Dept. of Animal Prduction
Fac. of Agriculture – Assiut University Assiut -Egypt

INFLUENCE OF DIETARY ZINC AND VITAMIN A LEVELS ON GROWTH PERFORMANCE, BLOOD CONSTITUENTS AND IMMUNO COMPETENCE OF NILE TILAPIA, *OREOCHROMIS NILOTICUS* UNDER UPPER EGYPT CONDITIONS

(With 4 Tables and 2 Figures)

By

S. Y. HUSSEIN and M. A. KOBEISY

(Received at 28/6/2001)

تأثير مستويات الزنك وفيتامين أ في العليقه على النمو ومكونات الدم والمناعة للبلطي النيلي تحت ظروف مصر العليا

سمير يوسف حسين ، مصطفي أحمد قبيصي

استخدمت في هذه الدراسة عدد ٢٧٠ سمكة من البلطي النيلي لدراسة تأثير الزنك وفيتامين أ على النمو ومركبات الدم والعوامل المناعية. ولقد وزعت الأسماك عشوائيا على ثلاثة مجاميع بكل منها ٩٠ سمكة وأعطيت هذه المجموعات التركيزات الآتية من فيتامين أ : صفر ، ٤٠٠٠ ، ٦٠٠٠ وحدة دولية / كجم عليقة وكل مجموعة من هذه المجموعات الثلاثة قسمت إلى ثلاثة مجموعات أدنى بكل منها ٣٠ سمكة واعطيت أحد مستويات الزنك الأتية: صفر ، ٣٠ ، ٤٠ مجم / كجم عليقة. تم تسجيل وزن الجسم والطول كل اسبوعين لمدة ١٦ أسبوع. وتم تجميع عينات الدم من كل الأسماك لتقدير الهيموجلوبين والهيماتوكريت ومحتوى السيرم من الجلسريدات الثلاثية والكوليستيرول الكلى والبروتين الكلى والألبيومين والجلوبيولين وانزيمات الكبد (ALT&AST) وانزيم الفوسفاتيز القاعدى وهرمونات الغدة الدرقية (T3 & T4) ، LDI.&HDL والمكونات المناعية (الفاجلوبيولين وبيتاجلوبيولين وجاماجلوبيولين). وفي نهاية التجربة ذبحت كل الأسماك وتم ازالة الأحشاء ووزنها وحللت المادة الجافة والبروتين الخام والدهن الخام والرماد في العضلات الظهرية. وأظهرت نتائج هذه الدراسة أن فيتامين أ والزنك ادى إلى تحسين في وزن وطول الجسم. وأن معامل الطحال الذى تأثر بإضافة الزنك وفيتامين أ (٤٠٠٠ وحدة دولية) ولقد أدت اضافة الزنك (٣٠ مجم) وفيتامين أ (٤٠٠٠ وحدو دولية) إلى زيادة بروتين العضلات. ولقد أوضحت الدراسة ان اضافة الزنك وفيتامين أ أدى إلى تحسين الهيموجلوبين والهيماتوكريت ومحتوى السيرم من البروتين والجلوكوز وهرمونات الغدة الدرقية بينما لايوجد فروق معنوية بين المجموعات في الجلسريدات الثلاثية. وان مستوى ٣٠مجم زنك في العليقة ادى إلى نقص في مستوى انزيم الكبد AST بينما المستوى ٦٠٠٠ وحدة دولية فيتامين أ أدى إلى زيادة كل من أنزيمات الكبد AST & ALT ولقد وجد أن

اضافة الزنك ليس له تأثير على سيرم الكولسترول بينما الزنك مع فيتامين أ (٦٠٠٠ وحدة دولية) أدى إلى نقص في تركيز الكوليسترول الكلى في السيرم بالإضافة إلى ذلك فان الزنك مع فيتامين أ أدى إلى زيادة HDL بينما أدى إلى نقص في LDL وان إضافة الزنك مع فيتامين أ في العليقة أدى إلى زيادة ألفا وبيتا وجاما جلوبيولين .ولقد وجد أن مجموعات الأسماك التي أعطيت مستويات ٣٠ مجم زنك أو ٣٠ مجم زنك مع ٤٠٠٠ وحدة دولية فيتامين أ قد استجابت لإعطاء بيض بعد أسبوعين من المعاملة .

SUMMARY

A total number of two hundred seventy Nile fish, *Oreochromis niloticus* were used to study the effect of zinc and vitamin A on growth performance, blood constituents and immuno competence. The fish were randomly divided into three treatment groups of 90 fish each. These groups received vitamin A at a concentration of 0, 4000 and 6000 IU/kg DM diet. Each group of the three main groups was divided into three subgroups (30 fish) and received zinc at a concentration of 0, 30 and 40 mg/kg DM diet. Body weight and length were recorded biweekly allover the experimental period (16 weeks). Blood samples from each fish were collected to determine each of hemoglobin (Hb), packed cell volume (PCV%), serum glucose, triglycerides, total cholesterol, total protein, albumin, globulin, AST, ALT, alkaline phosphatase, triiodothyronin (T3), thyroxin (T4), high density lipoprotein (HDL), low density lipoprotein (LDL) and immuno compentence (α , β and γ - globulin). All fish were scarified and visceral organs were weighed. Dorsal muscles were analyzed for dry matter (DM), crude protein (CP), crude fat (CF) and ash percentages. Dietary vitamin A or zinc improved body gain and length increment, Spleen somatic index (SSI) was significantly affected by dietary zinc with vitamin A (4000 IU). Muscle protein percentage was found to be higher in fish received 30 mg Zn and 4000 IU vitamin A.Dietary zinc and vitamin A improved hemoglobin, PCV%, serum proteins, glucose and thyriod hormones concentrations. However, no significant differences in serum triglycerides were observed among all the groups.Dietary Zn (30 mg) alone decreased serum AST level, while the combination of zinc (30 mg) and vitamin A (6000 IU) significantly increased ($P<0.05$) both serum AST and ALT levels. Zinc had no significant effect on serum cholesterol, however, zinc and vitamin A (6000 IU) reduced its concentration. Also, dietary zinc and vitamin A resulted in a significant increase ($P< 0.05$) in the serum HDL and decrease ($P<0.05$) in the LDL level. Moreover, serum α , β and γ – globulin concentrations were significantly increased with Zn and vitamin A supplementation. In addition, the fish groups either fed 30 mg

Zn alone or 30 mg Zn with 4000 IU vitamin A / kg diet responded early for spawning after two weeks of treatment.

Key Words:Zinc, vitamin A, growth,blood constituents, immuno competence and O.niloticus.

