Ficha catalográfica elaborada através do Programa de Geração Automática
da Biblioteca Central da UFSM, com os dados fornecidos pelo(a) autor(a).

Resende Costa, Juliana
COMPORTAMENTO AGONÍSTICO EM LAGOSTINS: MUDANÇAS AO LONGO DOS ESTÁGOS DE VIDA E SISTEMAS DE AVALIAÇÃO / Juliana Resende Costa.-2015.
68 p.; 30cm

Orientador: Sandro Santos
Dissertação (mestrado) - Universidade Federal de Santa Maria, Centro de Ciências Naturais e Exatas, Programa de Pós-Graduação em Biodiversidade Animal, RS, 2015

1. Comportamento animal 2. Lagostins 3. Disputa animal 4. Hábito de vida 5. Ecologia comportamental I. Santos, Sandro II. Título.

COMPORTAMENTO AGONÍSTICO EM LAGOSTINS: MUDANÇAS AO LONGO DOS ESTÁGOS DE VIDA E SISTEMAS DE AVALIAÇÃO

Juliana Resende Costa

Dissertação apresentada ao Curso de Mestrado do Programa de Pós-Graduação
em Biodiversidade Animal – Área de concentração em Bioecologia, da
Universidade Federal de Santa Maria (UFSM, RS), como requisito parcial para
obtenção do grau de
Mestre em Ciências Biológicas – Área de Biodiversidade Animal

Orientador: Prof. Sandro Santos

Santa Maria, RS, Brasil

2015

Universidade Federal de Santa Maria
Centro de Ciências Naturais e Exatas
Programa de Pós-Graduação em Biodiversidade Animal

A Comissão Examinadora, abaixo assinada,
aprova a Dissertação de Mestrado

COMPORTAMENTO AGONÍSTICO EM LAGOSTINS: MUDANÇAS AO LONGO DOS ESTÁGOS DE VIDA E SISTEMAS DE AVALIAÇÃO

elaborada por
Juliana Resende Costa

Como requisito parcial para obtenção do grau de
Mestre em Ciências Biológicas – Área de Biodiversidade Animal

COMISSÃO EXAMINADORA:

Sandro Santos, Dr.
(Presidente/Orientador)

Daiana da Silva Castiglioni, Dra. (UNISINOS)

Paulo Enrique Cardoso Peixoto, Dr. (UEFS)

Santa Maria, 20 de Março, 2015.

Agradecimentos:

Aos meus pais, à minha irmã e às minhas avós, que mesmo longe me deram apoio e amor incondicional. Sempre tive a ambição de ter uma vida independente e ser dona do meu próprio nariz, mas eu não teria chegado aonde cheguei sem a segurança do colo de vocês.

Ao CNPq, pela bolsa de Mestrado, e ao Programa de Pós Graduação em Biodiversidade Animal pela estrutura.

Aos componentes da banca, Dra. Daiana da Silva Castiglioni e Dr. Paulo Enrique Cardoso Peixoto, pela gentileza de aceitar o convite e a disposição de melhorar esse trabalho.

Ao meu orientador, Dr. Sandro Santos, por ter me aceitado como aluna de Mestrado. O senhor sempre atendeu meus pedidos de ajuda e respeitou minhas opiniões, fico muito feliz por ter sido sua orientada.

Ao Dr. Thomas Breithaupt e à Gabi Breithaupt, pela oportunidade de um estágio na Inglaterra e por toda a hospitalidade. Nunca serei grata o suficiente pela ajuda de vocês nos momentos mais difíceis que passei em um país estrangeiro. Obrigada por terem me acolhido em sua própria casa e como parte de sua família, pelos passeios, pelo aprendizado e pelas aulas de samba! *Danke schön*!

Ao Alexandre Varaschin Palaoro, por me ouvir, pelos puxões de orelha e por ser um exemplo. Mesmo se escondendo atrás de piadas não muito boas, você fez os meus dias mais alegres e me mostrou que até o pior problema pode ser encarado com um pouquinho de humor. Obrigada por sempre saber quando eu acordei com o pé esquerdo e por toda a sua ajuda (de boa e má vontade). Você foi a mistura de amigo e professor que permitiu a evolução desse trabalho e da profissional que eu pretendo ser.

Ao Marcelo Marchet Dalosto, pela paciência, pela ajuda e pela inspiração. Você foi o meu primeiro professor no mundo da pesquisa e, mesmo com as “marcelices”, me fez sentir capaz de dar meus primeiros passos sozinha. Obrigada por sempre estar ao meu lado e confiar em mim. Sempre pude contar com você e espero que saiba que você também sempre poderá contar comigo.

Alexandre e Marcelo, apesar dos meus esforços, vocês conseguiram cultivar um pedacinho da colônia nessa bárbara-nortenha e me mostraram a felicidade de trabalhar com o que se ama. Eu espero um dia encontrar tanta paixão pelo meu trabalho quanto vocês encontraram.

À Cristiane Biasi, à Glaucia Bolzan Cogo, ao Cláudio Ayres Peres, ao Fernando Benso e à Gabriela Zemolin por toda a felicidade que vocês me proporcionaram nas nossas junções,

saídas e mesmo no laboratório. Obrigada Cris pela parceria nas aulas de Muay-Thai e na longa estrada contra o sedentarismo na pós-graduação. Sem vocês a pós-graduação não teria tido graça nenhuma.

A todos os meus colegas de laboratório pelo convívio, junções, bebedeiras, comilanças, discussões, fofocas e risadas. Apesar das nossas diferenças, encontrei em vocês uma família.

À Mariana Borba Trevisan, pela simples e pura amizade. Não tenho palavras para descrever o que a sua amizade significa para mim. Nós dividimos todos os medos, ambições e felicidades de entrar na Graduação, no Mestrado e do que virá adiante. Independente da nossa frequência afetiva, eu tenho certeza que a amizade que nasceu no minuto que nos conhecemos vai durar para sempre. Obrigada por sempre cuidar de mim e brigar comigo quando eu mereci. Quando me mudei para Santa Maria, sabia que uma fase muito importante e única da minha vida iria começar, mas eu não imaginava a sorte que eu iria ter. Você é a irmã que Santa Maria me deu de presente.

À Fernanda Tomasi Sutil e à Gabriella Sommer Vaz, por mais de dez anos de amizade e amor. Vocês são a prova de que a verdadeira amizade não se desfaz com o tempo e com caminhos diferentes. Eu tenho muito orgulho de ter vocês na minha vida.

Obrigada a todos que fizeram e ainda fazem parte da minha jornada em me descobrir como profissional. Quero que saibam que eu amo vocês e que de alguma forma eu carrego um pouquinho de vocês sempre comigo.

“I highly recommend inviting the worse-case scenario into your life.”

- ***Portia de Rossi, Unbearable Lightness***

RESUMO

Dissertação de Mestrado
Programa de Pós-Graduação em Biodiversidade Animal
Universidade Federal de Santa Maria

COMPORTAMENTO AGONÍSTICO EM LAGOSTINS: MUDANÇAS AO LONGO DOS ESTÁGOS DE VIDA E SISTEMAS DE AVALIAÇÃO

Autora: Juliana Resende Costa
Orientador: Sandro Santos
Data e local da defesa: Santa Maria, 20 de Março de 2015

O presente estudo foi desenvolvido em duas etapas que abordam o desenvolvimento da agressão ao longo dos estágios de vida do lagostim escavador *Parastacus brasiliensis*; a determinação do tipo de informação utilizada e a comparação de estratégias de avaliação em interações agonísticas em *P. brasiliensis* e na espécie de águas abertas *Pacifastacus leniusculus*. Nosso objetivo foi demostrar como o hábito de vida pode levar a estratégias de luta e de avaliação distintas, utilizando espécies de lagostins como modelos. Na primeira etapa, nós pareamos os indivíduos de acordo com seu tamanho corporal dentre os diferentes estágios de vida de *P. brasiliensis* e analisamos a dinâmica dos confrontos para determinar o escalonamento da agressão. Nós testamos a relação entre o tamanho corporal e a duração do confronto, o número de confrontos, e a frequência de atos pouco e altamente agressivos. A duração do confronto e a frequência de atos pouco agressivos aumentaram com o tamanho corporal, enquanto o número de confrontos e a frequência de atos altamente agressivos não se mostraram relacionadas com o aumento do tamanho corporal. Lagostins podem ter motivações distintas para entrar em um confronto ao longo de seus estágios de vida. Uma vez que o limiar de tolerância acompanha o tamanho corporal, esse limiar pode influenciar a vontade de permanecer em um confronto. Enquanto indivíduos maiores persistem mais, indivíduos menores podem se perceber como potenciais perdedores e tentar se retirar de um confronto precocemente. Na segunda etapa, nós pareamos indivíduos em duplas intraespecíficas de *P. brasiliensis* e *P. leniusculus* seguindo três tratamentos baseadas na habilidade de luta (*Resource Holding Potential* - RHP) e na comunicação: duplas aleatórias; duplas pareadas pelo RHP que não podiam se comunicar; e duplas pareadas pelo RHP que podiam se comunicar. Para diferenciar entre os modelos de auto-avaliação (AA) e avaliação mútua (AM)/avaliação cumulativa (AC), fizemos regressões de preditores de RHP com a duração dos confrontos de duplas aleatórias, e de duplas pareadas pelo RHP que não podiam se comunicar. Quando as estratégias AM/AC eram mais adequadas, nós testamos a diferença na duração do confronto entre duplas pareadas pelo RHP que não podiam se comunicar com aquelas que podiam se comunicar, de forma a testar o papel da comunicação na tomada de decisão. As espécies diferiram quanto ao tipo de avaliação: a espécie de águas abertas contou com uma combinação das estratégias AM e AC, e a espécie escavadora contou com a estratégia AA. Sendo assim, o habitat pode ter influenciado a confiabilidade e a troca de informação, selecionando a estratégia mais adequada para cada espécie.

Palavras-chave: Astacidae; Parastacidae; Agressão; Disputa animal; Hábito de vida.

