Egyptian J. Nutrition and Feeds (2024), 27 (3):355-372

REPRODUCTIVE EFFICIENCY, BLOOD METABOLITES, AND PRODUCTIVITY OF HI-PLUS DOE RABBITS FED VARIOUS BIOLOGICAL PROBIOTIC ADDITIVES UNDER NORTH SINAI CONDITIONS IN EGYPT

A.S. El-Hawy

Animal and Poultry Physiology Department, Desert Research Center, Egypt.

E-mail: ***ahmedelhawy78@yahoo.com***

(Received 15/10/2024, accepted 10/11/2024)

SUMMARY

This investigation aims to study the effect of probiotics ZAD®, ZADO® and YEAST® supplementation on blood parameters, immunological responses, and reproductive and productive performance of Hi-Plus doe rabbits under North Sinai conditions. Sixty Hi-Plus doe rabbits, aged 5 months and body weight 3190.2±30.7 g, were used in this study from May to September 2022. Does were randomly divided into four similar treatments. The 1st treatment (Tr1) was served as a control without supplementation. Does received 10 g of ZAD® / kg basal diet (1.0 %) in the 2nd treatment (Tr2), 10 ml of ZADO® /kg basal diet (1.0 %) in the 3rd treatment (Tr3), and 10 g of YEAST/ kg basal diet (1.0 %) in the 4th treatment (Tr4) during the experimental period. The obtained results demonstrated that applying biological additives increased (P<0.05) glucose, total protein, globulin, triiodothyronine (T3), immunoglobulin (IgA), and total antioxidant levels, while decreased (P<0.05) total cholesterol and triglycerides in blood plasma of the treated doe rabbits. Progesterone (P4) significantly increased in the treated groups compared to control doe rabbits. Indeed, immunoglobulin levels of IgG and IgM were increased (P<0.05) only in the doe rabbits of Tr4 compared with doe rabbits in Tr1. Conception rate, milk yield, litter size and weight at birth and weaning as well as productive efficiency index was higher (P<0.05) in the treated doe rabbits than in control ones. Feed conversion value was improved (P<0.05) in the treated doe rabbits compared with control group. Activity of plasma transaminase (ALT) decreased (P<0.05) only in the doe rabbits of Tr2 and Tr4 compared to doe rabbits in Tr1. In conclusion, under heat stress conditions, dietary supplementation of biological probiotics (ZAD®, ZADO® and YEAST®) at a level of 1% could enhance the reproductive and productive performance, increase milk yield, reduce pre-weaning mortality, improve blood constituents and immunological status of doe Hi-Plus rabbits. Supplementation of 10 g YEAST /kg diet showed the best impacts as compared to other supplements.

Key words: *Rabbits, probiotics, reproductive and productive performance, immunity.*

INTRODUCTION

In Egypt, rabbit production are suffering from heat stress which considered as one of the important environmental stressors challenging rabbit productions, where it causes deleterious effects on physiological responses, productive and reproductive performance of rabbits (Amer *et al.*, 2013; Sakr *et al.*, 2019). Under severe heat stress, rabbits have a depression in feed intake and efficiency of feed utilization, disturbances in water, protein, energy and mineral metabolism balances, enzymatic reactions, hormonal secretions, and blood metabolites as well as a depression in immune function (Abd El-Hamid *et al.*, 2022). Nowadays, probiotics are used as growth promoters on a large scale to avoid the presence of residues of antibiotics in animal products used in human consumption. Probiotics have been reported to positively affect growth performance and health of rabbits (Ezema and Eze, 2012; Bhatt *et al.*, 2017). However, studies on the use of yeast as a probiotic in rabbit does are scarce. Indeed, the results of Ayyat *et al.* (1996) suggested that the inclusion of the probiotic (Lacto-Sacc) in diets of lactating doe rabbits increased milk production, litter size, and weight at weaning. A large number of natural growth promoters, including immune-boosters, probiotics, and prebiotics are currently available on the market. Under this point of view, the anaerobic probiotic technology (ZAD)® may be a substitute for antibiotic growth promoters in the feeding idea (Gado *et al.*, 2017). The ZAD® is a patented product manufactured by the Academy of Scientific Research and Technology, Egypt. It is an enzyme biotechnology product from natural sources to raise the anaerobic bacteria cellulase enzyme levels that can transform polysaccharides by the enzyme catalytic process to monosaccharides. The product is designed in a liquid form to provide tools to improve the nutritional value of fibrous materials and to improve the overall digestion of animals. It includes the subsequent activity of enzymes such as hemicellulose (6.2 U/g), cellulose (8.2 U/g), protease (12.3 U/g), and amylase (64.4 U/mg) (Gado *et al.*, 2011, 2013 and 2017), besides the anaerobic bacteria that create these enzymes (Abdel-Aziz *et al.*, 2015). Anaerobic probiotic (ZAD®) is a dietary 1.5 liter for rabbits that helps animals to overcome heat stress. It augmented production of milk, raised the daily average gain, enhanced throughout the physiological aspects and lowered the rabbit mortality rate (Gado, 2020).

The probiotic product of ZAD® is a liquid biotechnical product made from natural sources of bacteria to elevate the level of multi-enzyme such as cellulase, xylanzses etc. The product is intended to provide tools to improve the nutritive value for fibrous materials through ZAD® and ZADO® is designed to improve the rumen kinetics and overall animal digestion. Gado *et al.* (2015 and 2016) revealed that the addition of ZADO to commercial diet increased conception and kindling rates, litter size, weights at birth and weaning and milk yield, while decreased abortion and preweaning mortality rates. The addition of 1 g of YEAST (Actisaf Sc47, LESAFFRE)/ kg of diet enhanced fertility and reduced mortality rate of rabbit does and improved the viability rate of kits at birth (Belhassen *et al.*, 2016).

This research aimed at studying the efficacy of ZADO®, ZAD® and YEAST®, as biological additives, to enhance productive and reproductive performance as well as immunological and blood metabolites in Hi-Plus doe rabbits under the conditions of North Sinai Governorate.

MATERIALS AND METHODS

An experiment was carried out in accordance with the guidelines laid down by the Institute of Animal Ethics Committee for the use of animals (2010/63/EU of the European Parliament and of the Council of September 22, 2010).

Study region:

This investigation was implemented at a private rabbit farm (Latitude 31° 29′ N; Longitude 32° 34′ E), Governorate of North Sinai, in cooperation with Department of Animal and Poultry Physiology, Desert Research Center, Ministry of Agriculture, Cairo, Egypt, Systel Telecom Company and Egyptian Center of Excellence for Biosaline Agriculture.

Animals, management and experimental design:

A total of 60 virgin Hi-Plus doe rabbits, 5 months old with an average body weight of 3190.2±30.7 g were used in this experiment. The study was conducted from May to September 2022. The rabbits were housed in standard dimensions (50×60×40 cm) wired metallic cages attached with nest box (40×30×27 cm) for kindling and nursing. All does were fed a commercial concentrate pelleted diet containing 18.0% crude protein, 15.0% crude fiber, 2.5% fat, 0.6% minerals mixture and 2600 kcal/kg digestible energy according to NRC (1994). Fresh water was available all days through nipples drinker system.

Doe rabbits were divided into four similar treatments: The 1st treatment (Tr1) wasserved as control without supplementation. Doe rabbits received 10 g of ZAD® / kg basal diet (1.0 %) in the 2nd treatment (Tr2), 10 ml of ZADO® /kg basal diet (1.0 %) in the 3rd treatment (Tr3), and 10 g of YEAST®/ kg basal diet (1.0 %) in the 4th treatment (Tr4).

Probiotic ZAD® is a mixture of anaerobic bacteria (Ruminococcus flavefaciens 1×10^9 CFU/L) and exogenous enzymes. Probiotic ZADO® is a mixture of anaerobic bacteria (Ruminococcus flavefaciens 1×10^{10} CFU/L) and exogenous enzymes. Both ZAD® and ZADO® were kindly provided by Bactizad Company. A commercial *saccharomyces cerevisiae* was used in the current study.

Meteorological data:

Ambient temperature (°C), relative humidity (RH, %) were recorded at 8 a.m. and 2 p.m. using hygro-thermometer. According to Marai *et al.* (2002), temperature-humidity index (THI) was calculated using the following equation: THI= (0.8×AT, °C) + [(RH/100) × (AT, °C–14.4)] + 46.4. Where, db °C = dry bulb temperature in centigrade and RH = relative humidity (%). The THI values were classified as absence of heat stress (<27.8), moderate heat stress (27.8- 28.8), severe heat stress (28.9- 29.9) and very severe heat stress (>30.0). Meteorological data during different experimental months are presented in Table 1.

Table (1): Indoor ambient temperature, relative humidity, and temperature-humidity index throughout experimental period.

Months	**Min AT (ºC)**	**Max AT (ºC)**	**Min RH (%)**	**Max RH (%)**	**Min THI**	**Max THI**
May	25.7	32.1	42.5	54.1	24.1	28.9
June	25.5	32.4	41.0	51.3	23.8	29.1
July	27.3	33.6	41.0	53.8	25.4	30.1
August	27.8	34.8	45.5	62.8	26.2	31.5
September	27.5	33.6	45.5	67.0	26.2	30.3
Overall	25.7	33.3	43.1	57.8	25.1	30.1

AT= ambient temperature, RH= relative humidity, THI= temperature humidity index

Thermo-respiratory responses:

At 2.00 pm, fife doe rabbits per experimental group were used to measure rectal, fur, ear, and skin temperatures (RT, FT, ET, and ST, °C) and respiratory rate (RR, breath/minute). Rectal temperature as body temperature indicator was measured using a calibrated clinical thermometer (Model Incoterm, Porto Alegre /RS, Brazil with ± 0.1 °C accuracy), with a temperature range of 32–43.9 °C. Temperature degrees of skin, fur and ear were recorded using digital tele-thermometer. Respiration rate (RR, breath/minute) was measured by counting the wave cycles of the breast up and down per 30 s, and the obtained value was multiplied by 2 to give the number of breaths per minute. Thermos-respiratory values were measured two times per month and the monthly averages of RT, FT, ET, ST, and RR were calculated in the four experimental groups.