INTRODUCTION

Zinc is obviously involved in metaloenzymes, i.e. carbonic anhydrase, alkaline phosphatase, various dehydrogenases, pancreatic carboxypeptidases A and B, pyridoxal phosphokinase and DNA polymerases (Larvor 1983). For this reason, in Zn-deficient animals including fish the corresponding enzymatic activities are generally decreased (Kirchgessner et al., 1976), resulted in reducing growth in rainbow trout (Ogino and Yang, 1978), common carp (Ogino and Yang, 1979) and channel catfish (Gatlin and Wilson, 1983), high incidence of cataracts in rainbow trout (Ketola, 1979), and high mortality in *Oreochromis niloticus* (Eid and Ghonim 1994).

In fact, zinc participates in DNA synthesis through two zinc-dependent enzymes, terminal deoxynucleotidyl transferase and DNA polymerases. Accordingly decreased protein synthesis certainly explains the decrease growth rate observed in zinc deficiency and more specifically the decrease in collagen synthesis which results in slower wound healing (Wacker, 1976).

In addition, zinc is competed with plasma albumin (Larvor, 1983) and might have a protective function toward the insulin molecule (Kirchgessner et al., 1976). Also, zinc is necessary to maintain normal concentrations of vitamin A in plasma and may be required for mobilization of vitamin A from the liver (Harper et al., 1979 and Berzin, 1988). Also, zinc activated immunity protection (Gross et. al. 1979 and Bires et. al. 1993). In this respect, vitamin A is an essential nutrient for all animal species including fish (Haiqi He et. al., 1992). Moreover, Hilton (1983) and Takeuchi et. al. (1998) showed that vitamin A improved growth of the fish. Also, Thompson et. al. (1995) and David et. al. (2001) in their experiments on fish stated that dietary vitamin A supplementation had immunostimulatory agents which stimulates immunoglobulin synthesis particularly γ – globulin. Due to the lack in the literature explaining the effect of combination of vitamin A and zinc in the diet on fish performance, this study was run to investigate the interaction among the combination of different dietary zinc and vitamin A levels and to determine their possible effects on growth performance,

blood constituents and immuno competence of Nile tilapia, *Oreochromis niloticus* under Upper Egypt conditions.

MATERIAL and METHODS

Fish and experimental design:

Two hundred seventy fish, *Oreochromis niloticus* were collected from the ponds of fish farm belongs to Faculty of Agriculture, Assiut University and transferred to the fish laboratory. All the fish appeared to be clinically normal and in a good health status. Average body weight and body length were 17.19± 1.25 g and 9.88 ± 0.60 cm, respectively. They were reared in aquaria 180 x 60 x 70 cm. with water flow of 6 L per hour. Water temperature during the experiment was recorded three times daily, its average was 26.04 ± 1.71 °C. The water zinc concentration was 9.00±2.29 ppb, measured by GBC model 300 atomic absorption spectrophotometer. After two weeks adaptation period, the fish were randomly divided into three experimental groups of 90 fish each. These groups received vitamin A at concentrations of 0, 0.54g (4000 IU) or 0.81g (6000 IU) per Kg DM diet. Each group of the three main groups was divided into three subgroups, each of 30 fish and received zinc at concentrations of 0, 30 or 40 mg/kg DM diet, respectively. The treated groups were distributed as follows:

Treatment	T1	T2	T3	T4	T5	T6	T7	T8	T9
Zn (mg/kg DM diet)	0.0	30	40	0.0	30	40	0.0	30	40
Vitamin A (IU/kg DM diet)	0.0	0.0	0.0	4000	4000	4000	6000	6000	6000

The fish were fed twice daily at a rate of 3% of wet body weight; the amounts of feed were readjusted biweekly according weight of the fish in each treatment.The basal diet was formulated according to Eid and El-Gamal (1996). The levels of zinc and vitamin A used in the present experiment were according National Research Council (NRC, 1983) and Eid and Gohenim (1994), respectively.

Zinc sulphate was used as a source of zinc while vitamin A was purchased as powder from Sigma Inc. Rations were prepared by mixing the ingredients with zinc sulphate and/or vitamin A as powder. After that the rations were pelleted by using suitable level of molasses and water.

Growth performance:

Body weight and length of *O. niloticus* were recorded biweekly along the experimental period (16 weeks). Then body weight gain, body length increment and feed conversion were calculated.

Blood samples and analyses:

Individual blood samples were collected at the end of the experiment by severing the caudal peduncle. Adequate amounts of whole blood in small plastic vials containing heparine as anticoagulent were used for determination of hemoglobin (Hb) by using suitable Kits (Diamond Diagnostics, Egypt), and the hematocrit (PCV%) was determined after Stoskopf (1993). Serum was separated by centrifugtion at 3000 r.p.m. for 15 min. for the rest of the blood samples and then kept in glass vials at - 20 °C until biochemical analyses. Serum glucose (mg/dl), triglycerides concentrations (mg/dl) and total cholesterol (mg/dl) were determined colorimetrically using commercial test kits supplied by Biocon (Germany). Serum total protein (g/dl), albumin (g/dl), aspartic amino transferase (AST, IU), alanine amino transferase (ALT, IU) and alkaline phosphatase concentrations were measured using kits supplied by Diamond, Diagnostics, Egypt. Serum globulin (g/dl) was calculated by difference between serum total protein and albumin concentrations. Serum triiodothyronine (T_3) and thyroxine (T_4) were analyzed using kits supplied by Chino (California, USA). Serum high-density lipoprotein (HDL) and low-density lipoprotein (LDL) levels were determined using test kits (Biocentric, France). Serum zinc concentrations were determined according to Zilva (1973). Serum β-cartone and vitamin A concentrations were estimated according to Carr and Prica (1926). Serum protein fractions (α , β and γ- globulin) as immune indices were separated by electrophoretic technique. The serum proteins were electrophorised according to the procedure mentioned in the Helena Laboratories Publications (1984). In such a procedure, Titan III cellulose acetate plates, Electra HR buffer and Ponceau S stain were used. The electrophoretic protein patterns were scanned and graphed by Auto Scanner Flur-Vis to reveal the densitometric tracinge then serum protein fractions (α , β and γ- globulin)were identified.

All fish were scarified and soon eviscerated. The liver, spleen and gonads were weighed at once. The liver, spleen and gonads somatic indices were calculated as percentages of body weights.

Dorsal muscle samples from individual fish were taken to determine the percentages of dry matter, crude protein, fat and ash according to AOAC (1984). Also, zinc concentration of whole body in experimental fish was detected after digestion in nitric acid and measured using GBC model 300 atomic absorption spectrophotometer according to APHA (1985).

Statistical analysis:

The data were analyzed using the GLM procedures of SAS (1987). Treatment effects were also examined by one-way ANOVA (Steel and Torrie, 1980).

