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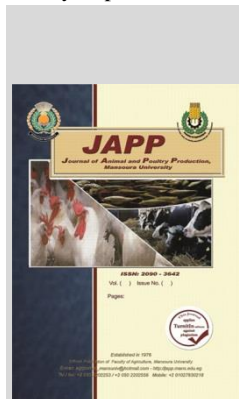
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### Does Dietary Yeast Extract Improve the Performance and Health of Quail Breeders Reared under High Stocking Density?

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#### ABSTRACT

This study was designed to study the effects of stocking density without or with yeast extracts supplementation on productive and reproductive performance, digestive enzymes, blood biochemical parameters, and antioxidant and immune measurements of quail breeders. A total number of 336 Japanese quail birds at 8 weeks old were used. Quails were randomly distributed into 6 groups and 4 replicates maintained / each group in a complete randomized design. The 1<sup>st</sup> group contained 36 birds (24 females and 12 males) was stocked at a rate of 9 birds /replicate (control, ND; 36 quails/m<sup>2</sup>) and fed the basal diet without any supplementation; the 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup>, 5<sup>th</sup> and 6<sup>th</sup> groups contained 60 quails (40 females and 20 males) and stoked at a rate of 15 chicks/replicate (HD; 60 quails/m<sup>2</sup>) and fed on a basal diet supplemented with 0, 1, 2, 3 and 4 mg YE /kg diet, respectively. Egg number, egg weight and egg mass were significantly decreased (P<.0001) due to increasing stocking density from ND (9/cage) to HD (15/cage) and fed without dietary YE supplementation. The activity of SOD (p<0.0001), TAC (p<0.0001) and GPX (p=0.0047) was improved with HD plus YE when compared to HD. Digestive enzymes (amylase, lipase, and protease) activity was decreased with HD when compared to ND. It could be concluded that addition of yeast extract can positively mitigate the stress applied to quail raised under high stoking density by enhancing productive and reproductive performance, lipid profile, antioxidants and immunity as well as egg quality criteria.

**Keywords:** yeast, stocking density, performance, blood, quail breeders.

#### INTRODUCTION

Japanese quail is considered one of the important alternative resources of animal protein. Quails have many advantages such as early sexual maturity, small size and high egg production, fast growth, short incubation period, low feed requirements, and less floor space compared with the different species of birds (Padmakumar *et al.*, 2000). Moreover, quails are widely distributed in different countries of the world (Roshdy *et al.*, 2010 and El-Tarabany *et al.*, 2015).

Stocking density increases animals' welfare but lowering it without guaranteed optimal environmental circumstances is of minimal importance (Utnik-Banaś *et al.*, 2014). The optimal housing condition for growing poultry is of considerable interest to researchers, and adequate production conditions are necessary to boost poultry production and improve welfare and profit (Lewko and Gornowicz, 2011; Mesa *et al.*, 2017). Housing conditions can also affect animal welfare and mortality of laying hens (Weimer *et al.*, 2019; Schuck-Paim *et al.*, 2021).

Worldwide quail production has contributed to the poultry sector and will have a bright future due to low production costs and satisfy customer expectations (Mohamed *et al.*, 2019; Abou-Kassem *et al.*, 2019). The objective of the quail producers is to raise the stocking density to obtain further reductions in production costs. However an excessive crowding of chicks might affect performance and wellbeing of chickens maintained in cages (Tayeb *et al.*, 2011; Utnik-Banaś *et al.*, 2014). To this regard, no clear advice is provided in the literature on

ideal space allowance for quail raising. Stocking stress linked with rising animal density can be alleviated by adding feed supplements with significant antioxidant potential (Attia *et al.*, 2021).

The significance of feed supplements/additives in the chicken sector is quite essential (Bolacali and Irak, 2017; El-Kholy *et al.*, 2019). The proper usage of feed additives can improve feed utilization, production and public health. Probiotics such as yeasts (*Saccharomyces cerevisiae*; SC) are one of the prominent feed additives and have been used as feed additives for improving animal health and performance (Ogbuewu *et al.*, 2018). Probiotics are live microorganisms that boost birds' health by competing with bad bacteria, improving the gut balance of microbiota and absorption of nutrients (Al-Khalafah, 2018; Tomaszewska *et al.*, 2018).

The SC is classified as one of the most yeast species that are introduced to chicken feeds during diet formulations (Duarte *et al.*, 2012). The SC contains high levels of nutrients such as proteins, vitamins and minerals, as well as it contains polysaccharides  $\alpha$ -D-mannan,  $\beta$ -D-glucan and chitin (Elghandour *et al.*, 2019) which play a key role in improving microbial balance in poultry intestine (Alizadeh *et al.*, 2016). Therefore, this work was designed to study the effects of stocking density without or with yeast extracts supplementation on productive and reproductive performance, digestive enzymes, blood biochemical parameters, antioxidant and immune measurements and egg quality of laying quail.

