



## Evaluation of biochemical parameters and productive performance of japanese quail in response to the replacement of soybean meal with canola meal

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**ABSTRACT.** The present study was conducted to evaluate the different levels of canola meal (CM) replacement by soybean meal (SBM) in Japanese quails' diets on productive performance, egg quality traits and blood parameters including blood protein, mineral concentration, liver enzymes and thyroid hormones. In this study, 160 Japanese quails in the second laying phase from 46 to 56 weeks of age were divided into four treatments with four replicates and 10 quails in each per replicate, on a completely randomized design (CRD). Treatments consisted: T1 control (without CM), T2, T3 and T4 replacing 30, 60 and 90% of SBM with CM respectively. The results were shown no significant variation in body weight between treatments in initial and end of the experiment ( $p > 0.05$ ). There was a significant decrease in feed intake by increasing level of CM ( $p < 0.05$ ). Results indicated that significant increase in egg production, egg weight and egg mass in the T1 (control group) and T2 (contained 10% CM) ( $p < 0.05$ ). No significant differences were found in egg quality parameters in the all period of experiment ( $p > 0.05$ ). However, the egg shell thickness showed significantly decreased with increasing levels of canola meal in the diet ( $p < 0.05$ ). According to this study results, 30% replacement of SBM by CM in the diet (contained 10% CM) has no adverse effect on quail's performance.

**Keywords:** Japanese quail, canola meal, egg quality, performance, liver enzymes, soybean meal.

## Avaliação de parâmetros bioquímicos e desempenho produtivo de codornas japonesas como resposta à substituição de farelo de soja por farelo de canola

**RESUMO.** Este estudo avaliou a substituição de diferentes níveis de farelo de canola por farelo de soja nas rações de codornas Japonesas sobre o desempenho produtivo, qualidade de ovos e parâmetros sanguíneos, inclusive proteína sanguínea, concentração mineral, enzimas de fígado e hormônios de tireoide. Cento e sessenta codornas japonesas na fase da segunda postura, de 46 a 56 semanas, foram divididas aleatoriamente em quatro tratamentos, com quatro repetições e 10 codornas por repetição. Os tratamentos foram: T1 controle (sem FC), T2, T3 e T4, substituindo 30, 60 e 90% de FS com FC, respectivamente. Os resultados não revelaram variação significativa no peso corporal entre os tratamentos no início e no final do experimento ( $p > 0.05$ ). Houve uma redução significativa no consumo de dieta com o aumento de nível de FC ( $p < 0.05$ ). Os resultados mostram um aumento significativo na produção de ovos, peso de ovos e massa de ovos em T1 (controle) e T2 (com 10% de FC) ( $p < 0.05$ ). Não houve diferença significativa nos parâmetros de qualidade de ovos durante todo o experimento ( $p > 0.05$ ). Todavia, a espessura da casca dos ovos reduziu-se significativamente com níveis crescentes de canola na dieta ( $p < 0.05$ ). Os resultados mostram que a substituição de 30% em FS por FC na dieta (com 10% de FC) não tem efeitos adversos no desempenho das codornas.

**Palavras-chave:** codorna japonesa, farelo de canola, qualidade de ovos, desempenho, enzimas do fígado, farelo de soja.

### Introduction

Researchers have been concerned over the recent years to find out solutions for poultry feeding which to support high performance and lead to lower feeding costs (Khoramabadi, Akbari, Khajali, Noorani, & Rahmatnejad, 2014). Soybean meal is

the main source of plant protein which is used in poultry feeding by the high protein level and suitable amino acid balanced in particularly the highest lysine digestibility (91%) (Kerley & Allee, 2003). Soybean meal may be replaced in the poultry ration by another protein sources such as canola meal with the lower price and desirable nutrient

