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# Meat quality and performance of broilers fed diets containing selenium yeast and sodium selenite

Abstract – The objective of this work was to evaluate performance parameters, carcass and cut yields, meat quality, plasma glutathione peroxidase (GSH-Px) activity, and selenium tissue deposition for broilers fed with Se from organic (Se-yeast) and inorganic (sodium selenite) sources. A total of 1,200 Cobb 500 chicks, males with one day of age, were randomly distributed into four treatments with ten replicates. The treatments consisted of two Se-yeast levels (3,000 and 2,000 ppm), sodium selenite (45.7%), and the combination between sodium selenite and 3,000 ppm Se-yeast. All diets, based on corn and soybean meal, were supplemented with 0.3 ppm Se. Among treatments, there were no differences for performance, carcass and cut yields, and meat quality. Diets containing only Se-yeast provided meat with lower values of thiobarbituric acid reactive substances. A higher GSH-Px activity was observed with 3,000 ppm Se-yeast and a greater deposition of Se in the muscle tissue with Se-yeast. The sources of Se do not affect performance parameters and carcass yield; however, the organic source 3,000 ppm Se-yeast results in a greater deposition of the mineral in the muscle and in a greater oxidative stability in the meat.

Index terms: glutathione peroxidase, organic selenium, oxidative stability.

## Qualidade da carne e desempenho de frangos de corte alimentados com dietas contendo selênio levedura e selenito de sódio

Resumo – O objetivo deste trabalho foi avaliar parâmetros de desempenho, rendimento de carcaça e cortes, qualidade de carne, atividade plasmática de glutationa peroxidase (GSH-Px) e deposição tecidual de selênio em frangos de corte alimentados com Se de fontes orgânica (Se-levedura) e inorgânica (selenito de sódio). Um total de 1.200 pintos Cobb 500, machos com um dia de idade, foram distribuídos inteiramente ao acaso em quatro tratamentos, com dez repetições. Os tratamentos consistiram de dois níveis de Se-levedura (3.000 e 2.000 ppm), de selenito de sódio (45,7%), e da combinação entre selenito de sódio e 3.000 ppm de Se-levedura. Todas as dietas, à base de milho e farelo de soja, foram suplementadas com 0,3 ppm de Se. Entre os tratamentos, não houve diferenças quanto a desempenho, rendimento de carcaça e de cortes, e qualidade de carne. Dietas apenas com Se-levedura proporcionaram carnes com menores valores de substâncias reativas ao ácido tiobarbitúrico. Observaram-se maior atividade de GSH-Px com 3.000 ppm Se-levedura e maior deposição muscular de Se com Se-levedura. As fontes de Se não afetam os parâmetros de desempenho e o rendimento de carcaça; no entanto, a fonte orgânica 3.000 ppm de Se-levedura resulta em maior deposição do mineral no músculo e maior estabilidade oxidativa da carne.

Termos para indexação: glutationa peroxidase, selênio orgânico, estabilidade oxidativa.

#### Introduction

After microbial deterioration, lipid oxidation is the main cause for decreases in meat nutritive values and physicochemical traits (Ahmad et al., 2012). To maintain meat sensory characteristics, the use of organic sources of selenium has been investigated intensively (Boiago et al., 2014; Oliveira et al., 2014; Li et al., 2018; Silva et al., 2019a).

Selenium stands out as a micromineral that maintains redox status and membrane integrity through the plasma glutathione peroxidase (GSH-Px) enzyme, which protects cells from damage caused by free radicals (Rajashree et al., 2014). One of the components of GSH-Px is selenocysteine, which is responsible for catalyzing the transition from the reduced to the oxidized glutathione, releasing hydrogen molecules that neutralize peroxides harmful to cell membranes (Rover Jr et al., 2001).

