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INFLUENCE OF SALINITY ON THE WEIGHT-DEPENDENT METABOLISM OF THE TADE MULLET, *LIZA TADE* (FORSSKAL) (PISCES: MUGILIDAE)

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Abstract: Studies on the salinity tolerance of Tade Mullet *Liza tade* have shown that they can live well in salinities ranging from freshwater to 45 ppt salinity. The relationship between total oxygen consumption and body weight in various salinities recorded progressive increase in total oxygen consumption with weight. The mean oxygen consumption was 2.3782 mlh^{-1} in 10 ppt salinity and 3. 3683 mlh^{-1} in freshwater and differed significantly in various salinity ranges. The relationship between total ammonia excretion and body weight was positively correlated and the mean ammonia excretion differed in various salinity ranges. The mean ammonia quotient (AQ) was 0.099 in 10 ppt salinity and 0.124 in freshwater; it differed significantly among different test salinities. The weight exponents obtained for *L. tade* was less than one indicating that the total metabolism increases more slowly than weight and that the weight specific metabolic rate is inversely related to total metabolism in all weight groups studied. Oxygen consumption, ammonia excretion and AQ were lowest in 10 ppt salinity. The minimum energy needed for osmoregulation in *L. tade* was recorded as 15 ppt and isosmotic salinity, 10 ppt.

***Key words*:** Salinity tolerance, ammonia excretion, oxygen consumption, osmoregulation, mullet

INTRODUCTION

Fishes belonging to the family Mugilidae, popularly known as mullets, are known for their ability to live in waters of varying salinity (Oren, 1981). They are a major source of subsistence protein requirements of the peoples of many countries (ICLARM, 1980), and represent ideal candidate species for brackishwater aquaculture. Growth and metabolism of euryhaline species is often affected by salinity because the energy used for osmoregulation is not available for growth (Wootton, 1990). The influence of environmental factors on the metabolism of different groups of teleosts has been studied (Nordlie *et al.,* 1991). The index for estimating the metabolic rate of fishes is often considered as its oxygen consumption (Winberg, 1956; Fry, 1957). The oxygen consumption of fish in relation to salinity has been studied in teleosts (DeSilva *et al*., 1986; Barton and Barton, 1987; Walsh *et al.,* 1989, 1991). Among mullets, the studies on the influence of salinity in oxygen consumption are generally limited to the grey mullet, *Mugil cephalus* (Marais, 1978; Mathew, 1976; Walsh *et al.,* 1989). Information regarding salinity tolerance and metabolism of the Tade mullet *Liza tade* is not available.

Ammonia can be extremely toxic to fish if allowed to accumulate in the body and ammonia production must be balanced by excretion. Since most of the nitrogen excreted by fish is in the form of ammonia, it is felt that a reasonable pattern of protein degradation can be obtained from the measurements of ammonia. Ratio of ammonia quotient (AQ) is often used as an index of substrate utilization in aquatic animals (Conover and Corner, 1968; Corner and Cowey, 1986; Mayzaud, 1973). AQ has also been used as index of energy utilization from carbohydrate and/or protein in fishes (Stroganov, 1956; Kutty, 1972). This paper reports on the oxygen consumption and ammonia excretion of *L. tade* acclimated to different salinities.

MATERIALS AND METHODS

Specimens of *Liza tade* collected from Veli Lake and acclimated to laboratory condition were used for the study. The Veli Lake is a small (9.56 ha waterspread area) brackish water body situated 12 km northwest of Thiruvananthapuram city (8° 28¢ N lat.; 76° 57' E long.) in India, which remains separated from the Lakshadweep Sea by a sand bar for the major part of the year.

Salinity tolerance: For this study, the juveniles ranging from 0.40 - 0.50 g wet weight and adults ranging from 3-20 g wet weight were acclimated in salinity as that of collection site (5 ppt.) for about two weeks and fed with formulated feed of 24 per cent protein. Oxygen content was kept near air saturation by continuous aeration and temperature kept remained 29 ± l^{0}C.

