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Improving the utilization of distilled dried grains with soluble by enzymes and yeast supplementations for production and economic sustainability and digestibility of laying hens

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Abstract. a study was conducted to test the influence of different concentrations of distillers dried grains with soluble (DDGS) with or without a commercial enzyme preparation (Kemzyme plus dry®) at 0 and 500 g/ ton feed and *Saccharomyces cerevisiae* at 1 kg/ton feed in native Inshas laying hens' diets to evaluate the effects on production and economic sustainability, nutrients digestibility, nitrogen, and phosphorus retention. A total number of 270 Inshas laying hens, local 28-week-old were randomly divided into nine experimental groups, each group containing three replicates of 10 hens. The experimental treatments include three levels (0, 10, and 20%) of DDGS without or with two additives supplementation. The study revealed that body weight and the productive performance of laying hens were not significantly affected by dietary treatments. Feed intake decreased progressively with increasing DDGS levels; however, enzyme and yeast supplementation completely recovered the adverse influence of DDGS. Both nitrogen and P retention increased with increasing DDGS, in contrast, N and P excretion decreased as DDGS increased, but enzymes or yeast had no effects within each DDGS level. Moreover, there was a significant increase in the relative economic efficiency due to feeding laying hens with diet containing 10% DDGS compared to the control. It was concluded that corn DDGS could be provided to laying hens up to 20% without adverse effects on productive and economic efficiency while adding enzymes or *Saccharomyces cerevisiae* decreased N and P excretion, suggesting positive environmental impacts.
Keywords: Distillers dried grains with soluble; laying hens; Enzymes; *Saccharomyces cerevisiae*

1. Introduction

Poultry nutrition plays an important and major role in poultry production. Recently, an increase in the price of conventional feed ingredients and their shortage are the major limiting factors for the uplift and development of the poultry industry in developing countries. Therefore, there is an urgent need to search for alternative ingredients which could be fed as cheap sources of either partial or full replacement for conventional feedstuffs such as corn and soybean meal in developing countries (US.Grains Council, 2011; Salim *et al*., 2010).

Distillers Dried Grain with Soluble (DDGS) can be an effective alternative. It is a by-product of ethanol production. Because of the high content of readily fermentable starch, corn is the main grain used in the ethanol industry. During fermentation, equal amounts of ethanol, carbon dioxide (CO_2), and DDGS are formed (Lumpkins *et al*., 2005; Jones *et al*., 2022). Ethanol production from 100 kg of corn is about 34.4 kg of ethanol, 34.0 kg of CO2, and 31.6 kg of DDGS (Renewable Fuels Association, 2005). Using DDGS in poultry diets was essentially a source of unidentified growth factors at low inclusion levels (Noll *et al*., 2007; Shirisha *et al*., 2021; Prasad, 2022) and can enhance the sustainability of poultry breeding during a crisis such as COVID-19 and recent Ukraine-Russia war (Hafez and Attia., 2020).

Research shows that DDGS is higher in non-starch polysaccharides (NSP) than in the parent grain. Instead, mono-gastric such as chickens do not efficiently digest feedstuffs high in NSP. As a result, the metabolizable energy (ME) of DDGS was lower than in corn (2820 vs. 3350 kcal/kg; NRC, 1994). Therefore, NSP enzyme addition could improve nutrient availability of DDGS diets for poultry (Ward *et al.*, 2008). In this connection, Ghazalah *et al.* (2011) showed that laying hens could be fed diets containing 40 % DDGS as a replacement for corn (24 % of diet) with supplemented enzymes without any adverse effect on laying hen performance. Ghazalah *et al.* (2012) concluded that broiler chicks could tolerate different levels of DDGS up to 60 % as a replacement of soybean meal with or without supplemental enzymes, without any adverse effect on broiler performance.

Probiotics are other approaches that have the potential to reduce enteric disease in poultry and subsequent contamination of poultry products (Gibson and Fuller, 2000). A probiotic has been defined as a live microbial feed supplement which beneficially affects the host animal by improving gut health (Fuller, 1989). Dietary supplementation of *B. subtilis* has been observed to reduce NH_3 emissions in poultry by improving the activity of enzymes and the utilization of N (Tanaka and Santoso, 2000; Miles *et al.*, 2004). However, the efficacy of probiotics could be affected by many factors, such as the age of animals, the strain of microorganisms, and inclusion level (Chen *et al.*, 2006).

Therefore, this feeding trial was conducted to evaluate the utilization of different levels of DDGS (0, 10, and 20% of diet) without or with Kemzyme enzymes and /or *Saccharomyces cerevisiae* supplementation on productive performance, nutrient digestibility, nitrogen, and phosphorus retention and economic efficiency of laying hens during 28-48 weeks of age.

2. Materials and Methods

2.1. Ingredients

The tested material, DDGS golden in color, suggesting reasonable heating during processing. The other ingredients used in formulating the experimental diets were obtained from a local market.

2.2. Enzyme and yeast preparation:

The products used are Kemzyme and yeast. Kemzyme plus dry is an enzyme produced in Kemin Europa N.V. Kemin Industries, Inc. ® ™ Trademarks of Kemin Industries, Inc., USA (Kemzyme® PLUS Dry, euro code E 1620, and containing five different enzymes: 2,350 U/g endo-1,3(4)-beta-glucanase 4,000 U/g endo-1,4-beta-glucanase, 400 U/g alpha-amylase, 450 U/g bacillolysin 20,000 U/g endo-1,4-beta-xylanase). The *Saccharomyces cerevisiae* preparation used in the present study is a commercial product by Alltech, NicholaSiville, Kentucky, USA, containing active yeast at 7×10^8 CFU/gram.

