Effect of onion meal and commercial prebiotic on growth and survival of common carp, *Cyprinus carpio* juveniles cultivated in earthen ponds

Majid M. Taher [1*], Ahmed M. Mojer[1], Sadiq J. Muhammed[1], Adel Y. Al-Dubakel[1], Osamah A. Salih[2] and Zeki A. Sabti[1]
[1]Aquaculture Unit - College of Agriculture- University of Basrah
[2]Diroctorate of Basrah Agriculture
*Corresponding Author E-mail: maj61ae@yahoo.com

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Abstract

The current experiment was conducted in earthen ponds at Agricultural Research Station belong to Aquaculture Unit- Agriculture College at Basrah University, Al-Hartha District from 16th May to 30th July 2022. Eight small earthen ponds (600 m2) were used for current experiment, and each pond stocked with 3000 common carp, Cyprinus carpio juveniles at average weight of 0.572 g. The purpose of current experiment is to investigate the differences in survival rate and growth performance with different additives [without additives (C), addition of 1.5 g probiotics/kg feed (T1), addition of 15 g dry onion meal/kg feed as prebiotic (T2), addition of 0.75 g probiotics and 7.5 g onion/kg feed as symbiotic (T3)]. The results revealed that survival rate for different treatments was more than 90%. Highest average final weight (13.58 g) and weight gain (13.01 g) were reached by fishes in T2. Statistical analysis of the results proved that there were no significant differences (P>0.05) in survival rate, growth criteria and feed conversion rate among different treatments. All treatments have a positive allometric growth pattern, with T2 exhibiting the maximum slope value (b) of 3.8097 and T1 exhibiting the lowest of 3.0421. Statistical analysis of condition factors proved that there were significant differences (P≤0.05) in modified condition factor between C and T2 with T1 and T3 and between T1 and T3. For relative condition factor there were significant differences (P≤0.05) between C and other treatments, while there were no significant differences (P>0.05) between T1, T2 and T3. The results of Fulton's condition factor appeared significant differences (P≤0.05) between T2 and T3 with C and T1 and also between C and T1.

Keywords: Cyprinus carpio, Onion, Daily growth rate, Feed conversion rate, Probiotic.

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Introduction

It is well known that common carp, *Cyprinus carpio* is one of the famous species that play significant role in inland freshwater fish production. For previous reason common carp introduced to inland waters in different regions around the world (Vilizzi *et al*., 2015; Ljubojević *et al*., 2016; Khan *et al*., 2016). Common carp was the fourth important cultivated species at 2020 contributing 8.6% of total major world production after grass carp, *Ctenopharyngodon idella* (11.8%), silver carp, *Hypophthalmichthys molitrix* (10%) and Nile tilapia, *Oreochromis niloticus* (9%) (FAO, 2022). Common carp production in Iraq is much lower than other countries and the main aquaculture system was earthen ponds followed by floating cages.

The survival and success of larvae and juveniles in natural habitats depend mainly on food availability, water quality and escaping from predators. It is well known that *Artemia nauplii* used in intensive carp culture, but Dabrowskii *et al*. (1984) stated that great attention was given to their replacement by a more practical inert diet. It had been stated that in culture conditions, the success rate of fish larvae and juveniles is high due to regulated feed supply and absence of predators, but the mortality rate is still high (Research Council of Norway, 2009). Rathore *et al*. (2016) pointed out that it was necessary for designing larval diets that meet the larval requirements for optimal ingestion, digestion and absorption that lead to good growth and survival rates. Probiotics are beneficent microorganisms added to the feeds to accelerate the growth and enhance the health of cultivated animals (Bajagai *et al*., 2016), and Dietary supplementation with probiotics could increase growth rate in aquaculture (Wang *et al*., 2020).

