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# Effect of Pelleted Fermented Feed-in Egg Quality of Laying Hens

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## Abstract

lactobacilli, Bacillus subtills, Bifidobacterium, Saccharomyces cerevisiae were applied to produce fermented feeds (FF), The FF was pelleted to investigated for its influences on egg quality for laying hens, the (FF) was ferment with 10 g / kg feed of the probiotic with a wetting ratio of half a liter/kg of feed for 48 hours where it was used at rates 0, 25%, 50%, 75%, and 100% for the treatments T1, T2, T3, T4, and T5 respectively, and the duration of the study lasted for seven periods, each period was two weeks, as for the following study results: Significant superiority (P≤0.05) for all FF treatments during the second period and treatment T5 during the third and sixth periods, and for treatments T3, T4 and T5 during the fourth period in the shell weight. As for the shell thickness, a significant superiority (P<0.01) was obtained for treatment T3 in the fourth period and for treatment T2 in the fifth, sixth, and seventh periods, and in the yolk height, there were a significant superiority ( $P \le 0.01$ ) for the treatment T4 during the first, second and seventh periods, and for the treatments T2 and T5 during the fourth and fifth periods, as for the yolk diameter, a significant superiority (P≤0.01)was obtained for the treatments T2, T3 and T4 in the fourth and fifth periods, and T2 during the seventh period, and in albumen height, a significant (P≤0.01) was increased for the treatments T3, T4, and T5 during the second period, and a significant superiority was obtained for treatment T5 in the fifth period and a significant superiority for the two treatments T1 and T2 in the seventh period.

Keyword: Egg quality, Fermentation feed, lying, Pelleting.

# Introduction

The poultry industry is one of the fastest-growing animal industries in the world (Wang *et al*, 2017). Poultry meat and eggs are the most important source of dietary protein for the world population (Magdelaine, 2011). At the same time, the poultry industry faces some problems, including that egg production is now hampered by a shortage. Also, the layers of laying poultry projects aim to increase production to the maximum at the lowest cost (Godfray *et al*, 2010; Wang *et al*, 2017), as the problem

<sup>1</sup>The article is part of Ph.D. Thesis of the first author Dates: Received: 04 March 2021 Accepted 14 April 2021 Published: 30 June 2021 of nutrition and mycotoxins remains in the first place considering the control of other conditions such as genetics and environmental conditions, may nutrition is the key to the aforementioned problems, as modern biotechnology provides new feeding methods to closely suit the nutritional needs of laying hens and mothers, thus reducing the cost of feed, which constitutes about 65-70% of the production cost (Nagy et al, 2016). Intensive egg production and mating or a weak immune system make birds vulnerable to disease (Cherian, 2013), and thus it became necessary to improve the quality of feed provided to birds to reach the best level of production. (FAOSTAT, 2015), fermentation technology is one of the most important techniques that improve the level of forage, as fermentation with the probiotic causes inhibition of the growth of pathogenic bacteria in the regimen of food (Cotter et al, 2013; Khan et al, 2020), increasing the protein content in it (El-Hag et al, 2002: Doudu et al, 2003; Pranoto et al, 2013) Besides, the fermentation process raise of the amino acids, inclusive methionine and lysine (Pranoto et al, 2013; Nkhata et al, 2018), which increases the absorbed and protein digested, by the bird. Fermentation increases the digestibility of FF grain protein (Yousif and El Tinavi, 2001 and 2003; Osman, 2004) as well as removes trypsin inhibitors (Osman, 2011) and reduces phytic (Osman, 2011; Hassan, 2015) and the production of organic acids (Van Immerseel et al., 2005). 2006), increased availability of mineral elements (Pranoto et al., 2013; Nkhata et al, 2018) as well as production of antioxidant vitamins (LeBlanc et al, 2011; Nagy et al, 2016) which is reflected in the productive and reproductive performance of laying hens. Among the novelty technologies for using a FF to convert it into the pellets to facilitate its storage, transportation, and handling (Yeh et al, 2018), the researcher indicated that it is can be to use an FF to reduce the price of feed due to the increase in the efficiency of using it, also for using the probiotic for fermentation of feed materials, also, the FF increase shell weight, eggs weight, and shell stiffness in laying hens are increased via fermented feed complement(Engberg et al, 2009). Meanwhile, Zhu et al, (2020) showed that feeding on fermented fodder improved the growth performance of laying hens, as well as the immune status by increasing the level of interleukins and the number of T- Lymphocyte, so the aims of a study to detect the effect of FF after the pelleting process in the eggs quality of the layer hens.

