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# A by-product of pinto beans in diets for quail in the laying phase

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**ABSTRACT.** The aim of this study was to evaluate levels of inclusion of pinto beans in diets for quail in the laying phase on their performance and egg quality. A total of 280 Japanese quail in the laying phase were distributed into five treatments in a completely randomized design with eight replicates and seven birds per plot. Treatments consisted of five levels (0, 2, 4, 6, and 8%) of inclusion of pinto beans in the diets, which remained isoenergetic and isoproteic. The experiment began with the quail at 70 days of age and lasted 63 days, with their performance assessed every 21 days and egg-quality parameters on the last two days of each cycle. The following parameters were evaluated: final weight, feed intake, egg-laying rate, feed conversion (per dozen eggs and per egg mass), egg weight, eggshell thickness, specific gravity, yolk color, and weights of shell, yolk, and albumen. No difference was detected between the bean inclusion levels for any of the performance and egg-quality traits assessed or the birds' final weight. In conclusion, up to 8% raw pinto beans may be added in diets for Japanese quail in the laying phase without compromising their performance or internal-external egg quality.

**Keywords:** eggs; leguminous; nutrition; by-product.

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## Introduction

The inclusion of waste or by-products from the agro-industry in quail diets may lead to a reduction of feeding costs, making them more compatible with the reality of quail farming, given the partial substitution of corn and soybean meal.

The rising demand for protein sources and their high cost have stimulated research studies to discover new options to replace the traditional protein sources, mainly soybean meal. The common bean (*Phaseolus vulgaris L.*) is one of such alternatives, as it contains considerable amounts of protein, which range from 20 to 30%, and high concentrations of lysine, dietary fibers, complex carbohydrates (60 to 65%), minerals like calcium and especially iron, in addition to B-complex vitamins (Toledo & Canniatti-Brazaca, 2008).

Brazil is considered the largest world producer and consumer of the species *Phaseolus vulgares* (Mechi, Caniatti-Brazaca, & Arthur, 2005). Despite the existence of a regional preference for certain types of grain, beans of the pinto group are the most widely grown in the country, accounting for 70% of the national consumption and 53% of the cultivated area (Lovato et al., 2018). This leguminous species is also resistant to dry spells, which is an important trait for its growth in semi-arid regions.

However, like other legume plants, the pinto bean has anti-nutritional factors such as protease inhibitors (trypsin and chymotrypsin) and hemagglutinins that may reduce the digestibility of nutrients, affecting the animal performance when supplied raw (Alencar et al., 2014). Non-protein anti-nutritional factors (oxalates and phytates) may also be present in these legumes and compromise luminal absorption, given the possible presence of polyphenols and dietary fiber, which have the property of chelating calcium and other minerals (Pereira & Costa, 2002).

For beans to be used in animal feeding, their chemical composition must be first characterized and known so that it can be incorporated into the diet and the desired performance can be achieved (Magalhães et al., 2008). Little research has been done on the use of raw pinto bean in poultry feeding, and the few

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reports are conflicting about the levels of inclusion of this by-product and its effects on animal production. Therefore, the present study was proposed to evaluate the effects of including pinto bean in diets for quail during the laying phase on their production performance and egg quality.

### Material and methods

The experiment was conducted in the Quail Farming Unit of the Department of Animal Science at the Federal University of Alagoas (UFAL), Arapiraca *Campus*, located in the municipality of Arapiraca - AL, Brazil.

This study involved 280 female Japanese quail (*Coturnix japonica*) fed corn- and soybean meal-based diets during the developer and grower phases, as recommended by Rostagno et al. (2011). Upon completing 70 days of age, the quail had an average weight of 176.3 g ( $\pm$  8,6 g) and a laying rate of 90.8% ( $\pm$  5,5 %). Birds were selected to standardize the plots and distributed into five treatments in a completely randomized design with eight replicates and seven quail per plot, totaling 40 experimental units.

Quail were housed in galvanized-wire cages equipped with a trough feeder and a nipple drinker, with an available area of 47 cm<sup>2</sup>/bird. Birds had free access to feed and water.