RESULTS and DISCUSSION

1. Effect of dietary zinc and vitamin A on growth performance of *O.niloticus:*

Fish fed diets containing Zn (30 and 40 mg/kg DM diet) had higher ($P<0.05$) body gain (g/fish) and length increment (mm) (T_2 and T_3 vs. T_1, Table 1). Dietary vitamin A improved body gain and length increment (T_4 and T_7 vs. T_1, Table 1). The highest value of body gain was recorded with the group fed on diet supplemented with 40 mg zinc and 6000 IU vitamin A (Table 1). Similary, Eid and Ghonim (1994) studied the effect of dietary Zn (0 to 100 mg/kg diet) on fingerling of *Oreochromis niloticus* for 70 days and found that levels of supplemental zinc above 30 mg/kg improved growth. These results were supported by those of Tan and Kangsen (2001) on *Haliotis discus* hannai. Takeuchi et al. (1998) found that growth of fish was improved by feeding vitamin A compounds. The obtained results on vitamin A are coincided with those of Hilton (1983) who showed that vitamin A had a positive effect on growth of rainbow trout.

In addition, zinc deficiency reduced growth in rainbow trout (Ogino and Yang, 1978), common carp (Ogino and Yang, 1979) and channel catfish (Gatlin and Wilson, 1983). Feed conversion (g feed / g gain) was improved ($P<0.05$) by either zinc supplementation (T_2 and T_3 vs. T_1, Table 1) or zinc with vitamin A at a level of 4000 IU.

Dietary vitamin A at a level of 6000 IU, recorded the most improvement among treatments particularly with zinc (T_8 and T_9 vs. T_1 to T_7, Table 1). Similarly, Hoza (1991) found that feed and nutrient utilization were improved with all levels of Zn supplementation (10, 20 and 30 ppm) in tilapia. On the other hand, Gatlin et al. (1989) found that feed efficiency values of catfish fed 20, 100 or 200 mg Zn/kg diet were not affected by these zinc levels. Also, Gatlin and Wilson (1984) stated that feed utilization in catfish did not affect by 150 mg Zn/kg diet. However, Eid and Ghonim (1994) reported that 30 mg Zn/kg dry diet is the minimum zinc requirement for normal feed efficiency of Nile tilapia fingerlings. However, Thodesen et al. (2001) found that changes of

growth in different fish species due to zinc supplementation might be related to genetic variation in zinc absorption.

Hepatopsomatic (HSI), male gonadosomatic (mGSI) and female gonadosomatic (fGSI) indices were not significantly ($P > 0.05$) affected by either zinc or vitamin A alone or even when combined together (Table 1). Similar results were obtained by Edwards and Brown (1966) who found no significant difference in H S I of rainbow trout exposed to 0.6–2.0 mg zinc / L for four months. Spleen somatic index (SSI) was significantly affected by zinc and vitamin A (4000 IU supplementation, Table 1). The reduction of spleen weight indicated that the experimental treatments did not exert any stress on the fish.

2- Effect of dietary zinc and vitamin A on body composition of *O.niloticus:*

Muscle composition of *O. niloticus* indicated no significant ($P > 0.05$) differences among treatments in both dry matter and ash percentages (Table 2). While, crude protein percentage tended to be higher, particularly in muscles of fish in groups (T3, 30 mg zinc /kg diet) and T4 (vitamin A 4000 IU), such increase was in account of a decrease in fat percentages in muscles of both groups (Table 2). In fact, high growth rate and/or protein synthesis in zinc-treated fish may be due to participation of zinc in DNA synthesis through two zinc-dependent enzymes, terminal deoxynucleotidyl transferase and DNA polymerases (Larvor, 1983). Decreased protein synthesis certainly explains the growth retardation observed in zinc deficiency (Wacker, 1976). In addition, Zn^{+2} ions might have a protective function toward the insulin molecule. Insulin and proinsulin aggregate soluble polymers by binding zinc atoms (crystalline insulin contatins 0.5% Zn). For this reason, zinc-depleted rats have a lower tolerance to intraperitoneal glucose than pair-fed animals (Kirchgessner et al. 1976). Insulin treatment increased body weight of rainbow trout (Ablett et al., 1981), eel (Huang et al. 1999) and tilapia (Al-Salahy and Hussein, 1995; Chen et al., 2000 and Silverstein, et al. 2000). Furthermore, insulin may enhance the incorporation of amino acids into protein in the absence of growth hormone, but growth hormone has no such anabolic effect in absence of insulin (Krahl, 1961; Al-Salahy, 1990 and Huang, et al. 1999).

The significant ($P < 0.05$) decrease of muscle crude fat in fish groups treated either with zinc or a combination of zinc and vitamin A (Table 2) may be due to the decrease of serum T. cholesterol (Table 3) recorded in these groups.

The significant interaction between dietary zinc and vitamin A may be related to the necessity of zinc to maintain normal concentrations of vitamin A in serum (Table 3). These results are coincided with the findings of Harper et al., (1979) and Berzin (1988) and to the anabolic effects, cell differentation and bone growth as reported by Moore (1957). Furthermore, zinc concentrations in whole body were significantly ($P< 0.05$) influenced by dietary zinc level (Table 2). Similar results were found by Hardy and Shearer (1985) ;Wekell and Shearer (1986) and Maage and Julshamn (1993). Eid and Ghonim (1994) stated that whole body zinc concentration was significantly correlated with dietary zinc levels.

3. Effect of dietary zinc and vitamin A on some blood constituents in *O. niloticus*:

Dietary zinc, particularly high level, significantly increased ($P<0.05$) hemoglobin and PCV%. Similar improvement was noticed with vitamin A supplementation. The most beneficial effect was recorded at a level of 30 mg Zn with 6000 IU vitamin A (Table 3). However, Eid and Ghonim (1994) found strong negative correlation between dietary zinc and whole-body iron. This may be due to the opposite absorption of these elements to each other (Settlemire and Marlron, 1967 and Davis, 1980). Contrary, Maage and Julshamn (1993) showed that there were no significant differences in iron levels for *Salmo salar* fed dietary zinc at 0,10,20,40 and 80 mg/kg for 8 weeks. Also, Gatlin et al. (1989) found that PCV% and hemoglobin decreased with the increase of dietary zinc level (20, 100 and 200 mg/kg diet). In this respect, Santos, et al. (2000) reported that the high dose of zinc led to reduction in oxygen consumption and food ingestion. However, the improvement of Hb and PCV% with dietary zinc in the present study may be due to the implication of the used levels within the normal requirement (Gatlin, et al. 1989;Hoza, 1991 and Eid and Ghonim, 1994).

