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## ANIMAL PERFORMANCE AND EGG QUALITY OF COMMERCIAL LAYING HENS FED MACAUBA (*ACROCOMIA ACULEATA* LODD.) OIL AND EMULSIFIERS

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ABSTRACT - The objective of this study was to investigate the effects of emulsifiers and partial replacement of soybean oil for macauba oil (Acrocomia aculeata Lodd.) on animal performance and egg quality of commercial laying hens. One hundred and eighty semi-weighted Hisex Brown hens, aged 35 weeks, were randomly allocated in one of the five following treatments: T1- control diet + 1% soybean oil (SO), T2 control diet + 1% SO + 0.012% emulsifier, T3- control diet + 0.2% macauba oil (MO) + 0.8% SO + 0.012% emulsifier, T4 – control diet + 0.4% MO + 0.6% SO + 0.012% emulsifier, or T5 – control diet + 0.6% MO + 0.4% SO + 0.012% emulsifier. Egg production, feed intake, feed conversion per mass and per dozen of eggs, and average egg weight were the performance variables evaluated. Regarding egg guality, the following variables were evaluated: egg yolk color, Haugh Unit, specific gravity, shell thickness, percentage of yolk, egg white, and shell, and viable eggs. The study was carried out in a completely randomized design and means were compared using the SNK test as 1% of probability. There was a statistically significant effect (P < 0.01) of macauba oil and emulsifier on egg yolk color and feed intake. No effect was observed (P > 0.01) on other variables evaluated. Therefore, emulsifiers and macauba oil can be used as a partial replacement for soybean oil in diet of laying hens.

Keywords: Egg production, fatty acids, oil levels

## USO DE EMULSIFICANTE E ÓLEO DE MACAUBA (ACROCOMIA ACULEATA LODD.) EM DIETAS PARA POEDEIRAS COMERCIAIS E SEUS EFEITOS SOBRE DESEMPENHO E QUALIDADE DE OVOS

objetivo com o presente trabalho foi verificar os efeitos da RESUMO - O suplementação de dietas com uso de óleo de macaúba (Acrocomiaaculeata (Jacq.) Lodd.) e emulsificante no desempenho produtivo e na qualidade dos ovos de poedeiras. Utilizou-se 180 poedeiras distribuídas em 5 tratamentos, 6 repetições com 6 aves cada, totalizando 30 parcelas. As rações foram formuladas de acordo as exigências nutricionais dadas por Rostagno et al., (2005). Nas variáveis de desempenho foram avaliadas a porcentagem de produção de ovos, consumo de ração expresso em gramas/ave/dia, conversão alimentar por massa e dúzia de ovos e peso médio do ovo. Já com relação à qualidade dos ovos realizada as análises de cor da gema, Unidade Haugh, gravidade especifica, espessura de casca, porcentagens de gema, clara, casca e ovos viáveis. A inclusão do óleo de macaúba em substituição ao óleo de soja em dietas com a suplementação de emulsificante não influenciou significativamente (P>0,05) as variaveis analisadas em relação ao tratamento controle com o uso de óleo de soja e suplementação com emulsificante. De acordo com os resultados pode-se utilizar o óleo de macaúba em substituição do óleo de soja em dietas suplementadas com emulsificantes para poedeiras comerciais nos níveis e períodos utilizados na pesquisa.

Palavras-chave: Níveis de óleo, qualidade interna, qualidade externa.

#### INTRODUCTION

Nutrition plays an important role in animal production, since animal performance is directly related to it, especially in poultry farming. Using the right percentage of oil or fat in animal rations is beneficial. This ingredient is essential to not only meet animal's energy needs, but also to meet the requirements in essential fatty acids, transport fat-soluble vitamins, improve ration palatability among others (BERTECHINI, 2006).

Currently, there has been some difficulties meeting the high demand for soybean oil, which increased the search for alternative oil sources such as macauba, palm, corn, sunflower among others. Macauba (*Acrocomia aculeata* Lodd.) is a native Brazilian species found in the Cerrado. Although there is not many studies evaluating macauba oil on animal nutrition, it can be a feasible alternative due to its nutritional properties and fatty acid content as well as its high concentrations of carotenoids (MOTTA, 2011).

The former is responsible for the orange color of the oil and may benefit the pigmentation of the egg yolk if used in laying hens feeding. Oliveira et al. (2010) found that egg yolk pigmentation is the result of carotenoid deposition on it. In general, a darker egg yolk color is desirable in commercial laying hens. Then, it is important to feed pigments to hens since they can absorb from 20 to 60% of the pigments from the diet.

