Bull. Inst. Zool., Academia Sinica **21**(1): 21-26 (1982)

INFLUENCE OF SALINITY ACCLIMATION ON ROUTINE METABOLIC RATE PATTERN IN DIFFERENT SALINITIES OF THE PERCOID FISH, *GIRELLA NIGRICANS* (AYRES)

LEE-SHING FANG

Scripps Institution of Oceanography, University of California, San Diego, La Jolla, California 92093, U. S. A.

(Received November 12, 1981)

Lee-shing Fang (1982). Influence of Salinity acclimation on Routine Metabolic Rate Pattern in Different Salinities of the Percoid Fish, *Girella nigricans* (Ayres). *Bull. Inst. Zool., Academia Sinica* **21**(1): 21-26. *Girella nigricans* osmoregulated and maintained a stable metabolic rate over a range of salinities from 10 to 48‰. Yet, blood hematocrit values increased in 48‰ acclimated fish. Blood osmolarity value changed differently when different acclimated groups were transfered into the same salinity. And the stable metabolic rate pattern with respect to salinity change was altered after acclimation to 10‰ and 48‰. This phenomenon and its possible ecological significance are discussed.

Numerous studies have demonstrated the tolerance of salinity variations, and the osmotic and ionic regulation capacity of fishes[1,2,3,4]. However, the energetic cost of salinity adaptation needs more attention[5]. Previous works in this field such as Framer and Beamish[7], Job[8], Muir and Nimii[9], Nordlie[10], Igram and Wares[11] revealed that. a very important question with respect to the physiological ecology of fishes has received essentially no study, that is, how does metabolic rate affected by acute salinity change following salnity acclimation? Marine fish can easily be trapped in semiclosed lagoons, mashes, swamps, canals and river mouths and becomes partly or fully acclimated to a different salinity region before returning to the ocean. In such cases how would a fish respond metabolically to normal sea water?

Tests were carried out with opaleye, *Girella nigricans*, which as juveniles, live intertidally for one to two years before becoming subtidal[12]. This fish occurs in habitats of isolated tide pools or estuaries, where both hyper- and hyposalinities were found.

MATERIALS AND METHODS

Experimental fish were obtained from the intertidal pools of Dyke rock near the Scripps Institution of Oceanography. They were kept in running sea water and were fed every other day with anchovy and boiled beef liver. Fish weighing from 1.31 to 5.92 gm were used for osmolarity and salinity tolerance experiments, those weighing between 2.02 to 3.20 gm were used for oxygen consumption measurements. Fish were fasten for 24 hrs before use in an experiment.

Salinity tolerance and acclimation

Fish were segregated in 20 groups and transferred into 10 L aquarium providing different salinities (0.5‰, 10‰, 20‰, 34‰, 48‰, 60‰). The salinities were implemented by either diluting the sea water with deionized fresh water or by adding NaCl to elevate the concentration.

Oxygen consumption with respect to salinity change

The oxygen consumption measurement apparatus was basically a circulating, closed system in which the dissolved oxygen can be continuously recorded (YSI 5700 dissolved oxygen probe) and a modified salinity equilibrium reservoir was attached on it[13]. All measurements were made at 21±0.5°C.

Fish was put in a dimly lighted chamber at the appropriate salinity 18 to 24 hrs before the measurements were made. Oxygen content was continuously recorded until the oxygen tension in the system dropped to about 5 ppm. Each time aerated water of new salinity was introduced into the system, about half an hour was allowed for achievement of equilibrium before starting a measurement. All readings were obtained between 0900 am to 1700 pm to minimize possible diurnal metabolic fluctuations. The tidal rhythm should not appear after 2-3 weeks acclimation[14]. Usually a fish was left in the chamber for 3 days to accomplish a test over a salinity range from 10 to 66‰.

Blood osmolarity and hematocrit

Blood was collected from the caudal artery by a heparinized microtubes[6]. The hematocrit was read by a Spiracrit reader and the blood serum osmolarity was measured with a vapor pressure osmometer (Wescor Inc. 5100).

These measurements were made on fish of each acclimated group after 30 days of acclimation as well as made on fish during they were in oxygen consumption rate measurements.

