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Effects of *In-Ovo* Injection of Manganese on Some Organ Weights and Lengths in Quail Hatching Eggs

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ABSTRACT

This study investigated the effects of in-ovo injection of manganese on some organ weights and length in quail hatching (*Coturnix coturnix japonica*) eggs. In total, 400 quail hatching eggs were randomly divided into four treatment groups of 100 eggs per treatment with four replicates of 25 eggs each. On the 14th day of incubation, eggs from group 1 were not injected (control(C)), group 2 was injected with Mn-bioplex at 20 µg per egg (Mn20), group 3 was injected with Mn-bioplex at 40 µg per egg (Mn40), and group 4 was injected with Mn-bioplex at 80 µg per egg (Mn80). There were no significant differences among treatments for chick weight, yolk sac weight, liver weight, and heart weight. The highest chick length was obtained from the C treatment. No significant differences were found in chick length among Mn treatment groups. The beak length of C treatment were lower compares with that of the Mn80 treatment group ($P<0.05$). But no significant differences were observed in beak length among Mn treatments groups. The leg length in C group was significantly lower than Mn40 and Mn80 treatments groups, except Mn20 treatment group ($P<0.01$). As a result, it can be said that injection of Mn into the hatching quail eggs causes the chick and leg lengths to decrease, and beak length to increase without affecting the weight parameters examined.

1. Introduction

In-ovo injection is applied in order to provide positive effects on hatchability, chick quality and chick performance by giving various substances to amniotic fluid during the development of the embryo (Uni and Ferket 2004; Abdulqader et al. 2017; Açıkgöz and Kırkpınar 2017; Peebles 2018). In-ovo application was first applied to turkey eggs for vaccination against Marek's disease by Sharma and Burmester (1982).

In recent years, in ovo application have received considerable interest from researchers and has been used in-ovo applications of substances such as carbohydrates (Tako et al. 2004; Salmanzadeh et al. 2012), proteins (Ohta et al. 1999; Bhanja et al. 2014), vitamins (Ipek et al. 2004; Selim et al. 2012; Bello et al. 2015), minerals (Ghobadi and Matin 2015; Ebrahimi et al. 2016; Jose et al. 2017), prebiotic/probiotics (Ashouri 2014; Pruszynska-Oszmalek et al. 2015; Majidi-Mosleh et al. 2017; Triplett et al. 2018), hormone (Moore et al. 1994; Kocamis et al. 1999) and bee products (Coşkun et al. 2014; Aygun 2016; Bozbay et al. 2016).

Manganese (Mn) is a trace element for animal nutrition. Its plays a vital role in growth, leg development and perosis prevention because it is involved in many enzyme systems in carbohydrate and lipid metabolism (Olgun 2017). Egg yolk is the main source of trace elements during the incubation period. However, trace elements as Mn, zinc and copper are thoroughly decreased in concentration in yolk which is the main reservoir of the trace minerals 17th incubation (Yair and Uni 2011). The in ovo injection of these elements in the last period of the incubation may support embryo development. In ovo injection of trace elements have been reported to improved post-hatch chicken growth, and bone properties (Oliveira et al. 2015). Yair and Uni (2011) reported that in ovo at the 17^{th} day of injection of mineral include Mn (0.36 mg/egg) increased their consumption by the broiler embryo.

The aim of this study were to determined effect of in ovo injection of Mn-bioplex as organic Mn sources into breeder quail eggs on hatching parameters and some measure of the body.

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2. Materials and Methods

Hatching Eggs

This research was carried out at the hatchery laboratory, Department of Animal Science, Faculty of Agriculture, Selcuk University, Konya, Turkey. A total of 400 quail hatching eggs with an average weight of 11-12 g was obtained from Japanese (*Coturnix coturnix japonica*) quails reared on a local farm (Konya, Turkey). The eggs randomly assigned to four treatment groups with 100 eggs per treatment with four replicates of 25 eggs each.

Incubation Management

Eggs were incubated in a commercial incubator at 37.5 °C and 55-60% relative humidity and the eggs were turned 12 times at 90° per day until d 14 of incubation. The incubator condition were changed to 37.2 °C and 75% relative humidity.

Prepare of the solutions

Manganese-bioplex (15 % Mn) was used as an organic manganese source in the experiment. Manganese-bioplex was dissolved in 0.9 % NaCl containing 20, 40 and 80 µg Mn per egg of the solution. The treatment groups were as follows: (1) Control (uninjected; C), (2) injected with Mn-bioplex at 20 µg per egg (Mn20), (3) injected with Mn-bioplex at 40 µg per egg (Mn40) and (4) injected with Mn-bioplex at 80 µg per egg (Mn80).