2.3. Experimental birds, design and diets:

A total number 270 Inshas laying hens were randomly weighted and then divided into nine experimental groups, each group containing three replicates of 10 hens. The experimental design included three levels (0, 10 and 20%, respectively) of DDGS without or with enzyme and/or *S. cerevisiae* supplementation (unsupplemented control, Kemzyme plus dry® supplementation at 500 g/ton and *S. cerevisiae* supplementation at 1000 g g/ton*)* under a 3 × 3 factorial arrangement. The additives was supplemented at the top of the diet. The chemical composition of feeds are shown in Table 1. Birds of each replicate were housed in floor pens (280 cm x 220 cm) furnished with rice hulls. Birds were provided with 16h light and 8h dark daily. All groups were reared under similar hygienic and managerial conditions. Throughout the experimental period (28-48 weeks), feed and fresh water were continuously available.

Table 1: Composition and calculated analysis of experimental diets

Ingredients	Control diet	10% DDGS	20 % DDGS
Yellow corn	64.00	60.69	57.0
Soybean meal 44% CP	23.70	18.30	12.76
Wheat bran	1.85	0.70	0.00
DDGS	0.00	10.00	20.00
Limestone	8.00	8.00	8.06
Dicalcium phosphate	1.63	1.53	1.41
NaCl	0.45	0.33	0.21
Vitamin and Mineral premix	0.30	0.30	0.30
DL- Methionine	0.07	0.06	0.05
L-lysine-HCl	0.00	0.09	0.21
Calculated			
CP (%)	16.04	16.01	16.01
ME (Kcal/kg)	2,700	2,706	2,704
CF (%)	4.64	4.93	5.25

Methionine	0.36	0.36	0.37
Methionine+Cystine	0.62	0.62	0.62
Lysine	0.79	0.79	0.79
Ca (%)	3.46	3.44	3.42
Av. P (%)	0.43	0.43	0.42
Na (%)	0.19	0.19	0.19
Determined analyses%			
Dry matter	89.80	89.56	89.68
Crude protein	16.54	16.48	16.41
Ether extract	2.70	3.52	4.38
Crude fiber	4.78	4.82	5.00

Supplied per kg of diet: Vit.A, 12000 IU; D_3, 2200 IU; Vit. E, 10mg; Vit.K_3, 2mg; vit.B_1, 1mg; Vit. B_2, 5mg; vit. B_6, 1.5mg; Vit. B_{12}, 10mcg; Niacin, 30mg; Pantothenic acid, 10mg; Folic acid, 1mg; Biotin, 50µg; Choline, 260mg; Copper, 10mg; Iron; 30mg; Manganese, 60mg; Zinc, 50mg; Iodine, 1.3mg; Selenium, 0.1mg and Cobalt, 0.1mg

Table 2. Chemical composition of the DDGS on dry matter basis (%)

Dry matter (%)	**Crude protein (%)**	**Ether extract (%)**	**Crude fiber (%)**	**Ash (%)**	**ME (kcal/kg)**
10.95	27.15	10.03	9.30	4.30	2,500

The calculated metabolizable [as feed basis (92.52% DM) 1772 kcal/kg] using the equation published by European Table of Energy Values for Poultry Feedstuffs 3rd edition (Ir. W. M. M. A. Janssen, 1989) for Distilled dried grains with soluble:

AMEnkj/kg= DM × 16.38 + ash × -16.38 + CP × -4.066 +crude fibre × -26.7.

2.4. Parameters studied:

Body weight, egg production, egg weight and mass, feed consumption and feed conversion ratio per egg mass (kg feed /kg egg) were assessed during the experimental period using the pen as experimental unit. Initial body weight at the beginning and the final body weight at the end of the experiment for laying hens were measured, and the average body weight gain was calculated as difference between the two body weights.

2.5. Digestibility trial

A total of 27 Inshas cocks in the last week of the experiment were used to determine the digestion coefficients of nutrients of the experimental diets. Birds were randomly selected (3 cocks/treatment) and housed individually in metabolic cages with the facility of excreta collection. The experimental diets and water were offered *ad libitum.* Feed intake was recorded, and excreta were collected quantitatively for 5 days. The excreta were sprayed with 1% boric acid to prevent any loss in ammonia during the drying period at 60°C for 24 hrs. Fecal nitrogen was estimated according to Jakobsen *et al.* (1960).

2.6. Determination of Nitrogen and Phosphorus Retention

The following steps were followed to determine N and P retention. Diets were mixed with Chromic oxide (Cr, 0.1% of the feed) as an indigestible marker and were fed for 1 week before the fecal collection. Clean excreta (from feathers and feed) were collected after a 24 h production period and samples were placed in -20°C freezers for 2 d. The apparent retention of N and P was determined using equations for the indicator method described by Schneider and Flat (1975) and Masadeh (2011): % Nutrient retention = 100 – 100 × ((% Cr analyzed in the diet (×) % nutrient analyzed in the feces) / (% Cr analyzed output in the feces (×) % analyzed nutrient in the diet)). Further, the total nutrients excreted per kilogram of DM consumed (DMI) were calculated using the ratio of Cr intake to Cr output (Dilger and Adeola, 2006 Masadeh, 2011).