Blood sampling:

Blood samples were collected biweekly from all doe rabbits from the ear vein in plasma vacutainer tubes (5 ml) throughout the experimental period to determined blood metabolites, triiodothyronine and progesterone. The blood samples were centrifuged for 15 minutes at 5000 rpm, then plasma samples were separated and stored at −20 °C for later analysis.

Blood metabolites analysis:

Plasma total proteins, albumin, glucose, cholesterol, tri-glycerides, creatinine, alanine aminotransferases (ALT), aspartate aminotransferase (AST), and urea were analyze using commercial chemical kits (Biomed diagnostics, Egypt). Globulin concentration was calculated. Total antioxidant capacity (TAC), was calorimetrically assayed using commercial kits (Biodiagnostic Research, Egypt).

Hormonal assay:

Triiodothyronine (T_3) and progesterone (P_4) hormones were analyzed using ELISA kits (Monobind, USA) according to Wheeler *et al.* (1994) and Abraham (1974), respectively. The intra -and inter-assay CV's were 9.3 and 8.83, respectively. However, blood samples for progesterone hormone analysis were collected on days 7, 14, 21 and 28 post-mating.

Immunological parameters:

Immunoglobulin G (IgG), immunoglobulin A (IgA), immunoglobulin M (IgM), were analyzed using commercial kits (Reactivos GPL Barcelona, España).

Reproductive and productive performance:

In this experiment, two parities were obtained from each doe in all groups. Conception rate (CR, %), number of services per conception (NSC), and gestation period length (GL, day) were recorded. Litter traits, mortality from birth to weaning and stillbirth rates were also recorded.

Economic indicators and efficiency:

Total feed intake (TFI, kg) = daily feed intake×120 days, feed conversion (FC) = feed conversion (TFI / PI)., and productive efficiency index (PEI, kg live weight) = litter size at weaning × number of parities (2 parities) × total weaning weight (kg), were calculated. The data were calculated based on the Egyptian market prices of diets at 2022 as follows: Total feeding cost (L.E.) = TFI (kg) × (cost of kg feed +cost of kg biological additive). Cost of one kit production = total feeding cost (LE) / litter size at weaning per doe.

Statistical analysis:

Data were analyzed by one-way analysis procedure (ANOVA) using the General Linear Model Procedure (SAS, 2004).

The model was as follows:

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

Where, Y_{ij}= any observations of i^{th} rabbit within j^{th} treatment, μ = overall mean, T_i = effect of i^{th} treatment, (i: 1-4), e_{ij} = standard error.

All statements of significance are based a probability of less than 0.05. Significant differences among means were tested using Duncan multiple range test (Duncan, 1955).

RESULTS AND DISCUSSION

Thermo-respiratory responses:

Dietary addition of ZAD, ZADO, and Yeast (Tr2, Tr3 and Tr4, respectively) decreased ($P \leq 0.05$) the thermos-respiratory responses (RT, FT, ET, ST, and RR) as compared to of the control diet in Tr1 (Table 2). No significantly differences were observed in thermos-respiratory responses among treatments (Tr2, Tr3 and Tr4). Thermoregulation is defined as the maintenance of the temperature(s) of the body within a restricted range under conditions of internal and/or external heat loads (Bligh and Johnson, 1973). The comfort zone for rabbits under Egyptian conditions is between 18 and 21 ºC (Marai *et al.*, 2002; Ahmed, Nagwa *et al.*, 2000 and 2005). Exposure to high ambient temperature induces rabbits to try to balance their excessive heat load by using different means to dissipate the latent heat. If such means fail, rabbits use strategies that including depression in feed intake and efficiency of feed utilization as well as disturbances in water, protein, energy, and mineral metabolism balances, enzymatic reactions, hormonal secretions, and blood metabolites (Habeeb *et al.*, 1992). Treated doe rabbits with biological additives during heat stress conditions might resulted in more controlled thermo-respiratory responses and/or a decrease of energy resulting in decreasing serum cholesterol and triglyceride concentration (Gado, 2020).

Table (2): Mean ± SEM of thermo-respiratory responses in Hi-Plus doe rabbits fed on biological treatments.

Items	Treatment				SEM	P value
	Tr1	Tr2	Tr3	Tr4		
RT (ºC)	39.77^{a}	39.03^{b}	39.22^{b}	39.26^{b}	0.08	0.001
FT (ºC)	31.27^{a}	29.79^{b}	29.68^{b}	30.03^{b}	0.42	0.037
ET (ºC)	33.20^{a}	31.74^{b}	32.20^{ab}	32.02^{b}	0.38	0.049
ST (ºC)	35.13^{a}	33.96^{b}	34.05^{b}	34.31^{b}	0.22	0.001
RR (breathes/minute)	174.95^{a}	143.15^{b}	141.95^{b}	135.20^{b}	5.74	0.001

Tr1=control, Tr2=rabbit fed 10 ml ZAD®/kg diet), Tr3= rabbits fed 10 g ZADO®/kg diet and Tr4= rabbits fed 10 g YEAST®/kg diet. RT=rectal temperature, FT=fur temperature, ET=ear temperature, ST=skin temperature, RR=respiration rate

$^{a-c}$ *Means bearing different superscripts within the same row are significantly different ($P \leq 0.05$).*

Total protein and its fractions:

Supplementing biological additives (ZAD, ZADO and Yeast) increased (P≤0.05) the levels of total proteins and globulin as compared to the control in Tr1 (Table 3). However, no significant differences were observed in albumin and albumin/globulin ratio among treatments.

These results are in agreement with Abd El-Latif *et al.* (2008), who found that rabbits fed enzymes achieved high concentrations of serum total protein, albumin, and globulin. They explained that the elevation of these blood parameters may be due to the positive effect of enzymes on liver function and the digestibility of crude fiber and organic matter. Also, Makled *et al.* (2005) suggested that the increase in plasma total protein concentration by feeding the Optizyme-supplemented diets is due to an increase in crude protein digestibility which was associated with an improvement in growth performance of rabbits. On the other wise, increased total protein and globulin of doe rabbits received biological treatments may be an indication of the relatively good quality and availability of the dietary protein. Higher total protein and globulin values, being within the normal range of the rabbits received 1 % ZAD, ZADO and YEAST (10 g / kg diet) may indicate a proper and save level of the dietary protein used in this study.

Table (3): Mean ± SEM of total protein and its fractions in blood plasma of Hi-Plus doe rabbits fed on biological treatments.

Items	Treatment				SEM	P value
	Tr1	Tr2	Tr3	Tr4		
Total protein (g/dl)	5.34[b]	6.14[a]	6.42[a]	6.34[a]	0.16	0.001
Albumin (g/dl)	3.33	3.69	3.35	3.50	0.15	0.307
Globulin (g/dl)	2.01[c]	2.45[b]	3.07[a]	2.84[a]	0.19	0.002
Albumin/Globulin ratio	1.72	1.61	1.23	1.48	0.15	0.143

Tr1=control, Tr2=rabbit fed 10 ml ZAD®/kg diet), Tr3= rabbits fed 10 g ZADO®/kg diet and Tr4= rabbits fed 10 g YEAST®/kg diet. TAC=total antioxidant capacity

[a-c] Means bearing different superscripts within the same row are significantly different (P≤0.05).

The blood protein and globulin are the part of immunity components that albumin-based antibodies are the main protein component of serum which synthesized in the hepatic tissues, which acts the response of humoral immune and that could support the augmentation of immune organs (Abdel-Azeem *et al.*, 2018). Accordingly, in the present study, findings of globulin levels in serum were supported by Girotti *et al.* (2004), who recommended that supplementing xylanase to wheat-based cockerel diets from 7 to 21 d of age augmented the response of humoral immune. The functions of albumin include regulation of the distribution of extracellular fluid, and transport agent of many substances as bilirubin, fatty acids, hormones, and vitamins (Attia *et al.*, 2016). These results, in turn, affected the albumin/globulin ratio as in the other treatment groups it descended from 1.72 in the control to 1.61, 1.23 and 1.48 in biological treatments. The reduction in the albumin /globulin ratio may indicate an augment in the immunity of doe rabbits.

Blood metabolites parameters:

Mean value of plasma glucose concentration increased (P≤0.05) in the rabbits of Tr2, Tr3 and Tr4 by 23.6, 31.7, and 25.2 %, respectively, as compared to rabbits in Tr1. In an opposite trend, total cholesterol and triglycerides concentrations were decreased (P≤0.05) in the treated doe rabbits (Tr2, Tr3 and Tr4) compared with control does (Tr1). So, no significant differences were noticed in plasma glucose, cholesterol and triglycerides levels among treated groups (Table 4).