RESULTS

Salinity tolerance

The LD_{50} time of *Girella nigricans* in an acute change to fresh water was 1.5 hrs and that

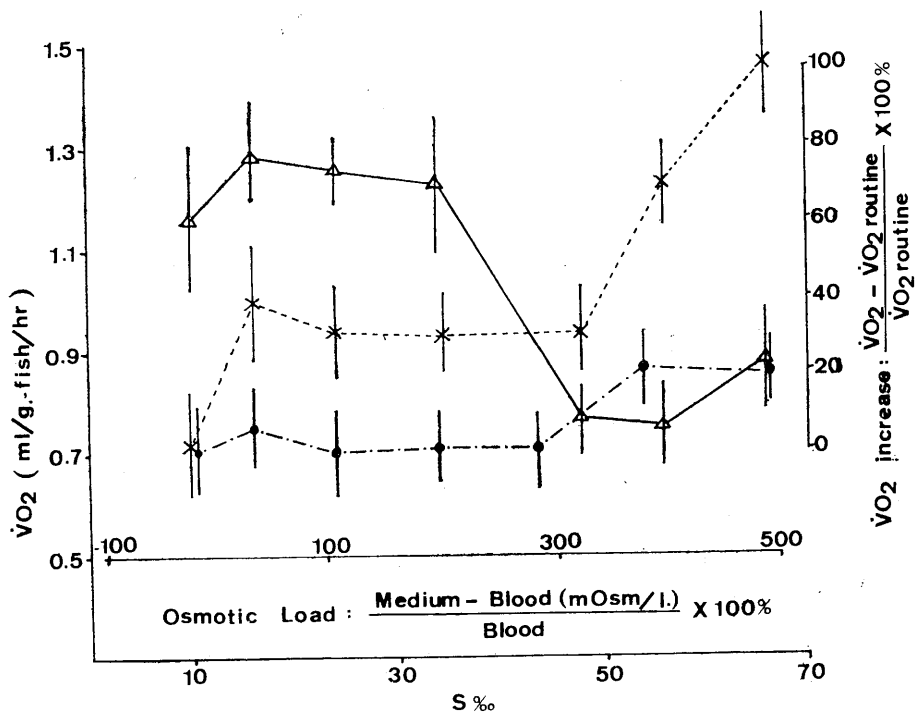

Fig. 1. The relationship of metabolic rate and osmotic load for different acclimated group with respect to salinities.
•: 34‰ acclimated group
△: 48‰ acclimated group
×: 10‰ acclimated group

to 60‰ was 9.1 hrs. Small fish died faster than larger ones. Fish in other salinity groups (10‰, 20‰, 34‰, 48‰) all survived for more than 4 weeks. However, after one month, those in 10‰, 20‰ and 48‰ began to die very slowly (about 1 fish/3 days), but those kept in 34‰ did not. Secondary effects of salinity on nutrition could have occurred[15].

Metabolic rate of different acclimated groups in different salinities

The oxygen consumption of fish from normal sea water was not significantly altered over a wide salinity range from 10‰ to 44‰ (Fig. 1), but increased by about 22% at 56‰ to 66‰. Fish acclimated to 48‰ had a slightly higher routine metabolic rate which increased both at higher and lower salinities. However, at 10‰ some of the fish were near death and had a low metabolic rate. Fish acclimated at 10‰ showed almost the same routine metabolic rate as those in normal sea water; yet at higher salinities the metabolic rate of the group increased; it was relatively constant between 16-48‰ but increased sharply after 48‰.

Blood plasma osmolarity

Fig. 2 shows that *Girella nigricans* is a very good osmoregulator. After 30 days acclimation to 10‰, the fish still had the same blood osmolarity as in normal sea water, And those kept in 48‰ had their plasma osmolarity raised only 13% above normal. This species displayed a typical homeostatic patterns as described by Gilles and Jeuniaux[3].

The blood osmolarity of acclimated fish exposed to different salinities is shown in Fig. 3. Fish acclimated to 48‰ did not have a regulation capacity in 10‰, and fish acclimated to 10‰ began to show a higher blood osmolarity at salinities above 48‰.

The effect of salinity acclimaation on hematocrit

The hematocrit level of 10‰ acclimated fish was slightly lower than those of the 20‰ to 34‰ groups but this difference is not statistically significant. However, the average hematocrit value for the 48‰ group was significantly higher than the other three groups. (Fig. 4) (Student T test, $p<0.05$).

