ORIGINAL RESEARCH

THE EFFECTS OF PROBIOTIC, PREBIOTIC AND SYNBIOTIC TREATMENTS ON EGG PRODUCTION AND EGG CHARACTERISTICS IN BROWN NICK HENS DURING THE LAST STAGE OF PRODUCTION IN WINETR SEASON

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ABSTRACT

The egg production trajectory of laying hens starts with the age of sexual maturity, and then ascends the peak, followed by regression in the last phase of production cycle in conjunction with quality deterioration making the enterprise commercially unsustainable to the breeder. It is more pronounced in winter season due to non-availability of required metabolic energy for maintenance of body temperature and sustenance of egg production. This study attempts to moderate it through administration of nutritional supplements, such as probiotic (Bio-plus2B®), and prebiotic (Techno Mos®) individually or in combination (synbiotic), focusing on the evaluation of (1) Hen-day egg production (%), egg weight (g) and daily egg mass (g); (2) Body weight (g) and body weight gain (g); (3) Feed intake (g/hen/day) and feed efficiency (feed intake/egg mass); (4) Egg quality in terms of egg weight (g), egg shape index (%), albumen (%), yolk (%), yolk/albumen ratio and haugh unit; (5) Shell quality in terms of shell weight (g), shell (%), shell thickness (mm), shell surface area (cm^2), SWUSA (mg/cm^2), and shell density (g/cm^3); (6) Chemical composition of inner egg components (yolk & albumen) in terms of protein (%), fat (%) and GE (kcal/kg), and (7) Economic efficiency of feed (%) and net return (LE.) /kg eggs in eighty H&N Brown Nick laying hens in their last phase of production (65-74 weeks of age) under winter conditions in south Sinai desert of Egypt. The hens were randomly divided into 4 equal treatment groups with 10 replicates per treatment and 2 hens per replicate. These treatments were T1 (control), T2 (Probiotic: 1g Bio-plus2B® / 1kg diet), T3 (Prebiotic: 1g Techno Mos® / 1kg diet) and T4 (Synbiotic: 1g Bio-plus2B® along with 1g Techno Mos® / 1kg diet). The results indicated positive and significant ($P<0.001$) superiority of the experimental treatments particularly the probiotic (T2) treatment on hen-day egg production (%) and daily egg mass (g), over the control group. The improvement in feed efficiency in terms of feed intake / daily egg mass were significantly ($P<0.001$) higher in T2, T3 and T4 groups compared to the control (T1) indicating the advantage of supplements. The gross energy content (kcal/kg) of the inner egg components (yolk & albumen) for hens fed on probiotic containing diet (T2) was higher ($P<0.05$) than other three treatments (T1, T3, T4). Feed cost / kg eggs (LE. 12.67) was the lowest in probiotic (T2) group, while the net return / kg eggs (LE. 6.33) was the highest in this group (T2) compared to the other three groups (T1, T3, T4). It is concluded that dietary supplementation of probiotic (1g Bio-plus2B® / 1kg feed) excelled other treatments in enhancing egg production (%), egg mass (g), gross energy (GE) content (kcal / kg) of the inner egg components, feed efficiency (g), economic efficiency of feed (%) and net return (LE.)/1kg eggs in Brown Nick hens during the last stage of egg production in winter season.

KEY WORDS

Bio-plus2B®, Egg production, Egg quality, Laying hens, Techno Mos®

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INTRODUCTION

Hen-day egg production of laying hens (CP 909) has been reported to increase to 87.45% in the second phase from 18.11% in the early phase, which subsequently declines to 44.75% in the final phase (Arifin, 2016). The quality of egg (Leeson, 2006) is focused externally on its shell, while the internal quality primarily refers to albumen and yolk. Adequate dietary calcium and phosphorus has been found to improve egg production, shell weight and shell thickness, besides decreasing mortality, particularly in the winter season, where the spread of diseases and pathogenic bacteria decline the immunity of hens and their ability to produce more eggs. Moreover, it has been experimentally proved that layers under cold conditions loose more energy to maintain their body temperature. Due to their reduced feed intake capacity, hens are not capable of increasing the availability of the metabolic energy required to maintain their body temperature and egg production, consequently resulting in economic losses (Alves et al., 2012).

In addition, thickness of the egg shell weakens due to the large size of eggs produced during the final stage of production cycle (Bar et al., 2002), due to decrease in the efficiency of older hens in absorbing calcium than the younger ones (Al-Batshan et al., 1994). Hens' eggs, particularly in the final stage of production cycle, exhibit deterioration of egg shell quality in terms of production of higher numbers of broken and defective eggs due to changes in the bird's body related to vitamin D3 metabolism (Pelicia et al., 2009). It is thus important to use some feed additives that help in increasing the egg production, and protecting the thickness of the egg shell from weakness as well to reduce the percentage of broken eggs which exceed 20% at the final stage of production cycle (Nys, 2001), by enhancing the absorption of nutrients and improving the health of hens by strengthening the immune system.

