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Research Article
Effect of Delayed Access to Feed on Physiological, Morphometric and Intestinal Parameters of Guinea Fowl Broiler

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Abstract

Background and Objective: After hatching, day-old chicks usually have delay in feed access for 48-72 h before they are placed on farms. This study was conducted to evaluate the effect of delayed access to feed on keets post-hatch performance, physiological and intestinal morphometric parameters. **Materials and Methods:** Four hundred day-old keets were distributed in a completely randomized design consisting of 2 treatments and 4 replicates with 50 birds per replicate. The treatments were: (1) Keets with immediate access to feed and (2) Keets with delay in feed access for 48h. Prior to feed access, the keets were weighed and reared for 11 weeks. Data were collected on feed intake, body weight. Also, blood samples were collected at week 11 from guinea fowls for determination of triiodothyronine, thyroxine, total cholesterol, triglyceride, glucose and total protein levels. Guinea fowls were later slaughtered and intestinal morphometry were determined. **Results:** From 3 weeks onward, body weights of guinea fowls with immediate access to feed were higher ($p<0.05$) than those with delayed access to feed. Also, the weight and length of intestine of birds with immediate access to feed was higher than those with delayed access to feed. Triiodothyronine and thyroxine of guinea fowl with immediate access to feed were higher ($p<0.05$) than those with delayed access to feed. Total cholesterol, total protein and glucose of the birds with delayed access to feed were lower ($p<0.05$) than those with immediate access to feed. **Conclusion:** Delay in feed access more than 48h adversely affected growth performance of guinea fowl.

Keys words: Guinea fowl, feed access, intestinal morphometric, thyroid hormone, biochemical parameters

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

The post-hatch growth of newly hatched chicks can be influenced by several factors including hatching egg characteristics, incubation conditions and chick holding conditions before their placement at broiler farm[1]. Indeed, in a normal condition, day-old chicks are deprived of feed for about 48 h before they are placed on farms. Potturi *et al.*[2] reported that under practical conditions, poults often gain access to feed only 36-48 h after hatching due to variations in hatching time, hatchling servicing and delivery to the farm. Moreover, before and immediately after hatch, chicks are dependent on the lipids and proteins of the egg yolk as main sources of nutrients. However, egg yolk is usually depleted within 5-6 day posthatch[3]. Therefore, early supply of feed is essential for maintenance, growth and production of chick[4,5]. Moreover, the physical and functional development of the gastrointestinal tract is necessary to enable newly hatched birds efficiently utilize nutrients supplied in the diet[3]. Thus, early access to feed and water after hatch has been shown to be an important factor for subsequent performance[6]. It has been reported that chicks deprived of feed and water for 48 h lost 10.7% of their initial post hatch weights[7] prior to placement. Thus, it has been observed that delayed access to feed has a negative impact on gastrointestinal development[8-12] retarded growth performance and reduced body weight and breast muscle[13-15]. Additionally, studies have shown that delay in feed access affected tri-iodothyronine (T_3) and thyroxine (T_4) concentrations and biochemical parameters. Newcombe *et al.*[16] and Kitaysky *et al.*[17] indicated that the levels of triiodothyronine (T_3) decreased in feed-restricted broilers, suggesting a lower metabolic rate during restriction. This observation indicates that post hatch developmental stage is significantly influenced by delayed feed access. To our knowledge, most studies have focused on effects of delayed feed access on chickens while guinea fowl has been given very little attention. It can be hypothesized that delay in feed access may be detrimental for post-hatch performance and physiological parameters of day-old guinea fowl. Therefore, this study aimed to investigate the effects of early and late access to feed on growth performance, intestinal morphometry, levels of T_3, T_4 and biochemical parameters of guinea fowl.

