

Investigation of the Effects of Different Levels of Dietary Clinoptilolite on Bone Development, Carcass Traits and Some Blood Parameters in Japanese Quails

Süleyman Ercüment Önel^{1,a,*}, Baran Erdem^{2,b}, Serkan İrfan Köse^{3,c}, Sema Alaşahan^{4,d}, Sevinç Ateş^{2,e}

¹Hatay Mustafa Kemal University, Faculty of Veterinary Medicine, Department of Animal Nutrition, Hatay, Türkiye

²Hatay Mustafa Kemal University, Faculty of Veterinary Medicine, Department of Anatomy, Hatay, Türkiye

³Hatay Mustafa Kemal University, Faculty of Veterinary Medicine, Department of Internal Medicine, Hatay, Türkiye

⁴Hatay Mustafa Kemal University, Faculty of Veterinary Medicine, Department of Animal Science, Hatay, Türkiye

^aORCID: 0000-0001-6599-0541; ^bORCID: 0000-0002-3319-6586; ^cORCID: 0000-0003-3189-6690;

^dORCID: 0000-0002-1144-7786; ^eORCID: 0000-0002-8381-0038

*Corresponding Author

E-mail: ercumentonel@gmail.com

Received: February 11, 2022

Accepted: April 20, 2022

Abstract

This study was aimed at determining the effects of dietary supplementation with 0%, 3%, 6% and 8% of clinoptilolite on live weight gain, carcass/carcass part traits and some blood and bone parameters in quails. For this purpose, forty 17-day-old Japanese quails (*Coturnix coturnix japonica*) were randomly assigned to four groups, each of 10 animals. The study groups were established as follows: BC (+0% clinoptilolite), B3C (3% clinoptilolite), B6C (6% clinoptilolite) and B8C (8% clinoptilolite). Live weight measurements were performed until 35 days of age, and after the measurement of the slaughter weight at 38 days of age, all animals were slaughtered. The live weight values measured in Groups B3C and B6C were similar to those of group BC, the control group. Even if slightly different, higher mean initial live weights in the males resulted in final live weights higher than those of the females. It was observed that the feed conversion rate had improved in the groups that received dietary clinoptilolite. The weight of abdominal fat was determined to have decreased with increasing dietary clinoptilolite levels. The weights of the edible visceral organs and abdominal fat were higher in the female quails, compared to the males. The osteometric values of the femur and humerus were higher in the males, compared to the females, in Group B3C. While calcium (Ca-CAL) and magnesium (Mg) levels were lowest in Group B8C, phosphorus (P-FOS) levels were lowest in groups BC and B3C. In result, it is suggested that dietary supplementation with clinoptilolite, up to a level of 6%, would not show any adverse effect on the fattening performance of animals. Thus, clinoptilolite can be used as a feed additive. Furthermore, good quality clinoptilolite supplied at low costs may also aid in preventing feed losses due to wetting, depending on the environmental conditions.

Keywords: Carcass parts, feed additive, humerus and femur, zeolite.

INTRODUCTION

Clinoptilolite is a natural zeolite. Zeolites are microporous minerals, which have a honeycomb-like spongy structure containing cations and water. In agriculture and animal production, zeolites are used for their water absorption capacity. A smaller particle size is associated with a greater water absorption capacity; and this feature is particularly important in soil and animal production. Zeolites are mainly used in the pharmaceutical industry, production of artificial fertilizers, construction industry, and water purification systems, and are also used as feed additives (Aksoy et al., 2018a). Initially used as a litter material in poultry production to prevent both foot pad lesions and breast meat damage, over time, zeolites have found use for multiple purposes, including performance enhancement, toxin binding, eggshell quality improvement, and owing to their low cost, dietary supplementation (Eleroglu and Yalcin, 2005; Schneider et al., 2017; Rajendran et al., 2020; Amad, 2021). A multitude of zeolites, comprising nearly 50 natural and 200 synthetic minerals, are available. Clinoptilolite is a highly preferred natural zeolite mineral, owing to its high absorption capacity, catalytic and dehydrating properties, and cation exchange capacity. Furthermore, it is also a

good pellet-binding and anti-caking agent for use in animal nutrition. Zeolites are reported to improve eggshell formation and bone development. It is indicated that zeolites improve eggshell quality by accelerating both the absorption of plasma calcium and the mobilization of bone calcium (Roland, 1990). They contain alkaline elements, including sodium, potassium, calcium, and magnesium (Celebi and Kaya, 2012). Berto et al. (2013) reported that the dietary supplementation of laying hens with 3% zeolite increased bone quality.

The present study was aimed at the investigation of the potential effects of clinoptilolite powder, incorporated into commercial broiler starter feed at different rates, on live weight, slaughter and carcass traits, some blood parameters, and the osteometric parameters of certain bones in quails.