ABSTRACT

Mastership Dissertation
Post-Graduation in Animal Biodiversity
Universidade Federal de Santa Maria

AGONISTIC BEHAVIOR IN CRAYFISH: CHANGES ALONG LIFE STAGES AND ASSESSMENT STRATEGIES

Author: Juliana Resende Costa
Advisor: Sandro Santos
Date and place of the defense: Santa Maria, March 20th 2015

The present study was developed by two steps which include respectively: the aggression development along the life stages of the burrowing species *Parastacus brasiliensis*; and the determination of the type of information used and the comparison of the assessment strategies in agonistic interactions in *P. brasiliensis* and in the open water species *Pacifastacus leniusculus*. We aimed to demonstrate how life habit can lead to different life and assessment strategies, using crayfish species as models. In the first step, we paired individuals according to their body size among different life stages of *P. brasiliensis* and analyzed the contest dynamics to determine how aggression escalates. We tested the relation between body size and contest duration, number of bouts, and frequency of low and highly aggressive behaviors. Contest duration and low aggressive behaviors increased with body size, whereas the number of bouts and frequency of highly aggressive behaviors were not related to body size. Individuals may have distinct motivations to fight throughout the life stages. As the individual threshold follows the body development, it could influence the willingness to persist in a contest. While larger individuals persist more, smaller individuals could perceive themselves as potential losers and attempt to withdraw from the contest earlier. In the second step, we assembled the individuals of *P. brasiliensis* and *P. leniusculus* in intraspecific pairs according to three experimental setups based on the Resource Holding Potential (RHP) and communication: random pairs; RHP-matched pairs that could not communicate; and RHP-matched pairs that could communicate. To differ between self-assessment (SA) and Sequential Mutual Assessment (SAM)/Cumulative Assessment (CAM) models, we regressed RHP proxies with contest duration of the random pairs, and the RHP-matched pairs that could not communicate. When SAM/CAM strategies were the best fit, we tested the difference in contest duration between RHP-matched pairs that could not communicate with those that could communicate, thus testing role of communication in decision-making. Species differed in their type of assessment: the open water species relied on a mixed strategy of SAM and CAM, and the burrowing species relies on a SA strategy. Therefore, the habitat could be influencing information reliability and exchange, selecting the most suitable strategy for each species.

Keywords: Astacidae; Parastacidae; Aggression escalation; Animal contest; Life habit.

LISTA DE ILUSTRAÇÕES

Introdução Geral

Figura 1. *Parastacus brasiliensis*. Foto tirada por Alexandre V. Palaoro. 18
Figura 2. *Pacifastacus leniusculus*. Foto tirada por Eilif Byrnak. 18

Artigo 01: Why are the kids fighting? Aggressive behavior in potentially subsocial crayfish (Crustacea: Decapoda)

Figure 1. Relation between the cephalothorax length and mean claw length mean in *Parastacus brasiliensis* (R = 0.9757, P <0.001). Dashed line represents the linear regression fit. 40

Figure 2. Relation between the cephalothorax length and the contest duration in *Parastacus brasiliensis* (R = 0.4145, P < 0.001). Dashed line represents the linear regression fit. 41

Figure 3. Relation between the cephalothorax length and the number of bouts in *Parastacus brasiliensis* ($\chi^2_{1,24}$= 1.5996, P = 0.206). Dashed line represents a linear model fit with a Poisson error structure. 41

Figure 4. Relation between the cephalothorax length and the frequency of low aggressive behaviors in *Parastacus brasiliensis* ($\chi^2_{1,24}$= 152.29, P = 0.006115). Dashed line represents a linear model fit with a binomial error structure. 42

Figure 5. Relation between the cephalothorax length and the frequency of high aggressive behaviors in *P. brasiliensis* ($\chi^2_{1,24}$ = 24.767, P = 0.3437). Dashed line represents a linear model fit with a binomial error structure. 42

Artigo 02: Does ecology affects decision-making during contests?

Figure 1. *Pacifastacus leniusculus*. A) Relation between cephalothorax length and the left and right claw height. The straight line represents the regression for the right claw length and the dashed line represents the regression for the left claw length. B) Relation between cephalothorax length and both claws height. The straight line represents the regression for the right claw height and the dashed line represents the regression for the left claw height. 60

Figure 2. *Parastacus brasiliensis*. A) Relation between cephalothorax length and the left and right claw height. The straight line represents the regression for the right claw length and the dashed line represents the regression for the left claw length. B) Relation between cephalothorax length and both claws height. The straight line represents the regression for the right claw height and the dashed line represents the regression for the left claw height. 61

Figure 3. Relation between cephalothorax length and contest duration in non-perforated divider and random pairs' treatment. The straight line represents the regression for winners and the dashed line represents the regression for losers 64

Figure 4. *Pacifastacus leniusculus*. Relation between cephalothorax length and contest duration in non-perforated divider and RHP-matched pairs' treatment. 65

Figure 5. *Pacifastacus leniusculus*. Relation between RHP-matched non-perforated and perforated divider pairs' treatment two treatments based on the divider type and contest duration (two-tailed t test: $t_{14} = 0.7888$; $P = 0.4431$; Levene test: $F = 1.6419$; $P = 0.2183$). 65

LISTA DE TABELAS

Artigo 01: Why are the kids fighting? Aggressive behavior in potentially subsocial crayfish (Crustacea: Decapoda)

Table 1. Ethogram codes for the agonistic behavior of *Parastacus* elaborated by DALOSTO et al. (2013). 40

Artigo 02: Does ecology affects decision-making during contests?

Table I. Predictions for the assessment models adapted from ARNOTT & ELWOOD, 2009. 62

Table II. Results from the AICc to determine the best traits to predict RHP. AICc: Akaike's Information Criterion corrected for small samples; Δ_i: difference in the AICc value between the most parsimonious combinations and combination *i*.; w_i: Akaike weight of model *I*; df: degrees of freedom. 62

Table III. Non-perforated divider and random pairs' treatment: results from the multiple linear regression using contest duration as the response variable and cephalothorax length (CL) and claw length (CwL) of winners and losers as the explanatory covariables. 63

Table IV. *Pacifastacus leniusculus*. Non-perforated divider and RHP-matched pairs' treatment: results from the multiple linear regression using contest duration as the response variable and cephalothorax length (CL) and claw length (CwL) of winners and losers as the explanatory covariables. 63

Table V. Relation between cephalothorax length and the claws (right and left) length and height: results from the ANCOVA analyzes using each claw measurement (length and height) as the response variable, cephalothorax length as the explanatory co-variable and the claw sizes (right and left) as a factor. 66

SUMÁRIO

1 INTRODUÇÃO GERAL 13
  1.1 Influências do habitat 13
  1.2 Comportamento agonístico 14
  1.3 Ecologia dos lagostins 16
2 OBJETIVOS 19
  2.1 Objetivos gerais 19
  2.2 Objetivos específicos 19
3 REFERÊNCIAS BIBLIOGRÁFICAS 20
ARTIGO 01 28
  Why are the kids fighting? Aggressive behavior in potentially subsocial crayfish (Crustacea: Decapoda) 28
    Abstract 28
    Introduction 29
    Materials and Methods 30
    Results 33
    Discussion 33
    Acknowledgements 35
    References 35
    Figures and Tables 40
ARTIGO 02 43
  Does ecology affects decision-making during contests? 43
    Abstract 43
    Introduction 43
    Materials and Methods 46
    Results 49
    Discussion 50
    Acknowledgements 53
    References 54
    Figures and Tables 60
4 CONCLUSÕES 67

1 INTRODUÇÃO GERAL

1.1 Habitat e estratégias evolutivas

“Quando uma variação constitui uma superioridade por pequena que seja para um ser qualquer, não se poderia dizer que parte convém atribuir à ação acumuladora da seleção natural, e que parte convém atribuir à ação definida das condições de existência. Assim, todos os peleiros sabem muito bem que os animais da mesma espécie têm uma pele tanto mais espessa e tanto mais bela, quanto mais setentrional é o país que habitam; mas quem pode dizer se esta diferença provém de que os indivíduos mais quentemente vestidos têm sido favorecidos e têm persistido durante numerosas gerações, ou se é uma conseqüência do rigor do clima? Parece, com efeito, que o clima exerce uma certa ação direta sobre a pele dos nossos quadrúpedes domésticos”, DARWIN (1859).

A resposta adaptativa em animais é fortemente conduzida por pressões ambientais, de forma que variações do hábitat (e.g. estações do ano, condições climáticas, disponibilidade de recursos) são capazes de afetar a vida dos animais em diferentes intensidades (DANKS, 2007). A plasticidade animal permite que respostas alternativas ao ambiente (i.e. fenótipos) sejam desenvolvidas a partir de um só genótipo, favorecendo o sucesso adaptativo em hábitats distintos (GHERARDI et al., 2013). O ambiente não só seleciona fenótipos como também influencia no desenvolvimento dos indivíduos de uma dada espécie (WEST-EBERHARD, 1989). Questões intrínsecas de uma espécie (e.g. reprodução, dispersão e obtenção de recursos) dependem tanto de condições climáticas quanto da qualidade do hábitat (BAKER, 2005). Desta forma, adaptações morfológicas, fisiológicas, ontogenéticas e comportamentais podem ser favorecidas pela seleção natural e garantir a sobrevivência dos seres vivos (LINSENMAIR, 2007).

A fase inicial de vida em muitas espécies é caracterizada pela habilidade de responder a um estímulo ambiental. Essa resposta pode ser chamada de desenvolvimento de plasticidade e é importante para gerar uma variedade de fenótipos adaptados a diferentes ambientes (BATESON et al., 2004; GLUCKMAN et al., 2005). Mudanças ontogenéticas durante a vida de um animal podem ser consequências de experiência prévia e aprendizado assim como de um resultado do desenvolvimento corporal (HESSELBERG, 2010). A ontogenia é uma grande fonte de diferenças entre indivíduos, já que juvenis se comportam de forma diferente dos adultos e o comportamento de juvenis muda ao longo do tempo (e.g. FENTRESS, 1983; BATESON, 1991). Artrópodos frequentemente apresentam reflexos comportamentais estereotipados (e.g. batimento do leque caudal) por possuírem um sistema nervoso central relativamente simples (NAGAYAMA et al., 1994). Entretanto, seus processos comportamentais podem ser complexos e um mesmo sinal sensorial pode desencadear diferentes comportamentos em um mesmo animal, dependendo do ambiente em que o indivíduo se encontra, seu estado fisiológico, idade e tipo de aprendizado (DAVIS, 1979; RITZMANN & TOBIAS, 1980; BELLMAN & KRASNE, 1983).

