



## Performance and intestinal development of chicks submitted to different rations and different types of incubation

[*Performance e desenvolvimento intestinal de pintos submetidos a diferentes rações e diferentes tipos de incubação*]

### **"Scientific Article/Artigo Científico"**

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#### **Abstract**

The objective of this work was to evaluate the effects of two types of incubation and two types of feeding on the performance of chickens and intestinal development of chicks during the pre-starter phase (1 to 7 days of age). One-year-old male Ross® chicks were used. The experimental design was a randomized complete block design, in a 2x2 factorial design, with two types of feed processing (bran and micropelleted) and two types of incubation machines (single and multiple stage), totaling four treatments, with four treatment repetitions each. Twelve birds per experimental unit were used, totaling 192 animals. Performance evaluations were conducted, such as mean weight, weight gain, feed intake, feed conversion, bowel weight and size, and percentage of intestine in relation to live weight. Histomorphometric tests on the height of the villi and depth of the duodenal crypts were also performed. The two incubation machines did not influence any of the analyzed variables. Even though both had the same composition, the micropelleted feed resulted in positive gains for the performance variables: average weight, weight gain, feed intake, and feed conversion when compared to the meal, but was unable to significantly influence the morphometric measurements of the duodenum of chicks in the pre-starter phase.

**Keywords:** duodenal; incubators; nutrition.

#### **Resumo**

O objetivo deste trabalho foi avaliar os efeitos de dois tipos de incubação e dois tipos de alimentação no desempenho de galinhas e no desenvolvimento intestinal de pintos durante a fase pré-inicial (1 a 7 dias de idade). Foram utilizados pintos machos Ross® de um ano de idade. O delineamento experimental utilizado foi o de blocos ao acaso, em esquema fatorial (2x2), com dois tipos de processamento de ração (farelada e micropelotizada) e dois tipos de máquinas de incubação (estágio único e múltiplo), totalizando quatro tratamentos, com quatro repetições de tratamentos cada. Foram utilizadas 12 aves por unidade experimental, totalizando 192 animais. Avaliações de desempenho foram realizadas, como peso médio, ganho de peso, consumo de ração, conversão alimentar, peso e tamanho do intestino e porcentagem de intestino em relação ao peso vivo. Além de testes histomorfométricos da altura das vilosidades e profundidade das criptas duodenais. As duas máquinas de incubação não foram capazes de influenciar nenhuma das variáveis analisadas. A ração micropelotizada trouxe ganhos positivos para as variáveis de desempenho: peso médio, ganho de peso, consumo de ração, conversão alimentar, em comparação com a farelada, mesmo ambas tendo a mesma composição, mas não foi capaz de influenciar significativamente os exames morfométricos do duodeno de pintos no período pré-inicial.

**Palavras-chave:** duodeno; incubadoras; nutrição.

## Introduction

Understanding the importance of adequate feed for broilers is definitely of fundamental importance for a quality end product so that not only the poultry chain can obtain good results, but also all Brazilian agribusiness (Troni et al., 2016). In this scenario, the choice of the physical processing of the food depends on the biological, genetic and, mainly, productive purpose of the birds (Melo et al., 2016). The pelleting procedure for animal feed is well recognized and used. Any imbalance during the manufacturing process can cause irreversible economic and nutritional damages (Andrade et al., 2016).

Carvalho et al. (2013) reported the consequences of nutritional restriction immediately after hatching on bird growth. The well-known hatch window is deeply related to this fasting, since all birds do not hatch at the same time. Thus, some animals can remain without food for longer periods. Failure to provide food immediately after hatching may disrupt the proper development of organs of the digestive tract. The incubation period of the eggs is of equal importance since any bad practices at this stage can cause issues throughout the animals' life. Temperature, humidity and ventilation are some of the most important parameters, since the embryos do not tolerate large oscillations in these variables (Santana et al., 2013).

Every productive cycle of broilers depends on several principles. Among them, the characteristics of the digestive system are very relevant since the maximum use of food by birds depends on a good development and good functionality of said system. The intestines are a fundamental part since they are responsible for the absorption of these nutrients (Sousa et al., 2015). About 80% of productive expenditures are related to nutrition, thus maintaining bowel integrity and ensuring bowel development is essential to ensure a bird absorbs most of the available food (Maiorka et al., 2002).

