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# Productive and Reproductive Performance of Doe Rabbits Fed Diets Supplemented with Spirulina, Yeast, and *Bacillus subtilis* as Natural Additives

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# ABSTRACT



This study was designed to investigate the effects of dietary adding *Spirulina platensis* (SP), live yeast (LY), and *Bacillus subtilis* (BS) on nutrient digestion, and the productive and reproductive performances of doe rabbits. Forty healthy doe rabbits aged nine months and weighing  $3223\pm5$  g were used for a 75 - day experimental period and split into four similar experimental groups according to their live body weight. Rabbits were fed a pelleted diet free from probiotics (control group, T1), while T2, T3, and T4 groups received the same diet supplemented with 1 g SP/doe/day, 1 g LY/doe/day, and 0.5 g BS/doe/day, respectively. Results showed a significant (p<0.05) increase in milk yield in T4 but didn't influence feed intake or live body weight of rabbits. All nutrient digestibility coefficients were the highest in T2. Most productive and reproductive performances of does had significantly enhanced in probiotic groups. Supplementation with probiotics significantly increased serum total protein, albumin, and globulin concentrations, while serum urea and creatinine concentrations appeared to be significantly (p<0.05) lower. Generally, the results obtained revealed that using SP, LY, and BS as feed additives in rations of doe rabbits, have positive effects on the productive and reproductive performance traits and economic feed efficiency. These supplements could be used as natural feed additives to improve the health status as well as the productive and reproductive performance fractions appeared to be significantly.

Keywords: Probiotics, rabbits, nutrient digestion, blood parameters, litter size

# INTRODUCTION

Probiotics are demonstrated to be an effective alternative to antibiotics for livestock producers, improving the productive performance and health status of animals (Kiczorowska et. *al.*, 2017). They also help support intestinal health by promoting good bacterial growth and producing chemical compounds to fight pathogenic bacterial growth (Sokale *et al.*; 2019; Liu *et al.* 2019). As explained by Krieg *et al.* (2010), gut microbiota plays a vital role in preserving gut health system by maintaining ecological intestinal structure and its function, which results in enhanced nutrients digestion and absorption (Zhang *et al.*, 2019).

On the other hand, several studies documented the effectiveness of spirulina, yeast, and B. subtilis in improving animal performance without any harmful effects on rabbit health (Guo et al., 2017; Shen et al., 2018; Abdou et al., 2024). Specifically, Bacillus species are bacteria that are highly safe and more effective in animal nutrition (EFSA, 2013). Moreover, Sanders et al. (2003) reported that Bacillus species can generate substances important as growth promoters such as vitamin K, B12 and amino acids and consume oxygen in the gut, leading to increases in the number of anaerobic bacteria (Zhou et al., 2013). Earlier studies observed that B. subtilis can enhance nutrient digestibility coefficients and improve growth performance as well as enhancing the immune system of rabbits (Makarenko et al., 2019), by increasing the beneficial of the lactic acid bacteria relative to the harmful one (Guo et al. 2017).

In animal nutrition, yeast is popularly used as a probiotic agent to improve the productive performance and overall health of animals (Ayyat *et al.*, 2018). Furthermore, spirulina in animal feeding has good nutritional and functional properties due to its high-quality vital compounds and health-promoting properties (Marquez- Rocha *et al.*, 1995). In addition, many studies have indicated that feeding on different probiotic species to experimental rabbits improved well-being, enhanced productive performance, and a decreased mortality rate (Makarenko *et al.*, 2019; Simonová *et al.*, 2020; Abd El-Aziz *et al.*, 2021; Abdou *et al.*, 2024). Thus, this study aimed to evaluate the impact of spirulina, live yeast, and Bacillus subtilis supplementation on nutrient digestibility coefficients, certain parameters in the blood, and the productive and reproductive performance of doe rabbits.

# MATERIALS AND METHODS

## **Ethical Statement**

This study was conducted at Sakha Animal Production Research Station, Animal Production Research Institute (APRI), Agricultural Research Centre, Ministry of Agriculture, Egypt. All implementation measures were taken to reduce animals suffering and safe the welfare conditions throughout the trial.

#### Feeding trail:

Forty New Zealand doe rabbits, having nine months of age and weighing an average LBW of 3223±5 g, were used for 75 days experimental period, chosen and randomly distributed to four similar groups of ten each, based on their LBW. Rabbits were fed a pelleted diet free from probiotics (control group, T1), while T2, T3, and T4 groups received the same diet supplemented with 1 g SP/doe/day, 1 g LY/doe/day and 0.5 g BS/doe/day, respectively. Spirulina is obtained from the National Institute of Oceanography and Fisheries (NIOF), in Egypt, which reports its chemical composition was 55.80, 6.20, 4.90, 23.00, and 10.10% for CP, EE, CF, NFE, and ash, respectively. While, like yeast additive have contained 2x109 cfu/g for Bacillus subtilis. The formulation and chemical composition of conventional ration are shown in Table 1. Rabbits were covered accurately the nutrient requirements according to NRC (1977). During the trial period, doe rabbits were housed individually in galvanized metal wire cages. The drinking fresh water was available via automatic nipple drinkers. All doe rabbits were preserved in the same administrative and hygienic trial conditions. Ingredients and chemical analysis of belted diet are shown in Table (1).

