;/sup&gt; ± 2.70</td>
<td>39.64&lt;sup&gt;b&lt;/sup&gt; ± 5.71</td>
<td>172.10&lt;sup&gt;ab&lt;/sup&gt; ± 11.93</td>
</tr>
<tr>
<td>T&lt;sub&gt;3&lt;/sub&gt;</td>
<td>116.30&lt;sup&gt;b&lt;/sup&gt; ± 0.03</td>
<td>176.75&lt;sup&gt;b&lt;/sup&gt; ± 3.84</td>
<td>95.68&lt;sup&gt;a&lt;/sup&gt; ± 4.21</td>
<td>59.87&lt;sup&gt;a&lt;/sup&gt; ± 6.04</td>
<td>97.74&lt;sup&gt;c&lt;/sup&gt; ± 10.66</td>
</tr>
<tr>
<td>T&lt;sub&gt;4&lt;/sub&gt;</td>
<td>115.88&lt;sup&gt;c&lt;/sup&gt; ± 0.77</td>
<td>198.75&lt;sup&gt;b&lt;/sup&gt; ± 2.24</td>
<td>96.52&lt;sup&gt;a&lt;/sup&gt; ± 5.50</td>
<td>22.42&lt;sup&gt;c&lt;/sup&gt; ± 2.39</td>
<td>157.02&lt;sup&gt;b&lt;/sup&gt; ± 3.52</td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01

## Table 4.20: Effect of nucleotide supplementation on serum protein profile and health status related parameters of Japanese quails

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Total Serum Protein** (g/dl)</th>
<th>Albumin* (g/dl)</th>
<th>Globulin** (g/dl)</th>
<th>A/G ratio</th>
<th>Serum creatinine (mg/dl)</th>
<th>Serum uric acid* (mg/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>T&lt;sub&gt;1&lt;/sub&gt;</td>
<td>3.42&lt;sup&gt;c&lt;/sup&gt; ± 0.32</td>
<td>0.95&lt;sup&gt;b&lt;/sup&gt; ± 0.07</td>
<td>2.47&lt;sup&gt;c&lt;/sup&gt; ± 0.14</td>
<td>0.39 ± 0.01</td>
<td>0.72 ± 0.03</td>
<td>1.09&lt;sup&gt;a&lt;/sup&gt; ± 0.06</td>
</tr>
<tr>
<td>T&lt;sub&gt;2&lt;/sub&gt;</td>
<td>5.85&lt;sup&gt;a&lt;/sup&gt; ± 0.45</td>
<td>1.23&lt;sup&gt;a&lt;/sup&gt; ± 0.06</td>
<td>4.62&lt;sup&gt;a&lt;/sup&gt; ± 0.24</td>
<td>0.27 ± 0.01</td>
<td>0.56 ± 0.06</td>
<td>0.76&lt;sup&gt;b&lt;/sup&gt; ± 0.11</td>
</tr>
<tr>
<td>T&lt;sub&gt;3&lt;/sub&gt;</td>
<td>4.77&lt;sup&gt;ab&lt;/sup&gt; ± 0.22</td>
<td>1.33&lt;sup&gt;a&lt;/sup&gt; ± 0.11</td>
<td>3.44&lt;sup&gt;ab&lt;/sup&gt; ± 0.31</td>
<td>0.39 ± 0.07</td>
<td>0.49 ± 0.05</td>
<td>0.73&lt;sup&gt;b&lt;/sup&gt; ± 0.01</td>
</tr>
<tr>
<td>T&lt;sub&gt;4&lt;/sub&gt;</td>
<td>4.31&lt;sup&gt;bc&lt;/sup&gt; ± 0.30</td>
<td>1.12&lt;sup&gt;ab&lt;/sup&gt; ± 0.02</td>
<td>3.19&lt;sup&gt;b&lt;/sup&gt; ± 0.12</td>
<td>0.35 ± 0.01</td>
<td>0.57 ± 0.10</td>
<td>0.90&lt;sup&gt;ab&lt;/sup&gt; ± 0.05</td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01

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Results and Discussion
Fig. 4.17: Effect of nucleotide supplementation on serum glucose and lipid profile of Japanese quails

Fig. 4.18: Effect of nucleotide supplementation on serum protein profile and health status related parameters of Japanese quails
4.6.3.1 (d) Albumin-Globulin ratio (A/G ratio)

There was no significant difference in the serum albumin – globulin ratio among different treatment groups. The A/G ratio was maximum in Japanese quails of group T₁ (0.39 ± 0.01) and T₃ (0.39 ± 0.07) and minimum (0.27 ± 0.01) in T₂ group of quails.