The present results showed a positive effect of ZAD, ZADO, and YEAST on decreasing the serum lipid profile in the rabbits, indicating that supplementation of biological or probiotic treatments could play a role in the lipid metabolism of rabbit. Our results showed significant decrease in total cholesterol and triglycerides levels. These results agreed with the results of Abdel-Azeem *et al.* (2018). By supplementing enzymes, it can inhibit the merger of 14C-labeled acetate to the non-saponifiable lipid fraction and therefore decrease the biosynthesis of lipid profiles and/or may have indirect inhibitory effects in the lipid biosynthesis enzyme hydroxymethyl-glutaryl coenzyme-A reductase levels (Fukushima and Nakano, 1995). This might indicate to the important of probiotic supplementation of rabbit to reduce the blood lipid profile and thus aiding to reduce total cholesterol deposition in the muscles and skin. This also means the possibility of incorporating the probiotic which will lead to products of an animal with the decreased cholesterol. The decline in rabbit blood cholesterol levels received probiotic supplementation is likely to indicate an overall decline in the mobilization of lipid. On the other hand, *S. cerevisiae* cell wall component, beta glucan, had a cholesterol lowering effect as documented by some researchers (Dhewantara, 2016; Sudiana *et al.*, 2022). In this respect, Paryad and Mahmoudi (2008) reported a reduction in plasma cholesterol and triglyceride levels of chicks fed on yeast supplement. These researchers suggested that yeast may regulate the blood cholesterol concentrations by de conjunction of bile acids. However, Seyidoğlu and Galip (2014) observed that *saccharomyces cerevisiae* (SC) did not affect the serum lipid, triglyceride, and cholesterol.

Table (4): Mean ± SEM of blood metabolites in Hi-Plus doe rabbits fed on biological treatments.

Items	Treatments				SEM	P value
	Tr1	Tr2	Tr3	Tr4		
Glucose (mg/dl)	108.96[b]	134.70[a]	143.56[a]	136.49[a]	8.86	0.050
Cholesterol (mg/dl)	161.72[a]	148.68[b]	146.69[b]	145.21[b]	3.67	0.022
Triglycerides (mg/dl)	127.93[a]	97.74[b]	84.38[b]	87.46[b]	6.52	0.001

Tr1=control, Tr2=rabbit fed 10 ml ZAD®/kg diet), Tr3= rabbits fed 10 g ZADO®/kg diet and Tr4= rabbits fed 10 g YEAST®/kg diet.
[a-b] Means bearing different superscripts within the same row are significantly different (P≤0.05).

Liver and kidney functions:

Mean values of plasma alanine aminotransferase (ALT) activity decreased (P≤0.05) in the doe rabbits of Tr2, Tr3, and Tr4 by 31.0 and 24.1 %, respectively, as compared to doe rabbits in Tr1. The results showed that ALT activity did not differ significantly (P>0.05) in Tr3 from that in control (Tr1). However, AST activity and concentration of creatinine and urea in plasma of rabbits were not affected by treatments (Table 5).

Table (5): Mean ± SEM of liver and kidney functions in plasma of Hi-Plus doe rabbits fed on biological treatments.

Items	Treatments				SEM	P value
	Tr1	Tr2	Tr3	Tr4		
ALT (IU/L)	18.50[a]	12.76[c]	16.30[ab]	14.03[bc]	0.92	0.003
AST (IU/L)	41.56	35.60	39.16	40.16	2.10	0.233
Creatinine (mg/dl)	0.96	0.97	1.05	1.02	0.04	0.370
Urea (mg/dl)	39.29	43.02	39.87	38.20	1.78	0.269

Tr1=control, Tr2=rabbit fed 10 ml ZAD®/kg diet), Tr3= rabbits fed 10 g ZADO®/kg diet and Tr4= rabbits fed 10 g YEAST®/kg diet. ALT=alanine aminotransferase, AST=aspartate aminotransferase
[a-c] Means bearing different superscripts within the same row are significantly different (P≤0.05).

Serum liver enzymes activities values (ALT and AST) in the present study are within the normal range of rabbits (Ajayi and Raji, 2012). Activity of serum ALT and AST are commonly utilized specifically to diagnose domestic animal hepatic damage and utilized to detect bile obstruction (i.e. mild and progressive liver damage) (McGill, 2016). That none of these blood metabolites changed between biological treatments received ZAD®, ZADO®, YEAST® and that they all fell within normal ranges for rabbits suggest that no liver damage had occurred. Rabbits in the biological treatments indicated no clinical signs of toxicity or signs of morbidity, indicating safe and healthy effects of these biological treatments.

Immunological parameters:

Plasma concentration of IgA increased (P≤0.05) in treated doe rabbits (Tr2, Tr3 and Tr4) by 32.1, 35.8, and 48.4%, respectively, compared with control group (Table 6). Indeed, IgG and IgM levels increased (P<0.05) in the doe rabbits of Tr4 by 17.8 and 34.5 %, respectively, compared with control doe rabbits. Meanwhile, IgM level increased (P≤0.05) in Tr4 compared to the does in Tr3. Feeding rabbits on YEAST expand mucosal immunity by increasing IgM and IgA levels against pathogens, enhances intestinal development and function, adsorbs mycotoxins, modulates gut microbiota, and reduces post-weaning diarrhea (and Azoz and Al-Kholy, 2006; Elghandour *et al.*, 2019). These results agreed with the results of Fathi *et al.* (2017), who reported that the inclusion of a probiotic in the feed of rabbits reared under hot environmental conditions greatly enhanced humoral and cell-mediated immune responses. On the other hand, no significant differences were found in IgA an IgG levels among treated groups (Tr2, Tr3 and Tr4).

Table (6): Mean ± SEM of immunological parameters in Hi-Plus doe rabbits fed on biological treatments.

Items	Treatments				SEM	P value
	Tr1	Tr2	Tr3	Tr4		
IgA (mg/dl)	13.55^{b}	17.91^{a}	18.41^{a}	20.11^{a}	0.95	0.001
IgG (mg/dl)	221.54^{b}	245.10^{ab}	238.67^{ab}	261.13^{a}	8.58	0.018
IgM (mg/dl)	14.23^{b}	17.37^{ab}	15.18^{b}	19.15^{a}	1.32	0.049

Tr1=control, Tr2=rabbit fed 10 ml ZAD®/kg diet), Tr3= rabbits fed 10 g ZADO®/kg diet and Tr4= rabbits fed 10 g YEAST®/kg diet.
$^{a-b}$ Means bearing different superscripts within the same row are significantly different (P≤0.05).

Total antioxidant capacity:

The results exhibited that total antioxidant level increased (P≤0.05) in the doe rabbits of Tr2 by 36.4%, Tr3 by 70.1%, and Tr4 by 58.2% as compared to the control doe rabbits in Tr1 (Figure 1).

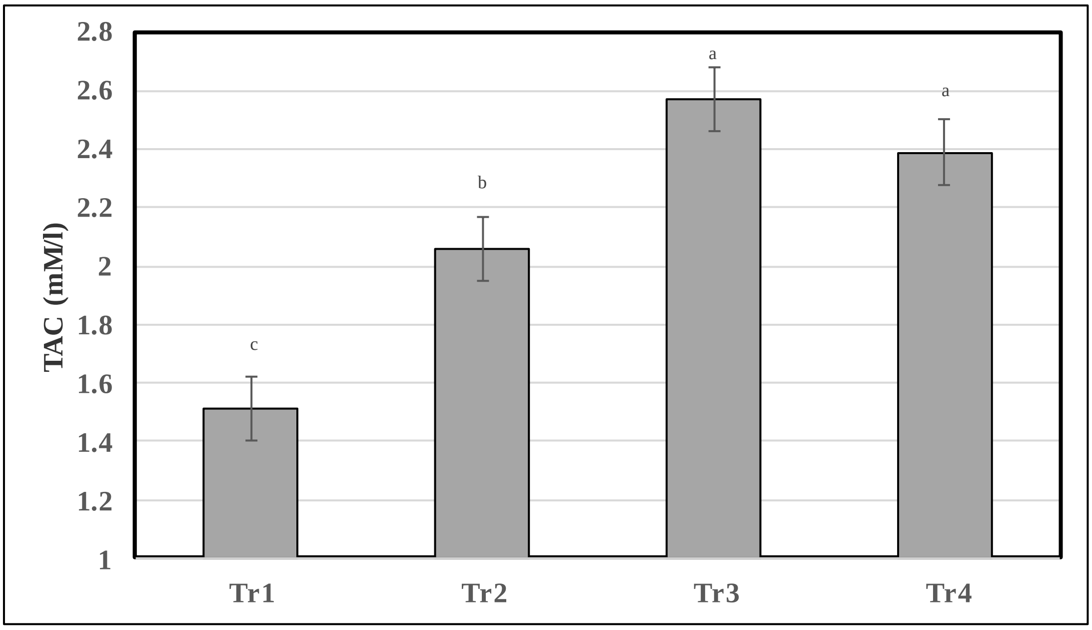

Figure 1. Total antioxidant capacity in plasma of Hi-Plus doe rabbits fed on biological treatments

Tr1=control, Tr2=rabbit fed 10 ml ZAD®/kg diet), Tr3= rabbits fed 10 g ZADO®/kg diet and Tr4= rabbits fed 10 g YEAST®/kg diet.

$^{a-c}$ Means bearing different superscripts are significantly different (P≤0.05).

The outcomes indicated that the changes in the activity of numerous antioxidant enzymes can be used in rabbits to evaluate the oxidative stress level and the total antioxidant status (Abdel-Azeem, 2019). An availability indicator of reduction agents in blood plasma is the total antioxidant capacity (TAC), and thus plasma's ability to scavenge free radicals of oxidation (Kambayashi *et al.*, 2009). Antioxidant enzymes can stop the oxidation either by scavenging the mainly reactive free radical *in vivo* or by steadying move metal radicals such as Cu^{+} or Fe^{2+} (Afolabi and Oloyede, 2014). Our results revealed that total antioxidant capacity in blood plasma of rabbits fed biological probiotics treatments (ZAD®, ZADO® and YEAST®) groups were significantly higher than the control, indicating improved scavenging capacity of the antioxidant defense system against oxidative stress processes in treatment groups.

