

# Potential influence of dietary synbiotic and fenugreek seed to improve the productive traits and economic cost in stressed broiler chickens

Cite as: AIP Conference Proceedings **2776**, 100008 (2023); <https://doi.org/10.1063/5.0135987>  
Published Online: 12 April 2023

Ahmed Jebur Dakhil and Karrar Imad Abdulsahib Al-Shammari



[View Online](#)



[Export Citation](#)



Time to get excited.  
Lock-in Amplifiers – from DC to 8.5 GHz

[Find out more](#)

 Zurich Instruments

# Potential Influence of Dietary Synbiotic and Fenugreek Seed to Improve the Productive Traits and Economic Cost in Stressed Broiler Chickens

Ahmed Jebur Dakhil <sup>a)</sup> and Karrar Imad Abdulsahib Al-Shammari <sup>b)</sup>

*Department of Animal Production Techniques, Technical College of Al-Musaib, Al-Furat Al-Awsat Technical University, Babylon City, Almusaiab, P.O. Box 00964, Iraq.*

<sup>a)</sup> ahmed.jebur.tcm.67@student.atu.edu.iq

<sup>b)</sup> Corresponding author: karrar.Al-Shammari@atu.edu.iq

**Abstract.** This study was conducted to evaluate the use powders of local synbiotic and fenugreek seeds as antioxidants in the broiler diet against the experimentally stimulated stress by hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) in drinking water and its influence on productive traits and economic cost up to 5 weeks old. In total, 300 one-day-old chicks were used, chicks were randomly distributed into five treatments, each treatment contained 60 chicks with 3 replicates/group. The treatments were divided into 1st treatment (T1) which involved the chicks fed a diet without any additives as negative control whereas 2nd treatment (T2) involved the chicks which were subjected to stress by 1 ml (0.5%) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water as positive control. In 3rd, 4th and 5th treatments, the chicks were subjected to stress by H<sub>2</sub>O<sub>2</sub> and fed 0.5% of synbiotic (T3), 0.5% of fenugreek (T4) and mixture of 0.25% each of synbiotic and fenugreek (T5), respectively. The results of the research indicated that T3, T4 and T5 caused a significant increase (P<0.05) in the mean body weight, cumulative weight gain and relative growth rate with high significance (P<0.01) in cumulative feed intake and production efficiency factor compared to T2. Also, the same treatments recorded highly significant superiority (P<0.01) in cumulative feed conversion ratio and high (P<0.01) dressing percentage without giblets and with giblets. Moreover, T3, T4 and T5 decreased the economic cost of the broiler rearing project. Thus, we recommended to using both powders of local synbiotic and fenugreek seed or their combination as promising antistress dietary materials for improving the productivity and reducing the economic cost of broilers. This will in turn support the prospects of sustainable development in the globally poultry industry in future directions.

**Keywords.** synbiotic, fenugreek, hydrogen peroxide, broilers, economic cost.

## INTRODUCTION

One of the crucial problems facing the poultry industry is an exposure of birds to oxidative stress, which is one of the most important obstacles to cellular metabolism, whose deleterious impacts are not limited to the welfare and health of the animal only, but also extends to the negative effects represented by impeding the productive performance and final products (meat and eggs) [1], [2]. Accordingly, oxidative stress can be defined as state of imbalance between the oxidation and the antioxidants system *in vivo*, which in turn causes lipid peroxidation, protein oxidation, DNA molecule damage, and obvious defects in the regulation of intracellular signaling [3], [4]. This led to find out the feed additives such as antibiotics for maximizing production, which have significant impacts on the development and growth of poultry. However, the intensive use of antibiotics was accompanied by side effects on the health for both humans and animal through the development of bacterial strains resistant to these antibiotics, as well as the accumulation of antibiotics residues in poultry products [5], [6]. Thus, to overcome the oxidative stress incidence and scavenging the free radicals, researchers have tended to use the alternative natural products such as, synbiotic and multiple medicinal plants as promising powerful antioxidants in the poultry diet.

The synbiotics is a synergistic mixture of probiotics and prebiotics that is used in different proportions as a feed additive to improve the public health and productive performance of poultry. The probiotics are consisting of beneficial microorganisms that are isolated from the intestinal flora of adult bird gut. These microorganisms envelop the receptors of intestinal epithelial cells, preventing pathogenic bacteria from obtaining adhesion sites of these cells and excluding them outside the host body. Also, the beneficial microorganisms contribute to the beneficial microbial balance of intestinal flora during exposure of adult birds to stressful conditions such as, high temperature,

toxic stress and various disease [7], [8]. The prebiotics are defined as a group of complex non-digestible carbohydrates by in vivo host enzymes such as, fructooligosaccharides and mannan-oligosaccharides, which are derived from the cell wall of bacteria, yeasts, molds and some medicinal herbs that has a beneficial medical effect [9]. These complex sugars are consumed by beneficial intestinal bacteria and have important roles in blocking the receptors located on the surface of pathogenic bacteria walls, thus preventing their adhesion to the epithelial cells lining the alimentary canal, reducing the chance of disease and promoting the animal health [10]. On basis that assumption, the synbiotic product proved its beneficial effect in histomorphological changes by increasing the thickness of the epithelial layer and villi length of intestine through its role in improving the microbial balance of intestinal flora [11],[12].

The fenugreek (*Trigonella foenum-graecum*) is one of the influential medicinal plants that is characterized by anti-stress, antitoxic, anti-microbial and hypoglycemic properties because of its richness in many polyphenols, flavonoids, saponins, alkaloids, vitamins, proteins, fatty acids and minerals such as phosphorous, iron, zinc, magnesium and calcium [13], [14].

The study aimed to the possibility of using powders of local synbiotic and fenugreek seeds as antioxidants in Ross 308 broiler diets after exposure to stress induced by H<sub>2</sub>O<sub>2</sub> drinking water and investigating their effect on productive performance and economic cost of birds until 5 weeks old of age.