# **Materials and methods**

## **Ethical approval**

The Scientific Ethical Committee of College of Agricultural Engineering Sciences-University of Baghdad, Iraq approved this study (Approval no: (572); October 2020).

## **Probiotics and Fermented Feed Preparation**

A commercial Iraqi probiotic content (*lactobacilli*, *Bacillus subtills*, *Bifidobacterium*, *Saccharomyces cerevisiae* used to ferment layer diet in table 1, aerobically fermentation with 10 g of a probiotic / kg of feed and a wetting ratio of half a liter of water/kg of feed for 48 hours after that the FF was dried and pelleted by pelleting machine.

### Experiment designing and chicken management

This farm experiment was in the poultry farm of the Faculty of Agriculture/ Al-Qasim green University for 14 weeks extended for the period from Oct-28-2020 to Jan-27-2021, wherein this experiment the 58-week-old Lohman brown chicken was used, and 150 laying hens were randomly divided into five treatments, each treatment 30 chickens were divided into three replicates each of 10 chickens (T1: control treatment, T2: FF at a rate of 25%, T3: FF at a rate of 50%, T4: FF at a rate of 75%, T5: FF at a rate of 100%), at seven periods each period of 2 weeks.

## Feed treatment

The chickens were fed on ration below, a feed ration of 115 gm/chicken/day.

Ingredients	%
yellow corn	36.5
wheat	12
barley	13.9
Soybean meal (44% protein)	25
*Premix	2.5
DCP Calcium Diphosphate	8.3
Sunflower oil	1.8
Total**	100
Metabolic energy (kilocalories/kg feed)	2758.68
Crude protein (%)	17.19
Crud fiber	3.24
Calcium (%)	3.81
Available phosphorus (%)	0.29
Methionine + cysteine (%)	0.73
Lysine (%)	0.95

 Table 1. The diet components in the study and their chemical composition

\*Premix Maxcare of Belgian origin Each 1 kg contains: crude protein 7.9%, lysine 2.4%, methionine 7.7%, methionine + cysteine (7.7%), calcium 23.1%, phosphorous 3.3%, sodium 5.5%, representative energy (2903 kcal/kg), vitamin A (400,000 IU), Vitamin D3 (300,000 IU), Vitamin D (20,000 IU), Vitamin E (800 IU), Vitamin K 80 ppm, Vitamin B1 40 ppm, Vitamin B2 (1600) ppm, Calcium Pantothenate (320) ppm, Niacin (600) ppm, Biotin (1600) ppb, vitamin B12(1000) ppb, folic acid (40) ppm, vitamin B6 (160) ppm, iron 2800 ppm, copper 600 ppm, zinc (2400) ppm, magnesium (4000) ppm, iodine (80) ppm, selenium 8 ppm.

\*\* Chemical analysis computed according to NRC (1994)

• Dicalcium phosphate with a concentration of calcium in 24%, phosphorus 18%.

• (9000) kcal/kg oil

### **Studied traits**

### Yolk and albumen height

Later than the egg was broken on a flat surfacing, the rising of the yolk and albumen was measured by a special three-base micrometer device (Ames micrometer), and a mean of two readings of the albumen of each egg were taken from the area close to the contact of the thick white with the yolk (Wilgus and Van Wangener, 1936). The height of the yolk was measured from the center after breaking the egg on a flat surface. The diameter of the yolk was also measured with a special Vernia, where the eggs were taken randomly and for all the replicates, and the height of the yolk was taken from the center. As for the height of the albumen, it was taken from the center and the far end, after which the rate was taken.

