

Chewing lice (Phthiraptera) Found on Wild Birds in Turkey

Bilal DİK *  Elif ERDOĞDU YAMAÇ ** Uğur USLU *

* Selçuk University, Veterinary Faculty, Department of Parasitology, Alaeddin Keykubat Kampusü, TR-42075 Konya - TURKEY

** Anadolu University, Faculty of Science, Department of Biology, TR-26470 Eskişehir - TURKEY

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Summary

This study was performed to detect chewing lice on some birds investigated in Eskişehir and Konya provinces in Central Anatolian Region of Turkey between 2008 and 2010 years. For this aim, 31 bird specimens belonging to 23 bird species which were injured or died were examined for the louse infestation. Firstly, the feathers of each bird were inspected macroscopically, all observed louse specimens were collected and then the examined birds were treated with a synthetic pyrethroid spray (Biyo avispray-Biyoteknik®). The collected lice were placed into the tubes with 70% alcohol and mounted on slides with Canada balsam after being cleared in KOH 10%. Then the collected chewing lice were identified under the light microscope. Eleven out of totally 31 (35.48%) birds were found to be infested with at least one chewing louse species. Eighteen lice species were found belonging to 16 genera on infested birds. Thirteen of 18 lice species; *Actornithophilus piceus piceus* (Denny, 1842); *Anaticola phoenicopteri* (Coincide, 1859); *Anatoecus pygaspis* (Nitzsch, 1866); *Colpocephalum heterosoma* Piaget, 1880; *C. polonum* Eichler and Zlotorzyska, 1971; *Fulicoffula lurida* (Nitzsch, 1818); *Incidifrons fulicia* (Linnaeus, 1758); *Meromenopon meropis* Clay ve Meinertzhagen, 1941; *Meropoecus meropis* (Denny, 1842); *Pseudomenopon pilosum* (Scopoli, 1763); *Rallicola fulicia* (Denny, 1842); *Saemundssonsonia lari* Fabricius, O, 1780), and *Trinoton femoratum* Piaget, 1889 have been recorded from Turkey for the first time.

Keywords: Chewinglice, *Anaticola*, *Fulicoffula*, *Incidifrons*, *Pseudomenopon*, *Meropoecus*, *Meromenopon*, *Rallicola*, *Colpocephalum*, *Actornithophilus*, *Saemundssonsonia*, *Trinoton*, Turkey

Türkiye'deki Yabani Kanatlılarda Bulunan Çiğneyici Bit (Phthiraptera) Türleri

Özet

Bu çalışma 2008-2010 yılları arasında Türkiye'nin İç Anadolu Bölgesinde yer alan Konya ve Eskişehir'deki bazı yabani kanatlıların bitlerini belirlemek amacıyla yapılmıştır. Bu amaçla 23 kanatlı hayvan türüne ait 31 ölü veya yaralı kuş örneği bit yönünden incelenmiştir. Kuşların tüyleri makroskopik olarak dikkatli bir şekilde incelenmiş, daha sonra sentetik pretroidli (Biyo avispray-Biyoteknik®) bir insektisit ile ilaçlanmıştır. Toplanan bitler içinde %70 alkol bulunan tüplere konulup %10 KOH'de saydamlaştırıldıktan sonra Kanada balsam ile lamlara yapıştırılarak ışık mikroskopunda teşhis edilmişlerdir. İncelenen 31 kuşun 11 (35,48%)'i en az bir bit türü ile enfeste bulunmuştur. Enfeste kuşlarda 16 cinse ait 18 bit türü tespit edilmiştir. Bu türlerden 13'ü; *Actornithophilus piceus piceus* (Denny, 1842); *Anaticola phoenicopteri* (Coincide, 1859); *Anatoecus pygaspis* (Nitzsch, 1866); *Colpocephalum heterosoma* Piaget, 1880; *C. polonum* Eichler and Zlotorzyska, 1971; *Fulicoffula lurida* (Nitzsch, 1818); *Incidifrons fulicia* (Linnaeus, 1758); *Meromenopon meropis* Clay ve Meinertzhagen, 1941; *Meropoecus meropis* (Denny, 1842); *Pseudomenopon pilosum* (Scopoli, 1763); *Rallicola fulicia* (Denny, 1842); *Saemundssonsonia lari* Fabricius, O, 1780) ve *Trinoton femoratum* Piaget, 1889 Türkiye'den ilk kez bu çalışmayla bildirilmiştir.

Anahtar sözcükler: Çiğneyici bit, *Anaticola*, *Fulicoffula*, *Incidifrons*, *Pseudomenopon*, *Meropoecus*, *Meromenopon*, *Rallicola*, *Colpocephalum*, *Actornithophilus*, *Saemundssonsonia*, *Trinoton*, Türkiye

INTRODUCTION

Chewing lice (Ishnocera, Amblycera) are permanent obligate ectoparasites mostly parasitic on bird species and they feed on feathers and skin scales. Although they have

mouthparts designed for chewing, some of the species of Ishnocera can cause skin irritations and suck blood. Chewing lice have harmful effects that lead to decrease in



İletişim (Correspondence)



+90 332 2232736



bdk@selcuk.edu.tr

productivity on the host.

About 4.000 lice species were recorded on birds in the world ¹. The number of louse species recorded from birds in Turkey is limited ². On the other hand, the studies on the identification of lice on bird species have been increasing over the last years in Turkey. Approximately 50 new records for the bird louse fauna of Turkey have been recorded in recent 10 years. Up today 82 chewing lice were described from birds in the limited studies in Turkey ².

In some of these studies, most chewing lice were collected from injured or died birds that were found accidentally ³⁻⁹. On the other hand, recent studies have focused on louse species of Songbirds (Passeriformes) ¹⁰ and Shorebirds (Charadriiformes) ¹¹. In spite of the records on chewing lice on birds have already indicated limited number of louse species, the studies on the lice have a big potential for new discoveries. The objective of this study was to investigate and identify louse species on birds and provide data on the chewing lice list from Turkey.

MATERIAL and METHODS

This study was performed in Eskişehir and Konya provinces between 2008 and 2010 years. The collection of chewing lice was carried out from 31 birds (of 23 species

in 19 genera, 16 families and 9 orders) that were found to be injured or died in the field (*Table 1*). In order to collect the lice, the feathers of each bird were carefully examined and rubbed over a white piece of paper with a synthetic pyrethroid insecticide (Biyo avispray®: 0.09 g tetramethrin and 0.45 g piperonyl butoxide 100 g (w/w); Biyoteknik Chemistry, Turkey) harmless to vertebrates. Then the birds were placed in breathable paper bags for 15 min. All lice preserved in 70% alcohol, cleared in 10% KOH, then washed with distilled water, passed in alcohol series 70%, 80%, 90% and 99% in four consecutive days. They were mounted on slides in Canada balsam and examined by a light microscope. The identification of the louse species was carried out according to relevant literatures ^{1,12-28}.

RESULTS

Eleven out of totally 31 (35.48%) examined birds were found to be infested with at least one chewing louse species and 18 lice species from 16 genera were identified. Although, no louse was found on 9 birds of 8 species belong to Passeriformes and Strigiformes, 11 of 22 birds belong to 15 species in 7 orders were parasitized (*Table 2*).

