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Carbohydrase and phytase supplementation in diets for semi-heavy laying hens

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ABSTRACT. This study was conducted in order to evaluate the association of phytase with an enzymatic complex comprised of carbohydrases (α-galactosidase, galactomannan, xylanase and β-glucanase) in nutrition reduction diets for semi-heavy laying hens and its effect on egg performance and egg quality. Four hundred Isa Brown laying hens with 42 to 57 weeks of age were distributed in an entirely random experiment with five treatments and 8 repetitions, during five production periods of 21 days. Variables studied: egg production, feed intake, mean egg weight, feed conversion, Haugh unit, percentage of yolk, egg white and albumen, yolk color, eggshell thickness and specific gravity. There was a significant interaction ($p < 0.05$) between treatments and experimental periods for feed intake. There were no significant effects ($p > 0.05$) of treatment on production, egg weight or internal and external egg quality. Treatment effects on feed conversion showed better values for hens fed with the control diet. The levels of nutrient reduction used in the diets with or without enzyme supplementation did not provide good results with regard to feed conversion and feed intake. However, they did not affect the other parameters for egg production and internal and external egg quality.

Keywords: enzymes, monogastrics, nutrient availability, performance, egg quality.

Suplementação de carboidrases e fitase em dietas para poedeiras semi-pesadas

RESUMO. O estudo foi conduzido com o objetivo de avaliar a associação de fitase com um complexo enzimático composto por carboidrases (α-galactosidase, galactomananase, xilanase e β-glucanase) em dieta com redução nutricional para poedeiras semipesadas e seus efeitos sobre o desempenho e qualidade de ovos. Foram distribuídas 400 poedeiras Isa Brown de 42 a 57 semanas de idade em delineamento inteiramente casualisado com cinco tratamentos e oito repetições, sendo cinco períodos de produção, com 21 dias cada. As variáveis estudadas foram: produção de ovos, consumo de ração, peso médio dos ovos e conversão alimentar, unidade Haugh, porcentagens de gema, casca e albúmen, cor da gema, espessura da casca e gravidade específica. Houve interação significativa ($p < 0,05$) entre tratamentos e períodos experimentais para o consumo de ração. Não houve efeitos significativos ($p > 0,05$) dos tratamentos sobre produção e sobre as variáveis de peso médio e de qualidade interna e externa dos ovos. Houve efeito dos tratamentos sobre a conversão alimentar, com melhor valor para aves que receberam o tratamento controle. Os níveis adotados de redução nutricional das dietas com ou sem suplementação enzimática não proporcionaram bons resultados no que diz respeito à conversão alimentar e consumo de ração, não afetando, porém, os demais parâmetros produtivos e de qualidade interna e externa dos ovos.

Palavras-chave: enzimas, monogástricos, disponibilização de nutrientes, desempenho, qualidade de ovos.

Introduction

In the field of nutrition, a great deal of research is being conducted in the search for alternatives to enable the formulation of more efficient and economical feeds, enabling better use of nutrients of the ingredients as well as lower feed cost (STRADA et al., 2005).

Diets of laying hens and broilers are mainly based on corn and soybean meal. These ingredients have good digestibility and also an indigestible part that can be leveraged with supplementation of exogenous enzymes in the formulations.

Specific enzymes such as carbohydrases enable the use of fibrous ingredients with higher content of soluble non-starch polysaccharides (NSPs), thus providing better use of energy (MATHLOUTHI et al., 2002). The effect of enzyme supplementation is well studied and established (SILVA; SMITHARD, 2002), and the use of xylanases and b-glucanases has

been shown effective in improving the performance of chickens fed diets containing ingredients such as wheat and barley, which increase viscosity, or even with corn and soybean meal, grains considered as unable to increase viscosity (MATHLOUTHI et al., 2003a and b). Phytase is regularly used in poultry production because it reduces the need for adding inorganic sources of phosphorus to the feed. This way, it also helps to avoid environmental contamination, a major concern in this century.

Several authors have been studying enzyme supplementation with carbohydrases and phytases in diets whose nutrient levels are below energy intake requirements, as recommended by poultry farming guidelines for chicken strains. These enzymes act on antinutritional factors and/or act on complex molecules not previously digested by poultry because of low or non-existent production of enzymes capable of acting on such complexes. There is still considerable controversy over the results of this supplementation in diets whose nutritional values are below the recommendations by animal farming guidelines.

