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Studies on tolerance and serum biochemical indexes of juvenile *Pseudosciaena crocea* in different temperature

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ABSTRACT

Both high temperature and low temperature test are included in this essay, studying the tolerance on temperature of juvenile *Pseudosciaena crocea*. At the same time, we will compare the data in normal water temperature, high temperature limit and low limit to analyze the mechanism of their reaction to temperature. The result indicates that the high limit is 37℃ while the low one is 8℃, we also find that the fish's serum enzyme and blood fat in normal water condition are so different from they are in limit temperature conditions through the study on the serum biological data(P<0.05). There is a possibility that the change of water will cause some damage on the fish entrails, leading to the change for serum enzyme and blood fat.

KEYWORDS

Juvenile *Pseudosciaena crocea*; Temperature; Tolerance; Serum; Biochemical indexes.

Pseudosciaena crocea Richardson belongs to the Perciformes、Sciaenidae and Pseudosciaena[1]. The fish is very tasty and has a high content of protein and unsaturated fattyacid. As one of the most sea commercial fish, it has an ability of getting used to the temperature. But in recent years, *Pseudosciaena crocea* raised in ocean district died in a large quantity because of the low water temperature[2, 3]. Although the breeding is mostly processed in net cages along the coastline, the search on industrial manufacture is also on the way[4]. Hence, it seems to be very important to study the tolerance on temperature of this fish to settle down the high and low limit in the breeding process.

When animals are involved in uncomfortable temperature environment, their bodies will become stressed due to the threat of both hot and cold climate, leading to the change in blood biological index[5, 6], especially for the serum enzyme, blood fat and serum inorganic ions. Through the analyze on the blood biological index, we could get a deeper understanding of the stress mechanism. Some researches in this area have also been done by domestic and overseas experts[7-9]. Yet there is no report on the blood biological index in different temperatures.

This study aims to find the high and low limit through the research on the temperature tolerance of juvenile *Pseudosciaena crocea* and the blood biological index.

MATERIALS AND METHODS

The tested fish

Fishes used in experiments are Min-Yue Dong Zu large yellow croakers from government ocean zones, which can be purchased in Ken Yuan breeding nursery, Ningde, Fujian Province. Firstly, it was a kind of green-house spring seeds with a total length of about 1cm, raising in our eco-breeding test site. Some well-grown fishes, with thier strong body and regular eating diets, would be selected for the experiment. The average numbers of weight and length of those fish are 38.42±2.40g and 14.09±0.77cm,respectively.

Research method

The test will be held in a aquarium with its size of 70cm×55cm×45cm(40cm water) and we will set up 2 parallels. The water in this test was used for breeding these fishes, the temperature is 22℃, the salinity is 25 and the PH number is 6.5. In addtion, NH_4^+-N accounts for 1.13mg/L while NO_2^--N number is about 0.18mg/L. the test will be divided into two stages: one for the upgoing trend of temperature, the other for the downwards trend. During the former stage, we will use a heating rod to warm it,and bring it up to the death temperature with a rate of 1℃/h. However, in the second stage, we will use some icy seawater to reach the temperature at about 1℃ per hour. All the processes will be recorded to realize the conditions of these tested fishes.

Measures for data on blood and biochemistry

We decided to use 22℃ as our standard group to test the serum and biochemistry data on Pseudosciaena crocea in both high and low temperature. By extracting some blood from the fishtail vein through a one-time intravenous injector, and by placing it into a 1.5ml centrifuge tube for 5h at 4℃,with a rate of 4000r/min for 10 min,we could collect some upper serum and preserve it. All the data will be tested by a full-automatic biochemistry analyze machine(Ri Li 7600). The test index are :ALT, AST, LDH, ALP, TG, CHOL.

The data will be tested and analyzed by SPSS19.0. If there were some obvious differences(P<0.05),then a Duncan's will be used for multiple comparison analyze.

TEST RESULT

Juvenile yellow croakers' tolerance for temperature

TABLE 1 : Effects of high temperature on juvenile *Pseudosciaena crocea*

Tem. (℃)	Fish Condition
22～28	No Change
28～31	More active
31～34	Uncomfortable, swimming fastly, uploading and rolling over
35～36	Half of fishes are rolling over, lose their balance, uploading, lay on the water surface, one or two die.
37	Most fishes are rolling over,lay on the water surface, convulsion, the death rate is about 36.67±7.64%
38	The majority of fish are rolling over, lay on the water surface, convulsion, the death rate is about72.33±8.74%

From TABLE.1 we can conclude that in high temperature process, juvenile fishes are normal when the water temperature rises from 22℃ to 28℃. Then they become more active between 28℃ and 31℃. However, when the temperature is over 31℃, they feel uncomfortable and start to die beyond 35℃. The majority of fish will keep dying over 37℃. Thus, it is clear that in such an environment the critical upper temperature limit for juvenile Pseudosciaena crocea seems to be 37℃.