## MATERIALS AND METHODS

### Experimental Scheme

In total 300 unsexed broiler chicks Ross 308 (initial weight=42.2±0.5g) were procured from a commercial hatchery and were transported and reared in Poultry Farm which belongs to the Technical College of Al-Musaib, Babylon, Iraq from December 16th, 2021 till January 19th, 2022. Chicks were randomly divided into 5 equal treatments (n=60 per treatment) with 3 replicates each. All chicks were reared under environmentally controlled conditions with feeding on balanced diet (table 1) according to NRC [15] and also based on broiler chicken guide 308. The chicks were exposed to oxidative stress from 2 days old until the end of the experiment (35 days). The experimental treatments were assigned as follows:

- i. The 1st treatment (T1): *ad libitum* feeding without any dietary additives (negative control).
- ii. The 2nd treatment (T2): adding 0.5% (1 ml) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water (positive control).
- iii. The 3rd treatment (T3): adding 0.5% (1 ml) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.5% of synbiotic powder in diet.
- iv. The 4th treatment (T4): adding 0.5% (1 ml) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.5% of fenugreek seed powder in diet.
- v. The 5th treatment (T5): adding 0.5% (1 ml) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.25% of synbiotic powder in diet + 0.25% fenugreek seed powder in diet.

**TABLE 1.** Composition of the Experimental Diet and Chemical Composition.

<b>Feed stuff</b>	<b>Starter (%) (1-2 weeks)</b>	<b>Grower (%) (3-4 weeks)</b>	<b>Finisher (%) (5 weeks)</b>
Soybean meal (46%)	30.0	26.5	26.5
Wheat	25.0	28.0	10.0
Yellow corn	20.2	14.70	32.8
Flour	12.5	15.0	15.0
Wheat bran	8.00	9.00	9.00
Premix*	2.50	2.50	2.50
Limestone	0.80	0.90	0.90
Sunflower oil	0.50	2.70	3.00
Monocalcium phosphate	—	0.30	0.20
Anti-fungal agent (MINZEL PLUS)	0.10	0.10	—
Choline chloride + Sodium	0.10	—	—
Sodium bicarbonate	0.10	0.10	—
Table salt	—	0.05	—
Threonine	0.05	—	—
Emulsifier (APSA MOS)	0.05	—	—
Emulsifier (Lysoforte™ Extend Dry)	0.04	0.04	0.04
Allicine	0.03	—	—
Enzyme mixture (Kemzyme® Plus P Dry)	0.03	—	—
Total	100	100	100
Chemical analysis**			
Crude protein (%)	21.84	20.55	19.46
Metabolizable energy (kcal/ kg)	3025.3	3094.05	3144.30
Metabolizable energy to crude protein	138.5	150.56	161.57
Crude fiber (%)	2.73	2.65	2.865
Lysine (%)	1.342	1.248	1.161
Methionine (%)	0.71	0.519	0.470
Methionine + cysteine (%)	0.880	0.862	0.796
Calcium (%)	0.910	0.902	0.810
Available phosphorous (%)	0.501	0.932	0.837

\*Produced by Provimi 3110 Company (Jordan), contains 3800 kcal/kg metabolizable energy, 7% crude protein, 1.1% fat, 15% calcium, 4% lysine, 11% available phosphorus, 4.8% sodium, 5.4% calcium, 8.5% methionine, 8.5% methionine + cysteine, 0.55% threonine, 575000 IU/kg vitamin A, 201250 IU/kg vitamin D3, 3000 mg/kg vitamin E, 138 mg/kg vitamin K3, 138 mg/mg vitamin B1, 345 mg/kg vitamin B2, 1840 mg/ kg vitamin B3, 552 mg/kg vitamin B5, 184 mg/kg vitamin B6, 46 mg/kg vitamin B9, 1000 mg/kg vitamin B12, 6900 mg/kg biotin, 20000 mg/Kg choline chloride, 2760 mg/kg iron, 3680 mg/kg zinc, 3680 mg/kg manganese, 9.2 mg/kg selenium, 50 mg/kg iodine.

\*\*Calculated by using NRC [15].

## Dietary Additives

The synbiotic powder (Iraqi synbiotic, Iraq) was locally made and suitable for using in animal feed and scientific protocols. This product was obtained from the College of Agricultural Engineering Sciences/University of Baghdad, Iraq and was an equal mixture of prebiotic and probiotic. According to manufacturer's instructions, each 1 gm of probiotic contains  $10^8$  *Lactobacillus acidophilus*,  $10^9$  *Bacillus subtilis*,  $10^8$  *Bifidobacterium* and  $10^9$  *Saccharomyces cerevisia*. With respect to prebiotic, it consists of complex sugars isolated from dry yeast *Saccharomyces cerevisiae* after damaging its cell wall by adding the chloroform at an amount of 4% and incubating in a shaker at a temperature of 30°C for 48 hours at 125 rpm.

Fenugreek seed powder was analyzed to its bioactive compounds (table 2) using the high performance liquid chromatography (HPLC) technique according to the methods coined by Anhwange *et al.* [16], Trease and Evans [17], Mradu *et al.* [18], Ghorai *et al.* [19], Abdelkader *et al.* [20], Zare *et al.* [21], Babaa and Malikba [22].

Hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>, Panreac Quimica S.L.U., Barcelona, Spain, 50%) was used as an oxidizing agent to induce the oxidative stress. H<sub>2</sub>O<sub>2</sub> was diluted with sterile distilled water to obtain concentration of 0.5% (prescribed dose). The preparation of H<sub>2</sub>O<sub>2</sub> was daily carried out to maintain its purity and effectiveness and then stored after dilution in dark plastic containers away from sunlight. 1 ml of 0.5% was added to 1 liter of drinking water and provided to birds daily [2], [3].