In broiler diets, most Se supplementation is derived from inorganic sources, such as sodium selenite, which cost less than the organic ones. However, organic sources of Se are more bioavailable, providing a greater deposition of the mineral in tissues (Dalia et al., 2017) and a greater oxidative stability in meat, which will consequently have a longer shelf life (Cao et al., 2014; Rajashree et al., 2014; Calvo et al., 2017; Silva et al., 2019a). Of the organic sources, Se-yeast is the most used. It is produced by the fermentation of strains of the Saccharomyces cerevisiae yeast, and selenomethionine and selenocysteine are its main components (Kelly & Power, 1995). However, there are differences in the bioavailability of some forms of Seyeast due to variations both in the strains themselves and in their growth conditions (Surai & Fisinin, 2014), as well as to the used Se concentrations, which affect how Se-yeast in animal feed improves meat quality. Therefore, it is necessary to understand how Se concentrations from organic sources can affect meat quality without compromising performance and also to verify the possibility of using organic and inorganic sources simultaneously.

The objective of this work was to evaluate performance parameters, carcass and cut yields, meat quality, GSH-Px activity, and selenium tissue deposition for broilers fed with Se from organic (Seyeast) and inorganic (sodium selenite) sources.

#### **Materials and Methods**

The study was conducted in the municipality of Lavras, in the state of Minas Gerais, Brazil (21°14'43"S, 44°59'59"W, at 919 m above sea level). The experimental procedures were approved by the ethics committee on animal use of Universidade Federal de Lavras, under protocol number 006/16.

For the experiment, 1,200 Cobb 500 chicks, one-day old males weighing  $39.3\pm2$  g, were randomly distributed into four treatments with ten replicates each, totalizing 40 experimental units with 30 birds each. The treatments consisted of Se supplementation using the following organic (Se-yeast) and inorganic (sodium selenite) sources alone and combined: 3,000 ppm Se-yeast, 2,000 ppm Se-yeast, 45.7% sodium selenite, and 0.15 ppm sodium selenite + 0.15 ppm of 3,000 ppm Se-yeast.

Water and feed were provided ad libitum in a pendant and tubular feeder, respectively. A furnace was used for heating, with an automatic temperature control until the broilers were 14 days old. The light program used was 24 hours light:0 hour dark from 1 to 14 days of age and, then, 16 hours light:8 hours dark. The average maximum and minimum temperatures were 30.6 and 15.4°C, respectively, and the average relative humidity was 65%.

In all experimental treatments and in all chick developmental stages, the supplementation level of 0.3 ppm Se approved by National Research Council (NRC, 1994) was adopted.

A nutrition program was implemented from 1 to 22 and from 22 to 42 days of age. The diets were formulated on the basis of corn and soybean meal, following the nutritional requirements recommended by Bertechini (2013). The used mineral and vitamin supplements were free of Se (Table 1).

The following performance parameters were evaluated from 1 to 21 and from 1 to 42 days of age: weight gain, feed intake, and feed conversion rate on the basis of bird weight per pen and of feed (provided and leftover).

At 42 days, two birds per pen were randomly sampled, fasted for 8 hours, slaughtered by cervical dislocation, bled, scalded for 3 min at 54°C, plucked, and eviscerated. After the carcasses were weighed, breasts, wings, legs (thigh and drumstick), and abdominal fat were separated and weighed to determine carcass and cut yields.

Carcass yield was obtained by relating the weight of the eviscerated carcass without feet, head, and neck to the live body weight prior to slaughter. The yield of the breast, leg, and wing cuts, as well as the abdominal fat of the animals, was determined in relation to carcass weight.

To evaluate the quality of breast meat, breasts of two birds per experimental unit were deboned and kept on ice for 90 min and then subjected to physicochemical analyzes. One of the breasts from each pen was used to assess pH, drip loss, and meat color, whereas the other

