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EFFECT OF SPIRULINA AND PREBIOTIC (INMUNAIR 17.5®) ON NEW-ZEALAND WHITE RABBITS PERFORMANCE

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ABSTRACT: Twenty-four weanling New Zealand white rabbits (NWZ) at five weeks of age and nearly similar average body weight (682-693 g) randomly divided into four treatment groups (6 rabbits per group), each group was subdivided into three replicates (two rabbits per replicate). The 1st group fed the basal diet and given tap water without supplements as a control group, the 2nd group was fed the same basal diet and given tap water supplemented with Prebiotic (Inmunair 17.5®) at level (1 ml/ liter drinking water) three days in beginning of the experiment, the 3rd and 4th groups were given tap water and fed diet supplemented with Spirulina at levels of 0.1 and 0.2% diet, respectively at all the period of experiment (8 weeks). Growth performance (live body weight, feed intake, daily weight gain and feed conversion ratio) and carcass traits were studied. Results obtained showed that, significant ($P<0.01$) promoting effect on growth performance at 13 weeks of age of rabbits received Prebiotic (Inmunair 17.5) at 1 ml/ liter in drinking water for three days. The average of feed intake and feed conversion ratio of rabbits supplemented with Spirulina at levels 0.1 and 0.2% in the diet were improved significantly ($p<0.05$ or $p<0.01$), respectively when compared with control. The studied carcass traits (Fore part, Hind part, liver, head, heart, lung, kidney and dressing) except spleen and lion were significantly higher in growing rabbits received drinking water supplemented with Inmunair 17.5 and rabbits fed on diet contained 0.1 and 0.2% spirulina compared with control group.

Key words: Rabbits, growth, performance, Inmunair 17.5®, prebiotics, spirulina.

INTRODUCTION

Feed management is a cornerstone in ensuring pet rabbits welfare, and this is supported by the fact that the main reason pet rabbit owners consult veterinarians is because of health problems caused by incorrect nutrition **(Harrenstien, 1999).**

Recently, manufactures produced microorganisms commercially as growth promotors to substitute antibiotics in animal feeds to avoid its harmful effect on human health. The addition of prebiotic to the rabbit diets has been found to improve growth performance and feed conversion ratio in growing and breeding rabbits **(El-Hindawy *et al.,* 1993; Abdel-Azeem *et al*., 2009)**.

A prebiotic substance has been defined as a non-digestible food ingredient that beneficially affects the host by selectively stimulating the growth and/or activity of limited number of bacteria in the colon (**Choudhari *et al*, 2008)**. Several studies have shown that addition of prebiotic to the ration of broilers, layers and pigs improved performance through improving gut micro flora **(Xu *et al.,* 2003)**. Using of compounds that may have prebiotic effects is a possible way to improve intestinal health and animal performance in the absence of antibiotic growth promoters **(Kim *et al*., 2011)**. By adding Prebiotics to poultry diets, Zoo technical performance body weight and feed conversion ratio were significantly improved. Moreover, Prebiotics improve immune system to reduce

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colonization by pathogens **(El-Habback *et al.*, 2015)**.

Inmunair17.5® (Propionibacterium acnes, and coli lipopolysaccharides) is one of the commercial products available in the Egyptian market as nonspecific immune-stimulant for the chicken farms. Propionibacterium acnes are a Gram positive, non-spore forming opportunistic bacteria (**Perry and Lambert, 2006)**. Propionibacterium acnes are an effective activator of macrophage, lymphocyte, natural killer cells and cytokine release in the examined lab animals (**Tizard, 2009).**

Lipopolysaccharides are the main components of the outer membrane of Gram-negative bacteria, induce strong immune responses Lipopolysaccharides of *Escherichia coli* (LPS)had a role in releasing IL-1, IL-6, or tumor necrosis factor (TNF) by macrophages (**Silhavy *et al.*, 2010).**