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## MATERIALS AND METHODS

### Experimental birds and management

The present study was carried out at the quail unit, Poultry Department, Faculty of Agriculture, Zagazig University, Egypt. A total number of 336 Japanese quail birds (224 females and 112 males) at 8 weeks old were used. Quails were randomly distributed into 6 experimental groups and 4 replicates maintained / each group in a complete randomized design. The 1<sup>st</sup> group contained 36 birds (24 females and 12 males) was stocked at a rate of 9 birds /replicate (control, ND; 36 quails/m<sup>2</sup>) and fed the basal diet without any supplementation; the 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup>, 5<sup>th</sup> and 6<sup>th</sup> groups contained 60 quails (40 females and 20 males) and stoked at a rate of 15 chicks/replicate (HD; 60 quails/m<sup>2</sup>) and fed on a basal diet supplemented with 0, 1, 2, 3 and 4 mg YE /kg diet, respectively, (HD + yeast extract). Quails were housed in wire cages of an open farm during the experiment which started at 8 weeks till 16 weeks of age. Feed and water were available throughout the experiment. All laying hens were subjected to 16 hours of continuous light per day during the laying period. All experimental birds were raised under similar environmental, hygienic and managerial conditions.

### Experimental diets

The basal experimental diets were formulated to cover the nutrient requirements of laying Japanese quail as recommended by NRC (1994). The composition and calculated analyses of the experimental basal diets are presented in Table 1.

**Table 1. Composition and calculated analysis of the experimental basal diet.**

Items	Ingredient	(%)
Yellow Corn (8.5%)		58.70
Soybean meal (44%)		27.50
Gluten meal (62%)		4.60
Soybean oil		1.80
Limestone		5.54
Di-calcium phosphate		1.20
Salt		0.30
Premix <sup>1</sup>		0.30
L-Lysine		0.06
DL-Methionine		0.00
Choline chloride (50%)		0.00
Calculated composition <sup>2</sup> (%)		
ME, Kcal /Kg		2901
Crude protein		20.00
Calcium		2.50
Nonphytate P		0.35
Lysine		1.00
Total sulfur amino acids (M+CY)		0.70

<sup>1</sup>Layer Vitamin-mineral Premix Each 1Kg contains of vit. A, 8000 IU ;vit. D3, 1300 ICU, vit. E 5 mg ;vit. K, 2 mg ;vit B1 ,0.7 mg; vit. B2, 2 mg; vit.B6, 1.5 mg; vit. B12, 7 mg; Biotin 0.1 mg; Pantothenic acid , 6 g; Niacine, 20 g ; Folic acid, 1 mg , manganese , 60 mg ; Zinc, 50 mg, Copper , 6 mg ; Iodine, 1 mg , Selenium, 0.5 mg ; Cobalt, 1mg.

<sup>2</sup>Calculated according to NRC (1994).

### Measurements investigated

#### Egg production

Egg number per hen was calculated by dividing the number of eggs laid by the number of hens for each replicate. Each day, eggs were weighed individually to the nearest 0.01 g for each replicate and the average was

calculated. Egg mass was calculated by multiplying egg number by average egg weight.

#### Feed utilization

Weight of feed consumed (g) per each replicate per period was calculated by subtracting the amount of feed left from that supplied. Regarding feed conversion ratio, the amount of feed (g) required to produce one gram of eggs) was calculated by dividing the amount of feed consumed by egg mass produced.

#### Egg quality traits

Egg produced in the last 4 days of each period (2week) were collected and then used for egg quality measurements. All egg quality traits (egg shape index, egg shell percent, shell thickness, albumen weight percentage, yolk index, yolk weight percentage, Haugh units, and unit surface shell weight (USSW) (mg/cm<sup>2</sup>)) were determined according to Romanoff and Romanoff (1949), Yalcin *et al.*, (1990).

#### Reproductive Performance

At the end of each month, eggs from each treatment were collected and incubated. After hatching, all chicks were counted to compute fertility and hatchability percent, and also non-hatched eggs were broken (Reda *et al.*, 2020).

#### Biochemical characteristics

Individual blood samples were obtained into EDTA tubes from 4 birds within each treatment (on individual basis) at 5 weeks of age to determine the distinct biochemical features. Each blood sample was spun at 4000 rpm for 15 minutes to separate blood plasma. The colorimetric determination of cholesterol was carried out utilising kit provided by Bio-system according to Allain *et al.* (1974). (1974). The idea of the approach is that cholesterol forms a colourful complex with acetic anhydride and concentrated sulfuric acid and the coloured complex is measured photometrically. The colorimetric determination of triglycerides was carried out by a particular diagnostic kit provided by Bio diagnostic according to Allain *et al.* (1974). (1974). After centrifugation, the high-density lipoproteins (HDL), low density lipoproteins (LDL) and the very low-density lipoprotein and the very low-density lipoprotein (VLDL) were determined by enzymatic methods (Myers *et al.*, 1994).

#### Immunity indices

The levels of immunoglobulin G (IgG) and A (IgA) in the plasma were determined spectrophotometrically using a commercial kit from Biodiagnostic Company (Giza, Egypt) according to (Akiba *et al.*, 1982). The authors determined the level of complement (C<sub>3</sub>) in quail plasma, using the ELISA Kit from MyBiosource.com.

#### Antioxidant parameters

Plasma samples were subjected to the measurement of superoxide dismutase (SOD) and glutathione peroxidase (GPX) activity and total antioxidant capacity (TAC) according Koracevic, *et al.* (2001). Plasma malondialdehyde (MDA) was determined according to Mihara and Uohiyama, (1978).

#### Digestive enzymes

Digestive enzymes including the activity of amylase, lipase and protease were determined by the method of Tietz and Fiereck (1966) and Lynn and Clevette-Radford (1984), respectively.