These biological and probiotic treatments cause increases the number of beneficial bacteria (Abdel-Azeem *et al.*, 2018), which may be used the free radical and reactive oxygen species (ROS) during their metabolic processes and ⁄or indirectly because of the mixture of enzymes that positively improved growth performance, nutrient digestible coefficient and nitrogen balance as well as mortality rate (Gado *et al.*, 2017; Bhatt *et al.*, 2017; Abdel-Azeem *et al.*, 2018; Sherif, 2018), which might reflected on the improvement of the state of public health, increases the activity of cells to produce antioxidants and resistance to increase the production of free radicals. The ZAD® and ZADO® may be called probiotics to contain the mixture of anaerobic bacteria that create these enzymes. Probiotics antioxidant mechanisms may be attributed to ascorbate autoxidation reduction activity and inhibition, ROS scavenging, enzyme inhibition, and metal ion chelation (Abd El-Hamid, 2018). Probiotic metabolic operations may have an antioxidant impact by the scavenging or banning of oxidant compounds in the gut (Azcárate-Peril *et al.*, 2011). Moreover, Lin and Yen (1999) assumed that beneficial gut bacteria create several factors able to inhibit cytotoxic activity, remove free radicals, and catch ROS.

Hormonal profiles:

Thyroid hormone:

Mean concentration of plasma triiodothyronine hormone (T_3) was increased ($P \leq 0.05$) in the doe rabbits of Tr2, Tr3 and Tr4 by 33.3, 28.0 and 42.1%, respectively, as compared to doe rabbits in Tr1. Meanwhile, triiodothyronine hormone was increased ($P \leq 0.05$) in the doe rabbits of Tr4 by 10.9 % as compared to the rabbits in Tr3 (Figure 2).

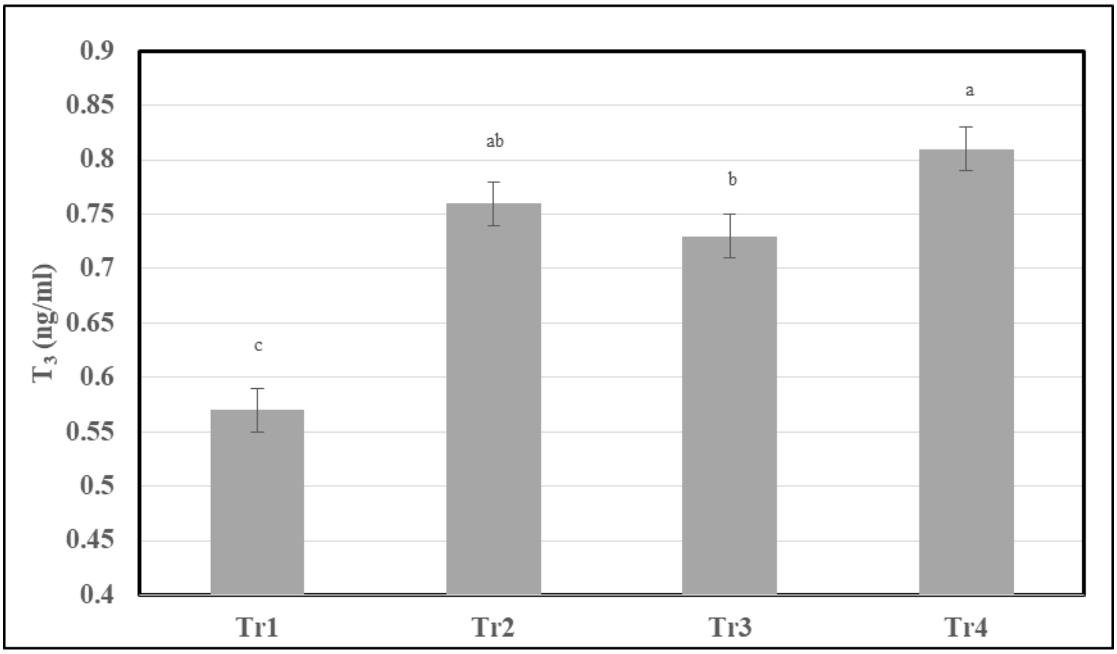

Figure (2): Concentration of plasma triiodothyronine hormone in Hi-Plus doe rabbits fed on biological treatments

Tr1=control, Tr2=rabbit fed 10 ml ZAD®/kg diet), Tr3= rabbits fed 10 g ZADO®/kg diet and Tr4= rabbits fed 10 g YEAST®/kg diet.

[a-c] Means bearing different superscripts are significantly different ($P \leq 0.05$).

Triiodothyronine hormone plays an important role in regulating metabolism and thermogenesis (Tao *et al.*, 2006). Concentration of T_3 hormone is highly correlated to feed intake and heat stress (Liang *et al.*, 2022). Exposure of rabbits to heat stress conditions causes a decrease in T_3 level and heat production, and sustain homoeothermic (Uni *et al.*, 2001; Attia *et al.*, 2016; Sakr *et al.*, 2019). The increase in thyroid hormones in ewes fed ration supplemented with probiotic mixture as compared to the control group indicated that these probiotics may increase protein level, and improve dry matter and other nutrient digestibility (Gado *et al.*, 2006). In this way, Abou-Zeina *et al.* (2015) found that T_3 and T_4 concentrations were higher in goats fed diets supplemented with high doses of exogenous enzyme than the control group. Abd El-Hamid *et al.* (2022) suggested that the enzymes directly or indirectly promote and enhance the activity of deiodinase in liver and kidney tissues, promoting the transformation of T_4 into the biologically active hormone (T_3).

Reproductive and productive performance:

The results of Table 7 reveled that CR (%), MY, LSB, LSW, LWB and LWW were increased (P≤0.05) in the doe rabbits of Tr2 (by 19.6, 26.9 ,32.4, 46.9, 19.8 and 40.3 %, respectively), Tr3 (by 25.6, 40.0 ,42.9, 53,9, 42.1 and 58.8 %, respectively) and Tr4 (by 28.6, 41.8 ,38.8, 53.2, 50.0 and 65.8 %, respectively) as compared to control group (Tr1).

Table (7): Mean ± SEM of reproductive and productive performance of Hi-Plus doe rabbits fed on biological treatments.

Items	Treatments				SEM	P value
	Tr1	Tr2	Tr3	Tr4		
CR (%)	62.18	74.41	78.10	79.97	4.85	0.052
NSC	1.66	1.50	1.37	1.33	0.10	0.116
GL (day)	31.80^{a}	30.73^{b}	30.81^{b}	30.60^{b}	0.19	0.002
LSB	5.33^{b}	7.06^{a}	7.62^{a}	7.40^{a}	0.50	0.007
LSW	4.26^{b}	6.26^{a}	6.56^{a}	6.53^{a}	0.44	0.001
LWB (g)	265.06^{b}	317.73^{b}	376.81^{a}	397.66^{a}	19.57	0.001
LWW (g)	2643.80^{c}	3709.66^{b}	4200.62^{ab}	4384.00^{a}	220.15	0.001
MRBW (%)	17.53	11.26	12.92	11.13	3.27	0.481
Stillbirth (%)	13.91	9.58	3.64	3.24	3.57	0.053
OMRBW (%)	31.44	20.84	16.56	14.37	5.22	0.057

Tr1=control, Tr2=rabbit fed 10 ml ZAD®/kg diet), Tr3= rabbits fed 10 g ZADO®/kg diet and Tr4= rabbits fed 10 g YEAST®/kg diet.

CR=conception rate, NSC=number of services per conception, GL=gestation length, LSB=litter size at birth, LSW=litter size at weaning, LWB=litter weight at birth, LWW=litter weight at weaning, MRBW=mortality rate from birth to weaning, OMRBW=overall mortality rate from birth to weaning.

$^{a-c}$ *Means bearing different superscripts within the same row are significantly different (P≤0.05).*

These results agree with the results of Gado *et al.* (2016), they reported that feeding enzyme increased conception rate with increased effect as the enzyme level was increased. Moreover, compared to the control rabbits, litter size and weight at birth and litter size and weight at weaning were improved with feeding enzyme additive, also, milk yield increased with the increasing level of enzyme additive compared with the control treatment. On the other hand, they noticed that the pre-weaning mortality was decreased with addition of enzyme additive compared with control. Improved reproductive and productive performance in the doe rabbits fed biological probiotic treatments may be related to the improved nutritional status of the mother does. Indeed, ZAD, ZADO and YEAST additives feeding decreased pre-weaning mortality.

Improved fertility and nutritional status of the doe rabbits were the results of the stimulatory effect of nutrients made available to the animals, as the improved nutrition enhances productive status of doe rabbits. Whereas numerous studies have been carried out with ruminants to investigate the potential benefits of feeding EZ (Rojo *et al.* 2015; Morsy *et al.* 2016), very few studies have been carried out on rabbits (Gado *et al.*, 2016). Indeed, the results of Ayyat *et al.* (1996) suggest that the inclusion of a probiotic (Lacto-Sacc) in diets of lactating rabbit does increased milk production, litter size and weight at weaning. However, biological probiotics treatment gained substantial interest in recent years in rabbit nutrition (Abdel-Aziz *et al.* 2015). Because the large intestine with cecum of the rabbit is a fermentation system similar to the rumen (De Blas and Wiseman 2010; Cunha and Cheeke 2012), some of our explanations would borrow from studies with ruminant animals. The direct results of such actions could have improved the nutrition status of rabbit does receiving the enzymes preparation. Improved feed utilization as a result of enzymes inclusion reported previously by Abo-Eid *et al.* (2016) is due to improved digestion rate of fiber fractions and altered fermentation kinetics as well as improved synergism between exogenous and endogenous enzymes (Wang *et al.* 2008). Feeding enzymes additive, in most cases, is paralleled with enhancing effect on microflora growth in gut and cecum and improving total and individual short chain fatty acids and nutrients (e.g., organic matter) digestibility.