**TABLE 2.** Some Bioactive Compounds Present in fenugreek seed powder.

Item	Unit	Amount
Total phenols	mg gallic acid/gm	30.69
Total flavonoids	mg rutin/gm	20.45
Total alkaloids	%	13.25
Total terpinoids	%	11.36
Total tannins	%	5.36
Total saponins	%	0.58
Trigonelline	ppm	135.69

## CHARACTERISTICS STUDIED

### Productive Performance

The productive attributes of chicks were registered and presented weekly and accumulatively in each replicate in treatment. The productive parameters involved live body weight, weight gain (difference between initial and final body weight), feed intake, feed conversion ratio (feed intake / body weight gain), relative growth rate (differences between initial and final body weight × 100 divided by 0.5 × sum of initial and final body weight), periodical mortality and productive efficiency factor [23]. At age of 35 days, all birds were deprived from feed and water for 3 hours, 1 male and 3 females were selected from each replicate in treatment to obtain their final body weights and slaughtered to calculate carcass yield with giblets (heart, gizzard and liver) or without giblet and calculation ratio of abdominal fat. All the carcasses were dissected and calculated into primary and secondary parts (breast, thighs, drumsticks, wings, neck and back) [3].

### Economic Cost

The economic cost of project for all treatments was evaluated at the end of experiment (35 days) and calculated based on Iraqi dinar (IQD) currency which includes the following items:

Total Feed cost = sum of consumed feed per bird (kg) × number of birds × price of feed – [1]

Total expenses = total feed cost, dietary additives, total cost of chicks, litter, gas and medical treatments...etc – – – – – [2]

Selling revenue = body weight × price of kg for live body weight – – – – – [3]

Net revenue = difference between total expenses and selling revenue – – – – – [4]

$$\text{Economic efficiency} = \frac{\text{net revenue}}{\text{total expenses cost}} \times 100 \quad \text{--- [5]}$$

### Statistical Analysis

Data analysis was performed by using the statistical analysis system [24] to analyze the effect of different treatments on the characteristics studied by following a completely randomized design (CRD), and the mean differences among the treatments were compared according to Duncan's multiple range test [25].

## RESULTS AND DISCUSSION

### Consumed Feed

Table (3) indicated a highly significant ( $P < 0.01$ ) in the average of feed consumed for T3, T4, T5 and T1 during the periods (1-3 weeks), (4 - 5 weeks) and (1- 5 weeks) compared with T2.

**TABLE 3.** The effect of adding powders of synbiotic and fenugreek seed to the diet on feed intake (g) of stressed broilers (mean  $\pm$  standard error)

Treatments	feed intake		
	1-3 week	4-5 week	1-5 week
T1	946.17 $\pm$ 3.87 b	2206.48 $\pm$ 3.86 b	3296.93 $\pm$ 11.86 a
T2	865.56 $\pm$ 8.94 c	1847.07 $\pm$ 5.84 c	2856.94 $\pm$ 15.97 b
T3	950.08 $\pm$ 9.87 ab	2222.73 $\pm$ 2.96 a	3318.62 $\pm$ 14.82 a
T4	972.38 $\pm$ 3.95 a	2220.01 $\pm$ 6.97 ab	3339.45 $\pm$ 17.85 a
T5	972.01 $\pm$ 8.53 a	2257.38 $\pm$ 4.86 a	3369.42 $\pm$ 13.87 a
Significance level	**	**	**

T1: negative control , T2: positive control, T3: 1 ml (0.5%) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.5 % synbiotic; T4: 1 ml (0.5%) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.5 % fenugreek seed in diet ; T5: 0.5% (1 ml) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.25% of synbiotic in diet + 0.25% fenugreek seed in diet.

\*\*Different letters in the same column of treatments indicate to significant difference at ( $p \leq 0.01$ ).

### Body Weight and Weight Gain and Relative Growth Rate

The results of the statistical analysis shown in table (4) showed a significant superiority ( $P < 0.05$ ) in the average body weight in the 3rd and 5th week for T1, T3, T4 and T5 over T2 which constituted the lowest significant differences. Also, it is evident from table (4) and table (5) that there is a highly significant differences ( $P < 0.01$ ) in weight gain and relative growth rate in T1, T3, T4 and T5 for the periods (1-3 weeks), (4-5 weeks) and (1-5 weeks) compared with T2 that recorded the lowest significant differences.

**TABLE 4.** The effect of adding powders of synbiotic and fenugreek seed to the diet on body weight and weight gain (g) of stressed broilers (mean  $\pm$  standard error).

Treatments	body weight		weight gain		
	3 weeks	5 weeks	1-3 week	4-5 week	1-5 week
T1	873.06 $\pm$ 9.31 a	2381.87 $\pm$ 42.57ab	714.78 $\pm$ 2.87 ab	1508.80 $\pm$ 6.98 ab	2340.87 $\pm$ 7.98 ab
T2	765.55 $\pm$ 13.55 b	1819.03 $\pm$ 18.04 c	608.55 $\pm$ 6.98 c	1053.48 $\pm$ 8.97 c	1776.83 $\pm$ 8.98 c
T3	826.03 $\pm$ 32.19 a	2354.36 $\pm$ 19.89 b	671.18 $\pm$ 4.87 b	1528.32 $\pm$ 9.87 a	2312.15 $\pm$ 4.87 b
T4	878.68 $\pm$ 4.46 a	2366.78 $\pm$ 13.28 b	719.40 $\pm$ 7.98 a	1488.12 $\pm$ 7.54 b	2324.58 $\pm$ 8.33 b
T5	879.51 $\pm$ 10.40 a	2447.13 $\pm$ 10.82 a	722.81 $\pm$ 6.98 a	1567.61 $\pm$ 8.87 a	2404.93 $\pm$ 9.54 a
Significance level	*	*	**	**	*

T1: negative control, T2: positive control, T3: 1 ml (0.5%) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.5 % synbiotic; T4: 1 ml (0.5%) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.5 % fenugreek seed in diet ; T5: 0.5% (1 ml) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.25% of synbiotic in diet + 0.25% fenugreek seed in diet.

Different letters in the same column of treatments indicate to significant difference, \* at ( $p \leq 0.05$ ), \*\*at ( $p \leq 0.01$ ).