The objective of this study was to evaluate the association of phytase with an enzyme complex consisting of carbohydrases (α-galactosidase, galactomannan, xylanase and β-glucanase) in nutrient reduction diets for semi-heavy laying hens and its effects on egg performance and egg quality.

Material and methods

The experiment was conducted at the Instituto Federal de Minas Gerais - Bambuí campus, from August to November 2012, and used 400 Isa Brown laying hens at 42 weeks of age, housed during egg production in a laying house with a gable roof covered with clay tiles. The experiment was completely randomized (CRD) in a split plot design (five periods of 21 days each), with five treatments and eight replicates per plot. Each plot consisted of five galvanized wire cages, with housing capacity for two hens each, totaling ten hens per plot. The treatments were the following: Positive control feed (PC) – 2,697 kcal ME kg^{-1}, 15.35% CP, 4.37% calcium (Ca), 0.354% available phosphorus (aP), without enzyme addition; Negative control feed (NC): 2,589 kcal ME kg^{-1}, 14.77% CP, 4.39% calcium (Ca), 0.355% available phosphorus (aP) without enzyme addition; T1. Diet with nutritional levels of 2,590 kcal ME kg^{-1}, 14.72% CP, 4.21% calcium (Ca), 0.231% available phosphorus (aP), supplemented with 200 g ton^{-1} carbohydrase and 30 g ton^{-1} phytase with inclusion of 12.7% wheat bran; T2. Diet with nutritional levels of 2,547 kcal ME kg^{-1}, 14.58% CP, 4.26% calcium (Ca), 0.237% available phosphorus (aP), supplemented with 300 g ton^{-1} carbohydrase, with inclusion of 15.9% wheat bran;. T3: Diet with nutrient levels of 2,532 kcal ME kg^{-1}, 14.49% CP, 4.26% calcium (Ca), 0.240% available phosphorus (aP), supplemented with 400 g ton^{-1} carbohydrase and 30 g ton^{-1} phytase, with inclusion of 17.2% wheat bran. The composition and activity of the enzyme carbohydrase used in the supplementation was the following: Alpha-galactosidase: 35 U g^{-1}; Galactomannan: 110 U g^{-1}, Beta-glucanase: 1,100 U g^{-1}; Xylanase: 1,500 U g^{-1} and the enzyme phytase: 10,000 FTU g^{-1}. The experimental feeds were based on corn, soybean meal and wheat meal, and dicalcium phosphate was used as a phosphorus source. Feed composition was based on the recommendations of Rostagno et al. (2005) and the requirements were in accordance with the animal farming guidelines for chicken strains (ISA, 2007). The percentage and calculated compositions of the experimental feeds are shown in Table 1.

The hens were kept under a light regime of 16h day^{-1}. Daily assessments were made of maximum and minimum temperature in the laying house. Feed intake (g hen^{-1} day^{-1}), feed conversion (kg feed kg^{-1} of egg mass) and mean egg weight (g) were assessed. The feeds and eggs were weighed at the end of each week, yielding, at the end of the experimental period, the mean values for feed intake, feed conversion and mean egg weight evaluated for three weeks (cycle or experimental period). The eggs were collected daily (at 12 p.m. and 3 p.m.) to determine egg production (% egg hen^{-1} day^{-1}), and the cracked, broken and abnormal, shelled and soft-shelled eggs were registered. On the last three days of each cycle, specific gravity, eggshell percentage and eggshell thickness were analyzed. Egg specific gravity was obtained by dipping the eggs in different salt solutions with densities of 1,066; 1,070; 1,074; 1,078; 1,082; 1,086; 1,090; 1,094; 1,098; and 1,102 g mL^{-1}. The salt solutions were adjusted with the use of an oil hydrometer and calibrated periodically. After specific gravity was determined, two eggs were collected, weighed and broken. The shells were washed in tap water, dried at room temperature for 48 hours and weighed on an analytical balance (0.01 g precision) to determine eggshell weight and then calculate eggshell percentage. Eggshell thickness was measured with a digital micrometer (Mitutoyo®) by taking four measurements in the central region of the shell, where calcium carbonate crystals are more evenly distributed. Yolk color was measured by comparison with the standard color fan (Yolk Color Fan with comparative scale of 1 to 15 colors). Albumen height was measured to determine the Haugh unit

with a digital caliper (0.02 mm precision) and the yolk was separated from the white and cleaned on paper towel for weighing and subsequent determination of yolk percentage in the egg.

