Hematological and enzymatic response to salinity in juvenile common carp *Cyprinus carpio* with potassium chloride and growth hormone supplementation in the diet.

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Abstract

The juveniles of common carp fish Cyprinus carpio with a weight of 45 g ± 2.12 g were gradually exposed to three salt concentrations: liquefaction water (1.2), 7 and 15 g/L for the purpose of studying the effect of salt acclimatization on the osmotic regulation of juveniles of common carp fish Cyprinus carpio By measuring some blood parameters such as total auxin concentration, total energy level, glucose, total proteins and liver enzymes in the blood plasma. The results showed that the fish that took potassium chloride as an additive to the feed experienced less changes in blood glucose levels than those that took the growth hormone as an additive in the feed, and this indicates that the effect of potassium chloride was greater and clearer in reducing osmotic stress. With regard to the effect of the two additives, potassium chloride and growth hormone, on quantitative proteins, there are differences between one treatment and another without a clear and consistent pattern. In terms of the amount of energy consumed, fish in the case of exposure to salinity of 7 PSU, there were no significant differences in energy consumption between fish in the control group and other groups that took potassium chloride salts or growth hormone, and the scene differs clearly when exposed to salinity of 15 PSUWhere differences appear, most parameter significantly between the fish in the control group that consumed the largest amount of energy and consumed the largest amount of oxygen compared to the other groups that consumed two levels of salt and two levels of the hormone, and the best of them was the consumption of high concentration of salt and also high concentration of hormone. The enzymes alkaline phosphatase (ALP) and enzyme (ALT) did not witness significant changes despite the exposure of fish to increased salinity, whether by adding potassium chloride and growth hormone or not.As for the only indicator that witnessed some kind of fluctuations in its levels in the blood, it was the enzyme (AST), which witnessed a mostly insignificant difference between some different concentrations of potassium chloride or growth hormone addition treatments in fish.

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Keywords: enzymatic response, juvenile common carp, *Cyprinus carpio*, potassium chloride, growth hormone

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Introduction

Global fish farming has grown steadily in recent years, to become of great economic importance among the agricultural sectors, as this sector is characterized by being the most productive and the fastest developing in the world due to its potential to meet the nutritional requirements of the consumer. This led to the emergence of specialized and accredited companies for the manufacture of feed for animals, especially fish. (Gaus, 2010).

Common carp is one of the most important fish species used in aquaculture worldwide. Abbass (2007) is the selected species due to its high growth rate.

Ease of breeding, tolerance to environmental conditions and stress and consumer palatability Feed additives have the potential to help improve growth rates or reduce fish farming time.

Bio-additives and hormones are widely used in fish farming to improve fish growth by modifying the environment that in live.

The process of osmotic regulation in fish is one of the environmental matters that must be taken into consideration. It is one of the energy-consuming processes that negatively affect growth. This process is under hormonal control that controls the movement of ions within and through the organs of osmotic regulation, directly or indirectly (Tait). et al., 2017).

Some studies have addressed the role of fish hormones in adapting to low and high osmotic environments because hormones in the blood plasma of organoid fish such as growth hormone and cortisol are accompanying the ionic regulation process when fish adapt to sea or fresh water (Froese and Pauly, 2018),

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Whereas, growth hormone plays an important role in the process of acclimatization towards water salinity by increasing the activity of the Na+/K+ ATPase enzyme and the number and size of the chloride cell (Dang et al., 2000).

materials and methods

Oxygen consumption rate:

The measurement of oxygen consumption in fish is a good indicator of metabolic rate. The method of measuring it was based on the amount of decrease in the amount of dissolved oxygen in the water, using the closed containers method for measuring oxygen consumption, based on Nordlie and Leffer (1975).

In which one fish of a known weight is placed in a conical, opaque glass container of one liter capacity, filled with water saturated with oxygen, by pumping air into it until saturation. The ration-fed fish (T1, T2, T3, T4 and T5) were transported at the following saline concentrations (fresh water, 7 and 15 g/L) in closed containers for 24 hours for the purpose of acclimatization to detention. When the experiment begins, the ventilation is stopped and the containers are closed tightly to prevent the leakage of oxygen into it.

Blood glucose level:

To calculate the percentage of glucose in the blood of fish, the Irish-made ACCU-CHEK ACTTIVE device (GNO 2278729) was used.

Where the caudal peduncle of the fish is cut with a sharp scalpel to make the blood flow continuously, then a drop of blood is placed on the tape of the device and fixed in the place designated for it, and the result is read directly by the appearance of the number on the screen of the device.

. Total proteins in blood plasma:

The total proteins in the blood plasma were estimated by Beuret method by Henry et al. 1974)) using the Biuret Kit produced by the French company RANDOX. In this method, copper ions react in the alkaline medium with the protein peptide, forming a colored complex. Then, the

absorbance is read by spectrophotometry at a wavelength of 546 nm, and the blood plasma proteins are estimated in g/100 ml or g/L units, according to the following equation.

$$\text{Concentration of total proteins} = \frac{\text{Sample absorbance}}{\text{absorbance of standard solution}} \times \text{concentration of standard solution}$$

Alkaline phosphatase (ALP) enzyme concentration measurement.