TABLE 2 : Effects of low temperature on juvenile *Pseudosciaena crocea*

Tem. （℃）	Result
22～15	normal
15～11	Less active
11～9	Nearly no swiming, some are
9～8	Most fishes are lying on one side at the bottom, unbalanced, one or two die
8～7	Most fishes are rolling over、convulsion,the death rate is 28.33±5.77%
7～6	Most fishes are rolling over、convulsion,the death rate is 41.67±5.77%

From TABLE 2, we can conclude that during the low temperature process,the fishes are normal when the temperature decreased from 22°C to 15°C and were less active between the are of 15°C to 10°C. They became uncomfortable when the water temperature went down below 10°C. Some fishes lied on their side and began to die at 8°C, they continued to die in a large quantity below 8°C. Hence, it is clear that in such an environment the critical lower temperature limit for juvenile Pseudosciaena crocea seems to be 8℃.

The change of serum and physiology data on juvenile Pseudosciaena crocea in different temperatures

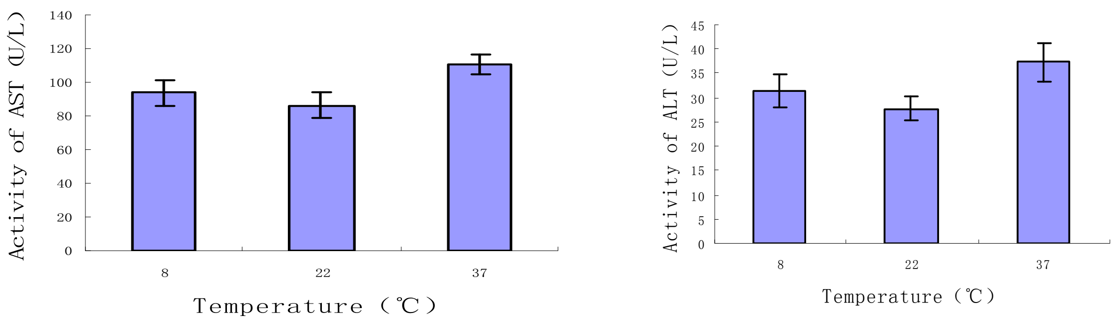

Figure 1 : Effects of different temperature on ALT and AST activities of juvenile *Pseudosciaena crocea*

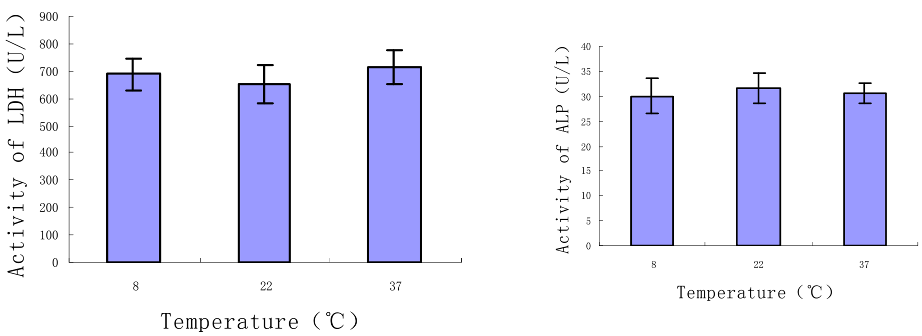

Figure 2 : Effects of different temperature on LDH and ALP activities of juvenile *Pseudosciaena crocea*

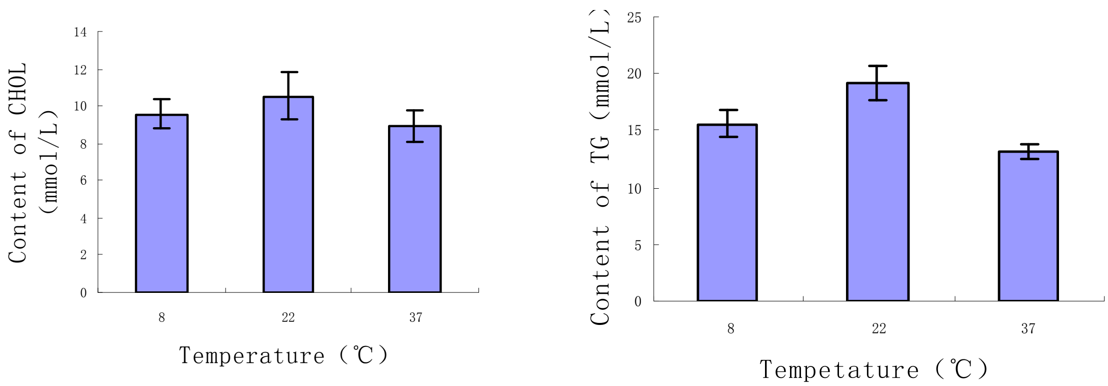

Figure 3 : Effects of different temperature on content of LDH and ALP in serum of juvenile *Pseudosciaena crocea*