From the result of serum protein profile it is clear that the supplementation of nucleotides significantly increased the serum total protein, serum albumin and serum globulin levels whereas there was insignificant effect on serum albumin-globulin ratio.

The results of the present experiment are in accordance with the findings of Choudhury et al. (2005) who found significant increase in serum protein, albumin and globulin levels in L. rohita juveniles supplemented with nucleotides. Similarly, Prayad and Mahmoudi (2008) and Shareef and Al-Dabbagh (2009) also found significant increase in serum protein, albumin and globulin levels of broilers supplemented with yeast. Recently, Shankar et al. (2012) also observed proportional increase in the serum protein level of fresh water prawns with increased concentration of nucleotide in the feed.

In contrast to the findings of present experiment Gheisari and Kholeghipour (2006) noted insignificant increase in serum protein of broilers supplemented with powdery yeast compared to the control.

Higher value of serum globulin indicates higher amount of immunoglobulin. Since the gamma fractions make the largest portion of globulin, it can be inferred that the dietary supplementation of nucleotide might enhance the immune response.

4.6.3.2 Serum uric acid

The mean values of serum uric acid showing the effect of nucleotide supplementation in Japanese quails are shown in Table 4.20 and Fig. 4.18.

Nucleotide supplementation had marked effect on serum uric acid levels in Japanese quails. The uric acid level of Japanese quails in T₂ and T₃ groups were significantly (P<0.05) lower than the control. The serum uric acid level among all supplemented groups as well as between T₁ and T₄ groups were statistically (P>0.05)
similar. However, insignificant decrease in serum uric acid level was noted in T₄ group compared to the control. Numerically, maximum (1.09 ± 0.06 mg/dl) concentration of uric acid was found in the Japanese quails of control whereas, minimum (0.73 ± 0.01 mg/dl) concentration was observed in the quails of T₃ group.

The findings of present experiment on serum uric acid are supported by Shareef and Al-dabbagh (2009) who found insignificant decrease in the uric acid level of broilers supplemented with yeast. While, Wu et al. (2011) observed significant reduction in blood urea nitrogen (corresponding end product of protein metabolism in mammals) in breast fed infants (breast milk is rich in nucleotides) compared to the formula fed infants.

The concentration of urea (uric acid in birds) in serum is inversely correlated to the net utilization of proteins and reflects the balance between intake, usage, and degradation of proteins and the renal excretion of protein metabolites (Taylor et al., 1974). Increased protein intake induces an increase in amino acid oxidation and the subsequent excretion of nitrogen, mainly as urea (uric acid in birds). The concentration of nitrogen in the urine and serum increased linearly with the amount of metabolizable protein that is fed to lactating dairy cows, indicating the decreased efficiency of nitrogen utilization (Wang et al., 2007). In this study, the reason for lower concentration of serum uric acid in groups supplemented with nucleotides may be the lower intake and presumably more efficient use of proteins in the diet.

4.6.3.3 Serum creatinine

The mean values of serum creatinine showing the effect of nucleotide supplementation in Japanese quails are shown in Table 4.20.

The quails of control group had numerically higher (0.72 ± 0.03 mg/dl) value of serum creatinine compared to the treatment groups but no significant difference could be noted. The minimum (0.49 ± 0.05 mg/dl) level of serum creatinine was noted in the quails of T₃ group.

Effect of nucleotide supplementation on serum creatinine of birds and animals has not been studied. Therefore, literature pertaining to this aspect is scanty, rather more experimentation on other aspects of serum biochemical parameters have been documented in the literature. The serum creatinine as well as uric acid levels of
different groups was similar in the present experiment which clearly indicate that there is no adverse effect of nucleotide supplementation on renal function of Japanese quails.

4.6.3.4 Enzymatic profile

In the present study, effect of supplementation of nucleotide on SGOT and SGPT levels of Japanese quails were studied at 42\textsuperscript{nd} day of trial. The results obtained are shown in Table 4.21 and Fig. 4.19.

4.6.3.4 (a) Serum glutamate oxaloacetate transaminase (SGOT)

The SGOT values in the present investigation showed a significant (P<0.01) effect of nucleotide supplementation in Japanese quails. The SGOT concentration in Japanese quails of T\textsubscript{1} group (166.90 ± 8.99 IU/L) was significantly (P<0.01) higher than the supplemented groups. However, the SGOT concentrations in Japanese quails of supplemented groups were statistically similar among themselves. Minimum (132.19 ± 0.53 IU/L) concentration of SGOT was noted in the Japanese quails of T\textsubscript{3} group.

