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SINGLE AND SYNERGISTIC EFFECTS OF LYSINE AND PHYTASE SUPPLEMENTATION ON GROWTH PERFORMANCE MEAT QUALITY AND BONE CHARACTERISTICS IN JAPANESE QUAIL

Waqas Haroon[*1], Saima Ashraf[2], Sana Saeed[3], Syeda Asloob Fatima[4], Muhammad Ali[5], Samia Mushtaq[6], Syeda Fakhra Waheed[7], Maria Fiaz[8], Muhammad Asif[9], Fazal Ur Rehman[10]

*[*1,2,3] Department of Anatomy and Histology, University of Veterinary and Animal Sciences, Lahore, Pakistan*
[4]Institute of Physiology and Pharmacology, University of Agriculture Faisalabad, Pakistan
[5]Department of Pathobiology, University of Layyah, Layyah, Pakistan
[6,7]Department of Veterinary Sciences, AJ and K University of Bhimber, Bhimber AJK, Pakistan
[8]Department of Food Safety and Quality Management, Bahauddin Zakariya University, Multan, Pakistan
[9]Department of Clinical Sciences, University of Layyah, Layyah, Pakistan
[10]Department of Veterinary Medicine and Surgery, Lasbela University of Agriculture, Water and Marine Sciences, Uthal, Pakistan

[*1]waqasharoon978@gmail.com

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Corresponding Author: *
Waqas Haroon[*1]

Abstract

Background

Supplementation of both lysine and phytase is important to improve growth performance and carcass yield in poultry birds.

Objectives

The purpose of current study was to evaluate the single and synergistic effect of lysine and phytase on growth performance, visceral organs weight, meat quality, and bone characteristics in Quails birds.

Materials and Methods

This study was carried out on 240 quails. The duration of trial was 35 days at Avian Research and Training (ART) Centre UVAS, Lahore. The quails were randomly divided into 8 groups and each group comprised of five replicates and each replicate consisted of six birds. Birds were fed with commercial diet (CD) supplemented with lysine sulphate and phytase with ad libitum access of fresh water for 35 days. On day 35, two birds from each replicate (10 birds /group) were selected for sampling. The feed conversion ratio (FCR), alkaline phosphates enzyme activity, gross parameters of visceral organs and various other meat parameters were studied. The data was analyzed with one way ANOVA using SPSS software version 20.0.

Results

The effects of lysine and phytase supplementation individually and in combination on body weight gain results showed that body weight gain, FCR, feed intake, muscle diameter, bone length, weight and diameter, and meat parameters such as pH, color, and drip loss showed significant difference (P≤0.05) except alkaline phophatase enzyme (P≥0.05). The lysine and phytase supplements showed positive effects on these parameters.

Conclusion

It was concluded that dietary supplementation lysine and microbial phytase individually and in combination microbial phytase in combination with 8g lysine generally improved meat quality and bone morphometric characteristics in Japanese Quails.

INTRODUCTION

Protein is the main nutrient which contributes to the development of muscle tissue and in turn the carcass (Yaremchuk et al. 2022). Amino acids are the building blocks of proteins. Amongst twenty essential amino acids Lysine is the second limiting amino acid in poultry diets after methionine. Also, enzymes are biological catalysts which accelerate the chemical reactions. The supplementation of enzymes in poultry diets has dramatically increased all over the world (Cheng et al. 2020). During the formation of a balanced poultry diet percentage of all other amino acids is calculated with reference to the lysine (Macelline et al. 2021).The chicks diet supplemented with essential amino acids have the greater performance (Kidd et al. 2021).

The use of phytase is to increase the phosphorus availability. The phosphorus and calcium are required in correct proportions to the birds (Walters et al. 2019). The efficiency of microbial phytase is to increase nutrient bioavailability (Zouaoui et al. 2018). The use of phytase improves the digestive health and protein digestibility (Vashishth et al. 2017).The supplementation with phytase increases the digestibility of nutrients and utilization of energy and also increases the utilization of minerals (Yang et al. 2016).

Quails have been used for biological studies because of their small size, lesser disease risk, easy handling and less space requirement for rearing. The current study was done to evaluate the single and synergistic effect of lysine and phytase on growth performance, visceral organs weight, meat quality, and bone characteristics.

Materials and Methods

Experimental Design

This study was carried out on 240 quails. The duration of trial was 35 days at Avian Research and Training (ART) Centre UVAS, Lahore. The quails were randomly divided into 8 groups and each group comprised of five replicates and each replicate consisted of six birds.