The pre-starter phase is very important because at this time, changes occur in the physiological and morphological part of the gastrointestinal tract of the animals. During this period, there is an enlargement of the surface involved in absorption and digestion of food. This is evidenced by the fact that the altometric increase of the small intestine is up to four times higher when compared to the live weight growth of the broilers (Nunes et al., 2011).

The objective of this study was to evaluate the effects of feeding micropelleted and bran rations, with the same composition and different incubation machines (single stage x multiple stage), on the productive and development performance of the intestines of chicks during the pre-starter phase (1 to 7 days).

## Materials and Methods

The experiment was carried out in the Poultry Sector of the *Universidade Estadual de Goiás* (UEG), Campus São Luís de Montes Belos (SLMB).

One-year-old male Ross® chicks, from hens of a same age, were incubated in different types of incubators (single x multiple stage). The period in which the experiment occurred was the pre-starter stage (1 to 7 days). The experimental design was a randomized complete block design in a factorial design (2x2), using two types of feed processing (bran and micropelleted) with the same nutritional composition and two types of incubators (single and multiple stages), totaling four treatments, with four replications each. Twelve birds per experimental unit were used, totaling 192 animals, as described below:

Treatment 1: chicks from single-stage incubators fed with meal in the experimental shed. Treatment 2: chicks from single-stage incubators fed with micropelleted ration. Treatment 3: day-old chicks from multiple-stage incubators fed with meal. Treatment 4: day-old chicks from multiple-stage incubators fed with micropelleted feed.

All birds were fed with micropelleted feed immediately after hatching in the hatchery.

From the second day of accommodation, the birds had available pre-starter feed and water ad libitum throughout the experimental period. The pre-starter diets offered on the farm were free of any chemicals, i.e. anticoccidials and growth-promoting antibiotics. The rations were formulated to meet the nutritional needs of broiler breeding during the study phase (2 to 7 days) according to the nutritional recommendations by Rostagno et al. (2011) as described in Table 1.

The birds during the experimental period remained in a plastic box measuring 1.2 m<sup>2</sup> and bedding composed of rice straw. A 60-Watt incandescent bulb per box was used for heating the birds and to ensure that the zone of neutrality was maintained, always following the farming manual for the respective lineage. The lighting program established in the aviary in the pre-starter breeding

phase for ages 0 to 7 days had 22 hours of artificial light and 2 hours of dark per day (22L: 2D).

The performance variables evaluated were: mean weight (MW): obtained by dividing the total weight of the birds of each plot by the average number of birds in the plot,  $MW = PF / NB$ ; Weight gain (WG): calculated by the difference between the final weight and the initial weight of the birds added to the weight of dead birds divided by the average number of birds,  $WG = [(PF - IW) + DW] / NB$ ; feed consumption (CR): calculated by the ratio between total feed consumption (supplied minus leftover) and average number of birds; feed conversion (FC): calculated by the ratio:  $FC = CR / WG$ . The weighing of the birds and of the rations were carried out at seven days of age, to calculate feed intake, weight gain, and feed conversion (Lima et al., 2018).

To determine the biometric indexes, one bird per plot was euthanized on the seventh day of age after six hours fasting. The weight of the birds and intestines were recorded to calculate the body weight / weight ratio of the bird. In addition to weight, the length of the duodenum was also measured for calculation of the weight / length ratio.

At 7 days of age, fragments of duodenum were collected, which were stained by Hematoxylin-Eosin (HE). From each slide, the images were scanned using a light field optical microscope (Carl Zeiss model Jenaval) into the computer, via a digital video camera and capture card. Subsequently, the images obtained were used to calculate the morphometric indexes using Axion Vision 3.0 software.

Afterwards, quantified histomorphometry was performed in microns, adopting the criteria proposed by Uni et al. (1998). Villi height was measured from the apex of the villi to the base of the junction between villi and the crypt. Crypt depth was defined by the depth of the invagination of the crypt with adjacent villi. The ratio between villi height and crypt depth was also calculated.