Spirulina (Arthrospira) is microscopic blue green algae, and it is considered one of the richest sources of organic nutrients which make it a good nutritional supplement in human and animal feed worldwide. Spirulina could potentially be used in poultry and animal nutrition to improve the productivity and quality of obtained meat **(Peiretti and Meineri, 2008).** It can act as a good growth and immune-modulator **(Jamil *et al*., 2015)** Spirulina has been trialed in the feed rations of commercially farmed meat rabbits. Using of Spirulina as a feed additive in rabbit diets not affect rabbit growth (**Peiretti and Meineri, 2008),** or carcass yields **(Peiretti and Meineri, 2011).**

Therefore, the aim of the present study was to investigate the effect of adding Inmunair 17.5 as prebiotic in drinking water and Spirulina platensis in the diet on growth performance and carcass traits of growing NZW rabbits.

MATERIALS AND METHODS

The present work was carried out at department of Animal production, Faculty of Agriculture, Zigzag University, Egypt. The practical part was carried out at Middle East for Veterinary vaccines (ME-VAC) El-Salhiya El-Gadida city, Sharkia Governorate, Egypt.

Twenty-four weanling New Zealand white rabbits (NWZ) rabbits at five weeks of age and nearly similar average body weight (682-693g) were randomly divided into four treatment groups (6 rabbits per group). Each group was subdivided into three replicates (Two rabbits per replicate). The 1st group fed the basal diet and given tap water without any supplement as a control group. The 2nd groups was fed the same diet and given tap water supplemented with Inmunair 17.5 ®at level (1ml/1liter of drinking water) three days in beginning of the experiment. The 3rd and 4th group were given tap water and fed diet supplemented with Spirulina at levels of 0.1 and 0.2% in diet, respectively at all the period of experiment (8 weeks). The additives were kindly provided by Dr. Osama Abdel-Monem professor of animal breeding, Faculty of Agriculture, Zagazig University.

Animals were housed in galvanized wire cages, each cage was 40×30×25 cm in well ventilated place at laboratory animal house in ME-VAC. Tap water was automatically available all the time by stainless steel nipples. The rabbits were fed on basal diet that formulated to the level of the nutrient requirements of growing rabbits from 5 to13 weeks of age according to **AOAC (2000)**.

The composition and chemical analyses of experimental basal diet are presented in Table 1.

All rabbits were raised under the same management, hygienic and environmental conditions. Average ambient temperature and relative humidity during the experiment entire period (June-July, 2016) were (27-29 ^{o}C) and (55-70%), respectively. The experimental period was extended for 8 weeks (5-13weaks of age).

Individual live body weight (BW), feed intake (FI), daily weight gain (DWF) and feed conversion ratio were recorded weekly during the experimental period. At the end of the experiment (13 weeks age), three rabbits were randomly chosen from each treatment group. The assigned rabbits were fasted for 16 hours before slaughtering, individually weighted as pre-slaughtered weight. Animals were slaughtered by cutting the jugular veins of the neck. When completed bleeding was achieved, slaughter weight was recorded. After skinning, the carcass was opened down and all viscera were removed.

Table 1. Formulation and calculated chemical composition of the basal diet

Ingredient*	(%)	Calculated chemical composition **	
Clover hay	16.00	DE (Kcal/kg)	2562.46
Soybean meal (44% CP)	19.40	CF (%)	13.37
Yellow corn	16.00	CP (%)	17.29
Wheat bran	33.62	Lysine %	0.90
Wheat straw	12.00	Methionine + cysteine (%)	0.56
Vitam. and Min. mixture*	0.20		
Bone meal	1.30		
Limestone	1.10		
Salt	0.30		
DL-Methionine	0.08		
Total	100.00		

* Each 3 Kg of Vitamamins and minerals mixture contains: Vitam. A 10000 IU; Vitam. D3 2000 IU; Vitam. E 50 mg; Vitam. K3 1000 mg; Vitam. B1 1000 mg; Vitam. B2 5000 mg; Vitam. B6 1500 mg; Vitam. B12 10 mg; Pantothenic acid 60 mg; Niacin 150 mg; Folic acid 1000 mg; Biotin 50 mg; Choline 12000 mg; Iodine 20 mg; Manganese 90 mg; Zinc 210 mg; Copper 10 mg; Iron 30 mg; Iodine 1000 mg; Selenium 100 mg; Cobalt 100 mg and Magnesium 40 mg.