**Statistical analysis**

The differences among treatments were statistically analyzed by one- way ANOVA using the SAS General Linear Models Procedure (SAS. 2002) by adopting the following model:

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

**Where:**  $X_{ij}$  = An observation,  $\mu$  = Overall mean,  $T_i$  = Effect of treatments ( $i = 1, 2, \dots$  and 6).  $e_{ij}$  = The experimental random error. The significant differences between treatment means were separated by Tukey's Multiple Range-test ( $P < 0.05$ ).

**RESULTS AND DISSCUTION**

**Productive performance (egg number, egg weight and egg mass)**

Throughout the interval periods studied (8- 12, 12- 16 and 8- 16 weeks of age) the results in Table 2 showed that, means of egg number, egg weight and egg mass were significantly decreased ( $P < .0001$ ) due to increasing stocking density from ND (9/cage) to HD (15/cage) and fed without dietary YE supplementation. Adding YE to diets of HD groups led to increasing the aforementioned traits compared to group housed at normal density and HD without dietary YE supplementation (Table 2).

The results observed productive performance due to stocking density accordance with previous studies (Ratriyanto *et al.*, 2020; El-Sheikh *et al.*, 2016; El-Trarbany *et al.*, 2015) who found that, high stocking density decrease egg production, egg weight, egg number and egg mass in Japanese quail. Other observations in these birds (Italian quails) demonstrated that, increasing the stocking density from 38 to 47 qualis/m<sup>2</sup> did not affect their egg production, while increasing the stocking density to more than 56 quail/m<sup>2</sup> causes a decrease in their egg production (Faitarone *et al.*, 2005). Similarly, Mousavi *et al.* (2016) and Kang *et al.* (2016) showed, reducing floor space adversely impacted the laying hens egg production without affecting the egg weight and feed conversion ratio.

The unfavorable effects due to high stocking density may be attributed to the alteration of resting behavior due to disruptions from other birds (Attia *et al.*, 2012). In addition, a high stocking density results in a higher temperature cage due to overcrowding, reduced air quality due to inadequate airflow and increased ammonia (Abudabos *et al.*, 2013; Kang *et al.*, 2016).

The results obtained of YE supplementation in the diet of HD groups agree with Pinar *et al.* (2013) who reported that, dietary supplementation of 1 % inactivated. Yeast in brown layers at 22 weeks old for 14 days increased hen-day production percentages. Likewise, Swain *et al.* (2011) stated that, dietary yeast supplementation at 1.5 g/kg improved egg weight in Vanaraja layer. Moreover, Hassanein and Soliman (2010) demonstrated addition of 0.4 % or 0.8 % yeast culture in layer diets improved egg production.

Contradicting results were obtained by Sanaa (2013) who reported that use of dietary yeast at 0.3- 0.6 % lowered laying rate in birds aged 26 weeks. Also, Dizji and Pirmohammadi (2009) observe that laying hens fed 0.04 % has been reported to lay smaller eggs vs. the control group.

Liu *et al.*, (2002) found an improvement in egg weight for Hy-Line hens (20-wks of age) when fed on diets containing commercial yeast culture product at levels 0.1, 0.2 and 0.3%. Furthermore, Osman (2003) reported that feeding Hy-Line White hens on diets supplemented with the probiotic mixture at levels 1, 2, 3 and 4 g/kg diet resulted in higher egg rate, egg weight and egg mass compared to the control group. Moreover, Kurtoglu *et al.* (2004) found that supplementation of commercial probiotic at 250, 500 and 750 mg/kg increased production of eggs but decreased the ratio of damaged egg. Diets contain live yeast culture increased egg production and egg weight when turkeys were fed phosphorous-deficient diets (Thayer *et al.*, 1978). In addition, Songsak *et al.*, (2009) reported that supplementation of cassava yeast to 26-wk laying hens had a significant positive effect on egg weight but has negative effect on egg production. Abd EL-Maksoud *et al.* (2011) observed significant improved in egg production and egg mas with adding 250g Ag diet yeast to local hens.

Abu-Taleb *et al.* (2006) found that average egg production was significantly increased due to adding yeast by 1% or 1.5% into diets of laying quail when laying Japanese quail was fed on basal diets supplemented. Similar results were obtained by Al-Homidan (2007), who indicated that the addition of dried yeast (SC) to laying rations increased the percentage of egg production traits than control.

**Table 2. Egg production of laying Japanese quail as affected by stocking density without or with yeast extract supplementation.**

Items	ND	HD	HD + Yeast extract level (mg/kg diet)				SEM	P value
			1	2	3	4		
Egg number/bird								
8-12 weeks	23.57 <sup>b</sup>	18.89 <sup>d</sup>	23.01 <sup>b</sup>	20.32 <sup>c</sup>	24.76 <sup>a</sup>	20.55 <sup>c</sup>	0.337	<.0001
12-16 weeks	19.44 <sup>b</sup>	15.00 <sup>d</sup>	19.44 <sup>b</sup>	17.04 <sup>c</sup>	26.11 <sup>a</sup>	20.40 <sup>b</sup>	0.300	<.0001
8-16 weeks	21.51 <sup>b</sup>	16.94 <sup>c</sup>	21.23 <sup>b</sup>	18.68 <sup>d</sup>	25.44 <sup>a</sup>	20.48 <sup>c</sup>	0.179	<.0001
Egg weight (g)								
8-12 weeks	12.45 <sup>ab</sup>	12.06 <sup>c</sup>	12.27 <sup>b</sup>	12.41 <sup>ab</sup>	12.48 <sup>a</sup>	12.00 <sup>c</sup>	0.053	0.0002
12-16 weeks	13.44 <sup>b</sup>	13.10 <sup>c</sup>	13.33 <sup>b</sup>	13.33 <sup>b</sup>	13.96 <sup>a</sup>	13.06 <sup>c</sup>	0.052	<.0001
8-16 weeks	12.94 <sup>b</sup>	12.58 <sup>c</sup>	12.80 <sup>b</sup>	12.87 <sup>b</sup>	13.22 <sup>a</sup>	12.53 <sup>c</sup>	0.045	<.0001
Egg mass (g/bird)								
8-12 weeks	293.44 <sup>ab</sup>	227.73 <sup>d</sup>	282.29 <sup>b</sup>	252.12 <sup>c</sup>	308.95 <sup>a</sup>	246.70 <sup>c</sup>	4.832	<.0001
12-16 weeks	261.22 <sup>b</sup>	196.54 <sup>d</sup>	259.13 <sup>b</sup>	227.01 <sup>c</sup>	364.50 <sup>a</sup>	266.27 <sup>b</sup>	3.662	<.0001
8-16 weeks	277.33 <sup>b</sup>	212.13 <sup>c</sup>	270.71 <sup>b</sup>	239.56 <sup>d</sup>	336.72 <sup>a</sup>	256.48 <sup>c</sup>	2.677	<.0001