Nutrient output, g/kg of DMI = NcE × (Cr diet/Cr out), where NcE is the concentration of the respective nutrient in the excreta, Cr diet is the initial Cr concentration in the diet, and Cr output is the concentration of Cr in the excreta.

2.7. Economic efficiency

Economic efficiency of egg production was estimated from the input-output analysis that was calculated according to the price of the experimental diets and egg production. The values of economic efficiency were calculated as the net revenue per unit of the total cost (Soliman, 2002**)**.

2.8. Statistical analysis

Data were analyzed using a two-way analysis of variance with DDGS levels, feed additives, and their interactions using the General Linear Model procedure of SAS (2008).

, according to the following model:

$Y_{ijkl} = \mu + T_i + M_j + (T*M)_{ij} + E_{ijk}$

Where

Yijkl = Trait measured

μ = Overall mean

Ti = effect of DDGS levels (i=1, 2, 3)

Mj = effect of feed additives (j=0, 1, 2)

(T*M)ij = Interaction between DDGS and additive type

Eijk = Experimental error

3. Results and Discussion

3.1 Chemical analyses of DDGS

The chemical analyses of DDGS are shown in Table 2. The results indicated dry matter was 89.05%, CP 27.15, ether extract 10.03, crude fiber 9.30, ash 4.30% and ME is 2500 kcal /kg diet. The value agree with those reported in Cromwell *et al*. (1993); NRC (1994) and

Ghazalah *et al*. (2011; 2012).

3.2 Productive performance and mortality

Results in Table 3 showed that increasing DDGS level from 0 to 20% had no significant effect on body weight measurements, egg production traits, FCR and the number of dead hens. Body weight gain and body weight change indicated no significant differences due to dietary treatments and their interactions.

The laying rate ranged from (61.5-63.0%), egg weight (49.4-50 g) and egg mass (30.4-31.3 g). These changes in egg production (2.4%), egg weight (1.2%), and egg mass (3.0%) were not significant due to different DDGS, feed additives, and the interaction between DDGS and the additives.

Feed intake was significantly affected by the interaction DDGS x aditives, enzyme and SC supplementation to 10 and 20% DDGS diets significantly and similarly increased feed intake compared to their corresponding control (Table 3). The results indicated that enzymes and SC yield similar effects on feed intake.

These results agree with those reported by Lumpkins *et al.* (2005), who suggested that feeding laying hens on a diet containing 10-12% DDGS had no significant effects on laying performance. In addition, Roberson *et al*. (2005) reported that 15% of DDGS did not adversely affect the performance of laying hens and suggested that a low level of DDGS is preferred when included DDGS in the diet for laying hens. Also, Swiatkiwicz and Korelwski (2006) and Noll *et al.*, (2007a; b) reported that up to 15% DDGS could be included in layers' diets, and 10 % DDGS for laying hens had no negative effects on egg production parameters (Roberts *et al.*, 2007).

Table 3. Productive performance as affected by distilled dried grains with soluble, Kemzyme and *Saccharomyces cerevisiae*

Dietary treatments		**Body weight**				**Productive performance**					
		Initial BW, g	**Final BW, g**	**BW G, g**	**BW change, %**	**Rate of laying, %**	**Egg weight, g**	**Egg mass, g/h/d**	**Feed intake, g/h/d**	**FCR, g feed / g egg**	**Dead birds, *n***
Main effect of distilled dried grains with soluble											
DDGS %	0	1486	1518	31.4	2.11	62.1	49.8	30.9	94.3[a]	3.05	1
	10	1492	1522	29.8	2.00	62.7	49.6	31.1	93.9[b]	3.02	1

	20	1488	1515	26.8	1.80	61.9	49.5	30.6	93.5^{b}	3.06	0
Main effect of additives											
Additive	0	1488	1514	26.3	1.77	61.9	49.5	30.7	93.4^{b}	3.05	0
	Kemzyme	1491	1523	31.8	2.13	62.5	49.7	31.0	94.1^{a}	3.04	0
	SC	1488	1518	29.8	2.00	62.2	49.7	30.9	94.2^{a}	3.05	1
Interaction between DDGS and additive											
DDGS %	Additive										
0	0	1484	1511	26.4	1.78	61.8	49.6	30.6	94.3^{a}	3.07	0
0	Kemzyme	1486	1520	34.6	2.32	62.5	49.8	31.1	94.4^{a}	3.03	0
0	SC	1489	1523	33.2	2.22	62.0	50.0	31.0	94.3^{a}	3.04	1
10	0	1491	1518	26.8	1.80	62.5	49.6	31.0	93.4^{b}	3.01	0
10	Kemzyme	1498	1533	34.2	2.28	63.0	49.7	31.3	94.2^{a}	3.01	0
10	SC	1487	1515	28.4	1.91	62.5	49.5	30.9	94.2^{a}	3.04	0
20	0	1488	1514	25.7	1.72	61.5	49.4	30.4	92.7^{c}	3.05	0
20	Kemzyme	1488	1515	26.8	1.80	62.0	49.6	30.7	93.8^{ab}	3.06	0
20	SC	1487	1515	27.8	1.87	62.2	49.5	30.8	94.2^{a}	3.05	0
SEM		6.49	8.55	6.51	0.439	0.846	0.163	0.498	0.219	0.045	-
Probability											
DDGS %		0.544	0.601	0.681	0.685	0.523	0.138	0.508	0.002	0.608	-
Additive		0.815	0.500	0.584	0.597	0.728	0.497	0.656	0.001	0.929	-
DDGS × additive		0.806	0.717	0.966	0.972	0.992	0.534	0.982	0.050	0.954	-