### Shell thickness

Measure the thickness of a shell with the inner membranes, after deriding it from the tapered and convex sides of each egg with a micrometer device, where he taking two readings for the pointed side and the wide side average of the two readings, according to the equation below:

Shell Thickness of First End (mm) + Shell Thickness of Second End (mm)

2

shell thickness = \_\_\_\_\_

(Mm)

## Shell Weight (g)

Use a sensor scale for this, which reads to two decimal places, where the shell was weighed with its membranes after emptying the entire contents of the eggs.

### statistical analysis

The Statistical Analysis System -SAS (2012) was used in data analysis to study the effect of different treatments on the traits studied according to a Completely Randomized Design (C.R.D), and the mean differences among the treatments were compared according to Duncan multiple ranges test (Duncan,1955).

## $Yij = \mu + Ti + Eij$

Yij: The observation j of treatment i.

 $\mu$ : Overall mean.

Ti: effect of treatment i (the study included the impact of five treatments).

Eij: a random error that is normally distributed with an average of zero and a variation

of  $\sigma 2e$ 

## **Results and Discussion**

#### Shell weight

Table 2 shows the effect of the treatments on the eggshell weight the eggshell produced. It is noticed that there are no significant differences between the studied treatments during the first, third, and seventh periods. In the second period, significant superiority was observed (P $\leq$ 0.05) for all FF treatments compared with the control treatment, and there was no large difference. Among the treatments T2, T3, T4, and T5, and in the fourth period, a significant superiority was obtained (P $\leq$ 0.05) for the treatment T5 compared to the two cases T1 and T4, and there was no large difference among the treatments T2, T3, and T5 and also among the treatments T1, T2, T3, and T4. The fifth period showed significant superiority (P $\leq$ 0.05) for the treatments of T3, T4, and T5 compared to the treatment T1. No significant difference was recorded among the treatments of T2, T3, T4, and T5 as well between the two treatments of T1 and T2. In the sixth period, a significant superiority (P $\leq$ 0.05) was observed for the treatment. T5 compared to the rest of the treatments, and there was no a large difference was recorded at the treatment. T5 compared to the treat superiority (P $\leq$ 0.05) was observed for the treatment. T5 compared to the treat of T1, T2, T3, and T4.

#### Shell thick (mm)

Table 3 indicates the impact of the treatments on the eggshell thickness the eggshell, and it is noticed during the first, second and third periods that there are no a large differences between the treatments, while in the fourth period, a significant notability(P≤0.01) for treatment T3 compared to treatment T1 and there are no significant differences between the treatments T2, T3, T4, T5 and also between the treatments of T1, T2, T4, T5 and in the second and eighth period, a significant superiority (P≤0.01) was observed for treatment T2 compared to treatment of T1 and there were no significant differences between the treatments T1, T3, T4, T5 as well as between the treatments of T2 T3, T4, T5 and the significant superiority (P≤0.01) of treatment T2 withdrew in the sixth period compared to the two treatments T1, T3 and the superiority of the treatments of T3, T4, T5 over treatment of T1, and no a large differences were found between the treatments for treatment T2 through the seventh period compared to the treatments T1, T3, T5, and T4 outperformed treatment T1 and T3, and no a large differences were found between the treatments T2 and T4, as well as between the treatments T1, T3, T5 and also between the two treatments of the T4 and T5.

#### Yolk height

Table 4 shows the effect of the treatments on the yolk height of the eggs produced during the study period. It is noticed during the first and second periods that a significant (P $\leq$ 0.01) was obtained for treatment T4 compared to the treatments T1, T2, and T5Noa large differences were found between the treatments T3 and T4 as well as between the treatments T1, T2, T3, and T5, also in the third and sixth periods, there was no significant difference among the studied treatments, but in the fourth and fifth periods, it was lucid that there was a significant superiority (P $\leq$ 0.01) for the treatments T2 and T5 compared to the treatments T1 and T4, and there was no significant difference between the treatments T2, T3 and T5 also the treatments T1,

T3, and T4. In the seventh period, there was a large superiority ( $P \le 0.01$ ) for treatment T4 compared to the treatments T1, T2, and T5, and for the treatments T3, T5 over the treatments T1 and T2, and there was no a large difference between the treatments T1, T2 and the treatments T3 and T5 as well as the treatments T3 and T4.

