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Effect of Dietary Inclusion of Cassava Yeast as Probiotic Source on Egg Production and Egg Quality of Laying Hens

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Abstract: The study was conducted to investigate the effect of dietary inclusion of cassava yeast as a probiotic source on laying hens performance and egg quality. Two hundred and sixteen Roman breed laying hens (26 week of age), were used. The laying hens were randomly allocated to 24 pens containing 9 laying hens each with 6 replicates and assigned to receive one of 4 dietary treatments (1. Control, 2. *S. cerevisiae* 1×10^6 organisms/kg, 3. *S. cerevisiae* 1×10^7 organisms/kg, 4. *S. cerevisiae* 1×10^8 organisms/kg) in a completely randomized design. The results showed that feed intake, feed conversion efficiency, albumin weight, yolk weight and haugh unit were not significantly different among treatments ($p > 0.05$). Significant differences were observed in egg production, egg weigh and shell thickness. Cassava yeast as probiotic source had positive effect on egg weigh and shell thickness, but has negative effect on egg production. The results of the present experiment showed that dietary inclusion of cassava yeast as a probiotic to laying hens seems to have minimal influence on laying hens performance.

Key words: Probiotic, cassava yeast, egg production, egg quality, laying hens

INTRODUCTION

Probiotics are live microorganisms that, when administered through the digestive tract, has a positive impact on the host's health and production. Generally, probiotics are derived bacteria, fungi and yeast. *Saccharomyces cerevisiae*, one of the most widely commercialized types of yeast, has long been fed to animals. Live yeast addition to animal feed has been known to improve the nutritive quality of feed and performance of animals (Matin *et al.*, 1989). In addition, mannan oligosaccharides and fructo-oligosaccharide derived from the cell wall of the yeast *S. cerevisiae*, has shown promise in suppressing enteric pathogens and modulating the immune in studies with poultry (Santin *et al.*, 2001; Spring *et al.*, 2000; Iji *et al.*, 2001). In addition, to egg quality, there are trials showing that enrichment of diets with yeast could favorably improve the egg weigh shell weight yolk weight and height in pullets (Ayanwale *et al.*, 2006) and improve shell thickness in laying hens (Yousefi and Karkoodi, 2007).

Cassava yeast is a naturally produced live yeast culture of *S. cerevisiae* together with its growth medium of cassava, soybean, sugar cane molasses, urea, $MgSO_4 \cdot 2H_2O$, KH_2PO_4 and citric acid. Cassava yeast are composed of natural concentrate mixture of essential nutrients, live cell of yeast, mannan oligosaccharide derived from cell wall of dead cell and vitamin B-complex. This work therefore, has the objective of evaluating the supplemental effect of cassava yeast (*S. cerevisiae* plus the growth medium) on egg production and egg quality in laying hens.

MATERIALS AND METHODS

Preparation of cassava yeast: Cassava chip and soybean meal were grounded using hammer mill and the both flour were subjected to fermentation. Pure strain of *S. cerevisiae* was sub-cultured into 730 mL nutrient solution [yeast (5 g), sugar cane molasses (24 g), urea (10 g), $MgSO_4 \cdot 2H_2O$ (7 g), KH_2PO_4 (13 g) and citric acid (20 g)] and inoculated into 1 kg of flour [cassava (900 g) and soybean (100 g) and then allowed to ferment for 15 h. The product obtained was subsequently dry at room temperature. Therefore, cassava yeast composed of yeast (*S. cerevisiae*) and medium on which it was grown. Dried cassava yeast sample was analyzed in laboratory chemically for proximate constituents (AOAC, 1990). Viable cell counts on product was estimated using the methylene blue staining method (Lindegren, 1949). This product was containing, dry matter 88.28%, ash 8.69%, crude protein 9.94%, crude fiber 5.61%, ether extract 0.42%, nitrogen free extract 75.34% and living cell of yeast 1.23×10^6 organisms/g.

