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Some Studies on Immunostimulent in Marine Fish

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ABSTRACT

This study aimed to prove and study the effect of feeding fish with immunostimulants (Feed additives CA growth and orgazol zinc) on the characters of water quality through determination of the level of (dissolved oxygen, ammonia, nitrites, pH, salinity, organic matter and sulphate) and also on its effects on the concentration of heavy metals in waters that include (lead, arsenic, mercury, cadmium, copper and zinc). Immunostimulants represent a modern and promising tool in aquaculture, enhancing the resistance of cultured fish to disease and stress. The results indicated that, the effect of CA growth and Orgazal zinc in seabream is higher than that of seabass as it affect DO, ammonia , nitrite, pH , salnity, organic matter and sulphate higher than that of seabass. The orgazol zinc increased the level of lead, arsenic, mercury, cadmium, copper and zinc than that of CA growth treated fish and its effect higher in seabream than that of seabass treated fish.

Keywords:

INTRODUCTION

Immunostimulants are biological extracts and synthetic chemicals which stimulate the immune response by promoting phagocytic cell function, increasing their bactericidal activity and/or non-specific cytotoxic cells and antibody production *(Sakai, 1999).* Also immunostimulants represent a modern and promising tool in aquaculture, enhancing the resistance of cultured fish to disease and stress *(Smith et al., 2003).* The addition of immunostimulants can improve the immunity of the fish via improving lymphocyte, monocytes and neutrophils and its activity, improving resistance against infectious diseases of either bacterial or viral diseases, improving the body weight, body weight gain and feed conversion, also improve carcass characters and relative body weight of internal organs in relation to body weight (*Tang et al., 2008).*

MATERIALS AND METHODS

A- MATERIALS

1.Fish

In our investigation, a total number of 400 cultured marine fishes of body weight range (50±3 gm) of Seabream (*Sparus aurata L.*), and seabass fishes were collected showing clinical signs from private fish farm at Alexandria governorate, Egypt.

2. Fish diets

Fish were fed on a commercial fish diet containing 45% crude protein. The diet was daily provided at a fixed feeding

ratio of 3 % of body weight of fish as described by *Eurell et al. (1978*).

3. Feed additives

Two feed additives were used which include CA growth (New growth) and orgazol zinc.

A-CA growth (New growth)

B-Orgazol zinc

4- Examination of water collected sample

A secehi disc depth reading of 40-60 cm was maintained in different farms salinity PH, temperature, total ammonia Nitrogen (TAN), dissolved oxygen and unionized ammonia were analyzed using the salicylate method and diazolization method, respectively, using (the Hach DREL 2400 portable water quality laboratory).

5- Determination of heavy metals levels

The method for analysis of the heavy metals in the water was carried out according to *APHA (1995*) and in the fish tissues according to *Clesceri (1998*) that was carried out using Atomic Absorption Spectrophotometery. Atomic Absorption Spectrophotometer (Model Thermo Electron Corporation, S. Series AA Spectrometer with Gravities furnace, UK,) instrument was used to detect the heavy metals. The concentrations of heavy metals were expressed as mg/l for water and µg/g dry wt. for fish tissues. Fish specimens were digested according to *AOAC (1996).*

6- Growth performances measurements

According to *El-Shinawy (1999).*

$$\text{Feed conversion ratio} = \frac{\text{Amount of feed intake}}{\text{Body weight gain}}$$

$$\text{Feed efficiency ratio} = \frac{\text{Body weight gain}}{\text{Amount of feed intake}}$$

RESULTS

A-Growth parameters:

1-Effect of feeding different growth promoters to seabream fish on body weight measurements

The results cleared in Tables (1, 2 and 3) indicated the significant differences body weight, body weight gain and food conversion among different treatments, of orgazol zinc and CA growth and also among different weeks of experiments.

Our results indicated that, the body weight of fish of a higher level in the seabass fish than that of the seabream and in the group treated with orgazol zinc than that of CA growth.

2-Effect of feeding different growth promoters to seabream fish on economic return:

The results cleared in Table (4) indicated the significant differences of total returns/100 fish among seabass and seabream fish, also, among different treatments, of orgazol zinc and CA growth and also among different weeks of experiments.

Table (1): ***Effect of different feed additives on body weight in seabass and seabream.***

Feed additives	Fish species	Original weight	1st week	2nd week	3rd week	4th week
Orgazol zinc	Seabass	A	A	A	A	A
		80.50±4.55	80.50±5.88	93.50±3.55	104.60±7.88	115.20±7.12
	Seabream	C	C	C	C	C
		31.50±2.55	31.50±2.33	40.30±3.44	52.50±2.55	63.50±5.66
CA growth	Seabass	B	B	B	B	B
		64.50±3.55	64.50±3.55	71.30±3.77	82.20±3.88	98.50±5.99
	Seabream	D	D	D	D	D
		27.50±2.30	27.50±3.55	32.40±3.24	40.50±2.55	53.20±3.55

Means within the same column of different litters are significantly different at (P < 0.05).