Housing of Experimental Birds

The temperature was35°C at 1st day and gradually decreased (at the rate of 2.8°C per week) to 26.7°C by the end of third week and relative humidity was maintained at 65% till day 40. After third week temperature and humidity were maintained at this level until 35 days of age. Feed and water were provided adlibitum.

Dietary Treatments

Birds were fed with commercial diet (CD) supplemented with lysine sulphate and phytase with ad libitum access of fresh water for 35 days. Table 1:

Table 1: Dietary Treatments

Groups	Feed	Dose per kg of Feed
A (Control group)	CD	-
B	CD+Lysine	5g Lysine (Oliveira et al. 2013)
C	CD+Lysine	8g Lysine (Oliveira et al. 2013)
D	CD+Lysine	11g Lysine (Shivazad et al. 2013)
E	CD+Phytase	500 phytase units/kg (Selle and Ravindran 2007)
F	CD+Lysine+Phytase	5g Lysine +500 phytase units/kg
G	CD+Lysine+Phytase	8g Lysine +500 phytase units/kg
H	CD+Lysine+Phytase	11gm Lysine +500 phytase units/kg

CD= Commercial diet

Growth Performance

Initial body weight was measured on 1st day of the trial and weight was subsequently recorded on weekly basis for FCR calculation.

FCR = Feed consumed / body weight gain

Sampling

On day 35, two birds from each replicate (10 birds /group) were selected for sampling.

Alkaline Phosphatase Level (ALP)

Level of alkaline phosphatase was calculated at the end of trial by serum of the bird. Mass spectrometer with suitable kit was used for the calculation of alkaline phosphatase.

Gross Parameters of Visceral Organ

Filled and empty weights of proventriculus, gizzard, small intestine, large intestine were measured by digital weight balance soon after slaughtering. The length of small intestine and large intestine was measured by measuring tape.

Muscle Fiber and Fascicle Diameter

Segments of muscle Pectoralis Major were placed in labeled tissue bottles. Segments were fixed in 10% neutral buffered formalin. Tissue samples were dehydrated by transferring them through a series of increasing concentration of alcohol, placed into xylene and then processed through paraffin embedding technique. A microtome was used to make cuts that and slides were stained with H&E staining method. Muscle fiber diameter was calculated by Morphometry program (Progress capture Pro 2.7.7. Labomed USA). Diameter (µm) of 5 muscle fibers from 3 fascicles was measured. Similarly the diameter of fascicle was measured.

Muscle Tissue Processing:

Paraffin Embedding Technique

Light microscopic examination of muscle tissue through paraffin-embedding technique was done (Porter et al. 2017). Thin muscle sections are required for light microscopy i.e. approximately 4-6µm which were prepared by paraffin embedding technique. Following steps were performed:

Washing

Tissue was washed with raining tap water for eight hours after fixation in 10% neutral buffered formalin.

Dehydration

Ascending grades of ethyl alcohol were used for tissue dehydration. Samples were placed in tissue cassettes and then these cassettes were placed for two hours each in 60%, 70%, 80%, 90% and twice in 100% absolute alcohol solution gradually for completion of dehydration procedure.

Clearing

For clearing, tissue cassettes were placed in Xylene I and Xylene II for two hours each.

Infiltration

For infiltration procedure paraffin wax was used. Tissue cassettes were placed in paraffin wax for 3 hours at 60°C temperature. After completion of infiltration procedure, paraffin blocks were formed by using metallic molds.

Sectioning and Tissue Mounting

Sectioning of tissue paraffin blocks was performed on the semi-automatic Microtome. The block was fixed on the microtome. First of all rough cutting was done at the scale of 20µm for exposing the tissue. Subsequently, the scale for sectioning was fixed at 5µm for slide preparation. The temperature of water bath was maintained at 45-50°C and ribbon of sections was placed in water bath. The sections were mounted on the slide with gelatin powder added in water bath. Mounted slides were dried for 15 min in hot air oven at 90-100°C. One slide per muscle sample was prepared with Haematoxylin and Eosin (H & E) staining and was used for measurements of histomorphometric characteristics of muscle.

Haematoxylin and Eosin Staining Technique

Haematoxylin and eosin stainingwas carried out as described by Yao et al. (2020).