For each repetition, 25 fields for villus height and 25 for crypt depth were read using the image j® software, totaling 400 readings for villi height and 400 for crypt depth for each repetition, always from the left to the right of the cut. The quantitative data for performance, biometry, and histomorphometry were submitted to analysis of variance (ANOVA) and the means were compared via the Tukey test at 5%, using Sisvar software, version 5.6 (Ferreira, 2014).

**Table 1.** Composition of the feed provided to the chicks during the period of 1-7 days.

| Ingredients%                         | Pre-starter phase (1-7 days) |
|--------------------------------------|------------------------------|
| Grain corn                           | 60.75                        |
| Soybean meal 46%                     | 28.77                        |
| Offal flour                          | 4.53                         |
| Meat meal 45%                        | 1.40                         |
| Offal oil                            | 1.07                         |
| Lysine 64%                           | 0.59                         |
| Methionine                           | 0.39                         |
| Threonine                            | 0.15                         |
| Limestone 38%                        | 0.73                         |
| Salt                                 | 0.21                         |
| Sodium bicarbonate                   | 0.21                         |
| Additive, Suppl. vitamin and mineral | 1.20                         |
| <b>Total</b>                         | <b>100.00</b>                |
| <b>Nutritional levels</b>            |                              |
| Metabolizable energy (Kcal)          | 2.980                        |
| Crude protein                        | 23.9329                      |
| Crude fiber                          | 2.5719                       |
| Ethereal extract                     | 4.6859                       |
| Ashes                                | 4.4461                       |
| Calcium                              | 0.9400                       |
| Total phosphorus                     | 0.6772                       |
| Total lysine                         | 1.5204                       |
| Total methionine                     | 0.7493                       |
| Met. + Cist. Total                   | 1.1336                       |
| Total threonine                      | 1.0365                       |
| Total tryptophan                     | 0.2819                       |
| Digestible arginine                  | 1.3807                       |
| Digestible lysine                    | 1.3400                       |
| Digestible methionine                | 0.6901                       |
| Met. + Cist. digestible              | 1.0065                       |
| Digestible threonine                 | 0.8777                       |
| Digestible tryptophan                | 0.2524                       |
| Phosphorus available                 | 0.4821                       |
| Sodium                               | 0.2250                       |
| Chlorine                             | 0.1712                       |
| Potassium                            | 0.7902                       |
| Acid-base balance                    | 240.000                      |

## Results and Discussion

The performance results of the chicks at seven days of age are expressed by the mean values of the treatments and presented in Table 2.

It is observed that the animals from single-stage incubators presented higher body weight at birth. Similar results were described by Araújo et al. (2016) and Villanueva et al. (2016). A plausible explanation is due to the fact that this type of equipment operates with its parameters oscillating

according to the need, age and development of the embryos. This differs from the multiple stage method, which uses the mean values of the variables, since it contains birds at different growth ages (Araújo and Albino, 2011). However, this upper initial weight for chicks incubated in single-stage machines did not result in a difference ( $p > 0.05$ ) between treatments at seven days of age (Table 2).

**Table 2.** Initial weight in the hatchery (IWH) and on the farm (IWF), mean weight (MW), weight gain (WG), feed consumption (CR), and feed conversion (FC) of chicks incubated in two types of machines and that received two types of feed in the first 1-7 days of life.

|                | IWH   | IWF     | MW       | WG       | CR     | FC      |
|----------------|-------|---------|----------|----------|--------|---------|
| Multiple Stage | 45.75 | 47.31   | 201.08   | 151.12   | 146.92 | 0.942   |
| Single Stage   | 46.02 | 47.69   | 197.25   | 149.50   | 138.50 | 1.001   |
| M. Pelleted    | 45.87 | 49.25 a | 220.75 a | 171.50 a | 132.35 | 0.819 b |
| Meal           | 45.90 | 45.75 b | 177.58 b | 129.12 b | 153.17 | 1.124 a |
| Machine        | 0.473 | 0.561   | 0.718    | 0.867    | 0.433  | 0.231   |
| Feed           | 0.934 | <0.001  | <0.001   | <0.001   | 0.071  | <0.001  |
| Machine x Feed | 0.693 | 0.188   | 0.875    | 0.690    | 0.207  | 0.425   |
| CV (%)         | 8.40  | 2.46    | 9.92     | 12.64    | 12.41  | 8.73    |

