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Hypoxia tolerance of neotropical fish culture candidates

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Abstract

Diurnal O_2-variations with periods of severe hypoxia ($O_2<0.5 mg.l^{-1}$) overnight frequently occur in tropical fish ponds. Both, the neotropical herbivorous serrasalmids *Colossoma* spp. and the omnivorous characids *Brycon* spp. show excellent growth rates under these conditions. Ecological and experimental investigations demonstrate their extreme tolerance towards hypoxia because they are capable of utilizing the oxygen rich surface layer of the water for respiration. For this purpose an ecomorphosis has been developed, involving the formation of a dermal extension on the lower jaw. The physiological importance of the so called aquatic surface respiration (ASR) for the oxygen supply of the organism is demonstrated by blood gas analyses. The influence of O_2 concentration on the activity and routine circadian O_2 consumption is demonstrated for *C. macropomum*. Investigations on O_2-induced migrations between the zone of macrophyte cover and the open water reveal the ability of *C. macropomum* to survive severe hypoxia beneath macrophyte cover. The importance of the hypoxia tolerance for pond management is discussed.

KEYWORDS: Fish culture, Oyxgen, ASR, Respiration, Floodplain.

Introduction

It is a well known fact that oxygen is essential for fish metabolism. But the required oxygen conditions for obligatory gill-breathers are entirely different from air-breathers, which live permanently under constant oxygen conditions. The reason for this is the specific dissolving capacity of gases in water, depending on the atmospheric pressure and water temperature. In addition, the absolute O_2-concentration in water under saturation conditions is about 30 times lower than in the air. As respiration of fish and mineralization of organic material require large quantities of O_2, the O_2-concentration in water can easily become a limiting factor for the biological productivity of a fish pond.

The limited O_2-dissolving capacity especially concerns tropical fish ponds, dominated by water temperatures of about 30°C with about only 65% dissolved oxygen, compared to saturation conditions at 10°C. Under extensive and semi-intensive culture conditions with minimal water exchange diurnal O_2-fluctuations are normal. At daytime 150% supersaturation can be measured, while at night severe O_2-depletion with concentrations to below 5%, corresponding $0.5 mg.l^{-1}$, are frequent.

Periods of O_2-depletion of such extent cannot be tolerated by most temperate fish species. In Latin America, however, some extremely suitable indigenous aquaculture candidates are found which tolerate such hypoxic conditions. In the following sections, adaptation strategies to O_2-depletion of *Colossoma* and *Brycon*, two Characoids, are presented, and their importance for tropical aquaculture is discussed.

Culture conditions

At the present moment three mostly herbivorous serrasalmids of the genus *Colossoma* are considered as the most suitable indigenous candidates for fish culture in Latin America. *C. macropomum* and *C. brachypomum* naturally occur in the Orinoco and Amazon River systems, whereas *C. mitrei* inhabits the Paraná-Uruguay River. Attempts of culturing these species were summarized by Martinez (1984), Saint-Paul (1986), and Woynarovich (1986).

None of the mentioned species spawn naturally under culture conditions, but all species are reproduced artificially. Hypophysation is most common. However, positive results are reported as well with combined dosages of hypophysis extract and purified hormones.

There is only limited information on the nutritional requirements of *Colossoma* spp. All examined species can survive on both artificial as well as natural feed. But artificial feed should contain at least 30% protein.

The stocking densities under monoculture conditions range from about 1 200 to 10 000 fish.ha^{-1}. The maximum production per year of *C. macropomum* is about 9t.ha^{-1}. In Venezuela in a commercial farm with an annual production capacity of 300-400t, fry takes about 14 months to grow to a commercial size of 2-3kg. There are also promising polyculture attempts with *C. macropomum* together with tilapia and carp. At a combined stocking density of 10 000 fish.ha^{-1}, a production of 8.9t.ha^{-1} can be achieved.

The growth rate of *C. brachypomum* seems to be less promising. The highest production per year of 8.3t.ha^{-1} was achieved at a stocking density of 10 000 fish.ha^{-1}. The temperature optimum of both species lies between 28 and 32°C.

Under extensive and semi-intensive culture conditions, *C. mitrei* grows to a final weight of 557g within 1 year, corresponding to an annual production of 5.9t.ha^{-1}. The temperature optimum of this species is between 20 and 28°C.