Haematoxylin and Eosin

Haematoxylin (2.5 g), potassium alum (50 g), absolute alcohol (25 ml), mercuric oxide (1.25 g), glacial acetic acid (20 ml),distilled water (500 ml) and 2% eosin solution was used.

Staining Protocol

Slides were kept in Xylene I and Xylene II for 2 minutes each. This eliminated the wax from tissues and facilitated in penetration of aqueous solution of Haematoxylin stain in tissue. Slides were kept in descending series of alcohol (100%, 90%, 80%, and 70%) for 2 minutes each for the elimination of xylene. Slides were then placed in water for 5 min for rehydration. For primary staining, slides were placed in Haematoxylin solution for fifteen minutes. After that slides were cleared with tap water to wash away extra Haematoxylin stain. For differentiation slides were passed from 1% acid alcohol for 5-10 seconds. For washing the slides were placed under tap water for 3-4 dip. Then slides were passed from 1% Eosin for ten minute. The excessive stain from the slides was removed by running tap water for 1-5 minutes. For final dehydration, slides were passed through ascending alcohol series (70%, 80%, 90% &100%) for 2 minutes each. For final clearing of alcohol, slides were put in Xylene I and II for two minutes each. At the end, coverslips were placed on tissues by using DPX (A mixture of Distyrane, Plasticiser and Xylene). The slides were dried overnight for further analysis. Histomorphometric measurements of all slides were done under light microscope.

Histomophometry of Muscles

Muscle Diameters

For Muscle fascicles diameter of pectoralis muscles, picture of slides were captured at 4X. For muscle fiber diameter, images were captured at 10X from five different areas of slide. Diameter of five muscle fibers from three fascicles was measured from all samples in µm.

Parameters for Evaluation of Meat

Measurement of Drip Loss

Honikel's gravimetric drip loss method was used. Muscle samples were weighed initially, placed in a meshed pouch and suspended in special container equipped with lid to avoid evaporation, and left in refrigerator at 4-6°C for 48h. Muscle fiber direction of the samples was horizontal to gravity. After 24 hours samples were removed from refrigerator and weighed again. The percent difference in two weights was used for calculating water-holding capacity.

Measurement of Muscle pH

Muscle pH of thigh meat samples was measured with digital pH meter (Portable meat pH meter HI 99163 Hanna, Italy). A vertical deep incision was given in muscles and after calibration of pH meter the piercing knob of digital pH meter was inserted 1 cm deep into muscles and pH was measured within 15 minutes after slaughtering. Muscle pH was determined at time interval of 0, 12 and 24 hours.

Measurement of Other Muscle Parameters

The muscle Color was checked with Minolta meter.The tenderness of the muscles was checked with the texture analyzer.The cooking loss was checked by cooking. Marination uptake was checked with salt solution uptake.Electrical stimulation was checked with the electric stimulator. The vacuum packaging was checked with the removing of air.For bone analysis, tibia bone of bird were separated, boiled in water for ten minutes at 100°C and then air-dried at room temperature.Weight of tibiae bones

was measured by using digital weight balance and length of bones was measured with digital Vernier caliper.Outside diaphysis diameter of tibia bone was measured at the mid-point**.** The tibial bone MCD was measured by breaking the bone at mid-point and thickness of bony wall was measured by using digital Vernier caliper**.** The length of bone was measured by using digital Vernier caliper. Bone Tibiotarsal index was identified by using formula; [(diaphysis diameter-medullary canal diameter)/ diaphysis dia]100.

Growth Performance

Feed Conversion Ratio (FCR), Body Weight Gain and Feed Intake

The feed conversion ratio was measured on weekly basis and also measured the average feed conversion ratio of each group. Body weight gain and feed intake was measured on weekly basis.

Statistical Analysis

The data was analyzed with one way ANOVA using SPSS software version 20.0. The data was described as mean ± standard deviation with $p<0.05$ which was regarded as significant.

Results

Growth Performance

Body Weight Gain

The effects of Lysine and Phytase supplementation individually and in combination on body weight gain results showed that body weight gain was significant (P< 0.05) higher in supplemented groups as compared to the control group except in first week .Highest values were observed in G-group.

Feed Intake

The effects of Lysine and Phytase supplementation individually and in combination on feed intake results showed that feed intake was significant (P< 0.05) higher in supplemented groups as compared to the control group. Highest values were observed in G-group.