Ischnocera

Anaticola phoenicopteri: Examined materials: 14 ♀, 10

Table 1. Studied bird orders, families, genera and species

Tablo 1. İncelenen kuş takım, aile, cins ve türleri

Order	Family	Genus	Species	Number of Examined Bird
Ciconiiformes	Ciconiidae	<i>Ciconia</i>	<i>C. ciconia</i>	2
Phoenicopteriformes	Phoenicopteridae	<i>Phoenicopiterus</i>	<i>P. ruber</i>	1
Falconiformes	Accipitridae	<i>Accipiter</i>	<i>A. gentilis</i>	1
		<i>Circus</i>	<i>C. gallicus</i>	1
		<i>Buteo</i>	<i>B. buteo</i>	1
			<i>B. rufinus</i>	3
	Falconidae	<i>Falco</i>	<i>F. columbarius</i>	1
Gruiformes	Rallidae	<i>Fulica</i>	<i>F. atra</i>	3
Charadriiformes	Laridae	<i>Larus</i>	<i>L. genei</i>	1
	Recurvirostridae	<i>Himantopus</i>	<i>H. himantopus</i>	1
	Scolopacidae	<i>Calidris</i>	<i>C. alba</i>	1
		<i>Tringa</i>	<i>T. totanus</i>	1
	Sternidae	<i>Chlidonias</i>	<i>C. leucopterus</i>	1
Columbiformes	Columbidae	<i>Columba</i>	<i>C. livia</i>	3
Strigiformes	Strigidae	<i>Asio</i>	<i>A. otus</i>	1
Coraciiformes	Meropidae	<i>Merops</i>	<i>M. apiaster</i>	1
Passeriformes	Fringillidae	<i>Carduelis</i>	<i>C. carduelis</i>	2
		<i>Loxia</i>	<i>L. curvirostra</i>	1
		<i>Serinus</i>	<i>S. canaria</i>	1
	Paridae	<i>Parus</i>	<i>P. caeruleus</i>	1
	Passeridae	<i>Passer</i>	<i>P. domesticus</i>	1
	Sylviidae	<i>Sylvia</i>	<i>S. melanocephala</i>	1
	Turdidae	<i>Luscinia</i>	<i>L. megarhynchos</i>	1
Total				31

♂, 10 N, 03.07.2010, Akşehir, Konya (Fig. 1). Host: Greater Flamingo (*Phoenicopterus ruber*). It was reported for the first time in Turkey.

Anatoecus pygaspis: Examined materials: 5 ♀, 3 ♂, 03.07.2010, Akşehir, Konya (Fig. 2, 3). Host: Greater Flamingo (*Phoenicopterus ruber*). It was reported for the first time in Turkey.

Ardeicola ciconiae: Examined materials: 2 ♀, 1 N, 07.09.2009, Konya Metropolitan Municipality Animal Shelter, Selçuklu-Konya; 2 ♀, 1 ♂, 1 N, 01.04.2010, Selçuk University, Clinics of Veterinary Faculty, Konya. Host: White Stork (*Ciconia ciconia*).

Columbicola columbae: Examined materials: 1 ♀, 1 N, 25.09.2009; 2 ♀, 18.11.2009 Konya. Host: Rock Pigeon

(*Columba livia*).

Fulicoffula lurida: Examined materials: 7 ♀, 3 N, 13.08.2010-1, Akşehir, Konya, 9 ♀, 5 N 13.08.2010-2, Akşehir, Konya (Fig. 4). Host: Eurasian Coot (*Fulica atra*). It was reported for the first time in Turkey.

Incidifrons fulica: Examined materials: 1 ♂, 13.08.2010-1, Akşehir, Konya (Fig. 5). Host: Eurasian Coot (*Fulica atra*). It was reported for the first time in Turkey.

Meropoecus meropis: Examined materials: 2 N, 13.08.2010, Eskişehir (Fig. 6). Host Bee-eater (*Merops apiaster*). It was reported for the first time in Turkey.

Neophiloaterus incompletus: Examined materials: 1 ♀, 1 ♂, 07.09.2009, Konya; 1 ♀, 1 N, 01.04.2010, Konya. Host:

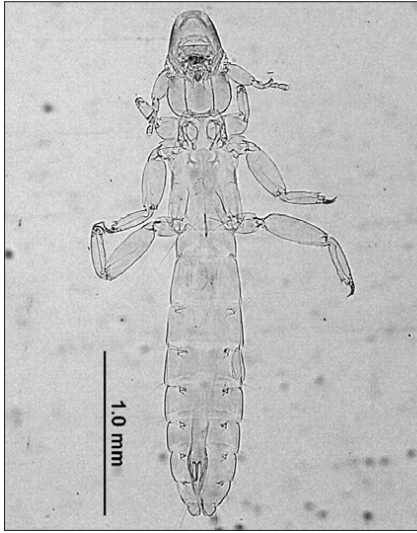


Fig 1. *Anaticola phoenicopteri*, male, orig.

Şekil 1. *Anaticola phoenicopteri*, erkek, orijinal

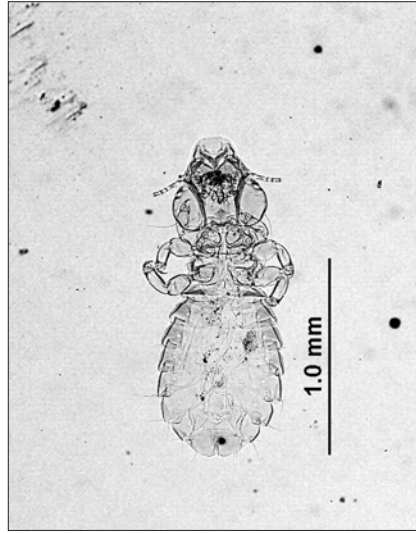


Fig 2. *Anatoecus pygaspis*, female, orig.

Şekil 2. *Anatoecus pygaspis*, dişi, orijinal

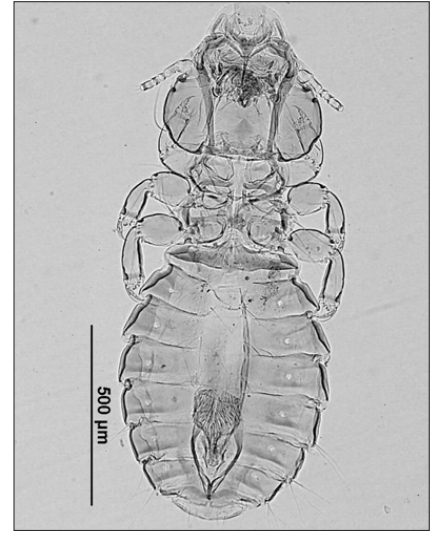


Fig 3. *Anatoecus pygaspis*, male, orig.

Şekil 3. *Anatoecus pygaspis*, erkek, orijinal

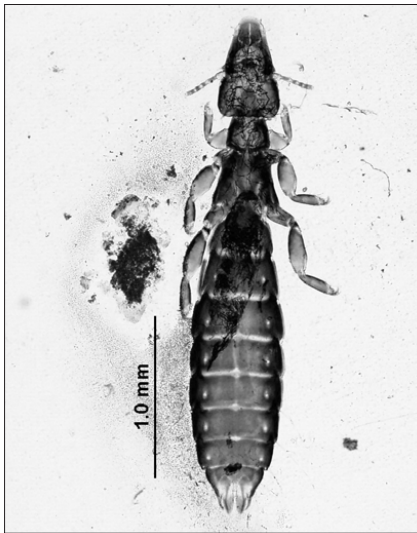


Fig 4. *Fulicoffula lurida*, female, orig.

Şekil 4. *Fulicoffula lurida*, dişi, orijinal

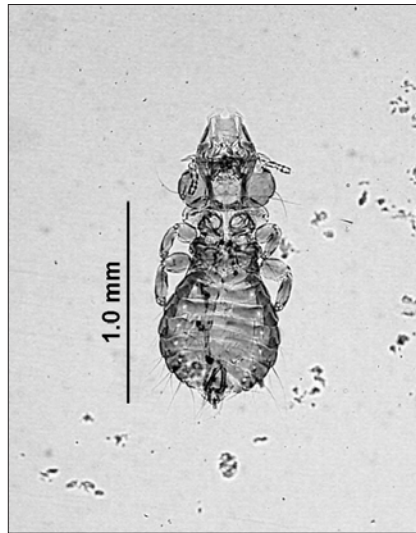


Fig 5. *Incidifrons fulicae*, male, orig.