$^{a-c}$Means within the same column within the same parameter not sharing similar superscripts were significantly different ($p \leq 0.05$). SEM: standard error of the mean, DDGS: distilled dried grains with soluble, SC: *Saccharomyces cerevisiae*, BW: body weight, BWG: body weight gain, FCR: feed conversion ratio

In addition, Cheon *et al.* (2008) reported that a 20% DDGS diet did not influence egg production, egg weight, egg mass, and feed intake.

Our results agreed with Masadeh *et al.* (2011), who found that increasing levels of DDGS from 0 to 25% for White Leghorn-type hens did not harm feed intake, egg weight, and egg production. Along the same line, Hassan and Al Aqil (2015) reported that DDGS can be safely added to diets as an alternative source of protein and energy up to 20% without negative effects on productive performance characteristics of Hisex laying hens from 30 to 42 weeks of age. On the other hand, Schideler *et al.* (2008) concluded that the inclusion of up to 25% DDGS in laying hens' diets decreased the egg weight of hens fed 20% and 25% DDGS because of amino acid deficiency. Further, Shalash *et al.* (2010) reported that increasing DDGS to 15 or 20 % in laying hen diets markedly decreased egg production %, egg number, and egg weight while not influencing feed intake compared to the other levels (0,5 and 10%). In addition, the inclusion of 16.5-20 % DDGS in laying diets adversely affected laying performance (Lumpkins *et al.,* 2005 Roberson *et al.,* 2005; Ghazalah *et al.* 2011; Świątkiewicz and Koreleski, 2006& 2008; Abd El-Hack *et al.,* 2017). These authors attributed the reduction in productive performance caused by inclusions of DDGS to the high crude fiber, unpalatable and sulfur content of the DDGS (Pineda *et al.*, 2008) and lysine deficiency (Sherr *et al.,* 1998) and Hansen and Millington (1997). These authors reported that the lysine deficiency may be due to the mallard reaction which reduces the digestibility of lysine by competing with the absorption of lysine or inhibition of carboxyl peptidases. Thus, a diet containing 20% DDGS had a low level of starch because most of the starch is converted to ethanol during fermentation (Creswell, 2006). This means that the hens relied solely on converting part of dietary amino acids to glucose through gluconeogenesis pathway to keep normal blood glucose concentration and relied increasingly on fatty acid oxidation to supply energy. The DDGS contained relatively high amounts of fat, protein, and fiber, but with variable availability (Abudabos *et al*., 2017; Saeed *et al.,* 2017). The contradiction in the previously mentioned results due to DDGS feeding may be due to different process conditions. Thus, the quality of DDGS and level of inclusion, the composition of the experimental diets, and consideration is given to balancing the amino acids by synthetic amino acids supplementations. SC significantly increased feed intake of the diets containing 10 and 20% DDGS showing complete recovery of feed intake. Moreover, the findings showed that SC supplementation numerically increased egg weight in the control diet, and this could be due to increasing feed intake and availability of nutrients for egg formation (Hajati *et al.,* 2009). Obviously, yeast culture, and its cell wall extract containing 1,3-1, and 6 D-glucan and Mannan oligosaccharide are essential natural growth promoters for modern livestock and poultry production (Van Leeuwen *et al.,* 2005a). The advantages of these promoters over the traditional antibiotic growth promoters are 1) no withdrawal time, 2) no residual effect, and 3) no causes of microbial mutation (Gibson and Roberfroid, 2008). *Saccharomyces cerevisiae* is observed as one of the live microorganisms' probiotics that, when given through the gut, has a positive impact on the host's health through its direct nutritional effect. Field reports (Banday and Risam, 2002) have suggested that probiotic addition improved broilers' performance. The different actions of probiotics suggested are nutritional effects by regulation of metabolic reactions that produce toxic substances, stimulation of endogenous enzymes, and by the production of vitamins or antimicrobial substances. Moreover, SC could act as a bio-regulator of the intestinal microflora and reinforce the host's natural defenses through the sanitary effect by raising the colonization resistance and stimulating the immune response (Line *et al.*, 1998). These impacts were largely reflected by using mannan oligosaccharide, the naturally derived extract from the cell wall of *Saccharomyces cerevisiae*. This oligosaccharide content is approximately 50% of the carbohydrate fraction and enhance body weight gain in broiler chickens. That this influence can be attributed to the trophic effect of this product on the intestinal mucosa, as it increases villus height, particularly during early chickens' life (Santin *et al.*, 2001). Oligosaccharides are supplied to control pathogenic scours of all kinds in livestock caused by *Salmonella*, *E. coli*, etc (Laegreid and Bauer, 2004). Mannan oligosaccharides are thought to block the attachment of pathogenic bacteria to the animal's intestine and colonization that may result in disease while acting as a nutrient to other beneficial bacteria. It is also thought to stimulate the animal's immune system, thereby further reducing the risk of disease (Firon and Ofek, 1983). Oyofo *et al.* (1989) observed that the adherence of *Salmonella typhimurium* to enterocytes of the small intestine of chicks *in vitro*, was inhibited in the presence of mannose. Later, they observed that inclusion of mannose in the drinking water of chickens' reduced *S. typhimurium* colonization of the cecum.