Means within the same row with different common superscripts differ significantly.

**Feed intake and feed conversion ratio**

Quail layers reared in HD without or with dietary YE supplementation at different levels consumed lower feed ( $P < .0001$ ) as compared with layers reared in ND

throughout all the experimental periods studied (Table 3). It is worth to note that, the lowest feed consumed through 12-16 weeks of age was recorded for layers housed at HD with dietary YE supplementation at 2 mg/kg diet. The

reduction in feed consumption may be attributed to reduced feeder space per bird in each cage.

Our results agree with the previous researches (Daniela *et al.*, 2018; EL-Sheikh *et al.*, 2016; El-Shafei *et al.*, 2012) who indicated that feed consumption for Japanese quail significantly decreased with increasing cage density. Likewise, the effect of YE on feed consumption agree with Liu *et al.* (2002) who found reduction feed consumption in quails at 0.2 % yeast. Also, Sanaa (2013) reported that, feed consumption was decreased due to dietary yeast at 3- 6 g/kg in laying hens. However, Sehu *et al.* (1997) noticed similar feed consumption in laying quails fed 1.5 % inactivated brewer's yeast.

Regarding to feed conversion ratio (FCR), the results in Table (3) showed increasing stocking density of Japanese quail layers from ND (9/cage) to HD (15/cage) without dietary YE supplementation caused a significant inferior FCR values thought the interval periods. Statistical analysis indicated that, dietary YE supplementation at different levels of quail layers groups raised under higher density resulted better ( $P < .0001$ ) FCR values when compared with quail layers group raised under ND and HD without dietary YE supplementation. It could be noticed that, HD group with dietary 3 mg YE/kg supplementation recorded better FCR values compared with other treatment groups.

The effect of stocking density on FCR values in the present study are in agreement with those of EL-Sheikh *et al.* (2016) who reported that FCR values were significantly

poorer with increasing the number of quails/square meter throughout the production period. On the other hand, Santos *et al.* (2011); EL-Shafei *et al.* (2012) observed FCR/kg of egg produced was better ( $P < 0.05$ ) for quail layers raised under higher floor space. Moreover, Faitarone *et al.* (2005) stated that, FCR values of quails were not significantly affected among treatment when stocked at 246, 211, 176 and 151 cm<sup>2</sup>/quail.

The better FCR values in Japanese quail layers raised under high density due to YE supplementation may be due to increase of egg number and egg weight associated with decreased feed consumption and consequently lead to better FCR values. These results are in agreement with Hassanein and Soliman (2010) who found yeast supplementation at 0.02 - 0.04 % improved nutrient utilization in laying birds. Sehu *et al.* (1997) observed similar FCR in laying quails fed 15% inactivated brewer's yeast.

In the same line, Al-Mansour *et al.* (2011) and Abu-Taleb *et al.* (2006) found that feed intake was not affected, while feed efficiency was enhanced due to adding yeast to laying quail diets by 1 and 19%. Also, Yalcin *et al.* (2008) investigated the effect of adding 0.5, 1.0, 1.5 and 2.0 the dried yeast (*Saccharomyces cervisiae*) to Japanese quail diets on the performance from 1 to 6 weeks old. Results indicated that there was a reduction in feed consumption values of yeast fed groups. Feed conversion ratios improved by 6, 9, 12 and 3% due to adding yeast by levels of 1.5 and 2.0% into diets, respectively.

**Table 3. Feed intake and feed conversion ratio of laying Japanese quail as affected by stocking density without or with yeast extract supplementation.**