Previous studies revealed that certain probiotic strains could boost immunity prebiotics are complex indigestible saccharides added to the feeds to accelerate the growth and enhance the health of cultivated animals (Hutkins *et al*., 2016). Newman and Arshad (2020) mentioned the synbiotics are using probiotics together with prebiotics in the feeds of cultivated animals can have greater consequences compared to the activity of the prebiotic or probiotic alone . Yazawa *et al*. (1978) used for the first time many carbohydrates in the diets of mammals, while in aquaculture the first study on prebiotics done by Hanley *et al*. (1995). Many laboratory studies in Iraq deal with effects of different prebiotics on growth and health of common carp (Al-Atabi, 2012; Ahmed, 2014; Al-Faiz *et al*., 2014; Al-Faragi, 2014; Mustafa *et al*., 2014; Abdulrahman and Ahmed, 2015; Abdulrahman *et al*., 2016; Al-Muslimawi and Al-Shawi, 2016; Mohammad, 2016; Taher *et al*., 2018). Many studies in Iraq deal also with the effects of different probiotics on growth and health of common carp (Al-Saphar, 2012; Nasir *et al*., 2013; Al-Asha'ab *et al*., 2014; Al-Niaeem, 2019; Taher and Al-Niaeem, 2020; Al-Mhanawi *et al*., 2021; Mojer, *et al*., 2021; Al-Janabi, 2022; Taher *et al*., 2022). The present experiment aims to study the effects of addition probiotic, prebiotic and

synbiotics to the diets on growth and survival of common carp juvenile cultivated in earthen ponds.

Materials and Methods

The current experiment was took place in earthen ponds at Agricultural Research Station belong to Aquaculture Unit- Agriculture College at Basrah University, Al-Hartha District about 16 km northern-east of Basrah Governorate (30065`64.6"N, 470 74`79.5"E) from 16th May to 30th July 2022. Eight small earthen ponds (600 m2) were used and each pond stocked with 3000 common carp juvenile at average weight of 0.572 g. The current study conducted to investigate the differences in survival rate and growth performance for these juveniles fed diets with different additives [without additives in pond 1 and 2 (C), addition of 1.5 g probiotics/kg feed in pond 3 and 4 (T1); addition of 15 g dry onion meal/kg feed in pond 5 and 6 (T2), addition of 0.75 g probiotics and 7.5 g onion/kg feed in pond 7 and 8 (T3)].

Each pond was fertilized by 100 kg of organic buffalo manure, so at the beginning of the experiment fishes depend on natural food in the ponds for 13 days and after that they were fed at feeding ratio 10% of fish weight for 21 days, then decreased to 6% at another 20 days and finally fed at 4% feeding ratio at last 20 days of the experiment. The diets were manufactured by Agricultural Consultant Office belonging to Agriculture College using different ingredients (Fishmeal 45%, wheat meal 25%, wheat bran 18%, barley meal 10% and vitamins-minerals premix 2%).

Total weight of fishes were measured at the beginning and at the end of the experiment, while subsamples of fishes were weighed periodically and daily food changed after each weighing. Total length of fishes was measured only at the end of the experiment. Daily feed was divided into two meals, the first given early on the morning and the second at mid-day. At each sampling interval, the water's temperature, pH, and salinity were recorded.

Throughout this period, four sampling data were collected to calculate the following equations:

Weight increments (WI, g) = FW – IW

Daily growth rate (DGR, g/day) = FW – IW / days

Specific growth rate (SGR, %/day) = 100 *[(ln FW)-(ln IW)] / days

Where: FW = Final fish weight (g); IW = Initial fish weight (g)

Length-weight relationship and condition factor were calculated for fishes at the end of the experiment for each treatment. The following equation was used to calculate the length-weight relationship:

$W = aL^b$ (Pauly, 1983).

Where W= weight of fish in g, L= Length of fish in cm, a = describe the rate of change in weight with length (intercept), and b = weight at unit length (slope).

The condition factors (K) of the carps were estimated using the following equations:

1- Fulton's condition factor, the value of K was calculated according to Froese (2006):

$K3=100w/L^3$

2- Modified condition factor (Ricker, 1975) was estimated according to Gomiero and Braga (2005):

$Kb = 100\ w/L^b$

3- Relative condition factor 'Kn' (Le Cren, 1951) was estimated according to Sheikh *et al.* (2017):

Kn = W/ ^w

Where W= the actual total weight of the fish in g, ^w= the expected weight from length-weight equation formula. The results of current experiment were conducted with a completely randomized design, and the differences between the means were tested by analysis of variance (ANOVA) and the significant differences were tested by LSD test at 0.5% probability level by SPSS program Ver. 26.