With consideration of the type of microorganisms used in different experiments and the present one, Gallazzi *et al.* (2008) noted that there were no improvements in laying performance, egg weight, feed intake, or feed efficiency in hens fed diets supplemented with *Lactobacillus acidophilus*. On the other side, probiotics did not affect egg weight and FCR (Panda *et al.*, 2003), feed intake egg weight, and laying rate, (Panda *et al.*, 2008), egg weight, feed intake and feed utilization (Quarantelli *et al.*, 2008), egg weight and laying rate (Balevi *et al.*, 2009), feed intake, laying rate, and feed utilization (Ramasamy *et al.*, 2009), or laying rate and feed intake (Mikulski *et al.*, 2012).

Enzymes supplementation had a slight enhancing effect on egg production, egg weight, egg mass, and FCR, but markedly increased feed consumed in the diets containing 10 and 20% DDGS showing complete recovery of feed intake. The beneficial effects of exogenous enzyme addition on improving the performance of chickens could be due to increased feed intake found herein. In addition, enzymes were observed to decrease viscosity and reduce the anti-nutritional effect of NSP, leading to better performance (Van der Klis *et al.,* 1995; Mathlouthi *et al.,* 2003). Świątkiewicz *et al.* (2013) suggested that DDGS could be included up to 20% in laying hens' diets without adversely impacting egg performance. Moreover, xylanase and phytase additions, as well as inulin and chitosan, can positively affect the performance of layers given diets with high levels of DDGS. In literature, feed additives such as yeast and enzymes have been used to improve the sustainability of poultry production and overcome the anti-nutritional effects and environmental pollution due to improving nutrient utilization and decreasing excretion. In the present study, enzyme supplementation (depends on enzyme type, composition, dose and the target substrate, and diet composition) increased the nutritive value of DDGS for chickens (Oryschak *et al.*, 2010), and the feed efficiency of laying hens (Abd El-Hack *et al*., 2017) and broilers (Latorre *et al*., 2017). Similarly, Abudabos (2010) found a significant increase in growth for broilers fed corn-soy diets supplemented with enzymes. Also, Arce *et al.* (2010) reported that adding protease and xylanase enzymes to broilers' diets that contain maize, soybean meal, and DDGS yielded the best results in terms of broiler performance. However, no marked differences were observed in FCR values because of enzyme supplementation. In addition, Świątkiewiczand Koreleski (2006 & 2008) found that the addition of NSPases in the 44- to 68-week feed-in phase helped offset the drop-in laying rate and daily egg mass in diets with 20% DDGS versus the non-supplemented diet. Along the same line, Ghazalah *et al.* (2011) indicated that Avizyme supplementation relatively improved ($P \leq 0.05$) egg production, egg mass, and FCR for DDGS inclusion levels at 25 and 50% instead of SBM when compared to those fed diets without Avizyme supplementation. Also, results suggest that diets containing DDGS at less than 15.45% level (50% of SBM) with Avizyme 1500® addition could enhance the nutritive value of DDGS for layers. In addition, Al-Harthi *et al*. (2009) and Alsaffar *et al*. (2013) reported that egg laying performance significantly improved by enzyme addition. Moreover, a multi-enzyme preparation containing a variety of enzymes, which degrade cell walls and liberate nutrients, improved laying performance (Nelson, 1989). Instead, El-Deek *et al.* (2003) observed that egg production% was not significantly affected by enzyme addition or type of diet.

There was recorded low mortality rate during the different experimental periods due to feeding laying hens' feed containing different levels of DDGS. These results agree with those of Shalash *et al.* (2009a,b) and Fassbinder *et al.* (2010) who recorded no mortality due to using DDGS in both broiler and layer diets.

3.3 Nutrient digestibility

The effect of dietary DDGS treatments on nutrient digestibility is summarized in Table 4. Results showed that no substantial differences were observed in the digestion coefficients of OM, DM, CP, CF, and EE while NFE was not affected Instead, there were no significant effects of enzymes and SC on the digestibility of OM, DM, CP, CF, EE, and NFE. The current results support with those observed by Ghazalah et al. (2011), who reported no substantial effects due to DDGS levels observed on digestibility coefficient values for CP, CF, EE, and NFE, and this was like those also observed by Shalash *et al.* (2010). On the other hand, Abd El-Hack (2015) recorded that digestibility values of nutrients were preferable in hens' fed diets containing 5.5% DDGS than the control diet and other DDGS treatment groups containing 11, 16.5, and 22%. Diets with high digestion increase the performance of the animals and consequently decrease the excretion of nitrogen, lowering the risks of environmental contamination (Bertechini, 2003).