Items	ND	HD	HD + Yeast extract level (mg/kg diet)				SEM	P value
			1	2	3	4		
Feed intake (g/bird)								
8-12 weeks	968.50 <sup>a</sup>	856.98 <sup>b</sup>	854.17 <sup>bc</sup>	824.67 <sup>c</sup>	853.83 <sup>bc</sup>	845.25 <sup>bc</sup>	8.124	<.0001
12-16 weeks	932.50 <sup>a</sup>	825.98 <sup>c</sup>	855.67 <sup>b</sup>	775.67 <sup>d</sup>	953.83 <sup>a</sup>	883.50 <sup>b</sup>	8.598	<.0001
8-16 weeks	950.50 <sup>a</sup>	841.48 <sup>c</sup>	854.92 <sup>c</sup>	800.17 <sup>d</sup>	903.83 <sup>b</sup>	864.38 <sup>c</sup>	6.565	<.0001
Feed conversion ratio (g feed/g egg)								
8-12 weeks	3.30 <sup>b</sup>	3.77 <sup>a</sup>	3.03 <sup>c</sup>	3.27 <sup>b</sup>	2.76 <sup>d</sup>	3.43 <sup>b</sup>	0.044	<.0001
12-16 weeks	3.64 <sup>b</sup>	4.28 <sup>a</sup>	3.30 <sup>c</sup>	3.52 <sup>b</sup>	2.48 <sup>d</sup>	3.25 <sup>c</sup>	0.042	<.0001
8-16 weeks	3.43 <sup>b</sup>	3.97 <sup>a</sup>	3.16 <sup>d</sup>	3.34 <sup>c</sup>	2.68 <sup>e</sup>	3.37 <sup>bc</sup>	0.015	<.0001

Means within the same row with different common superscripts differ significantly.

### Reproductive performance (Fertility and hatchability percentages)

Data illustrated in Table 4 revealed that, fertility % through 8-12 and 8-16 weeks of age and hatchability % during 8-12 period were significantly ( $P < 0.05$ ) decreased with increasing stocking density without or with dietary YE supplementation when compared with layers stocked at ND. However, fertility % during 12-16 weeks of age and hatchability % through 12-16 and 8-16 weeks of age were not significantly affected due to increasing stocking density with or without dietary YE supplementation.

The decreasing fertility % of eggs produced from quail layers raised at high stocking density may be attributed to the decrease in feed consumption and competition among birds, and consequently lead to decreases in hatchability rate.

The obtained results of fertility and hatchability percentages in the present study agree with EL-Sheikh *et al.* (2016) who observed, the percentages of fertility and hatchability for Japanese quail were increased with

increasing cage floor space through 11-21 weeks of age. However, Askar *et al.* (2012) found that, the percentages of fertility and hatchability for Japanese quails were not significantly affected by stocking density through the period from 10- 18 weeks.

Attia *et al.*, (2021) found that the fertility rate was better ( $P < 0.01$ ) at the LD than HD. The reduction in fertility percentage at greater density under investigation agrees with Tollba *et al.* (2006). On the contrary, the non-significant effect of stocking density recorded for hatchability percentages disagrees with the previous authors' findings. There is little information available in the literature concerning the impact of stocking and its interaction with feed supplement (SC/or Vit E) on reproductive traits. Therefore, this kind of research is needed to improve animal welfare exposed to stocking stress. Housing stress was reported to influence performance, welfare, antioxidant, and poultry behavior (Narinc *et al.*, 2013).

**Table 4. Fertility and hatchability percentages of laying Japanese quail as affected by stocking density without or with yeast extract supplementation.**

Items	ND	HD	HD + Yeast extract level (mg/kg diet)				SEM	P value
			1	2	3	4		
Fertility %								
8-12 weeks	89.01 <sup>ab</sup>	67.22 <sup>c</sup>	81.52 <sup>b</sup>	82.40 <sup>ab</sup>	91.92 <sup>a</sup>	84.72 <sup>ab</sup>	2.937	0.0017
12-16 weeks	92.22	83.58	92.38	95.45	94.19	91.28	3.547	0.3264
8-16 weeks	90.61 <sup>a</sup>	75.40 <sup>b</sup>	86.96 <sup>a</sup>	88.93 <sup>a</sup>	93.05 <sup>a</sup>	88.00 <sup>a</sup>	2.239	0.0057
Hatchability %								
8-12 weeks	78.57 <sup>a</sup>	58.89 <sup>c</sup>	65.55 <sup>bc</sup>	64.78 <sup>bc</sup>	75.28 <sup>ab</sup>	70.89 <sup>ab</sup>	3.537	0.0209
12-16 weeks	79.11	69.25	79.29	83.07	83.42	77.38	3.431	0.1226
8-16 weeks	78.84	64.07	72.42	73.92	79.35	74.14	3.142	0.0573

Means within the same row with different common superscripts differ significantly.

**Egg quality traits**

The results in Table (5) showed that, increasing the stocking density without or with YE addition in their diets did not significantly affect all quail egg quality traits studied (Albumin, Yolk, Shell thickness, egg shape index, Yolk index, Haugh unit and USSW percentages). These results, agree with Daniel *et al.* (2018) who found, no significant differences were observed for any of the variables analyzed (specific gravity, egg color, shell thickness and Haugh unit). Also, Lopes *et al.* (2008) did not find any significant difference of specific gravity, albumin percentage, yolk percentage and bark percentage.

Contradicting results were obtained by Ratriyanto *et al.* (2020) who stated that, a high stocking density adversely effects egg quality in quail layers, particularly, the egg shell thickness. Likewise, El-Sheikh *et al.* (2016) reported that, quail layers housed at low density laid eggs had higher shell thickness with or without membranes and egg surface area as compared to those at high density. Moreover, El-Tarabary *et al.* (2015) indicated that quail layers reared at low density (200 cm<sup>2</sup>/ bird) significantly (P<0.05) increased shell thickness, shell percentage and

egg shell weight than those reared at higher density (167 and 143 cm<sup>2</sup>/ bird) at 14 weeks of age.

