The effect of in ovo ascorbic acid and glucose injection in broiler breeder eggs on hatchability and chick weight

Einfluss von in ovo-Injektionen von Ascorbinsäure und Glukose in Eier von Broilerelternieren auf die Schlüpfhäftigkeit und das Kükgewicht

A. Ipek, U. Sahan and B. Yilmaz


Introduction

An increase of one unit in hatchability of total eggs, a primary criterion of productivity in breeder farms, is of great economical value. Nutrient administration in ovo may provide poultry companies with an alternative method to increasing hatchability (OHTA et al., 2001).

Egg injections of vitamins, such as pyridoxine and pantothenic acid, were applied during incubation in several experiments to study the effects of vitamins on hatchability of turkey eggs (ROBEL and CHRISTENSEN, 1991; ROBEL, 1993). Ascorbic Acid (AA) as an anti-stress agent is the most important vitamin (BRAKE and PARDUE, 1998). Chick embryos may be subjected to stress caused by excessive production of heat during the latter part of egg incubation (TULLETT, 1990). Therefore, it may be expected that the addition of AA may be beneficial for compensating embryonic stress (ZAKARIA and AL-ANEZI, 1996). On the other hand, ELIBOL et al. (2001) reported that the injection of 3 mg of AA to the eggs in the weight group of 70 g and above on the 13th day of incubation significantly reduced the late term embryo mortality. AA was dissolved in deionized sterile water and administered in varying concentrations and volumes. Hatchability and body weight were not significantly affected by deionized sterile water injections. AA injections of 0.75 g/ml were found to be toxic and resulted in embryo mortality prior to hatching (INGRAM et al., 1997a).

Glucose is the major energy source of living organisms (STRYER, 1995). INGRAM et al. (1997b) investigated the effect of in ovo injection of glucose in varying levels to broiler eggs prior to the transfer to the hatchers on the hatchability, chick weight and subsequent body weight, and reported that low levels of glucose significantly improved hatchability.

The present study was carried out with the aim of compensating the stress factors caused by the increase in the metabolic heat of the embryo via AA application, and providing a supplementary energy source to the embryo prior to hatching through glucose application, and determining its effects on embryonic mortality, hatchability and chick hatch weight.

Material and Methods

The study was carried out in a private hatchery. The eggs used in the study were obtained from Ross parental broiler strain at 70 weeks of age. All eggs were obtained from the same breeder flock and laid within a 24 h period. Eggs were weighed on a balance with 0.1 g precision and eggs with a weight of 68–70 g were incubated at 37.8 °C and 60% RH. Two experiments were conducted utilizing 3,564 fertilized commercial eggs.

Experiment 1 was carried out on the 13th day of incubation after candling and live embryos were subjected to the following treatments, using the in ovo injection system.1 Eggs were injected with AA (Ascorbic Acid Powder, Item Catalog Number 0.80, Roche, Germany) in the center of the air cell: 1 – uninjected (control); 2 – eggs injected with 0.5 ml saline solution; 3 – eggs injected with 0.5 ml of saline solution containing 1, 3, 5 or 7 mg of AA per egg. Control eggs were removed from the incubator together with the treated groups, and kept in the same environment. For each treatment 324 fertilized eggs were used.

In experiment 2 glucose (Item Catalog Number 108337, Merck, Germany) injections at varying concentrations were applied to the eggs before hatching with the aim of providing a supplementary energy source to the embryo. For this purpose live embryos candled prior to transfer were subjected to the following treatments on day 18 using in ovo injection system. Eggs were injected with glucose in the center of the air cell: 1 – uninjected (control); 2 – eggs injected with 0.5 ml saline solution containing 5, 10 or 15 mg of glucose. The control group was kept in the same environmental conditions during treatments. For each treatment 324 fertilized eggs were used.

Cull chicks and dead ones as well as unhatched eggs were discarded after the others hatched. Embryo mortality was determined in unhatched eggs. Hatchability was calculated by considering the ratio of salable chicks hatched to the live embryos after the treatment. Chick hatch weight was determined by weighing all chicks hatched one by one.