The lighting program adopted was continuous, providing 17 h of light per, using a timer-type controller. Curtains were managed for the control of ventilation and temperature within the shed. The temperature and relative humidity of the air within the shed were measured using a digital thermo-hygrometer positioned at the height of the birds and located at a height corresponding to the cages. Throughout the experimental period, the average maximum and minimum temperatures were 28.3 and 21.8°C, respectively. The maximum and minimum relative humidity of the air were 85,4 and 50,1%.

The common bean variety carioca to be included in the experimental diets was purchased from a distributor located in Arapiraca, Alagoas, Brazil, resulting from the waste from the bagging of the bean for human consumption. This raw legume was ground and passed through a 2-mm sieve to make the particles uniform for later chemical analysis and inclusion in the experimental diets.

The energy and crude fiber contents in the bean were analyzed according to the methodology described by Silva & Queiroz (2002) presenting 3,786.55 kcal of crude energy kg<sup>-1</sup>, 2,269.35 kcal of metabolizable energy and 6.87% of crude fiber. Dry matter, crude protein, and ether extract were determined adopting the methodology described by Instituto Nacional de Ciência e Tecnologia (INCT, 2012), in which he presented, respectively, 86.70, 22.47 and 2.25%, base on natural matter.

To determine the metabolizable energy, the pinto bean was provided *ad libitum* for three days to 70 quail that were housed in 10 cages. Total excreta were subsequently collected from each experimental unit at an approximate interval of 12 h, over five days. The collected excreta were stored in plastic bags that were identified per replicate, weighed, and preserved in a freezer at  $-5^{\circ}$ C. For the analysis of metabolizable energy, a bomb calorimeter (Parr 1341) was used, as described by Silva and Queiroz (2002).

After the chemical analysis of the bean, the experimental diets were formulated according to the recommendations of Rostagno et al. (2011), considering the requirements for Japanese quail weighing 177 g, in the laying phase. Treatments consisted of a corn- and soybean meal-based control diet and diets including 2, 4, 6, or 8% pinto bean (Table 1).

The experimental period for the collections of production-performance and egg-quality data was 63 days, with three 21-day cycles. The quail's performance was evaluated at the end of each cycle. In case of mortality during the period, the average intake of the plot was corrected according to the methodology described by Sakomura and Rostagno (2016).

Eggs were collected daily, at 07h30, and the average egg production in the period was obtained by recording the number of eggs produced daily — including broken, cracked, and abnormal eggs — and expressed as a percentage over the average number of housed birds. Feed conversion was calculated in two ways: by dividing the average daily feed intake by the average daily production in dozen eggs (conversion as g/dz) and as average feed intake divided by the egg mass (conversion as g/dz)

In the last two days of each cycle, the eggs from each plot were collected and weighed on a 0.01-g precision scale to determine the average weight. Next, eggs were sent for evaluation of specific gravity by immersion in bucket with different saline solutions (NaCl) whose densities ranged from 1.050 to 1.100, with 0.005 intervals.

For the other analyses, the three most homogeneous eggs from each plot were selected and cracked. The yolk was separated from the albumen, its color was determined using a DSM® color fan, and then it was weighed on a precision scale. Shells were washed in running water, left to dry in the shade at room

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temperature for 48h, and weighed after drying. The albumen weight was determined as the difference between the weight of the egg and its parts.