MATERIALS AND METHODS

Experimental design: This study used four hundred (400) day-old keets. The keets were purchased from reliable hatchery and were randomly assigned to two treatment groups as follows: (1) Keets with immediate feed access and (2) Keets with 48 h delayed feed access. For each treatment group, keets were assigned to 4 replicates with 50 birds in each replicate. Keets with delayed access to feed for 48 h, were held in shipping boxes at room temperatures ranged from 25-27°C until 48 h. Prior to feed access, the keets were weighed. Birds were reared for 11 weeks, including starter (0-4 weeks), grower (4-8 weeks) and finisher (8-11 weeks) stages. The replicates were randomly distributed in an open-sided poultry house in order to avoid positional effects. They were raised on a floor litter with a stocking density of 20 birds per m^2 and photoperiod of 23 h of light during the first 4 weeks of age. But, from 5 weeks of age, the stocking density was 10 birds m^{-2} and light was reduced gradually to natural photoperiod of 16L/8D at the end of week 6 of age. Feed and water were supplied *ad libitum*. Body weights and feed intake were recorded weekly. Mortality was recorded according to treatment during the 11 weeks of rearing. At 11 wks of age, 20 guinea fowls (5 birds/replicate) per treatment were selected at random and slaughtered to determine carcass, gizzard, heart, liver and abdominal fat weights. Blood samples were collected for plasma T_3, T_4, cholesterol, triglyceride, glucose, total protein, urea and creatinine determination. Also, intestinal morphometric parameters were determined.

Body weights, weight gains, feed intake and FCR: The keets were weighed at hatch according to treatment and then weekly until 11 weeks of age. Feed intake was recorded daily and was used to calculate average feed intake per week per bird. Body weights and feed intake were used to calculate average body weight gain and feed conversion ratio.

Carcass and organ weights: At 11 weeks of age, 20 birds per treatment were slaughtered to determine the weights of carcass, heart, liver, gizzard and abdominal fat. These weights were used to determine the relative weights of carcass yield and the organs as:

$$\text{Carcass yield / organ} = 100 \times \frac{\text{Carcass weight or organ weight}}{\text{Body weight}}$$

Intestinal morphometry: The small intestine with the pancreas was carefully removed from the slaughtered birds and suspended vertically to apply uniform tension for length measurements. For length and weight measurements, the small intestine was divided into three segments: duodenum

(from gizzard to entry of the bile and pancreatic ducts), jejunum (from entry of the ducts to yolk stalk) and ileum (from yolk stalk to ileocecal junction). The pancreas was removed and weighed.

Serum tri-iodothyronine (T_3), thyroxine (T_4) and biochemical determination: Blood samples were collected from 20 guinea fowls at slaughter per treatment (5 birds/replication) for determination of T_3, T_4, total cholesterol, triglyceride, glucose, total protein, creatinine and urea levels. The blood samples were centrifuged at 3,000 rpm for 15 min and the obtained serum was stored in a freezer at -20°C. Afterwards, a volume of 100 µL of serum was used for T_4 and T_3 concentrations determination by using the automated VIDAS systems, which is an enzyme-linked fluorescent assay (ELFA) technique. Antibody anti-T_3 of sheep, labelled with phosphatase alkaline and sodium azide, antibody anti-T_4 labelled with phosphatase alkaline provided by VIDAS were used, respectively for T_3, T_4 concentrations determination.

Only 2 µL serum was used for total cholesterol (CHOL2), triglyceride (TRIGL), glucose (GLUC2), total protein (TP2), creatinine (Crea2) and urea (Ureal) concentrations determination by using the automated COBAS systems, an enzymatic colorimetric techniques. ACN 8798 (CHOL2), ACN 8781 (TRIGL), ACN 8767 (GLUC2) and ACN 8678 (TP2), ACN 8452 (CREA2) and ACN 8418 (UREAL) were *in vitro* tests for quantitative determination of serum concentrations of cholesterol, triglyceride, glucose and total protein respectively. For each of the parameter, the samples were run in the same assay in order to avoid inter assay variability.

Statistical analysis: Statistical software package Graph Pad PRISM 5 was used to analyze the data. T-test was used to analyze body weight and physiological organs (gizzard, heart, liver, carcass, spleen, kidney and pancreas) weights of guinea fowls, thyroid hormone (T_3, T_4), biochemical parameters (total cholesterol, triglyceride, glucose, total protein, urea and creatinine) and intestinal morphometry. Chi square (X^2) was used to analyze mortality. A probability value of 0.05 was used as the degree of significance.

RESULTS

Effect of delayed feed access on keet weight loss and Post-hatch body weights: Figure 1 shows the effect of delayed feed access on the body weights of guinea fowl. Overall, body weight decreased with delay in feed access. Day-old keets deprived of feed for 48 h lost 2.86 g of weight.