MATERIALS AND METHODS

Creation of working groups

This study was conducted pursuant to the 27/04/2021 dated and 2021/03-02 numbered approval decision of the Local Ethics Board for Animal Experiments (HADYEK) of Hatay Mustafa Kemal University (MKU). The study was carried out at the premises of the Alternative Poultry

Cite this article as: Önel SE, Erdem B, Köse Sİ, Alaşahan S, Ateş S. 2022. Investigation of the Effects of Different Levels of Dietary Clinoptilolite on Bone Development, Carcass Traits and Some Blood Parameters in Japanese Quails. International Journal of Veterinary and Animal Research, 5(2): 94-101.

Breeding Unit of the Application and Research Centre for Scientific Research of MKU. Seventeen-day-old brown-feathered Japanese quails (*Coturnix coturnix japonica*) constituted the study material. Animals underwent macroscopic examination, and quail chicks with no body deformity were selected for use in the study. After being weighed individually, the quail chicks were randomly assigned to four groups, ensuring that the mean initial live weights of the groups were similar. Each group included 10 quail chicks of both sexes. Thus, in total, $4 \times 10 = 40$ quails were included in the study. The study groups were established as follows: Control-BC (Commercial chick starter feed + 0% Clinoptilolite), Basal3Clinoptilolite-B3C (Commercial chick starter feed + 3% Clinoptilolite), Basal6Clinoptilolite-B6C (Commercial chick starter feed + 6% Clinoptilolite) and Basal8Clinoptilolite-B8C (Commercial chick starter feed + 8% Clinoptilolite). The feed provided to the study groups was prepared on a weekly basis and was given *ad libitum*. Throughout the study period, the feeders and waterers of the animals were checked at least twice a day and were cleaned and/or added feed and water as required. The study was ended, when the quails reached 38 days of age.

The composition of the starter feed provided to the study groups and the nutritive values of the feedstuffs are presented in Table 1. The dry matter, crude ash, crude protein and crude fat analyses of the feedstuffs were performed according to the methodology of the Association of Official Analytical Chemists (AOAC, 1990). Crude cellulose levels were determined as described by Crampton and Maynard (1983), and metabolic energy levels (kcal/kg) were determined as described by Larbier and Leclercq (1994). The zeolite mineral, which was incorporated into the feed of the study groups and was of 0.3-0.7 mm particle size, was supplied from a commercial company (Rota Mining Corp., Turkey). The morphological and chemical properties of the dietary supplement, clinoptilolite, are given in Table 2.

Table 1. The composition of the mixed feed and nutritive values of the feedstuffs

Values determined by the firm	%	Values Determined by Analysis	%
Crude Protein %	20	Crude Protein %	23.7
Crude Cellulose %	3.4	Crude Cellulose %	4.1
Crude Ash %	4.9	Crude Ash %	4.96
Crude Fat %	3.8	Crude Fat %	6.17
Methionine %	0.51	Dry Matter %	89.55
Lysine %	1.13		
Calcium %	0.88	Starch %	34.4
Phosphorus %	0.44	Sugar %	3.3
Sodium %	0.14	Metabolic Energy, kcal/kg	2859.3

Table 2. Components of the zeolite mineral (clinoptilolite)

Mineral Components ¹	Rate (%)
Clinoptilolite	90.2
Cristobalite	3.88
Tridimite 4.45	4.45
Chemical Components ²	Rate (%)
SiO ₂	65.8
Al ₂ O ₂	10.45
CaO	2.42
K ₂ O	2.57
Fe ₂ O ₃	0.78
MgO	0.96
Na ₂ O	0.11
MnO	0.08
Cr ₂ O ₃	0.01
P ₂ O ₅	0.02
SiO ₂ /Al ₂ O ₃	5.43

¹A semi-quantitative whole rock analysis (stack mineralogy) was performed with the X-ray diffraction method (Gundogdu and Yilmaz, 1984). ²Analysed with an XRF spectrometer (Vendemiato and Enzweiler, 2001). The zeolite incorporated into the diets was supplied from a commercial company (Rota Mining Corp., Turkey) (Table 2) and analysed with XRF spectrometry, using the X-ray diffraction method (Gundogdu and Yilmaz, 1984; Vendemiato and Enzweiler, 2001). The particle size of the zeolite ranged between 0.3–0.7 mm.

The procedures applied throughout the fattening period

In this study, the quails included in the different study groups were individually weighed at seven-day intervals for the monitoring of live weight gain. Weight measurements were performed using a digital balance accurate to 0.01 g. The measurements aided in determining the weekly and daily live weight gains, which were used for the calculation of the feed conversion rates. The last live weight measurement was performed at 35 days of age, and once the slaughter weight was measured on day 38 of age, all of the animals were slaughtered.

The feed consumption of each study group was determined on a weekly basis. Weekly feed consumption was calculated based on the amount of feed provided to the groups at the beginning of the week and the feed remaining in the feeders at the end of the week. Feed conversion rates were calculated by dividing the feed consumption by the mean live weight gain.

With an aim to determine the slaughter and carcass traits, in view of the mean live weight of each study group on day 38, all animals in each group (10 both sexes) were sacrificed by cervical dislocation, and slaughtered. The

slaughter and carcass traits investigated in this study were live weight at slaughter, hot carcass weight with edible offal, hot carcass weight without edible offal, breast weight, leg weight, neck+back+wing weight, heart weight, liver weight, gizzard weight and abdominal fat weight. These weights were measured on a digital balance accurate to 0.01 g.

