Is Growth Inhibition in Redclaw Crayfish *Cherax quadricarinatus* Regulated by Chemical Communication?

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**Abstract**

Due to high consumer demand for freshwater crustaceans, rearing technology is evolving towards intensive aquaculture. High stocking densities in such cultures result in close proximity of individuals. In this trial, we studied chemical communication and growth inhibition in order to clarify whether large males inhibit the growth of juveniles in Redclaw crayfish (*Cherax quadricarinatus*). No growth inhibition was observed in juveniles grown in the rearing water of adults. The experimental design can be modified to test behavioural and visual cues that may also interfere with juvenile growth.
Freshwater crustacean aquaculture has developed rapidly in recent years (FAO, 2014). Improvements in production and rearing techniques have resulted in higher survival rates, one of the most pressing problems, both in terms of economic and ecological sustainability (Walker et al., 2009). Intensive culture of crustaceans in ponds, constructed wetlands or recirculating aquaculture systems is often regarded as an emerging industry, particularly in more affluent countries due to the high prices obtained (Naylor et al., 2000).

Marine and freshwater lobsters, and crayfish, do not live in large social groups. They are considered rather solitary (Karnofsky et al., 1989; Davis et al., 2007). At higher densities, aggression results in injuries, cannibalism, infections, and significant mortalities, which limit the use of recirculating technology (Nga et al., 2005). The management of intraspecific aggression requires the provision of individual shelters, carefully adjusted rearing densities, and in some species, individual rearing conditions (New, 2002; Sastry et al., 1977). Aggressive behaviour is often directly or indirectly linked to size and growth of individuals and modulated by density-dependent hierarchies. In crayfish, growth is always inversely correlated to stocking density, even if resources are not limited (Morrissy, 1971; Mills and McCloud, 1989). A better understanding of the connection between aggressive behaviour and growth may provide innovative approaches to control of aggression which typically involves access to resources, or reproduction (Moore, 2007). In crustaceans, this is often modulated or regulated by pheromones which either induce specific behaviour patterns or, in the long-term, physiological changes such as gonad maturation or growth regulation within a population (Wuertz, 1997; Breithaupt and Thiel, 2010). *Cherax quadricarinatus*, known as the "freshwater lobster", is a candidate species for freshwater aquaculture due to its high fecundity, relative ease of reproduction, and tolerance for critical water parameters (Medley et al., 1994). It also grows relatively quickly, reaching 50 g within 120-160 days (Medley et al., 1994). *C. quadricarinatus* males grow faster and reach a larger size (Curtis and Jones, 1995) than females, suggesting that when reared in isolation, selection of males may increase efficiency in aquaculture production. Crayfish form hierarchies characterised by agonistic behaviour, and competition for limited resources (Issa et al., 1999; Goessmann et al., 2000; Herberholz et al., 2007). In several species, social status is communicated via pheromones as well as relative size (Zulandt Schneider et al., 1999; Breithaupt and Eger, 2002; Moore and Bergman, 2005).

Recently, reduced growth rate has been observed when small crayfish are with large crayfish, suggesting that size of the neighbour modulates growth rate (Barki et al., 2005). Although the mechanism of growth inhibition was unknown, several hypotheses to be considered are: social interactions such as touching of the antenna, visual contact, and priming pheromones that reduce conspecific growth. Such mechanisms assure sustainable habitat utilization by regulating densities. We investigated whether growth inhibition of individually reared juvenile crayfish is established by chemical signals derived from adults in a flow through system.

**Materials and Methods**

The growth experiment was carried out in a flow through system comprising five 20 L aquaria, each containing a single adult male crayfish, which received water from a 100 L reservoir (Fig. 1). Each aquaria supplied a cluster of five 250 ml containers (water exchange 12 ml/min per container), each one stocked randomly with a juvenile. One cluster, the control, was directly supplied with tap water from the reservoir.
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Figure 1. Flow-through system used to assess growth inhibition of rearing water derived from large males, which were kept in five 20L aquaria and supplied a designated cluster of five 250 ml containers with an individual juvenile crayfish each. Each aquarium was supplied from the 100L tap water reservoir. In addition, a control cluster was directly drained from the reservoir. All aquaria and containers were isolated with an opaque screen to exclude visual contact between individuals.