Two sets of experiments were conducted on juveniles in the first set. Acclimated fish were suddenly transferred to each test salinity (5, 10, 15, 20, 25, seawater and fresh-water) and in the second set fish were gradually acclimated and transferred to each test salinity and observed for seven days.

Respiratory metabolism: Specimens of 3-20 g wet weight (5.4-10.6 cm standard length) collected from Veli Lake were used for this study. The apparatus used for the study was a modification of Fry respirometer (Kutty *et al.,* 1971). Dissolved oxygen was measured by the Winkler method (APHA, 2005). Total ammonia was measured by direct Nesslerisation method using spectrophotometer (Martin, 1968).

The data relating oxygen consumption/ammonia excretion/AQ and weight groups in various salinities were subjected to regression analysis using the equation $y = ax^b$ and its $\log y = \log a + b \log x$, where 'y' the oxygen consumption/ ammonia excretion/AQ, 'x' the weight and 'a' and 'b' constants (Snedecor and Cochran, 1967). The data obtained on oxygen consumption, ammonia excretion and AQ in various test salinities were compared by the analysis of covariance (ANCOVA) (Snedecor and Cochran, 1967).

RESULTS

On direct transfer from 5 ppt salinity, 100 per cent survival was observed at 5-25 ppt salinity. In seawater only 70 per cent survival and in fresh-water, 100 per cent mortality within 72 hr itself was observed. The mortality percentage on gradual change of salinity showed that all fishes survived in seawater (33 ppt), 95 per cent each survived in freshwater and in 45 ppt salinity.

On direct transfer from 5 ppt salinity adults of *L. tade* were able to tolerate freshwater to seawater and 100% survival was found at 5 ppt seawater. However, only 90 per cent survival was found at freshwater. The second series of experiments showed that on gradual change, 100 per cent survival was obtained at freshwater.

The total oxygen consumption (mlh^{-1}) in relation to weight in test salinities were subjected to regression analysis and the regression equations and weight exponents are presented in Table 1. The relationship between total oxygen consumption and body weight in seven salinities studied were found to be significant ($P < 0.01$) and is positively correlated. Lines of best fit were drawn using the principle of least-squares. The relationship between total oxygen consumption and body weight in seven salinities recorded progressive increase in total oxygen consumption with weight.

The data was subjected to analysis of covariance and is represented in Table 2. The slopes of regression lines of test-salinities did not differ significantly ($F = 0.127$). However, the mean oxygen consumption differed significantly ($F = 219$; $P<0.01$). The mean O_2 consumption was 2.3782 mlh^{-1} in 10 ppt salinity and 3. 3683 mlh^{-1} in freshwater (Table 1).

The data on total ammonia excretion as a function of bodyweight in seven salinities were statistically analyzed and the regression equations, weight exponents and mean ammonia-excretion are represented in Table 3. The relationship between total ammonia excretion and bodyweight was positively correlated as weight increased, total ammonia excretion increased in all salinities. The relationship between total ammonia excretion

Table 1. Regression equations relating total oxygen consumption ('Y' in mlh^{-1}) and weight ('X' in g) in seven test salinities

Salinity (ppt)	Weight range (g)	Regression equation log Y = log a + b log x	Regression coefficient (b)	Correlation coefficient (r)	Mean Oxygen consumption (mlh^{-1})
Fresh water	3.15-19.62	log Y = 0.5519 + 0.9251 log X	0.9251	0.9985**	3.3683
5	3.00-19.00	log Y = 0.5164 + 0.9045 log X	0.9045	0.9999**	2.9093
10	3.12-20.00	log Y = 0.4186 + 0.8966 log X	0.8966	0.9988**	2.3782
15	3.10-20.05	log Y = 0.4435 + 0.9007 log X	0.9007	0.9989**	2.5333
20	3.21-19.15	log Y = 0.5197 + 0.9036 log X	0.9036	0.9997**	2.9426
25	3.05-19.87	log Y = 0.5452 + 0.9014 log X	0.9014	0.9993**	3.1706
Sea water	3.00-19.57	log Y = 0.5512 + 0.9224 log X	0.9224	0.9991**	3.3247