**TABLE 5.** The effect of adding powders of synbiotic and fenugreek seed to the diet on relative growth rate (%) of stressed broilers (mean  $\pm$  standard error).

Treatments	relative growth rate		
	1-3 week	4-5 week	1-5 week
T1	138.61 $\pm$ 1.98 a	92.70 $\pm$ 1.26 ab	193.03 $\pm$ 1.76 ab
T2	131.92 $\pm$ 4.98 c	81.52 $\pm$ 2.87 c	190.13 $\pm$ 4.98 c
T3	136.85 $\pm$ 2.54 b	96.10 $\pm$ 2.43 a	192.95 $\pm$ 5.98 b
T4	138.61 $\pm$ 1.53 a	91.70 $\pm$ 2.87 b	192.99 $\pm$ 3.87 ab
T5	139.51 $\pm$ 1.65 a	94.24 $\pm$ 4.87 a	193.21 $\pm$ 1.87 a
Significance level	**	**	*

T1: negative control , T2: positive control, T3: 1 ml (0.5%) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.5 % synbiotic; T4: 1 ml (0.5%) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.5 % fenugreek seed in diet ; T5: 0.5% (1 ml) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.25% of synbiotic in diet + 0.25% fenugreek seed in diet.

Different letters in the same column of treatments indicate to significant difference, \* at ( $p \leq 0.05$ ), \*\*at ( $p \leq 0.01$ ).

### Feed Conversion Ratio

The data presented in table (6) indicated there was a significant decrease (significant improvement) ( $P < 0.01$ ) in feed conversion ratio for T3, T4, T5 and T1 during the periods (1-3 weeks), (4-5 weeks) and (1 -5 weeks) in comparison to T2.

**TABLE 6.** The Effect of Adding Powders of Synbiotic and Fenugreek Seed to the Diet on Feed Conversion Ratio of Stressed Broilers (Mean  $\pm$  Standard Error)

Treatments	feed conversion ratio		
	1-3 week	4-5 week	1-5 week
T1	1.32 $\pm$ 0.01 b	1.46 $\pm$ 0.03 b	1.40 $\pm$ 0.01 b
T2	1.42 $\pm$ 0.02 a	1.75 $\pm$ 0.03 a	1.60 $\pm$ 0.01 a
T3	1.41 $\pm$ 0.02 a	1.45 $\pm$ 0.04 b	1.43 $\pm$ 0.02 b
T4	1.35 $\pm$ 0.03 b	1.49 $\pm$ 0.01 b	1.43 $\pm$ 0.02 b
T5	1.34 $\pm$ 0.02 b	1.44 $\pm$ 0.02 b	1.40 $\pm$ 0.01 b
Significance level	**	**	**

T1: negative control , T2: positive control, T3: 1 ml (0.5%) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.5 % synbiotic; T4: 1 ml (0.5%) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.5 % fenugreek seed in diet ; T5: 0.5% (1 ml) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.25% of synbiotic in diet + 0.25% fenugreek seed in diet. Different letters in the same column of treatments indicate to significant difference, \*\*at (p $\leq$ 0.01).

### Mortality and Productive Efficiency Factor

It is apparent from the results presented in table (7) that there are no significant differences in the percentage of total mortality among the different groups of birds.

The production efficiency factor (Table 7) reached the highest value at the significant level (P<0.05) in T1, T3, T4 and T5 in comparison to T2 which achieved the lowest value.

**TABLE 7.** The effect of adding powders of synbiotic and fenugreek seed to the diet on Total Mortality (%) and Production Efficiency Factor of Stressed Broilers (mean  $\pm$  standard error).

Treatments	total mortality	Production efficiency factor
T1	0.00 $\pm$ 0.00	484.24 $\pm$ 6.93 a
T2	0.02 $\pm$ 0.00	307.30 $\pm$ 8.83 b
T3	0.00 $\pm$ 0.00	468.85 $\pm$ 10.25 a
T4	0.00 $\pm$ 0.00	470.81 $\pm$ 7.33 a
T5	0.00 $\pm$ 0.00	499.33 $\pm$ 10.19 a
Significance level	N.S	*

T1: negative control , T2: positive control, T3: 1 ml (0.5%) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.5 % synbiotic; T4: 1 ml (0.5%) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.5 % fenugreek seed in diet ; T5: 0.5% (1 ml) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.25% of synbiotic in diet + 0.25% fenugreek seed in diet. Different letters in the same column of treatments indicate to significant difference, \* at (p $\leq$ 0.05), N.S: non significant.

### Carcass Quality

The results in table (8) referred that there was a highly significant (P<0.01) value in carcass yield dressing without giblets, with a significant superiority (p<0.05) in carcass yield with giblets for T1, T3, T4 and T5 compared with T2. With respect to percentage of giblets, there is a significant difference (P<0.01) in the relative heart weight in favor of T5 over T2. As for the relative weight of liver and gizzard, it is noted that there are no significant differences among the experimental treatments. A significant decrease (P<0.01) was noted by T3 and T4 over T2 in abdominal fat content.



Regarding carcass parts and in comparison to T2, there was a no significant differences among all experimental groups in proportional weights of breast, thighs, drumsticks, neck and back. However, T1 constituted low significant value ( $P < 0.05$ ) in proportional weight of wings compared to other treatments.

### **The Economic Cost of the Study**

There was a positive decrease in the economic cost with regard to the broilers rearing project (table 9) for T1, T3, T4 and T5 by achieving the high values accounted based on IQD currency of the mass of marketed broilers, selling revenue, net revenue and economic efficiency compared with G2 which registered negative increasing in general economic cost.