Şekil 5. *Incidifrons fulicae*, erkek, orijinal

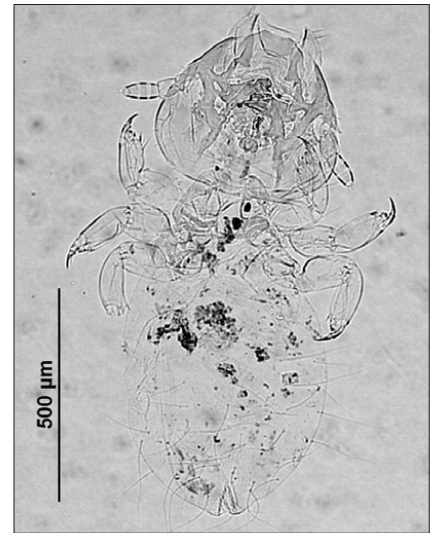


Fig 6. *Meropoecus meropis*, nymph, orig.

Şekil 6. *Meropoecus meropis*, nimf, orijinal

Table 2. Studied bird species, infestation rates and louse species on avian host**Table 2.** İncelenen kuş türleri, enfestasyon oranları ve saptanan bit türleri

Bird Species	Date/Locality	Number of Examined Bird	Number of Infested Bird	Louse Species
Nightingale (<i>Luscinia megarhynchos</i>)	10.05.2008 Beyyayla/Eskişehir (live)	1	-	-
Sardinian Warbler (<i>Sylvia melanocephala</i>)	11.05.2008 Beyyayla/Eskişehir (live)	1	-	-
Blue Tit (<i>Parus caeruleus</i>)	14.11.2009 Gürleyik/ Eskişehir (live)	1	-	-
Bee-eater (<i>Merops apiaster</i>)	13.08.2010 Eskişehir (died)	1	1	<i>Meromenopon meropis</i> <i>Meropoecus meropis</i>
Common Crossbill (<i>Loxia curvirostra</i>)	13.08.2010 Eskişehir (injured)	1	-	-
Goldfinch (<i>Carduelis carduelis</i>)	08.02.2008; 13.02.2008 Eskişehir (live)	2	-	-
White-winged Black Tern (<i>Chlidonias leucopterus</i>)	03.07.2010 Akşehir-Konya (died)	1	1	<i>Actornithophilus piceus piceus</i>
Buzzard (<i>Buteo buteo</i>)	01.07.2010 Eskişehir Zoo (injured)	1	-	-
Long-legged Buzzard (<i>Buteo rufinus</i>)	01.07.2010 Eskişehir Zoo (injured)	1	-	-
	17.05.2010 Konya (injured)	1	-	-
	21.06.2010 Konya (injured)	1	-	-
Goshawk (<i>Accipiter gentilis</i>)	08.08.2010 Kızılınler village-Eskişehir (injured)	1	1	<i>Colpocephalum polonum</i>
Greater Flamingo (<i>Phoenicopterus ruber</i>)	03.07.2010 Akşehir-Konya (died)	1	1	<i>Trinoton femoratum</i> <i>Colpocephalum heterosoma</i> <i>Anaticola phoenicopteri</i> <i>Anatoecus pygaspis</i>
Eurasian Coot (<i>Fulica atra</i>)	03.07.2010 Akşehir-Konya (2 lives, 1 died)	3	2	<i>Fulicoffula lurida</i> <i>Rallicola fulica</i> <i>Incidifrons fulica</i> <i>Pseudomenopon pilosum</i>
Slender-billed Gull (<i>Larus genei</i>)	03.07.2010 Akşehir-Konya (died)	1	1	<i>Saemundssonaria lari</i>
Merlin (<i>Falco columbarius</i>)	01.02.2010 Beyşehir-Konya (injured)	1	-	-
House Sparrow (<i>Passer domesticus</i>)	28.05.2010 Konya (live)	1	-	-
White Stork (<i>Ciconia ciconia</i>)	07.09.2009; 01.04.2010 Konya (injured)	2	2	<i>Colpocephalum zebra</i> <i>Ciconiphilus quadripustulosus</i> <i>Ardeicola ciconiae</i> <i>Neophilopterus incompletus</i>
Long-eared Owl (<i>Asio otus</i>)	19.09.2009 Karaman (died)	1	-	-
Rock Pigeon (<i>Columba livia</i>)	29.06.2009; 25.09.2009; 18.11.2009 Konya (live)	3	2	<i>Columbicola columbae</i>
Short-toed Eagle (<i>Circus gallicus</i>)	31.07.2009 Ürgüp-Nevşehir (died)	1	-	-
Canary (<i>Serinus canaria</i>)	30.12.2009 Konya (live)	1	-	-
Black-winged Stilt (<i>Himantopus himantopus</i>)	03.07.2010 Akşehir-Konya (died)	1	-	-
Sanderling (<i>Calidris alba</i>)	03.07.2010 Akşehir-Konya (died)	1	-	-
Redshank (<i>Tringa totanus</i>)	03.07.2010 Akşehir-Konya (died)	1	-	-

White Stork (*Ciconia ciconia*).

Rallicola fulica: Examined materials: 10 ♀, 11 ♂, 4 N, 13.08.2010-1, Akşehir, Konya, 5 ♀, 3 ♂, 2 N, 13.08.2010-2, Akşehir, Konya (Fig. 7, 8). Host: Eurasian Coot (*Fulica atra*). It was reported for the first time in Turkey.

Saemundssonaria lari: Examined materials: 1 ♀, 2 ♂, 3 N, 03.07.2010, Akşehir, Konya (Fig. 9). Host: Slender-billed Gull (*Larus genei*). It was reported for the first time in Turkey.

Amblycera

Actornithophilus piceus piceus: Examined materials: 2 ♀, 03.07.2010 Akşehir, Konya (Fig. 10). Host: White-winged Black Tern (*Chlidonias leucopterus*). It was reported for the first time in Turkey.