These feed additives include probiotics and prebiotics which are added in the diet individually or together for synbiotic action. Probiotics are live microorganisms which provide health benefits to the host when served in sufficient quantity (FAO/WHO, 2001). Probiotics benefit poultry by decreasing harmful bacteria (Salem, 2012), enhancing intestinal barrier functions and positively modulating the immune system (Ng et al., 2009). Prebiotics are feed ingredients that are not digested by the host and they can be used to promote the growth of beneficial bacteria (Kontula et al., 1999).

Probiotics and prebiotics affect the metabolic activity of the beneficial bacteria within the layers' intestine, which positively influence mineral absorption rate especially those of calcium and magnesium (Roberfroid, 2000). The purpose of this study is to evaluate the effect of adding both probiotic and prebiotic alone or in combination (synbiotic) on the productive performance and egg quality of H&N Brown Nick laying hens during the last stage of egg production in winter season.

MATERIALS AND METHODS

The current experiment was carried out in south Sinai experimental research station (Ras-Suder City) which belongs to the Desert Research Center during the winter season (from December, 2018 to February, 2019 for 64 days).

Birds' management and the experimental design: Eighty H&N Brown Nick laying hens at 65 weeks of age were randomly assigned into 4 treatments; each one has 10 replicates with 2 hens per replicate. The experimental treatments were T1 (control), T2 (1g Bio-plus2B®/1kg diet, probiotic), T3 (1g Techno Mos®/1kg diet, prebiotic) and T4 (1g Bio-plus2B® plus 1g Techno Mos®/1kg diet, synbiotic). The experimental diet (Table 1) was formulated according to NRC (1994) and was iso-nitrogenous (17% CP) and iso-caloric (2755 Kcal ME/Kg). Feed and fresh water were made available to the birds ad-lib all the time.

Table-1: The composition and calculated nutritive values of the experimental diets.

Constituents	T1 (Control)	T2 (Probiotic)	T3 (Prebiotic)	T4 (Synbiotic)
Feed Ingredients (%)				
Yellow corn	59.10	59.10	59.10	59.10
Soybean meal 44%	23.05	23.05	23.05	23.05
Corn gluten meal 60%	2.15	2.15	2.15	2.15
Sunflower oil	1.50	1.50	1.50	1.50
Limestone	9.00	9.00	9.00	9.00
Di-Calcium phosphate	1.45	1.45	1.45	1.45
Wheat bran	3.05	3.05	3.05	3.05
DL-Methionine	0.10	0.10	0.10	0.10
Salt	0.30	0.30	0.30	0.30
Vit. & Min. premix*	0.30	0.30	0.30	0.30
Total	100	100	100	100
Nutrient composition				
Crude protein %	16.98	16.98	16.98	16.98
ME (Kcal/kg)	2758	2758	2758	2758
Calcium (%)	3.82	3.82	3.82	3.82
Available P (%)	0.38	0.38	0.38	0.38
DL-Methionine (%)	0.39	0.39	0.39	0.39
L-Lysine (%)	0.81	0.81	0.81	0.81
Feed Additives**				
Probiotic/1 kg diet	-	1g Bio-lus2B®	-	-
Prebiotic/1 kg diet	-	-	1gTechnoMos®	-
Synbiotic/1kg diet	-	-	-	1g Bio-plus2B®+ 1g Techno Mos®

*Vitamins and minerals premix: Each 2.5 kg contain: Vit A 10,000,000 IU, Vit D3 2,000,000 IU, Vit E 10g, Vit K 1000mg, Vit B12 10mg, Vit B1 1000mg, Vit B2 5000mg, Vit B6 1.5g, Pantothenic acid 10g, Niacin 30g, Folic acid 1g, Biotin 50mg, Iron 30g, Manganese 70g, Choline chloride 10g, Copper 4g, Zinc 50g, Selenium 100mg and Iodine 300mg.

**The selected probiotic is Bio-plus 2B® (*Bacillus licheniformis* CH 200/DSM 5749 $1.6*10^9$ CFU/g and *Bacillus subtilis* CH 201/DSM 5750 $1.6*10^9$) and prebiotic is Techno Mos® (Mannanoligosaccarides (MOS) and 1,3 B-glucan that is derived from the cell wall of the yeast *Saccharomyces cerevisiae*).