The superiority in the productive performance in T3, T4 and T5 which is represented by the average live body weight, gain weight, relative growth rate and carcass yield might probably due to the increase the amount of feed consumed under stress conditions which reflected directly on improved feed conversion ratio. The high body weights achieved in the mentioned treatments of birds might belong to the contents of their diets for powder of synbiotic (probiotic+prebiotic) and fenugreek seeds and their mixture which proved their efficacy which are characterized by containing biologically active substances to support growth and suppress the stress. This was obvious that fenugreek powder contained many bioactive compounds such as, total phenols, total flavonoids and predominant compound, trigonelline, and others (table 2) which are involved in protein synthesis and enhancing feed utilization by stimulating the secretion of insulin hormone [26], [27]. Also, this improvement in the productive attributes might be attributed to the ability of the microorganisms present in the synbiotic to improve digestion and absorption of the nutrients and then increase the efficiency of feed utilization. It was proven that the intensive presence of beneficial bacteria in synbiotics could re-form the equilibrium of microflora and increase the villi length and function in intestine [8], [11]. This supremacy in mode of action in current synbiotic was due to its containing of multiple effective bacteria such as, *L. acidophilus*, *Bifidobacterium* and *B. subtilis* bacteria, in addition to *S. cerevisiae* yeast and sugars. It was well known that multiplicity of microorganisms in the probiotic had better positive effects and more efficient than containing one type of microorganisms in same blend [29]. The rapid spread of microorganisms on the surface of the mucus layer of the intestine plays an important role in the stimulation of intestinal microflora, providing nutrients and secretion of useful substances for growth such as, amino acids, involving lysine and other metabolites [30]. Plasma circulating lysine is an essential amino acid which is important for body protein synthesis and it is derived at 21% from intestinal flora sources [31]. These intestinal flora secrete also some crucial vitamins such as, B complex, K and E vitamins which are essential for the intestinal immune system of birds and increase the rate of digestion [30]. The results of this study are in agreement with Śliżewska et al. [32] who found that using synbiotic preparation comprised of *Lactobacillus* spp, *Saccharomyces cerevisiae* yeast (probiotic) and inulin (prebiotic) could improve the feed conversion ratio and European production efficiency factor and reduced the mortality through activation the growth of beneficial bacteria and limitation in growth of potential pathogens such as, *Salmonella* in the digestive system of broiler chickens [33]. Similarly, adding 1.5 g of a synbiotic (*Bacillus licheniformis*, *Bacillus subtilis*, *Clostridium butyricum*,

**TABLE 8.** The effect of adding powders of synbiotic and fenugreek seed to the diet on Carcass Yield and Carcass Parts (%) of stressed broilers (mean  $\pm$  standard error).

Treatments	carcass yield without giblets	carcass yield with giblets	breast	thighs	drumstick	back	wings	neck	abdominal fat	liver	heart	gizzard
T1	73.74 $\pm$ 0.30 a	78.09 $\pm$ 0.32 a	40.92 $\pm$ 0.16	17.1 7 $\pm$ 0.35	13.7 1 $\pm$ 0.61	15.4 4 $\pm$ 0.08	$\pm$ 8.95 $\pm$ 0.63 b	3.77 $\pm$ 0.10	1.19 $\pm$ 0.14 ab	2.19 $\pm$ 0.06	0.56 $\pm$ 0.04ab	1.58 $\pm$ 0.12
T2	71.33 $\pm$ 0.15 b	75.27 $\pm$ 0.29 b	38.04 $\pm$ 0.19	15.77 $\pm$ 0.05	13.3 0 $\pm$ 0.15	15.6 9 $\pm$ 0. 39	12.89 $\pm$ 0.18a b	3.84 $\pm$ 0.22	1.65 $\pm$ 0.06 a	2.15 $\pm$ 0.13	0.52 $\pm$ 0.03 b	1.26 $\pm$ 0.03
T3	73.04 $\pm$ 0.80a	77.34 $\pm$ 0.63 a	38.60 $\pm$ 0.51	16.0 9 $\pm$ 0.60	13.28 $\pm$ 0.76	15.91 $\pm$ 0.35	13.38 $\pm$ 0.84 a	3.10 $\pm$ 0.17	0.97 $\pm$ 0.21 b	2.23 $\pm$ 0.041	0.63 $\pm$ 0.01a b	1.43 $\pm$ 0.18
T4	73.92 $\pm$ 0.34 a	78.45 $\pm$ 0.39 a	37.96 $\pm$ 2.15	16.3 8 $\pm$ 0.94	13.01 $\pm$ 0.68	16.59 $\pm$ 1.05	12.68 $\pm$ 1.35 ab	3.36 $\pm$ 0.02	1.13 $\pm$ 0.25 b	2.53 $\pm$ 0.13	0.63 $\pm$ 0.04a b	1.35 $\pm$ 0.10
T5	73.16 $\pm$ 0.50 a	77.91 $\pm$ a 0.52	40.49 $\pm$ 1.65	14.43 1.74 $\pm$	13.14 $\pm$ 0.61	16.92 $\pm$ 0.68	11.10 $\pm$ 2.20 ab	3.77 $\pm$ 0.38	1.36 $\pm$ 0.05 ab	2.52 $\pm$ 0.15	0.65 $\pm$ 0.03a	1.56 $\pm$ 0.14
Significance level	**	**	N.S	N.S	N.S	N.S	*	NS	**	N.S	**	N.S

T1: negative control , T2: positive control, T3: 1 ml (0.5%) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.5 % synbiotic; T4: 1 ml (0.5%) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.5 % fenugreek seed in diet ; T5: 0.5% (1 ml) of H<sub>2</sub>O<sub>2</sub> per 1 L of drinking water + 0.25% of synbiotic in diet + 0.25% fenugreek seed in diet.

Different letters in the same column of treatments indicate to significant difference, \* at (p $\leq$ 0.05), \*\* at the level (p<0.01), N.S: non significant.

**TABLE 9.** The effect of adding powders of synbiotic and fenugreek seed to the diet on Economic Cost of stressed broilers (mean  $\pm$  standard error).