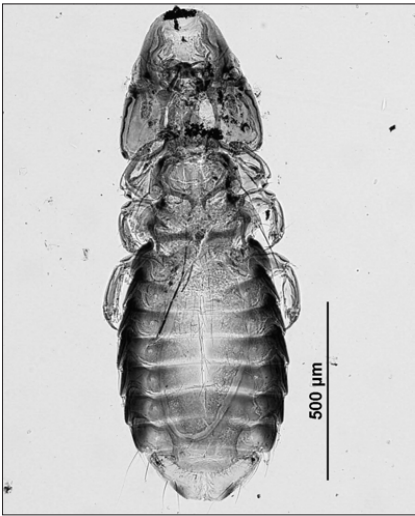
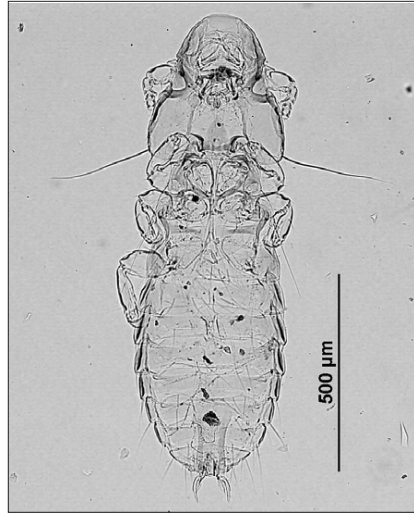
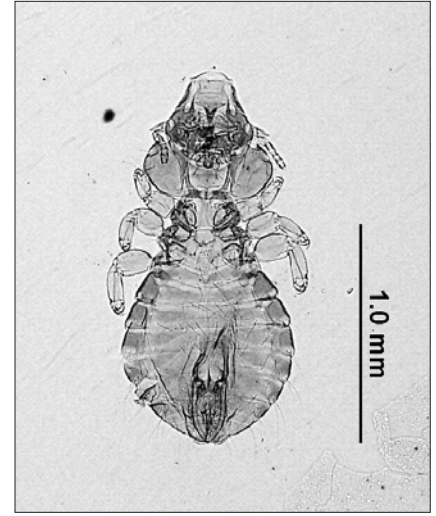
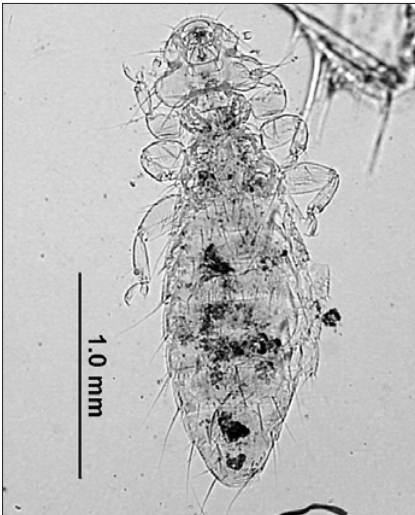
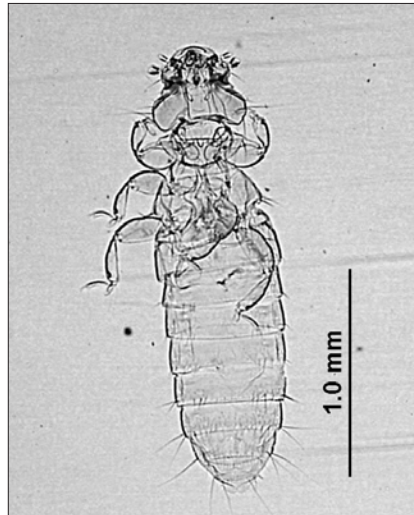
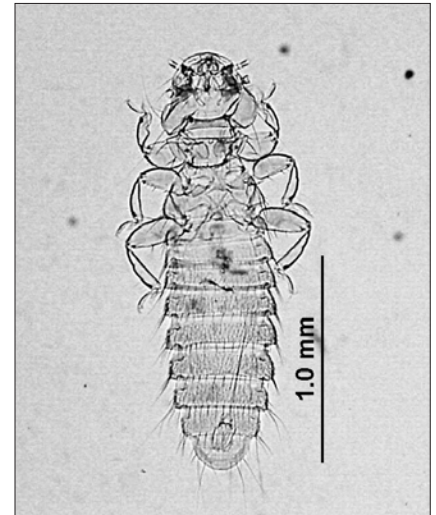
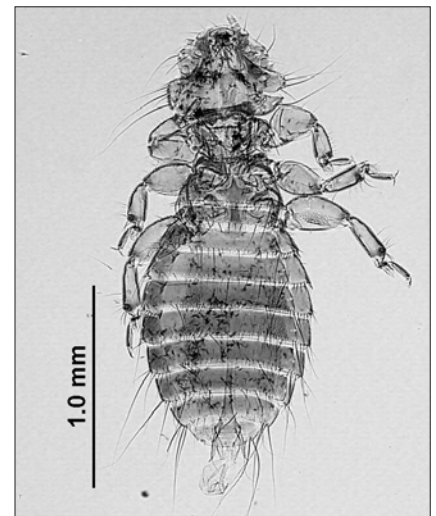
Ciconiphilus quadripustulosus: Examined materials: 29 ♀, 17 ♂, 6 N, 07.09.2009, Konya; 2 ♀, 1 ♂, 01.04.2010, Konya. Host: White Stork (*Ciconia ciconia*).

Colpocephalum zebra: Examined materials: 1 ♀, 07.09.2009, Konya. Host: White Stork (*Ciconia ciconia*).

Colpocephalum heterosoma: Examined materials: 13 ♀, 9 ♂, Akşehir, Konya (Fig. 11, 12). Host: Greater Flamingo (*Phoenicopterus ruber*). It was reported for the first time in Turkey.

Colpocephalum polonum: Examined materials: 16 ♀, 6 ♂, 12 N, 08.08.2010, Eskişehir (Fig. 13, 14). It was reported for the first time in Turkey. Host: Goshawk (*Accipiter gentilis*).

Meromenopon meropis: Examined materials: 1 ♂, 13.08.2010, Akşehir, Konya (Fig. 15). Host: Bee-eater (*Merops*

Fig 7. *Rallicola fulcia*, female, orig.Şekil 7. *Rallicola fulcia*, dişi, orijinalFig 8. *Rallicola fulcia*, male, orig.Şekil 8. *Rallicola fulcia*, erkek, orijinalFig 9. *Saemundssonina lari*, male, orig.Şekil 9. *Saemundssonina lari*, erkek, orijinalFig 10. *Actornithophilus piceus piceus*, female, orig.Şekil 10. *Actornithophilus piceus piceus*, dişi, orj.Fig 11. *Colpocephalum heterosoma*, female, orig.Şekil 11. *Colpocephalum heterosoma*, dişi, orj.Fig 12. *Colpocephalum heterosoma*, male, orig.Şekil 12. *Colpocephalum heterosoma*, erkek, orj.Fig 13. *Colpocephalum polonum*, female, orig.Şekil 13. *Colpocephalum polonum*, dişi, orijinalFig 14. *Colpocephalum polonum*, male, orig.Şekil 14. *Colpocephalum polonum*, erkek, orj.Fig 15. *Meromenopon meropis*, male, orig.Şekil 15. *Meromenopon meropis*, erkek, orijinal

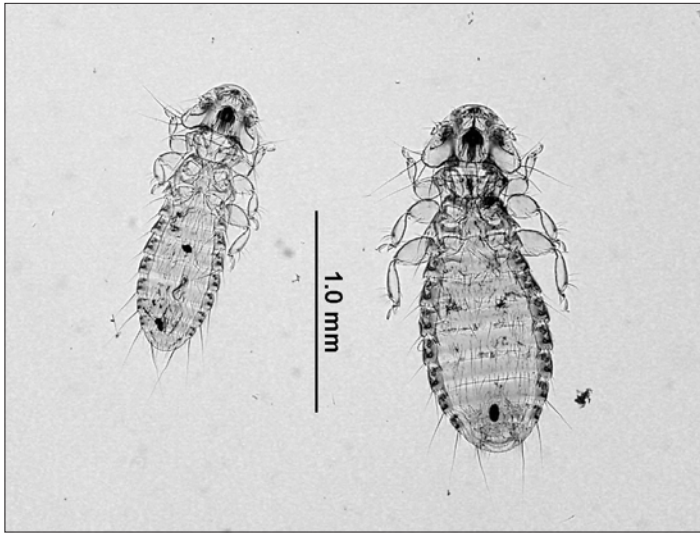


Fig 16. *Pseudomenopon pilosum*, male (at left), female (at right), orig.

Şekil 16. *Pseudomenopon pilosum*, erkek (solda), dişi (sağda), orijinal



Fig 17. *Trinoton femoratum*, female, orig.

Şekil 17. *Trinoton femoratum*, dişi, orijinal

apiaster). It was reported for the first time in Turkey.

Pseudomenopon pilosum: Examined materials: 8 ♀, 8 ♂ 2 N, 13.08.2010-1, Akşehir, Konya, 2 ♀, 4 ♂, 13.08.2010-2 Akşehir, Konya (Fig. 16). It was reported for the first time in Turkey. Host: Eurasian Coot (*Fulica atra*).