Enzyme concentrations in serum were measured using ready-made solutions Kit from the German company Human, and samples were read using a spectrophotometer at a wavelength of 400 nm.

According to the instructions of the supplied company, where the concentration calculation was from the sum of three readings and divided by their number and multiplied by the fixed factor as in the following equation

AΔ = absorbance variable x 3433

7.3.9. Measurement of the enzyme (GOT Glutamic Oxaloacetate Transaminase).

(AST) Aspartate Transaminase =

The enzyme concentrations in the blood serum were measured using ready-made solutions ((Kit) from the German company Human, and the samples were read using a spectrophotometer at a wavelength of 340 nm.

According to the instructions of the supplied company, where the concentration calculation was from the sum of three readings, divided by their number and multiplied by the fixed factor as in the following equation:

AΔ = absorbance variable × 1667

7.3.10. Glutamic Pyruvic Transaminase (GPT) Test

= Alanine Transaminase (ALT)

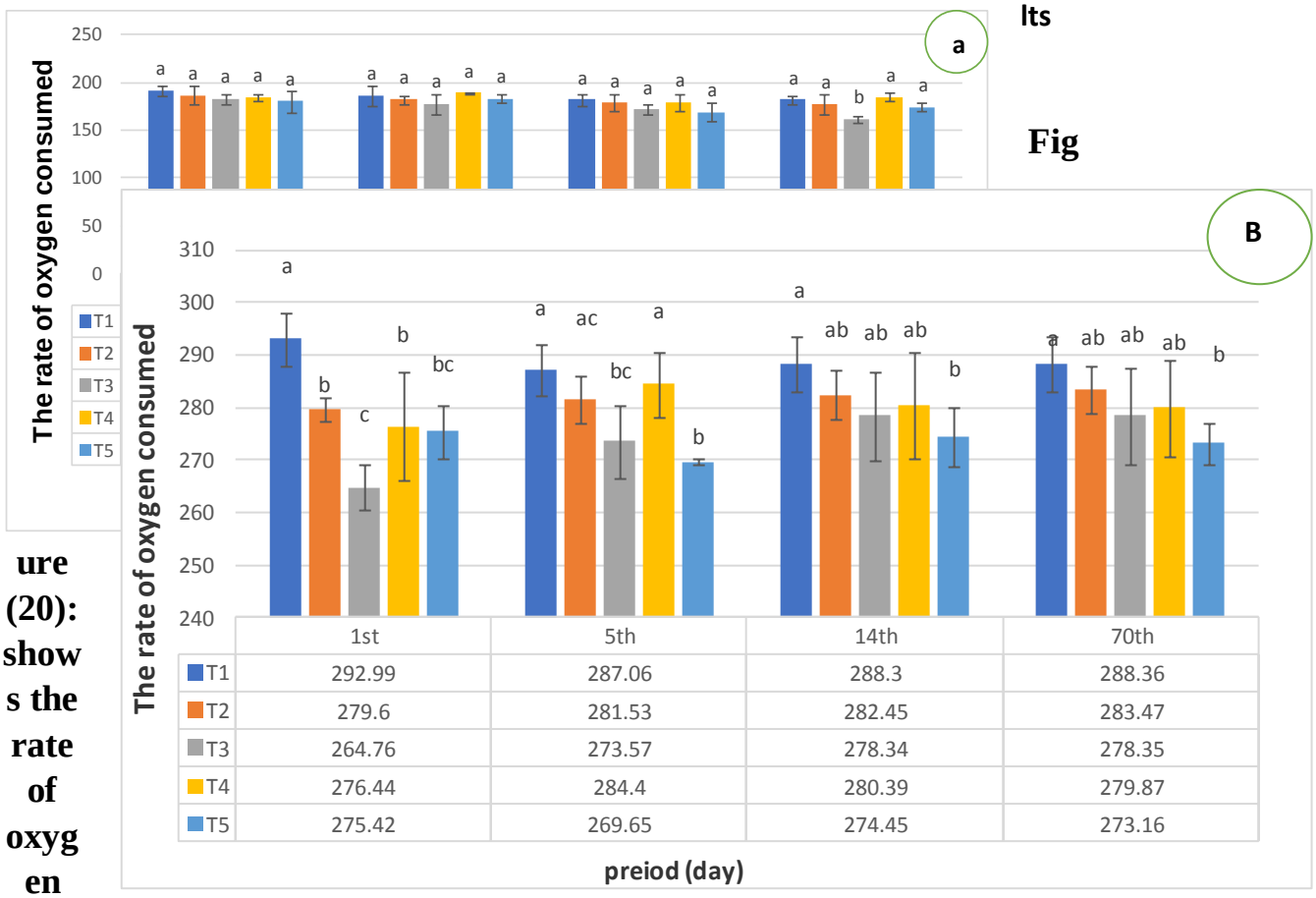
The enzyme concentrations in the blood serum were measured using ready-made solutions (Kit) from Randox Company in England, and the samples were read using a spectrophotometer at a wavelength of 550 nm according to the instructions of the

supplied company, where the concentration calculation was to compare the result with the table prepared by the company.

statistical analysis

Statistical analysis was carried out using a complete randomized design (CRD). The significant differences between the coefficients were determined by performing the least significant difference (LSD) test. All statistical tests were conducted using the Statistical package for social sciences (IBM SPSS), version 26.