content. Canola meal usage in poultry industry has increased rapidly in recent years, due to increases in the amount of canola grown and processed. There is a great potential for increasing the amount of CM used in the poultry diet. In economical aspect, it has a high protein concentration by high digestible. In addition, it is good source of energy, due to high level of fat. Canola is a rapeseed variety superior nutritionally value, with 20-22% crude protein, 40-42% fat, low levels of glucosinolates and uric acid (Huang, Ravindran, & Bryden, 2005). In comparison to the soybean meal, canola meal have a higher level of calcium and phosphorus and fat (Newkirk, 2009). In contrast, the presence of some anti-nutrients in canola meal is the most important limitation to its potential as a protein supplement. Among these compounds, glucosinolates, commonly referred to as goitrogens, are a uniform class of naturally compounds found exclusively in the plant kingdom and only in a limited number of dicotyledonous families (Ebrahimnezhad, Tajaddini, Ahmadzadeh, Aghdamshahryar, & Ghiasigalekandi, 2011). Glucosinolates adversely effects may appeared by liver and kidney function, erythrocyte oxygen-carrying or releasing capacity, respiration rate, feed intake, production and reproductive capacity (Elangoavan, Verms, Sastry, & Singh, 2001). So, Canola meal may negatively influence many physiological processes and a health status by reducing the level of thyroid hormones, decreasing mineral absorption and changing the activities of liver enzymes in the blood of poultry. Canola meal effect on plasma enzymes concentration is not well known. Some researchers showed the changes in liver enzyme activity of blood (Pearson, Greenwood, Butler, & Fenwick, 1983), but some others did not observe (Szymkiewicz, Jan, & Stepinska, 1988). For instance, Kloss et al. (1994) did not find any impact from feeding glucosinolates-extracted crambe meal on GGT, lipase and amylase. There is no agreement among scientists on glucosinolates levels impairing egg laying performance. This can probably be attributed to the source of the glucosinolates (Waldroup, Goodner, & Sloan, 1970). However some researchers have shown that canola meal could be used up to 20% level in the compound feed formulation and replacing by 25-30% of the dietary soybean meal, without significantly adverse effect on the laying performance and egg quality (Georgeta, 2009). The objective of this study was to determine optimum inclusion levels replacing of SBM by CM in the diet and their effects on egg weight, daily feed intake, feed conversion ratio, egg quality and blood parameters in Japanese quail.

## Material and methods

### Animals and treatments

In this study, 160 of Japanese quails in second laying phase were randomly divided into four treatments and four replications with 10 quails in each from 46 to 56 weeks of age. Diets were formulated to supply the nutrient requirements of Japanese quails (National Research Council [NRC], 1994). Experimental diets consisted of: T1) control (without CM), T2, T3 and T4) replacing 30, 60 and 90% of SBM with CM respectively (Table 1).

**Table 1.** Composition and calculated of the experimental diets composition.

Ingredients	Level of soybean meal replaced with of canola meal levels			
	0	30	60	90
Corn	57.523	55.925	54.253	52.582
Soybean meal	33.325	23.322	13.325	3.333
Canola meal	0	10	20	30
Gluten corn	1.322	3.043	4.832	6.635
Oyster Shells	5.720	5.610	5.500	5.400
Dicalcium phosphate <sup>1</sup>	1.030	1	0.970	0.930
NaCl	0.350	0.350	0.360	0.360
Vitamin premix <sup>2</sup>	0.250	0.250	0.250	0.250
Mineral premix <sup>3</sup>	0.250	0.250	0.250	0.250
L-Lysine	0	0.020	0.030	0.050
DL-Methionine	0.230	0.230	0.230	0.230
Total	100	100	100	100
Calculated values				
ME (kcal kg <sup>-1</sup> )	2900	2900	2900	2900
Crude protein (%)	20	20	20	20
Calcium (%)	2.5	2.5	2.5	2.5
Available Phosphorus (%)	0.35	0.35	0.35	0.35
Sodium (%)	0.15	0.15	0.15	0.15
Lysine (%)	1.1	1.1	1.1	1.1
Methionine (%)	0.50	0.50	0.50	0.50
Methionine + Cystine (%)	0.9	0.9	0.9	0.9