Results

Table (1) show the average of fish weight as well as some environmental factors during the experiment. Water temperature ranged between 25-30 ^oC, pH ranged between 7.7-8.0, while salinity ranged between 3.22-5.83 PSU. Highest average final weight (14.12 g) was reached by fishes in pond 6, while lowest final average weight (11.93 g) was reached by fishes reared in pond 7. Table (2) showed the survival rate and growth criteria of common carp juvenile fed diets with different additives. Highest survival rate (96.8%) was achieved by juvenile fed on diet with prebiotic additives (T2) and lowest survival rate (94.3%) was achieved by juvenile fed on diet with symbiotic additives (T3) (Fig. 1). Final average weights of juveniles were (13.49, 13.06, 13.58 and 12.73) g 1n control, T1, T2 and T3 respectively (Fig. 2). Highest weight increments (13.01 g) was achieved by juveniles fed on diet with prebiotic additives and lowest weight gain 12.16 g) was achieved by juveniles fed diet with symbiotic additives (Fig. 3).

Highest daily growth rate (0.173 g/day) was achieved by juveniles fed on diet with prebiotic additives and lowest daily growth rate (0.162 g/day) was achieved by juveniles fed diet with symbiotic additives (Fig. 4). Specific growth rate of juveniles were (3.84, 3.42, 3.47 and 3.88) %/day for control, T1, T2, and T3 respectively (Fig. 5). All feed conversion rates were acceptable and ranged between 1.13 for T2 and 1.29 for T3 (Fi. 6).

Statistical analysis of the results for survival rate and all growth criteria studied in current experiment proved that there were no significant differences (P>0.05) among control and other three treatments and also between these treatments.

Table (3) show that the averages and ranges of length and weight for common carp at the end of experiment. Highest average length (13.58 cm) reached by T2 and lowest (12.73 cm) reached by T3. Table (4) show the parameters of length-weight relationship for different treatments of common carp at the end of experiment. The growth pattern for all treatments is positive allometric, where highest slope value (b) was 3.8097 for T2, while lowest value was 3.0421 for T1. The statistical analysis of the results appeared significant differences (P≤0.01) between slope value and the number three in C and T2, while there were no significant differences (P>0.05) in T1 and T3.

Table (5) show the condition factors of different treatments of common carp at the end of experiment. There are high differences in the modified condition factor, where they range from 0.0877 in T2 to 1.3382 in T1. Relative condition factors were 1.0528, 1.0062, 0.9746 and 0.9953 for control, T1, T2 and T3 respectively. Fulton's condition factor range between 0.6457 to 1.4832. Statistical analysis of condition factors results proved that there significant differences (P≤0.05) in Kb between C and T2 with T1 and T3 and between T1 and T3. For relative condition factor there were significant differences (P≤0.05) between T1 and other treatments, while there were no significant differences (P>0.05) between T1, T2 and T3. The results of Fulton's condition factor appeared significant differences (P≤0.05) between T2 and T3 with C and T1 and also between C and T1.

It can be concluded from the results of current experiment that the addition of commercial probiotic, onion as prebiotic and both of them as symbiotic to the feed of common carp juveniles cultivated in earthen ponds don't have positive enhancements to the growth performance.

Table 1: Weights of common carp juveniles, as well as some environmental factors during the experiment.

Date 2022	Average Fish Weight (g)								Temp. (°C)	pH	Sal. (PSU)
	CP1	CP2	T1P3	T1P4	T2P5	T2P6	T3P7	T3P8			
16/5/	0.572	0.572	0.572	0.572	0.572	0.572	0.572	0.572	25	7.7	3.22
20/6	5.30	5.05	4.01	6.30	3.22	6.25	4.21	6.97	29	7.7	3.78
10/7	8.11	9.58	7.62	12.7	7.72	11.25	8.33	11.23	30	7.8	4.45
30/7	13.08	13.90	12.05	14.08	13.05	14.12	11.93	13.54	30	8.0	5.83

Table 2: Survival rate and growth criteria of different treatments in the experiment.