Table 1. Composition of experimental diets for Isa Brown laying hens fed diets supplemented with carbohydrases and phytase in the period of 42-57 weeks of age.

Ingredients	Positive Control[1]	Negative Control[1]	Treatment 1[2]	Treatment 2[3]	Treatment 3[4]
Corn grains (8.26%)	64.750	58.810	58.262	55.937	55.122
Soybean meal (45%)	21.400	17.900	17.200	16.200	15.700
Wheat meal	1.400	10.800	12.700	15.900	17.200
Calcitic lime	10.300	10.400	10.400	10.500	10.500
Dicalcium phosphate (18%)	1.400	1.300	0.600	0.600	0.600
Common salt	0.410	0.410	0.410	0.410	0.410
DL-Methionine (98%)	0.125	0.125	0.130	0.130	0.130
L – Lysine HCl (78%)	0.005	0.035	0.045	0.055	0.060
L – Tryptophan (98%)	0.005	-	0.005	0.005	0.005
L – Threonine (98%)	0.005	0.020	0.025	0.030	0.030
Carbohydrase[5]	-	-	0.020	0.030	0.040
Phytase[6]	-	-	0.003	0.003	0.003
Vitamin premix[7]	0.100	0.100	0.100	0.100	0.100
Mineral premix[8]	0.100	0.100	0.100	0.100	0.100
Total (kg)	100.000	100.000	100.000	100.000	100.000
Calculated nutritional composition[9]					
CP (%)	15.35	14.77	14.72	14.58	14.49
ME (kcal kg^{-1})	2,697	2,589	2,590	2,547	2,532
Digestible lysine (%)	0.692	0.658	0.656	0.649	0.644
Meth + Dig. Cys. (%)	0.580	0.562	0.565	0.560	0.558
Digestible Tryp. (%)	0.166	0.155	0.159	0.158	0.157
Digestible Thre. (%)	0.521	0.499	0.499	0.493	0.488
Sodium (%)	0.17	0.17	0.17	0.17	0.17
Calcium (%)	4.37	4.39	4.21	4.26	4.26
Available phosphorus (%)	0.354	0.355	0.231	0.237	0.240

[1]Positive control (2,697 kcal ME kg^{-1}) and negative control (NC – 2,589 kcal ME kg^{-1}), without enzyme addition. [2] Treatment 1 (T1) - Diet with nutrient levels of 2,590 kcal ME kg^{-1}, 14.72% CP, 4.21% calcium (Ca), 0.231% available phosphorus (aP), supplemented with 200 g ton^{-1} carbohydrase and 30 g ton^{-1} phytase with inclusion of 12.7% wheat bran; [3]Treatment 2 (T2) - Diet with nutrient levels of 2547 kcal ME kg^{-1}, 14.58% CP, 4.26% calcium (Ca), 0.237% available phosphorus (aP), supplemented with 300 g carbohydrase ton^{-1} and ton^{-1} 30 g phytase, including 15.9% wheat bran. [4]Treatment 3 (T3) - Diet with nutrient levels of 2,532 kcal ME kg^{-1}, 14.49% CP, 4.26% calcium (Ca), 0.240% available phosphorus (aP), supplemented with 400 g ton^{-1} carbohydrase and 30 g ton^{-1} phytase, including 17.2% of wheat bran. [5]Composition of the enzyme carbohydrase: alpha-galactosidase: 35 U g^{-1}; galactomannan: 110 U g^{-1}, beta-glucanase: 1,100 U g^{-1}; xylanase: 1,500 U g^{-1}. [6]Activity of the enzyme phytase: 10,000 FTU g^{-1}. [7]Mineral Premix Compound per kg of product: 75,000 mg manganese, 50,000 mg iron, 1,500 mg iodine, 70,000 mg zinc, 8,500 mg copper, 200 mg cobalt. [8]Vitamin Premix Compound per kg of product: Vitamins: A = 800,000 µ, B12 = 1,000 mg, D3 = 2,000,000 µ, 15,000 mg, K3 = 2,000 mg, B2 = 4,000 mg, B6 = 1,000 mg, niacin = 19,900 mg, pantothenic acid = 5,350 mg, folic acid = 200 mg, selenium = 2,500 mg, antioxidant = 100,000 mg. [9]Nutritional composition of ingredients according to the tables designed by Rostagno et al. (2005).

