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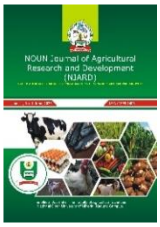

Original Article

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Investigating the Impact of Breed on Selected Fear Behaviors in four Meat type Chicken

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ABSTRACT

Fear is a critical welfare concern in poultry, often linked to reduced productivity. While growth and feed efficiency traits have been extensively studied in commercial broiler breeds, limited information exists on breed-specific differences in fear-related behaviors. This study evaluated fear responses in four broiler breeds using the tonic immobility (TI), inversion, and emergence tests at weeks 1, 2, and 3 of age. A total of 100 chicks (25 per breed), all from a common genetic background, were assessed. Results showed that emergence duration significantly differed by breed at all three time points, with Cobb 500 and Marshall generally emerging faster. Tonic immobility duration varied significantly by breed only at week 1, with Marshall exhibiting the longest duration. Inversion test results revealed significant breed effects across all weeks, with Marshall birds consistently showing the highest number of wing flaps, a strong fear indicator. Overall, Marshall chickens displayed the most pronounced fear responses across multiple tests and time points, indicating higher baseline fearfulness compared to other breeds. This study investigated breed-specific differences in fear behaviors among four commercial broiler breeds—Arbor Acres, Marshall, Ross 308, and Cobb 500—at weeks 1 to 3 of age. Using tonic immobility, inversion, and emergence tests, significant differences were observed in behavioral responses across breeds and ages. Marshall chickens exhibited the highest fear responses, as indicated by prolonged tonic immobility and increased wing flapping. The findings underscore the need to consider behavioral traits in broiler breeding programs to enhance welfare outcomes

Keywords: *Emergence; tonic immobility; inversion; meat-type chicken*

1.0 INTRODUCTION

In recent years, research on meat-type chickens has primarily focused on improving growth performance and feed efficiency. While these traits are vital for optimizing meat production, behavioral traits such as fear responses have received comparatively less attention. Yet, fear is a critical welfare concern, as excessive fear can reduce productivity, compromise immune function, and increase susceptibility to stress (Durosaro *et al*., 2023).

Fear in farm animals is an affective state often triggered by stressors such as novel environments, predators, handling, or social isolation (Grigor, 1993). These responses are expressed through various behaviors including immobility, flight, or struggle, which serve as survival mechanisms (Jones, 1986). In poultry, passive avoidance behaviors like tonic immobility and active responses like wing flapping or escape attempts are commonly observed indicators of fear (Miller *et al*., 2006).

Although environmental stimuli contribute to fear reactions, genetic predisposition also plays a significant role. Studies have shown that fear-related traits in poultry are moderately heritable (heritability estimates ranging from 0.07 to 0.49), making them amenable to selection (Grams *et al*., 2015). Genetic selection has already been used to reduce fearfulness in various livestock species, including poultry (Boissy *et al*., 2005; Agnvall *et al*., 2012). This supports the potential to improve behavioral resilience through breeding programs (Lord *et al*., 2020).

Several standardized tests are used to evaluate fear in poultry, including the tonic immobility test (for passive fear), emergence test (for neophobia), and inversion or wing flapping test (for struggle response) (Forkman *et al*., 2007). Despite their widespread application, these tests have rarely been used to compare fear responses across commercial broiler breeds.

Ross 308, Arbor Acres, Marshall, and Cobb 500 are among the most commonly used commercial broiler breeds globally, prized for their rapid growth and feed conversion efficiency (Mahmoud *et al*., 2011; Jeremiah *et al*., 2024). However, limited research has addressed whether these breeds differ in their fear-related behaviors. This represents a notable gap, particularly given that excessive fear during routine practices such as handling, transport, or vaccination can increase mortality and welfare concerns.

This study investigates breed-specific fear responses in Ross 308, Arbor Acres, Marshall, and Cobb 500 broilers at 1, 2, and 3 weeks of age using three validated behavioral tests: the emergence test, tonic immobility test, and inversion (wing flapping) test. These early developmental stages are critical windows for behavioral assessment and intervention. Although growth and feed efficiency have been extensively studied in broilers, fear-related behaviors—which impact both welfare and productivity—are understudied. This study hypothesizes that breed-specific differences exist in fear responses during early developmental stages in broiler chickens.

