

The Effect of Vitamin C in-ovo Injection on Incubation Results of Fertile Goose Eggs

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ABSTRACT

The research aims to determine the effect of vitamin C (Ascorbic acid) injected at different doses on the hatchability of eggs obtained from domestic geese raised in the Afyonkarahisar region. Injection doses of vitamin C, 8, and 10 mg were applied to the air cell of the eggs on the 24th day of incubation. As a result of the experiment, vitamin C given to fertile goose eggs positively affected hatch weight, chick length, and chick quality. There was no significant difference between the groups regarding hatchability and survival rate.

Döllü Kaz Yumurtalarında Vitamin C In-Ovo Enjeksiyonunun Kuluçka Sonuçlarına Etkisi

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ÖZ

Araştırmanın amacı, Afyonkarahisar yöresinde yetiştirilen yerli kazlardan elde edilen yumurtalara farklı dozlarda enjekte edilen C vitamininin (Askorbik asit) kuluçka kabiliyetine etkisini belirlemektir. Enjekte edilen C vitamini dozları, inkübasyonun 24. gününde 8 ve 10 mg olmak üzere yumurtaların hava hücreğine uygulanmıştır. Deneme sonucunda döllü kaz yumurtalarına yumurta içi verilen vitamin C'nin çıkım ağırlığı, civciv uzunluğu ve civciv kalitesi üzerine olumlu etki yapmıştır. Çıkış gücü ve yaşama gücü bakımından gruplar arasında anlamlı bir farklılık bulunmamıştır.

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1. Introduction

One of the most important problems in goose breeding is incubation. The low egg yield and the fact that they do not lay eggs during natural incubation increase the importance of artificial incubation in goose breeding. The hatching efficiency, which is reported as 75% in natural hatching in geese, drops considerably when the necessary conditions are not met in artificial hatching (Tilki and İnal, 2004; Pesmen and Yonetken, 2020).

The technique, also known as in-egg feeding, is applied by injecting nutrients such as protein, vitamins, minerals, and various substances such as hormones and antibodies into the embryonic sacs in the form of a liquid solution at any stage of incubation. The in ovo technique was first used at the end of the 1970s, and in 1995; automatic injection systems (in ovoject) started to become an alternative application that has come to the fore in recent years (Abdulqader et al., 2017). Introducing various nutrients (protein, minerals, vitamins etc.) into the egg increases the hatchability and ensures high survival and performance after hatching (İpek et al., 2003; Nowaczewski et al., 2012; Sözcü and Curabay, 2014; Zhu et al., 2019).

Vitamin C has anti-stress properties. It helps collagen synthesis, positively affects connective tissue, bone, and cartilage tissue strengthens the immune system and increases disease resistance (Santos et al., 2018). Vitamin C is absent in a newly laid egg and begins to be synthesized by the developing embryo on the 3rd-4th day of incubation due to endogenous biosynthesis. However, the quantities produced may not be sufficient towards the end of the incubation period, when the embryo is exposed to extreme overheating. This is mainly observed in ducks and geese but is also observed during broiler incubation. Waterfowl have exceptionally high vitamin C requirements and are more susceptible to vitamin C deficiency. Therefore, in ovo application of vitamin C to the egg during embryogenesis can reduce the negative effects of overheating (thermal stress) and, as a result, increase hatchability. Therefore, vitamin C injection may provide more effective results in the middle and late stages of incubation, when the embryo is expected to overheat (Nowaczewski et al., 2012).

In hot environmental conditions, the level of vitamin C in the blood and tissues decreases significantly, and exogenous vitamin C increases the level of vitamin C in the blood, reducing the rise in the birds' body temperatures under heat stress conditions. It is known that positive responses to vitamin C supplementation are further enhanced under adverse conditions such as high temperature and low protein levels (Pardue and Thaxton, 1986). Khan et al. (2012) reported that heat stress caused a decrease in feed consumption, food use, growth rate, egg production and quality, feed efficiency, immunity level, performance, and productivity. Decreased antioxidant levels can characterize heat stress due to increased oxidative stress.

Zhu et al. (2019), in the prenatal period, vitamin supply (in egg feeding of exogenous vitamins (IOF)) improves broiler chickens' hatchability and growth performance, antioxidation, and immune function. Brake and Pardue (1998) reported that the anti-stress feature of vitamin C is one of its most essential features. Giving additional ascorbic acid to animals in different ways, especially at high temperatures, reduces the negative effects that may occur in the yield characteristics.