Regarding the effect of yeast supplementation, Hosseini *et al.* (2006) observed to significant effect on egg shell thickness, egg shell quality and Haugh unit with using yeast at levels of 0.25, 0.5, 0.75 g/kg diet to layers Maziar *et al.* (2007) indicated no significant effete on egg quality parameters (shell thickness, shell percentage and Haugh unit) when Hy-Line hens were fed 1 g yeast of SC / kg diet under heat stress 33°C. Songsak *et al.* (2009) found that supplementation of cassava yeast to 26-wk laying hens had a positive effect on shell thickness, but had no significant effect on albumin weight, yolk weight and Haugh unit. Tollba and EL-Nagar (2008) reported no effect on egg quality in any term by adding *Saccharomyces cerevisiae* to 20-wk local hens reared under high density. Similar result was obtained by Soliman (2002) who added 4g active dried yeast /kg diet to 25-wk Bovans White laying hens. Also, Abd El-Maksoud *et al.* (2011) observed no effect of egg quality except it improved shell thickness when added 250 g/kg diet dried yeast to local hens.

**Table 5. Egg quality traits of laying Japanese quail as affected by stocking density without or with yeast extract supplementation.**

Items	ND	HD	HD + Yeast extract level (mg/kg diet)				SEM	P value
			1	2	3	4		
Albumin %	52.75	52.35	53.19	53.07	52.91	53.08	0.429	0.8185
Yolk %	32.09	31.69	32.92	31.75	33.19	33.05	0.507	0.2231
Shell %	15.17	15.98	13.89	15.18	13.89	13.87	0.486	0.0522
Shell thickness	0.23	0.21	0.22	0.21	0.22	0.20	0.007	0.4116
Egg shape index	78.85	78.67	80.57	78.89	79.76	78.92	1.666	0.9691
Yolk index	48.86	47.43	50.1	48.18	50.60	49.39	1.981	0.8761
Haugh unit	83.06	80.75	84.26	81.74	81.22	81.18	0.810	0.0761
USSW	47.67	47.12	47.66	47.62	47.98	48.00	0.401	0.7325

Means within the same row with different common superscripts differ significantly.

**Digestive enzymes**

Digestive enzymes (Amylase, lipase, and protease) activity was decreased with HD when compared to ND (Table 6). Dietary supplementation of YE to HD groups resulted in significant enhances in digestive enzyme comparatively with groups kept at HD fed diet without

addition of YE (Table 7). Yeasts including *S. cerevisiae* play positive roles in improving poultry productivity and public health which promote the efficacy of digestive enzymes (Yoon *et al.*, 2004).

**Table 6. Digestive enzymes of laying Japanese quail as affected by stocking density without or with yeast extract supplementation.**

Items	ND	HD	HD + Yeast extract level (mg/kg diet)				SEM	P value
			1	2	3	4		
Protease (U/L)	0.51 <sup>a</sup>	0.24 <sup>c</sup>	0.43 <sup>ab</sup>	0.26 <sup>bc</sup>	0.36 <sup>abc</sup>	0.41 <sup>abc</sup>	0.051	0.0336
Amylase (U/L)	18.80 <sup>b</sup>	10.71 <sup>c</sup>	13.55 <sup>c</sup>	24.05 <sup>a</sup>	21.55 <sup>ab</sup>	24.34 <sup>a</sup>	1.519	0.0002
Lipase (U/L)	8.45 <sup>ab</sup>	6.35 <sup>b</sup>	10.30 <sup>a</sup>	11.35 <sup>a</sup>	11.40 <sup>a</sup>	10.83 <sup>a</sup>	1.039	0.0478

Means within the same row with different common superscripts differ significantly.

**Blood constituents**

Results in Table (7) show that quails stocked under HD without dietary YE supplementation had higher values of serum ALB ( $p=0.0118$ ), GLO ( $p=0.0361$ ), A/B % ( $p<0.0001$ ), ALT ( $p=0.0044$ ), AST ( $p=0.0026$ ), urea ( $p=0.0251$ ), TC ( $p=0.0309$ ), TG ( $p<0.0001$ ), HDL ( $p=0.0013$ ), and VLDL ( $p<0.0001$ ) than those stocked under ND and HD with YE supplementation at different levels. However, the data revealed insignificant differences among all treatment groups for serum TP, LDL, creatinine and LDL concentrations. The addition of YE in HD diet alleviated the stress effect induced by high stocking density through decreasing the level of aforementioned parameters and achieved serum blood parameters values equal

significantly to quail layers stocked at ND compared with layers stocked at HD without YE supplementation.

The activity of SOD ( $p<0.0001$ ), TAC ( $p<0.0001$ ) and GPX ( $p=0.0047$ ) was improved with HD plus YE when compared to HD (Table 8). While, dietary supplementation of YE to HD groups resulted in significant decreases in MDA ( $P=0.0155$ ) comparatively with groups kept at HD fed diet without addition of YE. These results are confirmed by Abd El-Wahab *et al.* (2019) who found that dietary yeast supplementation in Japanese quail increased the activities of antioxidant enzymes (CAT and SOD). On the other hand, Cengiz *et al.* (2015) observed that no significant differences in blood MDA between low and high stocking densities.