The percentages of the slaughter and carcass traits were determined as follows:

- Hot carcass yield with edible offal (%) = (Hot carcass weight with edible offal / Slaughter weight) x 100
- Hot carcass yield without edible offal (%) = (Hot carcass weight without edible offal / Slaughter weight) x 100
- Leg percentage (%) = (Leg weight / Hot carcass weight with edible offal) x 100
- Breast percentage (%) = (Breast weight / Hot carcass weight with edible offal) x 100
- Wing percentage (%) = (Wing weight / Hot carcass weight with edible offal) x 100
- Neck+back+wing percentage (%) = (Neck+back+wing weight / Hot carcass weight with edible offal) x 100
- Liver percentage (%) = (Liver weight / Hot carcass weight with edible offal) x 100
- Heart percentage (%) = (Heart weight / Hot carcass weight with edible offal) x 100
- Gizzard percentage (%) = (Gizzard weight / Hot carcass weight with edible offal) x 100
- Abdominal fat percentage (%) = (Abdominal fat weight / Hot carcass weight with edible offal) x 100

Blood samples were collected by decapitation at slaughter for biochemical analyses. The blood samples taken were centrifuged at 3000 rpm and + 4 °C for 5 min for the separation of sera, and the serum samples were stored at -24 °C until being analyzed. Anatomical examinations were performed on the humerus and the femur after each carcass was weighed. The soft tissue remaining on the bones was removed with the aid of a forceps and scalpel. The humerus and femur were measured for the width of their proximal and distal ends and mid-corpus, as well as for their length (Figure 1). Measurements were performed with a digital caliper, and were checked and confirmed using the ImageJ software (Version 1.53e).

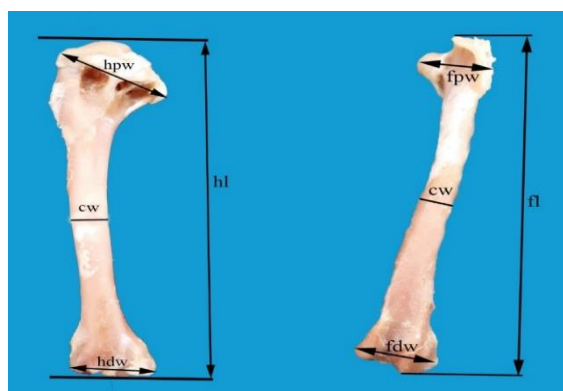


Figure 1. Measurement points of the humerus and femur in the Japanese quails. hl; length of humerus, hpw; width of the proximal end of the humerus, hdl; width of the distal end of the humerus, fl; length of the femur, fpw; width of the proximal end of the femur, fdw; width of the distal end of the femur, cw; width of the corpus.

Statistical analysis

Data analysis for the live weight values, slaughter and carcass traits, and blood and bone tissue values of the study groups was performed using the IBM SPSS Statistics 22 software. Whether the study group mean values differed for these parameters was determined using one-way analysis of variance (ANOVA). Duncan's multiple range test was used for the determination of any differences between the study groups.

RESULTS

The live weights measured for both sexes in the study groups are shown in Table 3. It was ascertained that the initial and weekly live weights of the study groups were similar, and the numerical differences between the groups were statistically insignificant ($P>0.05$). Throughout the study period, the weekly live weight values of Groups BC and B6C (females+males) were observed to be similar and higher than the mean values of the other two study groups ($P>0.05$). Until day 31 of age, the weekly live weights of the male quails in Group B3C were observed to be higher than those of the female quails in the other study groups, yet the differences between the groups were statistically insignificant (Table 3; $P>0.05$).

Table 3. The live weight values of the quails in the different study groups (g).

Group	Start of the Study - Day 17	Day 24	Day 31	Day 35
BC	74.69	127.32	173.55	193.60
B3C	75.53	124.64	171.22	191.10
B6C	73.27	121.04	169.28	192.57
B8C	72.49	119.83	163.62	184.63
OVERALL	73.99	123.21	169.42	190.48
SEM	1.751	2.224	2.584	3.041
P	0.927	0.625	0.575	0.726
For Sex				
BC Female	75.51	129.51	181.67	200.37
BC Male	74.15	125.87	168.13	189.09
B3C Female	73.71	121.66	168.72	190.47
B3C Male	79.76	131.61	177.05	192.58
B6C Female	74.97	128.96	176.65	193.20
B6C Male	70.72	109.15	158.23	191.64
B8C Female	71.76	124.68	174.72	194.58
B8C Male	73.21	114.99	152.52	174.68
OVERALL	73.99	123.21	169.42	190.48
SEM	1.891	2.183	2.415	3.166
P	0.983	0.233	0.075	0.684

The feed conversion rates of the study groups are shown in Table 4. The differences observed between the study groups for feed conversion rates in the selected time-periods were statistically significant ($P<0.001$, $P<0.01$). Throughout the fattening period, the feed conversion rates of Groups B3C, B6C and B8C were found to be better than that of Group BC.

