

## IN OVO ADMINISTRATION OF BITTER GOURD (*MOMORDICA CHARANTIA*) LEAF EXTRACT IMPROVED THE HATCHING PERFORMANCE OF BROILER CHICKS

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### ABSTRACT

*In ovo* administration of substances with potent bioactivities is a novel approach to improving poultry welfare and performance. This study focused on assessing the effect of *in ovo* injection of *Momordica charantia* leaf extract (MLE) into fertile eggs on the hatching process and physiology of broiler chicks. A total of 536 fertile eggs were randomly distributed into 4 groups of 4 replicates on the 15<sup>th</sup> d of incubation. The eggs were then injected with 0.5 µg/ml MLE, 2.5 µg/ml MLE, and 12.5 µg/ml MLE in the air space. The noninjected group served as the control. At the end of the hatching period, each group was assessed for embryo mortality, hatchability, hatching duration, and serum antioxidant activity. The results showed that *in ovo* injection of MLE significantly increased ( $P < 0.05$ ) hatchability, reduced embryo mortality. Serum antioxidant levels were significantly ( $P < 0.05$ ) higher in the extract-injected groups. Moreover, total incubation and hatch time were significantly lower ( $P < 0.05$ ) in the MLE-injected group compared to the control group. Overall, *in ovo* administration of *momordica charantia* leaf extract improved the hatching process and chick's welfare with the implication of improving performance.

**Keywords:** Antioxidant, Broiler chicken, *In ovo*, Hatching, leaf extract, *Momordica*

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### INTRODUCTION

The early developmental phase of broiler chickens is a critical determinant of their overall health, productivity, and post-hatch performance. Enhancing embryonic development through *in ovo* supplementation has gained attention as an effective strategy to improve hatchability and physiological resilience (Ferket & Uni, 2006). *In ovo* administration allows direct delivery of bioactive compounds to the embryo, facilitating nutrient absorption and providing an early metabolic advantage (Bhanja *et al.*, 2012). This technique has been applied with various nutrients, including amino acids, vitamins, probiotics, and plant-derived phytochemicals, to optimize growth and immune function in poultry (Tako *et al.*, 2004; Jha *et al.*, 2019).

Bitter gourd (*Momordica charantia*), a medicinal plant known for its pharmacological properties, is a candidate for *in ovo* application due to its diverse bioactive constituents. It is rich in flavonoids, tannins, terpenoids, anthraquinones, saponins, sterols, phlobatanins, glycosides, amino acids, carbohydrates, resins, fatty acids, and phenols, which exhibit antioxidant, anti-inflammatory, and immunomodulatory effects (Sathya *et al.*, 2012; Valyaie *et al.*, 2021). These phytochemicals are known to enhance metabolic efficiency, reduce oxidative stress, and modulate immune responses in various animal models (Grover & Yadav, 2004; Ahmed *et al.*, 2020). In poultry, dietary supplementation with *M. charantia* extracts has been associated with improved growth performance, feed efficiency, and disease resistance (Muhammad *et al.*, 2019). However, the potential benefits of its administration during embryogenesis remain largely unexplored.

The objectives of this study were to evaluate the effects of *in ovo* administration of *M. charantia* leaf extract on hatching and physiological performance of broiler chicks. We hypothesized that the bioactive compounds in bitter gourd would enhance embryonic survival and improve stress tolerance by enhancing antioxidant defense mechanisms. The findings from this study could provide insights into the application of phytochemicals in poultry production, contributing to sustainable, antibiotic-free, and environmentally friendly rearing practices.

### MATERIALS AND METHODS

Fresh leaves of *Momordica charantia* were collected and air-dried at 18°C, after which they were crushed into powder using a mortar and pestle. The leaf powder was macerated in ethanol (80%) and extracted (Kuka *et al.*, 2023). A quantity of 100 mg of *Momordica* leaf extract was weighed into a 100 ml volumetric flask and saline solution (0.9% NaCl) was added to the mark to produce 1000 µg/ml stock solution after homogenization. One ml of the stock solution was further diluted to obtain 0.5 µg, 2.5 µg, and 12.5 µg/ml respectively for injection (Kuka *et al.*, 2024).

A total of 560 hatching eggs from Abor Acres, aged 59 weeks, were weighed, coded for easy monitoring, and then incubated in a PasReform SmartPro-Combi TM incubator. The incubation temperature and relative humidity were set at 37.7°C and 60% during incubation. Eggs were candled on day 15 and randomly grouped into four experimental groups of 134 eggs each. Eggs from groups one to three were disinfected at the broad end and perforated using a 21g needle. A total of 0.2 ml of the various leaf extract concentrations were injected into the air space using an automatic syringe with a 22 g needle, and the holes were sealed with adhesive tape. All the eggs

were transferred into the hatcher in 4 replicates and monitored for hatching activities. The hatcher parameters were set at 37.5°C and 70% relative humidity.

Data was collected on internal pipping (IP), external pipping (EP), and total incubation duration. At the end of the hatching, two chicks per replicate were weighed and sacrificed for blood collection. Blood samples were collected early in the morning into dry tubes for serum antioxidant assays using the ferric reducing antioxidant power (FRAP) test. Unhatched eggs were broken for observation and determination of embryo mortality and hatchability.

$$\text{Hatchability} = \frac{\text{Number of hatched chicks}}{\text{Number of fertile eggs}} \times 100$$

$$\text{Rate of malposition} = \frac{\text{Number of malpositioned embryo}}{\text{Number of fertile eggs}} \times 100$$

$$\text{percentage mortality} = \frac{\text{Number of dead embryo}}{\text{Number of fertile eggs}} \times 100$$

#### Statistical analysis

The data collected were subjected to one-way analysis of variance in R console software. treatment means were compared using the Duncan multiple range test at 0.05 threshold. The results were presented as average and standard error of the mean. The graph and figures were created in GraphPad Prism 8.0.

