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Hatching Performance of Indonesian Native Chicken Supplemented by L-Glutamine at Different Days of Incubation

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Abstract

The present study aimed to determine the hatching performance of native chicken subjected to the supplementation of L-Glutamine at different days of incubation. A total of 240 fertilized eggs native chicken eggs with an initial weight of 48.85 ± 3.3 g, were subjected to injection of glutamine on the 7th, 9th and 11th day of incubation, while the control group received no injection. A total of 1.5% glutamine was dissolved in 0.5 mL of saline solution and injected at the pointed part of the egg with the target into the albumen. Hatchability, incubation time, and chick weight at hatch were determined during the study. The hatchability of native chicken treated with an injection of glutamine amino acid on different incubation days was still lower than of the control group. However, hatches were generally more substantial in size. The incubation time of the injected chicks was longer than that of the control. Chicks from injected of glutamine on the 11th day of incubation were 12.31% heavier than controls and did not differ from injections on the day 7th and 9th of incubation. The results of this study indicated that the administration of glutamine to obtain more massive chicks at the time of hatching could be conducted on the 7th, 9th and 11th days of incubation, even with lower hatchability.

Keywords: glutamine, in ovo-administration, native chicken, hatching performance

A. Introduction

The efforts to increase the productivity of the native chickens that are widely studied and applied today are the utilization of essential nutrients to maximize the embryo potential that develops in the egg during the incubation period. Through this method, chickens are expected to hatch with more excellent performance conditions, and these improvements can be sustained on the post-hatching period or even on the next generation without altering the genetic.

The supplementation of various nutrients look like (amino acids, fatty acids, carbohydrates, minerals, and vitamins) or other substances (hormones, vaccines, drugs, and other metabolites) were into the hatching egg during the incubation period. It had been widely known for beneficial effects on the productive performance of commercial line chickens (broiler or layer type), although these types of chicken have better genetic as the results of the selection for a long time. Reports of performance improvements in the commercial chicken can be seen in several studies such as Uni, Ferket, Tako, & Kedar (2005); Santos, Corzo, McDaniel, Filho, & Araujo (2010); Kornasio, Halevy, Kedar, & Uni (2011); Shafey, Sami, & Abouheif (2013); and Salmanzadeh, Ebrahimneshad, Shahryar, & Ghaleh-Kandi (2016). However, there were few studies on in ovo feeding of amino acids in Indonesian native chicken. Studies conducted by Asmawati (2014) and Azhar, Rahardja, & Pakiding (2016) involving in ovo administration of lysine, methionine, and arginine, improved not only the chicks weight at hatch but also post-hatch performance.

The administration time of glutamine during incubation varies among study based on the objective of the study. Injection of glutamine was done few days before hatching consistently improved growth and development of a digestive tract of duck (Chen, Wang, Xiong, Wan, Xu, & Peng, 2010) and broiler breeder (Shafey et al., 2013). Similar results of broiler breeder digestive tract improvement have been reported by Salmanzadeh et al. (2016) who injected glutamine on day 7th of incubation. There is no information about the effect of glutamine administration time during incubation on the native chicken performance. Hence, this research aimed to determine the hatching performance of native chicken as the result of glutamine supplementation during different days of incubation.

B. Methodology

1. Incubation and in ovo injection procedures

A total of 300 local chicken eggs were obtained from the university farm and selected based on the uniformity of weight, shape, and color of the shell. Eggs were numbered, weighed individually, and set in a forced-draft incubator equipped with automatic turning. Incubation temperature and humidity were maintained at 37-38 ^{o}C and 50-55% respectively. On the 7th day of incubation, each egg was candled to determine the embryo development. The infertile eggs or the eggs containing dead embryos were removed, and the remaining of fertile eggs were randomly divided into four treatments of glutamine injection with three replications and 20 eggs per replicate. The procedures of glutamine supplementation were performed respectively on the 7th, 9th, and 11th-day incubation while the uninjected group was treated as a control.

Before injection, the selected eggs were candled to identify the albumen position as the injection target. The point of the inserted needle was marked, cleaned with alcohol, sealed with sticky tape, and a small hole was made using the pointed tip of scissor on the marked point. A 0.5 mL solution (1.5% Gln dissolved in sterilized NaCl 0.9%) was injected into the albumen using 24 G needle to a depth of 1 cm. The injection process was conducted under aseptic conditions to avoid contamination. Furthermore, the injection hole was sealed with a nail-paint as a substitute for paraffin. The injected eggs were placed back into the incubator, and the incubation process continues until hatch at the 21st day.

2. Chicks care and data collections

On the 20th day of incubation, the eggs were observed to determine the hatching time, weight of chicks, and hatchability. The hatching time was marked on the observed chicks that had successfully left the shell. Incubation time (hours) is the time needed for the egg incubation process to hatch, calculated from the time the egg sets until it succeeds in getting out of the eggshell. The chicks were marked based on the number of the eggs in each treatments group. Chicks were weighed three h post-hatch to determine the chick weight at hatch and shown as absolute (g) and relative (%) weight. Hatchability (%) was calculated according to the number of hatched chicks from the number of eggs in each group.

3. Data Analysis

Data were analyzed by analyses of variance according to the completely randomized design using the general linear model procedure (Gasperz, 1991). Difference between treatments group was compared by multiple range Duncan test following the ANOVA, and values were considered difference statistically at 5% ($P<0.05$).

C. Result and Discussion

1. Hatchability

The hatchability is the main indicator of the success of chicken eggs hatching. The Effectiveness of external nutrient administration treatment during incubation, including glutamine, when conducted in the early incubation period, showed lower hatchability value compared to control as reported by Salmanzadeh et al. (2016). In the present study, injection of glutamine at different incubation days also showed the same phenomenon (table 1).