Table 4. The feed conversion rates of the quails in the different study groups (g / g)

Groups	Start of Study – Day 14	Start of Study – Day 21
BC	2.93a	3.69a
B3C	2.80b	3.51b
B6C	2.66d	3.47b
B8C	2.69c	3.49b
OVERALL	2.77	3.54
SEM	0.004	0.017
P	<0.001	0.002

^{a,b,c,d} Values within a column with different superscripts differ significantly at $P < 0.01$.

The differences observed between the study groups for the slaughter and carcass traits investigated, except for the weight of abdominal fat ($P < 0.01$), were statistically insignificant ($P > 0.05$) (Table 5). While live weight at slaughter was higher in Group B6C, the weight of abdominal fat was higher in Groups BC and B3C ($P > 0.05$). Liver and gizzard weights were highest in Group B6C and lowest in Group B8C ($P > 0.05$). An assessment for the percentages of the carcass parts showed that the study groups significantly differed for the neck+back+wing percentage, heart percentage and abdominal fat percentage ($P > 0.05$).

Table 5. The slaughter and carcass traits of the quails in the different study groups

Group	BC	B3C	B6C	B8C	SEM	P
Live weight at slaughter (g)	206.32	205.62	210.33	204.26	3.685	0.945
Carcass weight with edible offal (g)	151.19	152.34	156.85	151.67	2.966	0.901
Carcass weight without edible offal (g)	124.38	126.73	129.59	125.11	2.136	0.831
Breast weight (g)	46.76	51.63	50.98	49.40	0.925	0.266
Neck+back+wing weight (g)	47.68	44.29	44.93	44.33	1.060	0.635
Leg weight (g)	29.56	30.45	32.08	31.11	0.549	0.437
Heart weight (g)	2.39	2.15	2.24	2.11	0.042	0.111
Liver weight (g)	5.95	5.91	6.29	5.79	0.306	0.946
Gizzard weight (g)	4.40	4.46	4.61	4.36	0.123	0.890
Abdominal fat weight (g)	1.71a	1.88a	1.10b	1.19b	0.086	0.005
Carcass yield with edible offal (%)	73.28	74.02	74.54	74.15	0.253	0.367
Carcass yield without edible offal (%)	60.39	61.56	61.87	61.30	0.403	0.603

^{a,b} Values within a row with different superscripts differ significantly at $P < 0.01$

The assessment of the slaughter and carcass traits of the male and female quails included in the different study groups (Table 6) demonstrated that values pertaining to the female quails were higher than those of the male quails. The differences between the study groups, particularly for heart weight, liver weight and abdominal fat weight, were

statistically significant ($P < 0.05$). The highest heart weight, liver weight and abdominal fat weight were determined in the females of Group BC, the females of Group B6C, and both the males and females of Groups BC and B3C, respectively.

Table 6. The slaughter and carcass traits of the male and female quails in the different study groups

Features	BC Female	BC Male	B3C Female	B3C Male	B6C Female	B6C Male	B8C Female	B8C Male	SEM	P
Live weight at slaughter(g)	217.70	198.73	207.69	200.79	219.95	195.89	219.52	189.00	3.524	0.202
Carcass weight with edible offal (g)	159.14	145.88	153.30	150.08	164.09	145.99	163.30	140.05	2.901	0.300
Carcass weight without edible offal (g)	129.18	121.18	126.85	126.44	132.88	124.66	133.00	117.20	2.168	0.522
Breast weight (g)	46.99	46.66	52.26	50.16	52.97	48.00	52.64	46.16	0.940	0.256
Neck+back+wing weight (g)	51.25	45.30	43.24	46.74	45.90	43.47	47.30	41.36	1.083	0.486
Leg weight (g)	30.82	28.75	30.85	29.50	33.00	30.70	32.82	29.40	0.562	0.395
Heart weight (g)	2.54 ^a	2.29 ^{abc}	2.18 ^{bc}	2.07 ^{bc}	2.39 ^{ab}	2.00 ^c	2.21 ^{abc}	2.01 ^c	0.040	0.028
Liver weight (g)	7.12 ^{ab}	5.17 ^{abc}	6.49 ^{abc}	4.54 ^c	7.51 ^a	4.46 ^c	6.79 ^{abc}	4.80 ^{bc}	0.267	0.024
Gizzard weight (g)	4.93	4.04	4.78	3.70	4.83	4.28	4.76	3.95	0.112	0.062
Abdominal fat weight (g)	1.98 ^a	1.52 ^{ab}	1.99 ^a	1.63 ^{ab}	1.14 ^b	1.03 ^b	1.36 ^{ab}	1.02 ^b	0.089	0.028
Carcass yield with edible offal (%)	73.21	73.33	73.74	74.67	74.55	74.52	74.22	74.08	0.274	0.813
Carcass yield without edible offal (%)	59.52	60.70	60.98	62.90	60.62	63.74	60.62	61.97	0.402	0.305

^{a,b,c} Values within a row with different superscripts differ significantly at $P < 0.05$.