In contrast, the omnivorous *Brycon cephalus* and *B. erythropterus* seem to be the most promising species of the characid family (Saint-Paul, 1986). They can only be artificially reproduced by hypophysation. Artificial diets should contain about 35% protein, but the amount of animal protein in the feed may be quite small. In pond experiments, best individual growth rates of more than 500g.yr^{-1}, corresponding to an annual production of about 2t.ha^{-1}, were achieved under semi-extensive conditions. Under extensive conditions fish grow from 2 to 1 096g within 476 days.

Aquatic surface respiration

It was experimentally demonstrated, that the locomotory behavior of *Colossoma* and *Brycon* species is relatively constant at O_2-concentrations above 1mg.l^{-1} (Saint-Paul and Soares, 1987). However, when the concentration reaches 0.5mg.l^{-1} the fish come to the surface in order to practice aquatic surface respiration (ASR), which is indicated by increased swimming activity. A dermal swelling at the lower jaw appears after longer exposure to low O_2-concentrations. Such an unusual kind of lip formation is demonstrated so far for characoids of the genera *Colossoma*, *Brycon*, *Mylossoma*, and *Triportheus* (Braum and Junk, 1982; Saint-Paul and Soares, 1987, 1988). After areation of the water, the lip slowly retrogresses to its original size.

During ASR the top of the head protrudes out of the water and the lower jaw with its dermal extension was found to move rhythmically, while the body axis depending on the species, keeps a specific angle of 30 - 45° to the surface. The fish are, as a result, able to survive a couple of hours. As the histological sections show no unusual aggregations of blood vessels the dermal extension has obviously only a hydrodynamic function for utilizing the oxygen rich surface layer of the water for respiration (Braum and Junk, 1982; Saint-Paul and Soares, 1988).

The macrophyte zones of the Amazon floodplain lakes are very important as fish "nurseries" (Bayley, 1982) and tropical fish ponds are very often covered partly by floating macrophytes. The natural fish population is mostly found between and under the macrophytes. During normal O_2 concentrations, both *Brycon* and *Colossoma* prefer the macrophyte cover and show significantly less swimming activity than in the open water. It is interesting to examine to what extent the fish are forced to leave the area of macrophyte covering for ASR, since their access to the surface is blocked by the plants. By using sonar it was demonstrated that during night, when the O_2-concentration becomes extremely low, there is a measurable increased fish density in the open water, thus indicating an

O_2-induced diurnal pattern of horizontal migrations between the zone of macrophyte cover and the open water (Saint-Paul and Soares, 1987).

Supplemental experimental investigations seem to indicate that *C. macropomum* can deviate from this migration pattern. During longer periods of O_2-depletion, the fish return to the region of macrophytes and survive there, apparently without the usual kind of ASR. Mortality studies performed in net cages exposed in a natural lake environment, confirm its ability to survive severe hypoxia beneath macrophyte cover. This makes it very likely that *C. macropomum* can utilize the oxygen released from the macrophyte's rhizoid system (Jedicke, 1987).

Oxygen consumption

So far, the experiments yielded only detailed information on oxygen consumption of *C. macropomum*. The data are presented in the following exemplification. The influence of body weight on the oxygen consumption is demonstrated in Table I. The slope of the presented curve is temperature dependent, increasing from 0.64 at 25 and 30°C to 0.78 at 35°C. The metabolic rate of a fish weighing 100g increases from 103.7mg $O_2.kg^{-1}.h^{-1}$ at 20°C to 289.7mg $O_2.kg^{-1}.h^{-1}$ at 30°C, and decreases to 212.4mg $O_2.kg^{-1}.h^{-1}$ at 35°C (Saint-Paul, 1983).

However, the O_2-consumption clearly shows a circadian pattern (Saint-Paul, 1985, 1988). The diurnal O_2-consumption was found to be always significantly greater than the average for 24h, while it was significantly less at night-time. A circadian rhythm in the O_2-concentration of the water with a minimum during early morning does not change the circadian pattern in the O_2-consumption. It is significant, that the maximum O_2-consumption with an increase of about 60% coincides with the early morning O_2-depletion in the water.