Table (2): ***Effect of different feed additives on body weight gain in seabass and seabream.***

Feed additives	Fish species	Gain1	Gain2	Gain3	Gain 4	Gain total
Orgazol zinc	Seabass	A	A	B	D	A
		0	13±1.33	11.10±1.12	10.6±1.16	34.70±3.44
	Seabream	A	B	A	C	B
		0	8.80±1.88	12.20±1.23	11±1.13	32.00±5.33
CA growth	Seabass	A	C	C	A	A
		0	6.80±0.85	10.9±1.99	16.30±3.16	34±3.44
	Seabream	A	D	D	B	C
		0	4.90±0.99	8.10±1.18	12.70±2.77	25.70±3.55

Means within the same column of different litters are significantly different at (P < 0.05).

Table (3): ***Effect of different feed additives on food conversion ratio in seabass and seabream.***

Feed additives	Fish species	FCR1	FCR2	FCR3	FCR4	FCR total
Orgazol zinc	Seabass	A	B	A	A	A
		0	5.65	7.09	7.92	6.88
	Seabream	A	D	D	C	D
		0	3.57	3.01	3.81	3.46
CA growth	Seabass	A	A	B	B	B
		0	6.17	4.82	3.87	4.95
	Seabream	A	C	C	D	C
		0	4.28	3.89	3.30	3.83

Means within the same column of different litters are significantly different at (P < 0.05).

Table (4): ***Effect of different feed additives on economic return/100 fish body weight gain in seabass and seabream.***

Feed additives	Fish species	Total return1	Total return 2	Total return.	Total return4	Total return
Orgazol zinc	Seabass	A	A	B	C	A
		0	2600	2220	2120	6940
	Seabream	A	B	A	D	C
		0	1466	2033.33	1833.33	5333.34
CA growth	Seabass	A	C	C	A	B
		0	1372	2180	3260	6800
	Seabream	A	D	D	B	D
		0	816.66	1350	2116.66	4266.66

Means within the same column of different litters are significantly different at (P < 0.05).

B-Water quality parameters:-

1-Effect of feeding different growth promoters to seabream fish on water quality parameters at different periods of experiments

The results in Tables (5, 6 and 7) indicated that, the level of dissolved oxygen increased toward the end of the experiment. The results indicated that, the level of dissolved oxygen increase in the group treated with CA growth especially with higher dose (2ml/kg) followed by control group and the minimum level of dissolved oxygen observed in the orgasol zinc, especially at a higher dose of orgzol zinc (0.4 gm/Kg). The ammonia level (Tables, 5,6 and 7) cleared that, the level of ammonia increased significantly ($P < 0.05$). the higher level of ammonia increased with orgasol zinc especially the higher dose of orgazol zinc followed by the control group and the minimum levl of ammonia observed in CA growth especially a the higher dose of CA growth zinc. The Nitrate level also increased progressively from the 1st to the 4th week of the experiment. the higher nitrite level observed in the group treated with orgazol zinc especially the higher doses followed by control group and the lower level of nitrite observed in the group treated with CA growth especially the lower doses of CA growth. The pH level data presented in Tables (5, 6 and 7) cleared that the pH value increased progressively from the 1st to 4th week of the experiment. The pH level increased with the higher dose of CA growth followed by control group and decreased with the higher dose of orgazol zinc. The **salinity** level not changed at the all periods of the experiment and its level not changed among all treatment level.

The organic matter level increased progressively from the 1st week to the last week of the experiment. The higher level of organic matter observed in the group treated with Orgazol zinc especially the higher dose of orgazol zinc followed by control group and the lower organic matter level observed in the group treated with CA growth.

The sulphate level increased progressively from the 1st week of the experiment to the 4th week and the higher level of sulphate observed in the orgazol zinc especially at a higher doses followed by control group and the lower sulphate level observed in CA treated

C-Heavy metals parameters:-

2-Effect of feeding different growth promoters to seabream fish on heavy metals parameters at different periods of experiments:

The data presented in Tables (8, 9 and 10) cleared that, the level of lead, arsenic, mercury, cadmium, copper and zinc differ significantly ($P < 0.05$) among different periods of experiment and differ among fish of either seabream and seabass. The results indicated that, the level of lead increase in the group treated with orgazol zinc than the CA growth treated fish and the lead increased progressively from the 1st week to the last week of the experiment and in seabass higher than that of seabream. The arsenic l;evel increased in seabream treated fish than seabass treated fish and its level not changed from the 1st week to the 4th week of the

Table (5): ***Effect of feeding different growth promoters to seabass fish on water quality parameters at different periods of experiments.***