Growth Criteria	(Control)		T1 (Probiotic)		T2 (Prebiotic)		T3(Pro+Pre)	
	P1	P2	P3	P4	P5	P6	P7	P8
SR (100)	98.1	92.7	91.7	97.5	96.3	97.3	97.1	91.4
Average	**95.4 a**		**94.6 a**		**96.8 a**		**94.3 a**	
FW	13.08	13.90	12.05	14.08	13.05	14.12	11.93	13.54
Average	**13.49 a**		**13.06 a**		**13.58 a**		**12.73 a**	
WI (g)	12.51	13.33	11.48	13.51	12.48	13.55	11.36	12.97
Average	**12.92 a**		**12.49 a**		**13.01 a**		**12.16 a**	
DGR (g/day)	0.167	0.178	0.153	0.180	0.166	0.180	0.151	0.173
Average	**0.172 a**		**0.166 a**		**0.173 a**		**0.162 a**	
SGR (%/day)	4.17	3.51	3.32	3.53	3.42	3.53	3.30	3.47
Average	**3.84 a**		**3.42 a**		**3.47 a**		**3.38 a**	
FCR	1.15	1.20	1.14	1.43	0.93	1.34	1.16	1.42
Average	**1.17 a**		**1.28 a**		**1.13 a**		**1.29 a**	

Different letters in one row are significantly different (P≤0.05).

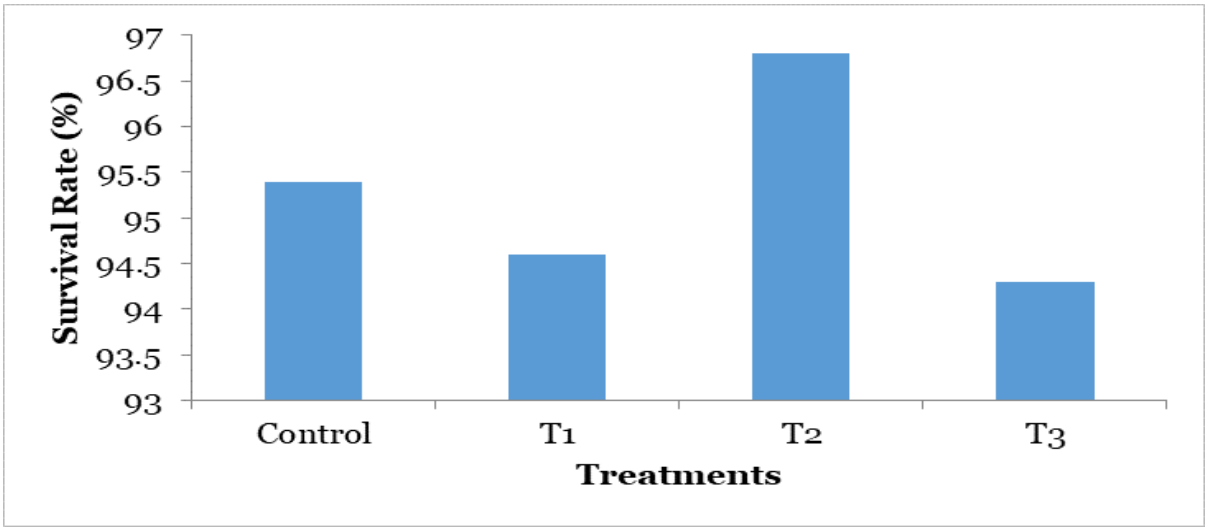

Figure 1: Survival rate of common carp juveniles fed feeds without and with different additives.