Results



	1st	5th	14th	70th
T1	292.99	287.06	288.3	288.36
T2	279.6	281.53	282.45	283.47
T3	264.76	273.57	278.34	278.35
T4	276.44	284.4	280.39	279.87
T5	275.42	269.65	274.45	273.16

Figure (20): shows the rate of oxygen consumed (mg O2 / kg / hour) for juvenile common carp fish in different treatments exposed to salt concentrations (A) (7PSU) and B (15PSU). (The values represent the rate ± standard deviation).

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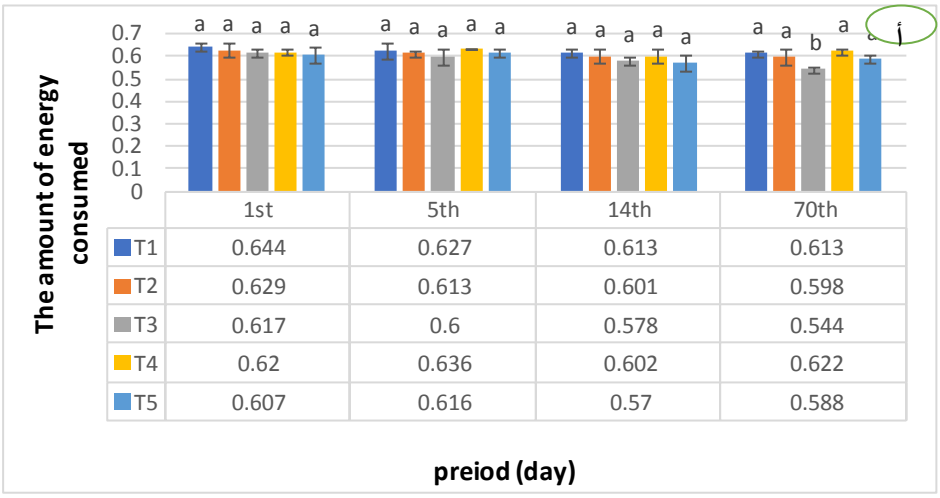
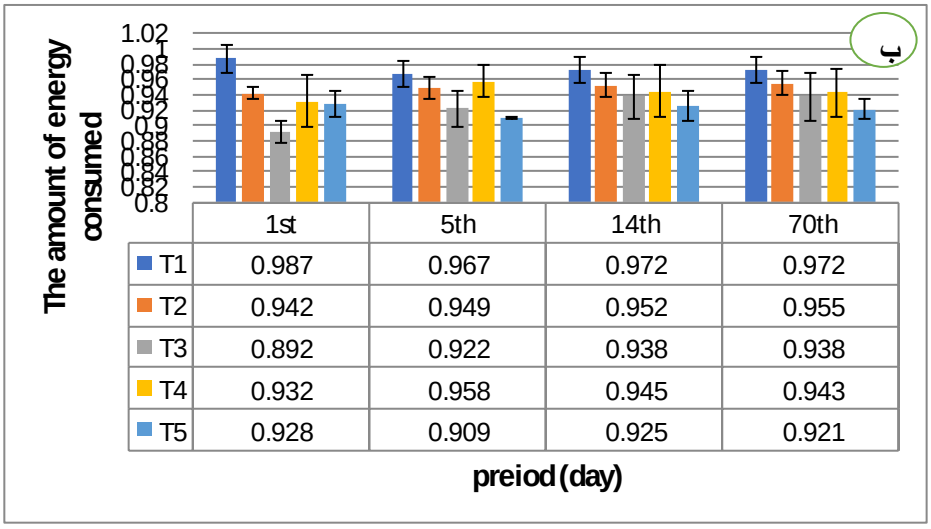


	1st	5th	14th	70th
T1	0.644	0.627	0.613	0.613
T2	0.629	0.613	0.601	0.598
T3	0.617	0.6	0.578	0.544
T4	0.62	0.636	0.602	0.622
T5	0.607	0.616	0.57	0.588



	1st	5th	14th	70th
T1	0.987	0.967	0.972	0.972
T2	0.942	0.949	0.952	0.955
T3	0.892	0.922	0.938	0.938
T4	0.932	0.958	0.945	0.943
T5	0.928	0.909	0.925	0.921

Figure (21): shows the amount of energy consumed (kilocalories/kg/hour) for juvenile common carp fish in different treatments exposed to salt concentrations (A) (7PSU) and B (15PSU). (The values represent the mean ± standard deviation).