The two experiments were subjected to analysis of variance (SAS, 1985), utilizing ANOVA procedures for balanced data. Analysis for percentage data were conducted

1 Embrex, Inc.

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after an arc sine transformation of the data. Significant differences among treatment means were determined by Duncan’s multiple range test.

**Results**

The effects of AA injections of different concentrations on the hatchability results in Experiment 1 are given in Table 1. Embryonic mortality between the 14th day and the 17th day of incubation were determined as 1.9, 2.2, 1.8, 1.7, 1.8 and 1.7% in control saline solution, 1, 3, 5 and 7 mg AA applications, respectively. The differences among the groups were not significant. The effect of applications on the embryonic mortality between the 18th day and the 21st day was found to be significant (P < 0.01). The lowest embryonic mortality during this period was determined as 5.2% in the group subjected to 3 mg of AA injection. Control, saline solution, 1, 5 and 7 mg AA injection groups did not differ statistically. No significant difference was observed among the application groups with respect to the number of dead embryos after breaking the pips, culls and dead chicks. The effect of applications on the hatchability of fertile eggs was found to be significant (P < 0.01). The highest hatchability was obtained from the group treated with AA at 3 mg concentration. Increasing the concentrations of AA applied to eggs did not have a positive effect on the hatchability and AA injected eggs did not differ from the control.

The incubation results obtained from Experiment 2 are given in Table 2. The effects of applications on the embryonic mortality between the 19th day and the 21st day of incubation, numbers of piped, dead and culled chicks, hatchability and chick hatch weight did not differ significantly (P > 0.01).

**Discussion**

The present study utilized injection during the latter part of incubation to study the effect of AA and glucose on embryonic mortality, hatchability, chick quality and chick weight of broiler chicken embryos. The effect of AA applications on the hatchability of fertile eggs was found to be significant (P < 0.01). The highest hatchability was obtained from the group treated with AA at 3 mg concentration. Increasing the concentrations of AA applied to eggs did not have a positive effect on the hatchability and AA injected eggs did not differ from the control. The results obtained from the study fall into line with the research results of Zakaria and Al-Anezi (1996).

Higher rates of embryonic mortality observed in heavy eggs especially in the later period leads to a decline in the hatchability of fertile eggs (French, 1994). Tulleit (1990) reported that chick embryos were subjected to stress factors due to the increase in metabolic heat after the 10th day of incubation and hence the ratio of embryonic mortality increased. Therefore, AA injection at 3 mg

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**Table 1.** Effect of ascorbic acid injection at day 13 of incubation on embryonic mortality, hatchability and chick hatch weight of commercial broiler chickens (mean ± SEM)

<table>
<thead>
<tr>
<th>Injection Dose of Ascorbic Acid (mg)</th>
<th>Unhatched egg with</th>
<th>Dead + Cull Chicks</th>
<th>Hatchability</th>
<th>Chick Hatch Weight</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>14–17 Day Embryo Died</td>
<td>18–21 Day Embryo Died</td>
<td>Piped</td>
<td>NS</td>
</tr>
<tr>
<td>Control</td>
<td>1.9 ± 0.8</td>
<td>10.4 ± 1.7a</td>
<td>6.9 ± 0.9a</td>
<td>4.6 ± 0.8</td>
</tr>
<tr>
<td>Saline Solution</td>
<td>2.2 ± 0.9</td>
<td>9.8 ± 1.5a</td>
<td>6.7 ± 0.8a</td>
<td>4.5 ± 0.8</td>
</tr>
<tr>
<td>1</td>
<td>1.8 ± 0.7</td>
<td>9.7 ± 1.2a</td>
<td>7.0 ± 1.1a</td>
<td>4.4 ± 0.7</td>
</tr>
<tr>
<td>3</td>
<td>1.7 ± 0.7</td>
<td>5.2 ± 1.0n</td>
<td>5.4 ± 0.6b</td>
<td>4.2 ± 0.8</td>
</tr>
<tr>
<td>5</td>
<td>1.8 ± 0.8</td>
<td>8.3 ± 1.3a</td>
<td>5.3 ± 0.6d</td>
<td>4.0 ± 0.6</td>
</tr>
<tr>
<td>7</td>
<td>1.7 ± 0.8</td>
<td>8.8 ± 1.4a</td>
<td>7.4 ± 1.2a</td>
<td>4.7 ± 0.9</td>
</tr>
</tbody>
</table>