Growth of the juveniles within each cluster was determined weekly by measuring carapax length after molting to the nearest 0.1 mm with a digital calliper (Mahr MarCal 16ER). All juveniles were fed a commercial carp diet (37% crude protein, 12% fat, 20 MJ gross energy), once a day independent of the adult crayfish as shown in Table 1. Before the start of the experiment, equipment was repeatedly sterilized 3 times with EtOH as well as distilled water.

Table 1. Growth of juvenile *Cherax quadricarinatus* (mean ± STD, n= 5 per cluster) kept in the rearing water of adults and compared to the control reared in tap water over an experimental period of 35 days. The feeding ratio was adjusted after each week, all clusters (Cl) received the same amount of feed. No significant differences were observed between groups (ANOVA, Kruskal-Wallis test, p>0.05).

<table>
<thead>
<tr>
<th>diet fed [g/ind.]</th>
<th>carapax length [mm]</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>control  Cl 1  Cl 2  Cl 3  Cl 4  Cl 5</td>
</tr>
<tr>
<td>0d</td>
<td>0.102  8  8.6  8  8.4  8  8.4</td>
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<tr>
<td>7d</td>
<td>0.171  9  9.2  8.4  9.2  8.2  10.2</td>
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<tr>
<td>14d</td>
<td>0.238  9.6  9.2  9  10  9  10.6</td>
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<tr>
<td>21d</td>
<td>0.272  11  10.6 11  11.4 11.2 11.8</td>
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<tr>
<td>28d</td>
<td>0.306  13  12.2 12.2 12.8 11.6 12.8</td>
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<tr>
<td>35d</td>
<td>0.341  13  12.8 13.4 14.2 12.8 13.8</td>
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Results

Carapax length is frequently used as indicator of growth in crayfish, increased after six weeks from initial size of 8.2±0.3 mm to 13.3±0.6 mm in the freshwater control. In the five treatment clusters (Cl) crayfish size ranged 12.8 mm to 14.2 mm after 35 days. Individuals almost doubled in size (2-3 molting). No differences in growth between clusters, or between clusters and the control, were observed (Table 1).
Discussion

No differences in growth were observed between the five clusters of juveniles supplied with rearing water of the dominant males as compared to control juveniles kept in tap water. In contrast, reduced growth in individuals reared in close proximity to large individuals in a compartment experiment was reported (Barki et al. 2005). In conditions where tactile interaction was allowed, effects were attributed to touching behaviour. Growth is a physiological, long term process and social interactions can be expected to be part of the regulation of crustacean growth. On the other hand, chemosensory organs (clusters of hair-like setae) that cover most regions of the body in crustaceans (concentrated on the antennae and pheomonore interaction has been reported to control aggression, reproduction, and growth in decapod crustaceans, particularly if long-term effects are involved (growth, gonad development). In freshwater prawn Macrobrachium rosenbergii the disparity of growth rates between members of the same cohort with fast-growing "jumpers" and severely growth-inhibited "laggards" is not present in individually raised prawns (Ra'anani and Cohen, 1984). Experimental evidence suggests that a chemically mediated growth inhibitor is progressively transmitted and accumulated (Juarez et al., 1987). Various opportunities to examine the phenomenon of growth inhibition have been suggested in intensive farming of Macrobrachium rosenbergii (Sagi et al., 1986). Some of these have been establishing delays in water recycling to permit dissipation of the growth-inhibiting factor, and the introduction of spatial heterogeneity, allowing the separation of individuals (Johnston et al., 2006), and technical approaches such as the application of ozone (Ritar et al., 2006). Increased growth rates may compensate for increased costs and potential interference with handling procedures, but the development of appropriate methods requires confirmation and identification of growth-inhibiting factors and study of the chemical and biological properties such as mode of action, persistence, and minimum threshold. Similar effects have also been described in lobster (Nelson et al., 1983). An experimental setup that allows the differentiation of chemical, social, or tactile cues as well as in combination, similar to the one presented here, will be valuable in identifying the mechanism of growth inhibition. In the future, experiments need to differentiate the potential cues as compensatory strategies differ substantially.

References