**Table 7. Total protein and its fractions, liver, kidney functions and lipid profile of laying Japanese quail as affected by stocking density without or with yeast extract supplementation.**

Items	ND	HD	HD + Yeast extract level (mg/kg diet)				SEM	P value
			1	2	3	4		
TP (g/dL)	3.77	3.59	3.68	3.46	3.92	3.84	0.182	0.6498
ALB (g/dL)	2.07 <sup>b</sup>	2.44 <sup>a</sup>	2.06 <sup>b</sup>	1.84 <sup>b</sup>	2.07 <sup>b</sup>	2.12 <sup>b</sup>	0.080	0.0118
GLOB (g/dL)	1.70 <sup>a</sup>	1.15 <sup>b</sup>	1.63 <sup>a</sup>	1.62 <sup>a</sup>	1.85 <sup>a</sup>	1.73 <sup>a</sup>	0.116	0.0361
A/G (%)	1.24 <sup>b</sup>	2.18 <sup>a</sup>	1.27 <sup>b</sup>	1.15 <sup>b</sup>	1.12 <sup>b</sup>	1.23 <sup>b</sup>	0.080	<.0001
ALT (IU/L)	8.12 <sup>b</sup>	16.02 <sup>a</sup>	9.04 <sup>b</sup>	6.86 <sup>b</sup>	8.59 <sup>b</sup>	9.68 <sup>b</sup>	1.173	0.0044
AST (IU/L)	188.55 <sup>c</sup>	332.31 <sup>a</sup>	286.35 <sup>ab</sup>	264.55 <sup>b</sup>	246.20 <sup>bc</sup>	307.90 <sup>ab</sup>	18.373	0.0026
Creatinine (mg/dL)	0.37	0.44	0.33	0.33	0.31	0.36	0.039	0.3405
Urea (mg/dL)	3.72 <sup>b</sup>	5.84 <sup>a</sup>	3.46 <sup>b</sup>	3.41 <sup>b</sup>	3.19 <sup>b</sup>	3.67 <sup>b</sup>	0.443	0.0251
Lipid profile:								
TC (mg/dL)	406.00 <sup>ab</sup>	448.78 <sup>a</sup>	366.85 <sup>b</sup>	389.55 <sup>b</sup>	402.25 <sup>b</sup>	397.20 <sup>b</sup>	13.142	0.0309
TG (mg/dL)	162.93 <sup>b</sup>	232.81 <sup>a</sup>	108.54 <sup>cd</sup>	131.25 <sup>c</sup>	91.40 <sup>d</sup>	127.25 <sup>c</sup>	9.209	<.0001
HDL (mg/dL)	40.79 <sup>bc</sup>	32.97 <sup>d</sup>	38.90 <sup>cd</sup>	42.44 <sup>bc</sup>	46.79 <sup>ab</sup>	49.20 <sup>a</sup>	1.915	0.0013
LDL (mg/dL)	332.62	369.25	306.25	320.87	337.18	322.55	13.375	0.1213
VLDL (mg/dL)	32.59 <sup>b</sup>	46.56 <sup>a</sup>	21.71 <sup>cd</sup>	26.25 <sup>c</sup>	18.28 <sup>d</sup>	25.45 <sup>c</sup>	1.842	<.0001

Means within the same row with different common superscripts differ significantly.

TP: total protein; Alb: albumin, GLOB: globulin; A/G: albumin/ globulin ratio; AST: aspartate aminotransferase and ALT: alanine aminotransferase; TC: total cholesterol; TG: triglycerides; HDL: high density lipoprotein; LDL: low density lipoprotein; VLDL: very low density lipoprotein.

There were no significant differences in lysozyme or Complement 3 activity due to increasing stocking density in quail breeders from ND to HD fed diets supplemented with YE. While, the values of IgG ( $p=0.0065$ ), and IgA ( $p=0.0112$ ) were improved with HD plus YE when compared to HD (Table 8). The results of stocking density are supported by Gholami *et al.* (2020) who stated that broiler chickens reared under high stocking density (20 chicks / m<sup>2</sup>) had the lowest immune

parameters than those reared under 10 and 17 chicks / m<sup>2</sup> at d 42. Likewise, Heckert *et al.*, (2002) reported that the high stocking density suppression of immunity in broiler chickens. Houshmand *et al.* (2012) pointed out that broiler chickens subjected to a high stocking density had a lower antibody titer against New castle disease than those subjected to a normal stocking density (Heckert *et al.*, 2002). So, it could be concluded that the high stocking density in the current study resulted in poorer immunity.

**Table 8. Antioxidant and immunological indices of laying Japanese quail as affected by stocking density without or with yeast extract supplementation.**

Items	ND	HD	HD + Yeast extract level (mg/kg diet)				SEM	P value
			1	2	3	4		
Antioxidants								
SOD (U/mL)	0.30 <sup>b</sup>	0.13 <sup>d</sup>	0.19 <sup>cd</sup>	0.23 <sup>c</sup>	0.33 <sup>ab</sup>	0.40 <sup>a</sup>	0.023	<.0001
MDA (nmol/mL)	0.45 <sup>b</sup>	0.57 <sup>a</sup>	0.36 <sup>b</sup>	0.40 <sup>b</sup>	0.39 <sup>b</sup>	0.34 <sup>b</sup>	0.038	0.0155
TAC (ng/ml)	0.35 <sup>b</sup>	0.12 <sup>d</sup>	0.24 <sup>c</sup>	0.38 <sup>ab</sup>	0.41 <sup>ab</sup>	0.46 <sup>a</sup>	0.029	<.0001
GPX (ng/ml)	0.31 <sup>bc</sup>	0.20 <sup>c</sup>	0.25 <sup>c</sup>	0.45 <sup>a</sup>	0.43 <sup>ab</sup>	0.43 <sup>ab</sup>	0.038	0.0047
Immunity								
IgG (mg/dl)	1.02 <sup>ab</sup>	0.42 <sup>c</sup>	0.91 <sup>ab</sup>	1.32 <sup>a</sup>	0.62 <sup>bc</sup>	0.73 <sup>bc</sup>	0.118	0.0065
IgA (mg/dl)	0.83 <sup>ab</sup>	0.42 <sup>c</sup>	1.07 <sup>a</sup>	0.88 <sup>ab</sup>	0.55 <sup>bc</sup>	1.03 <sup>a</sup>	0.110	0.0112
Lysozyme	0.31	0.15	0.23	0.22	0.13	0.22	0.037	0.0888
Complement 3	107.55	92.75	105.05	114.25	115.55	110.03	5.973	0.1983

Means within the same row with different common superscripts differ significantly.