**Calculated composition according to NRC (1977).

The empty carcass, heart, liver, kidney and spleen were separately weight, dressing percentage was calculated according to **(Ayyat *et al.,* 1995).**

Statistical Analysis

The differences among treatments were statically analyzed with a one–way ANOVA test in a completely randomized design according to **Snedecor and Cochran (1982)** as the following model:

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

Where:

Y_{ij}= defiened molel, μ= the overall mean, T_i = the fixed effect of treatment, e_{ij}=residual error. The significant differences among means were compared using Duncan's new multiple- range test **(Duncan, 1995).**

RESULTS AND DISCUSSION

Growth performance of Inmunaire 17.5 based prebiotic and *Spirulina platensis* on growth performance [average body weight (BW), daily body weight gain (DBWG), feed intake (FT) and feed conversion ratio (FCR)] are presented in Table 2.

Live Body Weight and Weight Gain

Concerning BW, the results obtained showed, significant ($p<0.01$) promoting effect on the live body weight and weight gain at 13 weeks of age in rabbit received 1m/1liter in drinking water for three days and 0.2% *Spirulina platensis* in the diet compared with those of the control group. On the other hand, the group fed diet supplemented with 0.1% *Spirulina platensis* showed insignificant higher final BW (13weeks of age) than in control. The results showed that the average BW of rabbits given drinking water supplemented with 1 ml Inmunair 17.5/1 liter and diet supplemented with 0.1or 0.2% *Spirulina platensis* were heaver by about 14.3, 9.2 and 10%, respectively when compared with control group.

With regard to DBWG, results in Table 2 show that, addition of Inmunair 17.5 (1 ml/1 liter)

Table 2. Growth performance ($\bar{X}$ ± SE) of New Zealand White rabbits as affected by Inmunair and Spirulina addition

	Treatment				
	Control	**Inmunair17.5 1ml/litter**	**Spirulina 0.1%**	**Spirulina 0.2%**	**Sign.**
Live body weight (g)					
5 week	695±69.8	690.3±61.6	683.7±51.4	689.3±52.2	NS
9 week	14.09±91.2	1461.5±73.6	1391.8±50.3	1420.1±50.0	NS
13 week	1809.0±112.6^{a}	2068.0±62.5^{b}	1993.9±59.4^{ab}	2010.5±61.2^{b}	*
Daily body weight gain (g/day)					
5-9 week	25.5±.99^{a}	27.5±1.4^{ab}	25.5±0.7^{a}	28.5±0.2^{b}	*
9-13 week	14.3±1.2^{a}	21.7±1.1^{c}	21.5±1.1^{c}	25.1±0.4^{b}	*
5-13 week	19.9±0.9^{a}	24.6±0.4^{bc}	23.4±0.7^{b}	23.6±1.1^{a}	**
Feed intake (g/day)					
5-13week	75.72±6.3^{b}	80.22±5.8^{a}	81.4±7.2^{a}	82.65±6.9^{a}	*
Feed conversation					
5-13week	3.98±0.02^{a}	3.26±0.01^{b}	3.47±0.03^{b}	3.50±0.01^{b}	*

Means in the same column within each classification bearing different letters are significantly different.
NS = Not significant and ** ($P \leq 0.01$).

in drinking water and Spirulina platensis (0.1 and 0.2%) in the diet, significantly ($p<0.05$ or $p<0.01$) higher DBWG through 5-9 and 5-13 weeks of age with insignificant affect between them than in the control. However at 5-9 weeks of age, DBWG was not significantly affected by addition of Inmunair 17.5 in drinking water or adding 0.1 and 0.2% *Spirulina platensis* in the diet while DBWG was significantly ($P<0.01$) higher in rabbits received diet, supplemented with 0.2% *spirulina* when compared with control and other treatment groups.