As adaptações abrangem desde táticas comportamentais de aquisição de recursos a variações intrínsecas dos indivíduos (WEST-EBEHARD, 1989). Em isópodos, um conjunto de adaptações morfológicas e fisiológicas permitiu que esse grupo obtivesse sucesso na conquista do ambiente terrestre. Os sistemas de respiração e reprodução, juntamente com o mecanismo de comunicação desenvolvido através de pressões do meio, resultaram em crustáceos terrestres bem adaptados (LINSENMAIR, 2007). Acerca de obtenção de recursos, a sua disponibilidade é potencialmente capaz de alterar as estratégias de vida entre grupos de animais. A hipótese "*food-aridity*" em ratos toupeira explica a preferência por ambientes de temperatura moderada dentre os ratos toupeira solitários, enquanto ratos toupeira sociais tendem habitar ambientes mais áridos. Nesse grupo, a disponibilidade do recurso alimentar relacionada às condições do ambiente faz com que as espécies tenham estratégias de vida distintas (HERBST & BENNET, 2006). O oposto pode ser visto em certas aranhas (e.g. *orb-weaver spider*). Apesar de aranhas não sociais se agregarem em ambientes de pastagens onde os recursos são escassos, as aranhas sociais são encontradas em regiões tropicais e com disponibilidade alta de recursos (HODGE & UETZ, 1994). Sendo assim, a capacidade de adaptar estratégias de vida de acordo com o ambiente determina a trajetória evolutiva dos organismos.

1.2 Comportamento agonístico

A disputa por recursos (e.g. abrigo, parceiros sexuais, alimento) entre indivíduos de uma mesma espécie é chamada interação agonística (MOORE, 2007). O principal objetivo dessas interações é estabelecer a dominância entre os indivíduos envolvidos e, posteriormente, a posse de um dado recurso (FERO et al., 2007). Uma vez que um indivíduo tenha decidido engajar em um confronto, a próxima decisão pode levar a uma luta mais intensa, ou a desistir da mesma. A desistência pode se manifestar como uma tentativa de fuga, liberando o recurso, ou como uma interrupção da agressão. Vários fatores influenciam o quanto os animais persistem em confrontos (e.g. BRIFFA & ELWOOD, 2004). Competidores maiores ou mais pesados tendem a ser vitoriosos com mais frequência. Assim, o tamanho corporal é comumente usado como indicador de potencial de assegurar um recurso (*Resource Holding Potential* – RHP *sensu* PARKER, 1974), já que é geralmente correlacionado à força e/ou habilidade necessárias para causar injúrias (ARCHER, 1988). Entretanto, em alguns casos, o menor competidor pode ser o vitorioso, particularmente quando a diferença do tamanho corporal entre os indivíduos é pequena (FABER & BAYLIS, 1993; HUGHES, 1996). Outros fatores podem influenciar a probabilidade de um indivíduo ganhar ou perder um confronto, as quais incluem experiência (DAWS et al., 2002; HSU et al., 2006), armamento (SNEDDON et al., 2000; PALAORO et al., 2014) e estado fisiológico (BEECHING, 1992; BRIFFA et al., 1998; MORETZ, 2003; JENNINGS et al., 2005). Utilizar uma medida multivariada de RHP, na qual vários parâmetros são analisados como possíveis preditores, é uma das formas mais seguras de prever qual medida será a ideal para prever o resultado dos confrontos (ARNOTT & ELWOOD, 2009).

Combates pela posse de recursos limitados podem ser bastante dispendiosos em termos de gastos de energia e tempo, além de aumentarem os riscos de injúrias, predação e até mesmo morte (e.g. GLASS & HUNTINGFORD, 1988; KELLY & GODIN, 2001; BRIFFA & ELWOOD, 2005). Supõe-se que a seleção natural favoreça a coleta de informações precisas sobre os custos e benefícios do conflito contínuo, permitindo que os indivíduos envolvidos tomem decisões apropriadas (PARKER, 1974; PARKER & RUBENSTEIN, 1981). Nesse âmbito, a teoria dos jogos enfatiza o papel central da compilação de informações durante encontros agressivos. Os modelos de briga criados a partir da teoria dos jogos pressupõem que os participantes avaliem o valor do recurso e, em alguns casos, a habilidade de luta do oponente a fim de obter uma estimativa dos possíveis custos e benefícios da luta (MAYNARD SMITH & PRICE, 1973; PARKER, 1974; ENQUIST & LEIMAR, 1983; 1987). Informações sobre as assimetrias entre o RHP entre os indivíduos podem ser usadas na tomada de decisões entre diferentes táticas relativas à persistência e alteração dos níveis de agressão (THORNHILL, 1984). No caso de haver trocas de informação, o conhecimento disponível sobre um oponente durante os primeiros momentos de um confronto pode não ser suficiente, sendo esperado que os combatentes acumulem mais informações à medida que o confronto progride para aumentar a precisão da avaliação (ENQUIST & LEIMAR, 1983).

Em estudos sobre comportamento agressivo, crustáceos são ótimos modelos uma vez que realizam confrontos agonísticos altamente estereotipados e possuem sistemas quimiossensoriais bem desenvolvidos (ZULANDT SCHNEIDER et al., 1999; 2001). Dentro dos Decapoda, os lagostins são comumente utilizados dado que apresentam rápido engajamento em confrontos, mesmo na ausência de recursos óbvios (BOVBJERG, 1956; BRUDKI & DUNHAM, 1987) e apresentam padrões comportamentais similares tanto em experimentos de campo (BERGMAN & MOORE, 2003; FERO et al., 2007) quanto em experimentos laboratoriais (BOVBJERG, 1956; BRUSKI & DUNHAM, 1987; CROOK et al., 2004; DALOSTO et al., 2013). É através desses confrontos que a relação dominante-subordinado é determinada (DAWS et al., 2002; BERGMAN & MOORE, 2003) e que previsões de desempenho são comumente feitas a partir do valor de RHP de cada indivíduo (SNEDDON et al., 1997; PERCIVAL & MOORE, 2010).

1.3 Ecologia dos lagostins

Os lagostins são crustáceos da ordem Decapoda cuja morfologia se assemelha às lagostas marinhas. Porém, formam um grupo distinto e diverso com mais de 500 espécies. As superfamílias Astacoidea DE HAAN, 1841, e Parastacoidea HUXLEY, 1879, dividem esses animais em dois grandes grupos originados no hemisfério Norte e no hemisfério Sul, respectivamente (BUCKUP, 2003). Distribuídos pelo mundo, esses animais ocupam vários tipos de habitats, tais como galerias subterrâneas, riachos, rios, lagoas, lagos e cavernas. O hábito de vida conta com adaptações morfológicas que variam de acordo com as pressões do meio em que se encontram e abrangem desde modificações na morfologia externa (e.g. quelípodo mais robustos, olhos reduzidos, abdomes alongados) até alterações fisiológicas (e.g. perda de pigmentação, intolerância a baixos níveis de oxigênio) (CRANDALL & BUHAY, 2008). Apesar de diversos, a maior parte dos estudos feitos acerca do grupo se limita a espécies de águas abertas (i.e. riachos, rios, lagoas e lagos) (e.g. BRUSKI & DUNHAM, 1987; GHERARDI, 2002; BERGMAN & MOORE, 2003; HAZLETT et al., 2007; HUDINA et al., 2011), sendo que pesquisas sobre lagostins escavadores ganharam mais atenção na última década (e.g. NORO & BUCKUP, 2010; DALOSTO & SANTOS, 2011; DALOSTO et al., 2013; PALAORO et al., 2013). Como animais de grande interesse na aquicultura, algumas espécies de lagostins foram introduzidas em regiões não nativas e se tornaram um problema biológico pela sua alta adaptabilidade (e.g. GUAN & WILES, 1997; JOHNSEN & VRÅLSTAD, 2009). Porém, é pelo seu comportamento estereotipado durante encontros agonísticos e fácil manejo que esses animais têm sido cada vez mais utilizados em experimentos laboratoriais acerca de interações agonísticas.

Dentre os lagostins que habitam ambientes de águas abertas, a espécie *Pacifastacus leniusculus* (Figura 2) é globalmente conhecida pelo seu sucesso em colonizar novos ambientes. Nativa dos Estados Unidos da América, essa espécie foi introduzida na Suécia em meados de 1960 e irradiou-se por toda a Europa (JOHNSEN & TAUGBØL, 2010). Além de ocupar o mesmo nicho das espécies nativas, *P. leniusculus* transmite um fungo que é responsável pelo rápido declínio de espécies nativas na Europa em regiões infectadas (ALDERMAN, 1997; HILEY, 2003; JOHNSEN & TAUGBØL, 2010). Entretanto, esses lagostins também são estudados pelo seu padrão comportamental durante interações agonísticas. As interações agonísticas desses animais de vida solitária são caracterizadas por confrontos escalonados nos quais os indivíduos executam sequências de atos em intensidades crescentes de agressão (ver BERGMAN & MOORE, 2003). Estudos acerca da agressividade na espécie demonstraram sua dominância em relação a outras, o que a fez ser considerada altamente agressiva (e.g. SÖDERBÄCK, 1992; TIERNEY et al., 2000). A partir dos estudos sobre interações agonísticas, novas pesquisas na comunicação entre esses crustáceos puderam ser realizadas. Estudos evidenciaram o papel fundamental da troca de urina na formação e manutenção de hierarquias de dominância, o que sugere possíveis trocas de informação durante os confrontos (BREITHAUPT & EGER, 2002; BERRY & BREITHAUPT, 2008; 2010; BREITHAUPT, 2011).

Por sua vez, as espécies de lagostins escavadores se destacam pelas particularidades de seu hábito de vida. As galerias subterrâneas construídas por esses lagostins podem suprir a busca por recursos, uma vez que oferecem abrigo, alimento e condições favoráveis à sobrevivência desses animais. Entretanto, apesar de não precisarem de água em abundância para sobreviver, dependem das condições do ambiente (i.e. umidade) para dispersar e construir suas galerias subterrâneas (RICHARDSON, 2007). Estudos acerca de lagostins escavadores indicam que as pressões do ambiente são responsáveis por comportamentos gregários (i.e. construir tocas próximas de outras já existentes de seus coespecíficos; PUZALAN et al., 2001) e compartilhamento de tocas por coespecíficos de diferentes faixas etárias (BUCKUP & ROSSI, 1980; NORROCKY, 1991). Além disso, o cuidado parental com a prole já foi observado (e.g. AQUILONI & GHERARDI, 2008; DALOSTO et al., 2012). No caso do cuidado parental prolongado (RICHARDSON, 2007; DALOSTO et al., 2012), quatro impulsionadores ambientais são ditos capazes de criar condições favoráveis para a sua evolução, sendo eles: ambientes bem estruturados ou estáveis, ambientes fisicamente estressantes, recursos ricos e efêmeros, e predação (WILSON, 1975). De acordo com WILSON (1975), qualquer nível de comportamento social que vá além do comportamento sexual pode ser considerado um caso de pré-socialidade. Dessa forma, a dispersão limitada pela qualidade do meio juntamente com as condições ambientais ideais dentro das tocas podem ter favorecido uma redução da agressão entre coespecíficos em prol de sua coexistência.