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The level of glucose in the blood

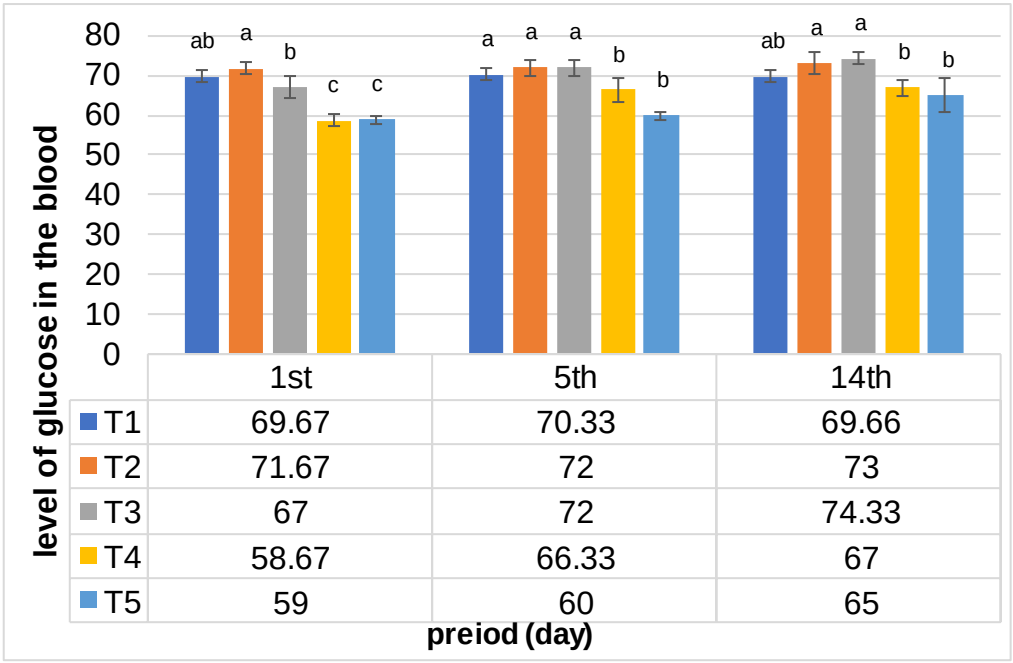
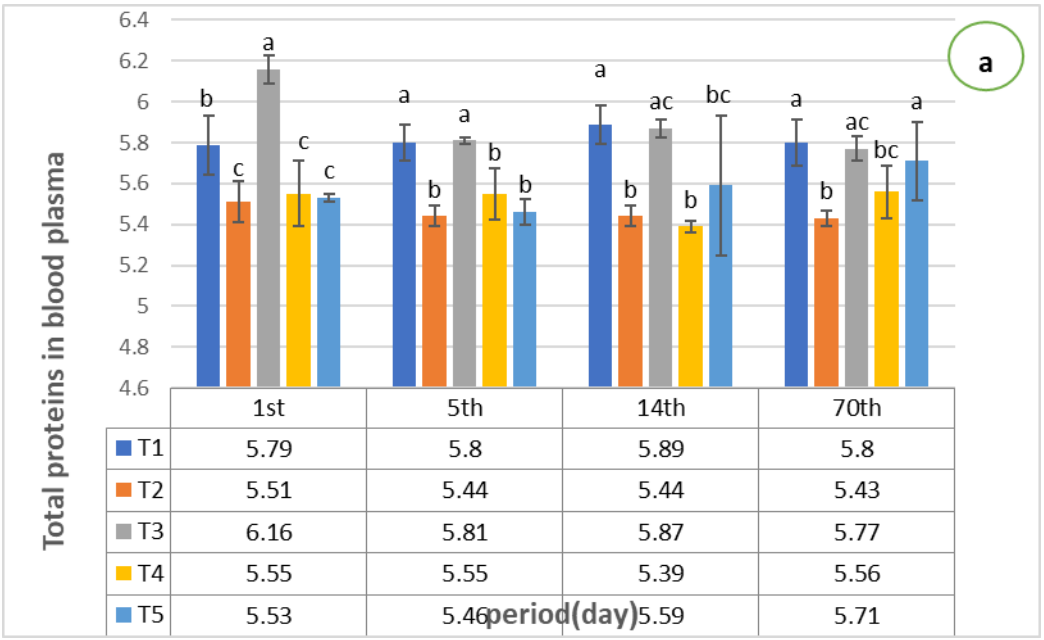


Figure (15) Level of glucose in the blood (mg/100ml of blood) of juvenile common carp fish in different treatments exposed to salt concentration... PSU (fresh water). (Values represent mean ± standard deviation).