Trinoton femoratum: Examined materials: 1 ♀, 3 N, 03.07.2010, Akşehir-Konya (Fig. 17). It was reported for the first time in Turkey. Host: Greater Flamingo (*Phoenicopterus ruber*).

DISCUSSION

Approximately 4000 valid lice species have been found on the birds in the world ¹. Until now, 465 bird species were recorded from Turkey ^{11,29}. However, almost all of the louse fauna of these birds are still unknown. Until today approximately 100 bird species were investigated for the louse infestation and 82 chewing lice species belonging to 37 genera were identified ². *Colpocephalum zebra*, *Ciconiphilus quadripustulosus*, *Ardeicola ciconiae* and *Neophilopterus incompletus* on white storks (*Ciconia ciconia*) ⁷, *Austromenopon atrofulvum*, *Quadriceps anagrapsus* and *Saemundssonina lobaticeps* on white-winged black tern (*Chlidonias leucopterus*) ¹¹, *Strigiphilus barbatus* on long-eared owl (*Asio otus*), *Kurodaia fulvofasciata* on buzzard (*Buteo buteo*), *Laemobothrion maximum*, *Colpocephalum nanum*, *Colpocephalum* sp, *Degeeriella fulva* and *Craspedorrhynchus platystomus* on long-legged buzzard (*Buteo rufinus*) were reported from Turkey in these studies. The presence of some louse species except *C. columbae* recorded from rock pigeons (*Columba livia*) was suspect and it has already been discussed by Dik ⁵ in a previous work. On the other hand, no louse specimen was found on sparrow (*Passer domesticus*) ⁵ and short-toed eagle

(*Circaetus gallicus*) ⁹. It was investigated some specimens of the bird species which their names were mentioned above in the present study, and only one species; *C. columbae* was found on rock pigeon, while all of the lice species recorded from white storks were detected. Two female samples of *Actornithophilus piceus piceus* were collected on white-winged black tern in this study, while *Austromenopon atrofulvum*, *Quadriceps anagrapsus* and *Saemundssonina lobaticeps* were recorded in a previous study ¹¹.

Clay ¹⁷ reported five lice species; *Colpocephalum heterosoma* Piaget, 1880; *Colpocephalum salimalii* Clay, 1951; *Trinoton* sp, *Anaticola phoenicopteri* (Coincide, 1859) and *Anatoecus keleri* Clay, 1962 from greater flamingo (*Phoenicopterus ruber*). In addition, Price et al.¹ recorded eight species; *Anaticola candidus* (Rudow, 1869); *Anaticola phoenicopteri* (Coincide, 1859), *Anatoecus keleri* Clay, 1962; *Anatoecus pygaspis* (Nitzsch, 1866); *Colpocephalum heterosoma* Piaget, 1880; *Colpocephalum rosei* Price and Emerson, 1974; *Colpocephalum salimalii* Clay, 1951 and *Trinoton femoratum* Piaget, 1889 on greater flamingo. Morphological features of the genus *Anatoecus* and some species were found in this genus studied in detail by Keler ²². Clay ¹⁶ described *A. keleri* from greater flamingo and reported that morphologic characters of this species similar to *A. pygaspis*, however distinguish from *A. pygaspis* by the characters of the male genital sac, the sclerite being long and stout and lying in the centre. It was detected that the morphological characters of abdominal tergites, lateral sclerites and male genitalia of the all male specimens examined in this study were observed to be similar to those of *A. pygaspis*.

Clay ¹³ described the morphological characters of *Colpocephalum heterosoma* Piaget, 1880 and *Colpocephalum salimalii* Clay, 1951 in detail, and reported that resemble of these species each other. This investigator ¹³ indicated

that the characters of male genitalia of *C. salimalii* are quite different from *C. heterosoma* and there are some differences in the shape of the metathorax and abdomen and the protergal, mesotergal and abdominal chaetotaxy. In addition, Price and Beer²⁵ explained the morphological characters and identification key of the *Colpocephalum* species found on Ciconiiformes and reported that there were many short or medium long seta on abdominal tergites IV-VIII of male *C. heterosoma* while male specimens of *C. salimalii* had rare and short seta on the abdominal segments IV-VIII. Same authors²⁵ recorded that abdomen was rounded in *C. heterosoma* while attenuated in *C. salimalii*, in the females. In the present study, it was detected that the morphologic characters of all *Colpocephalum* specimens collected from greater flamingo resemble to *C. heterosoma*.

Price et al.¹ stated that six lice species; *Colpocephalum nanum* Piaget, 1880; *C. polonum* Eichler and Zlotorzyska, 1971; *Craspedorrhynchus haematopus* (Scopoli, 1763); *Degeeriella vagans* (Giebel, 1874); *Laemobothrion maximum* (Scopoli, 1763) and *Nosopon lucidum* (Rudow, 1869) were found on goshawk (*Accipiter gentilis*). Price and Beer²⁴ reported that *Colpocephalum nanum* was occurred on goshawk in addition to the other prey birds. Eichler and Zlotorzyska²⁸ identified that the lice specimens were collected from goshawks as *C. polonum* (= *Neocolpocephalum polonum*). The morphologic characters of *C. polonum* were not described by Eichler and Zlotorzyska²⁸ in detail, however they were pointed out that this species resembles to *C. turbinatum* and *C. germanum*. Thus in this study, the all collected louse specimens from goshawk were identified as *C. polonum*. Some authors^{5,30} recorded that *Strigiphilus barbati* was found on long-eared owl (*Asio otus*) in Turkey. Dik et al.¹⁰ stated that the louse infestations were occurred on songbirds' (Passeriformes) very low rates, and five lice species were detected in their study in Turkey. Although, only one bird species (long-eared owl) belonging to the order Strigiformes was examined, no louse specimen was detected, in this study. Besides, eight bird specimens belonging to the order Passeriformes were examined but it was no louse obtained from these birds.

Three lice specimens collected from bee-eater were identified as *Meropoecus meropis* (two nymphs), and *Meromenopon meropis* (male). Other species occurring on bee-eater, *Brueelia apiastri* was not detected in this study. In previous studies were performed to detect lice of bee-eaters showed that the lice specimens on the bee-eaters were low numbers^{12,31}. Similarly, a few lice specimens were collected on the bee-eaters in this study.

It was determined that relatively more lice specimens were obtained on great flamingo, goshawk and Eurasian coot while a little numbers of the lice on the other examined birds which examined in the study. Sixty-eight lice samples from greater flamingo, 34 from goshawk and 84

from Eurasian coot, while two lice specimens from white-winged black tern, five from slender-billed gull, three from bee-eater, six from two white storks and four from two rock pigeon were collected in the present study.

In conclusion, 13 lice species; *C. polonum* Eichler and Zlotorzyska, 1971; *Meromenopon meropis* Clay and Meinertzhagen, 1941; *Meropoecus meropis* (Denny, 1842); *Actornithophilus piceus piceus* (Denny, 1842); *Trinoton femoratum* Piaget, 1889; *Colpocephalum heterosoma* Piaget, 1880; *Anaticola phoenicopteri* (Coincide, 1859); *Anatoecus pygaspis* (Nitzsch, 1866); *Fulicoffula lurida* (Nitzsch, 1818); *Rallicola fulicia* (Denny, 1842); *Incidifrons fulicia* (Linnaeus, 1758); *Pseudomenopon pilosum* (Scopoli, 1763) and *Saemundssonsonia lari* Fabricius, O, 1780) detected in this study were recorded for the first time in Turkey with this study.