2.0 MATERIALS AND METHODS

2.1 Study site

The Experiment was carried out at the Teaching and Research Farm, Federal College of Animal Health and production Technology, Moor Plantation, Apata, Ibadan. It experiences a temperature and humidity ranging from 35 – 40 °C and 76 – 78% respectively.

2.2 Experimental birds

One hundred broiler chickens of four different breeds were used in this study, and they were kept under an intensive system: each pen was fenced with wire gauze, and dry wood shavings were used as litter material, spread on the floor of each individual pen; the birds were given feeders, drinkers, clean water, and plenty of food; the litter was changed frequently to prevent the growth of microorganisms; each bird was tagged on its wing to make identification easier during the study; and strict sanitation protocols and immunization programs were put in place to prevent disease.

2.3 Housing

In this study, one hundred broiler chickens were used. The 25 Marshal, 25 Cobb 500, 25 Ross 308, and 25 Arbor acres chickens made up the group. From the first day until they were ten weeks old, these birds were raised under conditions of strict management. Every hen came from the same genetic family and was purchased from a respectable hatchery in Ibadan, Oyo State, Nigeria. On the day of hatching, all of the chicks were taken to the brooding pen after being hatched simultaneously. When they arrived, they were kept in the same brooding pen, which was 3.6 m by 3.6 m and had 200 Watt heating bulbs.

Five centimeters of wood shavings were spread out on the brooding pen's floor as litter substrate. The birds were kept in the brooding pen with a lighting schedule of 12 hours of light and 12 hours of darkness during the trial period. For days 1–7, the brooding temperature was kept at 34°C; for days 8–14, it was kept at 30°C; and for days 15–16, it was kept at 28°C. The birds were given a chicken starting meal in plastic trays with a diameter of 40 cm. It contained 28% crude protein and 2860 kcal/MEkg of metabolizable energy. Bell drinkers (25 cm in diameter and 30 cm in height) were used to supply clean water on demand. To stop illness outbreaks, strict adherence to a vaccination schedule and careful cleaning procedures were noted.

The birds were only exposed to brooding, feeding, water provision, immunization, medicine, and experimental handling during the experiment. The duration of the trial was six weeks.

2.4 Behavioural tests

A soundproof separate chamber (3.6 m × 3.6 m) next to the brooding pen was used for all of the terror tests. The person who brought the hens from the brooding pen, the person who conducted the fear test, and the timekeeper all participated in the fear testing. All 100 chickens were used in the fear tests at random, with no preference for a specific breed. One bird at a time was used for the terror test.

2.4.1 Emergence

When the birds were one, two, and three weeks old, the emergence test was carried out. Every bird was put into a dark box that was 48 cm by 32 cm by 26 cm, with an exit aperture that was 18 cm by 15 cm, through an inlet opening that was 10 cm by 10 cm. Each bird's exit time from the box was measured in seconds, with a 300-second maximum duration limit.

2.4.2 Tonic immobility

The birds were placed in tonic immobility at the ages of one, two, and three weeks. Using one hand on the chest and the other on the head, each bird was placed gently on its back and gently pressed for ten seconds. The researcher then removed their hands and stepped back, causing no more harm to the bird. Either 300 seconds of immobility or the bird's spontaneous righting after at least 10 seconds of inactivity marked the end of the test. Each bird's tonic immobility duration was carefully recorded in seconds, and every individual bird was able to achieve tonic immobility.

2.4.3. Inversion
At one, two, and three weeks of age, the birds underwent an inversion test. For a maximum of thirty seconds, each bird was inverted by holding its two legs together. Wing flapping time during inversion was measured in seconds.

2.5 Statistical Analysis
Shapiro-Wilks tests were used to check for normality in all of the data. Non-parametric tests (Kruskal-Wallis) were used to analyze the effects of breed on behavioral fear reactions because the data will not be regularly distributed, even after transformation. SAS v9 (2002) was used to perform the Shapiro-Wilks and Kruskal-Wallis tests using PROC UNIVATIATE and PROC NPAR1WAY, respectively.