### Digestibility trials:

Four digestibility trials were carried out at the end of the feeding trial on three rabbits from each group. The digestion experiment lasted 10 days, which as a preliminary period of 7 days and 3 days as collection period. Both feed and feces were collected quantitatively and recorded daily, individually for each doe, through the collection period. The various samples were chemically analyzed for CP, CF, EE, and ash according to AOAC (2005) after 48 hour drying period at 60°C in a forced air oven.

Table 1. ingredients and chemical analysis of basal diet.

Ingredient	%	Chemical analysis (% as DM):	%
Berseem hay	30.25	Dry matter (DM)	85.81
Barley grain	24.60	Crude protein (CP)	17.36
Wheat bran	21.50	Organic matter (OM)	91.42
Soybean meal (44% CP)	17.50	Crude fiber (CF)	12.37
Molasses	3.00	Ether extract (EE)	2.230
Limestone	0.95	Digestible energy (DE,kcal/kg)**	2412
Di-calcium phosphate	1.60	Calcium	1.243
Sodium chloride	0.30	Total phosphorus	0.808
Mineral-vitamin premix*	0.30	Lysine	0.862
Total	100	DL-Methionine	0.632

\* One kilogram of mineral-vitamin premix provided: Vitamin A, 150,000 UI; Vitamin E, 100 mg; Vitamin K3, 21mg; Vitamin B1, 10 mg; VitaminB2, 40mg; Vitamin B6, 15mg; Pantothenic acid, 100 mg; Vitamin B12, 0.1mg; Niacin, 200 mg; Folic acid, 10mg; Biotin, 0.5mg; Choline chloride, 5000 mg; Fe, 0.3mg; Mn, 600 mg; Cu, 50 mg; Co, 2 mg; Se, 1mg; and Zn, 450mg.\*\* Calculated according to De Blas and Mateos (1998).

#### Productive and reproductive traits:

Live body weight (LBW) and weight changes for each doe were recorded during different trial periods. The conception rate (CR), the size and weight of litter from birth to weaning (LSB and LWW), and the mortality rate (MR) was calculated as the following equations:

### Mortality rate (%) = [(number of kids at birth – number of kids at weaning) /number of kids at birth] x 100

The feed intake (FI) of pelleted diet was recorded, and then feed conversion ratio (FCR) was calculated as the following: FCR = Daily feed intake (g) / daily weight gain (g) of litter Milk intake by kids (MY) was determined by the kid weight differences prior and following suckling for 24 hours twice a week.

#### **Blood parameters:**

Blood samples were taken in clean tubes from the ear veins of three does in each group, at the end of the experimental period. Blood samples were allowed to clot, then a clear serum was separated by centrifugation at 4000 rpm for 20 minutes and kept in a deep freezer at 20  $^{0}$ C till the time of analysis.

Biochemical parameters were determined in serum using commercial kits of Bio-Merieus Lab, France in accordance with the manufacturer's instructions. Total protein (TP), albumin (Al), urea, and creatinine levels were estimated employing the techniques of (Henry *et al.*, 1974; Doumas *et al.*, 1971). The globulin (Gl) level was determined by positing the Al level from the TP level. The liver enzyme aspartate aminotransferases (AST) and alanine aminotransferases (ALT) levels were analysis using trade kits based on the methods of Reitman and Frankel (1957). **Statistical analysis:** 

The data were statistically analyzed using SAS (2003) program, General Linear Model's technique. A one-way analysis was carried out to assess the impact of the addition of various probiotics on the tested parameters by using the following model:  $Y_{ij}$ =  $\mu$ +T<sub>i</sub>+E<sub>jj</sub>,

Where:  $Y_{ij}$  = the observation of the parameter measured,  $\mu$  = the overall means,  $T_i$  = the effect of dietary treatment, (i= 1.2,3 and 4) and  $E_{ij}$  = the random error. Differences between means among all treatments were subjected to Duncan's Multiple Range– test Duncan (1955).

## **RESULTS AND DISCUSSION**

# Digestibility of nutrients:

All digestibility nutrients and feeding values of SPration were significantly (P<0.05) higher than those of the control one, except for NFE digestibility which recorded higher digestibility with no significant differences (Table 2). The highest values were associated with the T2 group followed by the T4 and T3 groups, whereas the lowest one occurred with the T1 group.