Data were statistically analyzed using the statistical software SISVAR (system for analysis of variance) (FERREIRA, 2000) by testing contrasts with Scheffé's method among the positive control and negative control treatments and the other treatments. For the experimental periods, regression analysis was used.

Results and discussion

There was a significant interaction ($p < 0.05$) between treatments and experimental periods for the variable feed intake. The values are shown in Table 2.

Table 2. Feed intake of Isa Brown hens fed diets supplemented with carbohydrases and phytase in the period 42-57 weeks of age.

Treatments	Feed intake (g hen^{-1} day^{-1})				
	Period (days)				
	21	42	63	84	105
CP	125.3	118.4	120.8	117.7	118.3
CN	130.7^{*1}	124.1^{*1}	125.2	126.2^{**1}	124.1^{*1}
T1	131.9^{*2}	127.5^{**2}	129.0^{**2}	128.8^{**2}	127.7^{**2}
T2	129.7	125.9^{**3}	127.2^{*3}	124.4^{*3}	125.4^{**3}
T3	129.6	122.8	123.5	126.0^{**4}	130.4^{**4*7}
	CV1 = 7.4 CV2 = 2.69				

[1,2,3,4,5,6, 7]Means followed by numbers in the rows are statistically different by Scheffé's test (* $p < 0.05$), according to the proposed contrasts: [1]y = mCP –mCN, [2]y = mCP – mT1, [3]y = mCP –mT2, [4]y = mCP –mT3, [5]y = mCN –mT1, [6]y = mCN –mT2, [7]y = mCN –mT3. CV1 = coefficient of variation for treatments. SMD = significant mean deviation. PC – positive control (2,697 kcal ME kg^{-1}) and negative control (NC – 2,589 kcal ME kg^{-1}), without enzyme addition; T1 - diet with nutritional levels of 2,590 kcal ME kg^{-1}, 14.72% CP, 4.21% calcium (Ca), 0.231% available phosphorus (aP), supplemented with 200 g ton^{-1} carbohydrase and 30 g ton^{-1} phytase with inclusion of 17.2% wheat bran; T2 - diet with nutritional levels of 2,547 kcal ME kg^{-1}, 14.58% CP, 4.26% calcium (Ca), 0.237% available phosphorus (aP), supplemented with 300 g ton^{-1}carbohydrase and 30 g ton^{-1} phytase, with inclusion of 15.9% wheat bran. T3 – Diet with nutrient levels of 2,532 kcal ME kg^{-1}, 14.49% CP, 4.26% calcium (Ca), 0.240% available phosphorus (aP), supplemented with 400 g ton^{-1} carbohydrase and 30 g ton^{-1} phytase with inclusion of 17.2% wheat bran.

It can be observed that feed intake, in general, was higher in hens whose diets had a reduction in nutrient levels (NC, T1, T2 and T3) with or without enzyme supplementation. This result indicates that the levels of enzyme supplementation did not adequately provide the energy levels needed to meet the dietary requirements of the hens. According to the farming guidelines for the strain (ISA, 2007), laying hens respond to energy reduction in the diet with increased feed intake to meet their daily energy needs. The study by Silva et al. (2012) on Isa Brown hens observed no effect of feed energy reduction (approximately 50 kcal kg^{-1} feed) and levels of digestible amino acids in feeds supplemented with carbohydrases and phytase on the feed intake of hens. The researchers concluded that the recovery of nutrients recommended by using the enzyme complex as well as the phytase matrix were technically efficient to maintain egg quality and performance of semi-heavy laying hens in the evaluated conditions. Jalal et al. (2007) did not observe significant effects, either, on feed intake and egg production in hens fed different dietary energy levels (2,810 and 2,900 kcal ME kg^{-1}) with or without phytase supplementation. Those energy levels are much higher than the ones used in this study.

There was no significant interaction ($p > 0.05$) between treatments and experimental periods for

the variables: egg production, feed conversion, mean egg weight, yolk color, percentages of yolk, albumen and shell, shell thickness, specific egg gravity and Haugh unit. There was no significant difference ($p > 0.05$) of the treatments on these variables, except for feed conversion ($p < 0.05$) (Table 3).