De fato, diferente de seus parentes de águas abertas, lagostins escavadores apresentam uma agressão marcadamente mais baixa durante suas interações (GUIAŞU et al., 2005; RICHARDSON, 2007). Outra particularidade marcante é a agressão súbita observada por DALOSTO et al. (2013) em duas espécies do gênero escavador *Parastacus*, na qual os confrontos agonísticos são constituídos por atos agressivos de diferentes intensidades sem uma sequência clara aparente. O gênero *Parastacus* é o único representante do grupo nativo no Brasil (BUCKUP, 2003), sendo que a espécie *Parastacus brasiliensis* (Figura 1) é mais facilmente encontrada e tem sido utilizada para responder questões acerca da influência do hábito de vida em estudos de comportamento (e.g. DALOSTO et al., 2013; PALAORO et al., 2013).

Figura 1. *Parastacus brasiliensis*. Foto tirada por Alexandre V. Palaoro.

Figura 2. *Pacifastacus leniusculus*. Foto tirada por Eilif Byrnak.

2 OBJETIVOS

2.1 Objetivos gerais

Esclarecer questões sobre a influência do hábito de vida no comportamento agonístico da espécie de lagostim escavador *Parastacus brasiliensis* através de relações intraespecíficas, além de comparar a sua dinâmica de confronto com o hábito de vida não escavador.

2.2 Objetivos específicos

- Testar a intensidade e o tempo em agressão ao longo dos estágios de vida em *P. brasiliensis*;

- Determinar o tipo de informação utilizada durante confrontos agonísiticos nas espécies *Pacifastacus leniusculus* e *P. brasiliensis*;

- Testar qual modelo de avaliação de confronto melhor se adequa aos confrontos de *P. leniusculus* e *P. brasiliensis*;

- Comparar os modelos de avaliação de confronto das espécies *P. leniusculus e P. brasiliensis* buscando influências do hábitat nas estratégias utilizadas.

3 REFERÊNCIAS BIBLIOGRÁFICAS

ALDERMAN, D. J. History of the spread of crayfish plague in Europe, in: Crustaceans: Bacterial and fungal diseases. **QIE Scientific and Technical Review**. 15: 15-23, 1997.

AQUILONI, L. & GHERARDI, F. Extended mother-offspring relationships in crayfish: the return behaviour of juvenile *Procambarus clarkii*. **Ethology**. 114: 946-954, 2008.

ARCHER, J. **The Behavioural Biology of Aggression**. Cambridge: Cambridge University Press, 1988.

ARNOTT, G. & ELWOOD, R. W. Assessment of fighting ability in animal contests. **Animal Behaviour**. 77: 991-1004, 2009.

BAKER, M. B. & RAO, A. Incremental costs and benefits shape natal dispersal: Theory and example with *Hemilepistus reaumuri*. **Ecology**. 85 (4): 1039-1051, 2004.

BATESON, P. Are there principles of behavioural development? In The development and integration of behaviour; essays in honour of Robert Hinde. Edited by P. Bateson. **Cambridge University Press, Cambridge**: 19-39, 1991.

BATESON, P. ET AL. Developmental plasticity and human health. **Nature**. 430: 419–421, 2004.

BEECHING, S. C. Visual assessment of relative body size in a cichlid fish, the oscar, *Astronotus ocellatus*. **Ethology**. 90: 177-186, 1992.

BELLMAN, K. L. & KRASNE, F. B. Adaptive complexity of interactions between feeding and escape in crayfish. **Science**. 221: 779-781, 1983.

BERGMAN, D. A.; KOZLOWSKI, C.P.; MCINTYRE, J. C.; HUBER, R.; DAWS, A. G. & MOORE, P. A. Temporal dynamics and communication of winner-effects in the crayfish, *Orconectes rusticus*. **Behaviour**. 140: 805-825, 2003.

BERGMAN, D.A. & MOORE, P.A. Field observations of intraspecific agonistic behavior of two crayfish species, *Orconectes rusticus* and *Orconectes virilis*, in different habitats. **The Biological Bulletin**. 205: 26-35, 2003.

BERRY F. C. & BREITHAUPT T. Development of behavioural and physiological assays to assess discrimination of male and female odours in crayfish, *Pacifastacus leniusculus*. **Behaviour**. 145: 1427-1446, 2008.

BERRY F. C. & BREITHAUPT T. To signal or not to signal? Chemical communication by urine-borne signals mirrors sexual conflict in crayfish. **BMC Biology**. 8: 25, 2010.

BOVBJERG, R. V. Some factors affecting aggressive behavior in crayfish. **Physiological and Biochemical Zoology**. 29: 127-136, 1956.

BREITHAUPT, T. Chemical communication in crayfish. *In*: BREITHAUPT, T. & THIEL, M. eds. **Chemical Communication in Crustaceans**: 257–276. New York: Springer, 2011.

BREITHAUPT, T. & EGER, P. Urine makes the difference: chemical communication infighting crayfish made visible. **Journal of Experimental Biology**. 205: 1221-1231, 2002.

BRIFFA, M. & ELWOOD, R.W. Rapid change in energy status in fighting animals: causes and effects of strategic decisions. **Animal Behaviour**. 70: 119-124, 2005.

BRIFFA, M., ELWOOD, R. W. & DICK, J. T. A. Analysis of repeated signals during shell fights in the hermit crab *Pagurus bernhardus*. **Proceedings of the Royal Society of London, Series B**. 265: 1467–1474, 1998.

BRUSKI, C. A. & DUNHAM, D. W. The importance of vision in agonistic communication of the crayfish *Orconectes rusticus*. I: An analysis of bout dynamics. **Behaviour**. 63: 83–107, 1987.

BUCKUP, L. & ROSSI, A. O gênero *Parastacus* no Brasil. **Revista Brasileira de Biologia**. 40 (4): 663-381, 1980.

BUCKUP, L. Família Parastacidae *In*: G. A. S. Melo (ed.), **Manual de Identificação dos Crustacea Decapoda de água doce do Brasil**: 117-141. São Paulo: Editora Loyola, 2003.

CRANDALL, K. A. & BUHAY, J. E. Global diversity of crayfish (Astacidae, Cambaridae, and Parastacidae-Decapoda) in freshwater. **Hydrobiologia**. 595:295-301, 2008.

CROOK, R.; PATULLO, B.W. & MACMILLAN, D.L. Multimodal individual recognition in the crayfish *Cherax destructor*. **Marine and Freshwater Behavioral and Physiology**. 37: 271-285, 2004.

DALOSTO, M. M. & SANTOS, S. Differences in oxygen consumption and diel activity as adaptations related to microhabitat in Neotropical freshwater decapods (Crustacea). **Comparative Biochemistry and Physiology A**. 160: 461-466, 2011.

DALOSTO, M. M.; PALAORO, A. V.; COSTA, J. R. & SANTOS, S. Aggressiveness and life underground: The case of burrowing crayfish. **Behaviour**. 150:3-22, 2013.

DALOSTO, M.M., PALAORO, A.V. & SANTOS, S. Mother-offspring relationship in the neotropical burrowing crayfish *Parastacus pilimanus* (von Martens, 1869) (Decapoda: Parastacidae). **Crustaceana**. 85: 1305-1315, 2012.

DANKS, H. V. The elements of seasonal adaptations in insects. **Canadian Entomologist**. 139: 1-44, 2007.

DARWIN, C. The origin of species. *In*: Murray eds. London, 1859.

DAVIS, K. M. & HUBER, R. Activity patterns, behavioural repertoires, and agonistic interactions of crayfish: a non-manipulative field study. **Behaviour**. 144: 229-247, 2007.

DAWS, A. G.; GRILLS, J.; KONZEN, K. & MOORE, P. A. Previous experiences alter the outcome of aggressive interactions between males in the crayfish, *Procambarus clarkii*. **Marine and Freshwater Behavioral and Physiology**. 35: 139-148, 2002.

ENQUIST, M. & LEIMAR, O. Evolution of fighting behavior: the effect of variation in resource value. **Journal of Theoretical Biology**. 127: 187-205, 1987.

ENQUIST, M. & LEIMAR, O. Evolution of fighting behaviour: decision rules and assessment of relative strength. **Journal of Theoretical Biology**. 102: 387-410, 1983.

FABER, D. B. & BAYLIS, J. R. Effects of body size on agonistic encounters between male jumping spiders (Araneae: Salticidae). **Animal Behaviour**. 45: 289–299, 1993.

FENTRESS, J. A view of ontogeny. **Special Publications American Society of Mammalogists**. 7: 24-64, 1983.

FERO, K.; SIMON, J. L.; JOURDIE, V. & MOORE, P. A. Consequences of social dominance on crayfish resource use. **Behaviour**. 144: 61–82, 2007.

GHERARDI, F. Behaviour. *In*: HOLDICH, D. M. eds. **Biology of freshwater crayfish**: 258-290. Oxford: Blackwell Science, 2002.

GHERARDI, F.; COIGNET, A.; SOUTY-GROSSET, C.; SPIGOLI, D. & AQUILONI, L. Climate warming and the agonistic behaviour in invasive crayfishes in Europe. **Freshwater Biology**. 58: 1958-1957, 2013.

GLASS, C. W. &HUNTINGFORD, F. A. Initiation and resolution of fights between swimming crabs (*Liocarcinus depurator*). **Ethology**. 77: 237–249, 1988.

GLUCKMAN, P. D.; HANSON, M. A.; SPENCER, H. G. & BATESON, P. Environmental influences during development and their later consequences for health and disease: implications for the interpretation of empirical studies. **Proceedings of The Royal Society**. 272: 671-677, 2005.

GUAN, R. -Z. & WILES, P. R.. Ecological impact of introduced crayfish on benthic fishes in a British lowland river. **Conservation Biology**. 11: 641-647, 1997.

HAZLETT, B.A.; LAWLER,, S. & EDNEY, G. Agonistic behavior of the crayfish *Euastacus armatus* and *Cherax destructo*r. **Marine and Freshwater Behavioral and Physiology**. 40: 257-266, 2007.

HERBST, M. & BENNETT, N. C. Burrow architecture and burrowing dynamics of the endangered Namaqua dune mole rat (*Bathyergus janetta*) (Rodentia: Bathyergidae). **Journal of Zoology**. 270: 420-428, 2006.