This favorable impact of SP supplement on digestibility nutrients is due to its high levels of unsaturated fatty acids, polysaccharides, carotenoids, growth factors, and β- glucan, which contribute to enhanced gut biodiversity and increased lactic acid bacteria numbers, according to Kholif et al. (2017) and Gomaa et al. (2018). These results are in agreement with those obtained by Amer et al. (2016) and Mahmoud et al. (2017), except for the reduction of CF and EE digestibility, respectively. Moreover, El-Deep et al. (2021) found that adding higher levels of Aspergillus awamori significantly improved CP, CF, and EE digestibility coefficients compared to the low level and control. A more recent study confirmed the positive effect of SP on nutrient digestibilities and nutritive values, as reported by Abdou et al. (2024) who added three levels of spirulina in rabbit diets. Otherwise, our findings didn't agree with those published by Peiretti and Meineri (2008) which suggested that spirulina supplementation in growing rabbit rations negatively affects nutrient digestibility. On the other hand, adding BS significantly (P<0.05) improved OM, CF, and CP digestibility coefficients and the nutritive value relative to the control one, with no notable differences in other nutrients. While adding LY led to somewhat improved nutrients digestibility and the nutritive values than that of the control one. Likely, the addition of probiotics can help support nutrient digestibility coefficients by encouraging digestive enzyme release (Hu et al., 2018). Additionally, it enhances the fermentation activity and gut structure adjusting, thereby increasing of homeostasis and overall gut function (Cartman

et al., 2008; Gu et al., 2015; El-Deep et al., 2021). Likewise, they can also improve nutrient absorptions across the intestinal barrier (Machida et al., 2005), activate the gut microbiota, improve the immune system, and boost gut mucosa structure (Rawski et al., 2016; Elghandour et al., 2019). These results were in line with those of Bhatt et al. (2017) and Elmasry et al. (2021), who stated that the addition of probiotics could improve all or some of nutrient digestibility coefficients. Likewise, another study on rabbits that conducted by El-Badawi et al. (2017), who found that the digestibility of nutrients was improved by adding yeast or BS compared to that of combination group (LY plus BS) and control one. However, yeast addition showed a slight effect on nutrient digestibility and nutritive values; the reason for this is not clear. Similarly, the results of Tag El Din et al. (2019) confirmed that supplementing yeast in rabbit rations didn't influence the digestibility of nutrients, except for crud fiber digestibility, which appeared to an improvement. Additionally, earlier results of Phuoc and Jamikorn (2017) found that nutrient digestibility coefficients with weaned rabbits were unchanged after fed dietary BS. Furthermore, Fathi et al. (2017) observed that the positive effects of probiotics on nutrient digestibility coefficients depend on some factors like kind, doses and duration of their use.

Table 2. Effect of probiotic supplementation on nutrient digestibility and feeding values of the experimental diets.

Itom		Experimental diet							
nem	T1	T2	T3	T4	SEM				
Digestion coefficients (%):									
DM	61.80 <sup>b</sup>	64.95 <sup>a</sup>	63.55 <sup>ab</sup>	64.56 <sup>ab</sup>	0.82				
OM	62.67 <sup>b</sup>	66.10 <sup>a</sup>	64.89 <sup>ab</sup>	65.45 <sup>a</sup>	0.72				
CP	71.88 <sup>c</sup>	78.36 <sup>a</sup>	73.56 <sup>bc</sup>	75.09 <sup>b</sup>	0.63				
CF	21.84 <sup>b</sup>	24.95 <sup>a</sup>	23.54 <sup>ab</sup>	24.53 <sup>a</sup>	0.60				
EE	74.80 <sup>b</sup>	79.45 <sup>a</sup>	77.80 <sup>ab</sup>	77.90 <sup>ab</sup>	0.97				
NFE	78.20	81.94	81.05	81.90	1.16				
Feeding values (%, DM basis):									
TDN	62.70 <sup>b</sup>	66.53 <sup>a</sup>	64.42 <sup>ab</sup>	65.82 <sup>a</sup>	0.68				
DCP	12.50 <sup>c</sup>	13.63 <sup>a</sup>	12.80 <sup>bc</sup>	13.07 <sup>b</sup>	0.11				

SEM = Standard error of means a, b and c: Means within rows with different superscript are significantly different (P<0.05). T1: control group; T2: 1 g of SP/h/d; T3: 1g of LY/d/h; T4:0.5 g of BS /h/d.

#### Feed intake:

Results obtained in Table 3, indicated that the additive supplementation had no significant impact on feed intake for pregnant and lactating does. These finding were consistent with the studies concluded by Seyidoğlu and Galip (2014), Tag El-Din (2019), Ribeiroa et al. (2020), Wang et al. (2020), Elmasry et al. (2021), and Colombino et al. (2022), which indicated the inclision of SP, Nannochloropsis oceanica or various probiotics in rabbit rations achieved comparable findings. However, the findings of Phuoc and Jamikorn (2017), Sikiru et al. (2019), and Abdou et al. (2024) found that supplementing probiotics, either alone or mixed with others, were appeared to be in contrast to our results. On the other hand, our results showed the daily feed intake through the lactation period increased by 44.84% compared to that of pregnancy period (Table 3), where such results are similar to the findings obtained by Amira M. Refaie et al. (2022).