Experimental laying hens: Two hundred and sixteen Roman breed laying hens were used in this experiment, which were 26 weeks of age and continued for two 28 day periods. Nine hens were grouped house and shared a common feed trough between them, forming one experimental unit. There were 6 experimental units for each of the 4 treatment groups following completely randomized design. The experimental house was evaporative cooling system. Experimental laying hens were maintained under standard management conditions.

Experimental diets: Four isonitrogenous and isocaloric laying hen diets in the form of mash were prepared (NRC, 1994). The different of cassava yeast such as 0, 0.08, 0.8 and 8.1% were used in laying hens diets (Table 1) fed to experimental groups T1, T2, T3 and T4, respectively. The control diets had no *S. cerevisiae* while diets T2, T3 and T4 contained *S. cerevisiae* at 1×10^6 , 1×10^7 , 1×10^8 microorganisms/kg. Broken rice, yellow maize, vegetable oil, poultry meat meal and soybean meal served as main energy and protein sources. Each diet was analyzed as described methods in AOAC (1990).

Parameter measured: Egg production was recorded daily. Feed intake was determined at the end of each week. On day 26, 27 and 28 of each period eggs were collected to measure egg weight, shell thickness, yolk weight, albumin weight and haugh unit.

Statistical analyses: All data obtained from the trials were subjected to the analysis of variance procedure of statistical analysis system (SAS, 1996) according to a completely randomized design. Means were separated by Duncan New's Multiple Range Test. The level of significance was determined at $p < 0.05$.

RESULTS AND DISCUSSION

Chemical composition of dietary treatment: The feed ingredients used in the experiment and chemical composition of dietary treatment are shown in Table 1. The ingredients of all treatments were the same, but different in levels of cassava yeast. All treatments had a similar chemical composition

Feed intake and production performance: Feed intake and production performance are shown in Table 2. During period 1 feed intake were significantly ($p < 0.05$) affected by cassava yeast level. Comparatively lower feed intake was observed in chicken fed diet with *S. cerevisiae* 1×10^8 organisms/kg compared to other diets. However, average feed intake was not significant different ($p > 0.05$) among treatments. The results are in agreement with previous studies in laying hens (Mohiti Asli *et al.*, 2007; Ayanwale *et al.*, 2006; Yousefi and Karkoodi, 2007) and broilers (Chumpawadee *et al.*, 2008; Karaoglu and Durdag, 2005) that observed feed intake was not affected by yeast inclusion the diet. Additionally, feed conversion efficiency was not significant different among treatment. These results are in agreement with those of Ergun *et al.* (2000); Mutus *et al.* (2006) who reported that the supplement of a probiotic did not have any effect on feed conversion ratio. Yousefi and Karkoodi (2007) also reported that feed conversion ratio was not affected by the dietary probiotic and yeast supplementation. In addition Chumpawadee *et al.* (2008) could not detect any difference in the feed

Table 1: Ingredients composition (%DM) of laying hen diets containing different levels of cassava yeast

Ingredients	Diets ¹			
	T1	T2	T3	T4
Broken rice	9.50	9.50	9.50	9.50
Yellow maize	45.00	45.00	44.20	37.90
Vegetable oil	4.00	4.00	4.00	4.00
Soybean meal (42% CP)	26.00	26.00	26.00	25.00
Poultry meat meal	3.00	3.00	3.00	3.00
Cassava yeast	0.00	0.08	0.81	8.13
Dicalciumphosphate	4.00	4.00	4.00	4.00
Limestone	7.50	7.50	7.50	7.50
Salt (NaCl)	0.25	0.25	0.25	0.25
Premix ²	0.50	0.50	0.50	0.50
DL-Methionine	0.25	0.25	0.25	0.25
Chemical composition				
Dry matter, %	93.43	92.32	94.75	93.26
Ash, %	7.84	7.37	7.59	7.60
Crude protein, %	18.77	18.77	18.47	18.04
Crude fiber, %	5.86	3.70	3.32	4.09
Ether extract, %	3.38	2.39	3.56	3.97
Calcium, %	6.41	6.30	6.64	6.18
Phosphorus, %	1.49	1.51	1.66	1.77
Lysine ³	1.00	1.00	1.00	1.00
Methionine and cystine ³	0.99	0.99	0.99	0.99
Metabolizable energy (Kcal/kg) ³	2908.00	2910.76	2909.24	2925.12