The different beneficial effect of Inmunair 17.5 in drinking water may be due, normal intestine micro flora by competitive exclusion and antagonism, altering metabolism by increasing digestive enzyme activity and decreasing bacterial enzyme activity and ammonia production, improving feed intake and digestion and neutralizing entertains and stimulating the immune system **(Boham and Srour, 1995; Jin *et al.*, 1997)**. Suppressing ammonia production and urease activity can be beneficial for improving animal and enhancing growth because ammonia produced by urealysis in the intestinal mucosa can exert significant damage to the surface of cells **(Li, 1995)** improvement of growth performance (BW and DBWG) results of *Spirulina platensis* supplementation to diet of growth rabbits may be attributed to the synergetic effect of the chemical constituents of Spirulina platensis dried supplemented has an excellent nutrition profile (high carotenoids, high proteins which includes all of the essential amino acids and rich in mineral and Vitamamins **(Ross and Dominy, 1990).** Spirulina has shown to enhance immune function, reproduction and growth as reported by **(Qureshi *et al*., 1994; Khan *et al.*, 2005).** Feeding *Spirulina* containing diet may increase the lactobacillus population and enhance the absorbability of dietary Vitamamins **(Mariey *et al*., 2012).** In this respect, the effect of *Spirulina platensis* levels on rabbits was studied by **Kaoud (2013)** who showed that supplement of dry powder *Spirulina platensis* (1 kg/ten of feed) for 6 weeks to Hubbard broils chickens diet were significantly increased body weight and

daily body weight gain as compared with control group. The same conclusion was reported by **(Shanmugapriya and Saravana 2014) and (Shanmugapriya *et al.,* 2015),** who showed that broiler chicken fed diet contained *Spirulina platensis* treatment (**Jamil, 2015).** Noted that the body weight of broiler chicks was significantly ($p<0.05$) increased in treatment groups fed with *Spirulina* diet (0.2, 4, and 8% *Spirulina*/kg) from 7^{th} to 28^{th} days. Contradicting results were obtained by **(Saad, 2007)** who revealed that, adding *Spirulina platensis* at level of 1 and 0.5 g/kg diet had insignificant effect on BW at 6, 8, 10 and 12 weeks of age and at the end of the experimental period (14 weeks of age). BWG during 4-8, 9-12 and 13-14 weeks of age. **Raach-Moujahed *et al.* (2011)** found that feeding Spirulina to diets to chicks for 38 days and increasing Spirulina rate in diet did not significantly affect body weight or daily body weight gain.

Moreover (**Abdel-Azeem *et al.,* 2009)** found that live body weight and body weight gain of rabbits significantly ($p< 0.01$) improved by prebiotics (400mg bioplus 2B/Kg feed) supplementation, on the other hand, **(Matusevicus *et al.,* 2006)** found that addition of Bioplus 2B at level of 400mg/kg did not affect significantly the body weight and daily weight gain of rabbits, during the period between 35and 66 days of age. **(El-Habback *et al.,* 2015)** showed that, dietary treatment of broilers chickens by Inmunair17.5 at inclusion rate of 1m/1litter showed significantly higher body weight in the treated birds than control group.