The findings of present experiment on serum SGOT concentration are in contrast with Shareef and Al-Dabbagh (2009) and Aluwong \textit{et al.} (2012) who found no significant difference in the SGOT values of broilers supplemented with yeast. Similarly, Krol (2011) also found non-significant effect of nucleotide supplementation on SGOT values in calves.

4.6.3.4 (b) Serum glutamate pyruvate transaminase (SGPT)

There was no significant effect of nucleotide supplementation on SGPT value in Japanese quails of different treatment groups. SGPT values noted for T\textsubscript{1}, T\textsubscript{2}, T\textsubscript{3} and T\textsubscript{4} group Japanese quails were 12.13 ± 0.68 IU/L, 11.76 ± 0.23 IU/L, 11.32 ± 0.36 IU/L and 11.59 ± 0.68 IU/L respectively.

The results of the present experiment on SGPT values are supported by Shareef and Al-Dabbagh (2009) who found no significant effect of yeast supplementation on SGPT values in broilers. However, Aluwong \textit{et al.} (2012) found significant reduction in SGPT values in broilers supplemented with yeast.
The decreased values of SGOT and non-significant difference in SGPT values of supplemented groups indicate that there is no adverse effect of nucleotide supplementation on hepatic function of Japanese quails.

4.7 Immunological Status

In the present investigation effect of supplementation of nucleotides on certain immunological parameters viz. cell mediated immune response (delayed type hypersensitivity test) and humoral immune response (zinc sulphate turbidity test) were analysed.

4.7.1 Cell mediated immune response (Delayed type hypersensitivity test)

The mean values of skin thickness showing the effect of nucleotide supplementation on cell mediated immune response in Japanese quails are shown in Table 4.22 and Fig. 4.20.

Cell mediated immune response in the present investigation showed a significant (P<0.05) positive effect of nucleotide supplementation in Japanese quails. At 24 hours post challenge, the mean skin thickness of supplemented group quails were significantly (P<0.01) higher than the control group quails. Significant and maximum (0.65 ± 0.01 mm) skin thickness was noted in the quails of T₃ group while minimum (0.43 ± 0.02 mm) skin thickness was noted in the control group quails. The mean skin thicknesses at 24 hours post challenge were statistically (P>0.05) similar in the quails of T₂ and T₄ groups.

At 48 hours post challenge, the mean skin thickness of supplemented group quails were significantly (P<0.01) higher than the control group quails. Maximum (0.94 ± 0.03 mm) skin thickness was noted in the quails of T₃ group which was statistically similar to the skin thickness of T₂ group quails, while minimum (0.68 ± 0.02 mm) skin thickness was noted in the control group quails. The mean skin thickness of T₂ and T₄ group Japanese quails were statistically similar.

From the above results it is clear that supplementation of nucleotides in Japanese quails caused increase in cell mediated immune response. These findings indicate the immunomodulatory role of nucleotide in Japanese quails.
The results of present experiment are in accordance with Deng et al. (2005) who noticed minimal cutaneous activity of toe web to phytohaemagglutunin (PHA) in leghorns supplemented with yeast nucleotides (RNA). Choudhury et al. (2005) and Shankar et al. (2012) also found significant influence of nucleotide supplementation on superoxide anion activity in rohu juveniles and prawns, respectively. Similarly, Superchi et al. (2011) reported that supplementation of nucleotides improved cell mediated immune response of piglets as indicated by the increase in the cytotoxic T lymphocytes in the peripheral blood mononuclear cell population.

### 4.7.2 Humoral Immune Response (Zinc sulphate turbidity test)

The mean values of serum immunoglobulin concentration (mg/dl) in different treatment groups of Japanese quails are presented in Table 4.22 and Fig. 4.21.

Supplementation of nucleotide in Japanese quails showed a significant impact on serum immunoglobulin concentration. The serum immunoglobulin concentration of supplemented group Japanese quails were significantly (P<0.01) higher than the quails of control group. Maximum (13.99 ± 0.06 mg/dl) and significantly higher serum immunoglobulin concentration was observed in the T$_2$ group quails while significantly lower and minimum (9.78 ± 0.06 mg/dl) serum immunoglobulin concentration was observed in the control (T$_1$) group.

The results of present experiment clearly reveal that dietary nucleotide supplementation improved humoral immune response in Japanese quails. These results are supported by findings of Gheisari and Kholeghipour (2006) who found that supplementation of powder form of yeast at higher levels in broilers showed increase in humoral immune response. Recently, Fathi et al. (2012) also noted that the supplementation of yeast @ 1.0 g and 1.5 g/kg of feed showed increased humoral immune response in broilers. In contrast, Deng et al. (2005) noticed minimal effect of nucleotide supplementation on humoral immune response in leghorn chicks which was indicated by lower value of antibody titers against sheep red blood cell antigens.