### Body weight changes:

During the different experiment stages, probiotic supplementation didn't significantly affect on rabbit body weight changes, except for the prepartum period, where weight changes in T2 and T4 groups showed significantly (P<0.05) higher than those of T1 group. The differences observed with the T3 group were not significant, as shown in Table 3. Moreover, there was a positive change in BW during gestation with the highest value in T4 followed by T2 then T3, but T1 had the lowest values. However, there was negative change in BW during lactation with the highest value in T1 followed by T3 and T2, while the lowest value recorded with T4.

These results are in line with those reported by Seyidoğlu and Galip (2014), Fathi *et al.* (2017), Emmanuel *et al.* (2019) and Tag El Din (2019),. Nevertheless, the findings of Wang *et al.* (2020), Elmasry *et al.* (2021), Abd El-Aziz *et al.* (2021), El-Deep *et al.* (2021), and Abdou *et al.* (2024), showed significant improvements in rabbit body weight after consuming probiotic rations. Moreover, during the lactation period, the lost body weight of rabbits in the experimented groups was -95.0,-103.3, and -84.2 g for T2, T3, and T4, respectively, compared to the - 118.3 g for the control ration (T1). These results were agreement with those reported by Abdou *et al.* (2024) after SP was added to the doe rabbit rations.

 
 Table 3. Impact of experimental rations on the productive performance of does during gestation and lactation periods.

Sebution and metation periods.							
Itom	]	SEM					
Item	T1	T2	T3	T4	SEAM		
Feed intake (g/h/d):							
Pregnant does	182.7	175.0	188.9	179.5	6.42		
Lactating does	264.0	251.5	270.5	265.6	11.70		
Average	223.35	213.25	229.7	222.55	9.06		
	Doe	weight (g	):				
At mating	3273.3	3281.7	3274.2	3265.0	77.39		
Pre-partum	3595.0 <sup>b</sup>	3697.5 <sup>a</sup>	3668.3 <sup>ab</sup>	3699.2 <sup>a</sup>	31.84		
Partum	3268.3	3290.0	3285.0	3297.5	93.44		
Weaning	3150.0	3195.0	3181.7	3213.3	93.22		
Doe weight change (g):							
Mating - pre-partum	321.7°	415.8 <sup>ab</sup>	394.2 <sup>b</sup>	434.2 <sup>a</sup>	58.78		
Mating -partum	-5.0°	8.3 <sup>b</sup>	10.8 <sup>b</sup>	32.5 <sup>a</sup>	1.16		
Mating-weaning	-118.3 <sup>a</sup>	-95.0 <sup>bc</sup>	-103.3 <sup>ab</sup>	-84.2 <sup>c</sup>	3.33		
SEM = Standard error of means <sup>a, b, and c,</sup> : Means in the same row with							

different superscripts are significantly different (P<0.05). T1: control group; T2: 1 g of SP/h/d; T3: 1g of LY/d/h; T4:0.5 g of BS /h/d.

### Milk yield (MY)

Data presented in Table 4 indicated that the dietary probiotic showed a significantly positive effect on milk yield during the 1<sup>st</sup> week. Additionally, during 2<sup>nd</sup> week, the T2 and T4 groups had significantly (P<0.05) higher milk yield compared to the T1 group, with no significant differences. Furthermore, the T2 and T4 groups recorded significantly (P<0.05) increased milk yield during 3<sup>rd</sup> week. In the 4<sup>th</sup> week, milk yield in the T4 group was significantly (P<0.05) greater than in the T1 group, with no significant differences observed between T2, T3 and T4 groups. Probiotic groups (T2, T3, and T4) appeared to higher milk yield as overall mean by 16.11, 11.26 and 18.50, respectively, based on the control group (T1).

The positive effect of SP on MY, might be due to its richness in bioactive compounds, including natural antioxidants along with other essential substances that enhance metabolic processes (Komoto *et al.*, 2018; Sharawy *et al.*, 2020). It also increased mRNA expression of glutathione peroxidases, which have antioxidant properties and help maintain animal health, productivity, and reproductive performance (Saleh *et al.*, 2011). Additionally,

it reduces stress and protects cells such as mammary gland cells from lipid peroxidation (Ohtsuka *et al.*, 1998). These findings were <del>are</del>-greatly supported by those outputted by Morsy and Abd El-Lateif (2017), Fahim *et al.* (2021), Amira M. Refaie *et al.* (2022), and Abdou *et al.* (2024). Moreover, milk yield increased gradually up to the third week of lactation and then decreased in the fourth one, as previously documented by El-Sayiad (1994), who reported that rabbits getting peak milk production in lactation during the third week.