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The Effects of Lower Levels of Organically Complexed Zinc, Copper and Manganese in Broiler Diets on Performance, Mineral Concentration of Tibia and Mineral Excretion ^[1]

Taylan AKSU *  Bülent ÖZSOY * Devrim SARIPINAR AKSU **
Mehmet Akif YÖRÜK *** Mehmet GÜL ***

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* University of Mustafa Kemal, Faculty of Veterinary Medicine, Department of Animal Nutrition and Nutritional Disorders, TR-31040 Hatay - TURKEY

** University of Mustafa Kemal, Faculty of Veterinary Medicine, Department of Physiology, TR-31040 Hatay - TURKEY

*** University of Atatürk, Faculty of Veterinary Medicine, Department of Animal Nutrition and Nutritional Disorders, TR-25240 Erzurum - TURKEY

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Summary

The effects of replacing inorganic with lower levels of organic trace minerals of Zn, Cu and Mn on growth performance, mineral excretion and mineral concentration in tibia were investigated in broilers. One-day-old Ross-308 broiler chicks were used. Chicks were divided into 4 different groups as 1 control and 3 experimental groups consisted of 50 chicks. All groups were also divided into 5 replicates containing 10 chicks. Starter (0-21 d) and finisher (21-42 d) broiler diets which not included Zn, Cu and Mn were used. Specially prepared mineral premix (containing at 40 mg Zn, 80 mg Cu and 60 mg Mn levels of inorganic minerals in each kg as sulphate form) added to control diet in the level of recommended by NRC. Mineral content of the control diet was prepared using standard inorganic mineral premix that reflects the normal supplementary levels and source of trace minerals for commercial broiler feed recommended by National Research Council. In the experimental diets, mineral premix was also prepared as inorganic forms except of Zn, Cu and Mn. Organically complexed Zn, Cu, and Mn were separately added into basal diet at 1/3, 2/3 and 3/3 proportions as BioplexTM instead of inorganic forms of those mineral recommended levels by NRC. Experiment lasted 42 days. Results showed that organically complexed trace minerals can be used at a much lower concentration than the current recommended as inorganic minerals, without a negative impact on performance, while also decreasing the excess mineral excretion. Further studies are needed to determine the proper level of organic trace minerals by taking tibia mineral levels into consideration.

Keywords: *Organically complexed mineral, Poultry, Performance, Mineral Excretion*

Etlik Piliç Diyetlerinde Daha Düşük Seviyelerde Organik Çinko, Bakır ve Manganın Performans, Tibia Mineral Yoğunluğu ve Dışkı Mineral Atılımı Üzerine Etkileri

Özet

Araştırma, etlik piliç rasyonlarına inorganik formları yerine daha düşük seviyelerde organik bakır, çinko ve mangan ilavesinin etlik piliçlerde performans, tibia mineral yoğunluğu ve dışkı mineral atılımı üzerine etkilerini belirlemek amacı ile yürütüldü. Araştırmada toplam 200 adet bir günlük yaşta Ross-308 civciv kullanıldı. Civcivler biri kontrol diğer üçü deneme grubu olmak üzere her biri 10'ar civcivli beş tekrardan oluşan dört gruba tesadüfi olarak dağıtıldı. Araştırmada bakır, çinko ve mangan içermeyen standart etlik piliç büyütme (0-21 gün) ve bitirme (21-42 gün) yemleri kullanıldı. Kontrol (inorganik) grubu yemlerine kg'ında sülfat formunda 40 mg Zn, 8mg Cu ve 60 mg Mn içeren özel olarak hazırlanmış mineral karışımı NRC tarafından bildirilen seviyelerde katıldı; deneme gruplarına ise bu minerallerin sırasıyla 1/3, 2/3 ve 3/3'ü oranında organik formlarını (BioplexTM) içeren özel olarak hazırlanmış mineral karışımı ilave edildi. Araştırma 42 gün sürdürüldü. Araştırma sonucunda, NRC tarafından etlik piliçler için bildirilen inorganik Zn, Cu ve Mn seviyeleri yerine daha düşük miktarlarda organik formlarının katılabileceği; mineral seviyelerindeki bu azaltmanın etlik piliç performansını olumsuz etkilemediği; dışkı ile atılan mineral yoğunluğunu azalttığı; etlik piliç diyetlerinde en uygun mineral seviyelerinin belirlenebilmesi için tibia mineral yoğunluğunun da göz önüne alındığı ileri çalışmalara ihtiyaç olduğu kanaatine varıldı.

Anahtar sözcükler: *Organik mineral, Kanatlı, Performans, Mineral atılımı*



İletişim (Correspondence)



+90 0326 2455845/1521



aksuturkiye@gmail.com

INTRODUCTION

Trace minerals such as copper, zinc and manganese play important roles in various bodily functions and are necessary to sustain life and maintain optimal health, and thus are essential nutrients for all animals. Minerals provide structure to bones and participate in many enzyme, hormone secretion pathways, and immune defense systems¹. In commercial poultry diets, majority of trace minerals are commonly supplemented in the form of inorganic salts, such as sulfates, oxides and carbonates, to provide levels of minerals that prevent clinical deficiencies and allow the birds to reach their genetic growth potential². In commercial poultry diets³, inorganic trace minerals supplementation is much higher than those recommended by National Research Council⁴. Excessive mineral supplementation leads to wasteful and the environmental contamination. Due to increasing concerns about potential mineral pollution, there has been considerable interest in discussion on how to reduce mineral excretion without any negative effect on production performance. The use of organically complexed or chelated minerals at a much lower concentration for livestock diets have been suggested as an answer to this discussion, based on the hypothesis that such mineral complexes have a higher bioavailability than inorganic salt analogues. Organically complexed or chelated minerals have a higher bioavailability than inorganic salts. Chelates are each molecule that consists of a non-covalent bond of a mineral (either dietary essential or not) with organic ligands. Animals absorb, digest and use mineral chelates better than inorganic minerals. This means that much lower concentrations can be used in animal feeds.

In previous experiment where lower levels of organically complexed minerals (Zn, Cu and Mn) were used instead of inorganic forms of those minerals, it was reported that using at a much lower levels of organically complexed minerals did not cause to a weakness in the plasma antioxidant defence system⁵ and created a negative impact on hematological and biochemical blood parameters⁶. In addition, the protective function of organically complexed zinc (Zn-proteinat) in the cell membrane against the lipid peroxidation is reported to be more effective than that of inorganic form of zinc⁷. In addition, animals fed chelated sources of essential trace minerals excrete lower amounts in their feces and the risk of environmental contamination may be reduced from manure^{8,9}. Therefore, the objective of the present study was to evaluate the substitution of much lower levels of organically complexed minerals (Zn, Cu and Mn) rather than inorganic forms of minerals those recommend by NRC on growth performance, mineral excretion and mineral concentration of the tibia in broilers.

MATERIAL and METHODS

Animals, Diets and Experimental Design

The experiment was conducted under protocols by The University of Mustafa Kemal, Ethical Commission Accession (No: 2009-1-1). A total of two hundred Ross-308 one-day-old broiler chickens were used in the feeding trial that lasted until the birds reached 42 d of age. The experimental animals in both sexes were divided into four groups comprising 1 control group, and three experimental groups each consisting of 50 chickens. All groups were also divided into 5 replicates containing 10 broiler chicks. The birds were given ad libitum access to feed and water. A lighting schedule of 23L: 1D was imposed throughout the experimental period. Newcastle disease vaccination was performed on d 10, whereas the Gumboro vaccination was administrated on d 18. The basal diet (*Table 1*) was formulated according to NRC⁴. Two phases were applied during the experiment: A starter (0-21 d) and finisher (21-42 d).