Table 3. Egg production (EP), feed conversion (FC), mean egg weight (MEW), yolk color (YC), albumen percentage (AP), yolk percentage (YP), eggshell percentage (ESP), eggshell thickness (ET), specific gravity (SG) and Haugh unit (HU) of eggs of Isa Brown laying hens fed diets supplemented with carbohydrases and phytase in the period of 42-57 weeks of age.

Characteristics	Treatments					
	PC	NC	T1	T2	T3	CV1(%)
EP (%/hen day^{-1})	91.54	91.71	93.36	90.66	92.04	10.13
FC (g feed g egg^{-1})	2.032	2.2135**1	2.1915*2	2.1782*3	2.2229**4	13.10
MEW (g)	63.23	62.54	63.61	63.14	62.31	5.25
YC	7.22	7.06	7.19	7.04	7.18	9.35
AP (%)	64.89	64.76	64.17	64.12	63.13	7.25
YP (%)	25.44	25.36	25.88	26.00	25.18	8.18
ESP (%)	9.64	9.86	9.78	9.91	9.72	5.59
ET (mm)	0.564	0.574	0.566	0.577	0.564	4.71
SG (g mL^{-1})	1.0894	1.0894	1.0894	1.0894	1.0889	0.18
HU	88.59	87.74	86.40	89.60	89.49	5.82

[1,2,3,4,5,6, 7]Means followed by numbers in the rows are statistically different by Scheffe's test (*$p < 0.05$, **$p < 0.01$), according to the proposed contrasts: [1]y = mCP –mCN, [2]y = mCP –mT1, [3]y = mCP –mT2, [4]y = mCP –mT3, [5]y = mCN –mT1, [6]y = mCN – mT2, [7]y = mCN –mT3. CV1 = Coefficient of variation for the treatments. PC – positive control (2,697 kcal ME kg^{-1}) and negative control (NC -2,589 kcal ME kg^{-1}), without enzyme addition; T1- diet with nutritional levels of 2,590 kcal ME kg^{-1}, 14.72% CP, 4.21% calcium (Ca), 0.231% available phosphorus (aP), supplemented with 200 g ton^{-1} carbohydrase and 30 g ton^{-1} phytase with inclusion of 17.2% wheat bran; T2- diet with nutritional levels of 2,547 kcal ME kg^{-1}, 14.58% CP, 4.26% calcium (Ca), 0.237% available phosphorus (aP), supplemented with 300 g ton^{-1} carbohydrase and 30 g ton^{-1} phytase with inclusion level of 15.9% wheat bran; T3 – diet with nutrient levels of 2,532 kcal ME kg^{-1}, 14.49% CP, 4.26% calcium (Ca), 0.240% available phosphorus (aP), supplemented with 400 g ton^{-1} carbohydrase and 30 g ton^{-1} phytase with inclusion of 17.2% wheat bran.

The experimental diets with their reductions in nutrient levels did not affect egg production, and further studies with more drastic nutrient reduction are required for assessment of the results. There was a significant effect of treatments on feed conversion ($p < 0.05$), where the positive control treatment showed better feed conversion compared with the other treatments, as shown by the contrasts. This reduced feed conversion is directly associated with increased intake of hens whose diet had nutrient levels below the recommended requirements of the Isa Brown hens, with or without enzyme supplementation. This shows that the enzymes did not effectively provide energy from the ingredients to meet the dietary needs of the hens. Silva et al. (2012) observed effects of experimental treatments on feed conversion, where the negative control treatments without enzyme supplementation showed worse FC values, confirming the beneficial effect of enzyme inclusion, possibly by the increased availability of energy, minerals and other nutrients from food.

Other researchers (NOVAK et al., 2008; PAN et al., 1998; ZANELLA et al., 1999) also reported benefits of enzyme supplementation on feed conversion, while Scheideler et al. (2005) found no effect of phytase supplementation on feed conversion; they only observed that the lowest energy level in the feed resulted in the worst feed conversion. The results obtained for feed conversion also disagree on those found by Freitas et al. (2000); by using isoproteic diets based on corn and soybean meal, and two energy levels - 2,850 and 2,750 kcal kg^{-1} - with and without the addition of 0.1% of enzymatic complex (alpha-amylase, xylanase and protease), they found no effect on feed conversion of laying hens. Silversides et al. (2006) found no differences in feed conversion values for hens fed diets with different inclusions of phytase and xylanase. In the study by Araujo et al. (2008), there was no significant effect on feed conversion in hens fed with different inclusion levels of wheat bran in their diet with or without supplementation of an enzyme complex.