In other studies, Maertens *et al.* (1994) and Ayyat *et al.* (1996) observed a significant improvement in litter size and weight at 21 d and at weaning due to Paciflor® and to Lacto-Sacc supplementation. However, supplementation of rabbit does' diet with Actisaf Sc 47 did not affect the kits' weight gain in the first 21 d of age and no marked changes were observed during the last week before weaning. These results showed firstly a possible lack of effect of Actisaf Sc 47 on milk production from birth to 21 d, when kits do not consume solid feed and their weight gain is related directly to the milk consumption, and secondly on solid consumption from 21 d until the weaning. However, Pinheiro *et al.* (2007) observed a higher weight gain in treatment group with Toyocerin® from 18 d of age to weaning in kits.

The decreased mortality of rabbits fed ZAD, ZADO and YEAST diets is another benefit of biological additive inclusion. Feeding rabbits on biological probiotics and balanced diets might help to control pre- and post-weaning mortality by limiting pathogen microbial populations (García- Ruiz *et al.* 2006) or by reduction of N flow at ileum, as observed by García *et al.* (2005). In an agreement with the present results, Eiben *et al.* (2004) obtained improved performance and decreased mortality of rabbits with the inclusion of cellulase in their diets.

Pinheiro *et al.* (2007) observed a decrease in mortality of kits from birth to 18 d of age. Moreover, Stamati *et al.* (2006), observed higher pre-weaning mortality among piglets in the control group than in the treated batch when applying 0.5 kg of Toyocerin/ton of feed.

No significant effects in NSC and MRBW (%) values among all groups (Tr1, Tr2, Tr3 and Tr4) were observed. However, values of GL (d), stillbirth (%) and overall MRBW (%) decreased ($P \leq 0.05$) in the treated doe rabbits (Tr2, Tr3 and Tr4) compared to control doe rabbits. No significant differences were observed in stillbirth (%) and overall MRBW (%) between the doe rabbits in Tr2 and Tr1 (Table 6).

Progesterone profile:

Plasma progesterone hormone (P_4) concentration increased ($P \leq 0.05$) in the treated does (Tr2, Tr3 and Tr4) as compared to control one (Figure 3). Progesterone hormone (P_4) has an essential role in physiology of reproduction in the mammalian species. Progesterone is a key hormone for maintenance of pregnancy, where it inhibits myometrial activity as well as it acts as regulator of hypothalamo-hypophyseal system (Ahmed, Nagwa *et al.*, 1994). These results may indicate that progesterone concentration increased significantly as litter size at birth increased in rabbits (Table 7). The high levels of progesterone at the 7th day of gestation which continued to 14th day plays an important role in the preparation of the uterus for implantation of the fertilized ovum and inhibition of uterine contractions during the first few days of pregnancy (Azoz, 1996).

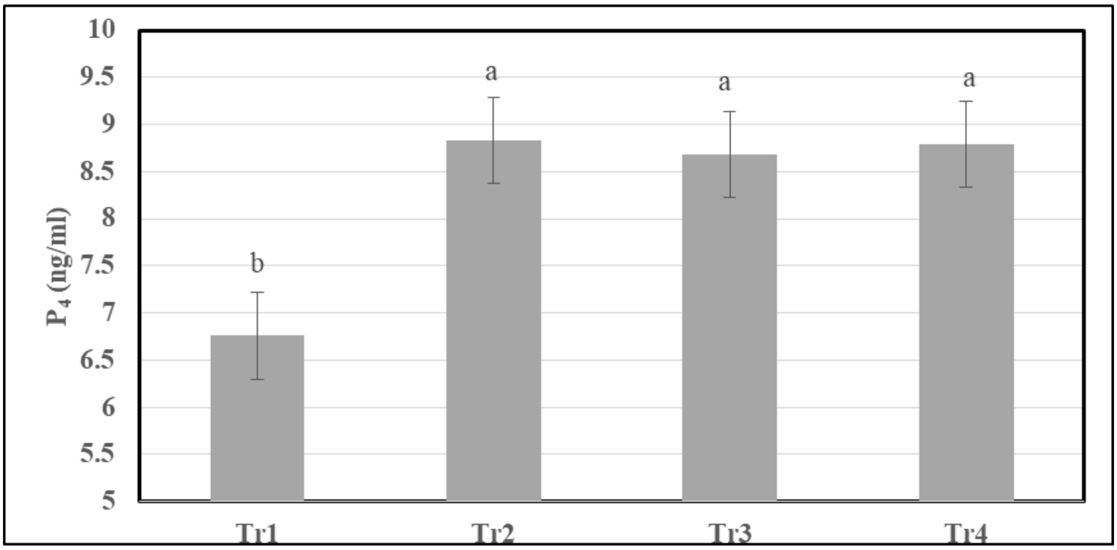

Figure (3): Plasma progesterone concentration in Hi-Plus doe rabbits fed on biological treatments

Tr1=control, Tr2=rabbit fed 10 ml ZAD®/kg diet), Tr3= rabbits fed 10 g ZADO®/kg diet and Tr4= rabbits fed 10 g YEAST®/kg diet.

[a-b] Means bearing different superscripts are significantly different ($P \leq 0.05$).

Milk yield:

The results showed that milk yield improved ($P \leq 0.05$) with biological treatments (ZAD, ZADO and YEAST) whereas treated doe rabbits of Tr2, Tr3 and Tr4 achieved higher values of milk yield by 26.9, 40.0, and 41.8%, respectively, as compared to control group (Tr1) (Figure 4). This increment in milk yield could be explained based on the increased intestinal metabolic activity, modified intestinal hindgut microbiota by the exclusive competition with intestinal pathogenic bacteria and modified structure and function of the intestinal epithelium causing stimulated immune system (Jeklova *et al.*, 2007). Another important factor affecting milk yield is the increased litter size. During the lactation, milk yield is higher when the litter size is larger (McNitt and Lukefahr 1990; Sakr *et al.*, 2019). Increased feed intake and utilization are important reasons for increased milk yield, as the milk produced by the doe is about 3 to 5% of the daily feed intake (Hassan *et al.*, 2013). Higher milk production and the nursing ability of the doe are key aspects for rearing young rabbits (Gado *et al.*, 2016). Increasing milk yield of biological supplemented groups might due to increased availability of nutrients and energy for lactogenesis. Probiotic additive feeding improved normal rabbit vital signs (e.g., rectal temperature, skin temperature, earlobe temperature, respiration rate, and pulse rate). The normal vital signs depend on recent activity, feed and water consumptions, and the physiological stage of the rabbits. The measured physiological traits indicate physiological ability of rabbit body thermoregulation with feeding enzyme additive. These results are due to the positive effects of enzyme on the respiratory rate and body metabolism (Gado *et al.*, 2016).

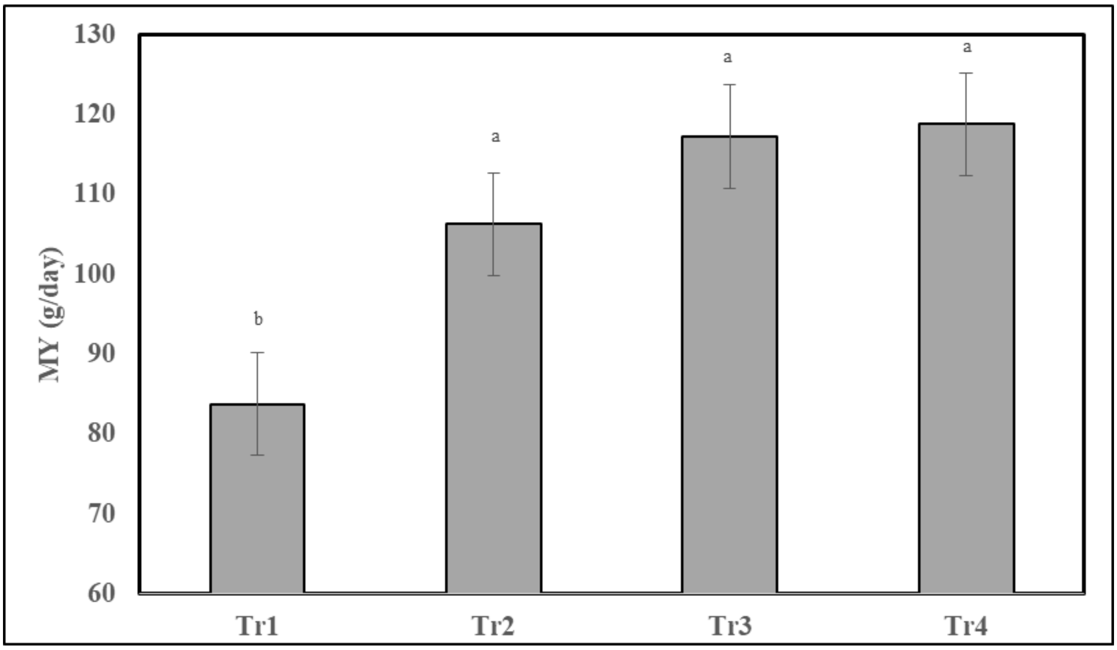

Figure (4): Milk yield in Hi-Plus doe rabbits fed on biological treatments

Tr1=control, Tr2=rabbit fed 10 ml ZAD®/kg diet), Tr3= rabbits fed 10 g ZADO®/kg diet and Tr4= rabbits fed 10 g YEAST®/kg diet.