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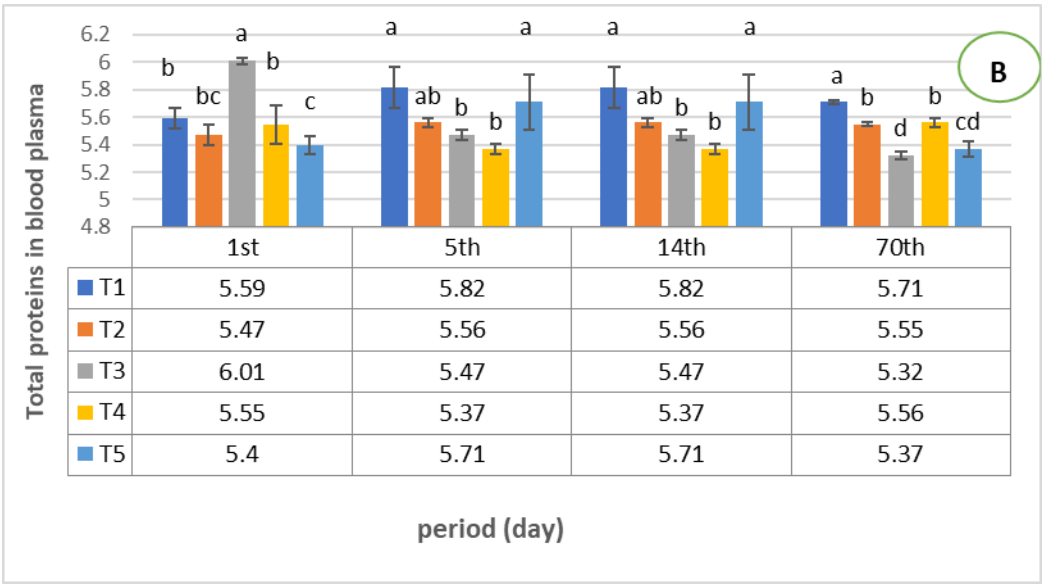
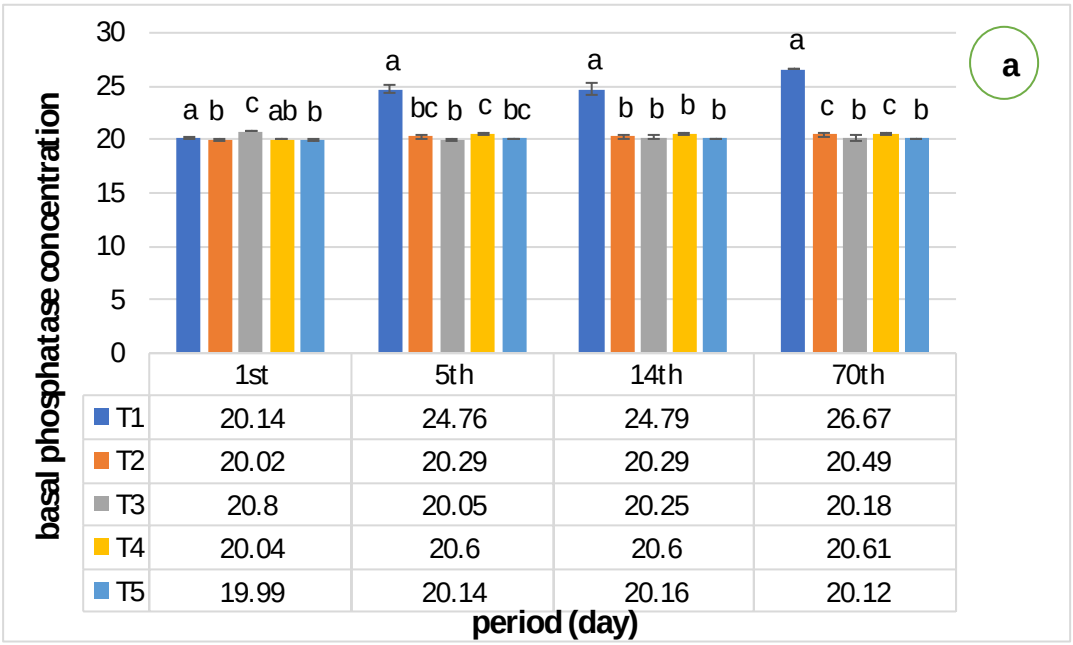


	1st	5th	14th	70th
T1	5.59	5.82	5.82	5.71
T2	5.47	5.56	5.56	5.55
T3	6.01	5.47	5.47	5.32
T4	5.55	5.37	5.37	5.56
T5	5.4	5.71	5.71	5.37

Figure (16): shows the concentration of total proteins in the blood plasma (mg/100ml) of juvenile common carp in different treatments exposed to salt concentrations (A) (7PSU) and B (15PSU). (The values represent the mean ± standard deviation).