Bird housing: All hens were housed in wire cages of triple deck batteries and they were exposed to 16 hr of continuous light during the production period. Average of indoor ambient temperature (19.90±0.20 ºC) and relative humidity, RH (56.33±1.44%) were recorded using electronic digital thermo-hygrometer. A temperature-humidity index (THI) was calculated according to Amundson et al. (2006) with the following formula:

$$\mathbf{THI = (0.8 \times T) + [(RH / 100) \times (T - 14.4)] + 46.4}$$

The average THI was 65.26±0.32

Performance measurements: Individual live body weights were recorded at 65 and 74 weeks of age. Body weight gain (g) was calculated as the difference between the initial and final body weights. Feed intake (g) was recorded weekly

Inner egg components analysis: The chemical composition of inner egg components (yolk and albumen) was determined according to A.O.A.C. (2005), while gross energy content (kcal/kg) was measured by completely combusting in a bomb calorimeter.

Egg quality and shell quality traits: Egg weight and number were recorded daily to calculate the egg mass (g/hen/day). Feed efficiency (g feed intake / g egg mass) was calculated as the amount of feed consumed divided by egg mass. A number of 8 eggs were taken from each treatment at the end of experiment for measuring egg quality traits which included egg weight, egg shape index, albumen (%), yolk (%), yolk/albumen ratio and haugh unit. Shell quality traits included shell weight, shell (%), shell thickness, shell density, shell surface area and shell weight per unit of surface area (SWUSA). The percentage of albumen, yolk and shell was calculated as their weights relative to egg weight (Carter, 1968). Yolk and albumen weights (g) were measured to calculate yolk/albumen ratio.

Egg dimensions (mm) were measured by using a digital caliper to calculate shape index (Panda, 1996) as follows:

$$\mathbf{Egg\ shape\ index = (Egg\ width / Egg\ length) \times 100}$$

Haugh unit was calculated according to Haugh (1937):

Haugh unit=100 x log $\mathbf{(H+7.57-1.7 \times W^{0.37})}$**,** where: H=Albumen height (mm), W=Egg weight (g)

Shell thickness (ST) was measured without membrane using micrometer.

Shell surface area (SA) was measured as $\mathbf{SA\ (cm^2) = 3.9782 \times EW^{0.7056}}$

Where: 3.9782 = constant factor, EW = egg weight (g)**.**

Shell weight per unit of surface area (SWUSA) was estimated as:

$\mathbf{SWUSA\ (mg/cm^2) = SW\ (mg) / SA\ (cm^2)}$**,** where SW= shell weight. Both SA and SWUSA were calculated according to Nordstrom and Qusterhout (1982).

Shell density (SD) in g/cm^3 was estimated by the equation of Curtis et al. (1985) as follows:

$$\mathbf{SD = SW\ (g)/SA\ (cm^2) \times ST\ (cm).}$$

Economic Efficiency: The economic efficiency of feed for egg production was calculated according to the costs of the experimental diets and the price of one kilogram of egg. The value of the economic efficiency was calculated as the net return per unit of total costs

Statistical analysis: The data was analyzed using General Linear Model (GLM) procedures by SAS program (SAS, 2002) using one-way analysis of variance according to the following model:

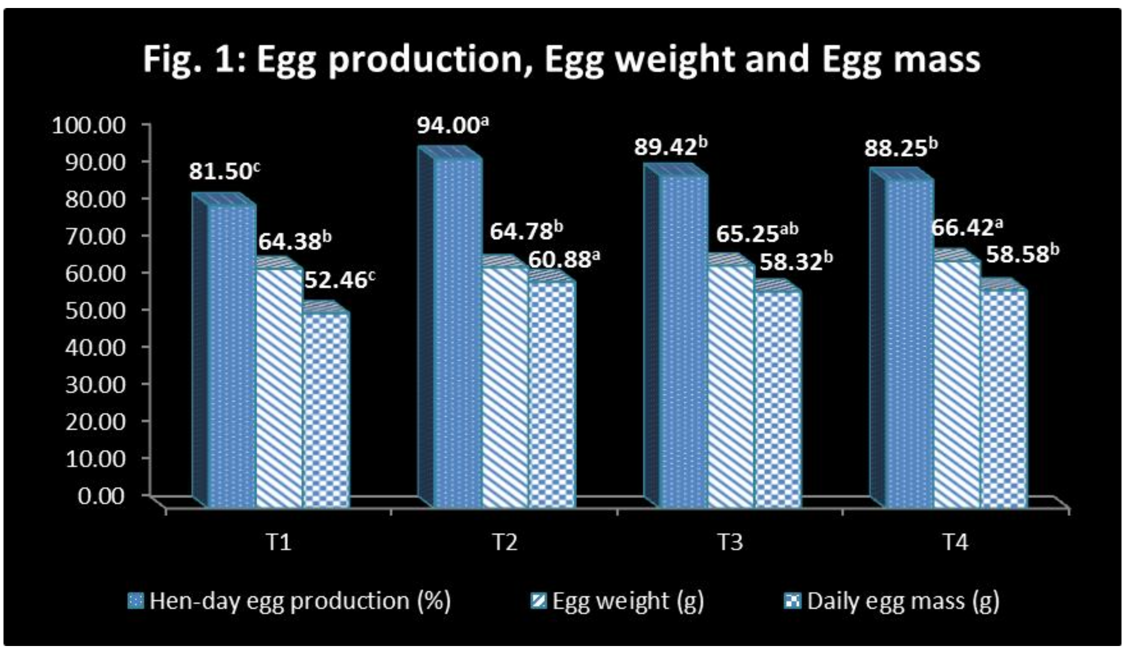
$$Y_{ij} = \mu + T_i + e_{ij}$$

Where: Y_{ij} = Observation. μ = General mean. T_i = Random effect of treatment (i = 1, 2, 3, 4). e_{ij}= Random error assumed to be independent and normally distributed. LSD test was conducted to test significant differences among treatment means.

RESULTS AND DISCUSSIONS

Productive performance: The results of hen-day egg production (%), egg weight (g) and daily egg mass (g) during the experimental period (65-74 weeks of age) are presented in Figure 1. All the recorded experimental treatments were highly significant (P<0.001).

Fig. 1: Hen-day egg production (%), egg weight (g) and daily egg mass (g) of the hens fed on control diet (T1), probiotic (T2), prebiotic (T3) and synbiotic (T4) fortified diets during the last stage of egg production (65-74 weeks of age).



Numerals with a, b, c superscripts were significantly (P<0.001) different.

Hen-day egg production (%) in T2 group (94%), was significantly (P<0.001) higher than in T3 (89.42%), T4 (88.25%), and T1 group (81.5%). The increase in egg production was 12.5%, 7.92% and 6.75% for T2, T3 and T4 groups respectively versus T1. Egg weight (g) was the highest (P<0.001) in T4 group (66.42 g) followed by T3 (65.25 g), T2 (64.78 g) and T1 (64.38 g). The differences were non-significant among T1, T2 and T3. Daily egg mass (g) was the highest in T2 group (60.88 g), and was significantly (P<0.001) higher than T4 (58.58 g), T3 (58.32 g) and T1 (52.46 g).

The daily egg mass (g) in T1 was significantly ($P<0.001$) lower than T2, T3 and T4. The differences between T3 and T4 were non-significant. The results indicated positive and significant ($P<0.001$) superiority of probiotic (T2) treatment on hen-day egg production and egg mass, over the other treatments.

These results were in conformation with Abdelqader et al. (2013a and 2013b) and Abdelqader (2017) who recorded a significant increase in egg production, egg weight and egg mass by adding probiotic (0.5 g and 1g *Bacillus subtilis* / kg diet), prebiotic (1 g inulin / kg diet) or synbiotic (1g *Bacillus subtilis* + 1g inulin / kg diet) in laying hens diet (64-75 weeks of age) compared to the control. It has been reported earlier that adding *Bacillus licheniformis* and *Bacillus subtilis* in rations of Brown-Nick layer hybrids increased egg production (Kurtoglu et al., 2004). Likewise, Nahashon et al. (1992) and Tortuero and Fernandez (1995) have exhibited that using probiotics as feed additives significantly improved the egg weight.

However, the improvement in egg production in this study may be due to the influence of probiotics and prebiotics on the metabolic activity of the beneficial bacterial colonies inside the layer's intestine. They could have improved the mineral absorption rate such as calcium (Roberfroid, 2000). It has been reported that calcium and phosphorus retention were improved in laying hens when the diet contained *Lactobacillus* (Nahashon et al., 1996). Therefore, probiotics and prebiotics could be considered as antimicrobial growth promoters, which have positive effects on the metabolic processes and utilization of nutrients (Yeo and Kim, 1997). Also, the synergism between probiotic and prebiotic together actually lead to better nutrients utilization, metabolism and good absorption.

On the contrary, Daneshyar et al. (2007) have reported that probiotic supplementation in rations of broiler breeder did not have significant effect on egg production and egg mass, however, the significant effect was observed in egg weight. Also, Haddadin et al. (1996), Nahashon et al. (1996) and Chen and Chen (2003) confirmed that probiotic addition had no significant effect on egg weight. Likewise, Elnagar (2013) had also observed that *Saccaromyces cerevisiae* at levels of 3 and 6 g / kg diet significantly decreased egg production.