<sup>1</sup>Dicalcium phosphate contained: 16.5% phosphorous and 23% calcium; <sup>2</sup>Vitamin premix supplied the following per kg of diet: vitamin A (retinol), 8400 IU; vitamin D3 (cholecalciferol), 1800 IU; vitamin E (tocopheryl acetate), 150 mg; vitamin K, 24 mg; B1, 8 mg; B2, 16.6 mg; B6, 13 mg; B12, 5 mg; pantothenic acid, 12 mg; niacin, 36 mg; biotin, 10 mg; folic acid, 2.2 mg; choline chloride, 128.8 mg; antioxidant, 100 mg; <sup>3</sup>Mineral premix supplied the following per kg of diet: Fe (FeSO<sub>4</sub>, 20.1% Fe), 95 mg; Mn (MnSO<sub>4</sub>, 32.5% Mn), 120 mg; Zn (ZnO, 80.5% Zn), 120 mg; Cu (CuSO<sub>4</sub>, 30.3% Cu), 35 mg; I (KI, 58% I), 5 mg; and Se (NaSeO<sub>3</sub>, 45.5% Se), 2.2 mg.

### Productive parameters

Feed intake was recorded daily in each replicate separately and then calculated per layer, and weekly average egg weight was determined. At the end of the experiment egg mass and feed conversion ratio (FCR) were calculated (g feed/g egg mass).

### Egg quality

Three eggs were randomly selected in each replicate for every two weeks, Albumen and yolk height and width were measured and analyzed by using EQM (Egg Quality Measurements) device (Harms & Russell, 2000). The egg quality traits evaluated including: Special egg weight, albumen

index, yolk index, egg shell thickness and Haughunits (Keener et al., 2006).

### Blood parameters

At the end of the experiment 12 quills in each treatment (three in each replicate) were randomly selected and bled from the brachial vein. Blood samples were taken in no anticoagulant tube for estimating some blood parameters. The bleeding procedure was limited to 1 min or less to minimize the influence of handling stress. Tubes containing blood were centrifuged at 5000 rpm during 15 min. Serum was collected and stored at -20°C for determination of total protein, albumin, uric acid, calcium and inorganic phosphorus by spectrophotometer (Hitachi, 911). Liver enzymes: Alanine Amino Transferase (ALT), Aspartate Amino Transferase (AST), Gamma-Glutamyl Transferase (GGT) and Alkaline Phosphatase (ALP) were measured by Vitros 350 autoanalyser (New York, USA; Product code 680-2153) using their accompanying commercial kits (Vitros Chemistry Products, Ortho-Clinical Diagnostics, Johnson-Johnson Company, New York, USA). Iron (Fe), Zn and Cu were measured (Zhang et al., 2008). Plasma T4 and T3 concentration were determined by Enzymun-Test T4 (Boehinger-Mannheim, Mannheim-Germany) and Enzymun-Test T3, Elisa-Auto T3 respectively (International Reagents Corporation, Kobe, Japan).

### Statistical analysis

Data were analyzed as a completely randomized design (CRD) using the GLM procedure (Statistical Analysis System [SAS], 2002). Duncan's multiple range test was used to compare treatments ( $p < 0.05$ ).