As the cells of immune system, lack the potential to synthesize nucleotides (Sanderson and Youping, 1994). Proliferation of immuno competent cells depends on the readily available nucleotides. This may be the reason for improved immune response in the supplemented groups.
4.8 Intestinal morphology

In the present investigation effect of supplementation of nucleotides on certain morphological parameters of intestine viz. relative intestinal length/100 g of body weight (from duodenum to cloaca), duodenal villous height, duodenal crypt depth and villous height : crypt depth ratio were analysed.

4.8.1 Relative intestinal length

The mean values of relative intestinal length (cm) in different groups of Japanese quails are presented in Table 4.23 and Fig. 4.23.

Supplementation of nucleotide in Japanese quails showed a significant impact on relative intestinal length. The relative intestinal length in Japanese quails of supplemented groups were significantly (P<0.01) higher than the control group quails. Maximum (32.31 ± 0.43 cm) and significantly higher relative length of intestine was recorded in Japanese quails of T₃ group while minimum (28.76 ± 0.12 cm) and significantly lower relative intestinal length was noted in Japanese quails of T₁ group. Relative intestinal lengths in Japanese quails were statistically similar between T₂ and T₄ groups.

The results of the present experiment are in contrast with the findings of Jung and Batal (2012) who found no significant effect of nucleotide supplementation on relative length of intestine in broilers.

4.8.2 Duodenal villous height

The mean values of duodenal villous height (µm) in different treatment groups of Japanese quails are presented in Table 4.23 and Fig. 4.22.

Supplementation of nucleotide in Japanese quails showed a significant impact on duodenal villous height. The villous height were significantly (P<0.01) different among various groups of Japanese quails. Maximum (823.75 ± 7.80 µm) and significantly higher villous height was recorded in Japanese quails of T₂ group whereas significantly lower and minimum (557.50 ± 8.78 µm) villous height was noted in Japanese quails of T₁ group.
Table 4.21: Effect of nucleotide supplementation on enzymatic profile of Japanese quails

<table>
<thead>
<tr>
<th>Treatment</th>
<th>SGOT** (IU/L)</th>
<th>SGPT (IU/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>T&lt;sub&gt;1&lt;/sub&gt;</td>
<td>166.90&lt;sup&gt;a&lt;/sup&gt; ± 8.99</td>
<td>12.13 ± 0.68</td>
</tr>
<tr>
<td>T&lt;sub&gt;2&lt;/sub&gt;</td>
<td>145.60&lt;sup&gt;b&lt;/sup&gt; ± 2.06</td>
<td>11.76 ± 0.23</td>
</tr>
<tr>
<td>T&lt;sub&gt;3&lt;/sub&gt;</td>
<td>132.19&lt;sup&gt;b&lt;/sup&gt; ± 0.53</td>
<td>11.32 ± 0.36</td>
</tr>
<tr>
<td>T&lt;sub&gt;4&lt;/sub&gt;</td>
<td>146.05&lt;sup&gt;b&lt;/sup&gt; ± 3.46</td>
<td>11.59 ± 0.68</td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01

Table 4.22: Effect of nucleotide supplementation on skin thickness in delayed type hypersensitivity test and total serum immunoglobulin values of Japanese quails

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Skin thickness (mm)</th>
<th>Serum immunoglobulins** (mg/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>24 Hours - Post challenge**</td>
<td>48 Hours - Post challenge**</td>
</tr>
<tr>
<td>T&lt;sub&gt;1&lt;/sub&gt;</td>
<td>0.43&lt;sup&gt;c&lt;/sup&gt; ± 0.02</td>
<td>0.68&lt;sup&gt;c&lt;/sup&gt; ± 0.02</td>
</tr>
<tr>
<td>T&lt;sub&gt;2&lt;/sub&gt;</td>
<td>0.56&lt;sup&gt;b&lt;/sup&gt; ± 0.02</td>
<td>0.86&lt;sup&gt;ab&lt;/sup&gt; ± 0.02</td>
</tr>
<tr>
<td>T&lt;sub&gt;3&lt;/sub&gt;</td>
<td>0.65&lt;sup&gt;a&lt;/sup&gt; ± 0.01</td>
<td>0.94&lt;sup&gt;a&lt;/sup&gt; ± 0.03</td>
</tr>
<tr>
<td>T&lt;sub&gt;4&lt;/sub&gt;</td>
<td>0.56&lt;sup&gt;b&lt;/sup&gt; ± 0.01</td>
<td>0.84&lt;sup&gt;b&lt;/sup&gt; ± 0.03</td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01

Table 4.23: Effect of nucleotide supplementation on intestinal morphological values of Japanese quails