These controversial results might be related to the dosage of probiotics and prebiotics in the diet, the species of probiotics, types of prebiotics, breed and age of birds, the stage of production, climatic conditions, degree of the surrounding environmental pollution and bacteria's tolerance of climatic conditions. These factors have an important role in increasing or decreasing the quantity and the quality of production.

Body weight and weight gain: The body weights and gains are illustrated in Table 2. During the whole experimental period, there was a non-significant increase (12.7%) in body weight gain of hens fed on synbiotic treatment (T4) compared to the control (T1). The current results agree with those of Sheoran et al. (2017) who observed that body weight gains in hens fed on different levels of prebiotic and probiotic feed additives were not significantly affected compared to the control. Similar results were reported by Nahashon et al. (1996). Laying hens fed on probiotic (*Lactobacillus* microorganisms) and prebiotic in their diets gained more body weight (Nahashon et al., 1994; Bozkurt et al., 2011).

Table-2: Body weights (g) and weight gain (g) of the hens fed on control diet (T1), probiotic (T2), prebiotic (T3) and synbiotic (T4) feed supplements during the last stage of egg production (65-74 weeks of age).

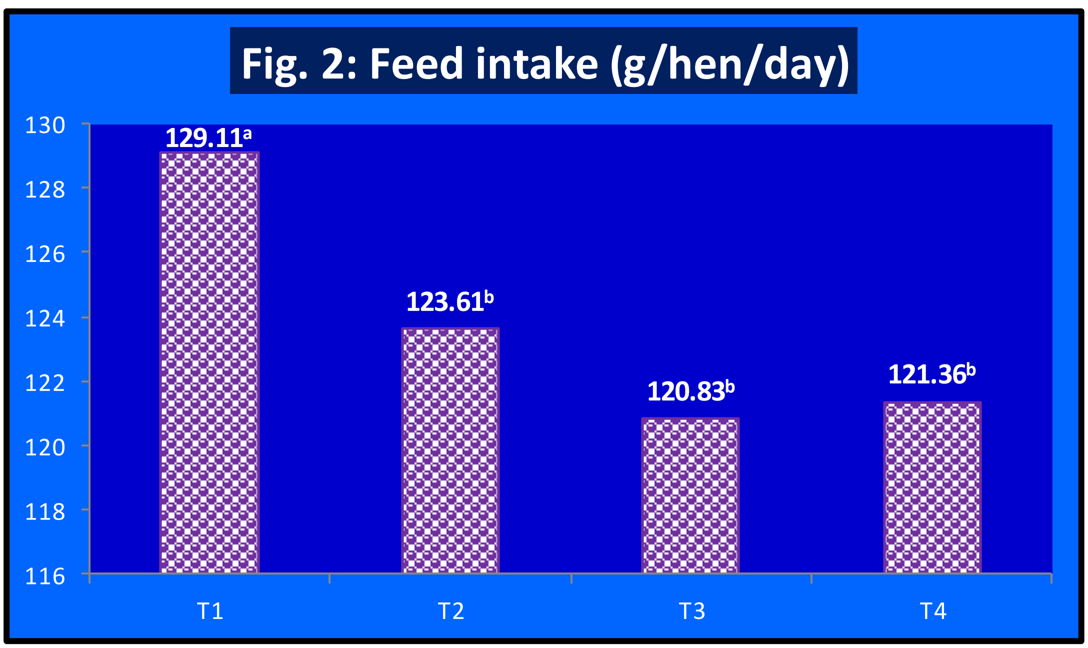
Body weight	T1	T2	T3	T4	SE	Sig
Initial weight (g)	1892.5	1879.5	1917.5	1892.5	37.49	ns
Final weight (g)	2050.0	2044.0	2077.5	2070.0	60.94	ns
Weight Gain (g)	157.5	160.5	160.0	177.5	70.45	ns

SE = Stander error, Sig = Significance, ns = not significant

Feed intake and Feed efficiency: Figure 2 and Figure 3 indicated that there were significant differences among treatments concerning the daily feed intake and feed efficiency (P<0.001), respectively. There was a significant decrease in feed intake of the experimental treatments compared to the control. The values of feed intake (g/hen/day) were 123.61, 120.83 and 121.36 versus 129.11 for T2, T3 and T4, respectively compared to T1.