Feed Conversion Ratio (FCR)

The effects of Lysine and Phytase supplementation individually and in combination on feed conversion ratio results showed that feed conversion ratio significantly (P< 0.05) better in supplemented groups during 4^{th} and 5^{th} week as compared to the control group. Best FCR values were observed in G-group.

Parameters of Muscle

Muscle Fascicle Diameter

The results of muscle fascicle diameter showed that the all treated groups had the significantly (P< 0.05) higher muscle fascicle diameter than control group. And G-group had highest muscle fascicles diameter as compared to all other groups.

Muscle Fiber Diameter

The effects of supplementation of Lysine and Phytaseseparately and in combination on muscle fiber diameter of the breast muscle of quails showed that the muscle fiber diameter was highest (P<0.05) in G-group as compared to all other groups.

Bone Parameters

Tibia Bone Length

The effects of Lysine and Phytase supplementation individually and in combination on length of the tibia bone showed that length of tibia bone was significantly (P< 0.05) higher in supplemented groups as compared to the control group. Highest values were observed in G-group.

Tibia Bone Weight

The effects of Lysine and Phytase supplementation individually and in combination on weight of the tibia bone showed that weight of tibia bone was significantly (P< 0.05) higher in supplemented groups as compared to the control group. Highest values were observed in G-group.

Bone Diaphysis Diameter

Lysine and Phytase supplementation individually and in combination increased the diaphysis diameter of the tibia bone as given. All the treated groups had significantly (P< 0.05) higher values of the said parameter when compared with the control group with maximum value observed in the G-group.

Medullary Canal Diameter and Tibio-tarsal Index of Tibia Bone

The effect of dietary supplementation of Lysine and Phytase on medullary canal diameter showed that all the supplemented groups had higher values (P< 0.05) when compared to the control group. The H-group

had highest diameter compared to other groups. Lysine and Phytase supplementation individually and in combination did not affect the tibio-tarsal index. All results are shown in below tables 2 to 4.

Table 2: Effect of Lysine and Phytase Supplementation on Feed Intake in Japanese Quails (Mean ± S.E)

	*GROUPS								
Parameters	Control	Lysine 5g	Lysine 8g	Lysine 11g	Phytse500units	Lysine 5g+Phytse500units	Lysine 8g+Phytse500units	Lysine 11g+Phytse500units	P-value
Feed intake 1st week	43.10±0.54^{ab}	46.30±0.59^{a}	44.00±0.63^{ab}	45.30±0.63^{ab}	43.40±0.60^{ab}	44.70±0.73^{ab}	44.60±0.70^{ab}	45.60±0.87^{ab}	<0.016
Feed intake 2nd week	85.80±0.77^{b}	77.10±0.83^{d}	74.60±0.74^{d}	92.30±0.55^{a}	84.90±0.87^{bc}	82.90±1.06^{bc}	83.00±1.12^{bc}	81.30±0.83^{c}	<0.000
Feed intake 3rd week	127.50±1.18^{g}	176.40±1.09^{a}	144.70±0.88^{f}	194.20±0.84^{a}	187.40±1.06^{b}	168.40±0.76^{d}	149.30±0.77^{e}	169.20±0.62^{d}	<0.000
Feed intake 4th week	167.90±0.70^{f}	203.90±0.73^{d}	198.90±1.11^{e}	215.80±1.28^{c}	213.80±1.27^{c}	221.30±1.70^{b}	232.70±1.33^{a}	207.50±0.88^{d}	<0.000
Feed intake 5th week	189.40±1.09^{f}	237.30±1.66^{d}	229.80±1.42^{e}	257.00±1.14^{b}	252.80±1.32^{b}	254.80±1.09^{b}	265.30±1.43^{a}	246.30±1.57^{c}	<0.000

Within the same row, different superscript indicates significantly different means

Table 3: Effect of Lysine and Phytase Supplementation on Weight Gain in Japanese Quails (Mean ± S.E)