There was no significant effect ($p > 0.05$) of the experimental treatments on mean egg weight, eggshell percentage and shell thickness, yolk color, percentage of albumen and yolk and egg specific gravity. The results are in agreement with those by Jalal et al. (2007), Sohail et al. (2003) and Scheideler et al. (2005), who also found no effect of either enzyme supplementation or levels of dietary energy, or both, on egg weight. Novak et al. (2008) also found higher egg weight in hens fed enzyme-supplemented diets (amylase, protease and xylanase), and no significant effects were observed on eggshell percentage, whereas egg weight was lower in poultry fed enzyme-supplemented diets. The authors did not provide further information on the cause of this result. Egg specific gravity was not affected by reductions in the levels of Ca and P in diets with phytase supplementation, which confirms the fact that this enzyme effectively makes P and other complexed minerals available in the molecule of phytate present in plant ingredients. Other researchers (ARAUJO et al., 2008; COSTA et al., 2004; JALAL et al., 2007; NOVAK et al., 2008; SOHAIL et al., 2003) also found no effect of enzyme supplementation on egg specific gravity in low-energy and low-phosphorus diets.

Internal egg quality depends, in part, on the presence and stability of the dense layer of albumen, which is given by the protein ovomucin. This quality can be influenced by several factors such as those related to hens (age and genetics), nutrition (raw materials, microingredients) and the environment (temperature, egg storage and

management) (LEANDRO et al., 2005). In this research, nutrient reduction of the diets and the different levels of enzyme supplementation did not affect internal egg quality, which is measured by the Haugh unit, whereas Murakami et al. (2007), when using supplementation of 400 ppm and 500 ppm of enzymatic complex in feeds for laying hens, found that the diet of 400 ppm increased the extent of albumen Haugh Units.

Experimental periods affected the variables egg performance and internal and external egg quality (Table 4).

Table 4. Effects of the experimental periods on egg production (EP), feed conversion (FC), mean egg weight (MEW), yolk color (YC), albumen percentage (AP), yolk percentage (YP), eggshell percentage (ESP), eggshell thickness (ET), specific gravity (SG) and Haugh unit (HU) for eggs of laying hens under different experimental treatments.

	Periods (days)							
Characteristics	21	42	63	84	105	CV2	Effect	R^2
EP (%/hen day^{-1})	91.65	95.20	92.93	93.14	86.41	6.61	Q**	0.8952
FC (g feed g^{-1} egg)	2.160	2.028	2.159	2.1519	2.331	7.83	Q**	0.8326
MEW (g)	62.2	64.3	62.8	62.2	62.8	2.83	T**	0.9134
YC	6.86	7.07	6.90	6.99	7.87	6.83	Q**	0.8056
AP (%)	64.43	62.08	65.39	64.09	65.11	6.97	S*	1.0000
YP (%)	25.58	26.35	24.93	25.81	25.21	7.65	S*	1.0000
ESP (%)	9.87	10.13	9.67	9.58	9.68	5.08	T**	0.8770
ET (mm)	0.5778	0.5477	0.5870	0.5610	0.5718	4.15	S*	1.0000
SG (g mL^{-1})	1.0898	1.0897	1.0884	1.0888	1.0897	0.16	T**	0.8523
HU	89.56	89.80	87.67	83.20	83.20	4.60	T**	0.9189

L - Linear effect, Q - quadratic effect, T – third-degree effect, S - fourth-degree effect. **($p < 0.01$) and *($p < 0.05$); CV2 - coefficient of variation for the periods.

There was a quadratic effect of experimental periods on the following variables: egg production ($p < 0.01$, $y = 86.593286 + 6.486626x - 1.290017x^2$, $R^2 = 89.52\%$), feed conversion ($p < 0.01$ $y = 2.264615 - 0.157423x + 0.034117x^2$, $R^2 = 83.26\%$) and yolk color ($p < 0.01$, $y = 6.976286 - 0.012337x + 0.0002x^2$, $R^2 = 80.56\%$). The remaining variables for the eggs - mean egg weight, albumen percentage, yolk percentage, eggshell percentage, eggshell thickness, egg specific gravity and Haugh unit - showed third-degree and fourth-degree effects, and there is no explanation for such equations.

Conclusion

The association of enzymes with the reduction of nutrient levels did not differ between parameters of production and internal and external quality, except for feed intake and feed conversion, which showed differences.

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