Dietary zinc (T_2 and T_3) or vitamin A (T_4 and T_7) significantly increased ($P<0.05$) serum protein and albumin concentrations. Fish received both vitamin A and zinc (T_5, T_6, T_8 and T_9) had higher values of total protein compared with control group (T_1, Table 3). This result may be related to that zinc dependent enzymes stimulate protein synthesis (Wacker, 1976). Similarly dietary zinc increased serum proteins in different animals as recorded by Bires et al. (1993); Daghash and Mousa (1999); Eldeeb and Afifi (2000) and Shetaewi (2000) in dairy cows, buffalo calves and rabbits, respectively.Also, El-Masry and Habeeb (1989) and Hegazy and Adachi (2000) refered this effect to the

elevation of anabolic hormone secretion e.g. insulin-like growth factor which increase amino acids uptake and protein synthesis. Otherwise, this effect of zinc may be due to decrease in the catabolic hormones such as glucocorticoids and catechalamine (Alvarez and Johnson, 1973).

Serum triglycerides showed no significant differences among treatments (Table 3). Serum glucose concentration was significantly higher in treated fish (T_2 to T_9) than in control (T_1). However, no significant differences among treated groups (T_2 to T_9) were observed (Table 3).

Zinc and vitamin A had significant effect on serum alkaline phosphatase (APase) concentration, particularly at the level of 30 mg zinc and 6000 IU vitamin A (Table 3). These results are coincided with the findings of Lan et al. (1995), who stated that (APase) significantly increased at 100 µg Zn / l in *Chrysophrys major*. Also, Tan and Kangsen (2001) showed that the increase of dietary zinc led to significant ($P<0.01$) increase in (APase) of Haliotisdiscus hannai.

Dietary zinc and vitamin A (6000 IU) reduced total cholesterol concentration (Table 3), however, zinc alone had no significant effect. Similarly, Shetaewi (2000) found that cholesterol in serum was not affected by zinc supplementation in rabbits. This decrease in T. cholesterol is related to the change of muscle crude fat in the treated groups either with zinc alone or its combination with vitamin A (table 2). Daghash and Mousa (1999) found an increase in serum cholesterol and triglycerides in buffalo calves treated with zinc. Similarly, Eldeeb and Afifi (2000) found an increase in serum cholesterol in rabbits fed either 100 or 200 mg zn/kg diet.

Zinc supplementation (30 mg) decreased significantly ($P<0.05$) serum AST level, while its supplementation with 6000 IU vitamin A increase significantly ($P<0.05$) serum AST and ALT (T_9, Table 3). The exact biochemical mechanism is not known. However, both enzymes, AST and ALT, are very important in glucose synthesis from noncarbohydrate metabolitie sources (Harper et al., 1979). It is clear that high AST and ALT may increase protein catabolism to synthesize glucose. High glucose level (85.11 mg/dl) of T_9 may support this view (Table 3).

Thyroid hormones (T_3 and T_4) secretion were improved significantly ($P<0.05$) in response to dietary vitamin A and zinc supplementations (Table 3). The mechanism of thyroid hormone stimulation is not known. However, this explain, at least partly, the higher growth rate of fish received Zn and vitamin A (Table 1). Since

the chief effect of T_3 and T_4 are stimulation of protein, fat and carbohydrate metabolism and growth and development of the body (Kutsky, 1981). These results are coincided with the findings of Eder and Kirchgessner (1996) who reported hypothyrodism in rats deficient in zinc i.e reduction in T_3 and T_4 levels. Also, Miyamoto et al. (1991) showed that zinc is involved in T3 binding to its nuclear receptor. Moreover, thyroid hormones (T3 and T4) increased the rate of cholesterol catabolism by the liver (Kaneko, 1989).

The dietary zinc and vitamin A combination significantly increased ($P<0.05$) high-density lipoprotein (HDL), while decreased ($P<0.05$) low-density lipoprotein (LDL) (Fig.1). These effects might be attributed to decline in serum total cholesterol (Table 3). Similar results were found by Yousef et al., (1996). In this respect, Choubert et al. (1991) found the distribution of lipoprotein fractions in immature rainbow trout were 28.3% LDL, 61.0% HDL and 10.6%. VHDL.

Table (3) showed that serum zinc concentration significantly increased ($P<0.05$) in Zn-treated groups in comparison to control. These results are in agreement with the findings of Gatlin and Wilson (1983) and Eid and Ghonim (1994). Serum β-carotone and vitamin A are positively corrlated with dietary Vitamin A levels (Table 3). Similarly, Thompson et al. (1995) found that rainbow trout fed vitamin A at a level of 18 mg/kg dry diet for four months increased serum vitamin A concentration (32–40 vs. 4 µg/dl). However, Borel et al. (1998) concluded that plasma β carotene was dramatically influenced by triglyceride chain – length.

4- Effect of dietary zinc and / or vitamin A on immuno competence:

Results in Table 4 and figure 2 show that both dietary zinc and zinc with vitamin A significantly increased ($P< 0.01$) serum globulin and its fractions (α , β and γ-globulin). Such improvement in serum globulins and its fractions (α , β and γ-globulins) confirm the results reported by Thompson et. al. (1995). Similary David et al. (2001) said that dietary vitamin A had immunostimulatory agents in the fish. In addition, Bires et al. (1993) and Kegley and Spear (1994) found that dietary vitamin A and zinc increased ($P<0.05$) γ – globulin in the fish. Also, Thompson, et al. (1995) stated that vitamin A supplementation stimulates immunoglobulin synthesis in the fish.

5- Effect of dietary zinc and / or vitamin A on spawning:

The fish groups treated with 30 mg Zn/kg diet (T_2) and 30 mg Zn + 4000 IU vitamin A (T_5) responded to spawning after two weeks of the treatments and fertilized eggs together with larvae could be obtained

from their buccal cavity. This may be due to the importance of vitamin A for egg maturation. Ando and Hatano (1986) reported that carotenoids are associated with the egg yolk protein "lipovitellin" in salmon. These findings are in harmony with the findings of Sivtseva, (1982) who showed that during sexual maturation of trout, 18% of the total body carotenoids may be present in their eggs. In addition, Czeczuga (1975) found that dietary carotenoids are important in spermatogenesis in rainbow trout.

However, the role of zinc in biological systems in fish is mainly limited (Watanabe et al., 1997).

Conclusion:

It could be concluded that, combination of dietary zinc and vitamin A may improve growth performance, body composition and blood constituents of *O. niloticus*. Also, zinc with vitamin A may modulate blood constituents and some aspects of immune response.

REFERENCES

Ablett, R.F.; R. O.,Sinnhuber; R.M., Homes and D.P., Selivonchick (1981): The effect of prolonged administration of bovine insulin in rainbow trout, *Salmo gairdneri*. Gen.Comp. Endocrinol. 43, 211-217.

Al-Salahy, M.B. (1990): Some metabolic effects of insulin and alloxan in the Nile fish, *Clarias lazera*, Ph. D. Thesis, Assiut University.