The osteometric measurements of the proximal and distal ends and corpus of the humerus and femur of the Japanese quails are summarized in Table 7 and Table 8. The differences between the study groups were found to be statistically insignificant ($P>0.05$). While the values measured in the control group were lower than those measured in the groups, which received dietary clinoptilolite, this difference was statistically insignificant

($P>0.05$). Assessment for sex showed that the length of the femur was greater in the females, compared to the males, in Groups BC and B8C, whilst greater in the males, compared to the females, in Groups B3C and B6C ($P>0.05$). On the other hand, the length of the humerus was greater in the males, compared to the females, in only Group B3C ($P>0.05$).

Table 7. Osteometric values of the femur of the quails in the different study groups

Group	Proximal width of the femur (mm)	Distal width of the femur (mm)	Width of the mid-corpus (mm)	Length of the femur (mm)	F (mm)
BC	7.81	6.61	3.00	42.95	60.36
B3C	7.41	6.62	3.00	43.62	60.64
B6C	7.43	6.45	3.01	43.87	60.76
B8C	7.46	6.59	3.06	43.53	60.64
OVERALL	7.53	6.56	3.02	43.49	60.60
SEM	0.061	0.050	0.024	0.219	0.300
P	0.083	0.591	0.791	0.507	0.971
Sex					
BC Female	8.18	6.83	3.05	43.76	61.82
BC Male	7.56	6.46	2.97	42.41	59.39
B3C Female	7.35	6.54	3.01	43.53	60.43
B3C Male	7.53	6.80	2.99	43.82	61.14
B6C Female	7.36	6.47	3.00	43.42	60.25
B6C Male	7.54	6.41	3.03	44.54	61.52
B8C Female	7.47	6.58	3.11	43.92	61.09
B8C Male	7.45	6.60	3.01	43.14	60.19
OVERALL	7.53	6.56	3.02	43.49	60.60
SEM	0.060	0.051	0.026	0.224	0.301
P	0.053	0.448	0.890	0.421	0.501

Table 8. Osteometric values of the humerus of the quails in the different study groups

Group	Proximal width of the humerus (mm)	Distal width of the humerus (mm)	Width of the mid-corpus (mm)	Length of the humerus (mm)	H (mm)
BC	9.87	6.36	2.81	39.18	58.22
B3C	10.23	6.37	2.91	39.53	59.05
B6C	10.32	6.42	2.93	39.54	59.20
B8C	10.39	6.61	2.90	39.52	59.41
OVERALL	10.20	6.44	2.89	39.44	58.97
SEM	0.092	0.058	0.036	0.209	0.299
P	0.221	0.399	0.654	0.912	0.523
For sex					
BC Female	10.25	6.53	2.90	39.66	59.33
BC Male	9.62	6.24	2.75	38.87	57.48
B3C Female	10.22	6.29	2.90	39.59	59.00
B3C Male	10.27	6.55	2.94	39.41	59.18
B6C Female	10.57	6.31	2.92	39.58	59.39
B6C Male	9.94	6.57	2.93	39.48	58.92
B8C Female	10.60	6.77	2.89	39.91	60.18
B8C Male	10.18	6.44	2.91	39.13	58.65
OVERALL	10.20	6.44	2.89	39.44	58.97
SEM	0.091	0.058	0.039	0.223	0.309
P	0.119	0.267	0.913	0.944	0.508

Some blood parameters of the quails in the different study groups are shown in Table 9. The differences between the study groups for the blood parameters investigated were statistically insignificant ($P>0.05$). However, assessment for sex demonstrated that the study groups significantly differed for Ca and Mg levels

($P<0.05$, $P<0.01$), such that these levels were higher in the female quails, compared to the males. Ca levels were lowest in the males of Group B8C and highest in the females of Group B8C. On the other hand, the highest Mg levels were detected in the females of Group BC and the males of Group B6C.

Table 9. Serum parameters of the different study groups.

Group	Alkaline Phosphatase (ALP) U/L	UREA mg/dl	Total Protein (TP) g/dl	ALBUMIN (ALB) g/dl	GLOBULIN (GLB) g/dl	ALB/GLB	Ca mg/dl	Mg mEq/L	P mg/dl
BC	3567.70	4.700	3.630	1.224	2.406	0.51	12.679	2.410	6.714
B3C	3324.80	5.800	3.360	1.288	2.072	0.81	13.146	2.340	6.628
B6C	3961.80	6.400	3.640	1.176	2.464	0.49	13.057	2.580	9.605
B8C	3864.90	5.700	3.650	1.234	2.416	0.52	11.912	2.330	8.326
OVERALL	3679.80	5.650	3.570	1.231	2.340	0.585	12.699	2.415	7.818
SEM	200.66	0.382	0.127	0.045	0.097	0.063	0.604	0.088	0.592
P	0.670	0.475	0.823	0.850	0.474	0.231	0.884	0.729	0.246
For Sex									
BC Female	3755.75	6.000	3.900	1.368	2.533	0.558	14.813 ^a	2.875 ^a	7.660
BC Male	3442.33	3.833	3.450	1.128	2.322	0.481	11.267 ^{ab}	2.100 ^{bc}	6.083
B3C Female	3101.20	6.600	3.420	1.344	2.076	1.034	14.804 ^a	2.740 ^{ab}	7.696
B3C Male	3548.40	5.000	3.300	1.232	2.068	0.595	11.488 ^{ab}	1.940 ^c	5.660
B6C Female	4412.50	7.000	3.983	1.325	2.658	0.512	14.237 ^a	2.850 ^a	11.730
B6C Male	3285.75	5.500	3.125	0.953	2.173	0.463	11.288 ^{ab}	2.175 ^{bc}	6.418
B8C Female	3722.25	6.750	3.775	1.238	2.538	0.492	15.765 ^a	2.600 ^{ab}	8.840
B8C Male	3960.00	5.000	3.567	1.232	2.335	0.539	9.343 ^b	2.150 ^{bc}	7.983
OVERALL	3679.80	6.5650	3.570	1.231	2.340	0.585	12.699	2.415	7.818
SEM	208.70	0.381	0.130	0.044	0.102	0.065	0.530	0.072	0.582
P	0.794	0.357	0.711	0.389	0.737	0.402	0.039	0.006	0.176