	*GROUPS								
Parameters	Control	Lysine 5g	Lysine 8g	Lysine 11g	Phytse500units	Lysine 5g+Phytse500units	Lysine 8g+Phytse500units	Lysine 11g+Phytse500units	P-value
Weight gain 1st week	24.30±0.74	24.40±1.15	22.70±2.53	25.10±0.99	23.70±0.91	24.90±0.99	26.60±0.83	25.30±1.07	>0.570
Weight gain 2nd week	46.50±0.98^{ab}	41.60±0.80^{cd}	39.80±0.98^{d}	50.00±0.94^{a}	44.40±0.77^{bc}	44.20±0.77^{bc}	45.60±1.36^{bc}	44.20±1.08^{bc}	<0.000
Weight gain 3rd week	67.60±1.54^{e}	94.60±0.99^{b}	77.30±1.39^{d}	105.10±1.25^{a}	102.00±1.61^{a}	91.20±1.04^{b}	84.50±1.00^{c}	91.90±0.80^{b}	<0.000
Weight gain 4th week	87.20±0.55^{e}	107.10±0.64^{d}	104.70±0.86^{d}	116.10±1.12^{bc}	115.60±1.11^{bc}	119.80±1.2^{b}	126.60±0.95^{a}	114.40±1.55^{c}	<0.000
Weight gain 5th week	95.90±0.90^{d}	122.00±1.37^{a}	118.50±0.98^{a}	135.30±1.17^{b}	131.60±1.11^{b}	134.40±1.27^{b}	150.10±1.39^{a}	131.60±1.14^{b}	<0.000

Within the same row, different superscript indicates significantly different means

Table 3: Effect of Lysine and Phytase Supplementation on Feed Conversion Ratio in Japanese Quails (Mean ± S.E)

	*GROUPS								
Parameters	Control	Lysine 5g	Lysine 8g	Lysine 11g	Phytse500units	Lysine 5g+Phytse500units	Lysine 8g+Phytse500units	Lysine 11g+Phytse500units	P-value
FCR 1st week	1.79±0.77	1.93±0.92	3.86±2.07	1.82±0.07	1.85±0.07	1.82±0.82	1.69±0.57	1.83±0.95	>0.464
FCR 2nd week	1.85±0.36	1.86±0.46	1.88±0.46	1.85±0.34	1.91±0.39	1.88±0.44	1.83±0.56	1.84±0.40	>0.914
FCR 3rd week	1.89±0.46	1.86±0.17	1.87±0.33	1.85±0.21	1.84±0.34	1.84±0.22	1.76±0.21	1.84±0.17	>0.127
FCR 4th week	1.92±0.01^{a}	1.90±0.01^{ab}	1.90±0.01^{ab}	1.86±0.01^{abc}	1.85±0.01^{abc}	1.84±0.02^{abc}	1.83±0.01^{bc}	1.81±0.03^{c}	<0.001
FCR 5th week	1.97±0.02^{a}	1.94±0.02^{ab}	1.94±0.02^{ab}	1.90±0.02^{ab}	1.92±0.02^{ab}	1.89±0.02^{ab}	1.76±0.02^{c}	1.87±0.01^{b}	<0.000

Within the same row, different superscript indicates significantly different means
FCR: feed conversion ratio

Table 4: Effect of Lysine and Phytase Supplementation on Morphometric Characteristicsof Bone in Japanese Quails (Mean ± S.E)

	*GROUPS								
Parameters	Control (A)	Lysine 5g (B)	Lysine 8g (C)	Lysine 11g (D)	Phytse500units (E)	Lysine 5g+Phytse500units (F)	Lysine 8g+Phytse500units (G)	Lysine 11g+Phytse500units (H)	P-value
Bone weight (mg)	526.40±4.83^{f}	537.80±3.29^{def}	543.80±3.67^{def}	559.00±3.63^{de}	565.60±5.41^{cd}	587.00±3.63^{c}	699.60±5.24^{a}	634.20±8.22^{b}	<0.00
Bone length (mm)	48.82±0.23^{e}	49.57±0.15^{de}	50.44±0.12^{d}	51.48±0.24^{c}	52.64±0.22^{b}	53.11±0.35^{b}	54.91±0.09^{a}	53.46±0.12^{b}	<0.00
Medullary canal diameter (mm)	1.26±0.02^{d}	1.35±0.02^{cd}	1.37±0.05^{cd}	1.48±0.04^{c}	1.67±0.03^{b}	1.84±0.03^{a}	1.85±0.04^{a}	1.97±0.01^{a}	<0.00
Diaphysis diameter (mm)	2.21±0.04^{d}	2.35±0.04^{cd}	2.63±0.03^{bc}	2.75±0.05^{abc}	2.92±0.02^{ab}	2.92±0.07^{ab}	3.05±0.05^{a}	2.95±0.21^{ab}	<0.00
Tibio-tarsal Index	42.62±1.52	43.07±0.74	47.66±1.92	45.88±1.75	42.75±1.47	41.71±5.28	42.86±3.02	31.47±6.13	>0.07
Serum alkaline phosphatase	545.85±296.33	304.40±158.15	51.00±26.75	406.75±176.07	290.00±78.79	393.80±42.13	374.80±75.12	593.00±283.27	>0.583