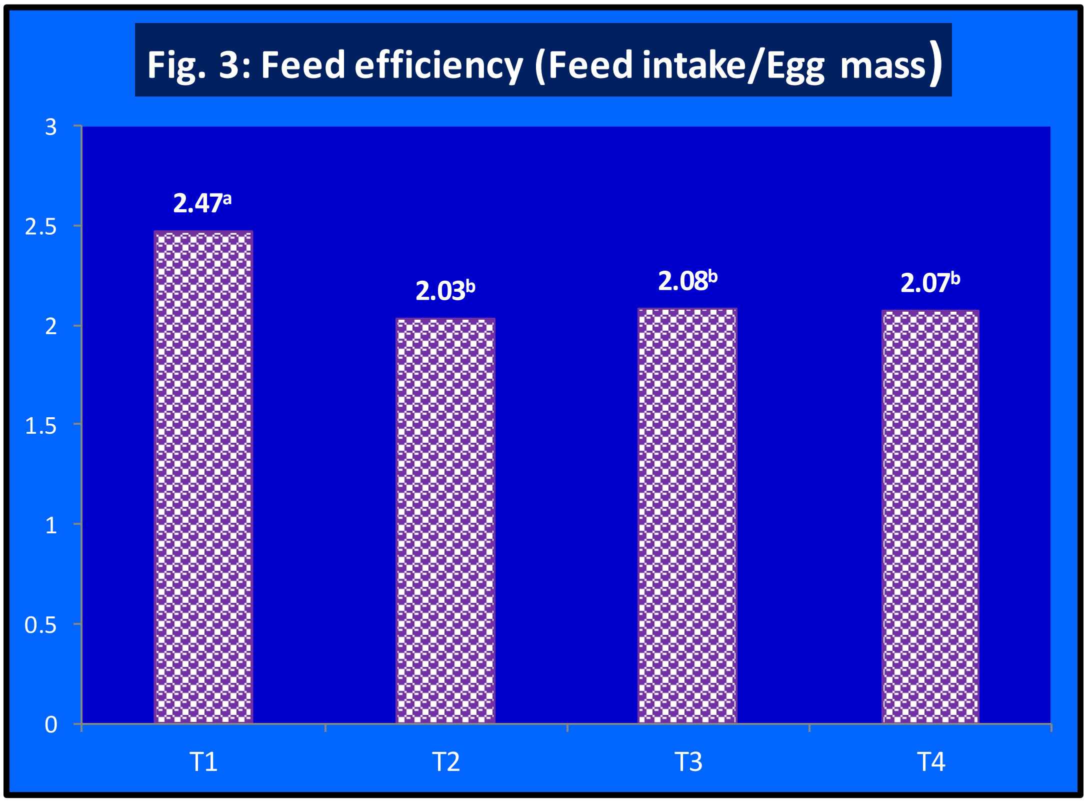
However, the experimental treatments showed a significant improvement in feed efficiency compared to the control. The values of feed efficiency were 2.03, 2.08 and 2.07 versus 2.47 for T2, T3 and T4, respectively compared to T1. This improvement may be attributed to probiotics and / or prebiotics which have beneficial effects in increasing the nutrient absorptive surface area in the gut (Amat et al., 1996: Salem, 2012).

Fig. 2: Feed intake (g/hen/day) of the hens fed on control diet (T1), probiotic (T2), prebiotic (T3) and synbiotic (T4) fortified diets during the last stage of egg production (65-74 weeks of age).



Numerals with a, b superscripts were significantly (P<0.001) different

Fig. 3: Feed efficiency (Feed intake/Egg mass) of the hens fed on control diet (T1), probiotic (T2), prebiotic (T3) and synbiotic (T4) fortified diets during the last stage of egg production (65-74 weeks of age).



Numerals with a, b, superscripts were significantly (P<0.001) different

Similar results were reported by Elnagar (2013) who showed that *Saccaromyces cerevisiae* significantly decreased feed consumption. Several other studies have proved that feed efficiency was improved by adding probiotic to the diet (Panda et al., 2008; Youssef et al., 2013; Chung et al., 2015). Likewise, Li et al. (2007) have confirmed that the lowest value of feed intake and feed efficiency was found when 2g / kg fructo-oligosaccharides was used, while the highest value was observed in the control group during 43-56 weeks of age in laying hens.

On the contrary, Fathi et al. (2018) have observed that there was insignificant decrease in feed intake in both groups fed on a diet containing probiotics and non-significant difference in feed efficiency compared to the control. Likewise, Nahashon et al. (1994), Mohan et al. (1995), Tortuero and Fernandez (1995), Salem et al. (2012) and Elnagar (2013) have shown that feed efficiency was not affected by probiotics.

Egg quality traits: The results in Table 3 showed the effect of dietary treatments on egg quality traits. Probiotic (T2) and synbiotc (T4) treatments recorded a non-significant increase in egg weight and the percentage of egg albumen. Prebiotic (T3) and synbiotic (T4) treatments showed a numerical increase in egg yolk (%) and yolk/albumen ratio. The best value of egg shape index was observed in T3. Haugh unit was higher in T4 group than the other three groups.