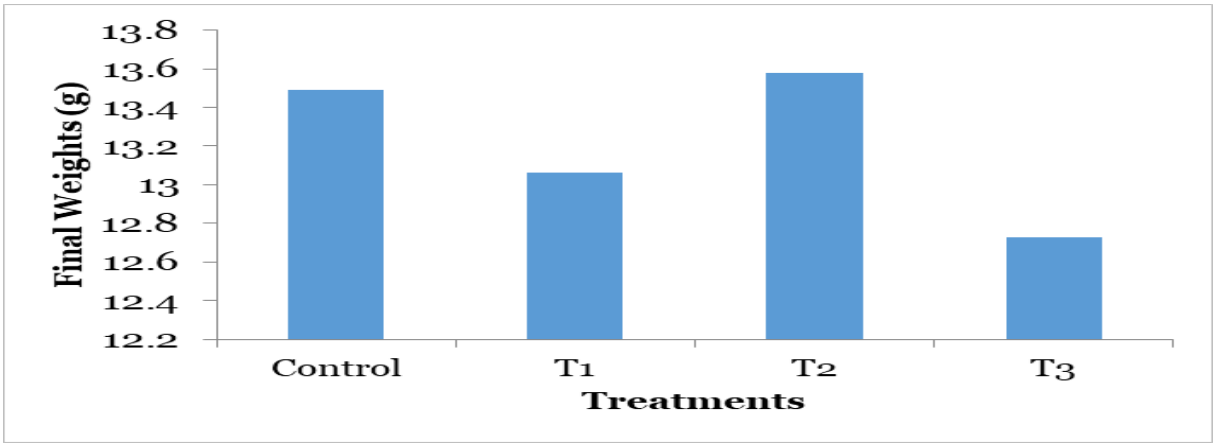

Figure 2: Final weights reached by common carp juveniles fed feeds without and with different additives.

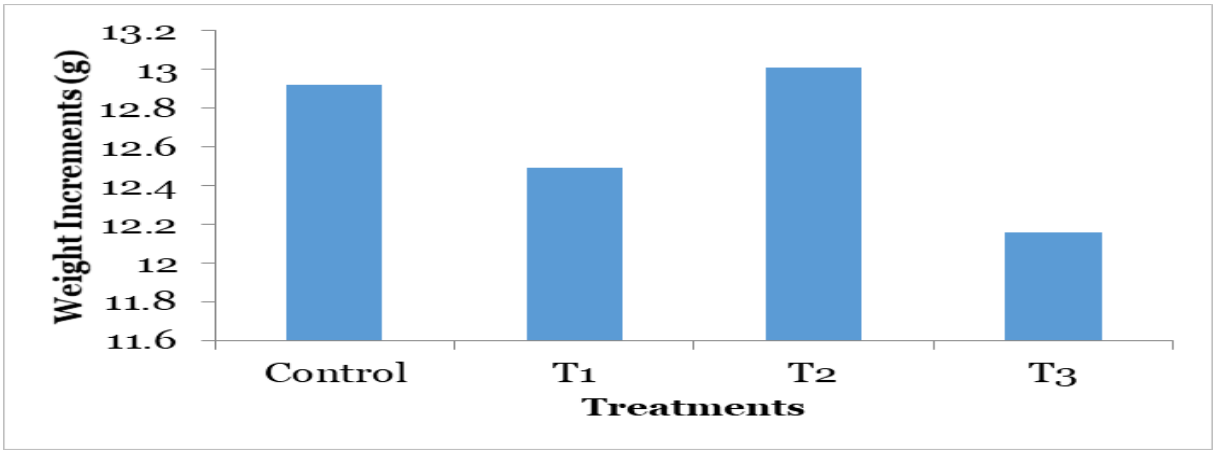

Figure 3: Weight gain of common carp juveniles fed feeds without and with different additives.

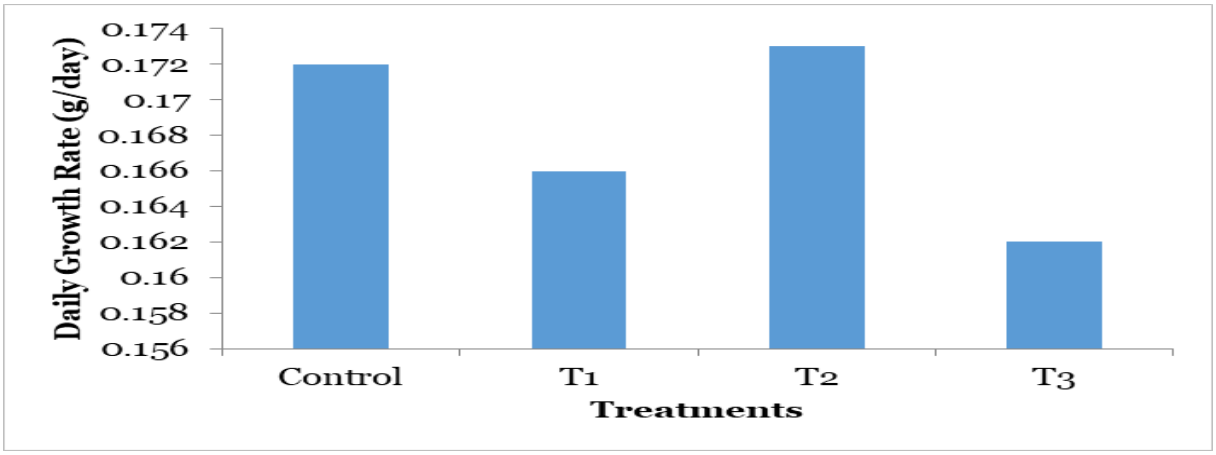

Figure 4: Daily growth rate of common carp juveniles fed feeds without and with different additives.

