Chemical modulation on heterogeneous growth in *Prochilodus lineatus* (Valenciennes, 1847) (Pisces; Characiformes)

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(With 3 figures)

Abstract

In the present study, the effect of chemical factors released by conspecifics on growth variability (heterogeneous growth – HetG) in a gregarious fish species (*Prochilodus lineatus*) was tested. HetG was assessed by the weight variation coefficient in two consecutive 21-day periods. The fish were grouped in tanks (4 fish in each) that received running water with constant draining. The tanks contained either conspecifics (C) or non-conspecifics (N). Four conditions were established in accordance with the tank water supply: a) water with previous contact with conspecifics throughout the experiment (CC); b) water without previous contact with conspecifics throughout the experiment (NN); c) water with previous contact with conspecifics in just the first period, 0 to 21 days (CN); and d) just in the period from 21 to 42 days (NC). At the end of the experiments, the occurrence of chemical modulation on the growth variability in *P. lineatus* was verified: the fish that received water with previous contact with a conspecific (C) presented exacerbation of HetG. This corroborates the notion that the predominant mechanism in the determination of intraspecific growth variation in the gregarious species is associated to chemical factors released by conspecifics.

Keywords: fish, growth, growth variability, chemical factors, chemical communication, *Prochilodus lineatus*.

Modulação química sobre o crescimento heterogêneo em *Prochilodus lineatus* (Valenciennes, 1847) (Pisces; Characiformes)

Resumo

No presente trabalho, testou-se o efeito de fatores químicos liberados por coespecíficos sobre o crescimento e sua variabilidade no grupo (crescimento heterogêneo, CHet), numa espécie gregária, o curimbatá, *Prochilodus lineatus*. O CHet foi avaliado pelo coeficiente de variação do peso dos animais, em dois períodos consecutivos de 21 dias. Os peixes foram agrupados em aquários (4 peixes cada) que receberam água corrente, com vazão constante, de tanques contendo (C) ou não (N) coespecíficos. Quatro condições foram delineadas de acordo com a água que abastecia os aquários: a) água com contato prévio com coespecíficos durante todo o experimento (CC); b) água sem contato prévio com coespecíficos durante todo o experimento (NN); c) água com contato prévio com coespecíficos apenas no primeiro período, 0 a 21 dias (CN); e d) apenas no período de 21 a 42 dias (NC). Ao término dos experimentos, verificou-se que ocorre modulação química sobre a variabilidade de crescimento em *P. lineatus*: os peixes que receberam água com contato prévio com coespecífico (C) apresentaram exacerbação do CHet. Fato que corrobora a idéia de que o mecanismo predominante da determinação da variação intra-específica do crescimento, em espécies gregárias, está associado à ação de fatores químicos liberados por coespecíficos.

Palavras-chave: peixes, variabilidade de crescimento, fatores químicos, comunicação química.

1. Introduction

Intraspecific growth variability is common in fish. Although this may occur because of growth enhancement in some fish (Wohlfarth, 1977), growth suppression is often observed among individuals within a group (Allen, 1972; Volpato et al., 1989; Karplus et al., 1992; Volpato and Fernandes, 1994).

Differences in growth rates have been found in isolated fish, which may be attributed to genetic (including sex) sources. However, heterogeneous growth is widely reported as a grouping-dependent phenomenon and thus expresses socially mediated growth variability. In the cichlid Nile tilapia, growth variability in terms of the body length coefficient of variation increased from 15% (isolated fish) to 75% when fish were grouped for 30 days (Volpato and Fernandes, 1994). This latter aspect calls special attention to social mechanisms underlying such growth behaviour.

Volpato and Fernandes (1994) suggest some mechanisms by which growth variability in fish may occur: a) decreased food intake derived from food competition and/or appetite suppression; b) social stress that shifts metabolic pathways to catabolic processes; and c) conspecific chemical factors promoting growth suppression.

The study of mechanisms involved in determining heterogeneous growth (HetG) is of great importance, especially in species used in fish farming, such as *P. lineatus*, as growth reduction in individuals from the group has a negative effect on the final biomass.

The effect of chemical factors released by conspecifics on growth has been described in fish (Francis et al., 1974; Pfuderer et al., 1974; Pfeiffer, 1982; Matty, 1985; Barbosa et al., 1996). However, such an effect on growth variability is not well known. Water-born substances have been shown to increase differences in growth rates among crowed tadpoles, with the larger individuals suppressing the smaller ones. Such an effect has not yet been consistently described for fish. Pereira-da-Silva (1990) suggests this chemically mediated growth suppression in the gregarious fish "pacu", *Piaractus mesopotamicus*, based on two cases where growth variability of crowded fish was decreased in more diluted water conditions. In the Nile talapia, *Oreochromis niloticus*, no effects of water communication and water dilution on interindividual growth variability was found for either isolated or grouped fish (Volpato et al., 1989).