 Table 4. Impact of experimental rations on the milk intake of kits from doe rabbits.

Itom	H	SEM						
Item	T1		T3	T4	SEIVI			
	Milk yiel	d (g/d) at	t:					
1 <sup>st</sup> week	93.7 <sup>b</sup>	117.0 <sup>a</sup>	113.0 <sup>a</sup>	117.1 <sup>a</sup>	3.416			
2 <sup>nd</sup> week	140.6 <sup>b</sup>	154.0 <sup>a</sup>	151.2 <sup>ab</sup>	158.3 <sup>a</sup>	4.151			
3 <sup>rd</sup> week	177.5 <sup>c</sup>	211.3ª	197.7 <sup>b</sup>	217.3 <sup>a</sup>	4.475			
4 <sup>th</sup> week	106.9 <sup>b</sup>	119.9 <sup>ab</sup>	115.5 <sup>ab</sup>	122.3 <sup>a</sup>	5.038			
Average milk yield (g/d)	129.7 <sup>c</sup>	150.6 <sup>a</sup>	144.3 <sup>b</sup>	153.7 <sup>a</sup>	1.591			
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SEM = Standard error of means <sup>a,b,andc</sup>, : Means in the same row with different superscripts are significantly different (P<0.05)

T1: control group; T2: 1 g of SP/h/d; T3: 1g of LY/d/h; T4:0.5 g of BS/h/d.

### **Reproductive performance:**

Data in Table 5 showed that there were no significant differences in the conception rate. The same significant results were found with litter size at birth. Meantime, mostly dietary supplementation groups had enhanced the size and weight of litter (p<0.05) than those of the control one, over at weaning. Treated groups T2, T3, and T4 showed that increased litter size by 25.42, 10.17, and 27.12% and increased litter weight by 26.64, 10.98, and 29.56%, respectively, compared to those of control group (T1).

The beneficial effect of SP on most reproductive parameters of doe rabbits can be linked to its high content of natural antioxidant pigments, vitamins and minerals (Abdelkhalek et al., 2015; Niu et al., 2017). These components have been strongly play a vital role in hormonal regulation, improving fertility, restoring antioxidant ovary status (Abadjieva et al., 2011; Yener et al., 2013), and immune system enhancement (Mirzaie et al., 2018). The results were agreement with those found by Abd El-Hamid et al. (2022). Moreover, Abdou et al. (2024) mentioned that adding SP in doe rabbit rations increased litter size and weight at birth and weaning periods. In contrast, Iatrou et al. (2022) found that SP supplementation at 100 mg/kg of body weight to female mink didn't affect their litter size or weight. Also, the addition of LY and BS supplements improved productivity and reproduction of doe rabbits through several mechanisms, like competition with harmful microbes and repression of their toxic effects, which led to enhanced intestinal and immunity functions (Elghandour et al., 2019; Abdel-Wareth et al., 2021). In this respect, the data were agree with those of Morsy and Abd El-Lateif (2017), Dimova et al. (2017), and Fahim et al. (2021), which have emphasized an increases in litter size at birth or weaning period. Recently, Amira M. Refaie et al. (2022) recommended that adding probiotics or higher levels of Aspergillus awamori led to increases in the size and weight of litter at the weaning phase.

On the other hand, the daily gain of kids in probiotic groups was significantly (p<0.001) higher than that of control group, being 108.50, 139.92, 121.89, and 143.90 for the T1,

T2, T3, and T4 groups, respectively. As reported by Pascual *et al.* (1996), there is a strong relationship between milk yield and improved birth litter size and health. These results were aligned with those recognized by kritas *et al.* (2008), Kadja *et al.* (2021), and El-Deep *et al.* (2021). Similarly, Abdou *et al.* (2024) observed that rabbits feeding on SP rations had improved the daily gain of their offspring at weaning. Conversely, Seyidoglu and Galip (2014) noted that the body weight of rabbits was not affected by adding 3g of SC or 5% of SP or their mixture in their rations.

Table 5 reveals that feed conversion rate as gm feed /litter size weight weaning was significantly (P<0.05) improved with SP and BS groups than that of LY and control one over 1-21 days of lactation. While, feed conversion rate for all tested rations was significant (P<0.05) improved than that of the control one (T1), during the period from 22 to 30 days of lactation. Results appeared to a harmony with those obtained by Sikiru et al. (2019), Wang et al. (2020), El-Deep et al. (2021), Kadja et al. (2021), and Abdou et al. (2024) when probiotics were added alone or in a mixture form. Also, Abbdel-Wareth et al. (2021) reported that rabbits received higher levels of mixed probiotics combined fenugreek could be enhanced gut health and growth performance as well as improved feed conversion. However, Lugar'a et al. (2022) reported that adding spirulina to the high-energy diets for sows didn't affect feed intake or FCR through the gestation and lactation periods. Also, Fathi et al. (2017) reported that FCR had no effect by adding two levels of BS to the diets of rabbits

Regarding the mortality rate (MR%) measurement, dietary supplementations significantly (P<0.05) lowered the mortality rate in comparison to the control group during the first period, while during the second period from 22 to 30 days, the groups (T2 and T4) had significantly (P<0.05) decreased MR% than those in the control group. While, there was an insignificant reduction in MR% with LY group with comparison with those of control one.