To summarize the results obtained in various studies by administering Vitamin C to fertile poultry eggs in ovo at multiple stages of incubation;

- High hatchability in broiler eggs with in ovo application of 3 mg Vit C to the air sac on the 18th day of hatching (İpek et al., 2003),

- In ovo application of 8 mg Vit C to the air sac on the 20th day of hatching in Pekin duck eggs, an increase in hatchability and a significant decrease in embryo mortality (Nowaczewski et al., 2012),
- In broiler eggs, with the application of 3 mg Vit C to the air sac in ovo on the 13th day of hatching, an increase in hatchability, a significant decrease in embryo deaths,
- In ovo application of 3 mg Vit C to egg yolk on the 15th day of hatching to broiler eggs increased hatchability and chick weight and improved immune function (Zhu et al., 2019),
- In broiler eggs, on the 14th day of hatching, an increase in hatchability and an increase in growth performance after hatching were observed with in ovo application of 6 µg Vit C to the air sac (Ismail et al., 2019).

This study was carried out to determine the effects of vitamin C in ovo injection during the incubation of fertile domestic goose eggs on hatchability, chick quality, chick weight, chick length, and 1-week survival rate.

2. Material and Method

A total of 82 fertile goose eggs were used in the study. Goose eggs were weighed and numbered with an accuracy of 0.01 g before being placed in the incubator. It was then incubated according to the method in Table 1. On the 10th day of incubation, a fertility examination was performed on the eggs with lamp control, and the infertile eggs were removed from the incubator. Eggs identified as fertile were placed back in the incubator to continue the normal incubation process. Until the 27th day of the incubation period, automatic rotation was performed at an angle of 45 degrees every hour.

Table 1. Incubation conditions of fertile goose eggs (Petersime, 1993).

Days	Heating (°C)	Humidity (%)	Cooling and water spraying
1-7	37.7	58	
8-10	37.7	58	5 minutes cooling+ water spraying
10	37.7	58	1 st control
11-14	37.7	58	5 minutes cooling+ water spraying
15-21	37.7	58	15 minutes cooling+ water spraying
22-27	37.7	58	25 minutes cooling+ water spraying
27	37.7	58	2 nd control and transfer
28-30	37.2	76	

During to incubation fertile eggs were subjected to treatments on the 24th days of incubation given in Table 2.

Table 2. Application procedures according to trial groups.

Gruplar	Uygulama
Control	K No application has been made.
Negative control	NK 0.1 ml of isotonic water was injected.
8 mg AA	AA1 Each egg was injected with a 0.1 ml solution containing 8 mg of Vit C.
10 mg AA	AA2 Each egg was injected with 0.1 mg of the solution containing 10 mg of Vit C.

K: Control, **NK:** Negative control, **AA1:** 8 mg Vit C, **AA2:** 10 mg Vit C

After the area above the air cell was wiped with 70% ethanol before in-ovo injection, a small 1 mm hole was drilled with a micromotor from the egg shell along the central axis at the top of the egg. 0.1 ml of a solution containing doses of Vitamin C (8 mg and 10 mg) was injected through the hole drilled in the eggshell, through the inner membrane of the eggshell, with a needle of 3-4 mm in length.

Figure 1. Drilling holes in fertile goose eggs with a micro motor and Vit C injection.



The hole was then sealed with a small drop of sterilized molten paraffin, and the eggs were placed back in the incubator. The eggs were transferred to the hatching section on the 27th day. At the end of the incubation period (30th day), hatchability, chick weight, chick length, chick quality, and 1-week survival rate were determined.

Hatchability; is evaluated as the ratio of chicks obtained from eggs determined to be fertile due to fertility control. The hatchability was determined by dividing the number of chicks hatched in each group by the number of fertile eggs (Aksoy, 1999).

Hatchability= number of hatched chicks/number of fertile eggs x 100

Chick weight; was determined with a digital scale with a sensitivity of 0.01 g.

Chick length; the length from the tip of the beak to the tip of the finger was measured in cm with the help of a ruler (Wolanski et al., 2005).

Chick quality; was evaluated and scored according to Tona et al.'s (2003) chick quality scale. General activity, feather condition, and appearance, remaining egg yolk, eyes, legs, navel region, and remaining, membrane parameters were evaluated in scoring.

Survival rate: Survival rate values were calculated according to the formula below by determining the ones that died at the age of 1 week among the goose chicks in each group (Şenköylü, 1991).

Survival rate (%) = (Initial number of animals of the group-number of animals that died)/initial number of animals of the group x100

Statistical analysis was made with the SPSS statistical package program (SPSS, 2018). ANOVA test was used to determine the differences between groups regarding chick weight, length, and quality. The chi-square test determined the differences between groups regarding hatchability and survival rate.

3. Results and Discussion

The Effects of Vitamin C injection in fertile goose eggs on the 24th day of incubation on chick weight, length, quality, hatchability, and survival rate are given in Table 3.

Table 3. Effects of Vitamin C injection in fertile goose eggs on the 24th day of incubation on chick weight, length, quality, hatchability, and survival rate.