DISCUSSION

The overall metabolic response pattern of *Girella nigricans* from normal sea water to different salinities corresponds to the model I pattern of Nordlie[10], i.e. the rate of oxygen consumption of *G. nigricans* is not significantly altered over a wide environmental salinity range. This permits the fish a euryhaline existence

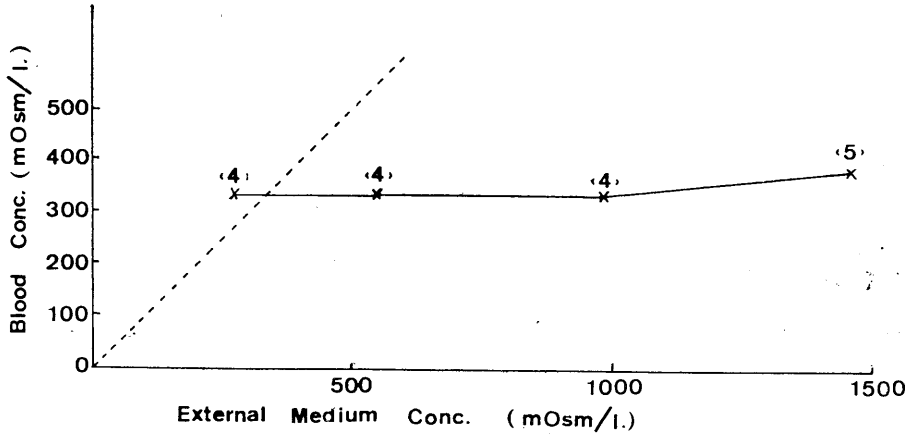

Fig. 2. The mean blood osmolarity of different salinity acclimated groups with respect to their external media osmolarity after 30 days of acclimation. Number in parenthesis represent number of fish sampled.

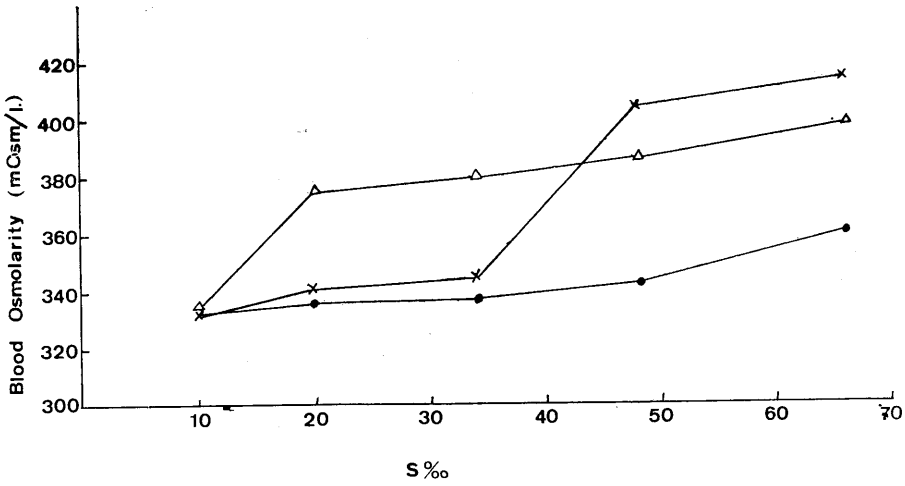

Fig. 3. The blood osmolarity change of each acclimated group after staying in the tested salinities for 1.5 hrs. Each point was the mean of 4 readings.
•: 34‰ acclimated group △: 48‰ acclimated group
×: 10‰ acclimated group

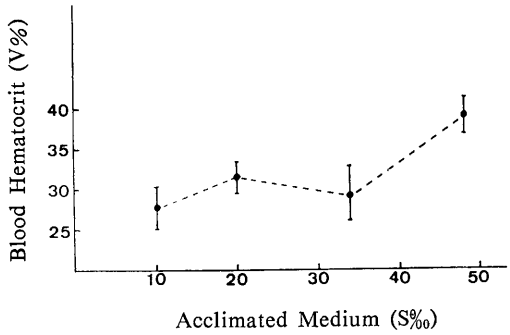

Fig. 4. The hematocrit of each acclimated group after 30 days of acclimation.