HESSELBERG, T. Ontogenetic changes in web design in two orb-web spiders. **Ethology**. 116:535–545, 2010.

HILEY, P. D. The slow quiet invasion of signal crayfish (*Pacifastacus leniusculus*) in England – prospects for the white-clawed crayfish (*Austropotamobius pallipes*). *In*: HOLDICH, D. M. & SIBLY, P. J. eds. **Management and Conservation of Crayfish**: 127-138. Bristol: Proceedings of a conference held on 7th November, 2002. Environment Agency, 2003.

HODGE, M. A. & UETZ, G. W. A comparison of agonistic behavior of colonial web-building spiders from desert and tropical habitats. **Animal Behaviour**. 50: 963-972, 1995.

HSU, Y., EARLEY, R.L. & WOLF, L.L. Modulation of aggressive behaviour by fighting experience: mechanisms and contest outcomes. **Biological Reviews**. 81: 33-74, 2006.

HUDINA, S.; GALIÉ, N.; ROESSINK, I. & HOCK, K. Competitive interactions between co-occurring invaders: identifying asymmetries between two invasive crayfish species. **Biological Invasions**. 13: 1791-1803, 2011.

HUGHES, M. Size assessment via a visual signal in snapping shrimp. **Behavioral Ecology and Sociobiology**. 38: 51–57, 1996.

JENNINGS, D.J., GAMMELL, M.P., PAYNE, R.J.H. & HAYDEN, T.J. An investigation of assessment games during fallow deer fights. **Ethology**. 111: 511-525, 2005.

JOHNSEN, S. I. & TAUGBØL, T. NOBANIS – Invasive Alien Species Fact Sheet – *Pacifastacus leniusculus*. **Online Database of the European Network on Invasive Alien Species**. NOBANIS, 2010 www.nobanis.org, Data of access 21/01/2015.

JOHNSEN, S. I. & VRÅLSTAD, T. Signal crayfish and crayfish plague in the Halden watercourse – proposition for a measurement plan. **NINA Report**. 474: 29, 2009.

KELLY, C. D. & GODIN, J. G. J. Predation risk reduces male–male sexual competition in the Trinidadian guppy (*Poecilia reticulata*). **Behavioral Ecology and Sociobiology**. 51: 95–100, 2001.

LINSENMAIR, K. E. Socialbiology of terrestrial Isopods. *In*: DUFFY J. E. & THIEL M. eds. **Evolutionary Ecology of Social and Sexual System-Crustaceans as Model Organisms**: 339-364. New York: Oxford University Press, 2007.

MAYNARD-SMITH, J. & PRICE, G. R. The logic of animal conflict. **Nature**. 246: 15-18, 1973.

MOORE, P. A. Agonistic behavior in freshwater crayfish: the influence of intrinsic and extrinsic factors on aggressive encounters and dominance. *In*: DUFFY, J.E. & THIEL, M. eds. **Evolutionary Ecology of Social and Sexual Systems – Crustaceans as Model Organisms**: 90-114. New York: Oxford University Press, 2007.

MORETZ, J.A. Aggression and RHP in the northern swordtail fish, *Xiphophorus cortezi*: the relationship between size and contest dynamics in male-male competition. **Ethology**. 109: 995-1008, 2003.

NAGAYAMA, T.; NAMBA, H. & AONUMA, H. Morphological and physiological bases of crayfish local circuit neurones. **Histology and Histopathology**. 9: 791-805, 1994.

NORO, C. K. & BUCKUP, L. The burrows of *Parastacus defossus* (Decapoda: Parastacidae), a fossorial freshwater crayfish from southern Brazil. **Zoologia (Curitiba, Impr.)**. 27: 341-346, 2010.

NORROCKY, M. J. Observations on the ecology, reproduction and growth of the burrowing crayfish *Fallicambarus* (*Creaserinus*) *fodiens* (Decapoda, Cambaridae) in north-central Ohio. **The American Midland Naturalist Journal**. 125: 75-86, 1991.

PALAORO, A. V.; DALOSTO, M. M.; COUTINHO, C. & SANTOS, S. Assessing the importance of burrows through behavioral observations of *Parastacus brasiliensis*, a Neotropical burrowing crayfish (Crustacea), in laboratory conditions. **Zoological Studies**. 52: 2-9, 2013.

PARKER, G. A. & RUBENSTEIN, D. I. Role assessment, reserve strategy, and acquisition of information in asymmetric animal conflicts. **Animal Behaviour**. 29: 221–240, 1981.

PARKER, G. A. Assessment strategy and the evolution of fighting behaviour. **Journal of Theoretical Biology**. 47: 223-243, 1974.

PERCIVAL, D.T. & MOORE, P.A. Shelter size influences self-assessment of size in crayfish, *Orconectes rusticus*: consequences for agonistic fights. **Behaviour**. 147: 103-119, 2010.

PUNZALAN, D.; GUIAŞU, R.C.; BELCHIOR, D. & DUNHAM, D.W. Discrimination of conspecific-built chimneys from human-built ones by the burrowing crayfish, *Fallicambarus fodiens* (Decapoda, Cambaridae). **Invertebrate Biology**. 120: 58-66, 2001.

RICHARDSON, A. M. M. Behavioral ecology of semiterrestrial crayfish. *In*: DUFFY J. E. & THIEL M. eds. **Evolutionary Ecology of Social and Sexual System-Crustaceans as Model Organisms**: 319-338. New York: Oxford University Press, 2007.

RITZMANN, R. E. & TOBIAS, M. L. Flight activity initiated via giant interneurons of the cockroach: evidence for bifunctional trigger interneurons. **Science**. 210:383-384, 1980.

SNEDDON, L. U.; HUNTINGFORD, F. A.; TAYLOR, A. C. & ORR, J. F. Weapon strength and competitive success in the fights of shore crabs (*Carcinus maenas*). **Journal of Zoology**. 250: 397-403, 2000.

SNEDDON, L.U.; HUNTINGFORD, F.A. & TAYLOR, A.C. Weapon size versus body size as a predictor of winning fights between shore crabs (*Carcinus maenas*). **Behavioral Ecology and Sociobiology**. 41: 237-242, 1997.

SÖDERBÄCK, B. Predator avoidance and vulnerability of two co-occurring crayfish species, *Astacus astacus* (L.) and *Pacifastacus leniusculus* (Dana). **Annales Zoologici Fennici**. 29: 253-259, 1992.

THORNHILL, R. Fighting and assessment in *Harpobittacus scorpionflies*. **Evolution**. 38: 204-214, 1984.

TIERNEY, A. J.; GODLESKI, M. S. & MASSANARI, J. R. Comparative analysis of agonistic behavior in four crayfish species. **Journal of Crustacean Biology**. 20(1): 54-66, 2000.

WEST-EBERHARD, M. J. Phenotypic plasticity and the origins of diversity. **Annual Review of Ecology and Systematics**. 20: 249-278, 1989.

WILSON, E. O. Sociobiology. **The New Synthesis**. Cambridge, Mass: Harvard University Press, 1975.

ZULANDT SCHNEIDER, R. A.; HUBER, R. & MOORE, P. A. Individual and status recognition in the crayfish, *Orconectes rusticus*: the effects of urine release on fight dynamics. **Behaviour**. 138: 137–153, 2001.

ZULANDT SCHNEIDER, R.A.; SCHNEIDER, R.W.S. & MOORE, P.A. Recognition of dominance status by chemoreception in the red swamp crayfish *Procambarus clarkii*. **Journal of Chemical Ecology**. 25: 781-794, 1999.

ARTIGO 01

Why are the kids fighting? Aggressive behavior in potentially subsocial crayfish (Crustacea: Decapoda)

Juliana Resende Costa; Marcelo Marchet Dalosto; Alexandre Varaschin Palaoro & Sandro Santos

Núcleo de Estudo em Biodiversidade Aquática, Programa de Pós-Graduação em Biodiversidade Animal, Universidade Federal de Santa Maria, Avenida Roraima 1000, Camobi, Santa Maria, RS, Brazil, Postal address 97105-900.

Abstract

The habitat has a great influence on the development of adaptations and survival of a species. Individuals within the same species may respond differently to environmental pressures depending on their life stage and the availability of vital resources. Burrowing crayfish juveniles are more susceptible to environmental pressures than adults, thus, selection could have favored adaptations towards a more social behavior decreasing aggression among young siblings. Herein, we hypothesize that individuals in early life stages will exhibit lower aggression towards conspecifics than in latest life stages. To evaluate this hypothesis we paired individuals according to their body size (used as a life stage measure) among different life stages of *Parastacus brasiliensis* and analyzed the contest dynamics to determine how aggression escalates. We tested the relation between body size and contest duration, number of bouts, and frequency of low and highly aggressive behaviors. Contest duration and low aggressive behaviors increased with body size, whereas the number of bouts and frequency of highly aggressive behaviors were unrelated to body size. Individuals may have distinct motivations to fight throughout the life stages. As the individual threshold follows body development, it could influence the willingness to persist in a contest. While larger individuals persist more, smaller individuals could hold a lower threshold and attempt to withdraw from the contest earlier.

Keywords: Life underground; Burrowing; Decision-making; Agonistic behavior.

Introduction

The early life stages of many animal species are usually characterized by the high ability to respond to environmental stimuli. This response is important in generating a range of suitable phenotypes for different environments (BATESON et al., 2004; GLUCKMAN et al., 2005). Most of the evidence focuses on the interplay between the developing organism and the environment (GILBERT, 2001; WEST-EBERHARD, 2003). As the organism develops, the overall body performance (e.g. locomotion, digging) grows. Thus, if juveniles compete for the same resources as adults and face the same difficulties, they may be in disadvantage because of their lower body performance. If this is true, animals could be selected towards their behavioral adaptations (HERREL & O'REILLY, 2006).

The habitat has a great influence on animals' life history, providing both resources and limitations (DANKS, 2007). This influence may be higher in animals that inhabit extreme seasonal environments and live under the ground (see further). Some animals, for example, inhabit burrows that protect them from the harshness of the environment outside (RICHARDSON, 2007). An underground life offers a stable and predictable microclimate, unlike the outside environment. Although burrows offer a protective environment, animals still need specializations in order to face competition, difficulties in dispersion and gregariousness (NEVO, 1979). Postulated for mole rats, the aridity-food distribution hypothesis predicts that the set of behavioral features will vary according to pressures imposed by distinct environments (JARVIS et al., 1994; FAULKES et al., 1997; BENNET & FAULKES, 2000). Thus, solitary species of mole rats usually inhabit environments with more predictable resources, while eusocial species will be found in more arid environments where rainfalls are unpredictable and food resources are scarce (HERBST & BENNET, 2006). This idea presents an interesting possibility: the habitat can affect different species, but also groups within the same species may respond differently to environmental conditions.