a, b, c; Values within columns with no common letter differ significantly

**Table 2.** Effect of glucose injection at day 18 of incubation on embryonic mortality, hatchability and chick hatch weight of commercial broiler chickens (mean ± SEM)

<table>
<thead>
<tr>
<th>Injection Dose of Ascorbic Acid (mg)</th>
<th>Unhatched egg with</th>
<th>Dead + Cull Chicks</th>
<th>Hatchability</th>
<th>Chick hatch weight</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>18–21 Day Embryo Died</td>
<td>Piped</td>
<td>NS</td>
<td>NS</td>
</tr>
<tr>
<td>Control</td>
<td>8.8 ± 1.3</td>
<td>7.2 ± 1.2</td>
<td>4.7 ± 1.0</td>
<td>79.3 ± 2.4</td>
</tr>
<tr>
<td>0.5 ml deionized sterile water</td>
<td>8.3 ± 1.4</td>
<td>7.4 ± 1.2</td>
<td>4.1 ± 0.8</td>
<td>80.2 ± 2.8</td>
</tr>
<tr>
<td>1</td>
<td>8.4 ± 1.4</td>
<td>7.3 ± 1.2</td>
<td>4.4 ± 0.9</td>
<td>79.9 ± 1.9</td>
</tr>
<tr>
<td>15</td>
<td>8.6 ± 1.5</td>
<td>7.2 ± 1.2</td>
<td>4.8 ± 0.8</td>
<td>79.4 ± 2.3</td>
</tr>
</tbody>
</table>
concentration might have a role in reducing the heat stress in the embryo. The increase in hatchability is an indication of this effect. This result is in accordance with the results of Zakaria and Al-Anizi (1996). Wilson and Jaworski (1992) stated that plasma AA concentration declined at the 15th day of incubation in White Leghorn chicken eggs. Elevation of blood corticosterone is usually associated with stress and with the reduction of AA biosynthesis in poultry (Kutlu and Forbes, 1994). Generally, AA may be regarded as an anti-stress agent (Pardue and Thaxton, 1986), because it may lead to the reduction of corticosterone (Satterlee et al., 1994). Additionally, AA has a role in collagen synthesis (Weiser et al., 1988), the metabolism of minerals (Roberson and Edwards, 1994), and vitamin D metabolism (Weiser et al., 1988). The effects of high concentrations of AA application on the hatchability results have been investigated by different researchers (Zakaria and Al-Anizi, 1996; Ingram et al. 1997a). However, reports indicate that increasing the concentration of AA is accompanied by a selective toxic action on the pancreatic beta cells (Meglasson and Hazelwood, 1982).

The effects of glucose applications on the embryonic mortality between the 19th day and the 21st day of incubation, numbers of piped, dead and culled chicks, hatchability and chick hatch weight were not found to be significant. Berdeanu (1997b) investigated the effects of in ovo injection of glucose at different doses to broiler breeder eggs during transfer, on hatchability, chick weight and subsequent body weight. They reported that glucose applied at levels lower than 25 mg increased the hatchability of fertile eggs. This result is different from the findings in our study. However, our results are in accordance with their conclusions in which they reported that different levels of glucose did not have a statistically significant effect on chick weight.