Ingredient			Treatments (%	)	·
	0.0	2.0	4.0	6.0	8.0
Corn	54.88	52.24	51.40	50.00	46.97
Soybean meal	34.92	34.23	33.22	32.54	31.60
Pinto bean	-	2.00	4.00	6.00	8.00
Dicalcium phosphate	1.04	1.06	1.08	1.10	2.20
Limestone	6.65	6.64	6.64	6.67	5.93
Common salt	0.32	0.32	0.32	0.33	0.33
Vegetable oil	1.57	2.84	2.61	3.13	4.00
DL-methionine	0.33	0.34	0.36	0.39	0.40
L-lysine HCL	0.09	0.13	0.16	0.21	0.25
Vitamin premix <sup>1</sup>	0.10	0.10	0.10	0.10	0.10
Mineral premix <sup>2</sup>	0.05	0.05	0.05	0.05	0.05
BHT	0.01	0.01	0.01	0.01	0.01
Choline	0.04	0.04	0.04	0.04	0.04
TOTAL	100.00	100.00	100.00	100.00	100.00
	Nutritional (9	%) and energy (kca	al kg <sup>-1</sup> ) compositio	on	
Met. energy birds	2,800	2,800	2,800	2,800	2,800
Crude protein	20.388	20.388	20.388	20.388	20.388
Calcium	2.909	2.909	2.909	2.909	2.909
Ava. phosphorus	0.303	0.303	0.303	0.303	0.303
Sodium	1.145	1.145	1.145	1.145	1.145
Lipid	4.158	5.347	5.114	5.575	6.389
Total lysine	1.174	1.174	1.174	1.180	1.187
Total met. + cystine	0.951	0.951	0.951	0.962	0.956
Total methionine	0.620	0.629	0.637	0.657	0.662

Table 1. Centesimal and nutritional composition of experimental diets.

 $^1Vitamin\ premix\ kg^{-1};\ vit.\ A\ 13,440.000\ IU;\ vit.\ D\ 3,200,000\ IU\ vit.\ E\ 28,000\ mg\ kg^{-1};\ vit.\ K\ 2,880\ mg\ kg^{-1};\ thiamine\ 3,500\ mg\ kg^{-1};\ riboflavin\ 9,600\ mg\ kg^{-1};\ pyridoxine\ 5,000\ mg\ kg^{-1};\ cyanocobalamin\ 19,200\ mcg\ kg^{-1};\ folic\ acid\ 1,600\ mg\ kg^{-1};\ pantothenic\ acid\ 25,000\ mg\ kg^{-1};\ niacin\ 67,200\ mg\ kg^{-1};\ biotin\ 80,000\ mcg\ kg^{-1};\ selenium\ 600\ ppm;\ antioxidant\ 0.40\ g\ kg^{-1}.\ ^2mineral\ premix\ kg^{-1}:\ Mg\ 150,000\ ppm;\ Zn\ 140,000\ ppm;\ Fe\ 100,000\ ppm;\ Cu\ 16,000\ ppm;\ I\ 1,500\ ppm.$ 

Shell thickness was measured after weighing the shell. Measurements were taken at two points in the center-transverse area of the egg using a caliper with 0.01-mm graduations (Lin et al., 2004). The value defined as shell thickness was obtained as the average of two measurements from each egg.

At the end of the experimental period, all birds were weighed to determine differences in quail weight during the development of the research.

Results were subjected to analysis of variance using the GLM procedure of the SAS software (Statistical Analysis System [SAS], 2003). When significance was detected (p < 0.05), a regression analysis was undertaken to better estimate the level of inclusion of the bean in the diets.

### Results and discussion

In the adult phase, the thermal comfort, or thermoneutral zone, for quail is between 18 and 22 °C, and the relative humidity of the air is between 65 and 70%. During the experiment, the quail were found to be under periods of moderate heat stress.

No difference was detected between the bean inclusion levels for any of the evaluated performance traits or the initial and final weights of the birds (Table 2), showing that in that phase the quail adapted to the diets containing raw bean.

Our results diverge from the findings of Alencar et al. (2014), who included 0 to 20% raw pigeon pea in the diet of late-maturing broilers and observed a decrease in performance only in the finisher stage. Saeed and Abdel Ati (2007) also observed a performance decline when only 5% raw pigeon pea was added to the diet of Ross broilers aged 1 to 42 days. These results indicate low performance in the presence of anti-nutritional factors such as protease and trypsin inhibitors in pigeon pea that may negatively influence the digestibility of the dietary protein and consequently weight gain and feed conversion (Alencar et al., 2014).

The contradictions of this study might have been due to difference in the species used, indicating that quail have a greater tolerance with respect to the anti-nutritional factors of the evaluated legumes and/or

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that pinto bean has a better digestibility when compared with pigeon pea, since the protein digestibility of the raw bean is around 25 to 60% (Batista, Prudêncio, & Fernandes, 2010).