Table-3: Egg quality traits of hens on control diet (T1), and probiotic (T2), prebiotic (T3) and synbiotic (T4) supplemented diets during the last stage of egg production (65-74 weeks of age).

Items	T1	T2	T3	T4	SE	Sig
Egg weight (g)	63.63	67.63	64.32	67.24	1.42	ns
Egg shape index (%)	76.84	76.41	78.14	76.60	0.76	ns
Albumen (%)	50.98	54.54	50.01	51.68	1.31	ns
Yolk (%)	31.18	28.56	33.26	32.92	1.36	ns
Yolk/Albumen ratio	61.71	52.67	68.03	63.91	4.21	ns
Haugh unit	64.67	65.69	65.24	67.91	3.00	ns
SE = Stander error, Sig = Significant, ns = not significant						

These results have been confirmed by some scientists such as Mahdavi et al. (2005) and Mohebbifar et al. (2013), who did not find any significant effects for dietary probiotic inclusion on egg quality. Fathi et al. (2018) have reported that there were non-significant differences among the dietary groups (control, 200 and 400 ppm *Bacillus subtilis*) with respect to yolk (%), haugh unit score and egg shape index. Likewise, *Saccharomyces cerevisiae* at levels of 3 or 6 g / kg feed had no significant effect on haugh unit, albumen (%) and egg shape index, but the percentage of yolk was improved (Elnagar, 2013), however, egg shape index was not affected by probiotics and prebiotics treatments (Sheoran et al., 2017).

Shell quality traits: Data of shell weight (g), shell (%), shell thickness (mm), shell surface area (cm^2), shell density (g/cm^3), shell weight per unit surface area (SWUSA, mg/cm^2) are represented in Table 4.

Table-4: Shell quality traits of hens fed on control diet (T1), and probiotic (T2), prebiotic (T3) and synbiotic (T4) supplemented diets during the last stage of egg production (65-74 weeks of age).

Items	T1	T2	T3	T4	SE	Sig
Shell weight (g)	7.72	7.81	8.43	8.18	0.258	ns
Shell (%)	12.16^{ab}	11.57^{b}	13.08^{a}	12.17^{ab}	0.328	…
Shell thickness (mm)	0.076	0.077	0.078	0.077	0.006	ns
Shell surface area (cm^2)	74.50	77.77	75.09	77.48	1.167	ns
SWUSA (mg/cm^2)	103.67^{b}	100.45^{b}	112.07^{a}	105.53^{ab}	2.819	…
Shell density (g/cm^3)	1.42	1.38	1.55	1.47	0.147	ns
Numerals with a, b, superscripts in the same row were significantly ($P<0.05$) different SE=Standard error, Sig=Significant, ns=non-significant						

Prebiotic treatment (T3) showed significant (P<0.05) improvement in shell (%) and SWUSA compared to T1 and T2, but did not differ from synbiotic treatment (T4). Shell weight was numerically improved by T3 and T4 compared to the other two treatments (T1 and T2). Other parameters were not affected by feed additives.

Nahashon et al. (1994), Mohan et al. (1995) and Li et al. (2007) showed non-significant differences in shell thickness and haugh unit which agreed with the current results. On the contrary, Sheoran et al. (2017) observed that there was a significant increase in shell thickness, shell weight and shell (%) with probiotics and prebiotics treatments compared to the control.

Abdelqader et al. (2013a) got the maximum increase in egg shell weight and egg shell thickness with 1g *Bacillus subtilis* / kg diet compared to the other treatments. Further, dietary supplementation with *Bacillus subtilis* at 400 ppm level (Fathi et al., 2018) and *saccharomyces cerevisiae* at levels of 3g or 6g / kg diet (Elnagar, 2013) significantly increased shell thickness compared to the non-treated laying hens.

The significant increase in shell (%) and SWUSA (mg/cm^2) in T3 group might be due to the increase in calcium and phosphorus content in serum of laying hens by prebiotic addition. This indicated that prebiotic addition provided favorable acidic environment inside the intestine, which helped in improving the digestion and absorption of calcium. Ashmead et al. (1985) have also reported that minerals such as calcium and phosphorus required a very low pH to be soluble. Thus, the acidic environment inside the intestine might have facilitated the ionization of minerals which boosted mineral absorption.