Environmental pressures affect juveniles and adults in different ways. In terrestrial isopods, for example, adults need to find suitable environments to dig burrows in order to care for their young as well as fend off intruders and find resources outside the burrow (BAKER & RAO, 2004). Meanwhile, juveniles suffer selective pressures that favor adaptations to crowded spaces, since they are unable to dig burrows of their own, and would not survive in the climatic conditions outside the burrow. Thus, selection favored adaptations such as social behaviors and conspecific recognition in the early life stages of the group, decreasing aggression among siblings and ultimately resulting in a cooperative society among family members (LINSENMAIR, 2007). Such observations indicate that different life stages show different behavioral adaptations according to the environmental pressures they face.

Limitations to obtain resources and dig burrows affect the life-history traits of a diverse number of burrowing animals. An interesting case occurs in juveniles of burrowing crayfish. Burrowing crayfish can inhabit environments kilometers away from water bodies (RICHARDSON, 2007). Even though they are morphologically well adapted to this semi-terrestrial life habit (RIEK, 1969), young individuals are incapable of digging a burrow by themselves (BOVBJERG, 1956). The inability to dig burrows makes juvenile's survival completely dependent of their natal burrow. Without it, they become unprotected and vulnerable towards any threat. Burrow-sharing and overlapping generations has been reported in burrowing crayfish species, even though adults are perfectly capable of digging burrows and defending themselves (RICHARDSON, 2007). Since juveniles face greater reliance to the burrow in comparison to adults (e.g. natal burrow dependency and under-developed appendages), we would expect lower levels of aggression in early life stages.

In this study, we aim to investigate the differences in aggression in different life stages of burrowing crayfish, using the Neotropical species *Parastacus brasiliensis* as a model. Our goal is to evaluate how aggressiveness develops during the life of *P. brasiliensis* using data from experiments with juveniles, along with data already known for adults of this species (DALOSTO et al., 2013). Since juveniles face stronger morphological and environmental pressures when compared to adults, we hypothesize that earliest life stages will exhibit lower aggression than latest life stages. Thus, we predict that smaller individuals will fight for a shorter period of time and at lower intensity levels than larger individuals will.

Materials and Methods

We collected *P. brasiliensis* in the municipality of Silveira Martins, Rio Grande do Sul state, Brazil (29°39' S; 53°37' W). The collection site is a small marshland characterized by an extensive coverage of aquatic macrophytes where crayfish and burrows are found. We used traps, manual searching and burrow excavation to collect non-adults during the autumn of 2011, 2012 and 2014.

Since the smallest ovigerous female found had 20 mm of cephalothorax length (CL - measured from the tip of the rostrum to the posterior edge of the carapace; DALOSTO et al., 2013), we considered all crayfish smaller than 20 mm CL non-adults. The CL of individuals ranged from 7 mm to 35 mm CL. We sampled 34 non-adults and then size-matched those into

17 pairs with a maximum of 15% CL difference (ZULANDT et al., 2007). We only sampled non-adults as we already have published data for adults using the same experimental protocol (DALOSTO et al., 2013).

In order to achieve a better comprehension of the body development in this species, we performed a linear regression to test the relation between CL and mean claw length. Since *P. brasiliensis* does not exhibit claw dimorphism (BUCKUP & ROSSI, 1980), we used the mean between left and right claw lengths for each individual as our response variable to diminish possible effects of claw length asymmetries in the results. Only intermolt crayfish with all appendages intact were used in the experiments. We did not sex the individuals because *P. brasiliensis* is an intersexed crayfish with no clear sexual dimorphism and occasionally exhibits hermaphroditism. Therefore, accurate sexing would only be possible by sacrificing the crayfish (ALMEIDA & BUCKUP, 2000; RUDOLPH & VERDI, 2010; DALOSTO et al., 2013). Both efforts to use previously published data and not to sex the individuals were conducted to avoid harming the low-density populations of *P. brasiliensis* (BUCKUP, 2003). After finishing the experiments, we released all individuals back in the collection site.

All crayfish were acclimated in the laboratory during seven days to eliminate any previous social experiences (ZULANDT et al., 2007). During the acclimation, all crayfish were kept in individual opaque aquariums with constant aeration, controlled temperature (20 ± 3 °C) and under natural photoperiod. We fed the crayfish with decaying leaves taken from the sampling site, and ceased five days before the beginning of the experiments to standardize fighting motivation (MOORE, 2007). We marked one individual of each pair with non-toxic paint (nail polish) to allow later identification of the contestants.

For the contests, we used a 21 x 13 x 15 cm aquarium (henceforth “arena”) filled with 2-L of filtered water. The arena was divided in three equal parts by two removable opaque dividers. We placed each member of a pair in one of the ends of the arena for 20 min to let the crayfish acclimate to the new conditions. After acclimation, we removed the dividers and let the contestants interact for additional 20 min. We recorded all contests using a Sony® Handycam HDR-CX560 camcorder placed perpendicularly above the arena (26 cm). We performed the experiments between 8 pm and 5 am since *P. brasiliensis* exhibits a preferably nocturnal activity (PALAORO et al., 2013b). We also used red incandescent light bulbs (40W) during the experiments because crustaceans are less sensitive to this wavelength (ZIMMERMANN et al., 2009; PALAORO et al., 2013b). We monitored all individuals for five days after the experiments.

Based on the recordings, we divided the 20 min interactions in 240 frames of 5 s each (see HUBER & KRAVITZ, 1995). We quantified and qualified the behaviors of all pairs. For that purpose, we used the ethogram table elaborated for *P. brasiliensis* by Dalosto et al. (2013). A bout was considered initiated when one of the contestants exhibited an aggressive behavior followed by its opponent response lasting at least 15 s (three frames). We deemed the bout finished when one of the contestants exhibited a submissive behavior (see Table I to the description of the behaviors) and did not engage in a new bout within 10 s (two frames). If more than one behavioral act occurred within the same frame, we selected the most representative act following the significance order of: -2 > -1 > 5 > 4 > 3 > 2 > 1 > 0 (submissive acts: -2; -1> aggressive acts: 5; 4; 3; 2; 1 > neutral acts: 0).

We calculated the frequency of low aggressive behaviors (i.e. scores 2 and 3, Table I), and highly aggressive behaviors (i.e. scores 4 and 5, Table I), by counting the number of frames in which the behaviors were observed and dividing it by the contest duration. We calculated the contest duration by summing the duration of the first three bouts. We used only the first three bouts because dominance seldom changes after the first encounters. Furthermore, crayfish could fight beyond three bouts only because they were confined in a limited space (BERGMAN & MOORE, 2003), and thus we chose to include only the bouts which were the most informative. The contestant that exhibited the submissive behavior was deemed the loser and its opponent the winner of the bout (GOESSMANN et al., 2000). The contestant that won most bouts was deemed the winner of the contest. When dominance was not clear despite acknowledging all bouts performed, we deemed the winner the individual with the highest total score sum.

To test the relation between the CL of the pairs (explanatory variable) and the aggression variables, we used data from non-adults and adults. We used different linear models, according to data error structure: contest duration was tested using a linear least squares regression; the number of bouts was tested using a generalized linear model (GLM) with a Poisson error structure and a log link function; and the frequency of low aggressive behaviors and highly aggressive behaviors were tested using GLMs with binomial error structure and a logit link function. We used the R environment (R DEVELOPMENT CORE TEAM, 2013) for all statistical analyses.

Results

We found a positive relation between CL and mean claw length ($R = 0.976$, $F_{1,34} = 1409.0$, $p < 0.001$; Figure 1). Contest duration increased with CL ($R = 0.415$, $F_{1,23} = 17.99$, $p < 0.001$; Figure 2). Even though the number of bouts did not show relation with the CL ($\chi^2_{1,24} = 1.600$, $p = 0.206$; Figure 3), the intensity of contests changed depending on the CL. The frequency of low aggressive behaviors increased with CL ($\chi^2_{1,24} = 152.29$, $p = 0.006$; Figure 4), while no relation was observed for highly aggressive behaviors ($\chi^2_{1,24} = 24.767$, $p = 0.344$; Figure 5).

Discussion

We demonstrated that the contest duration and the frequency of low aggressive behaviors increase with body size in *P. brasiliensis*. This corroborates our hypothesis that individuals in early life stages would have lower aggression when compared to older individuals. Thus, selection may be favoring adaptations in aggressiveness according to the life stage in *P. brasiliensis*. Since juveniles are compelled to share a burrow, being extremely aggressive would be disadvantageous. Highly aggressive juveniles would spend a large amount of energy on frequent aggressive encounters without obtaining resources that offset this expenditure (MAYNARD-SMITH & PRICE, 1973). Therefore, juveniles would reduce the fighting time aiming to reduce costs. Apart from digging, claws are also important as weaponries in crustacean agonistic interactions (PALAORO et al., 2014) and the weak weaponry could make younger individuals fight less when compared to older ones (BOVBJERG, 1956).

A possible explanation lies on the assessment strategies during contests. Among the assessment strategies, contestants may rely solely on their own information when deciding to withdrawal from a fight. In this case, no information from the opponent is taken into account and the individual persists in a contest until reaching its own cost threshold (MESTERTON-GIBBONS et al., 1996; PAYNE & PAGEL, 1996, 1997). Smaller individuals may have a lower threshold, when compared to bigger individuals, (ARCHER, 1988; SNEDDON et al., 1997). In addition, the risk of predation could lower even more the threshold of juveniles as the small size plus the weak weaponry could make them more prone to predation (GARVEY et al., 1994; JORDAN et al., 1996). Since cannibalism has been observed in this species (personal observations) and being inside a burrow may not completely prevent predators to enter, younger individuals could be reducing the fighting time to maintain vigilance (SHROEDER & HUBER, 2001).

Smaller individuals also used low aggressive behaviors less frequently. However, highly aggressive behaviors had no relation with body size. The fast escalation in earlier life stages complements the shorter contests duration: smaller individuals could be readily executing highly aggressive behaviors in order to determine dominance faster and cease the encounter without spending excessive time in fighting. Constrains imposed by the claws may reflect the importance of these appendages in social interactions and environmental costs experienced by burrowing crayfish. As larger claws are stronger and can inflict more injuries (SHROEDER & HUBER, 2001), larger individuals would avoid highly aggressive behaviors to protect a valuable tool (i.e. claws used in digging, foraging, aggression). Also, larger claws require more time and energy to grow back if they are lost (JUANES & SMITH, 1995). Therefore, the propensity to molt more often and the smaller claws in early life stages (SKINNER, 1985) may be one of the explanations for the quick escalation to highly aggressive behaviors of younger individuals. Moreover, since highly aggressive behaviors are energetically costly (GUIAŞU et al., 2005), larger individuals could be spending more time in low aggressive behaviors and use highly aggressive behaviors only if necessary (i.e. RHP-matched opponent). Even though the frequency of low aggressive behaviors differed along life stages, no difference was found in the number of bouts. Thus, we can assume that the strategy in contest resolution remains the same throughout development.