According to Pupa (2004), the use of lipids in hen diets is beneficial from an economic point of view because it has a high caloric value (2.25 times higher than other feedstuff), promotes ration economy due to improvement in feed conversion ratio, has a relatively low cost, and improves animal productivity.

The role of feed additives is another worth mentioning aspect since they improve feed digestibility. Emulsifiers are additives recently studied in poultry nutrition with the aim of improving the absorption of lipids by aiding the formation of micelles, thus forming an emulsion that is better absorbed by the animal.

Emulsifiers are amphipathic and surface-active agents with medium molecular weight, reason why they are absorbed in the interface between oil and water, reducing the surface tension and energy required forming the emulsion (ARAÚJO, 1995).

The objective of this study was to evaluate the effects of soybean oil partial replacement for macauba oil and the supplementation with emulsifier in diet of commercial laying hens aiming at improving fatty acid absorption and evaluating its effects on internal and external egg quality as well as animal performance.

#### MATERIAL AND METHODS

The study was conducted in the experimental sector of egg layers farming of the Animal Science Department of the Federal Institute of Education Science and Technology of Minas Gerais – *"Instituto Federal de Educação, Ciência e Tecnologia de Minas Gerais"* (IFMG – Bambuí Campus), from December 3, 2014 to December 31, 2014. All animal procedures were carried out in accordance with the guidelines described in the Guide for the Care and Use of Agricultural Animals in Research and Teaching (FASS 2010).

One hundred and eighty semi-weighted Hisex Brown hens aged 35 weeks were selected based on their average egg production. They were housed in 25 x 45 x 35cm galvanized wire cages with two animals/cage in a conventional laying barn. The barn had a clay tile roof, a central aisle, and one cage line on each side. The cages were arranged in double rows (pyramidal system), using feeder trough and automatic *nipple* drinkers.

A light program of 16 hours of light per day was adopted. The average maximum and minimum temperatures recorded in the barn during the experimental period were 29.5° and 21.7° C, respectively.

The study was carried out in a completely randomized design (4 periods with 7 days each) with 5 treatments and 6 repetitions. Three cages with two animals in each were considered the experimental unit, resulting in a total of 30 experimental units. Diets used in the experiment (Table 1) were formulated according to Rostagno et al. (2005) to have equal amount of calorie (2775 kcal/kg dry matter), protein (17.66% crude protein), phosphorus (0.46% P), and calcium (3.9% Ca). Five treatments were evaluated as follows: T1 – control diet (Table 1) + 1% of soybean oil (SO); T2 – control diet + 1% SO + 0.012% emulsifier; T3 – control diet + 0.2% of macauba oil (MO) + 0.8% SO + 0.012% emulsifier; T4 – control diet + 0.4% MO + 0.6% SO + 0.012% emulsifier; The emulsifier was composed of glycerol stearate, sucrose laurate, and sorbitan monolaurate. Animals had *ad libitum* access to experimental diets and water throughout the study.

Macro Ingredients	T 1	T 2	Т 3	T 4	T5
Corn 7.5% (%) F. soybean 46 (%)	60 28.4	60 28.4	60 28.4	60 28.4	60 28.4
Soybean meal (%)	1	1	0.8	0.6	0.4
Macauba oil	-	-	0.2	0.4	0.6
Emulsifier (%)	-	0.012	0.012	0.012	0.012
Limestone 38 (%)	4.3	4.3	4.3	4.3	4.3
Limestone thick (%)	4.4	4.4	4.4	4.4	4.4
Common salt (%)	0.45	0.45	0.45	0.45	0.45
Phosphate bicalcium (%)	1.34	1.34	1.34	1.34	1.34
Micro Ingredients					

 Table 1. Experimental rations used.

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Kaolin (%)	0.062	0.047	0.047	0.047	0.047
DL- methionine (%)	0.16	0.16	0.16	0.16	0.16
L- Lysine 78 (%)	0.005	0.005	0.005	0.005	0.005
Endopower Alpha C (%)	0.003	0.003	0.003	0.003	0.003
Unimix 01060 (%)	0.1	0.1	0.1	0.1	0.1
Unimix 01070 salts (%)	0.1	0.1	0.1	0.1	0.1
Total (kg)	100	100	100	100	100
Composition Calculated					
Nutrient (%)					
Protein gross	17.7	17.7	17.7	17.7	17.7
Calcium	3.7	3.7	3.7	3.7	3.7
Phosphor available	0.38	0.38	0.38	0.38	0.38
Total lysine	0.92	0.92	0.92	0.93	0.93
Met + total cis	0.71	0.71	0.71	0.71	0.71
Threonine	0.69	0.69	0.69	0.70	0.70
Energy metabolizable	2750	2750	2750	2750	2750