#### RESULTS

The effect of *Momordica charantia* on internal pipping, external pipping, total incubation duration, hatchability, and mortality of broiler chicks presents significant differences among the groups (Table 1). Group 4 (12.5µg) recorded the shortest ( $p < 0.05$ ) durations compared to the other groups. Similarly, embryonic mortality was less with higher hatchability in the same group. Therefore, *In ovo* administration of momordica leaf extract significantly reduced the incubation duration and embryonic mortality.

**Table 1: Effect of *Momordica charantia* on incubation and hatching of broiler chicks**

Settings	Treatments (Mormodica Leaf Extract) µg/ml				P-value
	0 µg	0.5 µg	2.5 µg	12.5 µg	
EP duration	10.19 ± 0.43 <sup>b</sup>	11.77 ± 0.72 <sup>a</sup>	11.76 ± 0.54 <sup>a</sup>	9.77 ± 0.53 <sup>b</sup>	0.014
H duration	10.89 ± 0.43 <sup>a</sup>	11.53 ± 0.43 <sup>a</sup>	9.472 ± 0.47 <sup>b</sup>	8.35 ± 0.50 <sup>c</sup>	<0.001
TI duration	480.10 ± 0.50 <sup>b</sup>	481.70 ± 0.61 <sup>a</sup>	479.60 ± 0.43 <sup>b</sup>	478.40 ± 0.40 <sup>c</sup>	0.0001
Hatchability (%)	86.56 ± 3.94 <sup>ab</sup>	69.89 ± 12.54 <sup>b</sup>	97.73 ± 1.31 <sup>a</sup>	93.25 ± 2.30 <sup>a</sup>	0.031
Early mortality (%)	2.20 ± 1.25	1.08 ± 10.30	3.72 ± 1.99	2.27±1.31	0.133
Late mortality (%)	11.23 ± 4.76 <sup>a</sup>	12.02 ± 2.82 <sup>a</sup>	3.03± 1.51 <sup>b</sup>	0.75 ± 0.75 <sup>c</sup>	0.024
Malposition (%)	1.51 ± 0.75 <sup>b</sup>	3.755 ± 1.532 <sup>a</sup>	0.75 ± 0.75 <sup>b</sup>	0.00 ± 0.000 <sup>c</sup>	0.054

<sup>a,b,c</sup> values with different superscripts within a row differ significantly at 0.05

EP: External pipping, H: hatch duration, TI: Total incubation

The effect of *Momordica charantia* on serum antioxidant capacity of broiler chicks at hatch revealed significant differences among the treatment groups. Momordica leaf extract at 2.5 and 12.5 µg significantly ( $p < 0.001$ ) increased the serum antioxidant capacity compared to the control group.

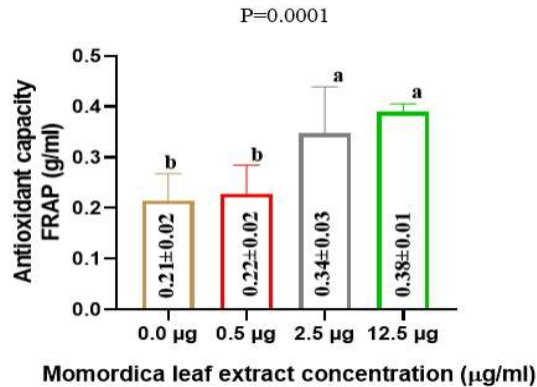


Figure 3: Effect of *in ovo* *Momordica charantia* leaf extract on serum antioxidant capacity of chicks at hatch

## DISCUSSION

*In ovo* administration of *Momordica charantia* leaf extract into the airspace of broiler chicken eggs significantly affected the incubation process, hatchability, and embryo mortality. This can be explained by the antioxidant and antibiotic properties of *Momordica* leaves in combating thermal and oxidative stress in the embryo at later stages of development, which weakens the chicks and leads to prolonged hatching. The decrease in incubation duration and hatching aligns with the findings of Bilalissi *et al.* (2019) and Kuka *et al.* (2024), who reported that injecting Moringa and soursop leaf extracts into the air chamber of incubated eggs reduced the incubation duration. This reduction is economically beneficial to farmers, as it lowers production costs by reducing energy consumption and wear and tear on machines. Furthermore, the reduced time will diminish the holding period of chicks at the hatchery and improve their welfare. Decuypere and Willemsen (2011) attributed the reduced incubation period to low partial pressure of CO<sub>2</sub> in the air chamber of eggs. The injected extract may have stimulated the production of corticosterone, which could increase the production of thyroid hormones that play a crucial role in the hatching process (Decuypere *et al.*, 2006). Antioxidants protect the body against chronic diseases and damage by reducing stress and neutralizing the harmful effects of free radicals and reactive oxygen species (ROS) in cells (Abuajah *et al.* 2015). The increased serum antioxidant activity in this study can be traced to the phytochemical composition of *Momordica charantia*, (Oliveira *et al.* (2018). This result supports the observed effect of *Momordica* leaf extract on the hatching performance of broiler chicks. The improved serum antioxidant activity correlates with reduced oxidative stress, enhanced immune function, and improved growth performance in the chickens. This implies that the *in ovo* administration of *Momordica* leaf extract would lead to decreased stress and mortality in embryos, resulting in improved hatching performance and physiology of broiler chicks.

## CONCLUSION

*In ovo* administration of *Momordica charantia* extract on the 15<sup>th</sup> day of incubation into the air space of broiler chicken eggs reduced the total incubation, reduced embryonic mortality, improved hatchability and serum antioxidant status of the chicks.

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