[a-b] Means bearing different superscripts are significantly different (P≤0.05).

In addition, this result might be due to better health condition due to positive actions of live yeast in their gut intestinal tract (Bontempo *et al.*, 2006). This observation agrees with the shortened interval from parturition to effective mating of New-Zealand × Californian rabbit does with Toyocerin® observed by Nicodemus *et al.* (2004). On the other hand, it contrasted with the results of Kalmus *et al.* (2009), who reported no effect of yeast supplementation on resumption of ovarian activity in dairy cows. However, the addition of ZAD®, ZADO® and YEAST® increased the ability of lactating doe rabbits to alleviate heat stress as evident by increased milk production during the summer season (Gado, 2020).

Economic feed efficiency:

Concerning the total feed intake (TFI), The present results demonstrated that there were no significant differences among experimental groups (Table 8), where productive efficiency index (PEI) was higher ($P \leq 0.05$) in all treated doe rabbits (Tr4, Tr3 and Tr2, in order) compared with control ones.

Table (8): Mean ± SEM of economic indicator of Hi-Plus doe rabbits fed on biological treatments.

Items	Treatments				SEM	P value
	Tr1	Tr2	Tr3	Tr4		
TFI (kg)	24.71	25.04	23.26	23.12	0.42	0.133
PEI (kg, live weight)	5.28^{c}	7.41^{b}	8.40^{ab}	8.76^{a}	0.44	0.001
FC	4.68^{a}	3.38^{b}	2.77^{b}	2.64^{b}	0.39	0.001

Tr1=control, Tr2=rabbit fed 10 ml ZAD®/kg diet), Tr3= rabbits fed 10 g ZADO®/kg diet and Tr4= rabbits fed 10 g YEAST®/kg diet.
TFI=total feed intake (daily feed intake × 120 days (experimental period)). PEI=productive efficiency index (litter size at weaning× number of parities × total weaning weight).
FC=feed conversion (TFI / PI).
$^{a-c}$ Means bearing different superscripts within the same row are significantly different ($P \leq 0.05$).

Moreover, efficiency index was higher ($P \leq 0.05$) in the doe rabbits of Tr4 as compared to doe rabbits in Tr2.

Feed conversion value improved ($P \leq 0.05$) as a result of biological additives supplementation where treated rabbits Tr2, Tr3 and Tr4 exceeded control ones by 27.8, 40.8 and 43.5 %, respectively. These improvements in feed conversion of rabbits could be due to the greater nutrient digestibility of probiotics and biological supplemented rabbit diets. Ezema and Eze (2010) and Bhatt *et al.* (2017) indicted that dietary supplementation with probiotics had positive effect on feed conversion ratio of rabbits. Oso *et al.* (2013) and Amber *et al.* (2004) suggested that the improvement in feed efficiency of rabbits in response to feeding probiotic-supplemented diets could be explained by increasing the beneficial microflora in the gut and improving nutrient digestion and absorption. They stated that the improvements were due to probiotic modification in caecal microflora. Kritas *et al.* (2008) also reported higher gains and better feed conversion ratio in rabbits supplemented with probiotics than their control counterparts. Also, Bhatt *et al.* (2017) reported that rabbits given diets supplemented with probiotics had superior feed conversion ratio to the control group.

Supplementation of biological activities increased the economic efficiency where doe- rabbits of Tr2, Tr3 and Tr4 had higher economic efficiency compared with control rabbits. Cost of one kit production of Hi-Plus rabbits decreased by 8.40, 12.72 and 12.77 L.E. in Tr2, Tr3 and Tr4, respectively as compared to their counterparts of control group (Table 9). These results agree those of with Amber *et al.* (2004) and Onu and Oboke (2010), they reported that performance index and the net revenue were significantly increased by the probiotic or enzyme diets, as compared to control diet. Moreover, Abdel-Azeem *et al.* (2009) observed the best net return, percentage of economic efficiency; relative economic efficiency and performance index due to supplementing probiotic on rabbit diets. Meanwhile, El-Katcha *et al.* (2011) recorded that dietary supplementation of probiotic at 0.1 or 0.15 g/kg diet improve economic efficiency by about 64.9% and 49.7% in two different groups. Ogunsipe (2014) found that lowest cost of feed /weight gain for rabbits was recorded when rabbits were fed enzyme supplemented-based diet.

Table (9): Economic efficiency of Hi-Plus doe rabbits fed on biological treatments.

Items	Treatments			
	Tr1	Tr2	Tr3	Tr4
Total feed intake/doe	24.71	25.04	23.26	23.12
Cost of kg feed (L.E.)	8	8	8	8
Cost of kg biological additive (L.E.)	0	1.5	1.5	1.5
Total feeding cost (L.E.)/doe	197.68	237.88	220.97	219.64
Litter size at weaning/doe	4.26	6.26	6.56	6.53
Cost (L.E.) of one kit production	46.40	38.00	33.68	33.63

Tr1=control, Tr2=rabbit fed 10 ml ZAD®/kg diet), Tr3= rabbits fed 10 g ZADO®/kg diet and Tr4= rabbits fed 10 g YEAST®/kg diet.

CONCLUSION

It could be concluded that under heat stress conditions, supplementation of biological probiotics treatments (ZAD®, ZADO® and YEAST®) at a level of 1% to the doe Hi-Plus rabbit's diets can enhance the reproductive and productive performances, increased milk yield, reduced pre-weaning mortality, and improve blood constituents and immunological status. The addition of 10 g YEAST /kg diet showed the best impacts as compared to other treatments (10 g ZAD and 10 g ZADO/kg diet).

ACKNOWLEDGEMENTS

The author is thankful to Dr. Hassan El-Shaer for facilitating this research work through the project "Utilization of Marine Algae for Salt Fodders, Milk, Meat and Fish production under saline conditions" which was funded by SYSTEL Telecom company, Egypt. Also, the authors gratefully acknowledge their appreciation to Dr. Ismail Mashhour, Chairman of SYSTEL Telecom, for his support.

REFERENCES

Abd El-Hamid, I. S., Khalifa, M.A., Shedeed, H.A., Rateb, S.A. (2018). Cryosurvival of Ram Spermatozoa after Supplementing the Diluent with L-Ascorbic Acid or α-Tocopherol. International Journal of Veterinary Science, 7(4): 197-204.

Abd El-Hamid, I.S., Fouda, W.A., Shedeed, H.A., Moustafa, S.A., Elbaz, A.M., Bakr, S.A., Mosa, B.H., Morsy, A.S., Hasan, A.M., Emam, K.R. (2022). Influence of Microalgae Nannochloropsis oculata on Blood Constituents, Reproductive Performance and Productivity in Hi-Plus Doe Rabbits Under North Sinai Conditions in Egypt. J. Anim. Health Prod. 10(2): 135-145 http://dx.doi.org/10.17582/journal.jahp/2022/10.2.135.145.

Abd El-Latif, S. A., Mohammed, K. A., Kawsar, A., Ghaly and Maha, A., Abd El-Latif (2008). Effect of using commercial enzymes on performance and some metabolic functions of rabbits fed grade levels of crude fiber. Egypt. Poult. Sci., 28: 1003-1022.

Abdel-Azeem, A.S.; A.A. Hassan; M.M. Basyony and S.H. Abu Hafsa (2018). Rabbit growth, carcass characteristic, digestion, caecal fermentation, microflora, and some blood biochemical components affected by oral administration of anaerobic probiotic (ZAD®). Egyptian Journal of Nutrition and Feeds, 21 (3): 693 - 710.

Abdel-Azeem, F.; N.A. Hashem; Y.K.E.H. Badawi and A. Farid (2009). Comparative study between probiotic (Bioplus 2B) and antibiotic (Lincofeed) on the performance of growing rabbits. Egyptian Journal of Rabbit Science, 19 (1): 7 - 22.

Abdel-Aziz, N.A.; M. El-Adawy; M.A. Mariezcurrena-Berasain A.Z. Salem; J. Olivares-Pérez; A.E. Kholif and B.E. Borhami (2015). Effects of exogenous enzymes, *Lactobacillus acidophilus* or their combination on feed performance response and carcass characteristics of rabbits fed sugarcane bagasse. Journal of Integrative Agriculture, 14 (3): 544 - 549.

Abo-Eid, H. A., Abousekken, M. S., El-Folly, I. A. M. (2016). Rabbit growth perfomance as affected by dietary levels of date waste meal. Egyptian Journal of Nutrition and Feeds, 19(2), 349-362.

Abou-Zeina, H. A., Nasr, S. M., Abdel-Aziem, S. H., Nassar, S. A., Mohamed, A. M. (2015). Effect of different dietary supplementation with antioxidants on gene expression and blood antioxidant markers as well as thyroid hormones status in goat kids. Middle-East Journal of Scientific Research, 23(6), 993-1004.

Abraham, G.E. (1974). Ovarian and adrenal contribution to peripheral androgens during the menstrual cycle. J. Clin. Endocrinol. Metab., (39): 340-346. https://doi.org/10.1210/jcem-39-2-340.

Afolabi, A.B. and O.I. Oloyede (2014). Antioxidant properties of the extracts of Talinum triangulare and its effect on antioxidant enzymes in tissue homogenate of Swiss albino rat. Toxicology international, 21 (3): 307 - 313.

Ahmed, Nagwa A. (2000). Diurnal and seasonal variations in thermoregulation and physiological reactions of pregnant and non-pregnant rabbits. J. Agric. Sci. Mansoura Univ., 25 (6): 3221 – 3233.