3.0 RESULTS
The effect of breed on latency to emerge, duration of tonic immobility and duration of wing flapping during inversion test at week 1 is shown in Figure 1. Breed had a significant (P<0.05) effect on latency to emerge from a dark box, duration of tonic immobility and number of wing flapping. The shortest latency to emerge was observed in Marshall, Arbor Acres and Cobb 500 (Figure 1), while the longest latency to emerge was observed in Ross 308. The longest duration of tonic immobility was observed in Marshall (Figure 1), but there was no significant difference among Arbor Acres, Cobb 500 and Ross 308. The highest number of wing flapping was observed in Ross 308 and Marshall, while there was no significant difference between Cobb 500 and Arbor Acres (Figure 1).

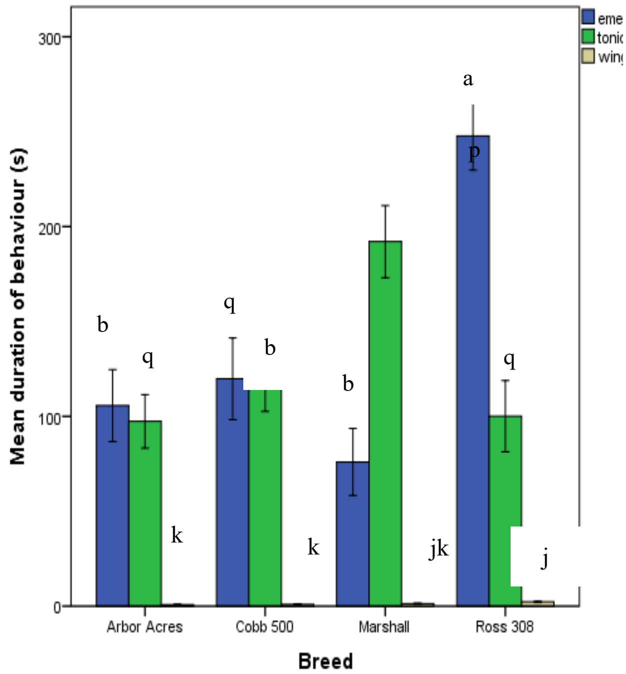

Figure 1. Effect of breed on latency to emerge, duration of tonic immobility and duration of wing flapping during inversion test at week 1. [ab,pq,jk] means with different letters are significantly (p < 0.05) different.

The effect of breed on latency to emerge, duration of tonic immobility and duration of wing flapping during inversion test at week 2 is presented in Figure 2. Breed had a significant (P<0.05) effect on latency to emerge from a dark box and number of wing flapping during. For latency to emerge, there was no significant difference among the breeds (Figure 2). The highest number of wing flapping was observed in Marshall, but there was no significant difference between Ross 308, Cobb 500 and Arbor Acres (Figure 2). However There was no breed significant (P>0.05) effect on duration of tonic immobility (Figure 2).

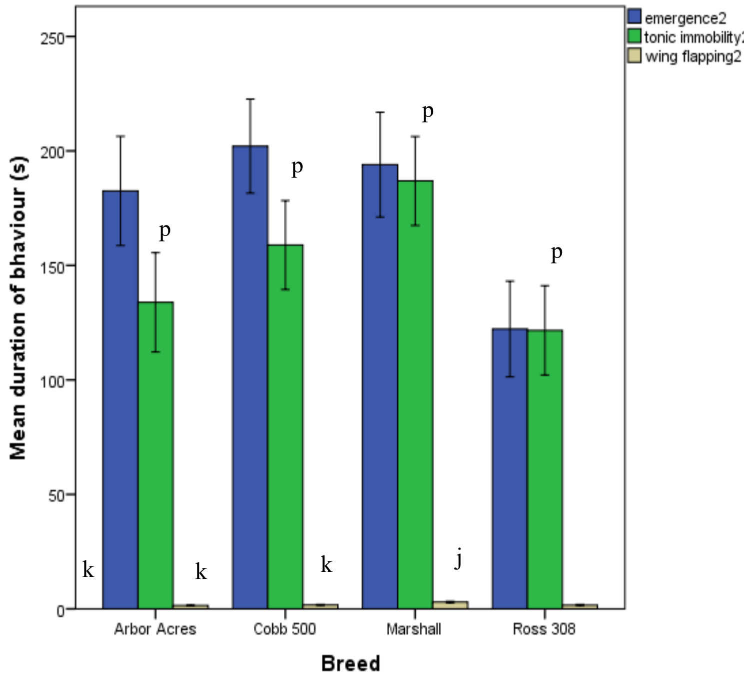

Figure 2: Effect of breed on latency to emerge, duration of tonic immobility and duration of wing flapping during inversion test at week 2. [pq, jk] means with different letters are significantly (p < 0.05) different.