Table 1. The hatching performance of the Native Chicken as the results of the supplementation of glutamine at different days of incubation.

Parameters	Supplementation time			
	control	Day-7th	Day-9th	Day-11st
Hatchability (%)	69,9 ± 16,2[b]	28,9 ± 20,6[a]	34,0 ± 13,0[a]	34,85 ± 17,2[a]
Incubation time (h)	490,4 ± 8,3	499,5 ± 3,3	495,7 ± 7,5	495,6 ± 6,6
Absolute hatching weight (g)	31,8 ± 1,0[a]	35,3 ± 2,2[ab]	34,1 ± 1,6[ab]	36,3 ± 2,0[b]
Relative hatching weight (%)	70,8 ± 2,6	72,6 ± 1,5	73,7 ± 0,6	73,0 ± 0,4

[abc]The difference superscript follows the value on the same row were significantly difference ($P < 0.05$).

The administration of glutamine at days 7th, 9th, and 11th of incubation showed hatchability value of treatments groups lower compared to the control group (32.5 vs. 70%). This low hatchability can occur, considering that the treatment applied is an invasive technique for embryos that develops in the first week of incubation. Risk of embryo death of in ovo administering of amino acids at the early of the incubation period is generally higher than the similar method at the end of incubation period as reported by some researchers (Foye, Uni, McMurtry, & Ferket, 2006; Kornasio et al., 2011; Asmawati, 2014).

Among the treatment of the supplementation of glutamine, the number of chicks hatched from the treatments on day 9th, and 11th of incubation groups tends to be higher than the injection that conducted on the day 7th. This condition indicated that embryos survived and have a better response to the glutamine additions, primarily when the injection was held on day 9th or 11th of incubation.

A similar trend of lower value of hatchability of broiler breeder eggs that injected glucose and magnesium on the day 7th of incubation was reported by Salmanzadeh, Ebrahimnezad, Shahryar, & Behesti (2012). This condition illustrated that any substance injected into the fertile eggs at the beginning of the incubation period may alter the internal environmental conditions of the egg and potentially harm the developing embryo's survival.

The albumen was absorbed in large quantities into the ectoderm embryo during the incubation period (Yoshizaki Ito, Hori, Saito, & Iwasawa, 2002). Interaction of albumen composition with exogenous amino acids can be destructive or protective through changes in the content of *Avidin* or *nuclease* (Lu, Killoran, & Riley, 2003). It can be a consideration in determining the proper of the injection day and take into the account that the albumen on the day 7th of incubation have a higher proportion compared to the following days.

2. Incubation Time

The incubation time required for a chick to hatch is averaged 504 hours with variations between 480 and 510 hours (Almeida, Vieira, Gallo, Conde, and Olmos, 2006; Kita, Ito, Sugahara, Kobayashi, Makino, Takahashi, Nakahara, Takahashi, & Nishimukai 2015). This value can be influenced by various factors such as breeder's age, storage factor, incubation temperature, egg weight, and poultry type (Wilson, 1991). Table 1 indicates that the time required by the embryo to develop in the injected eggs during the incubation period tends to be longer than in the control eggs, although statistically, these differences were negligible. The longer incubation time condition indicated that the developed embryos in the eggs from the injected group was better than embryo from the control group. The phenomenon of the long incubation time of larger

embryo has been shown earlier by some researchers (Almeida, Vieira, Reis, Berres, Barros, Ferreira, & Furtado, 2008; Shafey, Mahmoud, Alsobaye, & Abouheif, 2014).

Among the injected group, there were no differences observed in incubation time. Based on this result, it is known that the injection of the eggs in the early incubation period contributes relatively low to the length of incubation. Shafey et al. also reported similar results., (2013), who injects breeder broiler eggs with glutamine on day 16 incubation. However, another report suggests that the incubation process becomes shorter when supplemented glutamine amino acids on the day 16th of incubation (Pedroso, Chaves, de Almeida Martinez Lopes, Leandro, Cafe, & Stringhini, 2006).

3. Hatching Weight

In addition to hatchability parameter, the chick weight at hatch becomes an essential parameter in the effort to improve the performance of native chicken. The increase in initial weight will have an impact on achieving the ideal market weight economically, which is performed in a short time compared to the chicken that hatches with a lighter weight.

Table 1 indicated that the absolute chick weight of the injection of glutamine on different incubation days tends to be higher than the control group (not injected). The injection on the 11th day showed a significant increase of 12.31% over control, while the injection on the day 7th and 9th were respectively 9.9 and 6.52% heavier than the control. The relative chick weight data also shows similar results. Although not significant, chicks in the injected group have relative chick weight higher than the chicks in the control group.

This result is still consistent with the report of Salmanzadeh et al. (2016) which showed that chicks hatched in the treatment of glutamine administration in the early incubation period were markedly heavier than chicks in the control group. The study also showed that in-ovo administration of glutamine in albumen could be a useful tool for increasing the weight of chickens not only at the time of hatching but also in the growth period. Another report (Chen, Wang, Wan, Xiong, Peng, & Peng, 2009), suggests that administering the glutamine does not affect the body weight of the newly hatch ducks, but improves the development of the intestine and body weight in the growing period.

D. Conclusion

The study showed that the injection of glutamine to obtain the more massive chicks at hatch, can be conducted on the day 7th, 9th and 11th of incubation, even with lower hatchability. Further studies are still needed, primarily to determine the long-term effects of differences in the time of *in ovo* administration of glutamine to native chickens.

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