Overall, body weights increased with the age. Body weights were not affected by delay in feed access during the first two weeks post-hatch. But, from 3 weeks onward, body weights of guinea fowl with immediate access to feed were higher ($p<0.05$) than those of guinea fowl with delayed access to feed.

Effect of delayed feed access on feed intake, average daily weight gain, FCR and mortality: Table 1 shows feed intake, average daily weight gain, feed conversion ratio and post-hatch mortality according to treatment. Feed intake increased with the age of the birds in all treatments. However, feed intake was not affected by delay in feed access. Average daily gain and FCR were comparable in all treatments but, post-hatch mortality of guinea fowls with delayed access to feed was higher ($p<0.05$) than those of guinea fowls without delay in feed access.

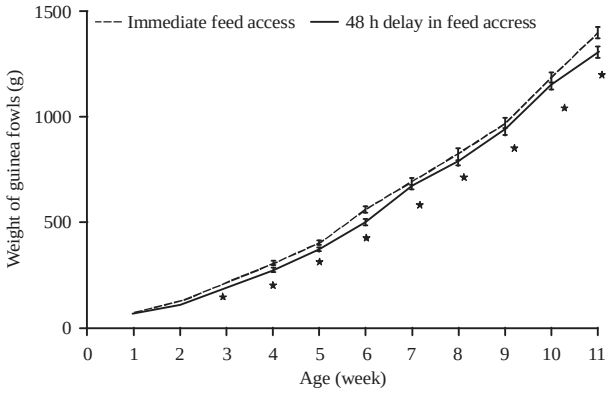

Fig. 1: Body weight according to treatment and age of guinea fowl. At each age, *indicates significant difference at $p<0.05$

Table 1: Feed intake, daily weight gain, feed conversion ratio and post-hatch mortality according to treatments

	Delay in feed access (h)	
Parameters	Immediate feed access	48 h delay in feed access
Feed intake (g day^{-1})	71.96±7,33	81.04±7,52
Average daily weight gain (g day^{-1})	17.36±1.45	16.85±1.62
Feed conversion ratio	3.17±0.51	3.55±0.59
Mortality (%)	3.9±0.01^{b}	4.23±0.61^{a}

^{a,b}Within row, data sharing different superscripts are significantly different ($p<0.05$). Mean±SEM

Effect of delayed feed access on organs' weights, live weights and carcass weights: Table 2 shows the effect of delayed feed access on relative weights of heart, liver, gizzard abdominal fat, spleen, kidney and pancreas of guinea fowl. The relative weights of heart, abdominal fat, spleen, kidney and pancreas were not affected by delayed feed access. However, the relative weights of liver and gizzard of birds subjected to immediate feed access were higher ($p<0.05$) than those of guinea fowl with delayed access to feed for 48 h.

Table 3 presents the live weights, carcass weights and carcass yield of guinea fowl affected by delayed feed access. Live weight, carcass weight and carcass yield of birds subjected to immediate feed access were higher ($p<0.05$) than those of guinea fowl with delayed access to feed for 48 h.

Table 2: Ratio of heart, liver, gizzard abdominal fat, spleen, kidney and pancreas at slaughter according to treatment

	Delay in feed access (h)	
Parameters	Immediate feed access	48 h delay in feed access
Heart	0.43±0.01	0.38±0.01
Liver	2.13±0.12[a]	1.70±0.07[b]
Gizzard	2.42±0.12[a]	2.01±0.07[b]
Abdominal fat	1.51±0.08	1.40±0.14
Spleen	0.10±0.01	0.08±0.01
Kidney	0.27±0.02	0.27±0.02
Pancreas	0.18±0.01	0.18±0.03

[a,b]Within row, data sharing different superscripts are significantly different ($p<0.05$). Mean±SEM

Table 3: Live weight, carcass weight and carcass yield at slaughter according to egg storage duration before incubation

	Delay in feed access (h)	
Parameters	Immediate feed access	48 h delay in feed access
Live weight (g)	1772.00±1.84[a]	1587.00±2.28[b]
Carcass weight (g)	1261.00±1.53[a]	1142.00±2.45[b]
Carcass yield (%)	72.25±0.44[a]	71.01±0.21[b]

[a,b]Within row, data sharing different superscripts are significantly different ($p<0.05$). Mean±SEM