^{a,b,c} Values within a column with different superscripts differ significantly at $P < 0.01$; $P < 0.05$.

DISCUSSION AND CONCLUSION

In the present study, it was ascertained that the alterations in the weekly and daily live weights of the different study groups (Table 3) were similar. Furthermore, it was observed that increased levels of dietary zeolite were associated with lower live weight values. Thus, it is important that dietary supplementation with zeolite does not exceed a level of 6% to ensure normal live weight gain in quails. In a study by Banaszak et al. (2020) on dietary supplementation with an aluminosilicate clay mineral, halloysite, and zeolite, the highest live weights on days 14, 28 and 42 were determined in the group that received 1% of dietary zeolite, yet the difference was statistically insignificant ($P > 0.05$). Yeter and Gokce (2018) divided the 42-day fattening period of hen chicks into four periods, which covered the intervals between days 1-14, 15-21, 22-35 and 36-42, and incorporated zeolite into the basal feed at levels of 0.5%, 0.75%, 1%, 1.25% (first zeolite group) and 1%, 1.5%, 2% and 2.5% (second zeolite group) to investigate whether increased levels of dietary zeolite improved live weight gain in broiler chickens, and eventually found no such positive effect of zeolite on live weight ($P > 0.05$). In a study by Tufan et al. (2014), in which quail basal feed was added 2%, 4% and 6% of clinoptilolite, no positive effect was observed on live weight alterations, such that the groups that received 4% and 6% of dietary clinoptilolite displayed live weights lower than that of the control group. In this respect, the results of the present study are in agreement with those reported in previous research on zeolites.

It is known that female quails have a greater body weight than male quails. In the present study, female quails reached a final live weight greater than that of male quails,

but this numerical difference was statistically insignificant (Table 3). Furthermore, in Group B3C, in which the male quails had initial live weights greater than those of the female quails, the males displayed higher live weights throughout the study until slaughter, which were close to those of the female controls. In a study in quails, Alasahan and Copur (2016) reported that the females weighed heavier than the males, and that the live weight difference between the two sexes gained significance as of 3 weeks of age.

In the present study, the feed conversion rates of the groups that received dietary clinoptilolite were better than those of the control quails in Group BC, which were fed on the basal ration alone (Table 4). Owing to its water absorption capacity, clinoptilolite reduces the humidity of feed and prevents it from sticking to the surface of the feeders and the feet and beak of animals. In return, both feed consumption and the feed conversion rate are reduced, which is for better. Ayed et al. (2008) reported that the incorporation of 0.5%, 1% and 2% of the clay mineral sepiolite into feed positively affected the feed conversion rate of broiler chicks, and indicated that the best results were achieved with 2% of dietary sepiolite. On the other hand, upon incorporating 1%, 3% and 5% of clinoptilolite into broiler feed, Eleroglu et al. (2011) reported to have observed no statistically significant difference for the feed conversion rate, with the best rates having been achieved in the controls and the group given 5% of dietary clinoptilolite. Similarly, Durak et al. (2017) determined no statistically significant difference in the feed conversion rates of quails, which were given 2.5% and 5% of dietary zeolite.

In the present study, when compared to the control group, the most adversely affected slaughter and carcass traits were determined in Group B8C (Table 5). Dietary supplementation with 3% and 6% of zeolite improved the weights of the carcass and carcass parts, but adversely affected the weight of abdominal fat. In their study in quails, Al-Tikriti and Al-Basha (2020) determined that dietary supplementation with 2%, 3% and 4% of zeolite improved the weights of the carcass and carcass parts, and reported the best results to have been achieved with the incorporation of 3% of zeolite into feed. In their research on two groups of ducks, one provided with dietary zeolite and the other not provided with dietary zeolite, Biesek et al. (2021) ascertained that the carcass weight, heart weight and gizzard weight were higher and the weight of abdominal fat was lower in the group, which received dietary zeolite.