The investigations on the influence of the O_2-concentration on the O_2-demand furthermore demonstrate that above $2mg.l^{-1}$ the respiration can be maintained independent from O_2-concentration in the water by regulation of the breathing pattern. Between 0.5 and $2.0mg.l^{-1}$ there is a reduction of the O_2-demand of 70% (Saint-Paul, 1984).

Data on the O_2-consumption during ASR are not available from the literature. However, it can be estimated that for instance a 500-g fish has an O_2-demand of 54 $mg.h^{-1}$ at 25°C. The efficiency of gas exchange during ASR is about 60% and the swimming speed is 190 body-lengths.h^{-1}. As the O_2-concentration in the surface layer is known during ASR, it can be calculated that *C. macropomum* meets about 10% of its O_2-demand during ASR. Preliminary measurements indicate an increase in the pO_2 of the blood during ASR of about 25%. In combination with the high O_2 affinity of the hemoglobin of *C. macropomum* this might help avoiding a toxic acidosis of the blood.

Another important adaptation for surviving low oxygen concentrations is the size of the respiratory surface area of the gills, which is unusually large in *C. macropomum* compared to other freshwater fish. A fish of 200g has a respiratory area of $350mm^2.g^{-1}$ (Saint-Paul, 1982).

Discussion

Investigations on the biology of *Brycon* spp. and *Colossoma* spp. have revealed that especially the juveniles, due to their nutritional requirements, migrate into the macrophyte belts of the floodplain lakes, where extremely low O_2-concentrations frequently occur (Bayley,

Table I. Influence of body weight (W) on the routine oxygen consumption (O) ($mg.h^{-1}$) of *Colossoma macropomum* at different ambient temperatures

25°C	$Q=1.01.W^{0.64\pm0.03}$
30°C	$Q=1.55.W^{0.64\pm0.03}$
35°C	$Q=0.78.W^{0.78\pm0.04}$

1982; Goulding and Carvalho, 1982). The information so far available confirms that these obligate gill- breathers are capable of surviving oxygen depletion by exploiting the water surface layer.

ASR seems to be a common adaptive reaction of gill-breathing fish to oxygen depletion. Gee et al. (1978) note such adaptation strategies among 85% of the 26 fish species in the Great Lakes of North America. In Panama, 93% of 27 non air-breathing fish from a forest stream show ASR under hypoxic conditions (Kramer, 1983). Kramer and McClure (1982) demonstrate the capability of this behavior in 94% of the 31 tropical ornamental fish species. Junk et al. (1983) found that about 30% of the 134 species in an Amazon floodplain lake are able to survive hypoxia and 27% of these display an ecomorphosis.

Sonar data clearly reveal that during night, when the O_2- concentration becomes extremely low, a measurable increase in the fish density in the open water of an Amazon floodplain occurs, thus indicating an O_2-induced horizontal migration between the macrophyte zone and the open water (Saint-Paul and Soares, 1987).

Additional experiments demonstrate that not all fish species necessarily take part of such migration. *C. macropomum*, e.g. is able to survive oxygen depletion below the floating macrophytes without displaying ASR (Saint-Paul and Soares, 1987). *Brycon* spp., however, need free surface for ASR. The importance of the macrophyte potential to release O_2 into the water, as discussed by Armstrong (1979) and positively checked out by Jedicke (1987) for *Eichhornia crassipes* and *Pistia* sp., needs more detailed ecological investigation, in order to understand how fish survive with a minimum O_2-supply under the macrophyte cover.

Short-term fluctuations of the O_2-concentration can be compensated for by changes in the ventilation rate. However, O_2- concentrations below a species-specific concentration can not be compensated any longer. A significant reduction of the O_2- consumption rate is the consequence. Specific information is available for only *C. macropomum*. Respiratory experiments confirmed dawning activity patterns, which are supported by ecological investigations as "sunrise-active-species" (Goulding, 1980; Saint-Paul, 1982, 1983). The actual oxygen consumption by *C. macropomum* shows, that the metabolic rate of this species is just about as high as those of other tropical fish species (Saint-Paul, 1983). Corresponding data for *Brycon* spp. and the other two *Colossoma* species are not available so far.