Week	Dose	N	DO	Ammonia	Nitrite	pH	Salinity	O M	Sulphate
			Mean ± Standard Error						
1st week	CA growth 1ml/Kg	3.0	G 4.70±0.01	E 0.12±0.01	D 0.11±0.01	D 7.80±00.01	A 28.00±0.01	F 2.13±0.00	C 30.17±0.03
	CA growth 2ml (Kg	3.0	G 4.70±0.01	E 0.12±0.01	D 0.12±0.01	D 7.77±00.03	A 28.00±0.01	F 2.13±0.00	C 30.17±0.07
	Orgazal zinc 0.2 gm/Kg	3.0	G 4.70±0.01	E 0.12±0.01	D 0.12±0.01	CD 7.87±00.03	A 28.00±0.01	F 2.12±0.01	C 30.13±0.03
	Orgazol zinc 0.4 gm *Kg	3.0	G 4.70±0.01	E 0.12±0.01	D 0.12±0.01	C 7.90±00.01	A 28.00±0.01	F 2.13±0.00	C 30.10±0.00
	Control	3.0	G 4.70±0.01	E 0.12±0.01	D 0.12±0.01	D 7.83±00.03	A 28.00±0.01	F 2.12±0.00	C 30.20±0.06
2nd week	CA growth 1ml/Kg	3.0	F 4.80±0.01	EF 0.11±0.01	DE 0.11±0.01	C 7.90±00.01	A 28.00±0.01	F 2.06±0.01	D 28.13±0.03
	CA growth 2ml (Kg	3.0	E 4.90±0.01	F 0.10±0.01	E 0.10±0.01	C 7.93±00.03	A 28.00±0.01	F 2.01±0.00	D 27.83±0.27
	Orgazal zinc 0.2 gm/Kg	3.0	G 4.73±0.03	D 0.13±0.01	CD 0.13±0.01	D 7.83±00.03	A 28.00±0.01	F 2.19±0.02	C 30.80±0.06
	Orgazol zinc 0.4 gm *Kg	3.0	I 4.63±0.03	C 0.14±0.01	CD 0.13±0.01	D 7.80±00.01	A 28.00±0.01	D 2.28±0.02	C 31.73±0.12
	Control	3.0	G 4.70±0.01	E 0.12±0.010	D 0.12±0.01	C 7.90±00.01	A 28.00±0.01	E 2.11±0.00	C 30.13±0.03
3rd week	CA growth 1ml/Kg	3.0	D 5.00±0.01	F 0.09±0.01	E 0.09±0.01	C 7.90±00.01	A 28.00±0.01	G 1.97±0.01	D 27.30±0.12
	CA growth 2ml (Kg	3.0	C 5.13±0.03	F 0.08±0.01	E 0.08±0.01	AB 8.10±00.01	A 28.00±0.01	G 1.84±0.01	E 25.57±0.09
	Orgazal zinc 0.2 gm/Kg	3.0	H 4.67±0.03	B 0.15±0.01	C 0.15±0.01	D 7.83±00.03	A 28.00±0.01	C 2.43±0.04	C 31.43±0.07
	Orgazol zinc 0.4 gm *Kg	3.0	K 4.60±0.01	B 0.15±0.01	C 0.15±0.01	D 7.80±00.01	A 28.00±0.01	B 2.60±0.02	B 35.60±0.38
	Control	3.0	G 4.70±0.01	E 0.12±0.01	D 0.12±0.01	C 7.90±00.01	A 28.00±0.01	F 2.11±0.01	C 30.13±0.03
4th week	CA growth 1ml/Kg	3.0	B 5.17±0.03	0.07±0.01	E 0.06±0.01	B 8.03±00.07	A 28.00±0.01	G 1.73±0.01	E 25.20±0.06
	CA growth 2ml/Kg	3.0	A 5.33±0.03	0.05±0.01	E 0.05±0.01	A 8.17±00.03	A 28.00±0.01	H 1.49±0.03	F 20.23±0.07
	Orgazal zinc 0.2 gm/Kg	3.0	K 4.57±0.03	B 0.16±0.01	B 0.16±0.01	D 7.73±00.03	A 28.00±0.01	B 2.60±0.04	B 35.33±0.89
	Orgazol zinc 0.4 gm *Kg	3.0	L 4.47±0.03	A 0.18±0.01	A 0.17±0.01	D 7.77±00.03	A 28.00±0.01	A 2.86±0.02	A 38.30±0.06
	Control	3.0	G 4.70±0.01	E 0.12±0.01	D 0.12±0.01	C 7.90±00.01	A 28.00±0.01	F 2.11±0.01	C 30.20±0.06

Means within the same column of different litters are significantly different at (P < 0.05).

Table (6): ***Effect of feeding different growth promoters to seabream fish on water quality parameters at different periods of experiments..***