SEM: standard error of the mean, DDGS: distilled dried grains with soluble, SC: *Saccharomyces cerevisiae*, DM: Dry matter, OM: Organic matter, CP: Crude protein, CF: Crude fiber, EE: Ether extract, NFE: Nitrogen free extract

3.4 Nitrogen and phosphorus retention

Apparent nitrogen (N) and phosphorus (P) retention and nutrients output in the excreta are displayed in Table (5). There were significant effects of only DDGS level on the N and P excretion and N and P retention. On the other hand, enzyme, SC, and the interaction between DDGS and feed additives had no significant effect on the % apparent retention and % N and % P output in the excreta. The results indicated that % N retention increased by 9.3 and 9.9% with the inclusion of 10 and 20% DDGS in the laying hens' diets. In addition, there was a stepwise decrease of 17.9 and 22.9% in % nitrogen excreted with increasing % DDGS in the laying hen diets. The results also indicated a stepwise decrease in the % phosphorus retention by 13.3 and 20.2% and phosphorus output in the excreta by 29.5 and 42.0%, with elevating DDGS to 10 and 20%, respectively. These present results agree with Santoso *et al.* (1999) and Tanaka and Santoso, (2000). They reported that dietary supplementation with *B. subtilis* has been observed to reduce NH3 emissions in poultry by enhancing the activity of enzymes and the utilization of N. Moreover, Masa'deh *et al.* (2011) observed that laying hens fed 25% DDGS increased N retention compared to hens on 0, 5, 10, 15, and 20% DDGS. In addition, there was a linear increase in N and P retention as DDGS levels increased, and the highest inclusion rate of DDGS (25%) resulted in the greatest ($p \leq 0.001$) N and P retention compared to lower levels of DDGS. In addition, Roberts *et al.* (2007a) reported that providing laying hens with 10% corn DDGS reduced N emission by 51% compared to corn-soybean meal diet. In addition, Wu-Haan *et al.* (2010) reported a decrease in daily ammonia emissions as the amount of DDGS increased from zero to 20%. The present results indicate that DDGS has a high bioavailability of N and P (Cromwell *et al.*, 1993; Shurson, 2003). In addition, DDGS is a good source of P, containing 0.72% total P (NRC, 1994) and the bioavailability of P is higher than the 25 to 35% that is typical of most plant ingredients.

Table 4.Nutrients digestibility as affected by distilled dried grains with soluble, Kemzyme and *Saccharomyces cerevisiae*

Dietary treatments		**Nutrients digestibility, %**					
		DM	**OM**	**CP**	**CF**	**EE**	**NFE**
Main effect of distilled dried grains with soluble							
DDGS, %	0	76.5	78.5	79.1	21.9	72.0	73.3
	10	76.5	78.5	79.0	22.2	72.6	72.9
	20	76.5	78.3	78.8	22.5	72.0	72.2
Main effect of additives							
Additive	0	76.3	78.8	78.7	22.0	71.7	72.9
	Kemzyme	76.5	78.3	79.2	22.1	72.3	72.8
	SC	76.7	78.3	79.0	22.4	72.5	72.7
Interaction between DDGS and additive							
DDGS, %	Additive						
0	0	76.2	78.5	79.00	21.4	71.9	73.1
0	Kemzyme	76.5	78.4	79.16	21.8	71.7	73.4
0	SC	76.9	78.8	79.10	22.4	72.3	73.4
10	0	76.5	79.0	78.60	22.1	72.0	73.6
10	Kemzyme	76.6	78.4	79.56	22.4	72.8	72.6
10	SC	76.3	78.1	78.83	22.2	72.9	72.4
20	0	76.1	78.9	78.40	22.4	71.3	72.0
20	Kemzyme	76.6	78.2	79.00	22.2	72.3	72.4
20	SC	77.0	78.0	79.00	22.7	72.3	72.2
SEM		0.264	0.775	0.408	0.485	0.445	0.516
Probability							
DDGS, %		0.937	0.948	0.680	0.363	0.206	0.064
Additive		0.123	0.705	0.250	0.487	0.142	0.867
DDGS × additive		0.270	0.939	0.811	0.822	0.765	0.555

It is well known that a major portion of N and P retention are used for supporting egg formation rather than maintenance (NRC, 1994). The N and P compounds of poultry fecal content are a potential pollutant-causing nitrite or nitrate water contamination, eutrophication, ammonia volatilization, and acid deposition in the air (Alagawany *et al.*, 2014, 2015). Therefore, reducing N and P excretion in poultry manure is an important concept for environmental sustainability (Alagawany and Abou-Kassem, 2014). Roberts *et al.* (2007b) found an high and positive correlation between the amount of DDGS in the diet and both consumption of N and excretion by the birds. However, the inability to digest fiber properly by poultry may provide environmental benefits when feeding DDGS. Microbes ferment undigested dietary fiber deriving from DDGS in the hindgut producing short-chain fatty acids that, in turn, lower the manure pH. Thus, minimized manure pH causes a lower production of the volatile ammonium form of N that does not evaporate and consequently has a less harmful influence on air quality (Babcock *et al.*, 2008; Bregendahl *et al.*, 2008). Abd El-Hack and Mahgoub (2015) reported that excreted nitrogen declined by 8.62 and 4.31% in laying hens fed 5 or 10 % DDGS, respectively, while excreted P depressed by 3.33, 7.22, and 10.56% in hens on 5, 10 or 15% DDGS, respectively compared to those fed the basal diet.