Enhancing the production and reproductive performance and decreasing mortality rate with the addition of SP, which might be able to reduce oxidative stress during fertilization, because it contains essential materials such as total phenolic acids, flavonoid compounds, antioxidants and immune modulatory enhancers (Mirzaie et al., 2018). Additionally, as explained by Ogawa et al (2001) feeding probiotics could be improved IgA released and protected against E. coli enteritis, resulting in a decreased mortality rate. Moreover, Kritas et al. (2008) reported that "feeding probiotics may have a growth-promoting activity by competing with harmful gut flora and stimulating the immune system". In addition, Kimsé et al. (2012) explained that probiotic supplementation could be modifying the physicochemical characteristics as an increase in the redox potential of caecum, which leads to improved digestive health. The same authors indicated that the mortality rate of growing rabbits fed S. cerevisiae rations was lowered by 22.5% compared to 45% for those fed a control ration. Furthermore, our results are consistent with those of Dimova et al. (2017) and Abdou et al. (2024) when probiotics were added to rabbit rations. Also, Belhassen et al. (2016) reported that yeast supplementation enhanced fertility and pup survival, as well as reducing the mortality rate of doe rabbits. Additionally, Morsy and Abd El-Lateif (2017) found that the mortality rate

of pups decreased with increased mannan oligosaccharides in rabbit rations. Contrary to our results Colombino *et al.* (2022) noticed that rabbit-fed Lactobacillus acidophilus rations recorded a higher MR% compared to that of control one.

Table	5.	Effect	of	experimental	rations	on	the
		reprod	lucti	ve performance	of doe ra	abbits	5

Térrere	F	Experimental diet					
liem	T1	T2	T3	T4	SEIVI		
Conceptions rate	70	80	80	80	5.03		
Litter size:							
At birth (alive)	7.5	8.3	7.8	8.2	0.428		
At weaning (30 days)	5.9 <sup>b</sup>	7.4 <sup>a</sup>	6.5 <sup>ab</sup>	7.5 <sup>a</sup>	0.372		
	Mortality	rate:					
Birth $-21$ days	21.7ª	8.7 <sup>b</sup>	10.8 <sup>b</sup>	7.3 <sup>b</sup>	3.690		
Birth – Weaning	21.33 <sup>a</sup>	10.84 <sup>b</sup>	16.67 <sup>ab</sup>	8.54 <sup>b</sup>	3.553		
I	itter weig	ght (g):					
Birth	405.5	437.5	405.5	452.5	12.26		
Weaning (30 days)	3660.5°	4635.0 <sup>a</sup>	4062.0 <sup>b</sup>	4742.5ª	49.97		
Daily gain of litter (g/d)	108.5°	139.92 <sup>a</sup>	121.89 <sup>b</sup>	143.9 <sup>a</sup>	37.48		
Daily	feed inta	ke (g/d) f	or:				
Lactating does	264.0	251.5	270.5	265.6	11.70		
Kids (22-30 day)	208.6	198.7	213.7	209.9	5.830		
Feed conversion ratio (g/g):							
1 to 21 days of lactation <sup>(1)</sup>	2.98 <sup>a</sup>	2.46 <sup>b</sup>	2.96 <sup>a</sup>	2.57 <sup>b</sup>	0.107		
22 to 30 days of lactation $^{(2)}$	3 16 <sup>a</sup>	1 99°	2 52 <sup>b</sup>	$2.04^{\circ}$	0.084		

SEM = Standard error of means<sup>a, b, c, and d</sup>: Means in the same row with different superscripts are significantly different (P<0.05).

T1: control group; T2: 1 g of SP/h/d; T3: 1g of LY/d/h; T4:0.5 g of BS/h/d. (1) As feed intake of does from 1 to 21 days (g) per litter weight gain from 1 to 21 days (g).(2) As feed intake of does from 22 to 30 days (g) per litters weight gain from 22 to 30 days (g).

#### **Blood parameters:**

Table 6 showed that the TP, AL, and GL concentrations were significantly (P<0.05) higher, while blood serum urea and creatinine levels were significantly (P<0.05) lower in the tested rations compared to those of the control one. Otherwise, no significant differences in respect of blood serum AST and ALT concentrations were observed among all experimental groups.