Al-Salahy, M.B. and S.Y. Hussein (1995): Effect of lon-term treatment of insulin on growth performance, lipid metabolism and electrolyte in *Oreochromis niloticus* Assiut Vet.Vol.33No.66: 70-83.

Alvarez, M.B. and H. D. Johnson (1973): Environmental heat exposure on cattle plasma catecholamine and glucocorticoid, J. Dairy Sci. 56: 189 – 194.

Ando, S. and M. Hatano (1986): Carotenoids in an egg yolk protein of chum salmon (Oncorhynchus Keta). Agric. Biol. Chem., 50: 1043 – 1`044.

APHA (American Public Health Association) (1985): Standard Methods for the Examination of Water and Wastewater 16 th edition. American Public Health Association, Washington, D.C.

Association of Official Analytical Chemists (1984): Mehhods of Analysis. AOAC, Washington, DC, pp. 152-169.

Berzin, N.I. (1988): Interaction between vitamin A and zinc in animals. Vestni Ksel, Skokhoznist Vennail Nauki, 1 : 106 – 111.

Bires, J;P. Bartko; H. Seidel; M., Sedovic; Z.Juhasovra and T. Weissava (1993): Zinc deficiency and possibilities of its supplementation by injection administration. Intrace element Nauki, : 327-328.

Borel, P. ; V. Tyssander ; N. Mekki ; P. Grolier ; Y. Rochette ; M.C. Alexandre – Gouabau ; D. Lairon and V. Azais – Braesco (1998) : Chyomicron β- carotene and retinly palmitate responses are dramatically diminished when men ingest β – carotene with medium-chain rather than long-chain triglycerides, J. Nutr. 128, : 1361-1367.

Carr, F.H. and E.A,. Prica (1926): Colometric determination of vitamin A and B-cartone. Biochemical J. : 420-498.

Chen., J.;J.,Chen;C.Chang;S Shen;M.ChenandJ.Wu(2000): Expression of recombinant tilapia insulin-like growth factor-I and stimulation of juvenile tilapia growth by injection of recombinant IGFs polypeptides.Aquaculture,Vol.181(3-4): 347-360.

Choubert, G.; de, la, J. Noue; J. M. Blanc. (1991): Apparent digestibility of canthaxanthin in rainbow trout: effect of dietary fat level, antibiotics and number of pyloric caeca, Aquaculture, 99, 323-329.

Czeczuga, B. (1975): Carotenoids in fish. IV. Salmonidae and Thumallidae from Polish water. Hydrobiologia, 46 : 223-239.

Daghash, H.A. and S.M. Mousa (1999): Zinc sulfate supplementation to ruminant rations and its effects on digestibility in lambs, growth, rectal temperature, and serum blood constituents in buffalo calves under heat stress. Assiut Vet. Med.J.40: 128-146.

David, J.C.M.; J.Polchana; J.H., Lilley; S.Kanchanakhn; K.D., Thompson and A.Adams (2001): Immunostimulation of striped snakehead *Channa striata* against epizootic ulcerative syndrome. Aquaculture, Vol. 195 (1-2) : 1-15.

Davis, G.K.(1980) : Microelement interaction of zonc, cooper, and iron in mammalian species.In O.A. Lavander and L.Cheng (Editors), Microelement Interactiions : Vitamins Minerals and Hazardous Elements. New York Acadamey of Sci. New York: `130-137.

Eder,k. and M.Kirchessner (1996): Zinc deficiency and the concentrations of thyroid hormones in serum of force-fed rats. J. of Animal Physiology and Animal Nutrition, 75 : 271-278.

Edwards, R.W. and V. M.,Brown (1966): Pollution and fisheries : A progress report. J. Inst. Wat. Pollut. Control, Lond. 66, 63-78.

Eid. A. and A.A. El-Gamal (1996): Effects of stocking density on growth performance of Nile tilapia (Oreochromis niloticus) reared in three different culture systems. Egyptian J.Anim. Prod., Suppl. Issue, :485-498.

Eid, A. and S.I. Ghonim (1994): Dietary zinc requirment of fingerling *Oreochromis niloticus.* Aquaculture, 119 : 259-264.

El deeb, M.A, and O.S. Afifi (2000): Effect of dietary zinc on semen quality, some blood constituents and urinary calcium of buck Rabbits.The 2nd Scientific Conf. Of Agric. Sci. Assiut: 935-945.

El-Masry, K.A. and A.A.Habbeeb (1989): Thyroid function in lactating Friesian cows and water buffaloes under winter and summer Egyptian conditions, Proceeding of 3 rd Egyptian–British Conf. On Anim.,Fish and Poult.,Prod.,Vol. 2 Alex. Egypt : 613 – 620.

Gatlin, D.M. and Wilson, R.R.(1984): Zinc supplementation of practical channel catfish diets. Aquaculture. 41 : 31-36.

Gatlin, D.M.; H.F.,Phillip and E.L., Torrans (1989): Effects of various levels of dietary copper and zinc on channel catfish. Aquaculture, 96 : 127-134.

Gatlin, D.M.and R.P. Wilson (1983): Dietary zinc requirement of fingerling channel catfish. J. Nutr. 113 : 630-635.

Gross, R.L.; N.Osolin; L.Fong and P.M. Newberne (1979): Depressed immunological function in zinc deprivefed rats as measured by nitrogen response of spleen, thymus and peripheral blood. American J.of Cilnical Nutrition , 32 : 1260-1266.

Haiqi, He; L.L., Addison and L. Ruiyu (1992): Evaluation of dietary essentiality of fat-soluble vitamins, A, D, E and K for penacid shrimp (Penaeus vannamei). Aquaculture, 103 : 177-185.

Hardy, R.W. and K. D. Shearer (1985): Effect of dietary calcium phosphate and zinc supplementation on whole body zinc concentration of rainbow trout (Salmo gairdneri) Can. J. Fish. Aquat. Sci., 42 : 181-184.

Harper, H.A., V.W. Rodwell and P.A. Mages (1979): Review of physiological chemistry 17th Edition pp. 511-555, Middle East Edition, lange Medical Publications, Beriut, Lebanon.

Hegazy, S.M. and Y. Adacht (2000): Comparison of the effects of dietary selenium, zinc and selenium and zinc supplementation on growth and immune response between chick groups that were

inoculated with Salmonella and aflatoxin or salmonella, Poultry Sci. 79 (3): 331-335.

Hilton, J.W. (1983): Hypervitaminosis A in rainbow trout (Salmo gairdneri): toxicity signs and maximum tolerable level. J. Nutr., 113 : 1737-1745.

Helena Laboratories Publications (1984): Serum protein electrophoresis procedure, Pro. 1: 9/84 (2).