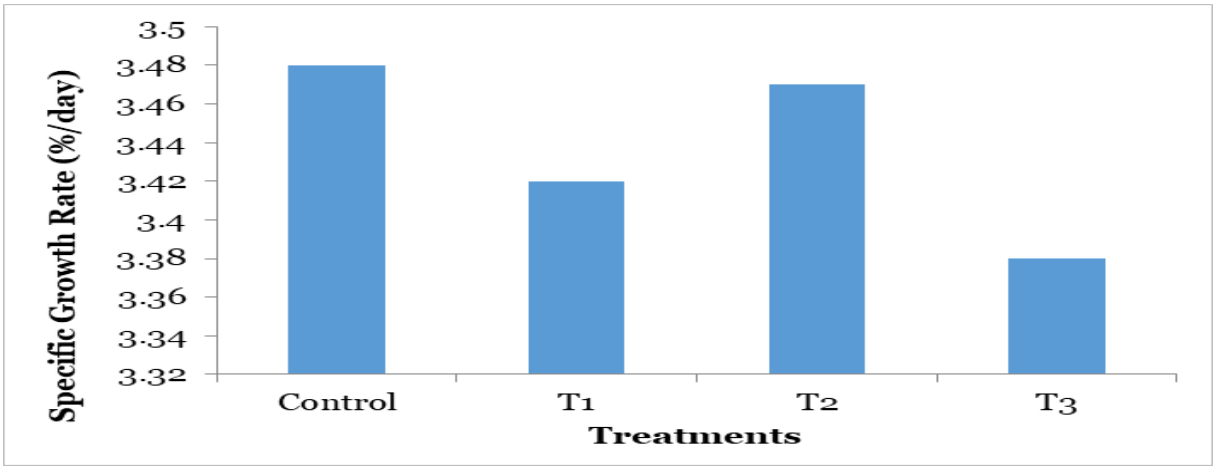

Figure 5: Specific growth rate of common carp juveniles fed feeds without and with different additives.

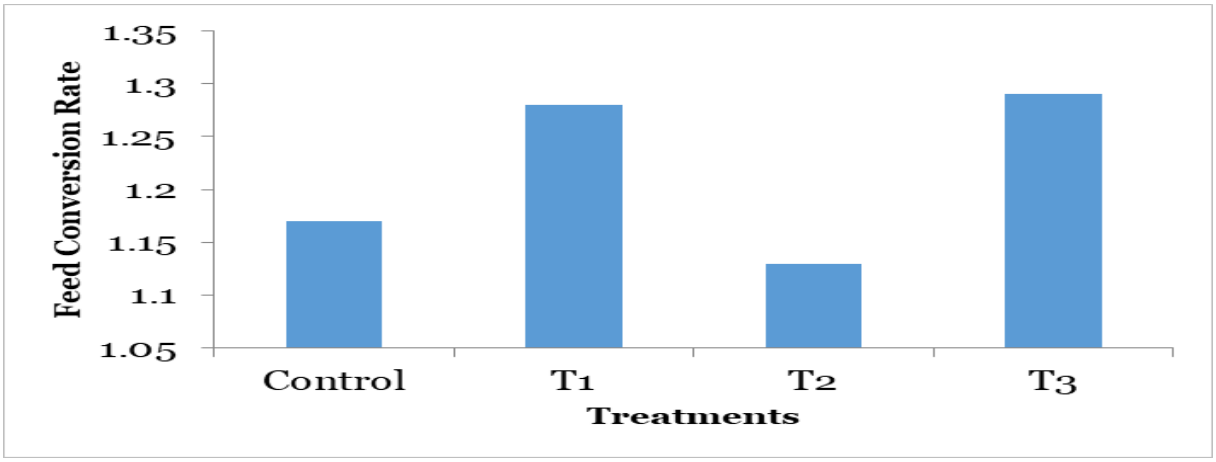

Figure 6: Feed conversion rate of common carp juveniles fed feeds without and with different additives

Table 3: Data on length and weight of common carp after the experiment.

