Guanidinoacetic Acid Over Reproductive Indexes and Performance of Progenies Using Meat-Type Quail’s Breeder

A.E.MURAKAMI¹*, R.J.B.RODRIGUEIRO², T.C.SANTOS¹, I.C.OSPINA-ROJAS¹, M.RADEMACHER³

¹ State Maringá University, Animal Science Department, Av Colombo, 5790, 87020-900, Maringá, PR, Brazil.  
² Evonik Degussa Ltda Brazil, Al. Campinas, 579 5 andar, 01404-000, São Paulo, SP, Brazil.  
³ Evonik Industries AG, Feed Additives, Rodenbacher Chaussee 4, 63457 Hanau, Germany.  
*Corresponding author: aemurakami@uem.br

Introduction

Creatine is considered an important element in energy metabolism in the cells of vertebrates and birds (MICHEILS et al. 2010). Studies have shown that guanidinoacetic acid (GAA), a precursor of creatine, can be classified as a conditionally essential nutrient because although birds endogenously synthesize creatine, approximately 25 to 33% of the total requirement of this component must be provided in the diet. Thus, GAA supplementation may be necessary to meet the needs for creatine and optimize performance of broilers fed vegetable diets (LEMMÉ et al., 2007). This fact has changed into a huge scientific interest because creatine sources can really be important for the animals once that the survivance and the best performance of animals depend on an efficient energetic metabolism.

If the birds in reproduction phase are fed with vegetable diet they may show a limited productive and reproductive performance. Studies about this topic were not found, what is even more limited when try to know the correlation between the creatine source and its effect on the reproductive performance or embryonic development of the domestic fowl.

Material and Methods

The trial was conducted in the quail house of Maringa State University, Brazil. In the first phase, a total of 240 meat-type quail breeders (25 weeks) were used, in a completely randomized experimental design of five treatments, with 8 replicates of 6 quails each (2 females : 1 male ratio).

The basal diets made based on corn and soybean meal were isocaloric and isonutritional, according to SILVA & PERAZZO (2009). The guanidinoacetic acid (GAA) in the basal diet replaced the inert creating the treatments: 0.0%; 0.06%; 0.12%; 0.18% and 0.24% of GAA. During the adaptation period and through all the experimental period, feed, in mash form, and water were supplied ad libitum. The light program was of 17 hours of light (natural + artificial) with 7 hours of dark.

Productive parameters were obtained weekly during 4 weeks to egg production and feed intake. The contents of three eggs per replicate were homogenized, weighed and lyophilized (Christ Alpha 1-4 LD plus, Osterode am Harz, Germany). The content of guanidine acetic acid, creatine and creatinine in the eggs were performed by the Company AlzChem AG in Germany using the ion-chromatography technique (Method-SOP 136-246/2).

Incubation performance was analysed in eggs produced in seven consecutive days (29th week), stored around 20°C, and incubated. Incubation were performed in an automatic incubator (37.6°C and relative humidity of 65%) until 15th day, when eggs were transferred to hatch machine (37°C and relative humidity of 70%), and, by the end, in the 17th day the hatchability period was established.

The number of sperm was determined per unit area in the outer perivitelline layer from six fertile eggs/replicate after preparation with DNA-marker DAPI (diaminodinophenilindole).
The progenies originated from those breeders were distributed according breeders treatments in four replicates of 20 quails. Quails were fed with vegetable diets (corn and soybean meal) without supplementation of GAA. The feeding programs of two phases (01 to 21 days and 22 to 35 days of age) were used.

The breast muscles of 8 chicks per replicate were collected at hatching. The samples were ground and pooled by replicate, and 1 gram of ground tissue was used to determine the creatine contents in the breast muscles according to the procedure described by Chamruspollett et al. (2002).

All data were analyzed by the GLM procedure of SAS software (SAS Institute, 2009).

Results and Discussion

Breeder Performance and Egg Quality

The levels of guanidinoacetic acid (GAA) did not have an effect (P>0.05) on the feed intake, production and weight of eggs. Also, the number of spermatozoa in the perivitelline layer did not show differences (P>0.05) among treatments (Table 1).

Table 1. Breeders egg production and GAA in the eggs of meat-type quail.

<table>
<thead>
<tr>
<th>GAA (%)</th>
<th>Feed intake (g/day)</th>
<th>Egg laying (%)</th>
<th>Egg weight (g)</th>
<th>GAA (µg/egg gram)</th>
<th>Creatine (µg/egg gram)</th>
<th>Creatinine (µg/egg gram)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.00%</td>
<td>32.87±0.44</td>
<td>85.48±5.30</td>
<td>13.38±0.27</td>
<td>0.00±0.00</td>
<td>19.63±1.15</td>
<td>2.25±0.16</td>
</tr>
<tr>
<td>0.06%</td>
<td>32.21±0.72</td>
<td>86.63±5.28</td>
<td>13.49±0.20</td>
<td>0.25±0.25</td>
<td>24.88±0.91</td>
<td>2.88±0.23</td>
</tr>
<tr>
<td>0.12%</td>
<td>32.79±0.62</td>
<td>90.45±3.04</td>
<td>13.54±0.32</td>
<td>1.50±0.19</td>
<td>24.13±1.06</td>
<td>2.63±0.18</td>
</tr>
<tr>
<td>0.18%</td>
<td>32.33±0.47</td>
<td>87.36±2.60</td>
<td>13.51±0.13</td>
<td>1.88±0.29</td>
<td>25.38±0.91</td>
<td>2.88±0.23</td>
</tr>
<tr>
<td>0.24%</td>
<td>32.45±0.60</td>
<td>85.04±3.18</td>
<td>13.37±0.20</td>
<td>3.00±0.46</td>
<td>26.75±1.52</td>
<td>3.25±0.16</td>
</tr>
<tr>
<td>CV (%)</td>
<td>5.04</td>
<td>11.94</td>
<td>4.88</td>
<td>60.34</td>
<td>13.26</td>
<td>19.85</td>
</tr>
</tbody>
</table>