unhampered by heavy metabolic cost. After acclimation, the metabolic response of the fish was completely changed. Fish acclimated to high (48‰) or low (10‰) salinities both had changed their metabolic salinity patten to model type III. Their rates of oxygen consumption were minimal in the acclimation salinity but increased with either increasing or decreasing salinity. Plateaus were found over certain salinity ranges in both acclimation groups, a result which was different from the strictly functional change predicted by model III. This modulation of pattern gives rise to an interesting ecological consideration: for example, fish which has been accidentally trapped in sandbar blocked river mouths could become acclimated to lower salinity. When next high tide coming, instead of returning to normal sea water, the fish should actively follow the isosalinity line that they were acclimated to, as predicated by their metabolic rate model. Thus, a physiologically different group is likely to be created by this behavior.

In this experiment the routine metabolic rate of fish acclimated to 10‰ was almost the same as in normal sea water, but that in 48‰ was slightly higher. This agrees with Nordlie's[6] argument that the cost of osmotic regulation is high in hyperosmotic media. He reasoned that the effective permeability in hyperosmotic media is significantly greater than in hypoosmotic media because the surface area across which the fluxes are occurring is greatly increased by the addition of the area of the gut wall (drinking rate higher) relative to that of the gill epithelium. The metabolic rate of *G. nigricans* after acclimation to high salinity (48‰) was lower than it was before. This was due to the inductive production of more osmoregulatory enzyme which

allowed for more efficient ion regulation[16,17]. However, in accordance with Fig. 1, it is clear that the picture was much more complicated. Fish, acclimated to 48‰ had a higher oxygen consumption rate not only when the osmotic load increased, but also when it decreased. Fish acclimated to 10‰ seemed to have lost the regulatory ability it used to have and could not maintain its metabolic rate when facing even small salinity changes. And for any given osmotic load, each acclimation group had consumed different amount of energy to maintain the same homeostatic internal status; while theoretically, the same amount of energy should be required for the same osmotic load. This phenomenon showed that during the period of acclimation, fish had developed some special methods of osmoregulation that allowing them to exist in an abnormal saline environment without having to consume an extra load of energy to regulate. By doing this, they traded with the loss of flexibility.

There was no decrease in the oxygen consumption observed in all acclimated groups when under severe hyper-osmotic stress which might happen due to the shrinkage of exchange surface of the gills[13]. This phase could occur in the first half hour equilibrium period and had not been recorded.

After 30 days, the hematocrit increased 38% in high salinity acclimated (48‰) fish. Grig[18] stated that the amount of oxygen carried by solution in fish blood is often a significant contribution to the total oxygen capacity. Fish acclimated to high salinity had a slightly higher routine metabolic rate and there was less oxygen dissolved in aqueous system of higher salinity. Therefore, oxygen transport capacity of the acclimated fish should increase. Moreover, the plasma osmolarity of this fish also was higher than normal; so less oxygen might be dissolved in the plasma. Thus, the hematocrit increase may act to offset these conditions to meet the oxygen demand. Assuming there is 6% blood volume in a 3 gram test fish, an estimated value approximately 35% plasma oxygen transport capacity loss due to the high salinity acclimation might be obtained. However, the calculated value of the oxygen carrying capacity increase due to hematocrit (hence total hemoglobin) increase is 60 times more than that necessary to compensate for the loss due to the plasma osmolarity increase (1.33×10^{-2} ml O_2/fish: 2.2×10^{-4} ml O_2/fish). Thus, a more likely explanation for the hematocrit volume increase may be that the animals had lost water in higher salinities and reduced the plasma volume[19].

Acknowledgements: The author wish to express his great thanks to Dr. J. Graham for encouraging and helping this work; Dr. G. Somero, Dr. D. Fo, Dr. R. Rosenblatt and Dr. L. Cheng for stimulating discussion and suggestions; Dr. K. H. Chang for valuable help. This work is partially supported by a scholarship from the Ministry of Education, R. O. C.