Hoza, M.M. (1991): The effect of different levels of cadmium and zinc on growth performance and feed utilization of tilapia (Oreochromis niloticus), A. Msc. Thesis, Alexandria University, Egypt.

Huang, Y.-S.; K. Rousseau ; N. Le Belle; B. Vidal; E. Burzawa-Gerard; J. Marchelidon and S. Dufour (1999): Opposite effects of insulin-like growth factors (IGFs) on gonadotropin (GtH-II) and growth hormone (GH) production by primary culture of European eel (Anguilla anguilla) pituitary cells. Aquaculture, Vol. 177 (1-4) : 73-83.

Kaneko, J.J. (1989): Thyroid function. In clinical biochemistry of domestic animals 4th Ed.pp. 630-649. Academic press division of harcourt brace. Company, N.Y. USA.

Kegley, E.B. and J.W., Spear (1994): Effect of zinc supplementation on performance and zinc metabolism of lambs fed forage based diet. J. of Agric. Sci., 123 (2): 287-292.

Ketola, H.G. (1979): Influence of dietary zinc on catracts in rainbow trout (Salmo gairdneri) J. Nutr. 109 : 965-969.

Kirchgessner, M.; H.P., Roth and E. Weingand, (1976): Biochemical changes in zinc deficiency, In : A.S. Prasad and D. oberleas (Editors), Trace Elements in Human Health and Disease, Vol. 1, : 189-225.

Krahl, M.E. (1961): (Ed.) Insulin and muscle, insulin and adipase issue. pp. 15-39, 69-79, in the action of insulin on cells, Academic Press, Inc. New York.

Kutsky, R.G. (1981): Iodine, In: Handbook of vitamins, Minerals and Hormones (2nd Ed.) pp. 125-145 Van Nostrand Reinhold Co., New York.

Lan, W.G.; M. K. Wong; N. Chen and Y. M. Sin (1995): Effect of combined copper, zinc, chromium and selenium by orthogonal array desigen on alkaline phosphatase activity in liver of the red sea bream, Chrysophrys major. Aquaculture, Vol. 131 (3-4) : 219-230.

Larvor, P. (1983): The peols of cellular Nutrients: Minerals, In PM. Riis, ed., Dynamic Biochem. story of animal production pp. 281-317, Elsevier, Amsterdam.

Maage, A. and K. Julshamn (1993): Assessment of zinc status in juvenile Atlantic salmon (Salmo salar) by measurement of whole body and tissue levels of zinc. Aquacultyre, Vol. 117 (1-2) : 179-192.

Miyamoto, T.; A. sakurai and L.J. DeGroot (1991): Effect of zinc and other divalent metals on deoxyriibonucleic acid binding and hormone –binding activity of human alpha-1 thyroid hormone receptor expressed in Escherichia coli. Endocrinology129: 3027-3033.

Moore, T. (1957): Vitamin A, Elsevier Publishing Company, Amsterdam.

National Research Council. (1983): Nutrient Requirement of Dpmestic Animals Nutrient Requirement of Warmwater Fishes and Shellfishes. National Academy Press. Washington, Dc, pp. 96.

Ogino, C. and G. Yang (1978): Requirement of rainbow trout for dietary zinc. Bull. Jap. Eoc. Sci. Fish., 44 : 1015-1018.

Ogino, C.and Yang, G.Y. (1979): Requirement of carp for dietary zinc. Bull. Jap. Soci. Su. Fish., 45 : 4567-967.

Santos, M.H.S; N.T. da Cunha and A. Bianchini (2000): Effects of copper and zinc on growth, feeding and oxygen consumption of Farfantepenaeus paulensis postlarvae (Decapoda: Penacidae).J. of Experimental Marine Biology and Ecology, Vol. 247 (2): 233-242.

SAS (1987): SAS/STAT Guide for personal computer (Version 6 End) SAS INST., Cary, N.C.

*Settlemire,L.TandG.Malron(1967):*In-vivo effect of zinc on iron turnover in rats and life span of the erythrocyte J.Nutr.92:159-164.

Shetaewi, M..M. (2000): Effects of zinc and corn oil supplementation on reproduction and growth of New Zealand White rabbits in North Sinai. J. of Agric Sci. Mansoura 25: 1991-2004.

Silverstein, J. T.; W.R.Wolters; M. Shimizu and W.W. Dickhoff (2000): Bovine growth hormone treatment of channel catfish: strain and temperature effects on growth, plasma IGF-I levels, feed intake and efficiency and body composition. Aquaculture, Vol. 190 (1-2) : 77-88.

Sivtseva, L.V. (1982): Qualitative composition and distribution of carotenoids and vitamin A in the organs and tissue of rainbow trout, *Salmo gairdneri*, J. Ichthyol., 22 : 96-100.

Steel, R.G.D. and J.H. Torrie (1980): Principle and procedures of statistics; A Biometrics Approach (2nd Ed). McGraw-Hill Book Co., New York.

Stoskopf, M.K. (1993): Fish Medicine, (1st). Pp. 113-131, W.B. Saunders Company Harcour Brace lovanovish, Inc.

Takeuchi, T.; J.Dedi; Y. Haga; T. Seikai and T. Watanabe (1998): Effect of vitamin A compounds on bone deformity in larval sapanese flounder (Paralichthys oliroceu) Aquaculture, 169: 155-165.

Tan, B. and M.,.Kangsen (2001): Zinc methionine and zinc sulfate as sources of dietary zinc for juvenile abalone Haliotis discus hannai Ino. Aquaculture, Vol. 192 (1) : 64-84.

Thodesen, J.; T. Storebakken; K.D. Shearer; M.Rye; B.Bjerkeng and B. Gjerde (2001): Genetic variation in mineral absorption of large Atlantic salmon (Salmo salar) reared in seawater. Aquaculture, Vol. 194 (3-4) : 263-271.

Thompson, I; G. Ghoobrt; D.F. Houlihan and C.J. Secombes (1995): The effect of dietary vitamin A and astatanthin on the immuno comptence of rainbow trout, Aquaculture, 133: 91-102.

Wacker,W.E.C. (1976): Role of zinc in wound healing: a critical review : In A.S. Prasad and D.Oberleas (Editors) Trace Elements in Human Health and Disease,Vol1.Academic Press,NewYork NY.

Watanabe, T.; V. Kiron and S. Satoh (1997): Trace minerals in fish nutrition. Aquaculture, Vol. 151 (1-4) : 185-207.

Wekell, J.C. and J.D. Shearer (1986): Zinc supplementation of trout diets. Tissue indications of body zinc status. Prog. Fish-Cult., 48 : 205-212.

Yousef, H.M.; K.A. el-Masry and A.A. Aboulnagea (1996): Effect of dried live yeast supplement on haemobiochemical levls and milk production responses of lactating buffaloes under hot summer conditions in Egypt. Egyptian J. of Animal Production, 33 (1) : 11 – 21.