Ahmed, Nagwa, A.; Amina, F.K.; Shafie, M.M. and Azoz, A.A. (1994). Effect of post-partum mating interval of rabbits on progesterone level through pregnancy. Egypt. J. Anim. Prod., 31 (suppl.): 463 – 473.

Ahmed, Nagwa A.; Elfar, A. A. and Sakr, O. G. (2005). Evaluation of sexual and maternal behaviour, hormonal pattern and reproductive performance of doe rabbits as affected by seasonal variation. The 4th Inter. Con. on Rabbit Prod., In Hot Clim., Sharm El-Sheikh, Egypt, 225-231.

Ajayi, A.F. and Y. Raji (2012). Hematological and serum biochemical indices of pre-pubertal male rabbits fed with graded level of blood-wild sunflower forage meal mixture. African Journal of biotechnology, 11 (35): 8730 - 8734.

Amber, K.H.; H.M. Yakout and S. Hamed Rawya (2004). Effect of feeding diets containing yucca extract or probiotic on growth, digestibility, nitrogen balance and caecal microbial activity of growing new zealand white rabbits. In Proc.: 8th World Rabbit Congress, Puebla, México, pp. 737 - 741.

Amer, F., Azoz, A. B., Deraz, T. (2013). Productive performance of growing rabbits fed diets containing fenugreek straw and supplemented with enzymes. Egyptian Journal of Rabbit Science, 23(1), 37-53.

Attia, Y.A.; F. Bovera; W.S. El-Tahawy; A.M. El-Hanoun; M.A. Al-Harthi and H.I. Habiba (2016). Productive and reproductive performance of rabbits does as affected by bee pollen and/or propolis, inulin and/or mannan-oligosaccharides. World Rabbit Science, 23 (4): 273 - 282.

Ayyat, M.S.; I.F.M. Marai and T.A. El-Aasar (1996). New Zealand White rabbit does and their growing offsprings as affected by diets containing different protein level with or without Lacto-Sacc supplementation. World Rabbit Science, 4 (4): 225 - 230.

Azcárate-Peril, M.A., Sikes, M., Bruno-Bárcena, J.M. (2011). The intestinal microbiota, gastrointestinal environment and colorectal cancer: a putative role for probiotics in prevention of colorectal cancer? Am J Physiol Gastrointest Liver Physiol 301: G401-424. doi: 10.1152/ajpgi.00110.2011/

Azoz A.A. (1996). Effect of different mating systems on productive and reproductive efficiency of rabbits. M. Sc. Thesis, Fac. Agric., Cairo Univ., Egypt.

Belhassen, T.; A. Bonai; Z.S. Gerencser; Z.S. Matics; T. Tuboly; R. Bergaoui and M. Kovacs (2016). Effect of diet supplementation with live yeast *Saccharomyces cerevisiae* on growth performance, caecal ecosystem and health of growing rabbits. World Rabbit Science, 24 (3): 191 - 200.

Bhatt, R.S.; A.R. Agrawal and A. Sahoo (2017). Effect of probiotic supplementation on growth performance, nutrient utilization and carcass characteristics of growing Chinchilla rabbits. Journal of Applied Animal Research, 45 (1): 304 - 309.

Bligh, J. and Johnson, K. G. (1973). Glossary of terms for thermal physiology. J. Appl. Physiol., 35: 941-960.

Bontempo, V., Giancamillo A.D., Savoini G., Dell'Orto V., Domeneghini C. (2006). Live yeast dietary supplementation acts upon intestinal morpho-fuctional aspects and growth in weanling piglets. Anim. Feed Sci., 129: 224-236. doi:10.1016/j.anifeedsci.2005.12.015.

Cunha, T.J. and Cheeke, P.R. (2012). Rabbit Feeding and Nutrition, (Elsevier).

De Blas, C. and Wiseman, J. (2010). Nutrition of the Rabbit. CABI.

Dhewantara, F. X. (2016). Cholesterol-lowering effect of beta glucan extracted from Saccharomyces cerevisiae in rats. Scientia pharmaceutica, 84(1), 153-165.

Duncan, D.B. (1955). Multiple Range and Multiple F-Test. Biometrics, 11 (1): 1 - 5.

Eiben, C.S., Mézes, M. Zijártó, N. Kustos, K., Gódor-Surmann, K., Erdélyi, M. (2004). Dose-dependent effect of cellulase supplementation on performance of early-weaned rabbit. In Proceeding: 8th World Rabbit Congress, Puebla, México, pp. 799 - 804.

El-Katcha, M.I.Y., Ismail, E.Y., Soltan, M.A. and El-Naggar, M.K (2011). Effect of dietary probiotics supplementation on growth performance, immune response, some blood parameters and carcass quality of growing rabbits. Alexandria Journal of Veterinary Sciences, 34 (1): 153 - 169.

Ezema, C., Eze, D.C. (2012). Determination of the effect of probiotic (Saccharomyces cerevisiae) on growth performance and haematological parameters of rabbits. Comparative Clinical Pathology, 21 (1): 73 - 76.

Fukushima, M., Nakano, M. (1995). The effect of probiotic on faecal and liver lipid classes in rats. British journal of nutrition, 73 (5): 701 - 710.

Gado, H. (2020). Utilization of anaerobic microbiology to improve animal production. Egyptian Journal of Animal Production, 57 (Suppl. Issue): 81 - 86.

Gado, H. M.; A.Z.M. Salem; N.E. Odongo and B.E. Borhami (2011). Influence of exogenous enzymes ensiled with orange pulp on digestion and growth performance in lambs. Animal Feed Science and Technology, 165 (1-2): 131 - 136.

Gado, H.; M. Mellado; A.Z.M. Salem; A. Zaragoza and T.S.T Seleem (2015). Semen characteristics, sexual hormones and libido of Hy-plus rabbit bucks influenced by a dietary multi enzyme additive. World Rabbit Science, 23 (2): 111 - 120.

Gado, H.M. and A.Z.M. Salem (2013). Anaerobic Enzymes as a New Technology in Animal Feed. In: Salem, A. Z. M. (Editor) Nutritional Strategies of Animal Feed Additives. Nova Science Publishers, Inc. New York, 2013 (1): 1 - 24.

Gado, H.M.; A. Khusro and A.Z.M. Salem (2017). Role of probiotics in animal nutrition: A review. Animal review, 4 (1): 8 - 20.

Gado, H.M.; A.E. Kholif; A.Z.M. Salem and M.M.M. Elghandour (2016). Fertility, mortality, milk output, and body thermoregulation of growing Hy-Plus rabbits fed on diets supplemented with multi-enzymes preparation. Tropical Animal health and production, 48 (7): 1375 - 1380.

Gado, H.M.; S.A. Nasr, K.B. Mobamed and A.A. Mahrous (2006). Effect of biological treatments on the nutritive value of rice straw. Egyptian J. Nutrition and Feeds, 9 (2): 207 - 219.

García, A.I., García, J., Corrent, E., Chamorro, S., García-Rebollar, P., De Blas, J.C., Carabaño, R. (2005). Effet de l'âge du lapin, de la source de protéineet de l´utilisation d´enzymessur les digestibilities apparentes de la matièresèche et de la protein brute sur un aliment lapin. In Proc.: 11èmes Journées de la Recherche Cunicole, Paris, France, 197–200. PMCid: PMC1201644.

García-Ruiz, A.I., García-Palomares, J., García-Rebollar, P., Chamorro, S., Caraba-O, R. and De Blas, J.C. (2006). Effect of protein source and enzyme supplementation on ileal protein digestibility and fattening performance in rabbits, Spanish Journal of Agricultural Research, 4, 297–303.

Girotti, S., Ferri, E., Fini, F., Bolelli, L., Sabatini, A.G., Budini, R., Sichertova, D. (2004). Automated and manual luminescent assay of antioxidant capacity: analytical features by comparison. Talanta 64:665-670. doi10.1016/j.talanta.2004.03.041.

Habeeb A.A.; Marai, I.F.M. and Kamal, T. H. (1992). Heat stress in farm animals and environment. Edited by C. Phillips and D. Piggin, pp 27 – 47. C. A. B. International U. K.

Hassan, M. A., Shaltout, F. A., Arfa, M. M., Mansour, A. H., & Saudi, K. R. (2013). Biochemical studies on rabbit meat related to some diseases. Benha Vet. Med. J, 25(1), 88-93.

Henry, R.J. (1964). *Clinical Chemistry: Principles and Techniques*. Harper and Row Publishers, NewYork.109 p.

Jeklova, E., Leva, L., Kudlackova, H., & Faldyna, M. (2007). Functional development of immune response in rabbits. Veterinary immunology and immunopathology, 118(3-4), 221-228.

Kalmus, P., Orro T., Waldmann, A., Lindjärv, R., Kask, K. (2009). Effect of yeast culture on milk production and metabolic and reproductive performance of early lactation dairy cows. Acta Vet. Scand., 51: 1-7. doi:10.1186/1751-0147-51-32.

Kambayashi, Y.; N.T. Binh; H.W. Asakura; Y. Hibino; Y. Hitomi; H. Nakamura and K. Ogino (2009). Efficient assay for total antioxidant capacity in human plasma using a 96-well microplate. Journal of clinical biochemistry and nutrition, 44 (1): 46 - 51.

Kritas, S.K., Petridou, E.I., Fortomaris, P., Tzika, E., Arsenos, G., Koptopoulos, G. (2008). The effect of probiotics on microbiology, health and performance of fattening rabbits. Asian Austral. J. Anim., 21: 1312-1317. doi:10.5713/ajas.2008.70186.