Treatments	Length range (cm)	Weight range (g)	Mean length (cm)	Mean Weight (g)
C	9.1-19.3	3.35-70.21	12.7	13.49
T1	7.3-18.0	6.00-94.00	11.7	13.06
T2	9.8-15.4	4.54-30.85	12.5	13.58
T3	11.0-15.7	8.00-30.24	11.5	12.73

Table 4: Equation parameters of length-weight for common carp after the experiment.

Treatments	a	B	R^2	t value (calculated)	Significance of t
C	0.0010	3.7533	0.9735	7.5496**	0.00001
T1	0.0133	3.0421	0.9778	0.2292	0.41006
T2	0.0009	3.8097	0.9342	4.4586**	0.00005
T3	0.0013	3.6409	0.9081	0.3645	0.35854

Table 5: Condition factors of common carp after the experiment.

Treatments	Condition factors		
	Modified condition factor Kb= 100 W/ Lb	Relative condition factor Kn= W/ W^	Fulton's condition factor K3= 100 W/ L3
C	0.1053 **c** ±0.0091	1.0528 **a** ±0.0909	0.7141 **b** ±0.0934
T1	1.3382 **a** ±0.1085	1.0062 **b** ±0.0816	1.4832 **a** ±0.1209
T2	0.0877 **c** ±0.0111	0.9746 **b** ±0.1235	0.6457 **c** ±0.0927
T3	0.1294 **b** ±0.0110	0.9953 **b** ±0.0849	0.6730 **c** ±0.0670

Different letters in one column are significantly different (P≤0.05)

Discussion

Probiotic and prebiotics added to fish diets in order to stimulate fish appetite, improvement of feeds by production vitamins and enzymes, helping in digesting some complex compounds, then increasing fish immunity and fish growth (Merrifild *et al.*,

2010). Results of current experiments proved that there were no effects for adding probiotics, prebiotics and synbiotics to the feed of common carp juveniles cultivated in earthen ponds. There were no significant differences (P>0.05) in survival rate, feed conversion rate and other growth criteria between control and other three treatments. This results can be attributed to the availability of natural foods found in earthen ponds consumed by these juveniles.

Venter (2007) stated that the continuous adding of prebiotics may create some problems such as the modifying of some diseases microorganisms to get benefits from the carbohydrates found in some prebiotics, while Olsen *et al.* (2001) pointed out that the positive or negative results of adding prebiotics related to the ability or an ability of microorganisms to leavening additional quantities of prebiotics. Al-Asha'ab *et al.* (2014) recorded that feed supplemented with 5 g FOS per kg feed hadn't any effects on growth of young common carp.

The results of current experiment are differ from the results of other studies in Iraq because most of them conducted inside the laboratories. Al-Saphar (2012) found that feed with *Saccharomyces cerevisiae* improved the growth of common carp. Ahmed (2014) and Abdulrahman and Ahmed (2015) referred that diets with the prebiotic FOS improve growth criteria of common carp. Muhsan and Al-Shawi (2016) stated that growth criteria of common carp juveniles were improved at feeding on diet supplemented with some organic acids. Taher *et al.* (2018) stated that highest weight increment (7.63 g) achieved by common carp fingerlings fed diet supplemented with 2% of bay laurel's (*Laurus nobilis*) leaf extract compared with the lowest (5.42 g) that achieved by control, and also better feed conversion rate was 4.56 compared with 6.59 for control, while highest daily growth rate (0.099 g/day) was achieved comparing with 0.070 g/day for control.

Probiotic can enhance immunity system of fishes and then increased survival rates of common carp cultivated inside laboratories (Al-Niaeem, 2019; Taher and Al-Niaeem, 2020). Al-Mhnawi *et al.* (2021) stated that the survival rate, condition factor and feed conversion rate of common carp was better with diet supplemented with 1g thepax per 1 kg feed comparing with diet without thepax. Al-Janabi (2022) stated that final weight average, weigh increment, daily growth rate were significantly (P<0.05) superior in diet with thepax treatment over the other treatments, while relative growth rate, specific growth rate and survival rate were better when diet supplemented with mix of thepax, bio boost and vitamin endo C, also best feed conversion rate (3.03) achieved for bio boost treatment then for (3.06) mix treatment comparing with control (3.69). Taher *et al.* (2022) stated that grass carp fed on formulated feed supplemented with Endo vit. C having high growth performance (WI, DGR and SGR) comparing with control and thepax additives. The results of current experiment are resemble to the result of Taher *et al.* (2024) who investigate the effect of adding different ratio of garlic as prebiotic on growth and survival of young common carp cultivated in earthen ponds.