Zilva, J.E. (1973): Toxicology in clinical chemistry. In: Diagnosis and Treatment, First Edition, London, Lioydbuke. pp. 314-332

Table (1): Effect of dietary zinc and/or vitamin A on growth and relative body weights of *O. niloticus*.

Treatment	1	2	3	4	5	6	7	8	9
Zinc (mg/kg DM diet)	0.0	30	40	0.0	30	40	0.0	30	40
Vitamin A (mg/kg DM diet)	0.0	0.0	0.0	4000	4000	40000	60000	6000	6000
body gain (g/fish)	2.17^{c} ±0.76	2.35^{ab} ±0.76	2.49^{ab} ±0.76	2.90^{ab} ±0.76	2.96^{ab} ±0.68	2.63^{ab} ±0.81	3.01^{ab} ±0.76	2.50^{ab} ±0.76	3.09^{a} ±0.76
Length increment (mm)	3.14^{d} ±1.20	3.33^{abc} ±1.30	3.81^{c} ±1.26	3.56^{bc} ±1.20	3.20^{cd} ±1.20	3.56^{bcc} ±1.20	3.82^{bac} ±1.30	3.26^{bc} ±1.20	3.80^{b} ±1.20
Feed conversion	3.30^{a} ±0.67	2.66^{c} ±0.67	2.57^{c} ±0.67	2.59^{c} ±0.67	2.77^{c} ±0.67	2.65^{c} ±0.67	2.54^{c} ±0.67	2.27^{b} ±0.67	2.25^{b} ±0.67
Hepatosomatic index (H S I)	2.15^{b} ±0.15	2.00^{b} ±0.15	2.09^{a} ±0.15	2.04^{a} ±0.15	2.13^{a} ±0.15	1.96^{b} ±0.15	2.00^{a} ±0.15	2.13^{a} ±0.15	2.15^{a} ±0.15
Male gonadosomatic index (mGSI)	0.88^{a} ±0.21	0.67^{a} ±0.17	0.99^{a} ±0.22	1.17^{a} ±0.21	1.07^{a} ±0.21	0.79^{a} ±0.23	0.67^{a} ±0.29	0.92^{a} ±0.19	1.07^{a} ±0.17
Female onadosomatic index (fGSI)	3.86^{a} ±0.92	4.18^{a} ±1.13	3.22^{a} ±0.71	3.29^{a} ±0.80	3.54^{a} ±0.80	2.69^{a} ±0.71	2.79^{a} ±0.60	3.86^{a} ±0.92	2.85^{a} ±1.13
Spleen somatic index (SSI)	0.20^{bcd} ±0.02	0.24^{abc} ±0.02	0.19^{bc} ±0.02	0.16^{d} ±0.02	0.15^{d} ±0.02	0.23^{bac} ±0.02	0.19^{bcc} ±0.02	0.28^{a} ±0.02	0.25^{ab} ±0.02

Means within rows differ (P< 0.05) when superscript differ.

Table (2) : Effect of dietary zinc and/or vitamin A on body composition of *O. niloticus*. (on DM basis)

Treatment	1	2	3	4	5	6	7	8	9
Zinc (mg/kg DM diet)	0.0	30	40	0.0	30	40	0.0	30	40
Vitamin A (mg/kg DM diet)	0.0	0.0	0.0	4000	4000	4000	6000	6000	6000
Dry matter (%)	25.46^{a} ±0.38	24.75^{a} ±0.38	24.84^{a} ±0.38	24.67^{a} ±0.38	25.25^{a} ±0.38	25.37^{a} ±0.38	25.05^{a} ±0.38	25.57^{a} ±0.38	24.86^{a} ±0.38
Crude protein (%)	69.88^{ba} ±1.08	70.29^{ba} ±1.08	71.39^{a} ±1.08	71.63^{a} ±1.08	67.70^{b} ±1.08	67.64^{b} ±1.08	67.26^{b} ±1.08	67.30^{b} ±1.08	68.56^{ba} ±1.08
Crude fat (%)	9.97^{ba} ±0.72	9.04^{b} ±0.72	9.71^{b} ±0.72	6.82^{c} ±0.72	9.77^{b} ±0.72	9.92^{ba} ±0.72	9.74^{b} ±0.72	10.90^{a} ±0.72	9.73^{a} ±0.72
Ash (%)	20.49^{a} ±0.40	20.71^{a} ±0.40	18.89^{a} ±0.40	21.56^{a} ±0.40	22.56^{a} ±0.40	22.43^{a} ±0.40	23.04^{a} ±0.40	21.47^{a} ±0.40	21.71^{a} ±0.40
Zinc mg/Kg	74.73^{bc} ±5.06	74.05^{bc} ±5.06	92.72^{a} ±5.06	78.57^{abc} ±5.06	88.17^{ab} ±5.06	94.08^{a} ±5.06	81.17^{abc} ±5.06	79.89^{abc} ±5.06	70.94^{c} ±5.06

Means within rows differ (P< 0.05) when superscript differ.

Table (3) :Influence of dietary zinc and vitamin A supplementation on some blood constituents in O. *niloticus*.