** P<0.01

Table 2. Analysis of covariance comparing relationship between total oxygen consumption and body weight among test salinities

Source	df	SS	MS	F
Within				
1. Fresh water	4	0.000944	0.000236	
2. 5 ppt salinity	4	0.000063	0.000016	
3. 10 ppt salinity	4	0.000975	0.000244	
4. 15 ppt salinity	4	0.000669	0.000167	
5. 20 ppt salinity	4	0.000183	0.000046	
6. 25 ppt salinity	4	0.00005	0.000013	
7. Seawater	4	0.000654	0.000164	
	28	0.003538	0.000126	
Pooled W	34	0.003636	0.000106	
Difference among slopes	6	0.000098	0.000016	
W + B	40	0.1432	0.00358	0.127
Difference among adjusted means	6	0.1395	0.023261	219**

** P<0.01

Table 3. Regression equations relating total ammonia excretion ('Y' in mlh^{-1}) in seven test salinities

Salinity (ppt)	Regression equation log Y = log a + b log x	Regression coefficient (b)	Correlation coefficient (r)	Mean Ammonia excretion (mlh^{-1})
Fresh water	log Y = 0.6910 + 0.8783 log X	0.8783	0.9978**	0.412
5	log Y = 0.6260 + 0.8619 log X	0.8619	0.9997**	0.336
10	log Y = 0.4518 + 0.8598 log X	0.8598	0.9982**	0.233
15	log Y = 0.5215 + 0.8609 log X	0.8609	0.9988**	0.274
20	log Y = 0.6298 + 0.8612 log X	0.8612	0.9995**	0.341
25	log Y = 0.6600 + 0.8644 log X	0.8644	0.9989**	0.375
Sea water	log Y = 0.6783 + 0.8778 log X	0.8778	0.9983**	0.398

** P<0.01

Table 4. Analysis of covariance comparing the relationship between total ammonia excretion and weight among various salinities

Source	df	SS	MS	F
Within				
1. Fresh water	4	0.00145	0.00036	
2. 5% salinity	4	0.00025	0.00006	
3.10% salinity	4	0.00108	0.00027	
4.15% salinity	4	0.00029	0.00007	
5.20 % salinity	4	0.00064	0.00016	
6.25 % salinity	4	0.00086	0.00021	
Seawater	4	0.00114	0.00029	
	28	0.00571	0.0002	
Pooled W	34	0.00588	0.00017	
Difference among slopes	6	0.00017	0.00003	
W + B	40	0.3271	0.00818	0.15
Difference among adjusted means	6	0.3212	0.05354	315**

** P<0.01

Table 5. Regression equations relating total ammonia quotient (Y) and body weight ('X' in g) in seven test salinities

Salinity (ppt)	Regression equation log Y = log a + b log x	Regression coefficient (b)	Correlation coefficient (r)	Mean Ammonia quotient (mlh^{-1})
Fresh water	log Y = 1.1390 -0.0466 log X	-0.0466	0.9516**	0.124
5	log Y = 1.1093 -0.0422 log X	-0.0422	0.9697**	0.117
10	log Y = 1.0304 -0.0340 log X	-0.034	0.9421**	0.099
15	log Y = 1.0778-0.0398 log X	-0.0398	0.9645**	0.109
20	log Y = 1.1101-0.0423 log X	-0.0423	0.9724**	0.117
25	log Y = 1.1149-0.0371 log X	-0.0371	0.9735**	0.119
Sea water	log Y = 1.1271-0.0446 log X	-0.0446	0.9452**	0.121

** P<0.01

Table 6. Analysis of Covariance comparing relationship between Ammonia Quotient (AQ) and body weight among test salinities