Week	Dose	N	DO	Ammonia	Nitrite	pH	Salninity	OM	Sulphate
			Mean ± Standard Error						
1st week	CA growth 1ml/Kg	3	E 4.70±0.01	D 0.12±0.01	F 0.11±0.01	E 7.80±00.001	A 28.00±0.01	C 2.23±0.01	B 36.97±6.57
	CA growth 2ml (Kg	3	E 4.70±0.01	D 0.12±0.01	F 0.11±0.01	F 7.77±00.03	A 28.00±0.01	C 2.20±0.05	C 31.10±0.46
	Orgazal zinc 0.2 gm/Kg	3	E 4.70±0.01	D 0.12±0.01	F 0.11±0.01	D 7.87±00.03	A 28.00±0.01	C 2.25±0.13	C 31.40±0.59
	Orgazol zinc 0.4 gm*Kg	3	E 4.70±0.01	D 0.12±0.01	F 0.11±0.01	C 7.90±00.001	A 28.00±0.01	D 2.16±0.03	C 30.77±0.48
	Control	3	E 4.70±0.01	D 0.12±0.01	F 0.11±0.01	E 7.83±00.03	A 28.00±0.01	D 2.15±0.01	C 30.40±0.06
2nd week	CA growth 1ml/Kg	3	D 4.87±0.03	E 0.10±0.01	G 0.10±0.01	C 7.90±00.001	A 28.00±0.01	D 2.10±0.01	D 28.13±0.03
	CA growth 2ml/Kg	3	D 4.87±0.03	E 0.10±0.01	G 0.10±0.01	C 7.93±00.03	A 28.00±0.01	D 2.07±0.02	D 27.97±0.07
	Orgazal zinc 0.2 gm/Kg	3	F 4.63±0.03	C 0.13±0.01	D 0.13±0.01	E 7.83±00.03	A 28.00±0.01	B 2.37±0.03	C 32.23±0.27
	Orgazol zinc 0.4 gm*Kg	3	F 4.60±0.01	C 0.13±0.01	E 0.12±0.01	E 7.80±00.001	A 28.00±0.01	B 2.34±0.02	C 31.77±0.49
	Control	3	E 4.70±0.01	D 0.12±0.01	F 0.11±0.01	C 7.90±00.001	A 28.00±0.01	D 2.12±0.01	C 30.17±0.03
3rd week	CA growth 1ml/Kg	3	C 5.03±0.03	E 0.10±0.01	G 0.10±0.01	C 7.90±00.001	A 28.00±0.01	E 2.02±0.01	D 27.47±0.03
	CA growth 2ml/Kg	3	B 5.10±0.01	E 0.09±0.01	A 0.36±0.27	AB 8.10±00.001	A 28.00±0.01	E 1.95±0.03	E 25.27±0.09
	Orgazal zinc 0.2 gm/Kg	3	F 4.63±0.03	C 0.13±0.01	C 0.14±0.01	E 7.83±00.03	A 28.00±0.01	E 2.08±0.00	C 32.67±0.49
	Orgazol zinc 0.4 gm*Kg	3	F 4.57±0.03	B 0.14±0.01	C 0.14±0.01	E 7.80±00.001	A 28.00±0.01	A 2.49±0.01	B 36.20±0.32
	Control	3	E 4.70±0.01	D 0.12±0.01	F 0.11±0.01	C 7.90±00.001	A 28.00±0.01	E 2.13±0.01	C 30.27±0.09
4th week	CA growth 1ml/Kg	3	B 5.10±0.01	E 0.09±0.01	0.10±0.01	B 8.03±00.07	A 28.00±0.01	E 1.93±0.01	F 24.87±0.12
	CA growth 2ml/Kg	3	A 5.27±0.03	F 0.08±0.01	0.08±0.01	A 8.17±00.03	A 28.00±0.01	F 1.85±0.01	G 21.90±0.12
	Orgazal zinc 0.2 gm/Kg	3	F 4.60±0.01	B 0.14±0.01	C 0.14±0.01	G 7.73±00.03	A 28.00±0.01	E 2.08±0.01	C 32.33±0.27
	Orgazol zinc 0.4 gm*Kg	3	G 4.50±0.01	A 0.15±0.01	B 0.15±0.01	F 7.77±00.03	A 28.00±0.01	A 2.49±0.01	A 39.10±0.12
	Control	3	F 4.73±0.03	D 0.12±0.01	F 0.12±0.01	F 7.90±00.001	A 28.00±0.01	D 2.14±0.01	C 30.20±0.06

Means within the same column of different litters are significantly different at (P < 0.05).

Table (7): ***Effect of feeding different growth promoters to seabass and seabream fish on water quality parameters of water at different periods of experiments..***

Species	Treatment	N	DO	Ammonia	Nitrite	pH	Salninity	OM	Sulphate
			Mean ± Standard Error						
Sea bass	CA growth 1ml/Kg	12	B 4.92 ±0.05	E 0.10 ±0.01	G 0.09 ±0.01	B 7.93 ±0.03	A 28.00 ±0.001	F 1.97 ±0.05	D 27.70 ±0.54
	CA growth2ml (Kg	12	A 5.02 ±0.07	E 0.09 ±0.01	G 0.09 ±0.01	A 8.03 ±0.05	A 28.00 ±0.001	G 1.8 7±0.07	F 25.95 ±1.11
	Orgazal zinc0.2 gm/Kg	12	C 4.67 ±0.02	B 0.14 ±0.001	B 0.14 ±0.01	D 7.78 ±0.01	A 28.00 ±0.001	C 2.33 ±0.06	B 31.93 ±0.64
	Orgazol zinc0.4 gm *Kg	12	C 4.60 ±0.03	A 0.15 ±0.01	B 0.14 ±0.01	E 7.68 ±0.07	A 28.00 ±0.001	A 2.47 ±0.09	A 33.93 ±0.97
	Control	12	C 4.70 ±0.00	D 0.12 ±0.001	D 0.12 ±0.001	BC 7.88 ±0.01	A 28.00 ±0.001	D 2.11 ±0.00	B 30.17 ±0.02
Sea bream	CA growth 1ml/Kg	12	B 4.93 ±0.05	E 0.10 ±0.001	F 0.10 ±0.001	B 7.91 ±0.03	A 28.00 ±0.001	E 2.07 ±0.03	C 29.36 ±1.96
	CA growth2ml (Kg	12	B 4.98 ±0.07	E 0.10 ±0.001	A 0.16 ±0.07	B 7.99 ±0.05	A 28.00 ±0.001	E 2.02 ±0.04	E 26.56 ±1.03
	Orgazal zinc0.2 gm/Kg	12	C 4.64 ±0.01	C 0.13 ±0.001	C 0.13 ±0.001	C 7.82 ±0.02	A 28.00 ±0.001	CD 2.20 ±0.05	AB 32.16 ±0.23
	Orgazol zinc0.4 gm *Kg	12	D 4.59 ±0.02	C 0.13 ±0.001	C 0.13 ±0.001	C 7.82 ±0.02	A 28.00 ±0.001	B 2.37 ±0.04	A 34.46 ±1.03
	Control	12	CD 4.71 ±0.01	D 0.12 ±0.001	E 0.11 ±0.001	BC 7.88 ±0.01	A 28.00 ±0.001	D 2.14 ±0.01	B 30.26 ±0.04
	Total	120	4.78 ±0.02	0.12 ±0.001	0.12 ±0.01	7.87 ±0.01	28.00 ±0.001	2.15 ±0.02	30.25 ±0.38