Despite the differences among life stages, very little is known to ensure what factors are decisive in the development of behavioral patterns in burrowing crayfish. The lack of research in the ecology of burrowing species limits the comprehension of behavioral patterns (RICHARDSON, 2007). In particular, the relation between individuals and burrows needs to be elucidated in order to comprehend the social interactions regarding the group. In this study, we used the same experimental protocol used for other crayfish species. However, since most studies are performed with species of different life habit (i.e. open water crayfish), our results may have been influenced by the experimental setup. Further studies adapting the experimental protocol and including aspects of the environment conditions (e.g. burrows) are necessary to obtain more information about the resource value and contest dynamics in burrowing crayfish. In this scenario, we hypothesize that younger individuals would not exhibit significant aggression towards each other and would also be tolerated by older individuals. On the other hand, older individuals would be less tolerating towards each other and would be more aggressive.

Acknowledgements

We would like to thank CNPq for the scholarship to J.R.C., CAPES/FAPERGS for the scholarship to M.M.D., CAPES for the scholarship to A.V.P. and productivity grant (process 308598/2011-3) to S.S. We would also like to thank Mr. Ângelo Baggio, Mr. Alderi Baggio and Mrs. Lúcia Baggio for kindly allow us to collect the animals in their property.

References

ALMEIDA, A. & BUCKUP, L. Occurrence of protandric hermaphroditism in a population of the neotropical freshwater crayfish *Parastacus brasiliensis* (Parastacidae). **Journal of Crustacean Biology**. 20: 224-230, 2000.

ARCHER, J. **The Behavioural Biology of Aggression**. Cambridge: Cambridge University Press, 1988.

BAKER, M. B. & RAO, A. Incremental costs and benefits shape natal dispersal: theory and example with *Hemilepistus reaumuri*. **Ecology**. 85: 1039-1051, 2004.

BATESON, P. ET AL. Developmental plasticity and human health. **Nature**. 430: 419–421, 2004.

BENNET, N. C. & FAULKES, C. G. **African mole-rats ecology and eusociality**. Cambridge: Cambridge University Press, 2000.

BERGMAN, D. A.; KOZLOWSKI, C.P.; MCINTYRE, J. C.; HUBER, R.; DAWS, A. G. & MOORE, P. A. Temporal dynamics and communication of winner-effects in the crayfish, *Orconectes rusticus*. **Behaviour**. 140: 805-825, 2003.

BOVBJERG, R. V. Some factors affecting aggressive behavior in crayfish. **Physiological and Biochemical Zoology**. 29: 127-136, 1956.

BUCKUP, L. & ROSSI, A. O gênero *Parastacus* no Brasil. **Revista Brasileira de Biologia**. 40: 663-381, 1980.

BUCKUP, L. Família Parastacidae *In*: G. A. S. Melo (ed.), **Manual de Identificação dos Crustacea Decapoda de água doce do Brasil**: 117-141. São Paulo: Editora Loyola, 2003.

DALOSTO, M. M.; PALAORO, A. V.; COSTA, J. R. & SANTOS, S. Aggressiveness and life underground: the case of burrowing crayfish. **Behaviour**. 150:3-22, 2013.

DANKS, H. V. The elements of seasonal adaptations in insects. **Canadian Entomologist**. 139: 1-44, 2007.

FAULKES, C. G.; BENNETT, N. C.; BRUFORD, M. W.; O'BRIEN, H. P.; AGUILA, G. H. & JA RVIS, J. U. M. Ecological constraints drive social evolution in the african mole-rats. **Proceedings of the Royal Society of Londom, B**. 264: 1619-1627, 1997.

GARVEY, J. E.; STEIN, R. A. & THOMAS, H. M. Assessing how fish predation and interspecific prey competition influence a crayfish assemblage. **Ecology**. 75: 532-547, 1994.

GILBERT, S. F. Ecological developmental biology: developmental biology meets the real world. **Developmental Biology**. 233: 1–12, 2001.

GLUCKMAN, P. D.; HANSON, M. A.; SPENCER, H. G. & BATESON, P. Environmental influences during development and their later consequences for health and disease: implications for the interpretation of empirical studies. **Proceedings of the Royal Society of Londom, B**. 272: 671-677, 2005.

GOESSMANN, C.; HEMELRIJK, C. & HUBER, R. The formation and maintenance of crayfish hierarchies: behavioral and self-structuring properties. **Behavioral Ecology and Sociobiology**. 48: 418-428, 2000.

GUIAŞU, R. C.; SALEH, N.; MOZEL, E. & DUNHAM, D. W. Low aggression in juvenile burrowing crayfish, *Fallicambarus fodiens* (Cottle, 1863) (Decapoda, Cambaridae). **Crustaceana**. 78: 421-428, 2005.

HERBST, M. & BENNETT, N. C. Burrow architecture and burrowing dynamics of the endangered Namaqua dune mole rat (*Bathyergus janetta*) (Rodentia: Bathyergidae). **Journal of Zoology**. 270: 420-428, 2006.

HERREL, A. & O'REILLY, J. C. Ontogenetic scaling of bite force in lizards and turtles. **Physiological and Biochemical Zoology**. 79: 31-42, 2006.

HUBER, R., & KRAVITZ, E.A. A quantitative analysis of agonistic behavior in juvenile american lobsters (*Homarus americanus* L.). **Brain, Behavior and Evolution**. 46: 72-83, 1995.

JARVIS, J. U. M.; O'RIAIN, M. J.; BENNET, N. C. & SHERMAN, P. W. Mammalian eusociality: a family affair. **Trends in Ecology and Evolution**. 9: 47–51, 1994.

JORDAN, F.; DELEON, C. J. & McCREARY, A. C. Predation, habitat complexity, and distribution of the crayfish *Procambarus alleni* within a wetland habitat mosaic. **Wetlands**. 16: 452-457, 1996.

JUANES, F. & SMITH, L. D. The ecological consequences of limb damage and loss in decapod crustaceans: a review and prospectus. **Journal of Experimental Marine Biology and Ecology**. 193: 197-223, 1995.

LINSENMAIR, K. E. Social biology of terrestrial Isopods. *In*: DUFFY J. E. & THIEL M. eds. **Evolutionary Ecology of Social and Sexual System - Crustaceans as Model Organisms**: 339-364. New York: Oxford University Press, 2007.

MAYNARD-SMITH, J. & PRICE, G. R. The logic of animal conflict. **Nature**. 246: 15-18, 1973.

MESTERTON-GIBBONS, M.; MARDEN, J. H. & DUGATKIN, L. A. On wars of attrition without assessment. **Journal of Theoretical Biology**. 181: 65-83, 1996.

MOORE, P. A. Agonistic behavior in freshwater crayfish: the influence of intrinsic and extrinsic factors on aggressive encounters and dominance. *In*: DUFFY, J.E. & THIEL, M. eds. **Evolutionary Ecology of Social and Sexual Systems – Crustaceans as Model Organisms**: 90-114. New York: Oxford University Press, 2007.

NEVO, E. Adaptive convergence and divergence of subterranean mammals. **Annual Review of Ecology, Evolution and Systematics**. 10: 269-308, 1979.

PALAORO, A. V.; DALOSTO, M. M.; COUTINHO, C. & SANTOS, S. Assessing the importance of burrows through behavioral observations of *Parastacus brasiliensis*, a Neotropical burrowing crayfish (Crustacea), in laboratory conditions. **Zoological Studies**. 52: 2-9, 2013.

PALAORO, A. V.; DALOSTO, M. M.; COSTA, J. R. & SANTOS, S. Freshwater decapod (*Aegla longirostri*) uses a mixed assessment strategy to resolve constests. **Animal Behaviour**. 95: 71-79, 2014.

PUNZALAN, D.; GUIAŞU, R.C.; BELCHIOR, D. & DUNHAM, D.W. Discrimination of conspecific-built chimneys from human-built ones by the burrowing crayfish, *Fallicambarus fodiens* (Decapoda, Cambaridae). **Invertebrate Biology**. 120: 58-66, 2001.

R DEVELOPMENT CORE TEAM. R: A language and environment for statistical computing. Vienna, Austria: R Foundation for Statistical Computing. 2013.

RICHARDSON, A. M. M. Behavioral ecology of semiterrestrial crayfish. *In*: DUFFY J. E. & THIEL M. eds. **Evolutionary Ecology of Social and Sexual System-Crustaceans as Model Organisms**: 319-338. New York: Oxford University Press, 2007.

RIEK, E. F. The Australian freshwater crayfish (Crustacea: Decapoda: Parastacidae), with the descriptions of new species. **Australian Journal of Zoology**. 17: 855-918, 1969.

RUDOLPH, E. H. & VERDI, A. C. Intersexuality in the burrowing crayfish, *Parastacus pilimanus* (von Martens, 1869) (Decapoda, Parastacidae). **Crustaceana**. 83:73-87, 2010.

SHROEDER, L. & HUBER, R. Fight strategies differ with size and allometric growth of claws in crayfish, *Orconectes rusticus*. **Behaviour**. 138: 1437-1449, 2001.

SKINNER, D. M. Molting and regeneration. In: BLISS D. E. & MANTEL L. H. eds. **The Biology of Crustacea**. 9: 43-146. Orlando: Academic Press, 1985.

SNEDDON, L.U.; HUNTINGFORD, F.A. & TAYLOR, A.C. Weapon size versus body size as a predictor of winning fights between shore crabs (*Carcinus maenas*). **Behavioral Ecology and Sociobiology**. 41: 237-242, 1997.

WEST-EBERHARD, M. J. Phenotypic plasticity and the origins of diversity. **Annual Review of Ecology, Evolution and Systematics**. 20:249-278, 2003

ZIMMERMANN, B.L.; AUED, A.W.; MACHADO, S.; MANFIO, D.; SCARTON, L.P. & SANTOS, S. Behavioural repertory of *Trichodactylus panoplus* (Crustacea: Trichodactylidae) under laboratory conditions. **Revista Brasileira de Zoologia**. 26: 5-11, 2009.