Treatments*	total feed cost (10 <sup>3</sup> IQD)	total expenses (10 <sup>3</sup> IQD)	mass of marketed broilers (kg)	selling revenue (10 <sup>3</sup> ID)	net revenue (10 <sup>3</sup> ID)	economic efficiency
T1	166.34	257.83	142.91	428.73	170.90	66.40
T2	144.14	235.63	103.68	311.05	75.42	32.03
T3	167.44	258.92	141.26	423.78	164.85	63.68
T4	168.49	259.97	142.00	426.02	166.04	63.87
T5	170.00	261.49	146.82	440.48	178.99	68.48

\*According to the prices of local market in IQD currency, one chick cost is 952.38 IQD, 1 kg of feed cost is 840.90 IQD, total cost of feed additives, litters, medical treatments and gas per group is 26000 IQD.

*xylooligosaccharide* and yeast cell wall) had positive impacts on growth performance represented by feed efficiency during days 1–21 and reduced the relative abdominal fat weight, however, no effect was recorded on daily feed intake, weight gain, feed efficiency from 1-50 day old of Partridge shank chickens [34]. Perhaps, that synbiotic extracts promoted antioxidant capacity, regulated nutrients metabolism and digestibility [34] and modulated the hematological and biochemical parameters of poultry [35]. The powerful activity of synbiotic might depend on dose incorporated in diet of broiler chickens. For instance, more recently, Nisar et al. [6] concluded the best dose was 1200 g/ton from synbiotic product (Synerall™) involving unique combination of *Saccharomyces cerevisiae*, galacto-oligosaccharide and mannanoligosaccharide) compared with 700, 1700 or 2200 g/ton. Thus, 1200 g/ton improved feed conversion ratio but reduced feed intake without any obvious changing in body weights and produced carcass quality. The improved performance of birds under stress condition in current data was probably that synbiotic extracts considered as the best preparations in comparison with an individual using of its components (probiotic or prebiotic). On basis that, these results were in line with Abdel-Fattah and Fararh [35] who suggested that dietary commercial synbiotic (0.5 kg of probiotic Lactic dry® /ton plus 1 kg of prebiotic Bio-MOS® /ton ) improved body weight gain, feed conversion ratio, breast muscle percentage and economic efficiency of broilers compared with singular using of either 0.5 kg of probiotic (*Saccharomyces cerevisiae*, *Lactobacillus acidophilus*, *Streptococcus faecium*, *Bacillus subtilis*) or 1 kg of prebiotic (phosphorylated mannanoligosaccharide derived from cell wall of *Saccharomyces cerevisiae*) per ton. Additionally, Cheng et al. [36] noticed that adding 1.5 g per kg of dietary synbiotic consisting (*Bacillus subtilis*, *Clostridium butyricum* and *Bacillus licheniformis* ) and prebiotics (xylooligosaccharide and yeast cell wall) increased average daily gain, reduced feed conversion ratio and abdominal fat in broilers from 1 to 42 days of age.

Similar series of previous studies proved the improvement in body weight, weight gain, feed intake and feed conversion ratio of broiler chickens fed 1% of fenugreek seed in diet [37], [38] or 2% of essential oil of fenugreek in diet [39]. Differently from what was obtained by Yesuf et al. [40], that adding 1 and 2 g/kg of fenugreek seed powder did not change the carcass yield at the age of 49 days, however, Toaha et al. [14] found that was significant increase in the dressing percentage of broiler fed 1, 2 and 3 % of fenugreek powder incorporated in experimental diet.

Lowering in economic efficiency for T2 in present data was inverted by the significant deterioration in overall productivity of birds due to chronic implications of oxidative stress. This was due to increase oral consumption of cytotoxic H<sub>2</sub>O<sub>2</sub> which is involved in overproduction of oxygen molecules and generation of other

harmful free radicals in stomach and functional tissues. Thus, free radicals can permeate easily the blood stream and causes augmentation the lipid peroxidation *in vivo* [41], [42]. They play detrimental effects by damaging biological molecules, such as proteins, lipids, and DNA which causes in turn deactivation the cellular membranes composition and related influences on transcription, translation and RNA processing [43], [44]. Oxidative stress is responsible for many pathological cases such as, cancer, obstructive pulmonary disease, neurological disorders, atherosclerosis, asthma, hypertension, diabetes, acute respiratory distress syndrome, and many cellular consequences [45]. H<sub>2</sub>O<sub>2</sub> is characterized by its diffusion and ubiquity in cells and therefore its excretion controlled by specific catabolism [46]. Although, the negative effect of H<sub>2</sub>O<sub>2</sub>, we did not registered any mortality incidence among all groups of stressed birds but the effects were obvious in final productive performance and economical point of view. Identical results were achieved by Al-Shammrai et al. [2], Al-Shammrai et al. [4] and Al-Shammari and Batkowska [47], who found that was a clear decrease in productive attributes and production efficiency factor of broilers or quails exposed to oxidative stress by 0.5% H<sub>2</sub>O<sub>2</sub> in drinking water or 200 ppm lead acetate in diet.

The amelioration in the productive performance obtained by the antistress feed additives (T3, T4 and T5) was positively reflected by the values of the production efficiency factor which is one of the important indicators in evaluating the productivity and the economic cost of the project. The high value in this indicator is due to the correlation among high live body weight and improved feed conversion ratio thus the increase in this value led to an increase in financial returns [23]. Therefore, superiority in productive characteristics of dietary antistress treatments of stressed birds was profitable for monetary savings which resulted in decreasing the total expenses cost of the rearing project with an augment in net revenue and overall economic efficiency accounted in IQD currency. There seems no doubt that powders of locally made synbiotic and fenugreek seed will optimize the highly mass production, lower the economic losses with inferior economic cost especially in case of feed prices which constitutes the most pivotal requirements in poultry nutrition.

## CONCLUSION

The oxidative stress induced experimentally by hydrogen peroxide posed physiological risks on live organisms which were reflected by impaired growth performance and productive features of broiler chickens until marketing age. However, the locally made synbiotics and fenugreek powder added to the diet alleviated the deleterious influence of stress by their specific mode of action. Also, these both feed additives had positive influence to reduce the economic cost of the rearing project which could open a new vista in understanding the sustainable development in poultry nutrition.