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Parameters</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Relative intestinal length** (cm)</td>
</tr>
<tr>
<td>T&lt;sub&gt;1&lt;/sub&gt;</td>
<td>28.76&lt;sup&gt;c&lt;/sup&gt; ± 0.12</td>
</tr>
<tr>
<td>T&lt;sub&gt;2&lt;/sub&gt;</td>
<td>30.62&lt;sup&gt;b&lt;/sup&gt; ± 0.25</td>
</tr>
<tr>
<td>T&lt;sub&gt;3&lt;/sub&gt;</td>
<td>32.31&lt;sup&gt;a&lt;/sup&gt; ± 0.43</td>
</tr>
<tr>
<td>T&lt;sub&gt;4&lt;/sub&gt;</td>
<td>29.94&lt;sup&gt;b&lt;/sup&gt; ± 0.28</td>
</tr>
</tbody>
</table>

* P<0.05 ; ** P<0.01
Fig. 4.19: Effect of nucleotide supplementation on enzymatic profile of Japanese quails

Fig. 4.20: Effect of nucleotide supplementation on skin thickness (mm) in delayed type hypersensitivity test of Japanese quails

Fig. 4.21: Effect of nucleotide supplementation on total serum immunoglobulin values (mg/dl) of Japanese quails
The findings of the present experiment are supported by Giancamillo et al. (2003) who found significant (P<0.01) increase in the villous height of ileum in weaned piglets supplemented with nucleotides. Similarly, Domeneghini et al. (2004), Puig et al. (2007) and Moore et al. (2011) also found increased intestinal villous height in piglets supplemented with nucleotides. Recently, Jung and Batal (2012) found increased villous height with nucleotide supplementation in broilers.

4.8.3 Duodenal crypt depth

The mean values of duodenal crypt depth (µm) in different treatment groups of Japanese quails are presented in Table 4.23 and Fig. 4.22.

Supplementation of nucleotide in Japanese quails showed a significant impact on duodenal crypt depth. The duodenal crypt depth in Japanese quails of supplemented groups were significantly (P<0.01) lower than the control group quails. Maximum (205.00 ± 2.50 µm) and significantly higher crypt depth was recorded in Japanese quails of T₁ group and minimum (180.00 ± 3.71 µm) crypt depth was noted in Japanese quails of T₂ group. Duodenal crypt depth of T₃ and T₄ groups as well as T₂ and T₄ groups were statistically (P>0.05) similar.

The findings of the present experiment are supported by Puig et al. (2007) who found decreased intestinal crypt depth in piglets supplemented with nucleotides. In contrast, Giancamillo et al. (2003) and Domeneghini et al. (2004) found significantly increased intestinal crypt depth in piglets supplemented with nucleotides. However, Zhang et al. (2005) found no significant effect of yeast supplementation on intestinal crypt depth of broilers. Jung and Batal (2012) also found non-significant influence of nucleotide supplementation on intestinal crypt depth in broilers.

4.8.4 Villous height-Crypt depth ratio (V/C ratio)

The mean values of villous height-crypt depth ratio in different treatment groups of Japanese quails are presented in Table 4.23 and Fig. 4.23.

The villous height and crypt depth ratio of Japanese quails showed a significant impact of nucleotide supplementation. The V/C ratio of Japanese quails were significantly (P<0.01) different among various treatment groups. Maximum (4.64 ±0.11) and significantly higher V/C ratio was recorded in Japanese quails of T₂ group while significantly lower and minimum (2.77 ± 0.09) V/C ratio was noted in Japanese quails of T₁ group.
Fig. 4.22: Effect of nucleotide supplementation on intestinal villous height and crypt depth (µm) of Japanese quails

Fig. 4.23: Effect of nucleotide supplementation on relative intestinal villous length and V/C ratio of Japanese quails
The results of the present experiment are supported by Zhang et al. (2005) who found increased V/C ratio in broilers supplemented with yeast. Recently, Jung and Batal (2012) found significant effect of nucleotide supplementation on V/C ratio in broilers. In contrast, Giancamillo et al. (2003) and Domeneghini et al. (2004) found that the supplementation of nucleotides in piglets resulted in decreased V/C ratio.

Intestinal mucosa is incapable to obtain its nucleotide requirement by de novo synthesis. Therefore, improvement in intestinal morphology may be attributed to high availability of nucleotides in quails provided with supplementary nucleotides.
Summary and Conclusion
Chapter 5

SUMMARY AND CONCLUSION

The beneficial effect of nucleotide supplementation in animal diet has drawn considerable research interests. Use of nucleotide supplement in poultry to enhance productivity and to produce safe food is relatively a new concept. Beneficial effects of nucleotide supplementation in animals, certain aquatic species and chicken have been studied. However, no such study is conducted in Japanese quails.