	1st	5th	14th	70th
T1	20.14	24.76	24.79	26.67
T2	20.02	20.29	20.29	20.49
T3	20.8	20.05	20.25	20.18
T4	20.04	20.6	20.6	20.61
T5	19.99	20.14	20.16	20.12

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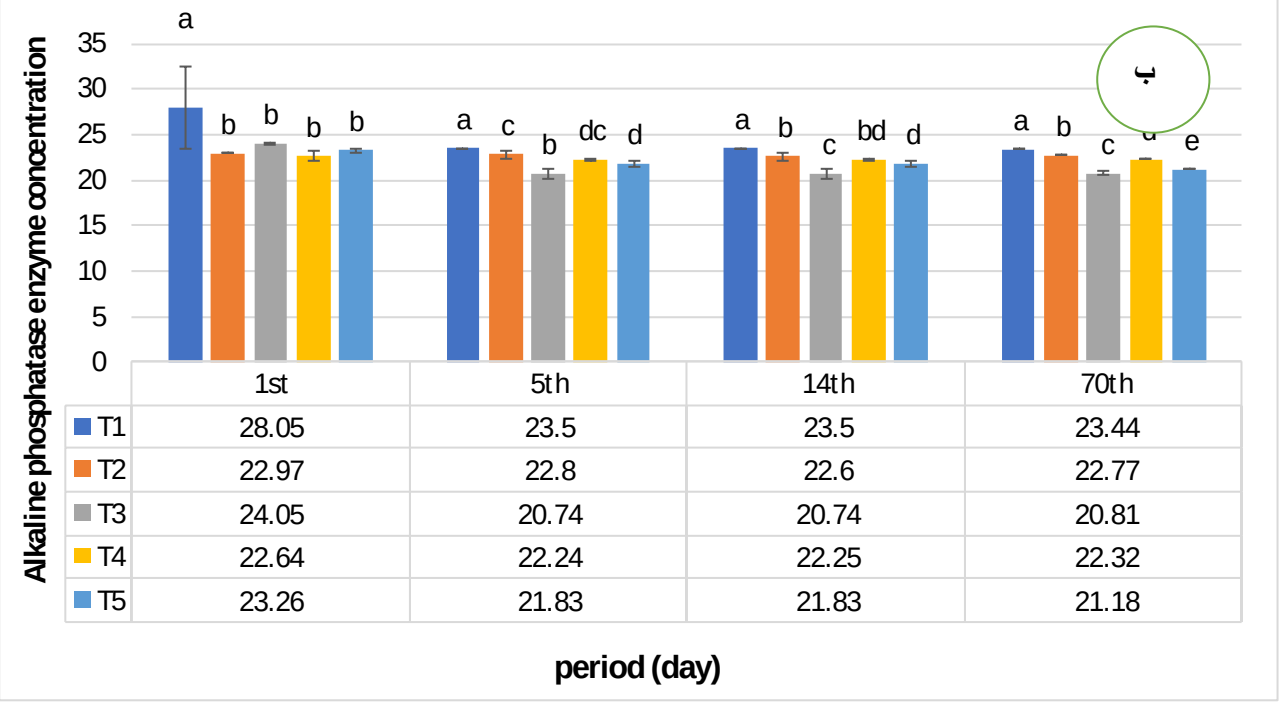
Figure (17): shows the concentration of alkaline phosphatase (ALP) (international unit/liter)



	1st	5th	14th	70th
T1	28.05	23.5	23.5	23.44
T2	22.97	22.8	22.6	22.77
T3	24.05	20.74	20.74	20.81
T4	22.64	22.24	22.25	22.32
T5	23.26	21.83	21.83	21.18

of juvenile common carp in different treatments exposed to salt concentrations (A) (7PSU) and B (15PSU). (The values represent the mean ± standard deviation).