Means within the same column of different litters are significantly different at (P < 0.05).

experiment. Mercury level (Tables, 8, 9) cleared that, the level of mercury increased significantly ($P < 0.05$) from the 1st week to the last week of the experiment and its level in seabream higher than that of seabass. And in orgazol zinc higher than that of CA growth. The cadmium level decreased progressively from the 1st week to the 4th week of the experiment and in orgazol zinc lower than that of CA groth treated group and in seabass higher than that of seabream treated fish. The copper level also increased progressively from the 1st to the 4th week of the experiment. The higher copper level observed in the group treated with orgazol zinc especially the higher doses followed by CA growth and all of them higher than that of the control group.

Data presented in Tables (8, 9 and 10) cleared that the zinc level increased progressively from the 1st to 4th week of the experiment. The zinc level increased with the higher dose of orgzol zinc than the CA growth and in the seabream higher than that of the seabass treated group.

C-Heavy metals parameters:-

Table (8): ***Effect of feeding different growth promoters to seabass fish on heavy metals of water at different periods of experiments.***

Week	Dose	N	Lead	Arsenic	Mercury	Cadmium	Copper	Zinc
			Mean ± Standard Error					
1st week	**CA growth 1ml/Kg**	3	**D 2.16±0.001**	**A 0.01±0.00**	**C 0.46±0.01**	**A 1.38±0.00**	**C 1.71±0.02**	**A 2.21±0.03**
	CA growth 2ml/Kg	3	**D 2.17±0.001**	**A 0.01±0.00**	**C 0.46±0.01**	**A 1.37±0.00**	**C 1.70±0.01**	**AB 2.19±0.01**
	Orgazal zinc 0.2 gm/Kg	3	**D 2.17±0.001**	**A 0.01±0.00**	**C 0.45±0.001**	**A 1.35±0.00**	**C 1.69±0.01**	**B 2.18±0.01**
	Orgazol zinc 0.4 gm*Kg	3	**D 2.17±0.001**	**A 0.01±0.00**	**C 0.45±0.001**	**A 1.38±0.01**	**C 1.70±0.01**	**A 2.20±0.01**
	Control	3	**D 2.17±0.001**	**A 0.01±0.00**	**C 0.46±0.001**	**A 1.37±0.01**	**C 1.70±0.03**	**AB 2.19±0.001**
2nd week	**CA growth 1ml/Kg**	3	**E 2.09±0.01**	**B 0.0001±0.00001**	**D 0.43±0.001**	**A 1.38±0.01**	**D 1.64±0.00**	**B 2.16±0.01**
	CA growth 2ml/Kg	3	**E 2.04±0.02**	**B 0.0001±0.00001**	**D 0.44±0.01**	**A 1.31±0.01**	**D 1.60±0.02**	**B 2.16±0.02**
	Orgazal zinc 0.2 gm/Kg	3	**C 2.25±0.01**	**A 0.02±0.001**	**C 0.48±0.001**	**B 1.22±0.01**	**C 1.74±0.01**	**B 2.17±0.01**
	Orgazol zinc 0.4 gm*Kg	3	**B 2.36±0.07**	**A 0.01±0.0001**	**C 0.49±0.001**	**C 1.14±0.01**	**C 1.76±0.02**	**A 2.22±0.05**
	Control	3	**D 2.17±0.001**	**A 0.01±0.0001**	**C 0.45±0.001**	**A 1.36±0.01**	**CD 1.69±0.02**	**B 2.18±0.001**
3rd week	**CA growth 1ml/Kg**	3	**E 2.03±0.001**	**B 0.001±0.00001**	**C 0.47±0.01**	**A 1.31±0.00**	**E 1.52±0.01**	**B 2.15±0.01**
	CA growth 2ml/Kg	3	**F 1.94±0.01**	**B 0.001±0.00001**	**C 0.49±0.001**	**B 1.23±0.03**	**E 1.50±0.01**	**A 2.20±0.01**
	Orgazal zinc 0.2 gm/Kg	3	**C 2.29±0.01**	**A 0.01±0.001**	**B 0.52±0.01**	**D 1.02±0.01**	**BC 1.77±0.02**	**B 2.17±0.01**
	Orgazol zinc 0.4 gm*Kg	3	**B 2.35±0.01**	**A 0.01±0.001**	**A 0.59±0.001**	**E 0.93±0.01**	**B 1.85±0.02**	**B 2.18±0.02**
	Control	3	**D 2.16±0.01**	**A 0.01±0.001**	**C 0.45±0.001**	**A 1.35±0.00**	**D 1.67±0.03**	**B 2.18±0.01**
4th week	**CA growth 1ml/Kg**	3	**C 2.01±0.01**	**B 0.001±0.00001**	**D 0.43±0.01**	**A 1.30±0.02**	**F 1.44±0.01**	**B 2.15±0.01**
	CA growth 2ml/Kg	3	**F 1.80±0.01**	**B 0.001±0.00001**	**D 0.41±0.001**	**B 1.27±0.02**	**G 1.43±0.02**	**AB 2.19±0.02**
	Orgazal zinc 0.2 gm/Kg	3	**B 2.35±0.01**	**A 0.01±0.0001**	**B 0.54±0.01**	**E 0.91±0.02**	**B 1.82±0.01**	**A 2.23±0.01**
	Orgazol zinc 0.4 gm*Kg	3	**A 2.72±0.33**	**A 0.01±0.0001**	**A 0.62±0.01**	**F 0.73±0.02**	**A 1.93±0.01**	**A 2.20±0.02**
	Control	3	**D 2.16±0.01**	**A 0.01±0.0001**	**C 0.44±0.01**	**A 1.35±0.01**	**D 1.68±0.03**	**2.17±0.02**