Blood serum TP and GL concentrations are considerably working as indicators of immune system activity, while the Al concentration in blood serum serves as indicator of liver function (Ismail et al. 2002; White et al., 2002; Selem et al., 2007). According to the studies of Wang et al. (2020), BS could be increased each of TP, and GL, which might be improving protein and carbohydrate metabolism (Stanley et al., 2002; Oh et al., 2008). Also, according to the results of Farag et al. (2016), the contents of TP, Al and GL were elevated with the addition of SP where such these increases might be due to its rich content of highquality protein (all amino acid contents), phospholipids and antioxidant substances, as well as excellent minerals and vitamins. Recently, Abo Ghanima et al. (2020) and Kadja et al. (2021) found that adding probiotics and prebiotics could enhance nutrient utilization and improve immune system functions by increasing globulin levels (Terciolo et al., 2019). These results are in harmony with those obtained by Seyidoglu and Galip (2014), Fahim et al. (2021), and Kadja et al. (2021). Furthermore, Helal et al. (2021) noticed that adding LY, SP, or their mixture in rabbit rations significantly raised TP, Al, and GL levels in comparison to the control one. Abdou et al. (2024) also mentioned that increasing SP levels in rabbit rations improved TP and Al levels and lowered Gl levels. In contrast, Beshara et al. (2018) reported that the addition of probiotics had lowered TP and Gl levels. On the other hand, all additive groups lowered blood serum creatinine and urea levels than those of control group, in order to it may have enhanced protein metabolism and improved kidney functions. Likewise, Mahmoud et al. (2017) and Abdou et al. (2024) noticed that similar findings in rabbits that were fed probiotic rations. In contrast, Seyidoglu and Galip (2014), Fahim et al. (2021), and Kadja et al. (2021) reported that the addition of probiotics to rations of rabbits didn't affect blood urea, or creatinine concentrations. Moreover, probiotic supplementation didn't affect blood serum ALT and AST levels, indicating these additives didn't negatively influence liver function and such findings are in line with those found by Seyidoglu and Galip (2014), Abdelhady and El-Abasy (2015) and Helal et al. (2021). Also, Abdou et al. (2024) observed that adding SP at high levels in rabbit rations didn't affect blood serum AST levels and decreased ALT levels significantly (P<0.05). Moreover, Belhassen et al. (2016) didn't find any effects on blood parameters when yeast was added to rabbit rations. Similarly, Hassanein et al. (2014) had observed like these results when added some algae species to rabbit rations.

Table 6. Blood serum constituents of doe rabbits fed the experimental diets.

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Item	T1	T2	T3	T4	SEM
Total protein (g/dl)	6.16 <sup>c</sup>	7.22 <sup>a</sup>	6.86 <sup>b</sup>	7.12 <sup>a</sup>	0.19
Albumin (g/dl)	3.13 <sup>c</sup>	4.58 <sup>a</sup>	4.42 <sup>a</sup>	4.50 <sup>a</sup>	0.13
Globulin (g/dl)	3.03 <sup>a</sup>	2.64 <sup>b</sup>	2.44 <sup>c</sup>	2.62 <sup>b</sup>	0.25
Creatinine (mg/dl)	1.19 <sup>a</sup>	0.97 <sup>b</sup>	1.00 <sup>b</sup>	0.98 <sup>b</sup>	0.05
Urea (mg/dl)	16.26 <sup>a</sup>	12.34 <sup>b</sup>	12.96 <sup>b</sup>	13.01 <sup>b</sup>	1.53
AST (IU/l)	22.51	22.05	20.47	17.24	0.4
ALT (IU/I)	25.41	23.34	22.16	19.41	4.62

SEM = Standard error of means<sup>a, b and c</sup> : Means in the same raw with different superscripts are significantly (p<0.05) difference.

T1: control group; T2: 1 g of SP/h/d; T3: 1g of LY/d/h; T4:0.5 g of BS/h/d.

### Economic feed efficiency:

The results presented in Table 7 revealed that although doe rabbits fed probiotic rations had higher total feed costs compared to the control, they showed the best values economically compared with the control.

 Table 7. Impact of probiotic supplementation on the economic efficiency of doe rabbit production.

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Itom	Experimental diets						
Itelli	<b>T1</b>	T2	T3	T4			
Feed cost of doe (L.E.)	117.6	6112.38	121.01	117.21			
Additive cost of doe (L.E.)	0.00	18.00	2.40	3.00			
Total feed cost of doe (L.E.)	117.6	6130.38	123.40	120.21			
Selling price of kids per doe (L.E.) <sup>1</sup>	146.4	2185.40	162.48	189.72			
Economic feed efficiency $(\%)^2$	124	142	132	158			
Economic efficiency relative to control (%)	<sup>3</sup> 100.0	0114.52	106.45	127.42			
T1: control group; T2: 1 g of SP/h/d;	T3: 1g	of LY/c	ł/h; T4:	0.5 g of			
BS/h/d Other conditions like mortality (	(%) and	manage	ement ar	e fixed.			
- Price (L.E. per kg) at 2021 was: 8.78 PD; 300 SP; 40 LY; 100 BS.1 Price of kg							
live body weight was 40 L.E.2 Economic efficiencies = (selling price/ total feed							
cost) x100.3 Economic efficiency relative to control, % = (economic efficiency							
of T2, T3 and T4 x 100/ economic efficiency	of T1) x	100.					