**Table 1.** Composition of the experimental diet used at different rearing stages of Cobb 500 male chicks.

Ingredient	1-21 days of age	22-42 days of age
Corn (%)	57.121	62.542
Soybean meal (%)	37.018	30.207
Vegetable oil (%)	2.026	3.199
Dicalcium phosphate (%)	1.807	1.870
Limestone (%)	0.148	0.783
Salt (%)	0.482	0.482
DL-methionine (%)	0.238	0.250
L-lysine HCl (%)	0.148	0.234
L-threonine (%)	0.020	0.080
Vitamin premix <sup>(1)</sup> (%)	0.100	0.100
Mineral premix <sup>(2)</sup> (%)	0.100	0.100
Choline chloride 60% (%)	0.040	0.040
Salinomycin 12% (%)	0.050	0.050
Zinc bacitracin 10% (%)	0.025	0.025
$Inert^{(3)}(\%)$	0.040	0.040
Calculated nutrient content (%)		
Metabolizable energy (kcal kg <sup>-1</sup> )	2,961	3,100
Crude protein (%)	21.52	19.00
Methionine (%)	0.54	0.53
Methionine + cysteine (%)	0.89	0.84
Lysine (%)	1.28	1.17
Threonine (%)	0.86	0.82
Tryptophan (%)	0.26	0.23
Arginine (%)	1.44	1.24
Isoleucine (%)	0.93	0.81
Glycine + serine (%)	2.04	1.78
Sodium (%)	0.21	0.21
Calcium (%)	0.85	0.85
Available phosphorus (%)	0.45	0.45
Analyzed selenium <sup>(4)</sup> (ppm)	0.080	0.075

<sup>(1)</sup>Amount per kilogram of feed: 12,000 IU vitamin A, 2,400 IU vitamin D3, 40 mg vitamin E, 31.8 mg vitamin K, 2.5 mg thiamine, 4.0 mg riboflavin, 2.0 mg pyridoxine, 15 µg vitamin B12, 60 µg biotin, 30 mg niacin, and 1.8 mg folic acid. <sup>(2)</sup>Amount per kilogram of feed: 80 mg Fe, 70 mg Zn, 70 mg Mn, 1.0 mg I, and 10 mg Cu. <sup>(3)</sup>Kaolin used for the inclusion of the treatments. <sup>(4)</sup>Analyzed by atomic absorption spectrometry using the 77 VGA hydride generator system (Varian Medical Systems Australasia, Belrose, Australia). one was frozen for further analysis of thiobarbituric acid reactive substances (TBARS), Se deposition, and cooking loss.

Four hours after slaughter, pH was measured using the DM-20 pH meter (Digimed: Instrumentação Analítica, São Paulo, SP, Brazil), by inserting a puncture electrode and temperature calibrating device into the muscle mass of the breast. The pH values for each sample were obtained from the average of three readings (triplicates) in different positions.

For drip loss measurements, two meat cube samples (2.5 cm<sup>3</sup>) per pen were taken from the pectoralis major muscle for analysis according to Rasmussen & Andersson (1996). Each cube was suspended by a stainless steel hook and attached to the lid of a sealed plastic pot to avoid losses to the outside environment; the cube was positioned so that the direction of the fibers was parallel to the bottom of the pot. After being sealed, the pots were placed in a refrigerator at 4°C. After 48 hours, the samples were removed from the refrigerator and weighed to calculate drip loss, expressed as percentage.

Meat color reading was taken on the dorsal surface of the pectoralis major muscle after exposure to room air for 30 min, using the CM-700d colorimeter (Konica Minolta Inc., Osaka, Japan) as described by Ramos & Gomide (2007). For the calculation of the color indices, illuminant A, the angle of 10° for the observer, the excluded specular reflectance, and the CIELAB color system were determined. The color indices for lightness, redness, and yellowness were obtained considering the average value of six readings taken at the cranial, middle, and distal points of the muscle.

For cooking loss determination, the breasts were deboned and the muscles were separated, weighed, and wrapped in aluminum foil. Then, the cuts were cooked in an electric plate previously heated to  $150\pm5^{\circ}$ C. After reaching 35°C, the samples were turned and cooked until reaching an internal temperature of  $72\pm2^{\circ}$ C. After cooking, the aluminum foil was removed and the samples were cooled at room temperature for 30 min. Then, they were weighed again, and cooking loss was obtained as the percentage of the difference between the weights before and after cooking. The samples used to determine the TBARS index were stored for 60 days at -18°C and then thawed in a refrigerator, at 4°C, for 24 hours prior to the analysis, which was carried out according to the methodology of Jo &