Feed Intake and Feed Conversion Ratio

The effect of experiment of drinking water and feed additives on the average of feed intake and feed conversion ratio significantly ($p<0.05$ or $p<0.01$) improved as a result of improved daily body weight gain (Table 2) and showed the similar pattern which observed with the results of body weight gain. It is worth noting that, average feed intake of rabbits given 1ml Inmunair 17.5 in dirking water or 0.1and 0.2% *spirulina* in the diet were improved by about 5.6, 6.9 and 8.3% respectively. The corresponding values of FCR were 18.12 and 12.1%, respectively when compared with control group. Also, the improvement effect of Inmunair 17.5 supplementation in drinking water was better than those of *Spirulina* (Table 2) **Fuller (1997).** Explained improvement of FCR values by prebiotics by the balance of microbial population reacted in the digestive tract and role of lactobacillus in preventing the harmful bacteria. **Ezzat *et al.* (1988)** reported similar result in lactobacillus preparation and expiration and explained that a possible increase in gut motility may occur in the presence of excessive number organism , there by altering nutrient availability for absorption, in addition to that other beneficial bacterial population may be altered, disrupting cohabitation of the established micro flora **(Miles,1993).** These results agree with **(Abdel-Azeem *et al.,* 2009),** who found that average feed intake and feed conversion ratio in the growing rabbits were improved in group fed 200 mg lincofeed/diet. **Saad (2007)** indicated that, the highest value of feed intake was recorded by chicks fed diet containing *Spirulina platensis* (0.5or1g/kg diet) compared with control group. **Shanmugapriya and Saravana (2014) and Shanmugapriya *et al.* (2015)** indicated that chick fed 10% of *Spirulina platensis* improved feed intake and feed conversion ratio compared with control group, however, **(Zahroojian; *et al.;* 2013)** showed that, feed intake and feed conversion ratio of laying hens were not significantly affected by the diet treatment of *Spirulina* levels (0, 1.5, 2.0 and 2.5%) at 63 weeks of age.

Carcass traits

The average value of some carcass traits of growing rabbits as affected by additives supplementation are shown in Table 3. From results in Table 3, it could be noticed that, most carcass traits studied (carcass weight, fore part**,** hind part, liver, head, heart and lung, kidney and dressing) except spleen and lion were significantly ($p<0.5$ or $P<0.01$) higher in growing rabbit received drinking water supplemented with 1ml Inmunair/liter and rabbit fed on diet contained 0.1 or 0.2% Spirulina, compared with control. Regarding to the effect of Inmunair, agreed with **El-Adawy *et al.* (2002)** reported that carcass traits and internal organs as percentages of live body weight were insignificantly affected by dietary supplementation with biogenic (as a prebiotic), except the dressing percentage which increased significantly with the supplementation.

Table 3. Carcass traits ($\bar{X}$ ± SE) of New Zealand White rabbits as affected by Inmunair 17.5 and Spirulina addition

	Treatment				
	Control	**Inmunair 17.5 1 ml/litter**	**Spirulina 0.1%**	**Spirulina 0.2%**	**Sign.**
Carcass traits					
Carcass weight	911.0±16.6^{a}	1337.0±19.7^{B}	1249.3±14.0^{B}	1258.8±10.1^{b}	*
Lion	221.7±14.8	319.7±18.3	286.3±16.8	289.1±19.3	NS
Forepart	240.0±10.0^{a}	299.7±14.9^{ab}	291.0±10.5^{ab}	291.3±12.3^{ab}	*
Hind part	263.3±10.2^{a}	448.7±13.5^{b}	399.3±14.9^{b}	402.4±12.9^{b}	*
Liver	38.3±3.3^{a}	51.3±4.8^{ab}	51.0±3.5^{B}	51.0±2.6^{b}	*
Head	130.0±12.9^{a}	189.3±11.0^{b}	191.7±12.5^{b}	193.±10.1^{b}	**
Heart and lung	8.7±0.7^{a}	15.3±0.7^{c}	16.3±2.9^{c}	16.5±1.0^{c}	*
Kidney	8.0±0.6^{a}	11.7±1.3^{B}	12.7±1.7^{b}	13.0±1.5^{b}	*
Spleen	1.0±02	1.3±0.3	1.3.0±0.3	1.4.0±0.4	NS
Dressing	57.0%	64.7%	65.7%	62.6%	*

Means in the same column within each classification bearing different letters are significantly different.
NS = Not significant and ** ($P \leq 0.01$).