REFERENCES

1. PARRY, G. (1966) Osmotic adaption in fishes. *Biol. Rev.* **41**: 392-444.
2. CONTE, F. P. (1969) Salt secretion. In "Physiology" Vol. 1, Academic Press, New York. 241-291.
3. GILLES, R. and JEUNIAUX, CH. (1979) Osmoregulation and ecology in media of fluctuation salinity. In "Mechanisms of osmoregulation in animals" Gilles ed., John Wiley and Sons. Chichester, N. Y., Brisban, Toronto. pp. 581-610.
4. EVANS, D. H. (1979) Fish. In "Comparative Physiology of Osmoregulation in Animals". Vol. 1. Maloiy, G. M. O. ed. Academic Press, London, N. Y. pp. 305-390.
5. MORRIS, R. W. (1974) Respiratory metabolism and ecology of fishes. In "Chemical Zoology", Vol. VIII, Florklin, M. and Scheer, B. ed. Academic Press, N. Y., London. pp. 447-469.
6. NORDLIE, F. G. and LEFFLER, C. W. (1975) Ionic regulation and the energetics of osmoregulation in *Mugil cephalus* Lin. *Comp. Biochem. Physiol.* **51A**: 125-131.
7. FRAMER, G. J. and BEAMISH, F. W. H. (1969) Oxygen consumption of *Tilapia nilotica* in relation to swimming speed and salinity. *J. Fish. Res. Bd. Can.* **26**: 2807-2821.

8. JOB, S. V. (1969) The respiratory metabolism of *Tilapia mossambica*; The effect of size, temperature, salinity and partial pressure of oxygen. *Marine Biology* **3**: 222-226.
9. MUIR, B. S. and NIMII, A. J. (1972) Oxygen consumption of the euryhaline fish aholehole (Kuhlia sandvicensis) with reference to salinity, swimming and food consumption. *J. Fish. Res. Bd. Can.* **29**: 67-77.
10. NORDLIE, F. G. (1978) The influence of environmental salinity on respiratory oxygen demand in the euryhaline teleost, Ambassis interrupt Bleeker. *Comp. Biochem. Physiol.* **59A**: 271-274.
11. IGRAM, R. and WARES, W. D. (1979) Oxygen consumption in the fathead minnow (*Pimephales promelas*)- Effect of PH, osmotic pressure and light level. *Comp. Biochem. Physiol.* **62A**: 895-897.
12. NORRIS, K. S. (1963) The functions of temperature in the ecology of percoid fish *Girella nigricans* (Ayres). *Ecol. Mono.* **33**: 23-62.
13. BATH, R. N. and EDDY, F. B. (1979) Ionic and respiratory regulation in rainbow trout during rapid transfer to seawater. J. *Comp. Physiol.* (B) **134**: 351-357.
14. GIBSON, R. N. (1967) Experiments on the tidal rhythm of *Blemmius pholis*. *J. Mar. Biol. Ass. U. K.* **47**: 97-111.
15. KINNE, O. (1960) Growth, food intake, and food conversion in euryplastic fish exposed to different temperatures and salinities. *Physio. Zool.* **33**:288-317.
16. KAMIYA, M. and UTIDA, S. (1968) Change in activity of Na-K activated ATPase in gill during adaptation of the Japanese eel to sea water. *Comp. Biochem. Physiol.* **26**: 675-685.
17. KAMIYA, M. and UTIDA, S. (1969) Na-K activated ATPase in gill of fresh water, marine and euryhaline teleosts. *Comp. Biochem. Physiol* **31**: 671-674.
18. GRIGG, G. C. (1974) Respiratory function of blood in fish. In "Chemical Zoology: VIII, Florklin, M. and Scheer, B. ed. Academic Press, N. Y. London. pp. 331-367.
19. BITTNER, A. and LANG, S. (1980) Some aspect of the osmoregulation of Amazonian freshwater stingrays. (*Potamotrygon hystrix*)-I. Serum osmolarity, sodium and chloride content, water content, hematocrit and urea level. *Comp. Biochem. Physiol.* **67(A)**: 9-13.

鹽度馴化時不同鹽度對瓜子鱲 *Girella nigricans* (Ayres) 基本新陳代謝之影響

方 力 行

本實驗在研究經鹽度馴化後不同鹽度對 *Girella nigricans* 基本新陳代謝之影響。*G. nigricans* 置於鹽度 10 至 48‰範圍內時，能調節其體內之滲透壓，並維持一穩定之新陳代謝率。若實驗魚先經過不同的鹽度馴化後，其原本對外界鹽度改變之新陳代謝守恒範圍將爲之減小。不同馴化羣置於同一鹽度下，其餘內滲透壓及新陳代謝率均不相同，馴化於 48‰之樣品羣，血紅球並有顯著增加。