Table 5. Apparent retention of N and P and excretion of fecal N and P as affected distilled dried grains with soluble, Kemzyme and *Saccharomyces cerevisiae*

Dietary treatments		***Apparent retention, %***		***Nutrient output, g/kg of DM intake***	
		N	***P***	***N***	***P***
Main effect of distilled dried grains with soluble					
DDGS, %	0	42.9[b]	21.8[c]	14.0[a]	4.57[a]
	10	46.9[a]	24.7[b]	11.5[b]	3.22[b]
	20	47.1[a]	26.2[a]	10.8[c]	2.65[c]
Main effect of additives					
Additive	0	45.8	24.2	12.1	3.6
	Kemzyme	45.9	24.2	12.2	3.4
	SC	45.7	24.2	12.1	3.4
Interaction between DDGS and additive					
DDGS, %	Additive				
0	0	42.8	21.8	14.2	4.76
0	Kemzyme	43.0	21.9	14.0	4.60
0	SC	42.9	21.8	14.3	4.36
10	0	46.8	24.8	11.6	3.30

10	Kemzyme	47.0	24.5	11.5	3.16
10	SC	46.9	24.9	11.5	3.20
20	0	47.1	26.2	10.8	2.80
20	Kemzyme	47.1	26.2	11.1	2.50
20	SC	47.2	26.2	10.7	2.66
SEM		0.221	0.327	0.221	0.228
Probability					
DDGS, %		0.0001	0.0001	0.0001	0.0001
Additive		0.809	0.932	0.858	0.458
DDGS × additive		0.981	0.964	0.800	0.908

[a-c] Means within the same column within the same parameter not sharing similar superscripts were significantly different ($p \leq 0.05$). DDGS: distilled dried grains with soluble, SC: Saccharomyces cerevisiae, SEM: Standard error of the mean, N: Nitrogen, P: Phosphorus, DM: Dry matter.

The high levels of P in DDGS-containing diets could be beneficial to minimize the costs of supplementation with this expensive macro element and reduce the need for supplementing diets with inorganic P (Lumpkins and Batal, 2005). In addition, Wu-Haan *et al.* (2010) reported a decrease in daily ammonia emission as the amount of DDGS increased from zero to 20%, which could bring additional benefits to the environment and poultry workers (Attia *et al.*, 2014). Instead, corn DDGS contains a relatively high sulfur level, leading to elevated hydrogen sulfide emissions that are voided in poultry letters. Pineda *et al.* (2008) hypothesized that both NH_3 and hydrogen sulfide emissions can harm egg production. Therefore, feeding laying hens on DDGS has positive environmental implications by reducing and lack of any adverse effects on air quality.

3.5 Economic efficiency

The effect of different dietary levels of DDGS with or without additives supplementation on the economic efficiency is presented in Table 6. The cost of feeding was reduced linearly with increasing DDGS level. Results showed a numerical increase in the average values of net revenue, economic efficiency, and a significant increase in relative economic efficiency due to feeding 10% DDGS to laying compared to the control. On the other hand, feed additives had no significant effect on all the economic traits.

There was no significant interaction between DDGS and feed additives on most of the economic traits except for feeding cost. The results showed that enzymes decreased the feeding cost of hens fed 10% DDGS compared to the enzyme-supplemented control group, while SC supplementation to 20% DDGS diets significantly decreased the cost of feeding compared to SC-supplemented control and 10% DDGS diets.

The highest economic efficiency and relative economic efficiency were from the enzyme-supplemented 10% DDGS group, followed by the enzyme-supplemented 20% DDGS group and 10% DDGS control. This indicates that DDGS could be supplied to laying hens up to 10-20% without enzyme supplementation or when supplemented with an enzyme. After the linear increase in prices of corn, soybean meal, and other feed ingredients, the poultry industry has been working to minimize feed costs of production. After the use of corn and soybean for biofuel production, alternative feed ingredients and feed additives are employed to sustain feed resources for animal production (Alagawany and Attia, 2015; Ashour *et al.*, 2015; Saeed *et al.*, 2017; Abudabos *et al.*, 2017; Latorre *et al.*, 2017). In addition, DDGS has been proven as an effective cost-saving ingredient (Schilling *et al.*, 2010; Lumpur, 2017). Studies in Egypt by leading poultry

Table 6. Economic efficiency as affected by distilled dried grains with soluble, Kemzyme and *Saccharomyces cerevisiae*

Dietary treatments		**Items**				
		Total feeding cost, LE[1]	**Total revenue, LE[2]**	**Net revenue, LE[3]**	**Economic efficiency**	**Relative economic efficiency,%[4]**
Main effect of distilled dried grains with soluble						
DDG%	0	78.2[a]	108.7	30.5	0.30	100.0[b]
	10	76.9[b]	109.7	32.8	0.42	108.7[a]
	20	75.9[c]	108.3	32.4	0.42	105.5[ab]
Main effect of additives						
Additive	0	77.0	108.4	31.4	0.40	100.0
	Kemzyme	77.1	109.4	32.3	0.41	107.1
	SC	76.9	108.9	32.0	0.41	102.6
Interaction between DDG % and additives						
DDG%	Additive					
0	0	77.9[a]	108.2	30.3	0.38	100.0
0	Kemzyme	78.3[a]	109.4	31.1	0.39	101.1
0	SC	78.4[a]	108.5	30.1	0.38	98.0
10	0	77.0[ab]	109.4	32.4	0.42	107.1
10	Kemzyme	76.4[b]	110.3	33.8	0.44	112.8
10	SC	77.2[ab]	109.4	32.2	0.41	106.2
20	0	76.1[b]	107.6	31.6	0.41	105.7
20	Kemzyme	76.5[b]	108.5	32.0	0.41	107.3
20	SC	75.2[c]	108.8	33.6	0.44	103.6
SEM		0.132	1.48	1.46	0.188	3.63
Probability						
DDGS%		0.0001	0.514	0.152	0.045	0.018
Additive		0.449	0.715	0.758	0.754	0.332
DDGS x Additive		0.0001	0.991	0.832	0.669	0.974