Regarding to the effect of *Spirulina*, results was in harmony with **Mariey *et al.* (2014)** who found that absolute weight of carcass, giblets and total edible part of broiler chicks was significantly ($p<0.05$) increased by dietary Spirulina groups compared with the control group. **(Shanmugapriya and Saravana, 2014)** reported that, represent meat weight , breast weight ,gizzard, liver and heart weight were increased and abdominal fat was decreased in chick fed diet containing 0.1% of Spirulina platensis compared with control and other treatment groups.

Conclusion

It could be use each of Inmunair17.5 (1 ml/ liter of drinking water) as water supplements and *Spirulina* (0.1% or 0.2%) as diet supplement for NZW rabbits which gave the best growth performance and carcass traits.

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تأثير الإسبيروليـنا والبيربيوتك (انميونير ١٧,٥) على أداء الأرانب النيوزيلندية البيضاء النامية

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تهدف هذه الدراسة إلى تقيم أثر المعضد الحيوي inmunair17.5 وطحلب الاسبيروليـنا على أداء النمو فى الأرانب النامية عند عمر ٥-١٣ أسبوع واستخدم فى هذه التجربة عدد ٢٤ أرنب نيوزيلندي أبيض فى عمر ٥أسابيع ومتساويين تقريبا فى وزن الجسم الإبتدائي ٦٩٥ جم وزعت عشوائيا إلى ٤ مجاميع (٣ مكررات ولكل مكرره ٢ أرنب) وقد تم إسكان كل مكرره في قفص مجلفن بأبعاد ٤٠×٣٠×٢٥سم التجربة ٨ أسابيع، ولقد أعطيت المجموعة الأولى عليقه وماء صنبور بدون إضافات (الكنترول) والمجموعة الثانية عليقه بدون إضافات وماء مضاف إليه المعضد الحيوي inmunair17.5 (بمعدل ١مل/١ لتر ماء) لمدة ٣ أيام فى بداية التجربة والمجموعة الثالثة والرابعة أعطيت طلحب الاسبيروليـنا لمستوى (٠,١ و٠,٢ عليقة على التوالي وماء الصنبور بدون إضافات وقد تم دراسة تأثير إضافة هذه المواد على أداء النمو (وزن الجسم – وزن الجسم المكتسب – معدل استهلاك الغذاء – ومعامل التحويل الغذائي) وكذلك على صفات أجزاء الذبيحة خلال الفترة العمرية (٥-١٣ أسبوع)، وقد أظهرت النتائج تحسن معنوي ($p<0.05$) فى وزن الجسم عند الأسبوع ١٣ ومتوسط الغذاء المأكول ومعدل الكفاءة التحويلية فى الأرانب التى أعطيت المعضد الحيوي inmunair17.5 (١مل/لتر ماء) فى ماء الشرب أو (٠,١ و ٠,٢%)/كجم عليقة طحلب الاسبيروليـنا عند مقارنتها بالكنترول، تحسنت معظم أجزاء الذبيحة المدروسة (الجزء الأمامى- الجزء الأوسط - الكبد - الرأس - القلب - الرئة- الكليتين- ونسبة التصافى) ماعدا الجزء الخلفى والطحال تحسنا معنويا فى الأرانب التى أعطيت المعضد الحيوي inmunair17.5 (١مل/١ لتر ماء) فى ماء الشرب أو (٠,١ و٠,٢%)/كجم عليقة طحلب الاسبيروليـنا، ونستنتج من هذه الدراسة أن استخدم المعضد الحيوي inmunair17.5 (١مل/١ لتر ماء) فى ماء الشرب أو (٠,١ و٠,٢%) /كجم عليقة طحلب الاسبيروليـنا أعطى أفضل أداء للنمو وخصائص الذبيحة.

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