Within the same row, different superscript indicates significantly different means

Parameters of Meat

Cooking and Drip Loss

The cooking loss of meat was lower significantly (P<0.05) in supplemented groups as compared to control group. The lowest values were observed in G-group.The drip loss of meat waslower (P>0.05) non significantly in supplemented groups as compared to control group except B-group and E-group which have the higher drip loss than control group. The lowest values were observed in G-group.

pH at Different hours

The PH of meat at 0 hour was lower (P<0.05) significantly in B-group, C-group, D-group, E-group and H-group. But G-group has the higher value than control group. And F-group almost has the same value to the control group.The PH of meat at 12 hour was lower (P>0.05) non significantly in supplemented groups as compared to control group. The lowest values were observed in D- group.

The Colors of Meat

The lightness of meat was lower significantly (P<0.05) in supplemented groups as compared to control group. Lowest values were observed in G-group as compared to the other groups. The yellowness of meat was significantly lower (P<0.05) in supplemented groups as compared to control group except C-group and H-group. Highest values were observed in C-group.

Serum Alkaline Phosphatase

The serum alkaline phosphatase level was not significant (P>0.05) among all the supplemented groups. Lowest values were observed in C-group as compared to the other groups.

All results are shown in table 5.

Table 5: Effect of Lysine and Phytase Supplementation on Meat Quality Parameters in Japanese Quails (Mean ± S.E)

	*GROUPS								
Parameters	Control	Lysine 5g	Lysine 8g	Lysine 11g	Phytse500units	Lysine 5g+Phytse500units	Lysine 8g+Phytse500units	Lysine 11g+Phytse500units	P-value
Cooking loss(gm)	0.22±0.02^{a}	0.19±0.03^{ab}	0.10±0.04^{ab}	0.16±0.04^{ab}	0.14±0.01^{ab}	0.09±0.02^{ab}	0.07±0.03^{ab}	0.21±0.04^{ab}	<0.011
Drip loss(gm)	1.42±0.13	1.80±0.24	1.22±0.18	1.36±0.24	1.58±0.23	1.32±0.25	1.16±0.12	1.35±0.25	>0.54
L*(Lightness)	46.20±0.87^{a}	44.81±0.32^{ab}	44.94±0.31^{ab}	43.98±0.45^{abc}	44.26±0.49^{abc}	42.87±0.64^{bc}	41.90±0.53^{c}	45.30±0.73^{ab}	<0.00
B*(Yellowness)	10.70±0.25^{b}	10.91±0.26^{ab}	12.14±0.45^{a}	10.68±0.38^{b}	10.46±0.26^{b}	10.12±0.24^{b}	10.10±0.20^{b}	11.40±0.21^{ab}	<0.00
pH at 0 hr	6.64±0.05^{a}	6.45±0.06^{b}	6.57±0.05^{ab}	6.56±0.02^{ab}	6.63±0.02^{a}	6.64±0.02^{a}	6.67±0.01^{a}	6.58±0.03^{ab}	<0.003
pH at 12 hr	5.76±0.05	5.44±0.07	5.55±0.06	4.88±0.52	5.44±0.06	5.46±0.06	5.37±0.06	5.46±0.07	>0.144
Muscle fiber diameter	17.60±.26^{e}	18.50±.47^{c}	20.40±.40^{d}	24.00±.36^{a}	25.20±.44^{a}	27.50±.22^{b}	30.70±.59^{a}	28.20±.41^{b}	<0.00
Muscle fascicle diameter	229.60±1.12^{d}	237.40±.84^{d}	246.30±.86^{c}	248.20±.74^{c}	234.10±4.86^{d}	250.70±.75^{c}	275.60±1.15^{a}	264.50±1.37^{b}	<0.00

Within the same row, different superscript indicates significantly different means

Discussion

The results offeed conversion ratio indicated that all the dietary inclusions of lysine and phytase significantly increased the feed conversion ratio in 4th and 5th week with better result observed in the group receiving microbial phytase in combination with 8g lysine. The results of body weight gain indicated that all the dietary inclusions of lysine and phytase are significant increased the body weight gain with best result observed in the group receiving microbial phytase in combination with 8g lysine. Comparable results of lysine supplementation were reported byErdaw et al. (2018) who reported that lysine supplemental groups had significantly improved weight gain and feed conversion ratio with best result observed in the group receiving 0.9% lysine with 23% crude protein. Comparable results of phytase supplementation were also reported by Yitbarek et al. (2016) who reported that phytase supplemental groups had significantly increased feed conversion ratio and body weight gain with best result observed in the group receiving phytase 500 units. Intestinal mucosa is responsible for the digestion and absorption of nutrients which regulate the growth in the animals.