Ahn (1998). Approximately 8.0 g of the samples were homogenized in 25 mL distilled water and 0.1 mL 7.2% butylhydroxytoluene using a Turrax-type disperser, at 1,4000 rpm. After homogenization, an aliquot of 1.0 mL was removed from the homogenate and placed in tubes to which a solution of 2.0 mL trichloroacetic acid + thiobarbituric acid was added. The tubes were kept in a boiling water bath for 15 min, cooled in cold water for 5 min, and centrifuged at 3,000 g for 15 min for reading on a spectrophotometer at 532 nm. The concentration of malonic dialdehyde (MDA) per kilogram of sample was determined by multiplying the absorbance reading by a conversion factor of 7.38, which was obtained by a standard calibration curve with 1,1,3,3-tetraethoxypropane.

For the determination of Se levels, a sample of the breast and liver was collected per pen, pre-dried in a forced-ventilation oven, at 65°C, for 72 hours, and then ground. Subsequently, approximately 0.5 to 1.0 g of the dehydrated sample was weighed and placed into a digesting tube containing 10 mL nitric acid. The mixture was kept in a refrigerator for 12 hours. Afterwards, the samples were heated mildly, at 80°C, for 2 hours and then, at 100°C, until the elimination of nitrous vapors. To complete digestion, 2.0 mL hydrogen peroxide were added, and the mixture was diluted to 50 mL for the pre-reduction of Se VI to Se IV using sodium bromide and sulfamic acid; the latter ensures the absence of nitrous compounds that interfere with the atomic absorption measurement. The total Se content was obtained by atomic absorption with hydride generation via quartz tube atomization, at 850°C, using the SpectrAA 200 atomic absorption spectrometer and the VGA 77 hydride generator (Varian Medical Systems Australasia, Belrose, Australia). The used reducing reagent was sodium borohydride (alkaline solution), and the source of electromagnetic radiation was the Hollow Cathode Lamp (Photron, Tokyo, Japan), at a wavelength of 196 nm.

To determine GSH-Px activity, blood samples from the brachial vein of the left wing of the birds were drawn, using disposable needles, directly into tubes that contained heparin and were kept on ice. Blood was collected from four birds per replicate, and each bird represented a measure of the experimental unit. The samples were centrifuged at 1,000 xg, for 10 min, at 4°C. Afterwards, the plasma was removed and stored at -80°C. Enzymatic activity was calculated using the Glutathione Peroxidase Assay Kit (Cayman Chemical, Ann Arbor, MI, USA), following the manufacturer's instructions. The results were expressed in  $\mu$ mol NADPH min<sup>-1</sup> mL<sup>-1</sup> plasma.

The obtained results were subjected to the analysis of variance according to the procedures of the SAS statistical package (SAS Institute, Inc., Cary, NC, USA), and, when necessary, means were compared by the Student-Newman-Keuls test, at 5% probability.

#### **Results and Discussion**

There were no significant effects of Se sources on performance parameters (Table 2) and on carcass, cuts, and abdominal fat (Table 3). This result reinforces that the nutritional requirement of 0.3 ppm Se established for broilers does not affect performance and carcass yield indexes, regardless of whether the used source is organic or inorganic. The results for performance are in agreement with those of other authors (Oliveira et al., 2014; Dalia et al., 2017; Silva et al., 2019b), whereas those for carcass, cuts, and abdominal fat are similar to those of Oliveira et al. (2014). However, these authors used levels of Se-yeast ranging from 0.15 to 0.6 ppm in broiler diets.

The lowest TBARS values in meat were found in the treatments with the inclusion of Se-yeast sources. This result is an indicative that organic sources are more efficient in protecting cell membranes against the potential damage caused by oxidation, contributing to an increased meat shelf life. Additionally, there was no significant difference between Se-yeast sources. Ahmad et al. (2012), Boiago et al. (2014), and Rajashree et al. (2014) also observed that Se-yeast provided lower TBARS values than the inorganic source.

Therefore, Se from organic sources, due to its greater bioavailability, is closely related to the preservation of cell membranes (Boiago et al., 2014; Oliveira et al., 2014; Rajashree et al., 2014; Dalia et al., 2017; Silva et al., 2019b). According to Puvača & Stanaćev (2011), Se maintains cell integrity by neutralizing free radicals, contributing to the decrease in the loss of intracellular fluids.