The control premix was also formulated to meet the requirement allowance of trace elements reported by

Table 1. Ingredient composition of the basal diets, %

Tablo 1. Temel diyetin bileşimi, %

Ingredients,%	Starter (0 to 21 d)	Finisher (21 to 42 d)
Maize	51.5	55.2
Wheat	7	7
Wheat Bran	4.5	4.5
Extracted Soybean Meal	27.5	24
Fish Meal	5.5	4.3
Vegetable Oil	1.5	2.5
Limestone	1	1
DCP	0.75	0.75
Salt	0.25	0.25
Vit-Min. Premix *	0.5	0.5
Calculated Nutrients		
ME, MJ/kg	12.6	13
Crude Protein, %	22.1	20
Ca, %	0.9	0.8
P, %	0.6	0.7
Lysine, %	1.1	0.8
Analyzed Nutrients		
Cu, mg/kg	9.63	8.71
Zn, mg/kg	35.65	30.23
Mn, mg/kg	38.84	36.63

* Supplied per kilogram of diet: Vitamin A 15.000 IU; cholecalciferol 1.500 ICU; vitamin E, 30 IU; menadion, 5.0 mg; thiamin, 3.0 mg; riboflavin, 6.0 mg; niacin, 20.0 mg; pantothenic acid, 8.0 mg; pyridoxine, 5.0 mg; folic acid, 1.0 mg; vitamin B₁₂, 15 µg; Fe, 30.0 mg; I, 2.0 mg; Se, 0.15 mg

NRC ⁴, using standard inorganic mineral premix (containing 40 mg Zn as ZnSO₄ and 8 mg Cu as CuSO₄ 60 mg Mn as MnO and others trace mineral per kg) that reflects the normal supplementary levels and source of trace minerals for commercial broiler feeds. The experimental premixes were also formulated using organically complexed Zn, Cu and Mn (organic) at 1/3 (L1), 2/3 (L2) and 3/3 (L3) levels instead of inorganic forms of those minerals as Bioplex™ (Table 2).

Table 2. Source and amounts of trace minerals fed to broilers

Tablo 2. İz minerallerin kaynak ve miktarları

Diet	Added Zn (mg/kg)	Added Cu (mg/kg)	Added Mn (mg/kg)
Inorganic (control) *	40	8	60
1/3 organic (L-1) **	13	2,5	20
2/3 organic (L-2) **	26	5	40
3/3 organic (L-3) **	40	8	60

* Supplied per kilogram of diet: Vitamin A 15.000 IU; cholecalciferol 1.500 ICU; vitamin E, 30 IU; menadion, 5.0 mg; thiamin, 3.0mg; riboflavin, 6.0 mg; niacin, 20.0 mg; pantothenic acid, 8.0 mg; pyridoxine, 5.0 mg; folic acid, 1.0 mg; vitamin B₁₂, 15 µg; Mn, 60.0 mg; Zn, 40 mg; Fe, 30.0 mg; Cu, 8.0 mg; I, 2.0 mg; Se, 0.15 mg

** Organically complexed Mn, Zn and Cu were provided as Bioplex-Mn™, Bioplex-Zn™ and Bioplex-Cu™

Bioplex containing amino acids from hydrolysed-soy protein is an amino acid-hydrate complex bonded with Cu, Zn, and Mn. Bioplex Cu™, Bioplex Zn™, and Bioplex Mn™ contain 100 g/kg of Cu, 150 g/kg of Zn, and 150 g/kg of Mn, respectively (Alltech UK, Standford, England).

Measurements, Sample Collection and Laboratory Analysis

Birds in each pen were individually weighed weekly for determine body weight (BW) and body weight gain (BWG). At the same days, feed intake (FI) was also recorded to determine feed conversion ratio (FCR), feed:gain (g:g). Mortality was recorded as it occurred. At the end of the experiment, the right tibia from 2 birds per each replicate was pooled and boiled for approximately 10 min in deionized water and were then cleaned of all soft tissue. At d 26, fecal samples were taken for mineral analysis. All birds per pen were placed in cages, where lighting was reduced for approximately 20 min, until sufficient droppings were produced (minimum of 50 g per 10 birds). After fecal collection, birds were returned to their original pens. Fecal samples from each pen were homogeneously mixed and frozen for analysis. Bone, basal diet and tibia samples were then ashed (550°C for 4 h). Approximately 1 g of ash samples was then dissolved in 10 ml of HClO₄ and boiled for 10 min, then the remainder mixed with 5 ml HNO₃. It was filtered and diluted to 50 ml flask. All samples

were then analyzed using Inductively Coupled Plasma Emission Spectroscopy (ICP) by a laboratory specializing in this assay.

Statistical Analysis

The appropriate Sum of Squares Method was selected for ANOVA ¹⁰. The data were analyzed by one-factor ANOVA using the general linear models procedure of SAS ¹¹ software for the main effect of treatments. Differences between means were determined by Duncan's multiple range test at a significance level of P<0.05.

RESULTS

Broiler Performance

During the experiment, any death was not recorded. No differences in response to addition of lower levels organic Zn, Cu and Mn instead of their inorganic forms were observed among any of the performance parameters measured. There was a tendency toward improved production performance for the birds fed at 3/3 level of organic minerals (L3) in d 21, but production performance was similar among all groups the end of the experiment (Table 3).

Mineral Concentration in Tibia

The mineral concentrations of tibia are shown in Table 4.

Trace mineral concentrations in tibia significantly decreased (P<0.05), when organic mineral inclusion was applied at 1/3 level of recommended inorganic levels. Except zinc concentration of tibia, this reduction in tibia mineral concentrations were restored in the birds fed at 2/3 and 3/3 level of organic minerals for Cu and Mn. The zinc concentration of tibia in the birds reached to the same value with control group only at 3/3 level of organic minerals. Not any findings were observed brings to mind the deformation of bone or articular during experiment.

Mineral Excretion

Mineral levels of basal diet and supplemented mineral levels from either the inorganic premix (control) or the organic source are shown in Table 1 and Table 2. The results from mineral analysis in manure samples at d 26 are also shown in Table 5.

The significantly lower excretion of Zn and Mn were observed in the birds fed at 1/3 levels of organic minerals (P<0.01). The excretion of Zn, Cu and Mn were not significantly different between the birds fed inorganic mineral and the birds fed at 3/3 levels of organic mineral. Higher levels of organic mineral did not lead to higher mineral retention (P<0.05).

Table 3. Performance of birds fed containing different levels of organic minerals**Tablo 3.** Farklı seviyelerde organik mineral içeren diyetlerle beslenen piliçlerin performansı

Parameters	Diets ¹					P
	Control	L1	L2	L3	SEM	
BW, g						
1 d	45.58	45.31	45.32	45.34	0.22	NS
21 d	664.40 ^b	642.76 ^b	656.96 ^b	705.60 ^a	6.23	*
42 d	1859.36	1875.24	1914	1910	14.74	NS
BWG, g/bird/week						
1 to 21 d	618.82	597.45	611.64	660.26	6.25	*
22 to 42 d	1194.96	1232.48	1243.18	1204.42	15.98	NS
1 to 42 d	1813.78	1829.93	1854.82	1864.68	14.73	NS
FI, g/birds						
1 to 21 d	993.64	911.10	970.44	953.88	9.45	NS
22 to 42 d	2288.06	2282.40	2265.62	2272.44	10.19	NS
1 to 42 d	3221.70	3193.50	3236.06	3226.32	12.54	NS
FCR, feed:gain, g:g						
1 to 21 d	1.51	1.53	1.59	1.45	0.01	NS
22 to 42 d	1.93	1.86	1.82	1.89	0.02	NS
1 to 42 d	1.78	1.73	1.75	1.73	0.02	NS

^{a,b} Means values within a row having differing superscripts are significantly different ($P < 0.05$), **NS**: non-significant