Treatment	1	2	3	4	5	6	7	8	9
Zinc (mg/kg DM diet)	0.0	30	40	0.0	30	40	0.0	30	40
Vitamin A (mg/kg DM diet)	0.0	0.0	0.0	4000	4000	4000	6000	6000	6000
Hemoglobin (g/dl)	5.36^{f} ±0.34	5.96^{bdec} ±0.34	6.79^{ba} ±0.34	5.62^{dc} ±0.34	5.72^{bdc} ±0.34	6.20b^{dca} ±0.34	6.28^{bdca} ±0.34	6.50^{bac} ±0.34	7.03^{a} ±0.34
Hematocrit (PCV) (%)	36.0^{e} ±3.16	38.^{bdcec} ±3.16	39.25^{bc} ±3.16	42.00^{dc} ±3.16	41.00^{bdc} ±3.16	41.5^{bdca} ±3.16	41.0^{bdca} ±3.16	41.50^{bac} ±3.16	42.50^{a} ±3.16
Total protein (g/dl)	5.97^{d} ±0.66	7.23^{ba} ±0.66	7.33^{a} ±0.75	7.69^{ba} ±0.71	7.37^{a} ±0.66	6.91^{bca} ±0.66	6.94^{c} ±0.75	6.33^{bc} ±0.71	6.03^{bc} ±0.66
Albumin (g/dl)	1.52^{bc} ±0.10	1.73^{bac} ±0.11	1.99^{a} ±0.12	1.84^{ba} ±0.12	1.98^{a} ±0.11	1.60^{bc} ±0.11	1.42^{c} ±0.12	1.53^{bc} ±0.12	1.56^{bc} ±0.11
Glucose (mg/dl)	59.32^{e} ±6.34	63.48^{eb} ±6.73	68.49^{eb} ±7.77	65.57^{eb} ±6.34	65.03^{eb} ±6.02	71.86^{ed} ±6.73	68.67^{eb} ±6.73	66.01^{eb} ±6.73	85.11^{a} ±6.73
Triglycerides (mg/dl)	199.4^{h} ±32.17	231.2^{g} ±32.17	219.47^{g} ±32.17	227.63^{g} ±32.17	261.05^{e} ±32.17	239.81^{f} ±32.17	219.21^{g} ±32.17	247.22^{e} ±23.17	235.53^{g} ±32.17
Alkaline phosphatase(U/l)	37.94^{f} ±2.68	39.43^{b} ±2.99	39.55^{g} ±3.20	39.65^{f} ±2.82	39.78^{c} ±2.68	40.00^{d} ±2.68	40.12^{c} ±2.99	40.27^{b} ±2.99	40.41^{a} ±2.99
T_3 (ng/ml)	1.866^{c} ±0.650	1.962^{e} ±0.650	3.478^{bc} ±0.650	3.257^{ba} ±0.650	2.436^{ba} ±0.650	3.626^{ba} ±0.650	2.054^{c} ±0.650	3.501^{ba} ±0.650	3.478^{ba} ±0.650
T_4 (ng/ml)	0.622^{e} ±0.195	1.973^{ba} ±0.195	2.420^{ba} ±0.195	2.134^{ba} ±0.195	1.925^{ba} ±0.195	2.756^{ba} ±0.195	4.699^{a} ±0.195	4.034^{ba} ±0.195	3.116^{ba} ±0.195
T_3/T_4 ratio	1.778^{a} ±0.60	1.162^{a} ±0.60	2.518^{a} ±0.60	2.094^{a} ±0.60	2.573^{a} ±0.60	1.674^{a} ±0.60	0.810^{a} ±0.60	0.960^{a} ±0.60	1.235^{a} ±0.60
T. cholesterol (mg/dl)	180.6^{a} ±16.34	185.2^{a} ±16.34	207.21^{a} ±16.34	175.0^{ba} ±16.34	167.2^{ba} ±16.34	172.1^{ba} ±16.34	160.9^{ba} ±16.34	142.95^{b} ±16.34	148.36^{b} ±16.34
AST (U/L)	34.00^{b} ±2.91	39.75^{b} ±2.91	26.00^{c} ±2.91	37.50^{b} ±2.91	38.50^{b} ±2.91	34.00^{bc} ±2.91	38.50^{b} ±2.91	36.25^{b} ±2.91	5250^{a} ±2.91
ALT (U/L)	22.00^{b} ±2.14	25.00^{e} ±2.14	24.00^{c} ±2.14	26.50^{b} ±2.14	23.00^{g} ±2.14	25.00^{dc} ±2.14	20.00^{b} ±2.14	21.00^{bc} ±2.14	27.00^{a} ±2.14
Serum zinc (ug/dl)	648^{c} ±28.79	562.4^{e} ±28.79	774^{b} ±28.79	446.4^{g} ±28.79	674^{c} ±28.79	512^{d} ±28.79	666^{c} ±28.79	1264^{a} ±28.79	1202^{a} ±28.79
B-cartone (ug %)	122.99^{g} ±56.20	109.20^{b} ±56.20	232.18^{b} ±56.20	100.00^{a} ±56.20	165.52^{c} ±56.20	112.64^{g} ±56.20	142.40^{f} ±56.20	151.61^{d} ±56.20	181.82^{c} ±36.20
Vitamin A (ug %)	7 311^{e} ±124.2	18 972^{b} ±124.2	17 186^{d} ±124.2	18 736^{b} ±124.2	35 750^{a} ±124.2	18 166^{c} ±124.2	17 406^{d} ±124.2	17 570^{d} ±124.2	18 421^{c} ±124.2

Means within rows differ (P< 0.05) when superscript differ.

Table (4) : Effect of dietary zinc and/or vitamin A on serum globulin and immune indices of *O. niloticus*.

Treatment	1	2	3	4	5	6	7	8	9
Zinc (mg/kg DM diet)	0.0	30	40	0.0	30	40	0.0	30	40
Vitamin A (mg/kg DM diet)	0.0	0.0	0.0	4000	4000	4000	6000	6000	6000
Globulin (g/dl)	4.37^{d} ±0.60	5.50^{bac} ±0.60	5.33^{ba} ±0.68	5.85^{a} ±0.63	5.39^{ba} ±0.63	5.27^{ba} ±0.73	5.56^{ba} ±0.73	4.79^{ba} ±0.68	4.45^{bd} ±0.68
α- globulin (g/dl)	1.30^{c} ±0.18	1.75^{bc} ±0.18	2.13^{ab} ±0.18	2.50^{a} ±0.18	1.69^{bc} ±0.18	1.73^{bc} ±0.18	2.23^{ab} ±0.18	1.44^{c} ±0.18	1.12^{ab} ±0.18
B - globulin (g/dl)	1.86^{ab} ±0.13	1.71^{ab} ±0.13	1.59^{b} ±0.13	1.50^{b} ±0.13	2.02^{a} ±0.13	1.67^{ab} ±0.13	1.86^{ab} ±0.13	1.87^{ab} ±0.13	1.74^{ab} ±0.13
γ - globulin (g/dl)	1.22^{c} ±0.13	2.03^{a} ±0.13	1.61^{bc} ±0.13	1.86^{ab} ±0.13	1.68^{ab} ±0.13	1.87^{ab} ±0.13	1.47^{bc} ±0.13	1.70^{ab} ±0.13	1.71^{ab} ±0.13

Means within rows differ ($P< 0.05$) when superscript differ.

Fig. (1): Effect of dietary zinc and/or vitamen A on serum high density lipoprotein and low density lipoprotein of O. niloticus.

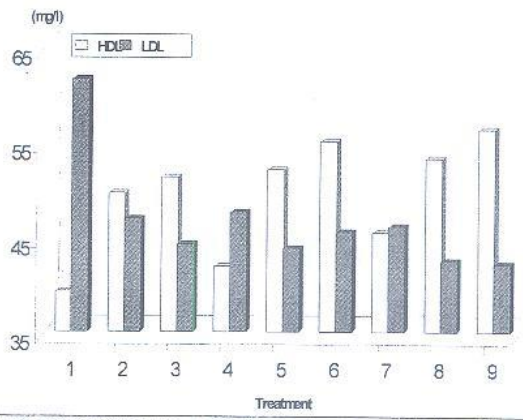

Fig. (2): Effect of dietary zinc and/or vitamen A on serum globulin and immune compor O.niloticus.