The present investigation was carried out at Instructional Poultry Farm (IPF), G.B. Pant University of Agriculture & Technology, Pantnagar, to study the effect of dietary nucleotide supplementation on the growth performance, nutrient retention, carcass traits, meat composition, haematological, certain serum biochemical and health status related parameters, intestinal morphology and immunological status in Japanese quails.

To achieve above objectives of the study, two experiments were conducted on Japanese quails.

**Experiment I**

Day old Japanese quail chicks were procured and kept on deep litter system. For first 3 days, chicks were kept on standard starter ration and plain water. On fourth day, one hundred and twenty Japanese quail chicks were selected on the basis of uniform average body weights and were randomly divided into 4 treatment groups (T₁, T₂, T₃ and T₄) and each group having three replications consisting of ten Japanese quails each. The nucleotide supplement was added in feed @ 0.5 per cent, 1.0 per cent and 1.5 per cent to the T₂, T₃ and T₄ groups respectively, excluding the control (T₁) group which was given feed without nucleotide supplement.

Japanese quails were offered feed and water *ad libitum* and kept on artificial light throughout the experimental period. Production parameters were recorded on weekly basis. Delayed type of hypersensitivity test was conducted on 21st day of trial by randomly selecting six Japanese quails from each group. The sensitized Japanese quails were challenged two weeks later and assessment of reaction was done 24 and 48 hours post challenge. A metabolic trial of 7 days duration was conducted from 36th
– 42nd days of experiment to study the effect of nucleotide supplementation on nutrient utilization. At the end of experiment on 42nd day, two Japanese quails from each replicate (six Japanese quails/treatment) were randomly slaughtered for carcass yield, processing losses, yield of cut up parts, organ weights and carcass composition. After evisceration the intestine of the birds were carefully separated and the length of the intestine from duodenum to end of the rectum was measured using a measuring tape to study the effect of supplement on the intestinal gross morphology. A sample of two cm from proximal jejunum was collected and preserved in 10 per cent formalin to study the histological changes in the villous height, crypt depth and villous height-crypt depth ratio of the intestine. Representative blood samples were also collected for haemato-biochemical parameters and humoral immune response.

Experiment II

On the basis of the experiment I results, the levels of the nucleotide supplement for the experiment II were fixed as 0, 0.25, 0.5 and 0.75 per cent in feed for the treatment groups T1, T2, T3 and T4 respectively.

In the experiment II, the performance of Japanese quails in terms of growth and nutrient utilization were studied similar to the experiment I using 120, four day old Japanese quail chicks.

The results obtained in the present investigation may be summarized as follows:

- The feed intake in Japanese quails revealed significant impact due to all levels of nucleotide supplementation with best results at 0.5 per cent and 0.25 per cent levels in experiment I and experiment II, respectively.
- The body weight gain of Japanese quails revealed significant (p<0.01) impact of nucleotide supplementation at all the levels with best results at 0.5 per cent level in both the experiments.
- FCR and Performance Index of broilers were significantly improved due to all levels of nucleotide supplementation with best performance at 0.5 per cent level in both the experiments.
- As regards nutrient utilization, per cent utilization of dry matter, crude fat and total carbohydrates was significant with all levels of nucleotide supplementation.
supplementation in experiment I while utilization of crude fat was not affected in experiment II. However, utilization of total carbohydrates was significant at 0.25 and 0.5 per cent levels of nucleotide supplementation in experiment II. In experiment I, 0.5 per cent level and in experiment II, 0.5 and 0.75 per cent levels of nucleotide supplementation significantly improved the crude protein utilization.

- The dressed yield without and with giblet revealed no significant influence at all levels of nucleotide supplementation.
- Supplementation of nucleotide did not significantly (P<0.05) affect the thigh, breast, drumsticks and neck weights while wing weight was significantly increased by 1.0 per cent level of nucleotide supplementation.
- Organ weights of quails revealed non-significant impact of nucleotide supplementation on heart weight. Whereas significantly higher liver weights at 0.5 and 1.5 per cent levels and gizzards weights at 0.5 and 1.0 per cent levels of nucleotide supplementation were noted.
- The processing losses as blood loss, feather loss, head and shank loss were not significantly affected by supplementation of nucleotides at different levels.
- Moisture, crude protein and total ash content of thigh and breast muscles were non-significantly affected by supplementation of nucleotide in Japanese quails.
- Fat content of thigh and breast muscles were significantly reduced by supplementation of nucleotides in Japanese quails.
- TEC, TLC, PCV, Haemoglobin and MCHC values in nucleotide supplemented groups were significantly (P<0.01) higher than the control group. Maximum values of all these parameters were noted in T4 group, in which nucleotide supplement was added @ 1.5 per cent in feed.
- MCV and MCH did not vary significantly due to the supplementation of nucleotides in Japanese quails.
- Nucleotide supplementation did not significantly affect the heterophil, lymphocyte, monocyte, basophil and eosinophil count in Japanese quails.
• Serum glucose level was significantly (P<0.01) reduced in the groups supplemented with higher levels of nucleotide with minimum glucose level in T4 group (1.5 per cent nucleotide).