[a-c] Means within the same column within the same parameter not sharing similar superscripts were significantly different ($p \leq 0.05$).DDGS: distilled dried grains with soluble, SEM: Standard error of the mean, L.E = Egyptian pound, 0= control, Kem=Kemzyme, SC=*Saccharomyces cerevisiae*[1]According to the price of different ingredients available in the market at the experimental time (2015). [2] According to the local market price at the experimental time (2015).[3] Net revenue per unit cost.[4]Compared to the economic efficiency of the control group.

nutritional labs indicated that high-quality DDGS is an economically viable feed ingredient and suitable alternative feedstuffs for poultry feeding (Ghazalah *et al.*, 2011). Laying hens fed a diet containing 50% DDGS substitution rate with Avizyme for soybean meal was economically viable (Ghazalah *et al.*, 2011). Recently, Abd El-Hack (2015) showed that laying hens fed a diet containing 11% DDGS substitution for soybean meal resulted in the best economic efficiency value compared with other 0, 5.5, and 16.5% DDGS. Moreover, net revenue values, economic efficiency, and relative economic efficiency were enhanced due to feeding broiler chicks 20, 40, or 60% DDGS replaced for soybean either without or with enzymes compared to the control group (Ghazalah *et al.*, 2012). Instead, feeding costs increased somewhat using DDGS, especially at the 20% level DDGS. However, the feed cost of 15% DDGS group was very much comparable to that of 0% DDGS group (Cheon *et al.*, 2008; Rew *et al.*, 2009).

Conclusion

Based on the findings of this study, it can be concluded that corn DDGS could be included in the laying hens' diets up to 20% Supplemented with enzymes (500 mg/ton) and yeat (1 kg/ton), without adverse effects on productivity, nutrient digestibility, and economic efficiency while decreasing N and P excretion, suggesting also a positive environmental impact and leading also to an improvement of poultry production sustainability.

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تحسين الاستفادة من نواتج تقطير الحبوب المجففة مع السوائل بإضافة الإنزيمات والخميرة لاستدامة الانتاج واقتصاديات التربية والهضم للدجاج البياض

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مستخلص. هدفت هذه الدراسة تحسين الاستفادة من نواتج تقطير الحبوب المجففة مع السوائل بإضافة مخلوط انزيمات تجارية أو خميرة الخبار الي علف الجاج البياض من سلالة انشاص وتأثيرهم على استدامة الأداء الإنتاجي، والكفاءة الاقتصادية، والهضم واحتجاز النتروجين والفوسفور. واستخدمت ٢٧٠ دجاجة بياضة عمر ٢٨ أسبوعا حبست في ٢٧ عش واحتوي كل عش على (١٠ دجاجات + ١ ديك). للتزاوج الطبيعي، وقسمت الدجاج عشوائياً إلى ٩ مجموعات تجريبية، كل مجموعة تحتوي على ثلاث مكررات تشمل المعاملات التجريبية ثلاثة مستويات (٠، ١٠ و ٢٠٪) من نواتج تقطير الحبوب المجففة مع السوائل بدون أو مع إضافة مستحضرات الإنزيم التجارية Kemzyme ®plus dry (بمعدل ٥٠٠ جم / طن علف) وهو خليط من "α-amylase و β-glucanase و cellulase و protease و xylanase) وخميرة الخباز بمعدل ١ كجم / طن علف وأظهرت النتائج أن وزن الجسم والأداء الإنتاجي للدجاج البياض لم يتأثروا بشكل معنوي بالمعاملات الغذائية، ولكن استهلاك العلف انخفض بشكل تدريجي مع زيادة مستوى نواتج تقطير الحبوب المجففة مع السوائل. ومع ذلك، فإن الإنزيم وخميرة الخباز اصلاحت تمامًا التأثير الضار لـ نواتج تقطير الحبوب المجففة مع السوائل. زادت معدلات الاحتجاز بشكل معنوي مع زيادة نواتج تقطير الحبوب المجففة مع السوائل، بينما انخفض إفراز النتروجين والفوسفور في الزرق مع زيادة نواتج تقطير الحبوب المجففة مع السوائل، ولكن لم يكن لـ Kemzyme و/أو خميرة الخباز أي تأثير داخل كل مستوى من نواتج تقطير الحبوب المجففة مع السوائل. زادت الكفاءة الاقتصادية النسبية مع تغذية الدجاج البياض على علف يحتوي على ١٠٪ من نواتج تقطير الحبوب المجففة مع السوائل مقارنة بمجموعة الكونترول. واتضح أن نواتج تقطير الحبوب المجففة مع السوائل يمكن استخدمه في تغذية الدجاج البياض حتى ٢٠٪ بدون إضافات الانزيمات أو الخميرة. ولكن الاضافات قللت التأثير الضار على الكفاءة الإنتاجية والاقتصادية مع انخفاض إفراز النيتروجين والفوسفور بالزرق، مما يشير إلى تأثيرات بيئية إيجابية للاضافات مثل الانزيمات والخميرة.

الكلمات الدالة: نواتج تقطير الحبوب المجففة مع السوائل، الدجاج البياض؛ الإنزيمات. خميرة الخميرة