## Results and discussion

### Productive parameters

Treatments had no significant differences in body weight change in the first and end of the experiment ( $p > 0.05$ ). There was significant decrease in feed intake with increasing level of canola meal (Table 2). The decrease in feed intake associated with dietary canola meal has been found by other researchers (Leslie & Summers, 1972). In some studies, feed intake was not affected by the inclusion of CM up to 100 g kg<sup>-1</sup> (Payvastagan, Farhoomand, Shahrooze, Delfani, & Talatpoh, 2012). The reason for decreasing in feed intake should be related to presence of some anti-nutrients in canola meal (McNeill, Bernard, & Macleod, 2004). Other results (Thanaseelaan, 2013) have indicated

that iodine supplementation was effective in ensuring normal thyroid function and offset the toxic effects of glucosinolates. No significant differences were found between treatment 2 and control in laying performance of quills in terms of egg production, egg weight, egg mass and feed conversion ratio (Table 2), and was matched by previous studies (Sadogpan, Johrir, & Reddy, 1983). The reasons could be related to nutrients in rapeseed meal were as available as those in SBM and amino acid especially lysine and methionine levels were also in required range (Elangoavan et al., 2001).

**Table 2.** Effect of different dietary levels of canola meal on productive parameters of Japanese quail in total period of experiment.

	T1	T2	T3	T4	SEM*
Feed intake(g)	37.07 <sup>a</sup>	36.89 <sup>b</sup>	36.48 <sup>c</sup>	35.98 <sup>d</sup>	1.04
Egg Production (%)	72.00 <sup>a</sup>	71.50 <sup>a</sup>	67.10 <sup>b</sup>	66.70 <sup>b</sup>	0.08
Egg weight(g)	11.46 <sup>a</sup>	11.44 <sup>a</sup>	10.93 <sup>b</sup>	10.35 <sup>c</sup>	0.09
Egg mass(g day <sup>-1</sup> )	8.25 <sup>a</sup>	8.17 <sup>a</sup>	7.33 <sup>b</sup>	6.90 <sup>c</sup>	0.46
FCR	4.49 <sup>c</sup>	4.51 <sup>c</sup>	4.97 <sup>b</sup>	5.27 <sup>a</sup>	0.03
Body Weight Change (g)	8.0	7.17	8.02	6.47	0.65

Values in the same row not sharing a common superscript are significantly different ( $p < 0.05$ ). T1) without CM, T2) replacing 30% of SBM with CM (contained 10% CM), T3) replacing 60% of SBM with CM (contained 20% CM), T4) replacing 90% of SBM with CM (contained 30% CM). \*SEM - Standard errors of the mean.

### Egg quality traits

Egg shell thickness and shell percentage significantly decreased by increasing canola meal levels in the diet ( $p < 0.05$ ). Treatments had no significant differences in egg quality parameters (Table 3). Eggshell thickness of quails fed 10% rapeseed meal were higher than other levels of canola meal, while control group had the best eggshell thickness. The results noted that there was not any significant difference in yolk index between dietary treatments.

**Table 3.** Effect of different dietary levels of canola meal on egg quality of Japanese quail in total period of experiment.

	T1	T2	T3	T4	SEM*
Egg shell Thickness (mm)	0.215 <sup>a</sup>	0.203 <sup>b</sup>	0.197 <sup>c</sup>	0.195 <sup>c</sup>	0.003
Yolk Index (%)	36.69	36.95	36.80	37.08	0.30
Albumen Index (%)	6.08	6.23	6.15	6.05	0.097
Haugh units	83.00	82.93	83.23	82.95	0.27
Special Egg weight (g mm <sup>-3</sup> )	1.08	1.08	1.08	1.08	0.007

Values in the same row not sharing a common superscript are significantly different ( $p < 0.05$ ). T1) without CM, T2) replacing 30% of SBM with CM (contained 10% CM), T3) replacing 60% of SBM with CM (contained 20% CM), T4) replacing 90% of SBM with CM (contained 30% CM). \*SEM - Standard errors of the mean.