• Higher levels of nucleotide supplementation reduced the serum total cholesterol and LDL cholesterol to different extents in Japanese quails. The maximum reduction of these was noted in 1.0 per cent nucleotide supplemented group of Japanese quails.

• Serum triglycerides and HDL cholesterol were significantly increased by supplementation of nucleotides in Japanese quails. Maximum serum triglycerides and HDL cholesterol levels were noted in Japanese quails supplemented with 0.5 per cent and 1.0 per cent of nucleotides, respectively.

• The protein profile viz. serum total protein, albumin and globulin were significantly increased in Japanese quails of all the supplemented groups. However, Albumin-Globulin ratio did not vary significantly due to inclusion of nucleotide supplement.

• Serum creatinine and SGPT contents in the present investigation showed non-significant impact of nucleotide supplementation.

• SGOT and serum uric acid contents of Japanese quails in nucleotide supplemented groups were found significantly (P≤0.05) lower as compared to control.

• Average skin thickness, both at 24 and 48 hours, post challenge were significantly (P<0.01) higher in all the nucleotide supplemented groups with maximum in 1.0 per cent nucleotide supplemented group of Japanese quails.

• Humoral immune response as measured by total serum immunoglobulin levels was significantly (P<0.05) increased in all the nucleotide supplemented groups with maximum in T2 group.

• As regards intestinal morphology, the relative intestinal length, duodenal villous height and villous height-crypt depth ratio were found significantly (P<0.01) increased in all the supplemented groups. Villous height and V/C ratio were highest at 0.5 per cent level of nucleotide supplementation whereas
relative intestinal length was maximum at 1.0 per cent level of nucleotide supplementation.

- Duodenal crypt depth was significantly (P<0.01) decreased in Japanese quails of all the supplemented groups compared to the control group quails with minimum in T₂ group, in which nucleotide supplement was added @ 0.5 per cent in feed.

**Conclusion**

From results of the experiment, it is concluded that the supplementation of nucleotides as yeast extract at 0.5 per cent level in the present investigation improved the production performance of Japanese quails in both the experiments. It also improved utilization of dry matter, crude fat, crude protein and total carbohydrates. Liver and gizzard weights were significantly improved by the nucleotide supplementation. Supplementation of nucleotide significantly reduced the crude fat content in thigh and breast muscles of Japanese quails. TEC, TLC, PCV, Hb and MCHC concentrations were best at 1.5 per cent level of nucleotide supplementation. Serum glucose, SGOT and uric acid were significantly decreased in groups supplemented with nucleotides. Serum protein, serum albumin and serum globulin levels were significantly increased with nucleotide supplementation while, albumin-globulin ratio, serum creatinine and SGOT were similar in all the groups. There was an improvement in the lipid profile by nucleotide supplementation viz. decreased serum cholesterol and LDL cholesterol and increased HDL cholesterol. Nucleotide supplementation was responsible for better cell mediated and humoral immune response. Relative intestinal length, duodenal villous height and V/C ratio were significantly increased due to nucleotide supplementation while, duodenal crypt depth was decreased with nucleotide supplementation.

Therefore, it can be finally concluded from the results of present investigation that dietary supplementation of nucleotides at 0.5 per cent level may be advised to improve the production performance, nutrient utilization, intestinal development and immune response of Japanese quails, whereas higher levels are advised for improvement of haemato-biochemical parameters in Japanese quails. Further studies on layer chicken, Japanese quail layers and other poultry species are recommended.
Literature Cited


Basic Animal Husbandry Statistics (BAHS), 2012, Goverment of India, Ministry of Agriculture, Department of Animal Husbandry and dairying, New Delhi, India. Web: http://dahd.nic.in


FAOSTAT 2012. Web: www.faostat.org


Sauer, N.; Eklund, M.; Bauer, E.; Gänzle, M. G.; Field, C. J.; Zijlstra, R. T. and Mosenthin, R. 2012. The effects of pure nucleotides on performance,
humoral immunity, gut structure and numbers of intestinal bacteria of newly weaned pigs. *J. Anim. Sci.* 90: 3126-3134