<sup>a,b</sup> Different letters in the same column show statistical difference via Tukey test ( $p < 0.05$ )

Controlling the weight of the chicks is indispensable, and their weight can be altered by several factors, such as nutritional grade and egg size, as well as inadequate incubation characteristics. All of these conditions are dependent on the level of embryonic development (Almeida et al., 2006). Weight of the birds in a hatchery is a predominant factor for good productivity. Leandro et al. (2006) shows that chicks with an initial weight between 40 and 50 g are those with the best capacity and higher probability for a better zootechnical performance within the appropriate time frame. The birds used in this study matched these characteristics, as presented in Table 2.

The hatch window is the time from the first hatching to the last one. Araújo et al. (2016) have shown that this factor can interfere in several ways, such as causing harm to birds born at the end of this period. This is because the chicks born toward the end of this period don't have enough time for their navel to heal properly, and many of them have their feathers still moist, leading to them being considered inadequate, which culminates in their being discarded.

The mean weight at seven days, when the different diets were evaluated, showed a difference between the treatments ( $p < 0.05$ ), where the birds fed with micropelleted feed had a 19.55% approximate increase in mean weight compared to

those fed with meal, a result also found by Boemo et al. (2016).

Results obtained by Silveira et al. (2010) corroborate those found in the present study, where weight gain and slaughter weight were higher and feed conversion more efficient ( $p < 0.05$ ) in those chicks fed with micropelleted feed, showing a statistical difference. This is probably due to the higher concentration of nutrients in the food because of the pelleting process, as well as uniform size which counters the chicks' preference for larger particles, a phenomenon that is quite common when feeding with meal and results in nutritional imbalance, and loss of productivity (Lorençon et al., 2007).

In relation to the feed intake variable ( $p > 0.05$ ), the animals that received a micropelleted diet eventually consumed a smaller amount of the food, over the evaluation time. However, this result contradicts findings by Oliveira et al. (2011), who concluded the opposite, with chickens fed with micropelleted feed had a higher food consumption, also in comparison with the meal.

According to Pucci et al. (2010), the cause of the lower consumption of micropelleted food is due to the glycostatic factor, where the food receives high temperatures during its production, which facilitates starch digestibility. There is also the breakdown of some organelles, making



carbohydrates and proteins more palatable for birds.

The lower consumption of micropelleted food still offers a certain environmental advantage. Therefore, a smaller amount of raw material will be used for formulation and preparation of these rations, mainly reducing the need and pressure for a greater production of grains like corn and soybean. This, in turn, favors the sustainable development of Brazilian agribusiness.

Comparing the feed conversion of chicks fed with bran and extruded and crushed feed, Faria et al. (2006) concluded that the animals that received the bran food had worse indicators for the same phase evaluated in this work and also had a higher consumption of this type of feed.

In their experiment, Freitas et al. (2009) concluded that if the birds are fed the same type of feed during and after the pre-starter phase, gains during the first seven days of life may be observed until 21 days, with a slight drop until slaughter.

Incubation systems were not able to significantly influence ( $p > 0.05$ ) the values found for intestine length and intestine percentage in relation to live weight (Table 3), similarly as observed by Villanueva et al. (2016) and Freitas et al. (2008). For these same variables, the different diets had similar implications, with results that did not show statistical difference ( $p > 0.05$ ), as found by Lara et al. (2008).

**Table 3.** Biometry of the duodenum of chicks incubated in two types of incubators and receiving two types of feed at seven days of age.

|                | Chick weight at slaughter (g) | Bowel Weight (g) | % Intestine in relation to live weight | Bowel length (cm) |
|----------------|-------------------------------|------------------|--|-------------------|
| Multiple Stage | 198.37                        | 12.00            | 5.94                                   | 83.04             |
| Single Stage   | 197.25                        | 12.12            | 6.12                                   | 86.58             |
| M. Pelleted    | 220.75 a                      | 12.37            | 5.87                                   | 85.71             |
| Meal           | 174.87 b                      | 11.75            | 6.18                                   | 83.92             |
| Machine        | 0.8981                        | 0.900            | 0.603                                  | 0.170             |
| Feed           | <0.001                        | 0.535            | 0.366                                  | 0.472             |
| Machine x Feed | 0.6208                        | 0.708            | 0.333                                  | 0.409             |
| CV (%)         | 8.64                          | 15.5             | 11.03                                  | 5.21              |

<sup>a,b</sup> Different letters in the same column show statistical difference via Tukey test ( $p < 0.05$ ).