SOD: superoxide dismutase; MDA: malondialdehyde; TAC: total antioxidant capacity; GPX: glutathione peroxidase; IgG immunoglobulin G; IgA: immunoglobulin A.

In this respect, El-Sheikh *et al.*, (2016) found significant reductions in plasma total protein fractions of Japanese quail layers by increasing stocking density

whereas, a significant increase in total lipids, cholesterol, ALT, plasma total calcium and phosphorous with increasing stocking density. On the other hand, they found

insignificant differences were observed for serum HDL, LDL and AST levels. Also, Tollba *et al.* (2006) manifested that, levels of plasma TP, albumin, globulin, A/G%, ALT and AST for Egyptian laying chickens were not affected significantly by different stocking densities. Simsek *et al.* (2014) displayed that, serum HDL and cholesterol levels were reduced for boiler chicks stocked at lower densities (7.5 bird /m<sup>2</sup>) when compared with those stocked at higher levels (22.5, 18.75, 15 and 11.5 bird/ m<sup>2</sup>).

Skrbic *et al.* (2009) reported that, no significant differences were observed of broiler chickens for cholesterol concentrations when stocked at 10, 13 or 16 birds/m<sup>2</sup>. Likewise, El-Deek and Al-Harhi (2004) denoted that, stock density had no significant effect on plasma constituents of broiler chicks, especially ALT and AST activity. While, Erisir and Erisir (2002) manifested that, no significant differences were observed for albumin and total proteins of quails stocked at different stocking density. The improvements of some serum blood constituents due to dietary YE supplementation in the present study agree with many researchers who declared that dietary supplementation of yeast reduced serum cholesterol and triglycerides (Yalcim *et al.*, 2010; Hassanein and Soliman 2010; Yalcin *et al.*, 2014).

## CONCLUSION

It could be concluded that use of yeast extract in quail breeder's diets can positively mitigate the stress applied to quail raised under high stoking density by improving productive and reproductive performance, enhancing the antioxidant status, immunological parameters, lipid profile and digestive enzymes.

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### هل يحسن مستخلص الخميرة أداء وصحة أمهات السمان المربي تحت كثافة تربية عالية؟ فايز محمد رضا ، محمود العجواني ، رشا محمد صبري ومحمد ممدوح المكاوي قسم الدواجن، كلية الزراعة، جامعة الزقازيق، مصر

صممت هذه الدراسة لدراسة تأثير كثافة التربية بدون أو مع إضافة مستخلص الخميرة (YE) على الأداء الإنتاجي والتناسلي، والإنزيمات الهاضمة، والقياسات البيوكيميائية في الدم، ومضادات الأكسدة والقياسات المناعية لامهات السمان. تم استخدام إجمالي عدد 336 طائر السمان الياباني بعمر 8 أسابيع ووزع السمان عشوائياً إلى 6 مجموعات بكل مجموعة 4 مكررات في تصميم عشوائي تام. وتضمنت المجموعة الأولى 36 طائرا (24 أنثى و 12 ذكور) تم تربيتها بمعدل 9 طيور / مكررة (المقارنة ، ND ؛ 36 سمان / م 2) وتم تغذيتها على العليقة الأساسية بدون أي إضافات. واحتوت المجموعات الثانية والثالثة والرابعة والخامسة والسادسة على 60 سمانة (40 أنثى و 20 نكراً) وتم تربيتها بمعدل 15 طائر / مكررة (HD ؛ 60 سمان / م 2) وتغذيتها على عليقة أساسية مع 0 ، 1 ، 2 ، 3 ، 4 ملجم من YE / كجم علف ، على التوالي. انخفض عدد البيض ووزن البيضة وكتلة البيض معنوياً ( $P < 0.0001$ ) نتيجة زيادة كثافة التربية من 9 / قفص إلى 15 / قفص. والتغذية بدون إضافة مستخلص الخميرة تحسن نشاط SOD ( $p < 0.0001$ ) و TAC ( $p < 0.0001$ ) و GPX ( $p = 0.0047$ ) باستخدام مستخلص الخميرة للسمان المربي تحت كثافة عالية مقارنة بـ HD. انخفض نشاط إنزيمات الجهاز الهضمي (الأميليز والليباز والبروتياز) مع الكثافة العالية بالمقارنة مع بالكثافة الطبيعية. يمكن الاستنتاج أن إضافة مستخلص الخميرة يمكن أن يخفف بشكل إيجابي من الضغط الواقع على السمان الذي يتم تربيته تحت كثافة تربية عالية من خلال تعزيز الأداء الإنتاجي والتناسلي، ومضادات الأكسدة والمناعة وكذلك معايير جودة البيض.