From Figure 1 we can conclude that when the water temperature arrived at37℃,the ALT and AST activity of juvenile Pseudosciaena crocea's serum was apparently higher than that in standard groups(P<0.05). But when the temperature went down to 8℃, the gap was disappearing though the activity of ALT and AST still increased.

From Figure 2, we can conclude that the LDH activity of juvenile Pseudosciaena crocea's serum will absolutely increase despite the change of water temperature, but the difference was not very clear(P>0.05). Instead, the ALP activity decreased in the same condition.

From Figure 3, we can conclude that the TG concentration will surely decline despite the change of temperature, but is obviously different to the standard group(P<0.05). Also, the CHOL concentration decreased but there is little distinction compared with the standard group(P>0.05).

DISCUSSION

Juvenile pseudosciaena crocea's tolerance on temperature

In this essay, when the temperature is between 15 and 28℃, juvenile Pseudosciaena croces seem to be normal so that it can be regarded as the thermophilic area to the fish, but when the temperature is up to 37℃ or below 8℃, the fish begin to die in a large quantity. Thus, the critical high limit is about 37℃ while the low one is 8℃. This conclusion is similar to the research result of Xu Zhen[3]. The critical low limit is a little higher than that in his study probably because we use the temperature mutation method in our research. In daily production, with the slight change of water temperature, the limit will also slowly increase or decrease. In addition, the limit temperatures in our study are recorded in our special water conditions. The content of NH_4^+-N and NO_2-N is more higher. According to the Shelford's tolerance law: when the ecological factor of a species is not matched with its most comfortable situation, its tolerance on other ecological factors will be relatively impaired[10]. Hence, if the quality of water could be improved, the tolerant water temperature will be probably widen.

Impact on enzyme in juvenile pseudosciaena croces serum in different temperatures

Enzyme is a catalyst for animal's metabolism. When animals are stressed, their body's metabolism is affected and therefore cause a change in the activity and content of enzyme. ALT and AST are both very important transaminases that are transformed between catalytic amino acid and ketonic acid. And when ALT is released into the serum, it will be more active. AST could also be a catalyst for the ammonia exchange between gluamic acid and oxaloacetic acid and more active in heart. ALT and AST are very essential serums for the damage of liver[11,12]. In our study, when the temperature reaches either high or low limit, the activity of ALT and AST in their body both increase, especially at 37℃. The damage by both high and low temperature to the fishes liver cells and myocardium cells may be cause by the emission of ALT and AST. This conclusion is similar to the Yi Weide[2] study on *Pseuosciaena crocea and* He Fulin[13] study on *Oncorhynchus mykiss.*

In our study, when the temperature reaches its high or low limit, the activity of LDH in their body increases. It is probably because in stressed situations, the fish body will firstly ensure the oxygen supply for heart and central nervous system and use the non-oxygen way in other organism. So the glycometabolism changed from oxygen to non-oxygen glycolysis area to improve the activity of LDH and the ALP metabolism rate to protein and lipid. In our study, when the temperature arrives at the limit, the activity of ALP decreases, which means the reduction of ATP break-down in muscular tissue due to the increase in anaerobic metabolism in stressed situations.

Impact on blood fat in juvenile pseudosciaena croces serum in different temperatures

Fat is involved in fish cells as TG, which is a main material in fish metabolism. CHOL exists in blood as a non-protein form. In this study, when the temperature reaches both the high and low limit, the concentration of TG sharply decrease (P<0.05), the concentration of CHOL also has a slight decrease probably because TG is largely consumed during the thermophilic process as a main storing material. Thus during the high temperature process, due to the large consumption of TG, the concentration at 37℃ is lower than that at 8℃. On the other hand, the damage of liver caused by stressed conditions will prevent the fat from re-absorbing leading to the decrease in TG and CHOL concentration. This conclusion agrees with the result of Yi Dewei research on Pseudosciaena croces and He Fulin[2] and Chang Yumei[14] research on *Cyprinus. carpiovar. wuyuanensis* and *Cyprinus. pellegrini pellegrini*, but is not similar to the result of Liu Bo[15] research on GIFT，*Oreochromis niloticus*. The reason may be the difference in test environment and fish spieces.

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