## ACKNOWLEDGEMENTS

The authors gratefully acknowledge Prof. Dr. Saad Abd Al-Hussien Naji, College of Agricultural Engineering Sciences/University of Baghdad, Iraq for providing Iraqi synbiotic powder as free donation for scientific research of the MSc degree for 1st author and his assistance is greatly appreciated.

## REFERENCES

1. Surai, P. F. Karadas, N. Sparks, "The importance of antioxidant in poultry". Annual north Carolina Poultry Nutrition conference, 38-56 (2003).
2. Al-Shammari, K.I.A., Batkowska, J. and Zamil, S.J., "Role of pomegranate peels and black pepper powder and their mixture in alleviating the oxidative stress in broiler chickens", *Int. J. Poult. Sci*, 18, 122-128 (2019a).

3. K.I.A Al-Shammari, PhD thesis, University of life Sciences in Lublin, Poland, 2017, The use of chosen plant extracts in various aspects of Japanese quail production.
4. Al-Shammari, K.I.A., Zamil, S.J. and Mohammed, E.M., "Influence of dietary epigallocatechin-3 gallate and L-arginine and its combination on early laying performance and physiological status of stressed Japanese quails", *Journal of Physics: Conference Series*, 1294, 9, 092014 (2019b).
5. Sahin, O., Morishita, T.Y. and Zhang, Q., "Campylobacter colonization in poultry: sources of infection and modes of transmission", *Animal Health Research Reviews*, 3(2), 95-105 (2002).
6. Nisar, H., Sharif, M., Rahman, M.A., Rehman, S., Kamboh, A.A. and Saeed, M., "Effects of dietary supplementations of synbiotics on growth performance, carcass characteristics and nutrient digestibility of broiler chicken", *Brazilian Journal of Poultry Science*, 23(2), 1-10 (2021).
7. Zinedine, A., Faid, M. and Benlemlih, M., "In vitro reduction of aflatoxin B1 by strains of lactic acid bacteria isolated from Moroccan sourdough bread", *Int J Agric Biol*, 7(1), 67-70 (2005).
8. Mahfuz, S.U., Nahar, M.J., Mo, C., Ganfu, Z., Zhongjun, L. and Hui, S., "Inclusion of probiotic on chicken performance and immunity: A review", *International Journal of Poultry Science*, 16(9), 328-335 (2017).
9. Markowiak, P. and Śliżewska, K., "The role of probiotics, prebiotics and synbiotics in animal nutrition", *Gut pathogens*, 10 (1), 1-20 (2018).
10. Kim, S., Jang, M.J., Kim, S.Y., Yang, Y., Pavlidis, H.O. and Ricke, S.C., Potential for prebiotics as feed additives to limit foodborne Campylobacter establishment in the poultry gastrointestinal tract. *Frontiers in microbiology*, 10, 91 (2019).
11. Beski, S.S.M. and Al-Sardary, S.Y.T., "Effects of dietary supplementation of probiotic and synbiotic on broiler chickens hematology and intestinal integrity", *International Journal of Poultry Science*, 14(1), 31-36, (2015).
12. Yaqoob, M.U., Abd El-Hack, M.E., Hassan, F., El-Saadony, M.T., Khafaga, A.F., Batiha, G.E., Yehia, N., Elnesr, S.S., Alagawany, M., El-Tarabily, K.A. and Wang, M., "The potential mechanistic insights and future implications for the effect of prebiotics on poultry performance, gut microbiome, and intestinal morphology", *Poultry Science*, 100(7), 101143, (2021).
13. Adil, S., Qureshi, S. and Pattoo, R.A., "A review on positive effects of fenugreek as feed additive in poultry production", *International Journal of Poultry Science*, 14(12), 664-669 (2015).
14. Toaha, S.M., Mollah, B.R. and Ahammad, M.U., "Use of dietary fenugreek (*Trigonella foenum-graecum* L.) seed for the production of safe broiler lean meat", *Research in Agriculture Livestock and Fisheries*, 3 (2), 305-314 (2016).
15. NRC. Nutrient Requirements of Poultry, National Research Council, 9th rev. ed, (National Academic Press, Washington, DC, 1994).
16. Anhawange, A.A., Ajibola V.O. and Oniye S.J. "Chemical studies of the Seeds *Moringa oleifera* (LAM) *Detarium microcarpum* (Guill and Perr)", *Journal of Biological Sciences*, 4, 711-715 (2004).
17. G.E Trease and W. C. Evans. Trease and Evans' Pharmacognosy. (Philadelphia: WB Saunders, Elsevier Science Limited., p. 336, 2009).
18. Mradu, G., Saumyakanti S., Sohini M. and Arup M., "HPLC profiles of phenolic compounds present in medicinal plants", *International Journal of Pharmacognosy and Phytochemical Research*, 4, 162-167 (2012).
19. Ghorai, N., Chakraborty S., Gucchait S., Saha S.K. and Biswas S., "Estimation of total terpenoids concentration in plant tissues using a monoterpene, linalool as standard reagent, Protocol Exchange, 27(11), (2012).
20. Abdelkader, M., Ahcen B., Rachid D. and Hakim H., "Phytochemical study and biological activity of sage (*Salvia officinalis* L.), "International Journal of Bioengineering and Life Sciences, 8, 1253- 1257 (2014).
21. Zare, K., Nazemyeh H., Lotfipour F., Farabi S., Ghiamirad M. and Barzegari A., "Antibacterial activity and total phenolic content of the *Onopordon acanthium* L. Seeds", *Pharmaceutical Sciences*, 1, 6-11 (2014).
22. Babaa, S.A. and Malikba S.A. "Determination of total phenolic and flavonoid content, antimicrobial and antioxidant activity of a root extract of *Arisaema jacquemontii* Blume", *Journal of Taibah University for Science*, 9, 449– 454 (2015).