There were no differences among treatments for drip loss, pH, meat color, and cooking loss (Table 4). However, other authors found positive results for meat quality when comparing organic sources of Se (Seyeast and selenomethionine) with an inorganic source (sodium selenite), at different inclusion levels (Downs et al., 2000; Boiago et al., 2014; Oliveira et al., 2014; Rajashree et al., 2014; Li et al., 2018; Silva et al., 2019b). In the present study, a greater muscle Se deposition was observed in the treatments containing organic sources; however, there was a greater deposition in the treatment with 3,000 ppm Se-yeast than in those with 2,000 ppm Se-yeast and with the combined sources. According to Navarro-Alarcón & López-Martínez (2000), organic Se in the form of selenomethionine and selenocysteine replaces sulfur in analogous molecules of methionine and cysteine, being absorbed through the sites of amino acid absorption, with an optimized deposition in the animal's body when compared with inorganic Se. These results are in alignment with those obtained in other works (Payne & Southern, 2005; Gomes et al., 2011; Boiago et al., 2014; Oliveira et al., 2014; Rajashree et al., 2014; Silva et al., 2019a, 2019b). Similar results were also found by Gomes et al. (2011), who compared two sources of Se-yeast (2,000 and 1,000 ppm) with sodium selenite. These authors concluded that the organic source, regardless of the

**Table 2.** Performance parameters of Cobb 500 broilers fed with organic (selenium yeast) and inorganic (sodium selenite) sources of Se at different days of age<sup>(1)</sup>.

Treatment	1–21 days of age			22–42 days of age			1-4	1–42 days of age		
	WG (g)	FI (g)	FC (g g <sup>-1</sup> )	WG (g)	FI (g)	FC (g g <sup>-1</sup> )	WG (g)	FI (g)	FC (g g <sup>-1</sup> )	
Sodium selenite (SS)	907	126	1.39	2,066	3,431	1.66	2,973	4,849	1.63	
3,000 ppm Se-yeast	894	124	1.39	2,044	3,390	1.65	2,938	4,796	1.63	
2,000 ppm Se-yeast	897	127	1.42	2,076	3,390	1.63	2,973	4,825	1.62	
3,000 ppm Se-yeast + SS	898	128	1.43	2,061	3,430	1.66	2,960	4,864	1.64	
Average	899	126	1.41	2,062	3,410	1.65	2,961	4,834	1.63	
CV (%)	3.23	3.04	2.82	2.96	2.35	1.98	2.67	2.28	1.84	

<sup>(1)</sup>WG, weight gain; FI, feed intake; and FC, feed conversion.

Table 3. Carcass and	d cut yields of Cob	b 500 broilers fee	d with organic	(selenium yeas	st) and inorganic	(sodium selenite)
sources of Se <sup>(1)</sup> .						

Treatment	Carcass	Breast	Legs	Wings	AF
Sodium selenite (SS)	78.38	40.10	26.53	9.30	1.11
3,000 ppm Se-yeast	78.50	39.21	26.82	9.40	1.25
2,000 ppm Se-yeast	78.90	40.45	26.58	8.90	1.18
3,000 ppm Se-yeast + SS	78.12	40.45	26.93	9.23	1.17
Average	78.72	39.77	26.72	9.21	1.18
CV (%)	1.73	4.95	5.42	7.12	23.30

(1)Legs, thigh + drumstick; and AF, abdominal fat.

**Table 4.** Physicochemical characteristics of the breast muscle of Cobb 500 broilers fed with organic (selenium yeast) and inorganic (sodium selenite) sources of Se<sup>(1)</sup>.