<sup>2</sup> Guarantee levels per kg of vitamin premix: folic Acid (min) 900.0 mg; Pantothenic Acid (min) 12, 000.00 mg; Biotin (min) 77.0 mg; Calcium (min - max) 130.0 - 143.7g; Niacin (min) 40, 000.0 mg; Selenium (min) 370.0 mg; Vitamin A (min) 8,800, 000.0 IU; Vitamin B1 (min) 2, 500.0 mg; Vitamin Growth 0.04 g; Antioxidant 0.02 g; Mn 75 mg; Zn 50 mg; Cu 8 mg; I 0.75 mg; Fe 50 mg.
<sup>3</sup> Guarantee levels per kg of mineral premix: copper (min) 7,000,0 mg; iron (min) 50.0 g; iodine (min) 1,500.0 mg; manganese

<sup>a</sup> Guarantee levels per kg of mineral premix: copper (min) 7,000,0 mg; iron (min) 50.0 g; iodine (min) 1,500.0 mg; manganese (min) 67.5 g; zinc (min) 45.6 g.

Eggs were collected twice a day (12:00 and 15:00). We recorded how many eggs were laid as well as if they were broken, cracked, thin-shelled, without the shell, or deformed. These data were used to calculate the percentage of laid and viable eggs. At the end of each experimental period, we analyzed the color of the egg yolk, Haugh unit (HU), and specific gravity (SG) as well as percentages of egg yolk, egg white, shell, and viable eggs.

Peroxide value analysis of the macauba oil was carried out to avoid possible rancidification of the diets. The analysis was performed in the physicochemical laboratory of the IFMG – Bambuí Campus. This analysis indicates all substances, in milliequivalents of peroxide per 1000g of sample, which oxidize the potassium iodide. These substances are generally considered as peroxides or other similar products resulting from fat oxidation.

According to Cecchi (2003), this method is mostly used to measure the oxidation state of oils and fats. Oils should not exceed 10 meq/1000g of sample, because it indicatives a low possibility of oxidative deterioration (MALACRIDA, 2003).

The method consisted of weighing  $5 \pm 0.05$  g of macauba oil in a 125 ml Erlenmeyer flask. Thirty milliliters of acetic acid-chloroform solution (3: 1) was added and stirred until complete dissolution. Next, we added 0.5 ml of saturated KI solution and let it in the dark for one minute. Titration with sodium thiosulfate solution 0.01 Mol/L was performed after 30 ml of distilled water was added. Titration was carried out until the disappearance of the yellow color. Then, 0.5 ml of starch indicator solution was added and the titration continued until the complete disappearance of the blue color.

We tested 3 samples and found an average of 4 meq/1000g, indicating that the peroxide level was within the limit of 10 meq/1000g of sample and macauba oil could be used without reducing the quality of the diet.

External and internal quality of the eggs was carried out during two days following each experimental period. Intact eggs from each experimental unit were collected in the morning, weighed, and calculated their average weight. Two eggs from each experimental unit were randomly selected after excluding eggs dirty and with problems in its shell (thin-shelled, deformities or cracked).

A total of sixty eggs was individually weighed and broken to analyze their yolk, shell, and albumen. Egg yolk color was obtained by comparison with the Yolk Color Fan - DSM color palette. This analysis was performed right after the egg was broken, choosing the closest color from color palette that ranged from 1 to 15. Because it is a subjective analysis, it was done by the same people and at the same place in order to avoid variations. An average per period was calculated. The shells were washed in water, dried at room temperature, and weighed. The thickness of the shell (ST) was measured using a digital micrometer (Digimess - 0.001mm precision). It was measured at three points in the middle region of the shell, obtaining an average for each egg.

Albumen height was measured in the median region of albumen using a digital caliper (Starret 150 mm). The HU was calculated using the following equation:

HU=100 ×log (h - 
$$1.7$$
w<sup>0.37</sup> + 7.6)

where: h = albumen height (mm) and w = egg weight (g) according to Brant et al. (1951).

The EG was obtained using the methodology described by Freitas et al. (2004) based on egg weight and the weight of the water displaced with the submersion of the egg. This method is called computer-assisted-displaced-water, and it obtains the SG using the following equation:

$$SG = \frac{EW}{EWW} \times Cf$$

where EW = weight of the egg in the air; EWW = egg weight in the water; and Cf = gravitational correction factor as a function of water temperature.