The effect of breed on latency to emerge, duration of tonic immobility and duration of wing flapping during inversion test at week 3 is shown in Figure 3. Breed had a significant (P<0.05) effect on latency to emerge and number of wing flapping. The shortest latency to emerge was observed in Cobb 500 (Figure 3), but there was no significant differences among Marshall, Arbor Acres and Ross 308. The highest number of wing flapping was observed in Marshall, followed by Arbor Acres, while there was no significant difference between Cobb 500 and Ross 308 (Figure 3). However, there was no breed significant (P>0.05) effect on duration of tonic immobility.

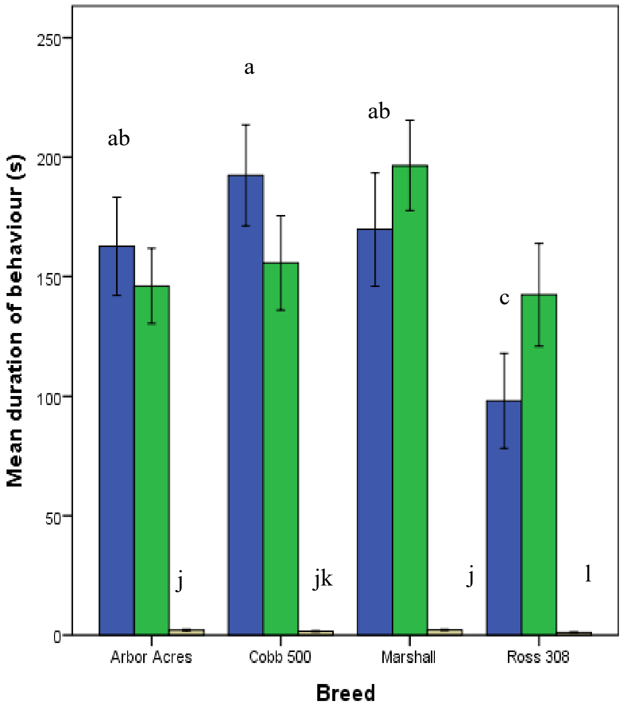

Figure 3: Effect of breed on latency to emerge, duration of tonic immobility and duration of wing flapping during inversion test at week 3. [abc, jkl] means with different letters are significantly ($p < 0.05$) different.

4.0 DISCUSSION

All behavioral data were initially assessed for normality using the Shapiro-Wilks test. As the data were not normally distributed, even after transformation, non-parametric Kruskal-Wallis tests were used to evaluate the effect of breed on fear-related behaviors. Analyses were conducted using SAS v9.0 (2002), employing the PROC UNIVARIATE and PROC NPAR1WAY procedures.

Fear responses in poultry have practical implications for welfare and productivity. Excessive fear has been associated with reduced feed intake, lower body weight, and heightened stress (de Haas *et al*., 2013). Although several studies have explored various behaviors in poultry (Durosaro *et al*., 2021; Durosaro *et al*., 2023), research on early-life fear responses in broilers remains limited.