Source	df	SS	MS	F
Within				
1. Fresh water	4	0.000078	0.000019	
2. 5 % salinity	4	0.000066	0.000017	
3. 10% salinity	4	0.000099	0.000025	
4. 15% salinity	4	0.000029	0.000007	
5. 20 % salinity	4	0.000003	0.000001	
6. 25 % salinity	4	0.000021	0.000005	
Seawater	4	0.000056	0.000014	
	28	0.000352	0.000013	
Pooled W	34	0.0004	0.000012	
Difference among slopes	6	0.000048	0.000008	
W + B	40	0.0395	0.00099	0.615
Difference among adjusted means	6	0.0391	0.00652	243**

** P<0.01

and bodyweight was positively correlated ; the relationship was statistically significant (P< 0.01) in all test salinities.

The data was subjected to analysis of covariance and the results are shown in Table 4. The slopes of regression lines did not differ among test salinities (F = 0.150). However, mean ammonia excretion differed among different - salinities studied and was significant (F = 315; P< 0.01). The mean ammonia excretion was 0.2334 mlh^{-1} in 10 ppt salinity and 0.4119 mlh^{-1} in freshwater (Table4).

The AQ for all weight class studied were subjected to statistical analysis (Table 5). The data were subjected to analysis of covariance (Table 6). The slopes of regression lines among test salinities did not differ significantly (F = 0.615). However, mean AQ differed among different test salinities and the relationship was significant (F = 543; P< 0.01). The mean AQ was 0.099 in 10 ppt salinity and 0.124 in freshwater (Table 6).

DISCUSSION

The distribution of wild animals is ruled by a combination of environmental, biotic and stochastic factors. Mullets are highly euryhaline and thrive in a wide range of salinities (Cardona, 2006). Results of the present study revealed that *Liza tade* can tolerate salinities up to 45 ppt and can also be used for rearing in such conditions. Studies on the salinity tolerance of other grey mullets have shown that they can live well in salinities ranging from freshwater to hypersaline conditions (Odum, 1970; Chervinsky, 1974; Wallace, 1974; Zisman and Ben-Tuvia, 1975). Adults and 95 per cent survival of fry in freshwater on gradual acclimation. However, on direct transfer both fry and adults showed mortality in freshwater, mortality being severe in fry (100%).

Increased survival rate of adults in freshwater can be attributed to the fact that in fishes increase in size or age is an important factor determining their ability to cope up with salinity fluctuations (Watanabe *et al.,* 1985, 1990). Studies have recorded strong preference of mullets for sites with a salinity level under 15, although adults may prefer more saline areas (Cardona, 2000, 2006; Chang *et al.,* 2004). Present results show that *L. tade* can be successfully cultured in various salinity regimes.

In *L. tade* the total oxygen consumption and ammonia excretion increased with increase in weight at a statistically significant level in all test salinities. However, weight specific consumption and ammonia excretion decreased with increase in weight. Similar findings were reported for other species of fish (Jobling, 1981, 1982; Hakim *et al.,* 1983; Eccles, 1985).

The weight exponents obtained for *L. tade* was less than one indicating the total metabolism increasing more slowly than weight and that the weight specific metabolic rate is inversely related to total metabolism. Intraspecific variation of weight exponents in fish may be due to various environmental/test conditions like temperature, salinity, etc (Marais, 1978; Hakim *et al.,* 1983).

In all weight groups studied, oxygen consumption, ammonia excretion and AQ were lowest in 10 ppt salinity. Hence in *L. tade* 10 ppt salinity can be considered to be the isosmotic salinity where the least or 'zero' energy expenditure for osmotic and ionic regulation is needed. That may be the reason for lowest oxygen consumption and ammonia excretion in 10 ppt salinity. The rate of oxygen consumption has been reported to be lowest in the near isosmotic salinity (Nordlie, 1978; Wohlschlag and Wakeman, 1978). In general, minimum energy needed for osmoregulation in *L. tade* was found in 15 ppt and isosmotic salinity, 10 ppt.

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