Treatment	TBARS <sup>(2)</sup>	DL <sup>(3)</sup>	pH	Meat color <sup>(4)</sup>		CL <sup>(5)</sup>	
	(mg MDA kg <sup>-1</sup> ) <sup>(6)</sup>	(%)		L*	a*	b*	(%)
Sodium selenite (SS)	10.66a	7.65	5.91	52.28	5.20	10.75	20.75
3,000 ppm Se-yeast	7.75b	7.18	5.89	50.74	5.23	10.91	22.84
2,000 ppm Se-yeast	7.81b	6.49	5.93	50.83	5.09	10.81	23.00
3,000 ppm Se-yeast + SS	12.09a	6.01	5.97	51.49	4.71	10.65	21.54
Average	9.58	6.83	5.92	51.33	5.06	10.78	22.03
CV (%)	12.80	34.81	2.49	5.74	20.09	16.76	22.21

<sup>(1)</sup>Means followed by equal letters, lowercase in columns, do not differ by the Student-Newman-Keuls test, at 5% probability. <sup>(2)</sup>Thiobarbituric acid reactive substances. <sup>(3)</sup>Drip loss. <sup>(4)</sup>L\*, lightness; a\*, redness; and b\*, yellowness. <sup>(5)</sup>Cooking loss. <sup>(6)</sup>Milligrams of malonic dialdehyde per kilogram of sample.

Se concentration, provided a greater deposition of Se in the muscle. However, for dietary levels of 0.15 and 0.3 ppm Se, 2,000 ppm Se-yeast provided higher deposition values than the less concentrated Se-yeast, whereas, for the level of 0.45 ppm Se, the two levels of the organic source did not differ statistically regarding the deposition of the mineral in the tissue.

There was no significant difference for Se deposition in the liver (Table 5). However, numerically, the ratio between Se deposition in the liver and muscle is much greater when using sodium selenite (6.82) than 3,000 ppm Se-yeast (3.12). Rajashree et al. (2014) attributes this result to the liver detoxification process, since the inorganic source is less bioavailable than the organic one. For Schrauzer (2003), this is explained by the organism's incapacity to distinguish between selenomethionine and methionine, integrating into selenomethionine some metabolic processes such as the formation of selenocysteine, which is an active component of GSH-Px.

GSH-Px showed a higher activity in the treatment containing only 3,000 ppm Se-yeast and a lower one in the combination of the organic and inorganic sources (Table 5). Payne & Southern (2005), using Se-yeast and sodium selenite, observed a greater GSH-Px activity in the treatment with a greater tissue Se deposition, as in the present study. Birds fed with Se-yeast after Se deprivation are able to maintain the levels of this micromineral through the turnover of proteins containing Se in the breast muscle. Therefore,

**Table 5.** Tissue selenium deposition and glutathione peroxidase plasma (GSH-Px) activity in Cobb 500 broilers fed with organic (Se-yeast) and inorganic (sodium selenite) sources of Se<sup>(1)</sup>.

Treatment	Se in breast	Se in liver	GSH-Px activity
	(mg kg <sup>-1</sup> ) <sup>(2)</sup>	(mg kg <sup>-1</sup> ) <sup>(2)</sup>	(µmol NADPH
			min <sup>-1</sup> mL <sup>-1</sup> )
Sodium selenite (SS)	0.045c	0.307	1.634ab
3,000 ppm Se-yeast	0.107a	0.334	1.644a
2,000 ppm Se-yeast	0.073b	0.355	1.637ab
3,000 ppm Se-yeast + SS	0.071b	0.357	1.588b
Average	0.074	0.338	1.63
CV (%)	19.29	13.69	1.19

<sup>(1)</sup>Means followed by equal letters, lowercase in columns, do not differ by the Student-Newman-Keuls test, at 5% probability. <sup>(2)</sup>Fresh sample.

Se can be recycled, maintaining its levels in the liver and plasma, as well as the GSH-Px activity in these tissues (Payne & Southern, 2005).

The treatment containing 3,000 ppm Se-yeast showed a greater deposition of the mineral in the muscle and a greater GSH-Px activity, indicating a greater bioavailability of Se in comparison with the other treatments.

### Conclusions

1. Organic (selenium yeast) and inorganic (sodium selenite) Se sources do not affect broiler performance, as well as carcass and cut yields.

2. The use of 3,000 ppm Se-yeast promotes the greatest muscle Se deposition and GSH-Px activity.

3. Both 2,000 and 3,000 ppm Se-yeast reduce the formation of peroxides in breast meat, leading to a greater oxidative stability in the meat.

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