Means within the same column of different litters are significantly different at (P < 0.05).

Table (9): ***Effect of feeding different growth promoters to seabream fish on heavy metals of water at different periods of experiments.***

Week	Dose	N	Lead	Arsenic	Mercury	Cadmium	Copper	Zinc
			Mean ± Standard Error					
1st week	CA growth 1ml/Kg	3	B 2.14±0.02	A 0.01±0.0001	A 0.57±0.03	A 1.44±0.02	C 1.67±0.09	D 2.29±0.08
	CA growth2ml (Kg	3	A 2.22±0.05	A 0.01±0.0001	B 0.53±0.02	A 1.46±0.03	C 1.72±0.01	C 2.42±0.05
	Orgazal zinc0.2 gm/Kg	3	B 2.17±0.00	A 0.01±0.0001	B 0.53±0.01	B 1.37±0.02	C 1.71±0.07	C 2.41±0.03
	Orgazol zinc0.4 gm *Kg	3	A 2.21±0.01	A 0.01±0.0001	C 0.49±0.00	A 1.45±0.02	C 1.68±0.02	C 2.41±0.12
	Control	3	A 2.23±0.04	A 0.01±0.0001	B 0.50±0.01	A 1.45±0.01	C 1.69±0.02	A 2.61±0.12
2nd week	CA growth 1ml/Kg	3	C 2.10±0.01	A 0.01±0.0001	B 0.51±0.02	C 1.35±0.02	C 1.76±0.09	D 2.28±0.09
	CA growth2ml (Kg	3	C 2.04±0.02	0.01±0.0001	B 0.52±0.01	C 1.32±0.01	D 1.61±0.02	C 2.43±0.04
	Orgazal zinc0.2 gm/Kg	3	A 2.28±0.03	A 0.01±0.0001	A 0.58±0.001	C 1.32±0.01	C 1.76±0.01	C 2.41±0.00
	Orgazol zinc0.4 gm *Kg	3	A 2.20±0.02	A 0.01±0.0001	A 0.57±0.001	D 1.24±0.02	B 1.82±0.01	B 2.48±0.01
	Control	3	B 2.19±0.01	A 0.01±0.0001	B 0.50±0.01	A 1.41±0.05	C 1.70±0.02	A 2.60±0.12
3rd week	CA growth 1ml/Kg	3	C 2.06±0.01	A 0.01±0.0001	B 0.50±0.01	B 1.37±0.01	C 1.69±0.07	A 2.61±0.40
	CA growth2ml (Kg	3	C 2.01±0.00	A 0.01±0.0001	C 0.49±0.01	B 1.38±0.01	1.51±0.05	C 2.42±0.05
	Orgazal zinc0.2 gm/Kg	3	A 2.29±0.04	A 0.01±0.0001	A 0.59±0.001	E 1.14±0.01	B 1.81±0.00	C 2.43±0.02
	Orgazol zinc0.4 gm *Kg	3	A 2.21±0.03	A 0.01±0.0001	A 0.59±0.001	F 1.10±0.01	A 1.89±0.00	B 2.49±0.01
	Control	3	A 2.19±0.01	A 0.01±0.0001	C 0.49±0.001	A 1.41±0.00	C 1.69±0.02	B 2.38±0.01
4th week	CA growth 1ml/Kg	3	C 2.03±0.01	A 0.01±0.0001	C 0.48±0.001	C 1.34±0.03	C 1.68±0.07	B 2.49±0.31
	CA growth2ml (Kg	3	D 1.96±0.01	A 0.01±0.0001	C 0.47±0.001	B 1.39±0.01	E 1.49±0.04	B 2.38±0.05
	Orgazal zinc0.2 gm/Kg	3	A 2.27±0.04	A 0.01±0.0001	A 0.58±0.001	G 1.01±0.00	A 1.93±0.01	B 2.39±0.04
	Orgazol zinc0.4 gm *Kg	3	B 2.13±0.01	A 0.01±0.0001	A 0.60±0.001	H 0.98±0.00	A 1.93±0.01	B 2.48±0.02
	Control	3	C 2.09±0.01	A 0.01±0.0001	C 0.49±0.001	AB 1.40±0.01	D 1.64±0.02	B 2.38±0.02

Means within the same column of different litters are significantly different at (P < 0.05).