In the present study, no statistically significant difference was determined between the study groups for the length and distal width of the humerus and the length and proximal width of the femur (Tables 7 and 8). In a study conducted by Demiraslan et al. (2014), in which 100 female quails were provided with 2%, 4% and 6% of dietary clinoptilolite, statistically significant differences were detected between the study groups for the length and distal width of the humerus and the length and proximal width of the femur. The results of the present study differing from those reported by these researchers (Demiraslan et al. 2014) was attributed to the difference in the numbers of animals included in these two studies. Nevertheless, both studies demonstrated that the proportional increase of dietary zeolite levels did not result in linear alterations in the osteometric values.

In the present study, the numerical alterations that occurred in the blood serum parameters with dietary zeolite supplementation did not reflect any statistically significant positive effect. However, the comparison of the male and female quails showed that, in particular the Ca and Mg levels were higher in the females, compared to the males (Table 9). While it has been suggested that zeolite reacts with phosphorus, and thereby reduces both the absorption and utilization of phosphorus (Aksoy et al., 2018b), no such effect was observed for zeolite in the present study. This could be related to the short time period and small sample size of the present study. The mean blood serum Ca level (12.699 mg/dl) determined in the present study is similar to the levels reported in previous research (Kabir, 2013; Jeevalakshmi et al., 2017; Saki et al., 2017), and falls within the reference range reported by Jeevalakshmi et al. (2017). While the blood calcium levels of the female quails were observed to be above the general average, they were still close to the levels of the control group. In view of the molecular structure of zeolite containing Ca, Mg and P, it was observed that dietary supplementation with 8% of zeolite led to a decrease in male and an increase in female blood Ca levels, whilst no significant alteration in male and a decrease in female Mg levels. It is indicated that increased levels of Mg in the ration increase the excretion of endogenous Mg (Ozek, 2016). Thus, the decrease observed, in the present study, in the Mg levels of the females was considered to may have occurred in relation to the Mg content of zeolite. Studies are available, which have shown the impact of zeolite on intestinal villi (Wu et al., 2013; Tufan et al., 2014). In view of this literature information, the differences observed between male and female quails for intestinal absorption, the elimination of minerals from the body via the kidneys,

and zeolite being capable of forming chelates with minerals were also considered to be involved in sex-related differences. However, a general assessment pointed out to dietary supplementation with 3%, 6% and 8% of zeolite not affecting the metabolism of the minerals Ca, Mg and P. A limitation of the present study was the composition of bone tissue not having been tested for these minerals (Ca, Mg and P).

Live weight gain in the study groups that received 3% and 6% of dietary clinoptilolite having been determined to be similar to that of the controls fed on the basal ration alone was considered to be significant within the context of not only reducing feed losses associated with high feed humidity levels, but also reducing feed costs. Furthermore, in the groups that were given 3% and 6% of clinoptilolite, the male quails were observed to display growth traits similar to those of the females. Dietary clinoptilolite was also observed to improve the feed conversion rate. Therefore, in quails, which are known to frequently attend feeders throughout the fattening period, both maintaining feed quality and minimizing feed losses are highly important. Dietary supplementation with 3-6% of clinoptilolite was concluded to be favourable for improving growth and slaughter traits, as well as for protecting quail health and achieving cost-effectiveness.

Conflict of Interest

The authors declare that they have no competing interests.

Authorship contributions

Concept: S.E.Ö, S.A., Design: S.E.Ö, S.A., Data Collection or Processing: S.E.Ö, B.E, S.İ.K., S.A., S.A. Analysis or Interpretation: S.A., Literature Search: S.E.Ö, B.E, S.İ.K., S.A., S.A. Writing: S.E.Ö, B.E, S.İ.K., S.A., S.A.

Financial Support

This research received no grant from any funding agency/sector.

REFERENCES

- Alasahan S, Copur AG. 2016. Hatching characteristics and growth performance of eggs with different egg shapes. *Brazilian Journal of Poultry Science*, 18(1): 001-008.
- Al-Tikriti SSA, Al-Basha SB. 2020. Addition of different level of zeolite powder on Japanese quail bird feed and its effects on carcass parameters. 2nd International Scientific Conference of Al-Ayen University (ISCAU-2020), 2020 IOP Conf. Ser.: Mater. Sci. Eng. 928 062001.
- Aksoy G, Avci M, Biricik HS, Kaplan O, Yerturk M. 2018a. Effect of zeolite mineral on animal welfare in natural shelters, sanliurfa region. *Harran Üniversitesi Veteriner Fakültesi Dergisi*, 7(2): 200-206.
- Aksoy G, Biricik HS, Avci M, Das A. 2018b. Prevention of left displacements of the abomasum in cattle by adding zeolite mineral to feeds. *Harran Üniversitesi Veteriner Fakültesi Dergisi*, 7(1): 32-39.
- Amad A. 2021. The effect of natural zeolite as feed additive on performance and egg quality in old laying hens. *Journal of Poultry Research*, 18(1): 13-18.
- AOAC. Official Methods of Analysis Association of Agricultural Acedemy Press. Ninth Revised Ed. Washington DC. 1990.
- Ayed MH, Zghal I, Rekik B. 2008. Effect of sepiolite supplementation on broiler growth performances and

carcass yield. Proceedings, Western Section, American Society of Animal Science, 59: 169-172.