Injection Procedure

After the blunt end of the egg was disinfected with the 70% alcohol, a hole was opened with a micromotor (Strong 210, Korea). The Mn solution were injected (0.20 ml) into the amnion fluid with a 26-gauge plastic disposable syringe. After the injection, the eggs were transferred to the hatch baskets after the hole was sealed with the adhesive.

Organ weights and lengths

After the incubation, 8 chicks from each treatment were randomly selected, weighed, some tissues and organ lengths (chick, beak, wing, and leg) were measured with a micrometer by adapting to Molenaar et al. (2008) and killed by cervical dislocation. The yolk sac, liver, and heart were removed; they, along with the yolk-free chick body, were weighed to the nearest 0.001 g. Organ weights were expressed as a percentage of chick body weight.

Statistical analysis

At the end of the experiment, the variance analyses were applied to all variables obtained from the trial groups (Minitab 2000), and the differences between means of the groups were determined by the Duncan test (Duncan 1955).

3. Results and discussion

The effects of in-ovo injection of Mn on chick weight, yolk sac weight, liver weight, and heart weight are shown in Table 1.

Table 1
The effects of in-ovo injection of Mn on chick weight, yolk sac weight, liver weight, and heart weight

Parameters	Manganese levels, µg/egg				SEM	*P*-value
	0	20	40	80		
Chick weight, g	9.02	8.40	7.74	8.41	0.334	0.095
Yolk sac weight, %	0.935	1.164	1.096	1.172	0.0928	0.278
Liver weight, %	0.182	0.192	0.166	0.190	0.0138	0.567
Heart weight, %	0.071	0.066	0.066	0.123	0.0287	0.526

There were no significant differences among treatments for chick weight, yolk sac weight, liver weight, and heart weight. Similarly, Oliveira et al. (2015) and Yair et al. (2015) reported that injection of mineral mixtures with different Mn (from 0.013 to 0.039 mg per egg) content into the egg did not affect hatching weight in broilers.

The effects of in-ovo injection of Mn on chick length, beak length, wing length, and leg length are presented in Table 2.

Table 2
The effects of in-ovo injection of Mn on chick length, beak length, wing length, and leg length

Parameters	Manganese levels, µg/egg				SEM	*P*-value
	0	20	40	80		
Chick length, cm	11.12^{a}	10.44^{b}	10.46^{b}	10.49^{b}	0.140	0.005
Beak length, mm	4.82^{b}	5.53^{a}	5.09^{ab}	5.03^{ab}	0.155	0.028
Wing length, mm	19.91	20.13	20.26	20.12	0.441	0.957
Leg length, mm	11.98^{a}	11.05^{ab}	10.01^{b}	10.17^{b}	0.338	0.002

^{a,b}Means within a line with different superscripts differ significantly ($P < 0.05$).

The highest chick length was obtained from the C treatment. No significant differences were found in chick length among Mn treatment groups. The beak length of C treatment were lower compares with that of the Mn20 treatment group ($P<0.05$). But no significant differences were observed in beak length among Mn treatments groups. The leg length in C group was significantly higher than Mn40 and Mn80 treatments groups, except Mn20 treatment group ($P<0.01$). In previous studies, no studies have been conducted to report the effects of treatments on chick length. However, Oliveira et al. (2015) and Yair et al. (2015) showed that injection of mineral mixtures with different Mn (from 0.013 to 0.039 mg per egg) content into the egg did not affect tibia length in broilers. Contrary to the present study, early studies have reported that Mn deficiency in the diet causes shortness of legs and beaks in chicks (Caskey and Norris 1939). (Favero et al. 2013a; Favero et al. 2013b) reported that broiler breeders fed diets containing different levels of Mn (164 or 204 mg/kg) did not affect tibia or chick length. Similarly, Bozkurt et al. (2015) reported that the addition of Mn at levels of 6.25, 12.50, 25 and 50 mg/kg to broilers did not affect tibia length and weight at the end of the trial. These differences may be due to the method of administration (addition of diet or injection into egg) of Mn. In addition, shortening of the leg length as a result of Mn injection into egg may result in an advantage in preventing bone abnormalities that may be caused by rapid growth during the breeding period. The long beak length may be advantageous in terms of feed intake after incubation. On the other hand, this situation may negatively affect animal welfare by the beak trimming and by increasing pecking during the production period.

As a result, it can be said that injection of Mn into the hatching quail eggs causes the chick and leg lengths to decrease without affecting the weight parameters examined and further studies are needed.

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