According to proposed mechanisms for grouping-mediated growth variability, social stress is not effective in gregarious species as it is for the territorial ones. Moreover, food competition may operate in both gregarious and territorial species. Conspecific chemical effects were observed only in animals with natural gregarious habits ("pacu" and tadpoles). Association between growth variability and social behaviour was first proposed by Yamagishi (1969) regarding the ontogeny of the intensity of size heterogeneity. Volpato et al. (1989) and Volpato and Fernandes (1994) hypothesised that the mechanisms involved in socially mediated growth variability were dependent on the social habits of the species. As data on this phenomenon is scarce, the aim of the present study was to contribute to the understanding of the effect of grouped conspecific chemicals on intraspecific growth in the gregarious Brazilian fish "curimatá", *Prochilodus lineatus*.

2. Material and Methods

Prochilodus lineatus fingerlings were held in separate tanks (2 m x 4 m x 0.5 m, 200 each) for one week before the experimental procedures were undertaken.

Tests were carried out in two consecutive 21-day periods. Test fish were kept in 25 cm x 30 cm x 25 cm glass tanks (4 fish/aquarium). Water from a natural source continuously flowed (5 $mL.s^{-1}$) through the aquaria. The conspecific odour condition was provided water flowing through a tank (30 L) with 15 conspecifics (held in a net tank) before reaching the glass aquaria. Four conditions (6 repetitions each) were set up according to the water supplied to the aquaria: 1) NN = with no conspecific odour in the two periods; 2) CC = with conspecific odour in the two periods; 3) CN = with conspecific odour only in the first 21-day period and 4) NC = with conspecific odour only in the second period.

The initial body weights (g ± sd) of the odour-source fish were: CC = 12.09 ± 2.02; CN = 12.13 ± 1.82 and NC = 14.66 ± 3.51. Weights of the experimental fish are shown in Figure 1.

The odour-source fish were fed every other day: the net tank was transferred to another tank where these fish were fed for 2 hours. After this period, the net was carefully washed and returned to the original tank. This procedure avoided food detritus in the water source system. The test fish, however, were fed daily (5% of the biomass) in their own glass aquaria. The bottom of the glass aquaria was siphoned every two days.

Water temperature and pH were monitored daily and dissolved oxygen (DO_2), ammonia (NH_4^{++}) and nitrite (NO_2^{--}) were quantified every two days. The physical-chemical conditions of the water were maintained stable in all the experimental groups: DO_2 concentration was 7.2 ± 0.1 ppm; NH_4^+ = 0.2 ± 0.0 ppm; NO_2^{--} = 0.2 ± 0.0 ppm and pH = 6.5 ± 0.1. The mean water temperature was 22.4 ± 1.0 °C.

The individual fish weight was quantified at the beginning and the end of each period. The variability in growth was evaluated from the coefficient of the variation (CV) of the mean weight for each group (CV = sd.100/mean).

Statistical analyses used a profile analysis test, including a previous test for normality and homoscedastic-

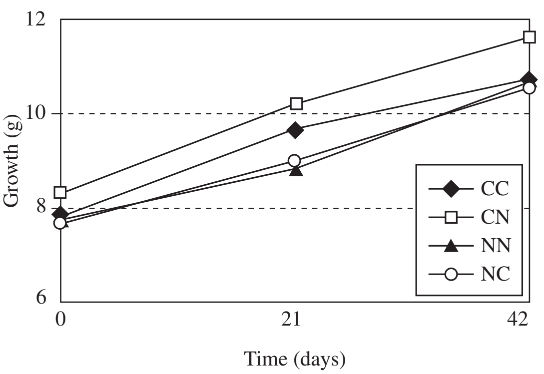

Figure 1. Effect of conspecific odour on mean weight gain *Prochilodus lineatus*. CC = with conspecific odour in both periods; NN = with no contact with conspecific odour; and with conspecific odour restricted to the first (CN) or the second (NC) period.

ity and quartiles analysis. Values are expressed as means (±sd). The critical significance level was considered at $\alpha = 0.05$.

3. Results

The body weight gain of *P. lineatus* is shown in Figure 1. For all experimental conditions, a significant increase of the mean body weight was observed ($F_{(2; 19)} = 278.02$, $p < 0.01$). The mean body weight was also not affected by conspecific odour in the water ($F_{(3; 20)} = 0.35$, $p > 0.05$).