The results of morphometric characteristics of tibia bone indicated that all the dietary inclusions of lysine and phytase improved the strength of tibia bone as indicated by increased length and weight and diaphysis and medullary canal diameter, with maximum effect observed in the group receiving microbial phytase in combination with 8g lysine. Comparable results of phytase supplementation were reported by Sarıca et al. (2025) who reported that phytase supplemental groups had significantly higher value of tibial bone weight and tibial bone length diaphysis diameter, medullary canal diameter and tibio-tarsal index than the control groups with the best results observed in the groups receiving 500 unit of phytase. It was found that phytase supplementation driven catalysis of the phytate hydrolysis improved the absorption of phosphorus from the intestine which in turn improved the gross characteristics of tibia bone (Jia et al. 2017). The tibio-tarsal index in groups receiving microbial phytase. We compare our results with results of Sedghi et al. (2024) who found that phytase supplemental groups had significantly higher diaphysis diameter and medullary canal diameter than control groups and non-significantly lower the tibio-tarsal index. There is an increased mineral absorption in intestine which improves the architecture of bone by increasing its strength and density. More deposition of minerals in bone was due to increasing intestinal absorptive surface area (Wang et al. 2021).

The results of serum alkaline phosphatase indicated that dietary inclusions of lysine and microbial phytase did not have significant effects on the level of serum alkaline phosphatase. The results of phytase supplementation were reported that phytase supplemental groups had increased value of serum alkaline phosphatase with maximum value observed in the group receiving 600 and 1200 phytase units.

Comparable results of lysine supplementation were reported by Paoletti et al. (2022) reported that lysine supplemental groups had decreased value of drip loss than control group with the best result observed in the group receiving 1.03% lysine. The results of cooking loss also indicated that all the dietary inclusions of lysine and phytase decreased the cooking loss associated with the quality of meat with minimum effect observed in the group receiving microbial phytase in combination with 8g lysine. Comparable results of lysine supplementation were also reported by Liao et al (2015) who reported that lysine supplemental groups had decreased in cooking loss than methionine groups with lowest cooking loss observed in the group receiving 1.216% lysine. It was found that lysine supplementation increased the solubility of myofibrillar : sarcoplasmic protein and decreased cooking loss (Pogorzelski et al. 2022). The results of muscle fiber diameter and muscle fascicle diameter indicated that all dietary inclusions lysine and phytase significant increased muscle fiber diameter and muscle fascicle diameter with maximum value observed in the group receiving microbial phytase with 8g lysine. It was recorded that deficiency in the lysine supplementation increased the fractional breakdown of muscles protein and resulted in the decreased in the synthesis of protein (Xiao et al. 2023).

The results of pH indicated that all the dietary inclusions of lysine and phytase significantly decreased pH at 0 hour and were not significant pH at 12 hour of slaughtering as indicated by pH

associated with the quality of meat which shows the preference for the consumer (Lomiwes, 2014)with minimum effect observed at 0 hour in the group receiving 5g lysine and at 12 hour in the group receiving microbial phytase 500 units. Comparable results of lysine supplementation were reported by Zhang et al. (2023) who reported that lysine supplemental groups had increased value of pH than control group with the best result observed in the group receiving 1.03% and 1.13% lysine. Comparable results of lysine supplementation were reported by Sarıca et al (2025) who reported that lysine supplemental groups had decreased the lightness and yellowness of meat with the best result observed in the group receiving 1.216% lysine. And this decrease in the lightness of meat indicated that improved the water holding capacity in meat and improved meat quality. The higher muscle pH preserves the myofibrils from degeneration and controlled the water loss from the gap between them.

Conclusion

From the above discussion, it was concluded that dietary supplementation lysine and microbial phytase individually and in combination microbial phytase in combination with 8g lysine generally improved meat quality and bone morphometric characteristics in Japanese quails.

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