These investigations are of great importance for the evaluation of the aquacultural suitability of these species. Especially tropical fish ponds are characterized by extreme diurnal and seasonal changes of the O_2-concentration. This is demonstrated among others by Braum and Junk (1982), da Costa et al. (1986), Nascimento et al. (1986), and Saint-Paul and Soares (1987). That is why aquaculture under extensive conditions can only be practiced with species tolerating such conditions.

The significance of these findings for practical aquaculture is obvious, if one attempts to calculate the maximum stocking density for a simple pond in which the oxygen concentration is the limiting factor. The results reported above show that among other factors the oxygen demand is dependent upon temperature, oxygen concentration, and weight of the fish. The amount of oxygen available for a fish depends on the flow rate of the water passing through a pond, the input of oxygen by diffusion from the atmosphere, and the amount of photosynthesis in the pond. The oxygen is, on the other hand, removed by respiration and by decomposition of organic material.

The oxygen release during photosynthesis, which is subject to large circadian and seasonal fluctuations is neglected in the model calculation, in order to simulate the most unfavourable situation. The following equation can be used to describe the relationship between the stocking density and the other factors mentioned above (Saint-Paul, 1982):

$$N = \frac{(Z-A).D + B}{2\,Q_T + O.W} + V\,(ha^{-1})$$

Where: N = stocking density (ha^{-1}); Z = oxygen concentration of the inflowing water ($mg.l^{-1}$); A = oxygen concentration of the out-flowing water ($mg.l^{-1}$); D = flow rate through the pond ($l.h^{-1}$); B = oxygen input from the atmosphere, set at

Table II. Maximum stocking densities.ha^{-1} for *Colossoma macropomum* based on temperature and desired weight

Temperature (^{o}C)	Desired final weight (g)				
	1	10	100	1 000	5 000
20	1 740 000	350 000	62 000	9 400	2 300
25	880 000	190 000	37 000	6 300	1 700
30	550 000	120 000	34 000	4 300	1 200

$6.25.10^5$mg $O_2.ha^{-1}.h^{-1}$ (Knösche, 1971); Q_T = routine metabolism ($mgO_2.h^{-1}$) of a fish weighing W (g) at a temperature of T°C, the routine oxygen demand is doubled in order to simulate the average natural metabolic level of the wild fish (Winberg, 1956); O = oxygen consumed in the decomposition of excrement and lost feed, set at 0.125 mg $O_2.h^{-1}.g^{-1}$ fish weight (Scherb and Braun, 1971); W = desired final weight (g) at stocking density N; and V = losses due to natural mortality (%).

The model pond used for the calculation is assigned a surface area of 1ha and an average water depth of 80cm. The flow rate of the water is set at the speed that will allow a volume of water equal to the total volume of the pond to be changed each day. The inflowing water is 80% saturated with oxygen, and the outflowing water contains a minimum concentration of $3mg.l^{-1}$. Under these conditions, the maximum stocking densities shown in Table II with their corresponding temperatures will allow the fishes to reach the desired final weight. For obtaining these values, no losses are assumed, since there is no empirical information on what the actual losses were. Additional investigations on the O_2 consumption of *C. macromomum* seem to indicate a lower rate by the factor 0.6-0.8 for pond fish (Saint- Paul, 1988). Thus, the stocking densities calculated are all below those that would theoretically be possible. However, the calculated maximum stocking densities correspond in a good way with the results of practical stocking experiments, which demonstrate individual growth rate depressions at stocking densities above 5 000 $fish.ha^{-1}$. Probably for *Brycon* spp. and the other *Colossoma* species similar stocking densities can be calculated.

Concerning pond management, there is therefore a significant difference between the species. The investigations on locomotory activity show, that all species are markedly less active beneath the macrophytes. This is an important advantage, since the fish waste less energy on swimming, and food conversion must hence be better. However, it is vital for the fish that not the whole pond surface is covered by macrophytes. If the ponds were completely covered by macrophytes, all species which relay on surface excess for ASR will, as a consequence suffocate during periods of oxygen depletion. This is demonstrated by Saint-Paul and Soares (1987) with net cages exposed in fish ponds and was observed by Graef (pers. commun.) in a fish pond totally covered with *Salvinia* sp.. During a severe oxygen depletion all *Brycon* died whereas *Colossoma* survived. More detailed research on this area may help to improve the culture techniques for these species in the future thus helping to make fish-culture more feasible in South America.

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