¹ Control: Inorganic Zn, Cu and Mn at NRC recommended levels as sulfate, L1, L2, and L3: Organically complexed Zn, Cu and Mn at 1/3, 2/3 and 3/3 proportions instead of inorganic forms of those minerals as Bioplex™, respectively

Table 4. Trace mineral concentrations in tibia of birds fed containing different levels of organic minerals**Tablo 4.** Farklı seviyelerde organik mineral içeren diyetlerle beslenen piliçlerin tibia iz mineral konsantrasyonları

Trace Minerals	Diets ¹				SEM	P
	Control	L1	L2	L3		
Zn (µg/g)	124.78 ^a	111.18 ^c	116.20 ^{bc}	120.79 ^{ab}	2.24	**
Cu (µg/g)	4.50 ^a	3.08 ^b	3.47 ^{ab}	4.35 ^a	0.22	*
Mn (µg/g)	7.46 ^a	4.09 ^b	6.27 ^a	6.66 ^a	0.34	**

Means represent from 10 birds per treatment

^{a,b,c} Means values within a row having differing superscripts are significantly different ($P < 0.05$)

* $P < 0.05$, ** $P < 0.01$

¹ Control: Inorganic Zn, Cu and Mn at NRC recommended levels as sulfate

L1, L2, and L3: Organically complexed Zn, Cu and Mn at 1/3, 2/3 and 3/3 proportions instead of inorganic forms of those minerals as Bioplex™, respectively

Table 5. Trace mineral excretion of birds fed containing different levels of organic minerals experimental diets with different levels of organic minerals at 26 d of age**Tablo 5.** Farklı seviyelerde organik mineral içeren diyetlerle beslenen piliçlerin 26. gündeki iz mineral atımı

Trace Minerals	Diets ¹				SEM	P
	Control	L1	L2	L3		
Zn (µg/g/bird)	33.03 ^a	19.25 ^c	25.50 ^b	30.82 ^{ab}	1.47	**
Cu (µg/g/bird)	2.85	2.19	2.91	3.14	0.15	NS
Mn (µg/g/bird)	16.24 ^{ab}	8.91 ^c	13.92 ^b	18.86 ^a	1.07	**

Means represent from 5 birds per treatment

^{a,b,c} Means values within a row having differing superscripts are significantly different ($P < 0.05$)

NS: non-significant, ** $P < 0.01$

¹ Control: Inorganic Zn, Cu and Mn at NRC recommended levels as sulfate.

L1, L2, and L3: Organically complexed Zn, Cu and Mn at 1/3, 2/3 and 3/3 proportions instead of inorganic forms of those minerals as Bioplex™, respectively

DISCUSSION

The use of organically complexed or chelated minerals at much lower levels in premixes has been suggested as a solution to environmental pollution originated from high mineral excretion, without jeopardizing animal health and performance. In the present study, we examined the organic minerals at much lower levels instead of NRC recommended levels of those mineral as sulfate. In the present study, BWG values of birds were slightly lower than those before reported studies. Environmental temperature and relative humidity levels during the current study ranged from 31.6-32.7°C and 62.8-65.5%, respectively. These unfavorable conditions on the period of present study may be caused to low BWG. On the other hand, lower levels inclusion of organic zinc, copper, and manganese did not affect animal performance. Similarly, some researchers^{2,12} reported that trace mineral at lower levels can be included the diets without any negative effect on broiler performance. Sacranie¹³ indicated that the total amount of inorganic minerals in a broiler premix could be totally replaced by 20% organic minerals without affecting growth performance and, at the same time reducing the environmental contamination. The use of organic mineral sources can improve intestinal absorption of trace elements as they reduce interference from agents that form insoluble complexes with the ionic trace elements¹⁴. In addition, Nollet et al.¹² reported that in the starter period feed conversion rate tended to improve in broiler fed organically complexed mineral diet. However, no significant differences were observed in any of the growth performance parameters measured during the trial. On the other hand, Leeson⁹ found that using trace minerals with greater bioavailability (Bioplex™ trace minerals) did not affect body weight gain and had little effect on feed efficiency of broilers even when fed at 20% of the inorganic trace mineral level.

Zinc and manganese play important role in development of bone and cartilage, and Mn is essential in the prevention of perosis in chicks^{15,16}. As shown in [Table 4](#), mineral concentration of the tibia dramatically decreased in birds fed at 1/3 level of organic minerals. This decline may be ascribed that the interaction between minerals. This interaction could be determined and interpreted if current minerals could be individually added to the diet. Whereas, the experimental design of the current study was not appropriate to thoroughly discuss and interpret this interaction. Except zinc concentration of tibia, this reduction in tibia mineral concentrations were restored in the birds fed at 2/3 and 3/3 level of organic minerals for Cu and Mn. However, during the current study, there was no macroscopical findings related to articular damage and bone deformation such

as perosis, lameness etc.

In agreement with the present study, Bao et al.² found that low levels inclusion of organic Zn, Cu and Mn (2 mg/kg of Cu and 20 mg/kg of diet each of Mn and Zn in each per kg of diet) into diet based mainly on sorghum and isolated soy with 20.38 mg/kg Zn, 4.20 mg/kg Cu and 14.82 mg/kg Mn from raw material, significantly decreased the concentration of Zn and Mn in tibia in comparison to the mid or high levels inclusion of those minerals. Similar to Bao et al.², reduction of tibia trace mineral concentration was restored when dietary levels of Zn, Mn and Cu were raised to 2/3 and 3/3 proportions of recommended inorganic levels in the current study. On the other hand, Loveridge¹⁷ expressed that bone growth is intimately connected with overall body growth, making tibia Zn concentration a good predictor of whole-body growth. Contrary to Loveridge¹⁷, body growth was not different among inclusion levels in the current study. These finding is an agreement with Ao et al.¹⁸, who reported that tibia Zn concentration was lower in the birds fed 20 ppm Bioplex Zn than the birds fed 40 ppm Bioplex Zn, although weight gain was similar among these supplemental organic mineral levels.

Reduction of the amount of trace minerals in the diet would therefore reduce mineral excretion to environment. Most of studies on minerals for animal performance^{19,20} which used conventional inorganic mineral mixtures, indicates that animal usually reduces the rate of growth and excretion to maintain normal tissue concentration if dietary mineral deficiency is moderate for growth. In the present study, reduction of the amount of Zn and Mn in manure of birds fed at 1/3 levels of recommendation in organic minerals were significant while the rate of growth was not different among inclusions levels. This could be due to the fact that organically complexed or chelated minerals have a higher bioavailability than inorganic salts^{2,12,21}. On the other hand, mineral excretion was higher in manure of birds fed at 2/3 and 3/3 levels of recommendation in organic mineral than in those of birds fed at 1/3 levels of organic minerals ([Table 5](#)). It is presumed that the body becomes "loaded" with these minerals²¹. Organically complexed minerals can be easily absorbed by intestines in comparison to inorganic salts but not be completely utilized in high levels. Similarly, Nollet et al.²¹ indicated that the mineral excretion of birds fed at 100% and 67% portion of organic mineral premix were not significantly different from the birds fed at 100% levels of recommended levels in inorganic minerals.

As a result, using at 1/3 level of organic minerals (at 1/3 level of Zn, Cu and Mn) in broiler diet instead of recommended level of inorganic forms of those

minerals did not adversely affect broiler performance and reduced the amount of mineral excreted in feces. Using at a much lower level of organic minerals (at 1/3 level of inorganic form) were decreased the tibia mineral levels but this decline was restored starting from at 2/3 level of organic minerals. Organically complexed minerals (Zn, Cu and Mn) can be supplied at a much lower levels into the broiler diets comparison to the current recommended of inorganic forms without a negative impact on performance. But further studies are needed to determine the proper levels of organic minerals by taking tibia mineral levels into consideration.

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