Lichovnikova, Klecker, and Zeman (2000) have shown the proportion of canola meal in the diets did not affect the haughunit. In contrast other studies (Blair, Robblee, Dewar, Bolton, & Overfield, 2006) have found significant effect of canola meal levels on haugh unit. Some studies have shown that the diet contained 10% canola meal had the best value in egg shell thickness and egg quality, while the other levels rapeseed meal have noted the poorest value

(Lichovnikova, Klecker, & Zeman, 2000; Blair et al., 2006). However, yolk index increased after feeding 20% canola seed in diet (Najib & Al-Khateeb 2004). Some results have stated feeding canola meal up to 20% did not have any adverse effects on egg weight, yolk weight and yolk weight ratio (Gheisari, Ghayor, Egbal-Saeid, Toghiani, & Najafi, 2011). These results suggested that the utilization of nutrients from canola meal diets was similar to soybean meal (Gheisari et al., 2011). The possibility of changes in fatty acid profile of egg yolk consequent to fed canola meal inclusion in layer diets and Japanese quail (Richiter, Lemser, & Andbargholz, 1996).

### Blood parameters

Total protein concentration, albumin, uric acid and urea in serum were not affected by increasing in canola meal level (Table 4). Xu et al. (2011) noted that 10 and 15% canola meal decreased levels of serum uric acid ( $p < 0.01$ ). According to Bielecka, Korol, and Puzio (2006), rapeseeds inclusion into broiler diets results in decreasing the serum uric acid level which can prove the beneficial protein metabolism. However, data from our study did not confirm the results cited above.

**Table 4.** Effect of different dietary levels of canola meal on blood protein concentration of Japanese quail at 56 weeks of age.

	T1	T2	T3	T4	SEM*
Albumin <sup>1</sup>	2.92	2.82	2.79	2.85	0.07
Total protein <sup>1</sup>	4.64	4.67	4.76	4.64	0.11
Uric acid <sup>2</sup>	11.57	11.97	11.29	11.76	0.22
Blood Urea <sup>2</sup>	681.2	570.4	661.7	739.4	0.09

mg day<sup>-1</sup> Lit, 2  $\mu$ g day<sup>-1</sup> Lit; Values in the same row not sharing a common superscript are significantly different ( $p < 0.05$ ); T1) without CM, T2) replacing 30% of SBM with CM (contained 10% CM), T3) replacing 60% of SBM with CM (contained 20% CM), T4) replacing 90% of SBM with CM (contained 30% CM). \*SEM - Standard errors of the mean.

The concentration of plasma enzymes of laying quills was unaffected by canola meal (Table 5).

**Table 5.** Effect of different dietary levels of canola meal on liver enzyme (Unit/ Lit) of Japanese quail at 56 weeks of age.

	T1	T2	T3	T4	SEM*
ALT	14.78	14.82	14.72	14.84	0.49
AST	168.37	170.63	168.69	171.93	3.82
ALP	421.12	416.68	427.08	427.67	9.86
GGT <sup>1</sup>	4.38	4.33	4.42	4.43	0.03

Alanine Amino Transferase (ALT), Aspartate Amino Transferase (AST), Alkaline Phosphatase (ALP), Gamma-GlutamylTransferase (GGT); Values in the same row not sharing a common superscript are significantly different ( $p < 0.05$ ); T1) without CM, T2) replacing 30% of SBM with CM (contained 10% CM), T3) replacing 60% of SBM with CM (contained 20% CM), T4) replacing 90% of SBM with CM (contained 30% CM). \*SEM - Standard errors of the mean.

Levels of plasma AST, ALT and GGT positively correlated with increased indicative of hepatic damage (Mondal, Das, Biswas, Samanta, & Bairagi, 2007). Thompson and Todd (1994) reported that serum AST activity begins to rise during the pre haemolytic period.

So it can be conclude that replacing 30% CM with SM could not able to produce any dystrophy in hepatic or others tissues containing these enzymes and signified that the birds were apparently healthy during the feeding trail. Some researchers have shown the changes in liver enzyme activity of blood (Pearson et al., 1983), but others did not observe this reaction (Payvastagan et al., 2012), for instance, they did not find any impact from feeding glucosinolates-extracted canola meal on AST, GGT, lipase and amylase.