The proportions of yolk, albumen, and shell in relation to the weight of the egg were evaluated during the 1st week after starting the treatments. Twelve eggs per treatment were randomly chosen. The eggs were weighed in a semi-analytical (Shimadzu BL 3200 H with capacity for 3200g), broken, and separated the albumen, yolk, and shell.

Egg yolk was manually separated and individually weighed. The shells were individually weighed after they were washed in tap water to remove residues from the albumen and left to dry at room temperature for 48 hours. Albumen weight was obtained from the difference between the weight of the whole egg and the weight of the yolk plus the weight of the shell.

The following performance variables were calculated: egg production (EP), feed intake (FI), feed conversion per mass (FCm) and per dozen (FCdz) of eggs, and average egg weight (AEG). Feed intake was obtained as the difference between offered and leftovers at the end of the each week. The average daily intake was calculated based on the number of animals in each repetition and the number of animals that have died during the week.

Feed conversion was estimated with FI over the average egg weight as well as how many dozen of eggs were produced in each experimental period using the following equations:

$$FCm = \frac{FI}{EW}$$
  $FCdz = \frac{FI}{DZ}$  and

where Ew = weight of the egg and DZ = dozen of eggs produced.

The data were submitted to analysis of variance and means were compared with Tukey test at 1% probability using SISVAR software.

#### **RESULTS AND DISCUSSION**

The inclusion of macauba oil and emulsifier did not influence (P > 0.01) the HU, ST, SG, and percentage of shell, albumen, and yolk as well as viable eggs (Table 2).

**Table 2**.Haugh Unit (HU), shell thickness (ST), yolk color (YC), specific gravity (SG), percentage of yolk (% Y), shell (% S), albumen (% A), and viable eggs of laying hens receiving diets with emulsifiers and different levels of macauba oil.

Item*	Treatment	Treatment					
	T1	T2	Т3	T4	T5	——CV (%)	
HU	87.13	88.64	89.2	89.31	88.33	7.96	
ST	0.557	0.552	0.554	0.537	0.552	5.72	
YC	5.37ab	5.44ab	5.60a	5.51ab	5.27b	7.15	
SG	1.093	1.093	1.081	1.089	1.089	1.26	
% Y	24.41	24.21	28.45	24.12	24.53	5.60	
% S	9.92	9.81	9.91	9.77	9.80	5.19	
% A	65.65	65.97	65.22	66.09	65.65	2.50	
Viable eggs	99.2	98.9	98.9	97.89	98.9	2.62	

\*Means followed by different letters in the line differ statistically by SNK test at 1% of probability.

<sup>1</sup>T1= control diet + 1% of soybean oil (SO); T2= control diet + 1% SO + 0.012% emulsifier; T3= control diet + 0.2% of macauba oil (MO) + 0.8% SO + 0.012% emulsifier; T4 = control diet + 0.4% MO + 0.6% SO + 0.012% emulsifier; and T5= control diet + 0.6% MO + 0.4% SO + 0.012% emulsifier.

Egg yolk color was different (P < 0.01) between treatments. Animals receiving the treatment with 0.2% inclusion of macauba oil showed darker color than animals receiving 0.6% of macauba oil. Filard et al. (2005) also reported that the 3% inclusion of canola, soybean, linseed, or sunflower oil in the diet of laying hens changed the color of their egg yolk.

On the other hand, Oliveira et al. (2010) did not find differences on HU and egg yolk color as well as percentage of yolk, albumen, and shell of animals receiving different vegetable oils. Different from our results, Santos et al. (2005), using oil of soybean, linseed, and cotton, did not observe significant differences in egg yolk color.

According to Keshavarz e Nakajima (1995), the addition of a lipid source in the diet of young laying hens reduces the passage rate of digesta, which influences the size of their egg yolk. Consequently, there is a better absorption of the nutrients from the diet increasing their egg weight. However, we did not observe these effects in our

study, which is probably because the animals we used were not young and their eggs were already of great size.

Grobas et al. (2001) evaluated the inclusion of the oil of fish, soybean, sunflower, linseed and canola as well as bovine tallow in diet of laying hens. They did not observe the effect of treatments on HU and egg yolk color, which is similar to our results. In addition, Leeson et al. (2003) did not find an effect of corn oil added to the diet of laying hens on the color of their egg yolk.