Effect of delayed feed access on weight, length and weight/length of intestine, jejunum, ileum and duodenum: Effects of delayed feed access on intestinal morphometry of guinea fowl are presented in Table 4. Intestine and jejunum weights of the birds subjected to immediate feed access were higher ($p<0.05$) than those birds subjected to delay in feed access for 48 h. But duodenum and ileum weights were comparable in the birds subjected to immediate feed access and birds subjected to delay in feed access for 48 h. Also, the length of intestine, jejunum and ileum of the birds subjected to immediate feed access were higher ($p<0.05$) than those of birds subjected to delay in feed access for 48 h. Similar length of duodenum was recorded in the birds subjected to immediate feed access and birds subjected to delay in feed access for 48 h. The weight/length was not affected by the treatment at the different segments of the intestine.

Effect of delayed feed access on biochemical parameters and thyroid hormone: Table 5 shows the effects of delayed feed access on serum cholesterol, triglyceride, glucose, total protein, creatinine and urea. The total cholesterol, glucose and total protein were affected by delayed access to feed. Indeed, the concentration of total cholesterol, glucose and total protein of birds with immediate access to feed was higher ($p<0.05$) than those of the birds subjected to delayed access to feed. But, the levels of triglyceride, glucose, creatinine and urea were not affected by delayed access to feed.

Table 6 presents the effects of delayed access to feed on serum concentrations of T_3, T_4 and $T_3/_{T4}$. The T_3 and T_4 concentration of guinea fowls with immediate feed access was

Table 4: Weight, length and weight/length of intestine, duodenum, jejunum and ileum according to treatment

		Delay in feed access (h)	
Parameters		Immediate feed access	48 h delay in feed access
Weight (g kg^{-1} of bird)	Intestine	24.140±0.71[a]	21.830±0.61[b]
	Duodenum	6.070±0.32	5.990±0.18
	Jejunum	9.730±0.58[a]	9.320±0.53[b]
	Ileum	6.770±0.3	6.580±0.31
Length (cm kg^{-1} of bird)	Intestine	77.680±2.07[a]	70.140±1.75[b]
	Duodenum	12.290±0.45	12.280±0.43
	Jejunum	36.310±1.11[a]	31.660±0.94[b]
	Ileum	29.570±0.9[a]	24.680±1.64[b]
Weight/Length (g cm^{-1})	Intestine	0.310±0.01	0.310±0.01
	Duodenum	0.49±0.01	0.49±0.22
	Jejunum	0.26±0.14	0.29±0.01
	Ileum	0.220±0.013	0.260±0.01

[a,b]Within row, data sharing different superscripts are significantly different ($p<0.05$). Mean±SEM

Table 5: Levels of cholesterol (Chol2), triglyceride (Trigl), glucose (Gluc2) and total protein (TP2), creatinine (Crea2) and urea (Ureal) in blood of guinea fowl at 11 week age according to treatment

Biochemical parameters	Delay in feed access (h)	
	Immediate feed access	48 h delay in feed access
Chol (g L^{-1})	1.74±0.12[a]	1.27±0.08[b]
Trigl (g L^{-1})	0.75±0.11	0.66±0.09
Gluc (g L^{-1})	3.11±0.15[a]	2.63±0.24[b]
TP2 (g L^{-1})	27.35±0.83[a]	23.92±0.69[b]
Crea2 (g L^{-1})	0.06±0.04	0.06±0.05
Ureal (mg dL^{-1})	0.01±0.01	0.03±0.08

[a,b]Within row, data sharing different superscripts are significantly different ($p<0.05$). Mean±SEM

Table 6: Levels of T_3, T_4 and T_3/T_4 in blood of guinea fowl according to treatment

Hormone parameters (pmo L^{-1})	Delay in feed access (h)	
	Immediate feed access	48 h delay in feed access
T_3	4.50±0.32[a]	3.2±0.29[b]
T_4	7.71±0.4[a]	6.26±0.6[b]
T_3/T_4	0.61±0.06	0.58±0.06

[a,b]Within row, data sharing different superscripts are significantly different ($p<0.05$). Mean±SEM

higher ($p<0.05$) than those of the birds with delayed access to feed. Also, T_3/T_4 levels tended to decrease with delay in feed access.