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In the present investigation, two experimental trials were conducted on 120, three day old Japanese quails (red plumaged) for a period of 6 weeks each, to study the effect of nucleotide supplementation as yeast extract on the growth performance, nutrient retention, carcass traits, meat composition, haematological, certain serum biochemical and health status related parameters, intestinal morphology and immunological status. In both of the experiments, experimental Japanese quails were randomly divided into four treatment groups (T1, T2, T3 and T4), each group having three replications consisting of ten Japanese quails each. The different groups were supplemented with nucleotides @ 0, 0.5, 1.0 and 1.5 per cent and 0, 0.25, 0.5 and 0.75 per cent in experiment I and experiment II, respectively. Growth parameters were studied at weekly intervals as well as for starter (I-II week), finisher (III -VI week) and overall periods (I-VI). Delayed type of hypersensitivity test was conducted on the 21st day of experiment I, by randomly selecting six Japanese quails from each group for the sensitization test. A metabolic trial of 7 days duration was conducted from 36th – 42nd days of both the experiments to determine the effect of nucleotide supplementation on nutrient utilization. At the end of experiment I on 42nd day, two Japanese quails from each replicate (six Japanese quails/treatment) were randomly slaughtered for carcass yield, processing losses, yield of cut up parts, organ weights and carcass composition. Representative blood samples were also collected for study of haemato-biochemical parameters and humoral immune response. After evisceration the intestine of the birds were carefully separated and the length of the intestine from duodenum to end of the rectum was measured using a measuring tape to study the effect of supplement on the intestinal gross morphology. A sample of two cm from proximal jejunum was collected and preserved in 10 per cent formalin to study the histological changes on the villous height, crypt depth and villous height-crypt depth ratio of the intestine. The results of the both experiments indicated that dietary inclusion of nucleotides in Japanese quails increased body weight gain, reduced feed intake, improved feed conversion ratio and performance index significantly (P<0.01), with best performance during all the periods was noted in group supplemented with 0.5 per cent of nucleotides. The nutrient utilization in terms of dry matter, crude protein and total carbohydrates were significantly (P<0.05) improved in Japanese quails of nucleotide supplemented groups in both the experiments. Utilization of crude fat content was significantly (P<0.05) improved in experiment I with all levels of nucleotide supplementation while utilization was not affected in experiment II. The dressed yield with and without giblet were not affected by nucleotide supplementation. Thigh, breast, drumstick and neck muscles weight were not affected by supplementation of nucleotides. Weight of back muscles were significantly (P<0.05) decreased in supplemented groups. However, wing muscles weight were significantly (P<0.05) improved in Japanese quails supplemented with 1.0 per cent level of nucleotide. Nucleotide supplementation significantly improved the liver and gizzard weight. Heart weight and processing losses were not affected by nucleotide supplementation. Fat content of the thigh and breast muscles were significantly (P<0.05) reduced by nucleotide supplementation. However, moisture, crude protein and total ash content of thigh and breast muscles were not affected. Haematological parameters such as TEC, TLC, PCV, Hb and MCHC values showed significant impact of nucleotide supplementation. Decreased concentration of serum glucose, serum cholesterol and LDL cholesterol and increased concentration of triglycerides and HDL cholesterol were noticed in nucleotide supplemented groups especially at higher levels. Protein profile showed significant improvement in total protein, albumin and globulin with higher levels of nucleotide. Serum creatinine and SGPT contents showed non-significant impact whereas, serum uric acid and SGOT contents were significantly reduced in nucleotide supplemented quails. Nucleotide supplementation significantly (P<0.01) improved both humoral and cell mediated immune response. Relative intestinal length, duodenal villous height and V/C ratio were significantly increased while duodenal crypt depth was decreased with all levels of nucleotide supplementation with best results at 0.5 per cent level of nucleotide supplementation. From the results of present study it can be concluded that, dietary nucleotide supplementation at 0.5 per cent level may be advised to improve growth performance, nutrient retention, lean meat production, intestinal morphology and immune status of Japanese quails.
उक्षी, विका विंयाण,

जापानी बंदर की उपयोगकर्ता पर न्यूजिल्यॉनड क्षेत्र के रूप में भूमिका का प्रभाव

अलावा,

नाम : एस.प्राकाश
परिचारक : 42782
व्यवसा एवं प्रशेष का वर्ष : प्रशासन, 2011-12
उपाधि : एम.बी.एसी.
मुख्य विषय : कुकुट्टि विघ्नान
विभाग : पर्यटन उद्यान विभाग
संस्थान : "जापानी बंदर की उपयोगकर्ता पर न्यूजिल्यॉनड स्थल के रूप में भूमिका का प्रभाव"