Analysis on variation in growth was evaluated by the coefficient of variation of the mean body weight (Figure 2). Fish size variability at the beginning of the experiments was very small ($F_{(3; 20)} = 1.85$, $p > 0.05$). This variability, however, increased over time.

A significant increase in growth variability occurred only in the condition with conspecific odour (21 days: $F_{(3; 20)} = 9.71$; 42 days: $F_{(3; 20)} = 16.12$, $p < 0.01$) (Table 1).

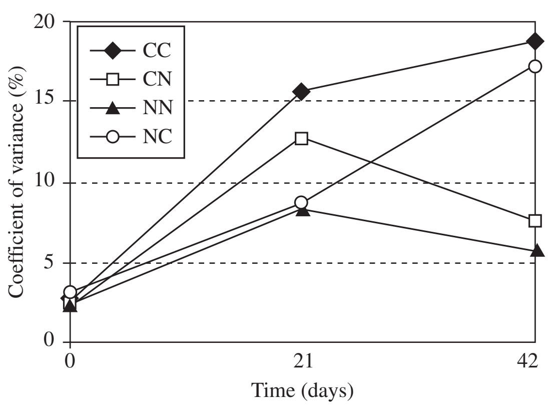

Figure 2. Effect of conspecific odour on mean growth variability. *Prochilodus lineatus*. CC = with conspecific odour in both periods; NN = with no contact with conspecific odour; and with conspecific odour restricted to the first (CN) or the second (NC) period.

As chemical communication affected growth variability in *P. lineatus*, but not mean body weight, a subsequent analysis was performed to clarify whether any growth suppression occurred.

Fish were not individually marked and distribution of the whole number of fish in each condition was analysed over time. The frequency of fish (24 for each condition) was distributed into the respective quartiles for

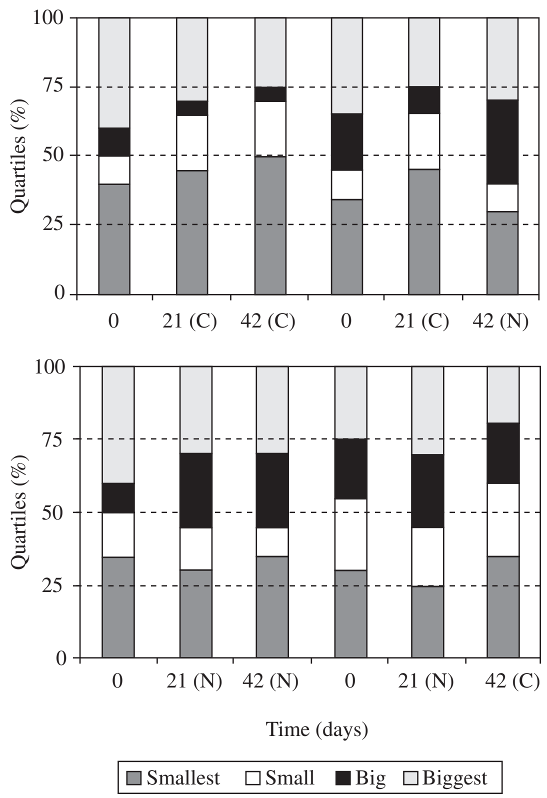

Figure 3. Size fish distribution (Smallest-Small-Big-Biggest) in the respective quartiles (0-25%; 25-50%; 50-75% and 75-100%) for each condition and time of grouping. CC = with conspecific odour in both periods; NN= with no contact with conspecific odour; and with conspecific odour restricted to the first (CN) or the second (NC) period.

Table 1. Statistical analyses of the effect of conspecific odour on mean growth variability. *Prochilodus lineatus*. CC = with conspecific odour in both periods; NN = with no contact with conspecific odour; and with conspecific odour restricted to the first (CN) or the second (NC) period.

0 - 21 days		21 - 42 days	
Odour condition	**Mean slope**	**Odour condition**	**Mean slope**
C	+2.61 [b]	C	+2.79 [ab]
C	+10.31 [b]	N	-5.23 [c]
N	+4.75 [a]	N	-2.58 [ac]
N	+4.40 [a]	C	+7.77 [b]
MSD = 4.00		MSD = 5.90	

Means with same lower case letter are not significantly different. Mean slopes represent an increase (+) or decrease (-) at the end (21-42 days) of the considered period in comparison with the beginning (0-21 days). MSD: minimal significant difference.

each condition and time of grouping. These results are shown in Figure 3. At the beginning of the experiment, size distribution was normal for all conditions. In the condition C (CC and CN at day 21; CC and NC at day 42), a higher frequency of smaller (small and smallest) individuals was observed and, consequently a lower frequency of bigger (big and biggest) individuals was observed, indicating possible growth suppression in most of the animals when receiving water with previous contact with conspecifics.