23. Lemme A., Frackenpohl U., Petri A., Meyer H. "Response of male BUT big 6 turkeys to varying amino acid feeding programs", [Poult. Sci.](#), 85, 652-660 (2006).
24. SAS, SAS/STAT User's Guide for Personal Computers. Release 9.1 (SAS Institute Inc., Cary, N.C., USA., 2012).
25. Duncan, D.B. "Multiple Rang and Multiple F -test", [Biometrics](#),11, 4-42 (1955).
26. Khosla, P., Gupta, D.D. and Nagpal, R.K., "Effect of *Trigonella foenum graecum* (Fenugreek) on blood glucose in normal and diabetic rats", *Indian journal of physiology and pharmacology*, 39, 173-173 (1995).
27. Sauvaire, Y., Petit, P., Broca, C., Manteghetti, M., Baissac, Y., Fernandez- Alvarez, J., Gross, R., Roye, M., Leconte, A., Gomis, R. and Ribes, G., "4-Hydroxyisoleucine: a novel amino acid potentiator of insulin secretion". [Diabetes](#), 47(2), 206-210 (1998).
28. Mahdavi, A.H., Rahmani, H.R. and Pourreza, J., "Effect of probiotic supplements on egg quality and laying hen's performance". [Int. J. Poult. Sci.](#), 4(7), 488-492 (2005).
29. Methner, U., "Administration of autochthonous intestinal microflora--a method to prevent Salmonella infections in poultry" *DTW. Deutsche Tierärztliche Wochenschrift*, 107(10), 402-408 (2000).
30. Gabriel, I., M. Lessire, S. Mallet and J.F. Guillot, "Microflora of the digestive tract: critical factors and consequences for poultry", *World's Poultry Science Journal*, 62, 3, 499-511 (2006).
31. Metges, C.C., "Contribution of microbial amino acids to amino acid homeostasis of the host", [The Journal of nutrition](#), 130(7), 1857S-1864S (2000).
32. Śliżewska, K., Markowiak-Kopeć, P., Żbikowski, A. and Szeleszczuk, P., "The effect of synbiotic preparations on the intestinal microbiota and her metabolism in broiler chickens", [Scientific reports](#), 10(1),1-13 (2020).
33. Sobotik, E.B., Ramirez, S., Roth, N., Tacconi, A., Pender, C., Murugesan, R. and Archer, G.S., "Evaluating the effects of a dietary synbiotic or synbiotic plus enhanced organic acid on broiler performance and cecal and carcass Salmonella load" [Poultry Science](#), 100(12), 101508 (2021).
34. Li, J., Cheng, Y., Chen, Y., Qu, H., Zhao, Y., Wen, C. and Zhou, Y., "Effects of dietary synbiotic supplementation on growth performance, lipid metabolism, antioxidant status, and meat quality in Partridge shank chickens", [Journal of Applied Animal Research](#), 47(1), 586-590 (2019).
35. Abdel-Fattah, F.A. and Fararh, K.M., "Effect of dietary supplementation of probiotic, prebiotic and synbiotic on performance, carcass characteristics, blood picture and some biochemical parameters in broiler chickens" *Benha Vet. Med. J.*, 20, 9-23 (2009).
36. Cheng, Y., Chen, Y., Li, X., Yang, W., Wen, C., Kang, Y., Wang, A. and Zhou, Y., "Effects of synbiotic supplementation on growth performance, carcass characteristics, meat quality and muscular antioxidant capacity and mineral contents in broilers", [Journal of the Science of Food and Agriculture](#), 97(11), 3699-3705 (2017).
37. Weerasingha, A.S. and Atapattu, N.S.B.M., "Effects of Fenugreek (*Trigonella foenum-graecum* L.) seed powder on growth performance, visceral organ weight, serum cholesterol levels and the nitrogen retention of broiler chicken", [Tropical Agricultural Research](#), 24(3):289-295 (2013).
38. Mamoun, T., Mukhtar, M.A. and Tabidi, M.H., "Effect of fenugreek seed powder on the performance, carcass characteristics and some blood serum attributes", *Advance Research in Agriculture and Veterinary Science*, 1(1), 6-11 (2014).
39. H. H. Hamid, Master thesis, Sudan University of Science and Technology ,Sudan, 2018, Effect of feeding of adding fenugreek oil on performance and blood serum profile of broiler chicks.
40. Yesuf, K.Y., Mersso, B.T. and Bekele, T.E., "Effects of different levels of turmeric, fenugreek and black cumin on carcass characteristics of broiler chicken", *Journal of Livestock Science*, 8, 11-17 (2017).
41. Mousa, Y.J. and Mohammad, F.K., "Effects of hydrogen peroxide on diazepam and xylazine sedation in chicks. [Interdisciplinary Toxicology](#)", 5 (4), 179–183 (2012).
42. Mousa, Y.J., "Anaesthetic properties of ketamine in chicks stressed with hydrogen peroxide", [Veterinari Medicina](#), 59 (8), 369–75 (2014).
43. Yoshikawa T. and Naito Y, "What is oxidative stress?", *Journal of the Japan Medical Association*, 45 (7) , 271–6 (2002).

44. Fellenberg, M.A. and Speisky, H., "Antioxidants: their effects on broiler oxidative stress and its meat oxidative stability", [World's Poultry Science Journal](#), 62 (1), 53-70 (2006).
45. Birben, E., Sahiner, U.M., Sackesen, C., Erzurum, S. and Kalayci, O., "Oxidative stress and antioxidant defense", [World allergy organization journal](#), 5 (1), 9-19 (2012).
46. Halliwell, B., Clement, M.V. and Long, L.H., "Hydrogen peroxide in the human body", [FEBS letters](#), 486 (1), 10-13 (2000).
47. Al-Shammari, K.I.A. and Batkowska, J., "The antioxidative impact of dietary vinegar and rocket salad on the productivity, serum oxidation system, and duodenal histology of chickens", [Animals](#), 11(8), 2277 (2021).