Agostinho et al. (2012) stated the positive aspects that providing pre-feed ration can bring to birds in the pre-starter phase, such as better development of the average intestinal weight and more efficient weight gain; however, with a worse feed conversion and higher feed consumption. Tavernari and Mendes (2009), explaining the speed and development of duodenal villi, reported that exactly between the sixth and eighth day of a chick's life, these structures reach the highest growth rate, thus further strengthening the need and seriousness with which food should be observed during the pre-starter phase.

It is shown in Table 4 that the different types of incubator machines did not significantly influence intestinal villi height and crypt depth in the duodenum. Although there was no statistical difference ( $p > 0.05$ ), the chicks from single-stage machines had higher values for villi height and depth of intestinal crypts. This may have been due to better sanitary control of these equipment. Valdo et al. (2020) reported that the eggs incubated in this

manner are better tracked and monitored, since they originate from the same batch depending on the time spent in the machines, which also facilitates cleaning and disinfection.

Among the other parts of the small intestine, the duodenum is one of the most sensitive to fasting after hatching, with only 37.1% of the villi remaining intact at the end of this type of management (Gomide Junior et al., 2004). As in the present study, all chicks received micropelleted feeding soon after hatching. Taking into account only the villus: crypt relationship, it is not necessary to continue with this type of nutrition during the pre-starter phase, since no statistical difference occurred between this type of food and the bran meal.

Flores et al. (2013) warned of the harmful occurrence of microclimates inside incubators, mainly due to poor airflow between the eggs, thus not allowing the eggs to have adequate temperature for the development of the embryo.

**Table 4.** Villi height (HV), crypt depth (DC) and villus:crypt ratio of the duodenum for chicks incubated in two types of incubators and that received two types of feed at seven days of age.

|                | HV ( $\mu\text{m}$ ) | DC ( $\mu\text{m}$ ) | Villus:Crypt |
|----------------|----------------------|----------------------|--------------|
| Multiple Stage | 1162.90              | 151.29               | 7.78         |
| Single Stage   | 1282.63              | 158.65               | 8.15         |
| M. Pelleted    | 1215.14a             | 158.91a              | 7.72a        |
| Meal           | 1230.13a             | 151.04a              | 8.22a        |
| Machine        | 0.062                | 0.464                | 0.580        |
| Feed           | 0.804                | 0.435                | 0.452        |
| Machine x Feed | 0.204                | 0.844                | 0.502        |
| CV (%)         | 9.45                 | 12.43                | 15.94        |

<sup>a,b</sup> Different letters in the same column show statistical difference via Tukey test ( $p < 0.05$ ).

The birds originating from the single stage machines were those that showed the highest ratio between villi height and crypt depth. It was also observed (Table 4) that although duodenal biometry and bird weights were greater for micropelleted feed, histomorphometric tests were very similar, both for crypt height and for intestinal villi. Results were similar to those obtained by Freitas et al. (2008), who observed the non-influence of the type of processing of the feed on these variables. This hypothesis can be sustained in parts by the fact that rations with the same granulometry, such as the micropelleted ones, begin to be digested in the proventriculus, arriving in the duodenum totally dissolved, extinguishing the effect of the physical form of the rations on this part of the intestine (Nir et al., 1994).

### Conclusion

The different types of incubators were not able to statistically influence any of the variables analyzed. The micropelleted feed, when compared to the meal, was able to positively interfere in the performance of the chicks at seven days of age. However, the same was not observed in relation to histomorphometric indices of the duodenum.

### Ethics Committee

The experimental protocol used in this study was approved by the Ethics Committee for the use of animals in research at the *Universidade Estadual de Goiás* (CEUA-UEG), under protocol n. 009/2018 and is in accordance with the Ethical Principles on Animal Experimentation, adopted by the Brazilian Society of Laboratory Animal Science (SBCAL).

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