Serum minerals (Table 6) were not affected by increasing in canola meal (Payvastagan et al. 2012). Leeson, Atteh, and Summers (1987) have shown that the content of serum total phosphorus and calcium were higher for birds fed canola meal rather than SBM. Leeson et al. (1987) reported that even 20% of dietary full-fat canola had no significant effect on calcium and phosphorus retention in broiler chickens. On the other hand, Talebali and Farzinpour (2005) state that the existence of phytic acid in canola seeds can cause reduction in calcium ability absorption and consequently, the feed consumption reduction.

**Table 6.** Effect of different dietary levels of canola meal on serum mineral Japanese quail at 56 weeks of age.

	T1	T2	T3	T4	SEM*
Ca <sup>1</sup>	11.63	11.24	11.00	10.88	0.70
P <sup>1</sup>	6.40	6.49	6.64	6.28	0.12
Fe <sup>2</sup>	92.14	93.05	93.56	92.76	1.17
Zn <sup>2</sup>	175.91	174.85	177.69	176.67	1.49
Cu <sup>2</sup>	21.32	20.63	21.38	21.21	0.57

mg day<sup>-1</sup> Lit, 2  $\mu$ g day<sup>-1</sup> Lit; Values in the same row not sharing a common superscript are significantly different ( $p < 0.05$ ); T1) without CM, T2) replacing 30% of SBM with CM (contained 10% CM), T3) replacing 60% of SBM with CM (contained 20% CM), T4) replacing 90% of SBM with CM (contained 30% CM). \*SEM - Standard errors of the mean.

Thyroid hormone concentrations were not affected by treatments (Table 7). The concentration of T3 and T4 were determined in order to obtain information on possible effects of isothiocyanates on the diets containing canola meal. Current results have shown that no significant differences were due to treatments ( $p > 0.05$ ) in this respect. It may be related to amount of isothiocyanates in canola meal used in this study. Therefore, this may be a major reason why T3 and T4 concentration unchanged. However previous reports stated consumption of isothiocyanates is known to cause decreased levels of thyroid hormones (Xu et al., 2011).

In economical aspect, in this experiment the maximum net returns noted in quills fed diet containing 10% CM, as a substitution for SBM, followed by quills fed diets with 0, 20 and 30% CM. Naseem, Khan, and Yousaf (2006) reported decreased average diet cost with increasing dietary

canola meal. Tadelles, Kijora, and Peters (2003) also reported the highest net benefit at 28% inclusion level of RSM as a replacement for Noug seed cake. Decreased net income at higher level of CM inclusion, regardless of its cheaper cost as compared to other ingredients is due to the adverse effects of CM on feed intake, egg production, egg weight, egg mass and feed conversion efficiency.

**Table 7.** Effect of different dietary levels of canola meal on Thyroid hormone (mmoL Lit<sup>-1</sup>) of Japanese quail at 56 weeks of age.

	T1	T2	T3	T4	SEM*
T3	4.32	4.18	4.04	4.14	0.14
T4	132.05	129.72	127.75	130.90	2.32
T3/T4	30.73	31.09	32.01	31.99	1.11

Values in the same row not sharing a common superscript are significantly different ( $p < 0.05$ ). T1) without CM, T2) replacing 30% of SBM with CM (contained 10% CM), T3) replacing 60% of SBM with CM (contained 20% CM), T4) replacing 90% of SBM with CM (contained 30% CM). \*SEM - Standard errors of the mean.

## Conclusion

In conclusion, the CM could be used up to 10% (replacing up to 30% of the soybean meal) in laying quails diets. However higher proportions of CM may be impaired production performance. This study results were shown the egg shell thickness decreased by increasing levels of canola meal in the diet. However, high levels of CM were not affecting egg quality and blood parameters including minerals, liver enzymes and thyroid hormone.

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