Consequently, supplying various nutrients into the eggs to increase the incubation success at hatching has become a highly practicable treatment thanks to the in-ovo injection (Embrex) system which has become widespread in recent years. Therefore, 3 mg AA injection into eggs during the period during which the heat production begins to increase will positively affect the hatchability of fertile eggs. No effect of glucose injection was determined on the hatchability and chick weight at transfer. Nevertheless, strain, age, egg size or breeder condition might have an effect on the influence of glucose. Therefore, it can be suggested that the effect of glucose should be examined with these traits in detail. More detailed studies directed towards applying different nutrients into the egg are needed in order to increase hatchability and incubation success via in ovo injection method, in future.

Summary

This study was carried out with the aim of eliminating the stress factors caused by the increase in metabolic heat of the embryo during incubation via AA application, and by providing a supplementary energy source to the embryo prior to hatching through glucose application, and determining its effects on embryonic mortality, hatchability and chick hatch weight. Experiment 1 was carried out on the 13th day of incubation after candling and live embryos were subjected to the following treatments, using the in ovo injection system: 1 – un.injected (control); 2 – eggs injected with 0.5 ml sterile saline solution; 3 – eggs injected with 0.5 ml of saline solution containing 1, 3, 5 or 7 mg of AA per egg. In experiment 2, glucose injection at varying concentrations was applied to the eggs before hatching. For this purpose live embryos candled prior to transfer were subjected to the following treatments on day 18 using in ovo injection (Embrex) system: 1 – un injected (control); 2 – eggs injected with 0.5 ml deionized sterile water; 3 – eggs injected with 0.5 ml of deionized sterile water containing 5, 10 or 15 mg of glucose. The effect of AA injection on the hatchability of fertile eggs was found to be significant (P < 0.01). The highest hatchability was obtained from the group treated with AA at 3 mg concentration. No effect of glucose injection was determined on the hatchability and chick weight.

Key words

In ovo injection, ascorbic acid, glucose, broiler breeder eggs, hatchability

Zusammenfassung

Einfluss von in ovo-Injektionen von Ascorbinsäure und Glukose in Eier von Bröllerrartenieren auf die Schlupffähigkeit und das Küken-gewicht

Die Untersuchung hatte zum Ziel, den Stress des Embryos während der Brut, der durch die Zunahme der metabolischen Abwärme ausgelöst wird, durch eine Injektion von Ascorbinsäure bzw. von Glukose als zusätzlicher Energiequelle in das Brutei zu kompensieren. Als Indikatoren für die Wirkung wurden die Bryonalersterblichkeit, die Schlupfrate und das Schupfgewicht der Küken ausgewählt. In Versuch 1 wurden am 13. Bruttag nach dem Schieren der Eier die lebenden Embryonen folgenden Behandlungen mittels in-ovo-Injektionstechnik unterzogen: 1 – keine Injektion (Kontrolle); 2 – Injektion mit 0,5 ml steriler Kochsalzlösung; 3 – Injektion mit 0,5 ml steriler Kochsalzlösung, die 1, 3, 5 oder 7 mg Ascorbinsäure enthielt. In Versuch 2 wurden am 18. Bruttag nach dem Schieren während der Umlage zur Schlupfbrut in die Eier mit lebenden Küken folgende Glukose-mengen mittels in-ovo-Injektionstechnik appliziert: 1 – keine Injektion (Kontrolle); 2 – Injektion mit 0,5 ml sterilem deionisiertem Wasser; 3 – Injektion mit sterilem deionisiertem Wasser, das 5, 10 oder 15 mg Glucose enthielt. Die Injektion von Ascorbinsäure hat sich signifikant auf die Schlupfrate der befruchtenen Eier ausgewirkt (P < 0.01). Die höchste Schlupfrate wurde bei einer Injektion von 3 mg Ascorbinsäure beobachtet. Dagegen hatte die Glukoseinjektion keinen Einfluss auf die Schlupfrate und das Küken-gewicht.

Stichworte

In-ovo-Injektionstechnik, Ascorbinsäure, Glukose, Bruteier, Schlupfrate

References


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