Treatments		Means ± standard error (gm)							
	first period	Second period	Third period	Fourth period	Fifth period	Sixth period	Seventh period		
T1	0.086.36±	b6.29±0.06	0.256.31±	0.24 b6.41±	0.06 b6.21±	0.08 b±6.43	0.05±6.52		
T2	0.086.53±	a6.66±0.07	0.136.57±	0.06 ab6.71±	0.12 ab±6.52	0.13 b6.58±	0.196.73±		
T3	0.126.76±	a6.69±0.08	0.296.72±	0.07 ab6.77±	0.04 a6.62±	0.22 b6.98±	0.126.86±		
T4	6.61±0.04	a6.57±0.06	0.356.74±	0.11 b6.29±	0.12 a6.67±	0.10 b6.77±	0.046.81±		
T5	6.56±0.21	a6.53±0.07	0.056.83±	0.15 a6.96±	0.12 a6.63±	0.04 a7.87±	0.066.87±		
Significant	N. S	*	N. S	*	*	*	N. S		

Table 2. Mean ±	standard error	of the effect	of fermented	food in	the eggshell	weight (gm)

Means with different letters indicate a significant difference in probability level 0.05, N. S: Not significant \* ( $P \le 0.05$ ). The treatment T1, T2, T3, T4, T5 are control treatments without addition, adding 25%, 50%, 75%, 100% fermented feed, respectively.

Treatments	Means ± standard error (mm)							
	first period	Second period	Third period	Fourth period	Fifth period	Sixth period	Seventh period	
T1	±0.090.50	0.05 0.41±	0.05 0.40±	0.01 b0.42±	0.08 b0.43±	0.02 c±0.36	c±0.010.43	
T2	0.08 0.41±	0.01 0.41±	0.03 0.45±	0.01 ab0.48±	0.02 a±0.57	0.15 a0.52±	0.01 a0.56±	
T3	0.03 0.49±	0.08 0.43±	0.03 0.41±	0.03 a0.55±	0.01 ab0.50±	0.02 b0.44±	0.07 c0.45±	
T4	0.06 0.53±	0.02 0.43±	0.06 0.44±	0.04 ab0.50±	0.05 ab0.54±	0.09 ab0.48±	0.06 ab0.52±	
T5	0.05 0.51±	0.01 0.44±	0.04 0.46±	0.02 ab 0.51±	0.05 ab0.50±	0.02 ab0.49±	0.08 bc0.47±	
Significant	N. S	N. S	N. S	**	**	**	**	

Table 3	. Mean	± standar	d error of t	the effect	of ferm	ented foo	d in t	the eggshell	thickness	(mm)
										<hr/>

Means with different letters indicate a significant difference in probability level 0.05, N. S: Not significant \*\* ( $P \le 0.01$ ). The treatment T1, T2, T3, T4, T5 are control treatments without addition, adding 25%, 50%,75%, 100% fermented feed, respectively.

## Yolk diameter

Table 5 shows the effect of the treatments on the diameter of the egg yolk produced during the trial period, and it is noticed that there are no significant differences between the studied treatments in the first, second, third, and sixth periods. In the fourth period, significant superiority ( $P \le 0.01$ ) was obtained for the treatments of T2, T3, T4 Compared with the treatment of T1, there was no significant difference between the treatments of T2, T3, T4, and T5, as well as between the treatments T1 and T5. In the fifth period, it was found that there was a large superiority ( $P \le 0.01$ ) for the treatments of T2 and T4 compared to the treatments of T1 and T5, and there was no significant difference between the treatments T1, T3, and T5 and the significant superiority ( $P \le 0.01$ ) of treatment of T2 continued during the seventh period compared to the treatments of T1, T3, T4 and the treatment T5 superiority the treatments T1 and T4 and the treatments T3 and T4 exceeded the treatment T1 and did not There was no a large difference between the treatments T2 and T5, as well as between the treatments T3 and T4 exceeded the treatment T1 and did not There was no a large difference between the treatments T2 and T5, as well as between the treatments T3 and T4.