Chemical composition of egg: Table 5 shows the percentage of crude protein and fat and the gross energy content GE (kcal / kg) of the inner egg components (yolk and albumen). There was an improvement in protein (%) for all the experimental treatments compared to the control, while fat (%) numerically decreased in T2 and T3. The synbiotic treatment recorded the highest percent of protein, while probiotic treatment showed the lowest percent of fat. Gross energy (kcal / kg) was higher (P<0.05) in T2 (Probiotic) compared to T3 (prebiotic) and T4 (synbiotic) treatment groups, but did not differ from the control (T1).

Table-5: Chemical composition of inner egg components (yolk and albumen) on dry matter basis (%) in hens fed on control (T1), probiotic (T2), prebiotic (T3) and synbiotic (T4) supplemented diets during the last stage of egg production (65-74 weeks of age).

Items	T1	T2	T3	T4	SE	Sig
Protein (%)	39.53	42.63	43.30	45.30	1.94	ns
Fat (%)	35.57	33.40	34.50	35.50	1.90	ns
GE (kcal/kg)	6709.1^{b}	6815.8^{a}	5840.9^{c}	6051.5^{bc}	221.6	…

Numerals with a, b, c superscripts in the same row were significantly (P<0.05) different. SE = Stander error, Sig=Significant, ns=non-significant, GE = Gross energy.

Trani et al. (2016) found that egg yolk obtained from laying hens fed on probiotic had a significantly lower fat content and higher protein content compared to the control group. There were no differences among treatments concerning the albumen composition of fat and protein.

The current results showed similar improvement in the composition of inner egg components (yolk and albumen) like Trani et al. (2016), but this improvement was non-significant. Therefore, there were no significant differences among treatments concerning total lipid contents of egg yolk by addition of prebiotic, probiotic and synbiotic (Tang et al., 2015) and *Lactobacillus* (Haddadin et al., 1996; Kalavathy et al., 2009) to the ration.

The reduction in the inner egg components of hens fed on probiotic may be a good indicator of low yolk cholesterol as reported by Slaugh (2002) and Aghaii et al. (2010) who found that adding probiotics to laying hens diet could lead to a 50% reduction of cholesterol in the yolk. About the gross energy of inner egg components, there are no available references.

The highest value of gross energy of the probiotic treatment may be related to probiotic bacteria that can breakdown the indigestible carbohydrates in the gut and produce the short chain fatty acids which can be used as fuels for the tissues of the host (Rémésy et al., 1992).

On the other hand, the low gross energy of the treatments containing prebiotic (mannanoligosaccarides and β-glucans) may be due to non-starch polysaccharides, which have strong negative effect on net energy utilization which are lost in the form of volatile fatty acids in the excreta (Choct, 1999).

Feed efficiency, cost efficiency and egg production economy: The data on feed efficiency and economic efficiency of the hens treated with experimental diets are shown in Table 6. Adding the current feed additives (probiotic, prebiotic and synbiotic) to laying hens' diets decreased the feed cost per kilogram of eggs and increased the net return of the production process during the late phase of egg production.

Table-6: Feed efficiency, feed cost efficiency and egg production economy in hens fed on control (T1), probiotic (T2), prebiotic (T3) and synbiotic (T4) supplemented diets during the last stage of egg production (65-74 weeks of age).

Items	T1	T2	T3	T4
Feed efficiency (g feed / g egg mass)	2.47	2.03	2.08	2.07
Cost of kg feed (LE.)	6.00	6.24	6.11	6.35
Feed cost / kg eggs (LE.)	14.82	12.67	12.71	13.14
Market price of one kg eggs (LE.)	19.00	19.00	19.00	19.00
Net return (LE.) / kg eggs	4.18	6.33	6.29	5.86
Economic efficiency of feed (%)	28.21	49.96	49.49	44.60
The price of 1 ton of feed = 6000 LE. (Egyptian Pound), 1 kg probiotic = 240 LE., and 1 kg prebiotic = 105 LE.				

CONCLUSION

It can be proved from the results that using probiotic (Bio-plus2B®) and prebiotic (Techno Mos®) individually or together in diets of laying hens during the last stage of egg production was beneficial in increasing egg production with improving feed efficiency and egg quality, besides the improvement in gross energy content of the inner egg components. In addition, it increased the net return of the produced eggs and improved the economic efficiency of feed.

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UNDERTAKING

It is certified that the research paper '**THE EFFECTS OF PROBIOTIC, PREBIOTIC AND SYNBIOTIC TREATMENTS ON EGG PRODUCTION AND EGG CHARACTERISTICS IN BROWN NICK HENS DURING THE LAST STAGE OF PRODUCTION IN WINTER SEASON'** is an original research work carried out by the authors in Animal and Poultry Nutrition Department, Desert Research Center, El-Mataria, Cairo, Egypt. It has neither been published nor contemplated for publication elsewhere.

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