Generally, doe rabbits fed T4 showed the highest economic feed efficiency followed by T2 and T3, but those fed the control diet showed the lowest values. Data were comparable with those found by Abdou *et al.* (2024) when adding SP in rabbit rations. Also, Fahim *et al.* (2021) noticed that the inclusion of Biogen with two levels in the rabbit rations improved economic efficiency compared to the unsupplemented ration. Moreover, Abd El-Aziz *et al.* (2021) stated that the best economic efficiency obtained when yeast was added to the rations of rabbits as growth promoter. Accordingly, to the results of Kalma *et al.* (2018), where they were added SC and *lactobacillus sporogenes* at 0.5g/kg feed and therefore such treatment was enhanced profitability of rabbit's production. Furthermore, under heat stress, Morsy and Abd El-Lateif (2017) found that the highest economic efficiency was recorded with increasing mannan oligosaccharides at levels up to 1.5 g in the diets of rabbit does.

# CONCLUSION

In the present study, adding *spirulina* or *Bacillus subtilis* to rabbit rations can be a positive strategy for enhancing productive and reproductive performance and health status, of doe rabbits and also improving the daily gains and economic efficiency of their offspring.

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# الآداء الإنتاجي والتناسلي للأرانب المغذاة على علائق مدعمة بالاسبير ولينا والخميرة والباسيللس ستليس كإضافات طبيعية

# على أحمد عبده ، شامه حسنى أحمد مرسى و محمد رفاعي محمود مصطفى

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# الملخص

تم تصميم هذه الدراسة للتعرف على تكثير إضافة اسبير ولينا بلاتنسيس والخميرة الحية والبلسيلاس سابتيليس على هضم العاصر الغالنية والأداء الإنتلجي والتناسلي لأمهات ا الأرانب. تم تقسيم أرجعين من إنك الأرانب يبلغ عمر هم ثمانية أشهر ووزن 5±3225 جم إلى أربع مجموعات تجريبية مماثلة وفقًا لوزن الجسم الحي. تم تغنية الأرانب على الطيقية الكنتر ول الخالية من البروبيوتيك (كمجموعة تحكم T1)، وتم تغنية الأرانب على الطيقية الكنتر ول الخالية من البروبيوتيك (كمجموعة تحكم 21)، وتم تغذية الأرانب على الطيقية الكنتر ول الخالية من البروبيوتيك (كمجموعة تحكم 21)، وتم تغذية المجموعات 12 و 31 على نفس النظلم الغذائي مع إضافة 1 جرام من الاسبيرولينا و 1 جرام من الخميرة الحية و 2.0 جرام من البسياس ستيلس إلى العلف المقول. تساد المي البليس ستيلس إلى العلف المائم الغذائية في موجوعة معام على التعربي والتناتيج إلى أن إنتاج اللين زاد بشكل ملحوظ (20.05) م) مع مكملات البروبيوتيك ولكنها لم تؤثر على كمية العلف المأكول. تحسنت قابلية هضم معظم العنصر الغذائية في مجموعة السبير ولينا ثم مجموعة البلسيل ستيلس إلى العلف المائم الحزائم مجموعة البليل سنيلس إلى العلق الغن الغذائية في مجموعة الي ألي الندين الذي مع معلم المائم الخراب عنه معام العنصر الغذائية في مجموعة السبير ولينا ثم مجموعة البليلاس سابتيلس مقارت مع من المائم المأكول. تحسنت قابلية هضم معظم الخام الغذائية في مجموعة السبير ولينا ثم مجموعة البليل المنولي بعن أن لار التب معلم المائم المائي للموري يوني والذائم مجموعة البير ويوتيك إلى والدين وي والمائم المنول في محموعة البير ويوتيك إلى ولا يتنا و ويوتيك المائم المائم المائم المائم ولادة حيث محمو علي أول الأر التب التنامي للموري والمو ولي المجموعات 21 و 13 ولم محموعة المائي الموط في المائمة للأمهات بشكل ملحوظ في مجموعات البروبيوتيك ولكن والولي ولي الموري علي الموري والمائين المائم والمائم المائم المائم المائم المائم ولدي المائم المائم واليا وو والبسيلاس مركز ال البر ويتين الكلى والأليومين والمويوني في المولية والمائم والمائم الموط في محموع مائر الروبية الى وليسير ول سابتيلس إلى علي والأليومين والجلوبيولين في المائم المائم المولية والمائم والمائم والمائم الحالية المائم والمائم والمائم والمائم المولية والم المولية المائم والمائم والم واليبير والي والل واليب وال