¹T1: 0% cassava yeast, T2: 0.08 % cassava yeast, *S. cerevisiae* 1×10^6 microorganisms/kg, T3: 0.8 % cassava yeast, *S. cerevisiae* 1×10^7 microorganisms/kg, T4: 8.1 % cassava yeast, *S. cerevisiae* 1×10^8 microorganisms/kg, ²calculated value, ³composition of premix: AD₃(500/100) = 2 g, riboflavin = 0.5 g, thiamin = 1.0 g, niacin = 5.0 g, D-calcium pantothenate = 1.0 g, choline chloride(50%) = 33 g, vitamin B12 (1000mg/kg) = 0.01 g, vitamin E50 (50%) = 2.0 g, vitamin K3 (51%) = 0.50 g, pyridoxine (96%) = 0.30, MnSO₄.5H₂O = 21.60 g, FeSO₄.7H₂O = 27.03 g, CuSO₄.5H₂O = 1.96 g, NaSeCO₃ (1%) = 0.20 g, COCl.6H₂O = 0.30, ZnO = 6.85 g, KI = 0.13 g, rice bran = 396.62 g.

conversion ratio of the broilers as compared to the control. Some studies show that probiotics supplementation in the feed of chickens improve the feed conversion ratio (Silva *et al.*, 2000; Day, 1997). Hen-day egg production was significant different ($p < 0.05$) among treatments. The dietary inclusion cassava yeast negatively affected egg production. The result disagree with Mutus *et al.* (2006); Ayanwale *et al.* (2006); Yousefi and Karkoodi (2007) that observed egg production was not affected by dietary inclusion *S. cerevisiae*. Additionally, Mohiti Asli *et al.* (2007) reported that egg production of laying hen fed dietary supplementation of yeast during heat stress was not significant different with control. The reason for the variable effect of biological additives may be confounded by variations in gut flora and environmental condition (Mahdavi *et al.*, 2005). Several researchers reported that when chicks were housed in a clean environment a probiotic was unaffection on performance (Gunal *et al.*, 2006; Aderson *et al.*, 1999).

Table 2: Feed intake and production performances of laying hen fed diets containing different levels of cassava yeast

Criteria	Diets*				SEM
	T1	T2	T3	T4	
Feed intake, (g/hen/day)					
Period 1(26-29 week)	98.50 ^a	97.92 ^a	99.42 ^a	94.59 ^b	0.63
Period 2 (30-33 week)	98.88 ^{ab}	97.21 ^b	99.10 ^{ab}	100.89 ^a	0.51
Average (26-33 week)	98.69	97.57	99.26	97.74	0.42
Feed conversion efficiency (g feed/g egg)					
Period 1(26-29 week)	2.31	2.34	2.53	2.39	0.03
Period 2 (30-33 week)	2.22	2.35	2.29	2.18	0.04
Average (26-33 week)	2.26	2.34	2.41	2.29	0.03
Hen-day Egg production, (%)					
Period 1(26-29 week)	86.14 ^a	82.19 ^{ab}	79.16 ^{bc}	76.43 ^c	1.09
Period 2 (30-33 week)	83.63 ^a	76.54 ^b	79.10 ^{ab}	83.46 ^a	1.21
Average (26-33 week)	84.88 ^a	79.37 ^b	79.13 ^b	79.95 ^b	0.92

*T1: 0% cassava yeast, T2: 0.08 % cassava yeast, *S. cerevisiae* 1x10⁶ organisms/kg, T3: 0.8 % cassava yeast, *S. cerevisiae* 1x10⁷ organisms/kg, T4: 8.1 % cassava yeast, *S. cerevisiae* 1x10⁸ organisms/kg.