The results of condition factors in current experiment revealed that there are too much differences in the modified and Fulton's condition factors with very little differences relative condition factor that consider the best for cultured species comparing with the two previous condition factors. Al-Janabi (2022) find different results where stated that there were no significant differences (P>0.05) in the Fulton's condition factor of common carp fed diet with different additives, and lowest value was 1.897 for control.

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تأثير مسحوق البصل والمعزز الحيوي التجاري في نمو وبقاء يافعات الكارب الشائع المستزرعة في الأحواض الأرضية

ماجد مكي طاهر[1]، احمد محسن موجر[1]، صادق جواد محمد[1]، عادل يعقوب الدبيكل[1]، أسامة عبد الهادي صالح[2]، زكي عبد الحسين سبتي[1]

[1]وحدة الاستزراع المائي، كلية الزراعة، جامعة البصرة، البصرة، العراق

[2]مديرية زراعة البصرة، البصرة، العراق

*Corresponding Author Email: maj61ae@yahoo.com

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المستخلص

أجريت التجربة الحالية في الأحواض الأرضية لمحطة بحوث الهارثة الزراعية التابعة لوحدة الاستزراع المائي في كلية الزراعة–جامعة البصرة، وذلك للفترة من 16 شهر مايس لغاية 30 من تموز2022. استعملت ثمانية أحواض صغيرة (600 متر مربع) وكل حوض وضع فيه 3000 يافعة كارب شائع *Cyprinus carpio* بمعدل وزن 0.572 غم. صممت الدراسة الحالية لفحص الاختلافات في معدلات البقاء وأداء النمو لهذه اليافعات المغذات على عليقة ذات إضافات مختلفة (عليقة السيطرة من دون إضافات، عليقة 1 بإضافة 1.5 غم معزز حيوي تجاري لكل كغم علف، عليقة 2 بإضافة 15 غم مسحوق البصل لكل كغم علف وعليقة 3 بإضافة 0.75 غم معزز حيوي تجاري مع 7.5 غم مسحوق البصل لكل كغم علف). بينت نتائج التجربة الحالية نسبة بقاء أكثر من 90% لجميع المعاملات، بينما سجل اعلى معدل وزن نهائي (13.58 غم) واعلى زيادة وزنية (13.01 غم) لأسماك المعاملة الثانية. اثبت التحليل الإحصائي للنتائج عدم وجود فروق معنوية ($P>0.05$) في نسبة البقاء ومعدلات النمو والتحويل الغذائي بين المعاملات الأربعة. أن نمط النمو المسجل لجميع المعاملات هو نمو غير متماثل إيجابي، إذ أن اعلى قيمة للانحدار (b) بلغت 3.8097 للمعاملة الثالثة واقل قيمة بلغت 3.0421 للمعاملة الثانية. اثبت التحليل الإحصائي لنتائج معامل الحالة وجود فروقات معنوية ($P\leq0.05$) في معامل الحالة المحور (Kb) بين المعاملة القياسية والمعاملة الثانية مع المعاملة الأولى والثالثة، وكذلك بين المعاملة الأولى والثالثة. وجدت فروقات معنوية ($P\leq0.05$) لمعامل الحالة النسبي (Kn) بين معاملة السيطرة وبقية المعاملات، بينما لا توجد فروقات معنوية ($P>0.05$) بين المعاملة الأولى والثانية والثالثة. أثبتت نتائج معامل حالة فولتن (K3) وجود فروقات معنوية ($P\leq0.05$)بين المعاملة الثانية والثالثة مع معاملة السيطرة والمعاملة الأولى وكذلك بين معاملة السيطرة والمعاملة الأولى.

الكلمات المفتاحية: الكارب الشائع، البصل، معدل النمو اليومي، معدل التحويل الغذائي، المعزز الحيوي.

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