Variable –		Treatments (%)					SEM	CV
	0.0	2.0	4.0	6.0	8.0	Prob.	SEIVI	(%)
FI (g/bird/day)	29.00	26.69	27.63	27.61	27.54	0.09	0.00	5.91
Egg weight (g)	12.00	11.89	11.82	11.85	11.76	0.16	0.05	2.53
Laying rate (%)	86.74	88.96	91.21	89.94	89.54	0.32	0.91	6.45
$FC (g dz^{-1})$	405.84	380.48	380.57	364.40	379.20	0.09	0.00	9.60
FC (g g <sup>-1</sup> )	2.60	2.51	2.54	2.55	2.60	0.66	0.04	9.23
Initial weight (g)	176.73	173.55	172.03	175.80	174.68	0.17	0.00	2.95
Final weight (g)	184.78	181.42	178.20	180.91	181.64	0.09	0.00	3.43

**Table 2.** Mean values for performance variables of laying quail.

FI = feed intake; FC = feed conversion; Prob. = probability; SEM = standard error of the mean; CV = coefficient of variation.

Shaahu, Carew and Dzungwe (2014) evaluated raw lablab bean and several types of processing, at an average inclusion rate of 20% in rabbit diets, and concluded that the processing types had no effect on the performance of the animals. Overall, raw grains have higher contents of anti-nutritional compounds compared with their cooked version, because the protein denaturation — the loss of a protein's three-dimensional structure — tends to improve digestibility as it improves the flexibility of proteins and facilitates the access of proteolytic enzymes. However, excess thermal treatment may reduce the protein digestibility due to the formation of crosslinks (Martín-Cabrejas et al., 2009).

No difference was observed between the treatments for the external and internal quality of the quail eggs (Table 3).

Variable -	Treatments (%)					Prob.	SEM	CV (%)
	0.0	2.0	4.0	6.0	8.0			
Specific gravity	1.074	1.073	1.086	1.072	1.074	0.31	0.00	1.61
Albumen (g)	7.50	7.36	7.28	7.27	7.29	0.13	0.03	2.85
Yolk (g)	3.55	3.59	3.60	3.65	3.55	0.16	0.02	3.53
Shell (g)	0.94	0.94	0.94	0.93	0.93	0.96	0.00	4.26
Shell thickness (mm)	0.228	0.230	0.232	0.228	0.233	0.27	0.00	2.34

**Table 3.** Mean values for the quail's egg quality parameters.

Prob. = probability; SEM = standard error of the mean; CV = coefficient of variation.

These findings corroborate those reported by Fonseca et al. (1995), who concluded that inclusion of raw pigeon pea at up to 10% in diets for layer hens does not affect any egg-quality parameter. However, those authors found that higher levels of the ingredient led to a gradual decrease in performance and egg quality.

The amino acid profile of the proteins from common bean is characterized by a deficiency of tryptophan and sulfur amino acids (methionine and cystine), whereas lysine is the amino acid found in the largest proportion in comparison with the other amino acids (Yin et al., 2010), indicating the need for supplementing these amino acids in diets — mainly poultry diets —, since methionine is the first limiting amino acid. In the present experiment, the synthetic amino acids lysine and methionine were supplemented, minimizing the impacts of an amino acid imbalance the bean might cause on the quail's performance and egg quality.

Possible anti-nutritional factors present in the raw pinto bean did not affect the nutrient utilization, performance, or internal-external egg quality parameters of quail at up to the level of 8% in the diet during 63 days of consumption. Another important factor to be considered is the age at which the birds started receiving the diets; in the present case, the quail were adult (from 70 days of age). Younger birds are less tolerant to these anti-nutritional factors.

However, because there are no known studies on the use of raw beans for quail, it is suggested that new research be conducted over a longer experimental period, since these birds may show later responses to the antinutritional factors. We also recommend experiments with higher levels of inclusion of pinto bean in quail diets.

### Conclusion

Raw pinto bean can be added at up to 8% in diets for Japanese quail in the laying phase without compromising their performance or internal-external egg quality.

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