Liang, Z. L., Chen, F., Park, S., Balasubramanian, B., & Liu, W. C. (2022). Impacts of heat stress on rabbit immune function, endocrine, blood biochemical changes, antioxidant capacity and production performance, and the potential mitigation strategies of nutritional intervention. Frontiers in Veterinary Science, 9, 906084

Lin, M.Y., Yen, C.L. (1999). Antioxidative ability of lactic acid bacteria. J. Agric. Food Chem. 1999, 47, 1460–1466.

Maertens, L. (1992). Influence of live yeast (BIOSAF) on rabbit performance. Cuniculture (Paris), (1992) 104: 97-98.

Makled, M.N.; M.A. Metwally; I.A. Soliman and H.A. Younis (2005). Impact of different levels of dietary multi-enzymes supplement on California rabbit performance. 3rd International Poultry Conference, Hurghada, Egypt, pp. 4 - 7.

Marai, I.F.M., Habeeb, A.A.M., Gad, A.E. (2002). Reproductive traits of male rabbits as affected by climatic conditions, in the subtropical environment of Egypt. Anim. Sci. J., (75): 451-458. DOI: https://doi.org/10.1017/S1357729800054394.

McGill, M. R. (2016). The past and present of serum aminotransferases and the future of liver injury biomarkers. Excli journal, 15 (15): 817 - 828.

McNitt, J. I., Lukefahr, S. D. (1990). Effects of breed, parity, day of lactation and number of kits on milk production of rabbits. Journal of Animal Science, 68(6), 1505-1512.

Morsy, T.A., Kholif, A.E., Kholif, S.M., Kholif, A.M., Sun, X. and Salem, A.Z.M. (2016). Effects of two enzyme feed additives on digestion and milk production in lactating Egyptian buffaloes, Annals of Animal Science, 16 (1), 209–222.

NRC (1994). Nutrient Requirements of Poultry. National Research Council, National Academy Press, Washington, D. C., 9th Revised Edition. pp. 234.

Ogunsipe, M. H. (2014). Effect of poultry litter with or without enzyme supplementation on the growth performance, nutrient digestibility and economy of rabbit production. International Journal of Livestock Production, Vol 5(2):23-29.

Onu, P.N. and S.A. Oboke (2010). Evaluation of enzyme and probiotic supplementation of maize processing waste-based diets on performance and nutrient utilization of weaner rabbits. Indian Journal of Animal Research, 44 (2): 87 - 93.

Oso, A. O.; O.M.O. Idowu; A.S. Haastrup; A.J. Ajibade; K.O. Olowonefa; A.O. Aluko, I.M. Ogunade, S.O. Osho and A.M. Bamgbose. (2013). Growth performance, apparent nutrient digestibility, caecal fermentation, ileal morphology and caecal microflora of growing rabbits fed diets containing probiotics and prebiotics. Livestock Science, 157 (1): 184 - 190.

Paryad, A., Mahmoudi, M. (2008). Effect of different levels of supplemental yeast (Saccharomyces cerevisiae) on performance, blood constituents and carcass characteristics of broiler chicks. African Journal of Agricultural Research, 3(12), 835-842.

Pinheiro, V., Mourão, J.L., Jimenez, G. (2007). Influence of Toyocerin® (Bacillus cereus var. toyoi) on breeding performance of primiparous rabbit does. World Rabbit Sci., 15: 179 -188. doi:10.4995/wrs.2007.590.

Rojo, R.., Kholif, A.E., Salem, A.Z.M., Elghandour, M.M.Y., Odongo, N.E., DE OCA, R.M., Rivero, N and Alonso, M.U. (2015). Influence of cellulase addition to dairy goat diets on digestion and fermentation, milk production and fatty acid content, The Journal of Agricultural Science, 153(08), 1514–1523.

Sakr, O.G., Mousa, B.H., Emam, K.R.S., Morsy, A.S., Nagwa, A. A. (2019). Effect of Early Heat Shock Exposure on Physiological Responses and Reproduction of Rabbits under Hot Desert Conditions. World's Vet. J., 9(2): 90-101. https://dx.doi. org/10.36380/scil.2019.wvj13.

SAS (2004). User's Guide: Statics. Release 9.1. SAS Institute Inc., Cary, NC. 536 p.

Seyidoğlu, N., Galip, N. (2014). Effects of Saccharomyces cerevisiae and Spirulina platensis on growth performances and biochemical parameters in rabbits. Kafkas Üniversitesi Veteriner Fakültesi Dergisi, 20(3).

Sherif, S.K. (2018). Effect of dietary additives on growth performance, carcass traits and some blood constituents of rabbits. Journal of Agricultural Science, 10 (1): 139-151.

Stamati, S., Alexopoulos, C., Siochu, A., Saoulidis, K., Kyriakis, S.C. (2006). Probiosis in sows by administration of bacillus toyoi spores during late pregnancy and lactation: effect on their health status/performance and on litter characteristics. International Journal of Probiotics and Prebiotics, 1: 33-40.

Sudiana, A. D. G., Kuswendi, H., Dewi, V. Y. K., & Balia, R. L. (2022). The potential of ß-glucan from Saccharomyces cerevisiae cell wall as anti-cholesterol. J Anim Health Prod, 9(1), 72-7.

Tao, X., Zhang, Z.Y., Dong, H., Zhang, H., Xin, H. (2006). Responses of thyroid hormones of market-size broilers to thermoneutral constant and warm cyclic temperatures Poult. Sci., (85): 1520-1528. https://doi.org/ 10.1093/ps/85.9.1520.

Uni, Z., Gal-Garger, O., Geyra, A., Sklan, D., Yahav, S. (2001). Changes in growth and function of chick small intestine epithelium duo to early thermal conditioning. Poult. Sci., (80): 438–445. https://doi.org/10.1093/ps/80.4.438.

Wang, Z.; Z. Gu,; Y. Huo; B. Chen; Y. Liu; Z. Zhao and G. Zhang (2008). Effects of probiotics and nanometre implement on growth performance of Rex Rabbit. In: *Proceedings of 9th World Rabbit Congress* held at Verona Italy, pp. 837 - 840.

Wheeler, M.H., Lazarus, J.H. (1994). Diseases of the Thyroid. London, Glasgo Weinheim, New York, Tokyo, Melbourme Madras: Chap. Hall. Medical., 107-115. https://doi.org/10.1016/S0140-6736(96)08015-4.

الكفاءة التناسلية، أيض الدم، والإنتاجية لأمهات أرانب الهاي بلس المغذاة على إضافات حيوية مختلفة تحت ظروف شمال سيناء في مصر

أحمد صبحى على الحاوى

قسم فسيولوجيا الحيوان والدواجن، مركز بحوث الصحراء، مصر.

يهدف هذا البحث إلى دراسة تأثير مكملات البروبيوتيك الزاد والزادو والخميرة على صفات الدم والاستجابات المناعية والأداء التناسلى والإنتاجي لأرانب الهاى بلس تحت ظروف شمال سيناء. تم استخدام 60 أرنب من نوع الهاى بلس عمر 5 أشهر ووزن الجسم 3190.2 ± 30.7 جم، في هذه الدراسة في الفترة من مايو إلى سبتمبر 2022. وتم تقسيمها عشوائيًا إلى أربع معاملات متساوية. المعاملة الأولى كانت كنترول بدون إضافات. المعاملة الثانية تم تقديم 10 جرام من الزاد / كجم من العليقة الأساسية (1.0%) وفى المعاملة الثالثة تم تقديم 10 جرام من الزادو / كجم من العليقة الأساسية (1.0%) وفى المعاملة الرابعة تم تقديم 10 جرام من الخميرة / كجم من العليقة الأساسية (1.0%) خلال الفترة التجريبية. أظهرت النتائج المتحصل عليها أن تطبيق الإضافات البيولوجية أدى إلى زيادة معنوية في مستويات الجلوكوز والبروتين الكلي والجلوبيولين وهرمون الثيرونين ثلاثى اليود والجلوبيولين المناعي (IgA) ومستويات مضادات الأكسدة الكلية، في حين انخفض معنوياً الكوليسترول الكلي والدهون الثلاثية في بلازما دم الأرانب المعاملة. ارتفع هرمون البروجسترون بشكل معنوي في المجموعات المعاملة مقارنة بأرانب المجموعة الكنترول. زادت مستويات الجلوبولين المناعي IgG وIgM معنوياً في أرانب المجموعة الرابعة مقارنة بأرانب المجموعة الكنترول. ارتفع معنوياً معدل الحمل وإنتاج اللبن وعدد ووزن الخلفات عند الميلاد والفطام وكذلك مؤشر الكفاءة الإنتاجية في أرانب الأرانب المعاملة (المعاملة الثانية والثالثة والرابعة) مقارنة بأرانب المجموعة الكنترول. تحسنت قيمة التحويل الغذائي معنوياً في الأرانب المعاملة مقارنة بالمجموعة الكنترول. انخفض معنوياً نشاط انزيمات الألانين الناقلة لمجموعة الأمين فى البلازما فقط في أرانب المعاملات الثانية والرابعة مقارنة بأرانب المعاملة الأولى (الكنترول).

تخلص الدراسة إلى أنه في ظروف الإجهاد الحراري، فإن المكملات الغذائية من البروبيوتيك البيولوجي (الزاد، الزادو، الخميرة) بمستوى 1٪ يمكن أن تعزز الأداء التناسلي والإنتاجي، وتزيد من إنتاج اللبن وتقلل من معدل النفوق قبل الفطام، وتحسن من مكونات الدم والحالة المناعية لأرانب الهاى بلس. أظهرت مكملات 10 جرام من الخميرة / كجم من العليقة الأساسية أفضل التأثيرات مقارنة بالمكملات الأخرى.