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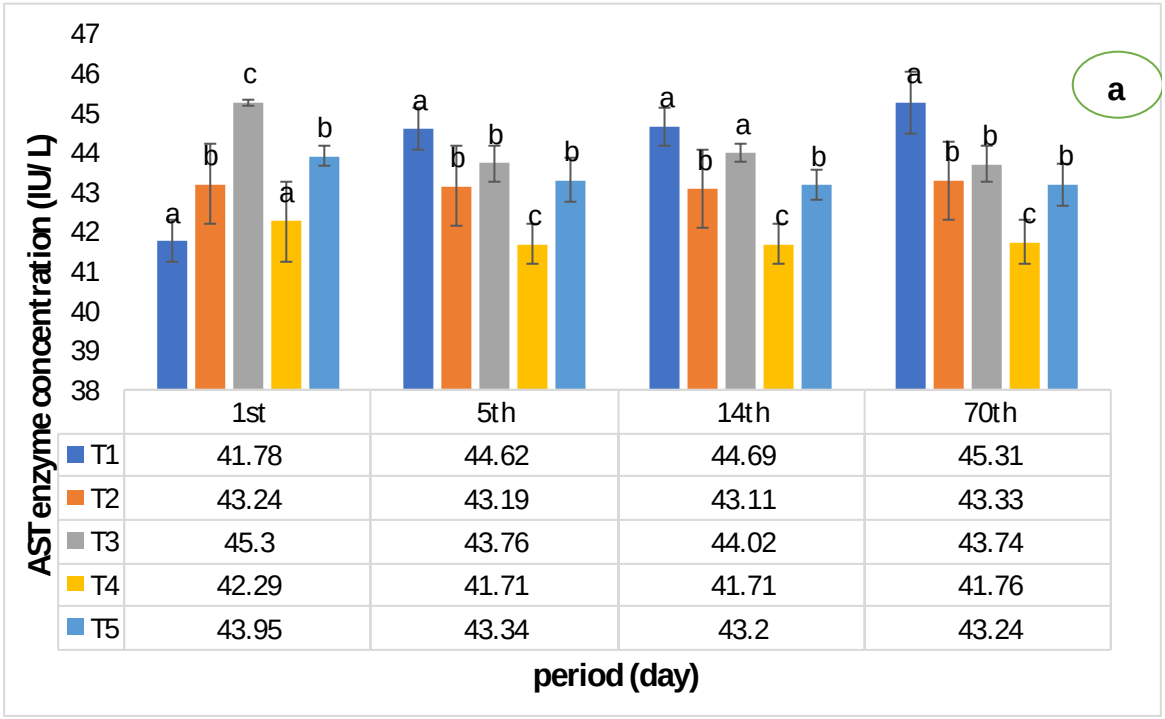
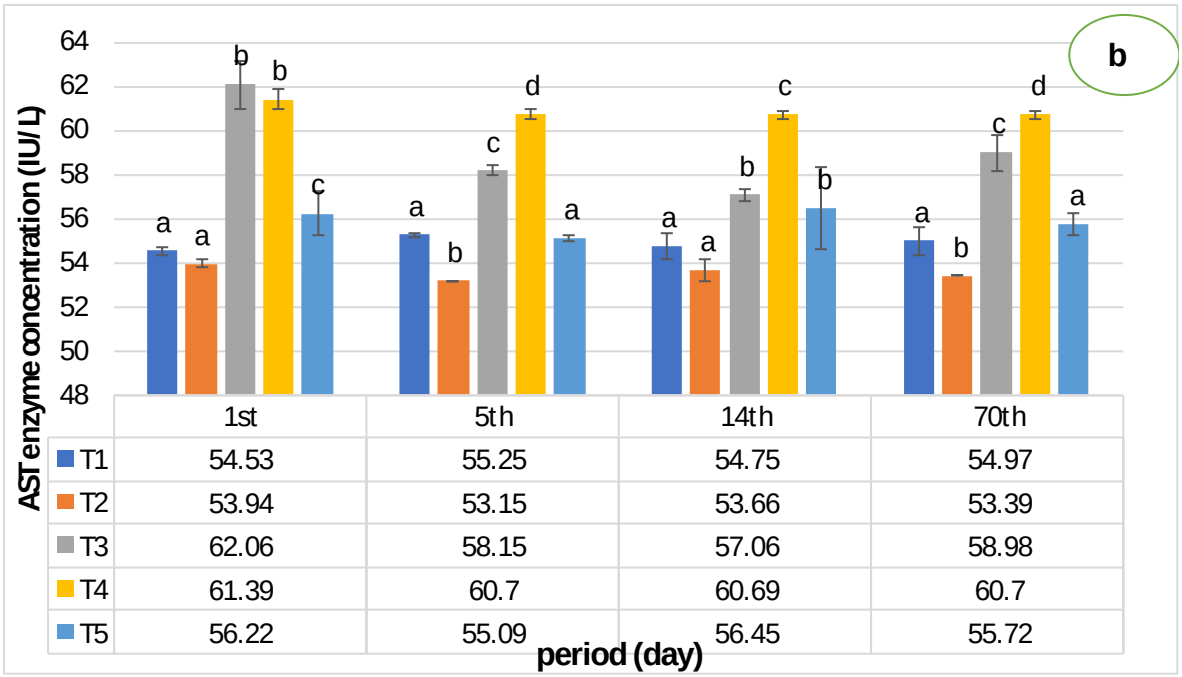


	1st	5th	14th	70th
T1	41.78	44.62	44.69	45.31
T2	43.24	43.19	43.11	43.33
T3	45.3	43.76	44.02	43.74
T4	42.29	41.71	41.71	41.76
T5	43.95	43.34	43.2	43.24



	1st	5th	14th	70th
T1	54.53	55.25	54.75	54.97
T2	53.94	53.15	53.66	53.39
T3	62.06	58.15	57.06	58.98
T4	61.39	60.7	60.69	60.7
T5	56.22	55.09	56.45	55.72

Figure (18): shows the concentration of AST enzyme (international units/liter) of juvenile common carp in different treatments exposed to salt concentrations (A) (7PSU) and B (15PSU). (The values represent the mean ± standard deviation).