ns – There was no difference by the F test (P>0.05). 1 CreAMINO® 96%. 2 GAA – Guanidine acetic acid analyzed in the egg. L- linear effect (P<0.05). 3 Regression analyses.

Increasing the dietary level of GAA, resulted in a linear effect (P<0.05) on GAA, creatine and creatinine content in the eggs (Table 1), showing that, different levels of GAA in the diet when consumed by fowl, are absorbed by the intestine and carried by the bloodstream to the liver, receiving a methyl group from S-adenosylmethionine catalyzed by the guanidinoacetate methyltransferase (GAMT) producing creatine that is transferred vertically to the egg. Lastly, the creatine produced can break down spontaneously to creatinine (LEMME et al., 2007).

There was transference from meat-type quail breeder of 19.63 micrograms of creatine per egg gram to the egg when they were fed with basal diet. On the other hand, quail breeders fed with 0.24% of GAA, transferred 26.75 micrograms of creatine per gram of egg, which is an increment of 36.27% of creatine per gram egg produced.

Incubation Parameters

The incubation of eggs parameters showed quadratic effect (P<0.05) to meat-type quail breeders fed with different levels of GAA.

Fertility had a quadratic effect (P<0.05; Y = 91.96 + 82.82X – 299.7X^2, R^2 = 0.88) with better values observed with 0.14% of GAA. This result suggested that the increment in fertility probably was a result of a better quality of spermatozoa. Inside the female reproductive tract the seminal liquid quality may help spermatozoa to synthesize or use the
ATP with a better efficiency, resulting in a better energy to gain access to ovum and hydrolyzing the perivitelline layer of the oocyte in the germinative disc. In this case fertility could not be directly correlated to the number of spermatozoa found in the perivitelline layer in the eggs, but by the quality of them.

The fluid of the reproductive tract of the animals contains high amount of creatine and phosphocreatine that can be used by the spermatozoa (LEE et al., 1988). The exposition of the spermatozoa with high concentration of creatine can, directly, improve their metabolism as well as the flagellar motility and consequently improves the fertility. Considering this argumentation, the GAA seems to be important to the avian reproduction, because, the spermatozoa naturally keep for long time in the uterovaginal glands. In quails, these cells can be found around laid eggs for 10 days, but fertility reduces considerably after 3 days of copulation (SANTOS et al., 2013).

The embryo mortality in the incubation period showed a quadratic effect (P<0.05, Y= 20.64 – 142.3X + 454.0X², R² = 0.64), estimating the level of 0.16% of GAA in the diet to give the lowest level of embryo mortality. Increasing levels of the GAA for the meat-type quail breeder during the reproduction phase provided quadratic effect (P<0.05) on the total hatchability and hatching fertile eggs. The regression analysis determined to these parameters the levels of 0.15% and 0.16%, respectively. The hatchability of eggs seems to be influenced directly by the fertility and concentration of the creatine in the egg that is provided by the GAA present in the diet of meat-type quail breeder. In this case, the embryo using the creatine in the egg produced by the meat-type quail breeder fed with GAA, may be used, with the best efficiency, the ATP form glucose and lipids source, improving the hatchability.

RAMIREZ et al. (1970) showed that the chicken embryos during the incubation period have body necessities for creatine. In fact, GAA, a creatine precursor, when supplemented in the diet, increased the creatine content in the egg contributing to the ATP availability to meet the embryonic metabolism and as consequence, improves the hatchability.

**Progenies Performance**

The meat-type quail breeder fed with different levels of GAA did not affect (P>0.05) the newborn progenies weight, but the creatine content found in the breast muscle of the newborn quails showed quadratic effect (P<0.07; Y= 0.450 + 1.152X -5.235X²; R² = 0.68), with 0.11% of the GAA in the diet of quail breeder determined the best level to provide the highest amount of creatine in the breast muscle of progenies.

The results of this trial showed that the meat-type quail breeder (males and females) when fed with the GAA can, not only, transmit the creatine to the egg, but also improve the fertility and reduce the quail embryo mortality. The result also indicated that the quail embryos uses the creatine present in the egg making their metabolism effective, increasing the content of muscular creatine, as well as, an improvement of the hatchability.

There were no differences (P>0.05) in the feed intake however the weight gain (Y= 205.3 + 281.2X – 939.9X², R² = 0.51) and the feed conversion (Y= 3.727 – 6.035X + 20.69X², R² = 0.69) presented quadratic effect (P<0.05).

**CONCLUSION**

Considering the reproductive and productive performance, it is recommended to use 0.15% of the GAA in the diet of meat-type quail breeders.

The results of this trial suggest the development of new studies to better understand the effect of GAA on the reproductive performance of farm avian breeders.
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REFERENCES