Banaszak M, Biesek J, Bogucka J, Dankowiakowska A, Olszewski D, Bigorowski B, Grabowicz M, Adamski M. 2020. Impact of aluminosilicates on productivity, carcass traits, meat quality, and jejunum morphology of broiler chickens. Poultry Science, 99:7169–7177.

Berto DA, Garcia EA, Vercese F, Santos GC dos, Barreiro FR, Molino A de B, Pelicia K, Silveira AF da, 2013. Effects of dietary clinoptilolite and calcium levels on uric acid and calcium blood concentrations and bone quality of commercial layers. Brazilian Journal of Poultry Science, 15(2): 145-150.

Biesek J, Banaszak M, Adamski M. 2021. Ducks' growth, meat quality, bone strength, and jejunum strength depend on zeolite in feed and long-term factors. Animals, 11: 2-16.

Celebi S, Kaya A. 2012. The Use of Zeolite in Laying Hens and Broilers Diets. Hayvansal Üretim, 53(2): 40-48.

Crampton EW, Maynard L. 1983. The relation of cellulose and lignin chromatography method for the simultaneous analysis of plasma retinol, α -tocopherol and various carotenoids. Analytical Biochemistry, 138-340.

Demiraslan Y, Tufan T, Sari M, Akbulut Y, Dayan MO, Kukurt A. 2014. The effect of clinoptilolite on long bone morphometry in Japanese quail (*Coturnix coturnix japonica*). Animal and Veterinary Sciences, 2(6): 179-183.

Durak MH, Bayril T, Simsek A, Alak I, Gokalp E, Gurgoze S. 2017. Dietary Zeolite, Fattening Performance and its Effect on Some Biochemical Parameters in Japanese Quail. Harran Üniversitesi Veteriner Fakültesi Dergisi, 6(1): 1-5.

Eleroglu H, Yalcin H. 2005. Use of natural zeolite-supplemented litter increased broiler production. South African Journal of Animal Science, 35(2): 90-97.

Eleroglu H, Yalcin H, Yildirim A, Aker A. 2011. The effects of dietary natural zeolite supplementation on performance of broiler. Hayvansal Üretim, 52(1): 24-32.

Gundogdu, MN, Yilmaz O. Clay mineralogy, I. Nat Clay Symp, Cukurova Univ., Adana, Proc. 1984; 319-330.

Jeevalakshmi. A, Sathya. C, Murugaian. P. 2017. Effect of chicory and inulin growth performance, serum biochemical analysis and intestinal microbial population in Japanese quail (*coturnix japonica*). International Journal of Advanced Research, 5(7): 699-707.

Kabir A. 2013. Blood chemistry analyses of Japanese quail (*Coturnix coturnix japonica*). Scholarly Journal of Agricultural Science, 3(4): 132-136.

Larbier M, Leclercq B. 1994. Nutrition and feeding of poultry. Nottingham (UK), Nottingham University Press.

Ozek K. 2016. Magnesium's functions and metabolism in poultry nutrition. Iğdır University Journal of The Institute of Science and Technology, 6(2): 165-173.

Saki AA, Goudarzi SM, Ranjbaran M, Ahmadi A, Khoramabadi V. 2017. Evaluation of biochemical parameters and productive performance of Japanese quail in response to the replacement of soybean meal with canola meal. Acta Scientiarum. Animal Sciences, 39: 51-56.

Schneider AF, Zimmermann OF, Gewehr CE. 2017. Zeolites in poultry and swine production. Ciência Rural, Santa Maria, 47(08), e20160344: 1-8.

Rajendran RM, Umesh B, Chirakkal H. 2020. Assessment of H- β zeolite as an ochratoxin binder for poultry. Poultry Science, 99: 76-88.

Roland DA, Sr Rabon HW, Frost TJ, Laurent SM, Barnes DG. 1990. Response of commercial Leghorns to sodium aluminosilicate when fed different levels and sources of available phosphorus. Poultry Science, 69(12): 2157-2164.

Tufan T, Arslan C, Sari M. 2014. Effects of clinoptilolite supplementation to Japanese quail diet on growth performance, carcass traits and some blood parameters. Journal of Lalahan Livestock Research Institute, 54(1):21-27.

Vendematto MA, Enzweiler J. 2001. Routine control of accuracy in silicate rock analysis by X-ray fluorescence spectrometry. Geo. News., 25/2-3: 283-291.

Wu Q. J., Wang LC, Zhou YM, Zhang JF, Wang T. 2013. Effects of clinoptilolite and modified clinoptilolite on the growth performance, intestinal microflora, and gut parameters of broilers. Poultry Science, 92(3): 684-692.

Yeter B, Gokce G. 2018. Determination of growth performances with addition of increased concentration zeolite (clinoptylolide) in broiler. KSÜ Tarım ve Doğa Dergisi, 21(5): 745-750.