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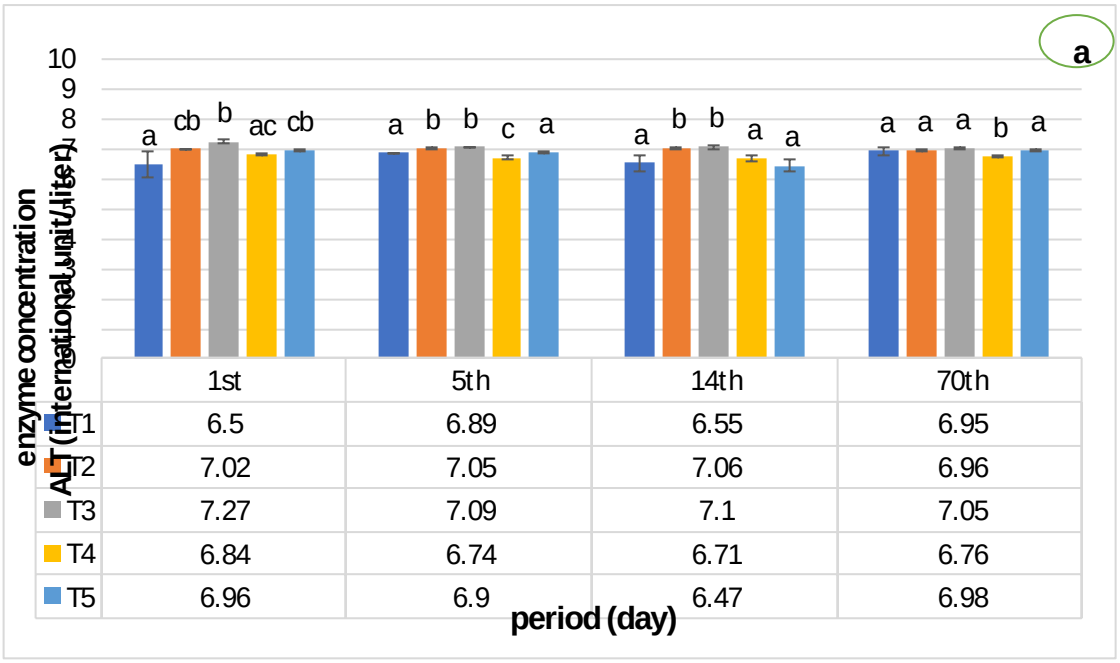
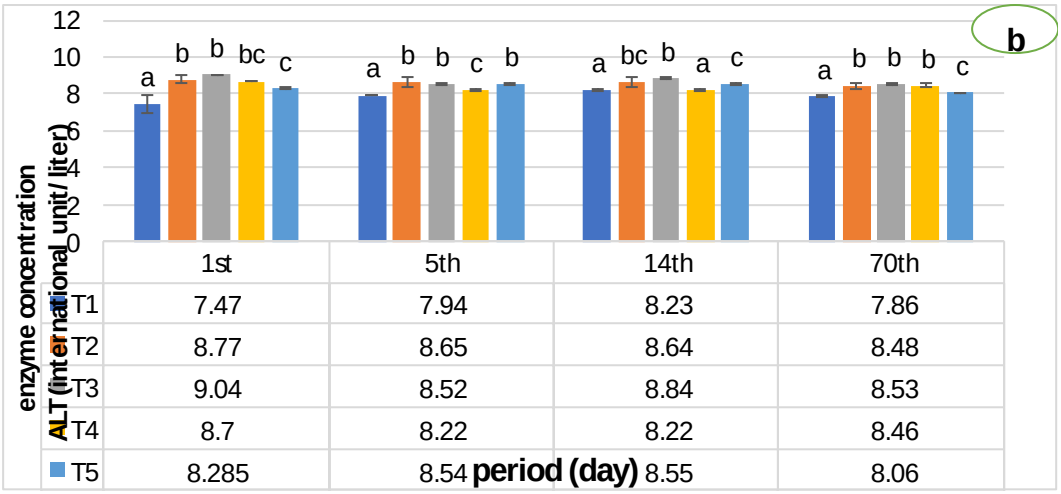


	1st	5th	14th	70th
T1	6.5	6.89	6.55	6.95
T2	7.02	7.05	7.06	6.96
T3	7.27	7.09	7.1	7.05
T4	6.84	6.74	6.71	6.76
T5	6.96	6.9	6.47	6.98



	1st	5th	14th	70th
T1	7.47	7.94	8.23	7.86
T2	8.77	8.65	8.64	8.48
T3	9.04	8.52	8.84	8.53
T4	8.7	8.22	8.22	8.46
T5	8.285	8.54	8.55	8.06

Figure (19): shows the concentration of ALT enzyme (international units / liter) of juvenile common carp fish in different treatments exposed to salt concentrations (A) (7PSU) and B (15PSU). (The values represent the mean ± standard deviation)

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