4. Discussion

An important aspect to be considered is the significant difference in weight gain shown by the fish in this experiment, which offers clear evidence of adequate conditions for fish growth found in the present study. Energy for growth is available only after other demands are met. Stress may occur due to poor water quality (Donaldson, 1981; Schereck, 1981; Pickering, 1992), handling (Mazeaud et al., 1977; Donaldson, 1981; Pickering, 1992) and grouping (Wohlfarth, 1977; Volpato et al., 1989; Fernandes and Volpato, 1993; Barbosa et al., 1996), all of which have been reported as growth suppressors.

In the present experiment, conspecific odour was the experimental variable acting upon the test fish. Food was not present in the aquarium water where animals were used as an odour source. Food was supplied in similar amounts for all experimental conditions

Water quality showed no change throughout the trials and in the experimental conditions. Water quality (in terms of temperature, pH, DO_2, ammonia and nitrite) was maintained similar within the experimental aquaria. Moreover, ammonia and nitrite remained below critical limits for fish: 0.05 for both. Oxygen saturated water is considered with DO_2 up to 6.0 ppm and pH = 6.5 is found in Brazilian rivers.

Given the considerations above, the fish kept in aquaria receiving water in contact with conspecifics may represent the effect of a substance released by these fish.

In the present study, we found that conspecific odour increases growth variability in *P. lineatus*. Effects of substances released by conspecific on growth have not been studied much in fish. The reported effects, however, mainly concern growth suppression in all individuals and have been related to grouping conditions (Francis et al., 1974; Liley, 1982; Pfeiffer, 1982). A clear effect of conspecific odour on fish growth variability was not found in the literature. However, Rose (1960) reported that waterborne growth inhibitors increased growth variability in crowded tadpoles of *Rana* sp. Pereira-da-Silva (1990) reported two cases where water dilution decreased growth variability in the Brazilian fish "pacu", *Piaractus mesopotamicus*, suggesting a chemical mediation on heterogeneous growth.

Pereira-da-Silva (1990) reports that conspecific chemicals usually require specific conditions for release. For instance, reproduction pheromones need a reproductive period (Liley, 1982; Matty, 1985); growth inhibitors released during crowding (Matty, 1985); alarm substance released by injury of the club cells (Peters et al., 1978) and aggression suppressor substances eliminated according to the level of stress of the source fish (Ribeiro, 1996). The substance shown in this study may also be regulated by grouping conditions (or derived from social stress).

A further aspect concerns the mechanisms involved in such chemical modulations of growth variability. Volpato et al. (1989) described the complexity of this mechanism; the supposed chemicals are in contact with all the fish, but growth is suppressed only in some individuals. In the present study, the mean growth was not affected, which means the effect of the odour on the conspecific growth was somewhat balanced for increasing or decreasing growth in the group. Since individual growth was not registered, this possibility could not be tested in the present study. Perhaps in a longer period of study, growth rate could be affected by conspecific odour.

Thus, the biological significance of such growth suppression has been assumed as decreasing the negative effects of the population increase on the habitat resources. In fact, the greater the number of fish, the higher their feeding, faeces and urine amounts are.

The present study, however, did not show any effect of conspecific odour on weight gain, as has been reported for the Nile tilapia (Volpato et al., 1989) and "pacu", *Piaractus mesopotamicus* (Pereira-da-Silva, 1990). One possibility is that these species solve their survival problems brought about by population increase by mechanisms other than chemically mediated growth suppression. Territorial fish, such as the Nile tilapia, use energy according to agonistic patterns (Alvarenga and Volpato, 1995), which can displace energy from growth processes to the strain imposed by social stressors (Fernandes and Volpato, 1993; Volpato et al., 1989; Volpato and Fernandes, 1994,). These mechanisms as a whole may help cope with environmental overexploitation. *P. lineatus*, however, is a gregarious species and may be more tolerant to crowding.

Despite the fact that no effect of odour occurred in weight gain for the species studied, a clear-cut effect of this chemical on growth variability in the group occurred. This effect accompanied the conspecific odour changes imposed for the experimental conditions and this shows a clear chemical modulation of growth variability in this species.

As a concluding remark, the present study points out the relevant effect of conspecific odour on modulation of growth variability for some fish species and emphasises the importance of further studies concerning the mechanisms underlying such an effect.

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