DISCUSSION

The present study revealed clearly that the delay in feed access affected post-hatch growth. Delayed access to feed for 48 h, adversely affected the weights of the keets. Day-old keets lost 2.86 g of weight due to lower yolk sac. The yolk sac serves as a temporary energy and protein source and it is important to support the early post-hatch life. The weight loss indicates that, the yolk sac of the day-old keets having delayed access to feed was exhausted by the birds to maintain the energy level. Similar findings were observed by Pinchasov and Noy[7] and Tong *et al.*[18] who reported that delayed access to feed and water for 48 h caused a loss in their initial post hatch weights and reduced yolk sac content. On the contrary, Franco[19] showed that 48 h delayed placement of chicks had no significant effect on yolk sac utilization. Interestingly, in the present study, the delayed feed access did not affect the body weights of guinea fowl during the first 2 weeks. However, beyond this point, the body weights of guinea fowl with immediate access to feed were higher than those of the birds with delayed access to feed. Results of the present study were similar to previous studies conducted by Almeida *et al.*[20] and Gaglo-Disse *et al.*[21] who reported that layer chicks with 48 h delayed feed access had lower body weights. Chamblee *et al.*[22] reported that the yolk sac is crucial in determining early growth of chicks. However, Bhanja *et al.*[23] and Shinde *et al.*[24] reported that yolk sac content was more rapidly utilised in chicks subjected to immediate placement when compared to those subjected to prolonged post-hatch holding time. Delayed feed access has been reported to cause detrimental effects on juvenile performance[21]. In the present study, feed intake and FCR were not affected by delayed feed access but post hatch mortality was higher in guinea fowls subjected to delayed feed access. Moreover, in the present study, final weight gain, carcass yield of guinea fowl with immediate access to feed were higher than those guinea fowls subjected to delayed access to feed. A similar opinion was expressed by Vieira and Moran[25] who indicated that 24 h of delayed placement was detrimental to early and final weight gain, carcass yield and overall mortality but had negligible effect on FCR. The results showed that the efficiency in feed utilization was higher in the birds with immediate access to feed. It has been shown that early nutritional status affected the percentage of muscle fiber type and changed the mRNA expression for the growth-related genes in muscle[26]. Interestingly, Kornasio *et al.*[14] reported that delayed feed access (36 h posthatch) decreased the number of the breast muscle cells and myofiber diameter in post hatch chicks. The depressed growth performance in chicks subjected to delayed placement has been associated with impaired morphological development of small intestine and thus impairment of nutrient utilisation[2,19,27,28]. Indeed, in the present study, the weight and length of intestine, jejunum were higher in guinea fowl with immediate feed access. The negative effect of delayed access to feed on intestinal villi length could be attributed to loss of enterocytes through apoptotic mechanisms or reductions in proliferations[2]. In accordance with Franco *et al.*[19] delayed placement had no effect on duodenal villi length and crypt depth at 21 day. There is a possibility that by 21 day, the chicks had undergone "compensatory" development in duodenal villi height. It is known that early access to feed results in a more rapid and efficient development of the intestine during the immediate post-hatch period[9]. Delayed feed access at the end of 11 week caused lowering of relative organ weights (liver, gizzard) and serum concentrations of total cholesterol, glucose and total proteins. Therefore, it is suggested that intermediary metabolism for proteins and fats has been adversely affected by early feed deprivation. Similarly, Gaglo-Disse *et al.*[21] reported that, layer chicks with 48 h delayed feed access had decreased serum glucose, total protein and triglycerides concentration. This may affect energy balance and mobilization of protein for growth, resulting in a lower body weights due to delayed feed access. In addition, it can be

hypothesised that the delayed feed access for a longer duration can negatively affects gastrointestinal tract by decreasing the absorption of nutrients. The concentration of T_3 and T_4 at slaughter age was lower in the birds subjected to delayed access to feed. The result of this research is in accordance with the findings of Careghi *et al.*[1], who observed lower plasma T_3 and T_4 levels immediately after a feed delay. This suggests that the depressed thyroid activity reflected the reduced metabolic rate and poor growth performance of the birds.

CONCLUSION

It can be concluded that early feeding is important for juvenile performance regardless of the goal of production. Also, delayed feed access is detrimental to post-hatch performance due to poor development of gastrointestinal tract of the birds.

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