Table (10): ***Effect of feeding different growth promoters to seabass and seabream fish on heavy metal level of water at different periods of experiments..***

Species	Treatment	N	Lead	Arsenic	Mercury	Cadmium	Copper	Zinc
			Mean ± Standard Error					
Sea bass	CA growth 1ml/Kg	12	E 2.07±0.02	B 0.001±0.00001	C 0.45±0.01	A 1.34±0.01	C 1.58±0.03	B 2.17±0.01
	CA growth2ml (Kg	12	E 1.99±0.04	B 0.001±0.00001	C 0.45±0.01	B 1.29±0.02	1.56±0.03	B 2.19±0.01
	Orgazal zinc0.2 gm/Kg	12	B 2.27±0.02	A 0.01±0.001	BC 0.50±0.01	C 1.12±0.05	B 1.75±0.01	B 2.19±0.01
	Orgazol zinc0.4 gm *Kg	12	A 2.40±0.09	A 0.01±0.001	A 0.54±0.02	C 1.05±0.07	A 1.81±0.03	B 2.20±0.01
	Control	12	C 2.16±0.00	A 0.01±0.001	C 0.45±0.00	A 1.36±0.00	B 1.68±0.01	B 2.18±0.01
Sea bream	CA growth 1ml/Kg	12	D 2.08±0.01	B 0.001±0.00001	A 0.52±0.01	A 1.37±0.01	B 1.70±0.04	A 2.42±0.12
	CA growth2ml (Kg	12	D 2.06±0.03	B 0.001±0.00001	BC 0.50±0.01	A 1.39±0.02	C 1.58±0.03	A 2.41±0.02
	Orgazal zinc0.2 gm/Kg	12	B 2.25±0.02	A 0.01±0.001	A 0.57±0.01	B 1.21±0.04	A 1.80±0.03	A 2.41±0.01
	Orgazol zinc0.4 gm *Kg	12	C 2.19±0.01	A 0.01±0.001	A 0.56±0.01	B 1.19±0.05	A 1.83±0.03	A 2.47±0.03
	Control	12	C 2.18±0.02	A 0.01±0.001	C 0.49±0.00	A 1.42±0.01	B 1.68±0.01	A 2.49±0.05
	Total	120	2.16±0.02	0.01±0.001	0.50±0.01	1.27±0.02	1.70±0.01	2.31±0.02

Means within the same column of different litters are significantly different at (P < 0.05).

DISCUSSION

Our results indicated that, the body weight of fish of a higher level in seabass fish than that of seabream and in the group treated with orgazol zinc than that of CA growth. Also, our results indicated that, the body weight gain decreased progressively toward the end of the experiment, the higher body weight gain observed in seabass than that of seabream fish.and the weight gain in the groups treated with orgazol zinc. The body

weight gain of a higher level in orgazol zinc than that of the CA growth.

The results of food conversion ratio cleared that, the food conversion ratio improved in seabream than that of seabass fish and in CA growth improved than that of orgazol zinc.

This results attributed to the constituents of CA growth (Calcium, magnesium, vitamins and organic matters) improved the feed utilization, body weight, body weight gain and feed conversion and also economic returns obtained from fish. This results agreed with those of (*Liebert, 2009)* where they indicated that the addition of feed additives in fish diet improve the body weight, body weight gain and feed conversion of fish. Also, the addition of feed additives in fish diet improve the immunity of fish against different diseases so it will improve the body weight and body weight gain with improvement of feed conversion and utilization *(Eduardo et al., 2009).* Our results indicated that, the level of dissolved oxygen increased toward the end of the experiment. Also, the results indicated that, the level of dissolved oxygen increase in the group treated with CA growth especially with the higher dose (2ml/kg) followed by control group and the minimum level of dissolved oxygen observed in the orgasol zinc, especially at a higher dose of orgzol zinc while, the results of ammonia level indicated that, the level of ammonia increased toward the end of the experiment and the higher level of ammonia increased with orgasol zinc especially the higher dose of orgazol zinc followed by the control group and the minimum level of ammonia observed in CA growth especially at the higher dose of CA growth zinc. The results also indicated that, the pH level increased with the higher dose of CA growth followed by control group and decreased with the higher dose of orgazol zinc. The results of salinity level indicated that, the salinity level not changed at the all periods of the experiment and its level not changed among all treatment level. The organic matter level increased progressively from the 1st week to the last week of the experiment. The higher level of organic matter observed in the group treated with Orgazol zinc especially the higher dose of orgazol zinc followed by control group and the lower organic matter level observed in the group treated with CA growth. The sulphate level increased progressively from the 1st week of the experiment to the 4th week and the higher level of sulphate observed in the orgazol zinc especially at a higher doses followed by control group and the lower sulphate level observed in CA treated groups, especially the lower dose of the CA growth.