## Albumin height

T is noted from table 6 the effect of the treatments on the albumin height during the trial period, and there was no a large difference among treatments during the first, third, fourth, and sixth periods, while in the second period, a significant superiority appeared ( $P \le 0.01$ ) for the treatments of T3, T4, and T5 compared to with the treatments T1 and T2, there was no a large difference between the treatments of T1, T2, as well as among the treatments of T3, T4, and T5. There was a significant difference between the treatments T2, T3, and T5 and also between treatments of T2, T3, T4, and T5 compared ( $P \le 0.01$ ) for the treatments T2, T3, and T5 and also between treatments of T2, T3, T4, and T5 and T2 compared to the treatments T3, T4, and T5, also there was no a large difference between the two treatments T1 and T2, as well as among treatments T1 and T2.

The improvement in the peel qualities of the fermented fodder treatments may be due to the feeding on the FF, as the fermentation of the fodder results in a higher protein

Treatments		Average ± standard error (mm)							
	first period	Second period	Third period	Fourth period	Fifth period	Sixth period	Seventh period		
T1	0.57 b±15.03	0.55 b±15.00	$0.53 \pm 15.30$	0.14 b±14.03	0.29 b±14.77	$0.54 \pm 15.60$	0.30 c±13.90		
T2	0.90 b±15.87	0.57 b±16.00	0.74 ±15.73	0.28 a±16.64	0.63 a±16.97	$0.07 \pm 16.28$	0.32 c±14.77		
Т3	1.05 ab±17.28	0.88 ab±16.66	$0.64 \pm 16.21$	0.92 ab±15.43	0.74 ab±16.07	$0.88 \pm 16.20$	0.12 ab±16.13		
T4	0.75 a±18.71	0.33 a±18.66	$1.04 \pm 15.31$	0.20 b±15.05	0.06 b±15.03	$0.10 \pm 15.46$	0.28 a±16.64		
T5	0.13 b±15.27	1.45 b±15.66	0.69 ±15.34	0.02 a±16.56	0.18 a±17.47	0.27 ±16.26	0.48 b±15.87		
Significant	**	**	N. S	**	**	N. S	**		

Table 4	Joon   standard	annon of the of	Faat of farmon	tod food in t	a valle haight (mm)
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Means with different letters indicate a significant difference in probability level 0.05, N. S: Not significant \*\* ( $P \le 0.01$ ). The treatment T1, T2, T3, T4, T5 are control treatments without addition, adding 25%, 50%,75%, 100% fermented feed, respectively.

Treatments		Average ± standard error							
	first period	Second period	Third period	Fourth period	Fifth period	Sixth period	Seventh period		
T1	3.18 ±40.08	0.40±39.78	1.55±40.35	1.27 b±40.65	0.90 b±40.18	0.20±40.05	0.55 d±39.77		
T2	1.72±40.69	0.91±41.45	1.37±41.77	0.70 a±43.82	0.07 a±42.76	0.68±41.62	0.09 a±42.77		
Т3	0.34 ±38.57	0.40±41.03	0.71±42.31	0.47 a±43.19	0.20 ab±42.16	0.27±40.96	0.35 bc±41.78		
T4	$0.85 \pm 40.52$	0.32±40.32	0.55±42.46	0.84 a±43.15	0.35 a±42.74	0.66±41.03	0.10 c±41.28		
T5	0.50±41.28	1.66±40.84	$0.47 \pm 40.87$	0.69 ab±41.72	0.16 b±40.15	0.45±41.57	0.26 ab±42.57		
Significant	N. S	N. S	N. S	**	**	N. S	**		

Table 5. Mean ± standard error of the effect o	f fermented food in the yolk diameter (m	m)
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Means with different letters indicate a significant difference in probability level 0.05, N. S: Not significant \*\* ( $P \le 0.01$ ). The treatment T1, T2, T3, T4, T5 are control treatments without addition, adding 25%, 50%,75%, 100% fermented feed, respectively.