Oliveira et al. (2010), March Bean e Leeson (2003), and Mazalli et al. (2004) evaluated the effect of oil added to the diet of old laying hens on egg constituents and they did not find a significant effect. They concluded that egg nutritional characteristics might be affected by the diet, but not the size of the egg nor its constituents, which is similar to what we found in our study.

There was no significant effect (P > 0.01) of including macauba oil and emulsifier on the EP, AEW, FCm, and FCdz (Table 3).

 Table 3. Feed intake (FI), egg production (EP), average egg weight (AEW), feed conversion per mass (FCm) and per dozen of eggs (FCdz) of laying hens receiving diets with emulsifiers and different levels of macauba oil.

Treatment					
T1	T2	Т3	T4	T5	— CV (%)
121.3ab	122.0ab	123.8ab	118.0b	127.3a	6.94
96.83	95.04	95.84	93.87	96.33	6.32
62.29	63.40	61.29	62.73	63.13	4.60
1.50	1.54	1.55	1.51	1.56	7.64
2.01	2.02	2.10	2.02	2.07	8.92
	96.83 62.29 1.50	T1T2121.3ab122.0ab96.8395.0462.2963.401.501.54	T1T2T3121.3ab122.0ab123.8ab96.8395.0495.8462.2963.4061.291.501.541.55	T1T2T3T4121.3ab122.0ab123.8ab118.0b96.8395.0495.8493.8762.2963.4061.2962.731.501.541.551.51	T1T2T3T4T5121.3ab122.0ab123.8ab118.0b127.3a96.8395.0495.8493.8796.3362.2963.4061.2962.7363.131.501.541.551.511.56

\*Means followed by different letters in the line differ statistically by SNK test at 1% of probability.

<sup>1</sup>T1= control diet + 1% of soybean oil (SO); T2= control diet + 1% SO + 0.012% emulsifier; T3 = control diet + 0.2% of macauba oil (MO) + 0.8% SO + 0.012% emulsifier; T4 = control diet + 0.4% MO + 0.6% SO + 0.012% emulsifier; and T5 = control diet + 0.6% MO + 0.4% SO + 0.012% emulsifier.

Similar results as ours have been reported by Oliveira et al. (2010) who evaluated the inclusion of 3.4% of soybean, sunflower, and linseed oil in diet of laying hens aging 20 to 28 weeks. They did not find effect of different oil source on feed intake, feed conversion, and egg weight.

Ferreira et al. (2015) studying semi-weighted laying hens concluded that the addition of emulsifiers in their diets did not influence the same performance variables measured in our study. Similarly, Costa et al. (2008) indicated that the inclusion of soybean or canola oil up to 3% of the diet of laying hens aged 18 weeks did not influence egg weight and feed conversion per mass of egg produced.

These results were similar to those obtained by Baucells et al. (2000), Grobas et al. (2001), and Galobart et al. (2001), who concluded that the lipid source did not affect the productive performance of laying hens at the beginning of their oviposition.

In our study, we observe different (P < 0.01) FI between the treatments. Animals receiving diet with 0.4% of macauba oil inclusion showed the lowest feed intake. SANTOS et al. (2009) did not find different FI between animals receiving diet with 4% inclusion of soybean, linseed, or cotton oil in their diet. Novak e Scheideler (2001), however, observed higher FI in laying hens receiving diet with the inclusion of linseed oil than receiving control diet.

According to Grobas et al. (2001), laying hens fed diets with soybean oil inclusion laid heavier eggs than those fed diets included with beef tallow, linseed oil, and olive oil. The effect of dietary lipid sources on egg weight observed in young laying hens can be attributed to the composition of unsaturated fatty acids in the diets, which seem to increase the synthesis of lipoproteins and their deposition in the ovum (GROBAS et al, 2001).

On the other hand, Galobart et al. (2001) did not observe a significant effect of adding different lipid sources in the diet of laying hens on egg weight, which is similar to what our data suggests.

In light of the results, macauba oil may be a possible replacement of soybean oil because it did not significantly affect performance variables in our study. Not many studies evaluated the addition of emulsifiers in the diet of laying hens as well as their effects, correct dosages in each phase, the time required for action among others, so further studies are required to verify their usefulness as an ally of the egg producer.

#### CONCLUSION

Macauba oil as partial replacement for soybean oil in diets with emulsifiers for semi-weighted laying hens does not influence internal and external quality of the eggs laid as well as animal performance. We, therefore, suggests the inclusion of 0.4% of macauba oil.

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