The results indicated that, the effect of CA growth and Orgazal zinc in its effect on water quality of that the fish live in it in seabream is higher than that of seabass as it affect DO, ammonia , nitrite, pH , salinity, organic matter and sulphate higher than that of seabass. This trsults attributed to the seabass have the ability to convert and absorb the vitamins, minerals and enzymes that found in CA growth and orgazol zinc than that of seabream and the residue of the feed additives causes increasing the level of DO, ammonia, nitrite, pH, salinity, organic matter and sulphate and increasing the growth of microflora and funa in water that live in it the seabream and decreased in water live in it seabass. Our results agreed with those of *Hirata and Niiro (2001)* where they reported that, the addition of the feed additives to the diet of the fish facilitated the growth of fauna

and flora that is one of the methods that achieved sustainable aquaculture and their growth that, commonly depended upon the season of production and increased in summer seasons than in winter seasons and causes increasing of organic matters and trace elements in water, and the intensive system the growth of funa and flora in it is decreased due to over-stocking rate of the fish.

The results of the effect of feeding different growth promoters to seabream fish on heavy metals parameters at different periods of experiments, indicated that, the level of lead increase in the group treated with orgazol zinc than the CA growth treated fish and the lead increased progressively from the 1st week to the last week of the experiment and in seabass higher than that of seabream. The arsenic level increased in seabream treated fish than seabass treated fish and its level not changed from the 1st week to the 4th week of the experiment. While, the results of mercury level cleared that, the level of mercury increased from the 1st week to the last week of the experiment and its level in seabream higher than that of seabass. And in orgazol zinc higher than that of CA growth. While, the cadmium level decreased progressively from the 1st week to the 4th week of the experiment and in orgazol zinc lower than that of CA growth treated group and in seabass higher than that of seabream treated fish. Also, the copper level increased progressively from the 1st to the 4th week of the experiment. The higher copper level observed in the group treated with orgazol zinc especially the higher doses followed by CA growth and all of them higher than that of the control group. While, the zinc level increased progressively from the 1st to 4th week of the experiment, and the zinc level increased with the higher dose of orgzol zinc than the CA growth and in the seabream higher than that of the seabass treated group. Our results concluded that the orgazol zinc increased the level of lead, arsenic, mercury, cadmium, copper and zinc than that of CA treated fish and its effect higher in seabream than that of seabass treated fish. Also, the results showed that the metal concentrations in fish diet of cultured Seabass and Seabream were closely associated with metal level and the level of contamination in water and this may be attributed to the fact that if an environment receives foreign pollutants (e.g. heavy metals), the organisms living in it could take up the pollutants from the water and/or food and concentrate it in their bodies and by feed the fish on this organisms causing increasing the level of heavy metals in fish body. (*Eiman and Zamzam, 1996)*. The heavy metal concentrations in Seabream gills and musculature were higher than that of Seabass and this may be attributed to the levels of heavy metals found in fish species generally vary depend on the various factors such as fish species, size, age, the region where fish was caught, seasonal changes, analytical methodology etc and due to the seabream of larger size and of high fat level than the seabass so the concentration and storage of the heavy metals in seabream higher than that of the seabass (*Minganti et al., 2010*). Our results concluded that, the addition of feed additives to the fish diet improve the body weight of the fish, food conversion and the economic efficiency of fish production farms , with improvement of fish immunity against fish diseases so it will improve the productive efficiency of fish production farms. This results agreed with those of (*Andrrews et al., 2013)* where they concluded that the addition of feed additives to the fish diet improve the

immunity, weight gain and improve the economic return of fish production farms.

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بعض الدراسات على محفزات المناعة فى أسماك المياه المالحة

رياض خليل – طلعت طلعت سعد – هانى على ناصف

قسم أمراض الدواجن والاسماك – طب بيطرى اسكندرية

زادت أهمية الاسماك فى الآونة الاخيرة فى حل الازمة الغذائية التى تعانى منها مصر ولذلك تم إستخدام بعض إضافات الاعلاف التى يدخل فيها الاملاح مثل الكالسيوم و الزنك . أجريت هذه الدراسة على عدد ٤٠٠ سمكة من أسماك الدنيس و القاروص و التى تتراوح بين (٥٠ جرام) ٢٠٠ سمكة دنيس و الاخرى ٢٠٠ سمكة قاروص حيث تم تجميعها من مزارع قطاع خاص مرباه غى أقفاص و تم نقلها الى معمل الاسماك بكلية الطب البيطرى – جامعة الاسكندرية فى أكياس تحتوى على (٣/٢) هواء .

وخلصت الدراسة إلى النتائج الآتية:- ١-أن إضافات الاعلاف مهمة جدا فى صناعة الاسماك ورفع الحالة المعتوية لها ضد الامراض المختلفة. ٢-أوضحت الدراسة أن الـ CA growth و الاورجازول زنك فى الدنيس يؤديان الى زيادة فى الاكسجين الذائب فى المياه ، و مستوى الاس الهيدروجينى ، و الملوحة والمواد العضوية و السلفات عنها فى المياه المربى فيها أسماك القاروص.

٣-أن الارجازول زنك يؤدى الى زيادة نسب تشمل الرصاص ، الزنك ، الزئبق ، و النحاس فى المياه المربى غيه الفاروص عن الدنيس. ٦-أن معدل الزيادة فى وزن الفاروص أعلى من معدل الزيادة فى وزن الدنيس عند إضافة الارجازول زنك عن الـ CA growth. ٧-أن معدل الحويل العذائى والعائد الاقتصادى يتحسن فى الفاروص و الدنيس عن المجكوعات الضابطة عند إضافة الارجازل زنك ، الـ CA growth.