content (El-Hag et al. 2002: Doudu et al. 2003; Pranoto et al. 2013). Besides, the fermentation process increases the amino acids, including methionine and lysine (Pranoto et al., 2013; Nkhata et al., 2018), which increases the protein digested and absorbed by the chicken, as well as the fermentation process increases the digestibility of forage grain protein (Yousif and El Tinavi, 2001 and 2003; Osman, 2004) They also remove trypsin inhibitors (Osman, 2011) and reduce phytic (Osman, 2011; Hassan, 2015) by activating the phytase enzyme, which increases the efficiency of utilizing the absorbed nutrients, which is reflected in the quality of the eggs produced by the chicken and the internal characteristics of the egg, Also, probiotic bacteria produce organic acids (Van Immerseel et al., 2006). These acids are produced by decomposing carbohydrates, thus reducing the pH of the feed and the chicken's intestine, and that lowering the intestinal ph improves the quality of egg albumin (Tortuero and Fernandez, 1995; Gallazzi et al, 2008), and the improvement in the quality of the shell may be caused by the fermentation of fodder, whereas a result of fermentation, the bioavailability of the mineral elements will increase. Available and not digested by the chicken's digestive enzymes (Nkhata et al., 2018) and that fermentation increases the availability of mineral elements for FF as it found an increase in the level of calcium, magnesium, iron, and zinc after fermentation (Pranoto et al., 2013) also the fermentation increases the availability of calcium, phosphorus, and iron due to decomposition the phytates and oxalates, which chicken to minerals and reduce their bioavailability (Sripriya et al., 1997). Also, amylase and phytase enzymes act on the excretion of probiotic bacteria during the fermentation process, which degrades carbohydrates and phytates, which increases the availability of phosphorous (Liang et al., 2008), all of these factors mentioned above improvement of shell weight and thickness in FF treatments, as well as a decrease in pH during the fermentation process. Aided in the absorption of mineral elements, causing phytic degradation and increasing phosphorous availability (Hemalatha et al. 2007; Reale et al., 2007). As Khan et al, (2020), have shown the enhanced bacteria improve the quality of eggs and their shells by increasing the efficiency of calcium transport through the wall of the gastrointestinal tract as well as compensating for the deficiency of minerals. Saccharomyces cerevisiae also produces a protein (Ferreira et al., 2010; Day, 2016). It was found that it increased the proportion of protein and mineral elements when fermenting corn (Day and Ruben, 2018). The fermented forage materials have a higher digestibility than the thermally treated forage materials (Duodu et al. 2003; Day and Ruben, 2018). Then drying it, after that, it is exposed to heat as a result of converting it into pellets, which will further break the bonds between the nutrients and thus be more available to the bird. Thus, this process is considered one of the most promising technologies in the poultry feed industry.

Treatments		Average ± standard error (mm)							
	first period	Second period	Third period	Fourth period	Fifth period	Sixth period	Seventh period		
T1	$0.14 \pm 3.76$	0.12 b±3.67	0.04±4.42	0.11±5.61	0.17 c±4.87	0.06±5.99	0.49 a±6.62		
T2	0.33±4.33	0.22 b±3.83	0.76±4.78	0.16±6.63	0.17 ab±6.06	0.14±5.80	0.20 a±6.47		
T3	0.47 ±4.23	0.06 a±5.21	0.20±4.73	0.38±6.14	0.16 ab±6.32	0.20±6.51	0.48 b±5.01		
T4	0.34 ±4.35	0.09 a±4.58	0.29±4.72	0.58±6.61	0.29 b±5.62	0.03±6.45	0.21 b±5.30		
T5	0.33 ±4.33	0.36 a±4.96	0.42±5.28	0.71±5.81	0.31 a±6.48	0.47±6.20	0.03 b±5.14		
Significant	N. S	**	N. S	N. S	**	N. S	**		

Table 6 Mean + standard error	• of the effect of	f fermented food in	the albumen	height (mm)
Table 0. Mean - Standard Critor	of the chect of	i ici menicu ioou m	the arbumen	neight (mm)

Means with different letters indicate a significant difference in probability level 0.05, N. S: Not significant. \*\* ( $P \le 0.01$ ). The treatment T1, T2, T3, T4, T5 are control treatments without addition, adding 25%, 50%,75%, 100% fermented feed, respectively.

# Conclusions

The fermentation of feed with probiotic and converting it into a pellet is one of the promising industries in poultry feeding, where fermentation and heat in the pelleting process reach the best level of crushing and digestion of nutrients and thus reaching the maximum benefit by the birds.

## **Conflict of interest**

there is no conflict of interest between authors

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