Grup		Properties				
		Chick Weight (g)	Chick Length (cm)	Chick Quality	Hatchability (%)	Survival Rate (%)
K	Mean	100.97	23.32	80.00	70.00	64.29
	Min-max	76.41-123.77	21.00-25.00	66.00-92.00		
	SE	3.93	0.26	2.32	0.10	0.13
	n	14	14	14	20	14
NK	Mean	93.90	23.70	86.00	50.00	70.00
	Min-max	71.11-108.25	22.50-24.50	80.00-90.00		
	SE	3.77	0.18	1.63	0.11	0.15
	n	10	10	10	20	10
AA1	Mean	110.75	24.78	96.00	76.19	100.00
	Min-max	94.92-121.65	23.00-26.00	86.00-100.00		
	SE	2.00	0.17	1.01	0.95	0.00
	n	16	16	16	21	16
AA2	Mean	112.92	24.36	86.00	52.38	72.73
	Min-max	87.10-140.98	23.00-25.50	80.00-92.00		
	SE	4.51	0.26	1.68	0.11	0.14
	n	11	11	11	21	11

($p \leq 0.05$).

Chick Weight: Chick weight in K, NK, AA1, and AA2 groups, respectively, was 100.97 g, 93.90 g, 110.75 g, and 112.92 g. A significant difference was found in chick weight between NK, AA1, and AA2 groups ($p \leq 0.05$). The highest chick weight was determined in the AA2 group and the lowest in the NK group.

Chick Length: Chick lengths in K, NK, AA1, and AA2 groups, respectively, were found as 23.32 cm, 23.70 cm, 24.78 cm, and 24.36 cm. A significant difference was found between the K and AA1 groups in terms of chick length ($p \leq 0.05$). The highest chick length was found in the AA1 group and the lowest in the K group.

Chick quality: Chick quality in K, NK, AA1, and AA2 groups, respectively; 80.00, 86.00, 96.00, and 86.00. A significant difference was found between the AA1 and the F, NK, and AA2 groups ($p \leq 0.05$). The highest chick quality was determined in the AA1 group and the lowest in the control group.

Hatchability: Hatchability power in K, NK, AA1, and AA2 groups was 70%, 50%, 80%, and 52%. There was no significant difference between the groups in terms of hatchability ($p \geq 0.05$). The lowest

hatchability was found in the saline-injected group (NK), and the highest in the 8 mg Vit C injected group (AA1).

Survival rate: The survival rate in C, NK, AA1, and AA2 groups was 64.29%, 70.00%, 100%, and 72.73%. There was no significant difference between the groups in terms of survivability ($p \geq 0.05$). The lowest survival rate was found in the control group and the highest in the AA1 group.

Various studies have observed that Vitamin C (8 mg) administered in ovo causes an increase in chick weight (İpek et al., 2003; Nowaczewski et al., 2012; İsmail et al., 2019; Zhu et al., 2019). It is consistent with other results obtained in the study.

The majority of Vit C administrations by in ovo injection were performed on fertile broiler eggs. In the literature researches, studies on ovo injection in geese were not found. In studies conducted on other poultry species, in ovo administration of Vit C increased hatchability and decreased the rate of dead embryos (İpek et al., 2003; Nowaczewski et al., 2012; İsmail et al., 2019; Zhu et al., 2019). Although the hatchability and survivability increased in the group injected with Vit C (8 mg), no significant difference was detected in the study performed. This situation may have resulted from the species characteristic and different hatching conditions of the poultry species. In addition, the place and time of in-ovo injection are important. More precise results will be obtained if vitamin C or other nutrients are applied at different hatching times.

Various researchers have found a high correlation between chick weight, chick quality, and chick length (Petek et al., 2008; Willemsen et al., 2008; Lin et al., 2018; Pesmen, 2021). In the study, both chick length and chick quality increased in the Vitamin C (8 mg) injected group. An objective and reproducible method is required to day-old chicks' quality and estimates their performance potential. Several methods have been developed in this regard, but not all methods have the same repeatability or predictive value. Chick length is one of the methods reported to be useful as it reflects embryo development during hatching. Studies have shown that there is a positive correlation between chick length and performance at older ages (Meijerhof, 2009).

As a result, 8 mg of Vitamin C injected in the study increased hatchability, chick weight, chick length, chick quality, and survivability in fertile goose eggs.

4. Conclusions

In various studies, high hatchability in broiler fertile eggs, decrease in embryo mortality, increase in chick weight, and increase in post-hatch growth performance have been achieved. Given that healthy and well-nourished broiler breeders produce eggs with complete nutritional characteristics for all embryo development, in-egg feeding is a technique that may show the best results for young broiler breeders who may have difficulties in passing adequate nutrients to the egg. Under these conditions, in ovo feeding can provide the chicks with corrective nutritional composition for eggs and make up for digestive deficiencies. In addition, more clear results will be obtained with intensive research on in-ovo injection in goose eggs.

Conflict of Interest Statement

The article's author declares that there is no conflict of interest.

Contribution Rate Statement Summary of Researchers

The author declares that she has contributed 100% to the article.

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