In the current study, latency to emerge from a dark box significantly varied by breed across all three weeks. At week 1, Marshall, Arbor Acres, and Cobb 500 exhibited the shortest emergence times, while Ross 308 had the longest. This suggests that Ross 308 birds may display higher fear levels or adopt a more reactive coping strategy, consistent with prior findings that longer emergence latency reflects heightened fearfulness (Durosaro *et al*., 2021; Jeremiah *et al*., 2024). Conversely, the shorter latency in the other breeds may indicate greater boldness or exploratory motivation. At week 2, although the Kruskal-Wallis test detected a significant breed effect, post-hoc comparisons revealed no significant pairwise differences. This may suggest subtle behavioral trends that did not reach individual statistical thresholds or indicate that fear responses began to converge across genotypes with age. Factors such as increasing environmental familiarity and habituation to handling may have contributed to this convergence (Campbell *et al*., 2016; Jones, 1996). By week 3, Cobb 500 birds continued to display the shortest emergence times, suggesting a possible proactive coping style. The similarity among Marshall, Arbor Acres, and Ross 308 could be due to age-related behavioral modulation or decreased novelty of the testing environment (Zidar *et al*., 2017). A significant breed effect on tonic immobility (TI) duration was observed only at week 1, with Marshall birds showing the longest durations. This suggests greater inherent fearfulness or a passive coping style in this breed. However, no significant differences were found among Arbor Acres, Cobb 500, and Ross 308, possibly reflecting similar behavioral selection histories. At weeks 2 and 3, no significant breed effects were detected for TI duration. This decline in breed-related variability may indicate habituation to handling or environmental conditions, leading to a more uniform fear response over time (Jones, 1996). It also supports the notion that TI may be less sensitive to detecting subtle breed differences in older chicks, as fear responses stabilize with age and exposure (Hemsworth and Barnett, 2000). These findings contrast with reports from native or less-selected breeds, where clear breed-based differences in TI duration have been noted (Yoshidome *et al*., 2021). The lack of significant variation among commercial broiler strains may be attributed to intensive selection for growth traits, which could inadvertently reduce behavioral diversity (Hemsworth and Coleman, 2011).

In the present study, breed significantly influenced the number of wing flaps during the inversion test across all three weeks. At week 1, Ross 308 and Marshall showed the highest wing flapping frequencies, which are typically interpreted as escape or agitation responses to restraint (Kunzmann *et al*., 2020). Cobb 500 and Arbor Acres displayed significantly lower frequencies, indicating reduced reactivity. By week 2, Marshall birds again exhibited the highest wing flapping, followed by Arbor Acres, suggesting a persistent heightened sensitivity to restraint. Ross 308, although reactive in week 1, did not significantly differ from Cobb 500 and Arbor Acres at this stage, possibly due to environmental habituation or individual variation in stress thresholds. At week 3, Marshall continued to exhibit the highest flapping activity, reinforcing its consistent reactivity. Arbor Acres followed closely, while Cobb 500 and Ross 308 showed comparable and lower responses. These patterns may reflect underlying temperament differences, possibly shaped by genetic factors or past selection for traits indirectly linked to behavior (Durosaro *et al*., 2021).

Although this study revealed significant breed differences in fear-related behaviors, several limitations should be considered. The relatively small sample size (25 birds per breed) may limit generalizability, and the short observation window (weeks 1–3) restricts insights into long-term behavioral patterns. Additionally, all birds were reared under similar environmental conditions, which may not reflect the full spectrum of commercial production settings. Despite these limitations, the findings highlight meaningful behavioral variation among broiler breeds. Marshall birds consistently showed heightened fear responses across multiple tests, suggesting the need for targeted welfare interventions. Conversely, the reduced reactivity in Cobb 500 may be advantageous in systems where birds experience frequent human handling. Understanding breed-specific behavioral profiles can guide welfare-friendly breeding programs and inform management practices such as handling, transport, and housing design. Future studies should incorporate larger, more diverse populations and consider longitudinal behavioral assessments to track fear response stability over time.

CONCLUSION

The findings from this study demonstrated that fear-related behaviors in broiler chickens are influenced by both genetic strain and developmental stage. Among the four commercial breeds evaluated, Marshall consistently exhibited the highest indicators of fearfulness, characterized by prolonged tonic immobility and a high frequency of wing flapping during inversion, indicating a more reactive behavioral profile. In contrast, Cobb 500 and Arbor Acres showed comparatively lower fear responses. The variation observed across different weeks underscores the importance of age-specific assessments in understanding the development of fear behaviors in broilers. These behavioral differences are likely rooted in genetic selection histories and highlight the potential for targeted improvement. These findings suggest that incorporating behavioral traits such as fear responsiveness into broiler breeding objectives could enhance animal welfare under intensive production systems. Selection for calmer, less fearful birds may reduce stress-related losses and improve management outcomes during handling, transport, and other routine farm practices. Consequently, this research supports the integration of behavioral assessments into welfare-focused breeding and management strategies for sustainable poultry production.

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