Table 3: Egg quality of laying hen fed diets containing different levels of cassava yeast

Criteria	Diets*				SEM
	T1	T2	T3	T4	
Egg weight, (g)					
Period 1(26-29 week)	56.27 ^b	58.03 ^b	57.12 ^b	58.17 ^a	0.30
Period 2 (30-33 week)	57.67 ^c	58.83 ^b	58.19 ^{ab}	59.65 ^a	0.22
Average (26-33 week)	56.97 ^c	58.43 ^{ab}	57.66 ^{bc}	58.91 ^a	0.24
Shell thickness (mm)					
Period 1(26-29 week)	0.32 ^{ab}	0.35 ^b	0.29 ^b	0.35 ^a	0.01
Period 2 (30-33 week)	0.32	0.32	0.32	0.33	0.01
Average (26-33 week)	0.32 ^{ab}	0.33 ^{ab}	0.30 ^b	0.34 ^a	0.01
Yolk weight (g)					
Period 1(26-29 week)	15.30 ^a	15.94 ^a	16.69 ^a	11.61 ^b	0.78
Period 2 (30-33 week)	15.19	14.47	16.42	15.32	0.45
Average (26-33 week)	15.24	15.21	16.56	13.47	0.51
Albumin weight (g)					
Period 1(26-29 week)	30.70	30.73	31.42	34.41	1.00
Period 2 (30-33 week)	31.98	31.90	31.21	32.95	0.72
Average (26-33 week)	31.34	31.32	31.32	33.68	0.63
Haugh unit					
Period 1(26-29 week)	86.00	85.83	84.00	82.83	1.34
Period 2 (30-33 week)	83.50	88.50	84.33	83.83	0.97
Average (26-33 week)	84.75	87.16	84.16	83.33	0.90

*T1: 0% cassava yeast, T2: 0.08 % cassava yeast, *S. cerevisiae* 1x10⁶ organisms/kg, T3: 0.8% cassava yeast, *S. cerevisiae* 1x10⁷ organisms/kg, T4: 8.1% cassava yeast, *S. cerevisiae* 1x10⁸ organisms/kg.

Egg quality: Egg qualities are shown in Table 2. Egg weight and shell thickness were significantly different ($p < 0.05$) among treatments. The results are in agreement with previous studies in pullets (Ayanwale *et al.*, 2006), laying quails (Ayasan *et al.*, 2006) that observed egg weight and shell thickness were affected by dietary inclusion probiotic. However, these observations disagree with those of Mohiti Asli *et al.* (2007) and Yousefi and Karkoodi (2007) who reported that egg weight were not affected by *S. cerevisiae*. In current experiment egg weight in all treatment was higher than control and this showed the positive effect of cassava yeast inclusion the diet of laying hen. Supplementation of the feeds with 8.1% cassava yeast improved egg weight due to improvement of FCE (Table

2). Furthermore, egg shell of hens fed T4 thicker than that of hens fed control diet. Thayer *et al.* (1978) found that yeast culture increased organic phosphorus utilization. The increased mineralization of eggs by yeast might be responsible for the high shell thickness observed in laying hens. Generally, poultry performance will be improved by adding *S. cerevisiae* to diets, because this product reduces stress responses of birds by increasing vitamin absorption, synthesis of enzyme and protein metabolism (Chumplen *et al.*, 1989). In contrast, (Hosseini *et al.*, 2006; Mohiti Asli *et al.*, 2007) reported that addition of yeast in layer hen diet had not positive effect on egg shell thickness. Inclusion of cassava yeast had no difference significant ($p > 0.05$) in yolk weight and albumin weight. Yousefi and

Karkoodi (2007); Ayanwale *et al.* (2006) reported that addition of yeast in layer hen diet had no effect on albumin weight, but had positive effect on yolk weight. Haugh unit is major indicator determining egg quality and did not change by dietary treatments. These findings were in agreement with Mohiti Asli *et al.* (2007) who observed that haugh unit was not affected by dietary inclusion of yeast.

Conclusion: The results of present study showed that dietary inclusion of cassava yeast as probiotic source had positive effect on egg weight and shell thickness of laying hens. In contrast, a dietary inclusion of cassava yeast has negative effect on egg production. Therefore, probiotic supplement of laying hens maybe more helpful during periods of nutritional and other stress, but under normal environmental and management condition it seems to have minimal influence on performance and egg quality.

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