ZULANDT, T.; ZULANDT-SCHNEIDER, R. A. & MOORE, P. A. Observing agonistic interactions alters subsequent fighting dynamics in the crayfish, *Orconectes rusticus*. **Animal Behaviour**. 75: 13-20, 2007.

Figures and Tables

Table 1. Ethogram codes for the agonistic behavior of *Parastacus* elaborated by Dalosto et al. (2013).

Score	Behavior
-2	Retreat with a tail flip.
-1	Retreat by walking away from the opponent.
0	Ignore the opponent/non-aggressive behaviors.
1	Approach without agonistic display.
2	Approach with meral spread and/or antennal whip.
3	Aggression with closed chelae: touching, punching and pushing the opponent.
4	Active use of the chelae to grab the opponent's appendages, or chela strike.
5	Intense combat: animals performing several agonistic acts simultaneously, trying to grab and pull the opponent's body parts, or attempting to turn/carry the opponent.

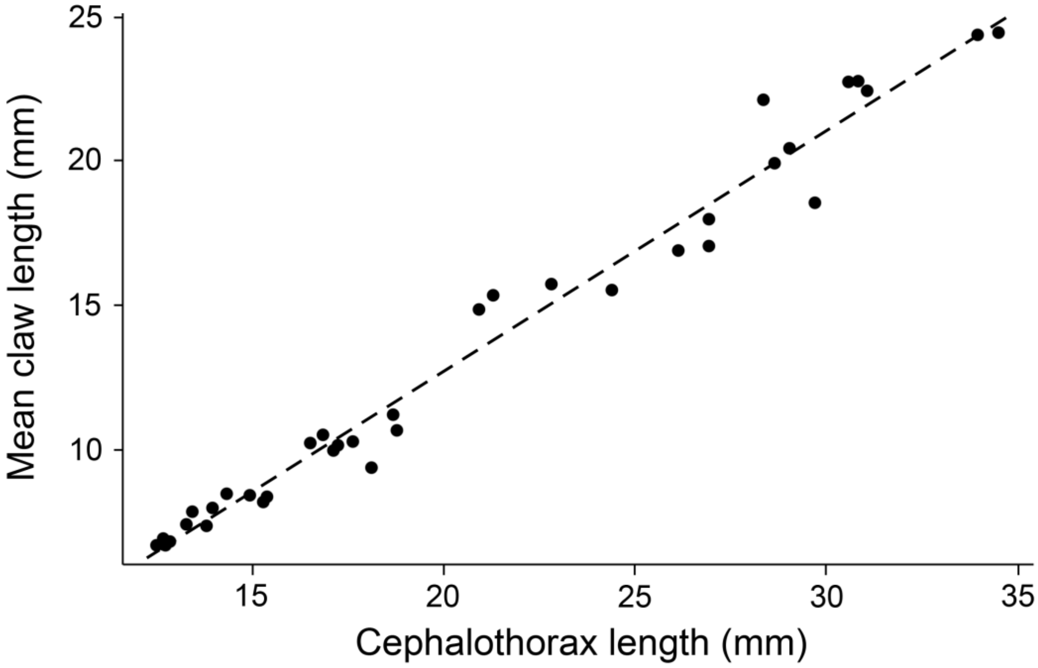


Figure 1. Relation between the cephalothorax length and mean claw length mean in *Parastacus brasiliensis* (R = 0.975, p <0.001).

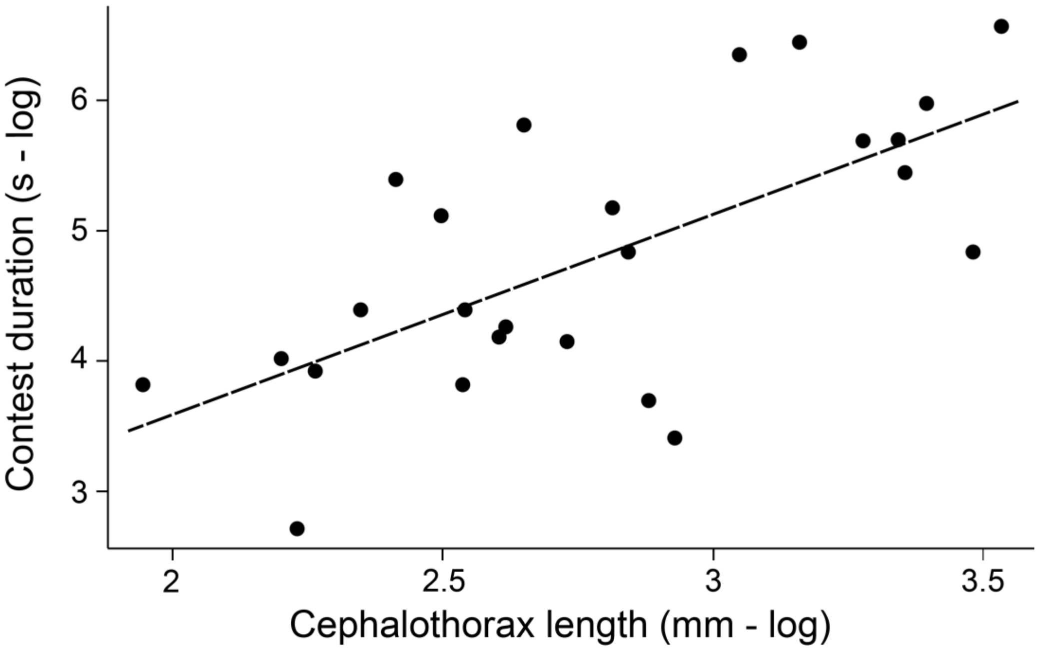


Figure 2. Relation between cephalothorax length and contest duration in *Parastacus brasiliensis* (R = 0.415, p < 0.001).

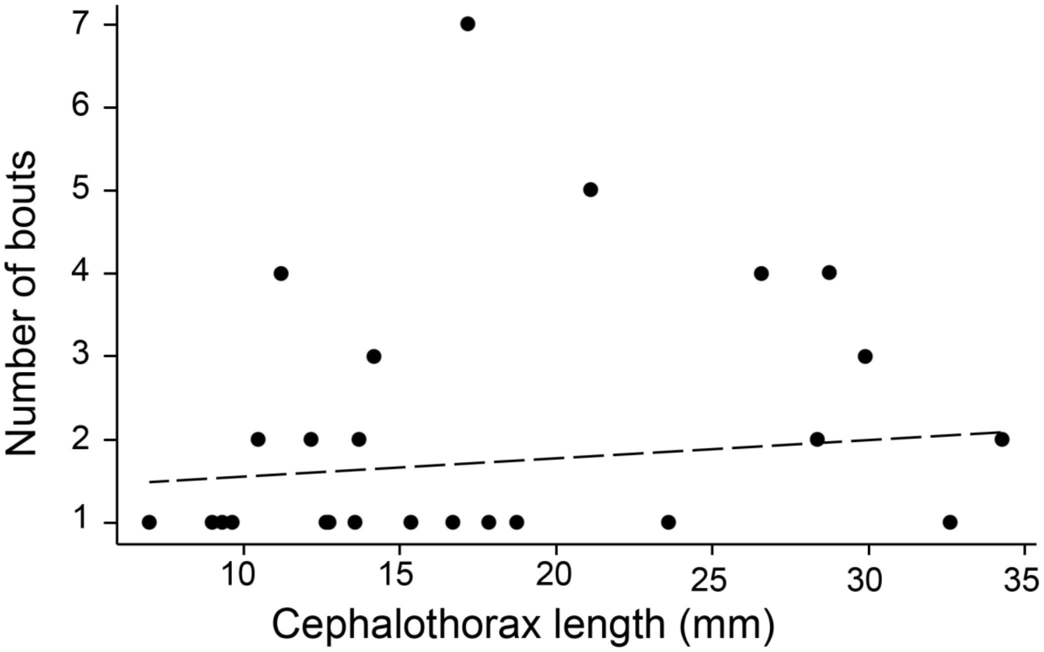


Figure 3. Relation between the cephalothorax length and the number of bouts in *Parastacus brasiliensis* ($\chi^2_{1,24}$ = 1. 600, p = 0.206).

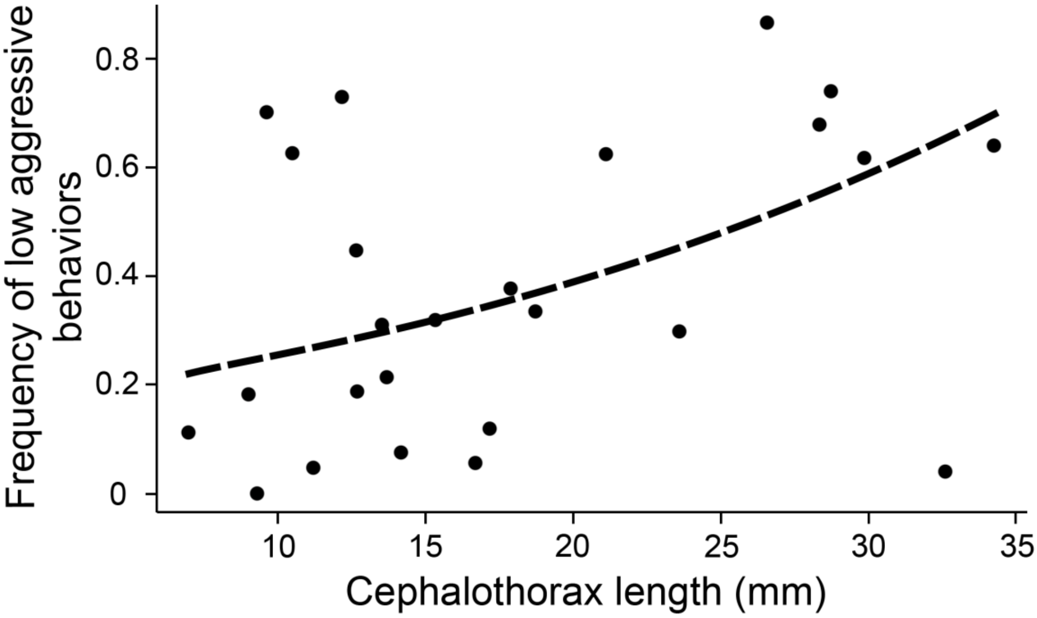


Figure 4. Relation between the cephalothorax length and the frequency of low aggressive behaviors in *Parastacus brasiliensis* ($\chi^2_{1,24}$ = 152.29, p = 0.006)..

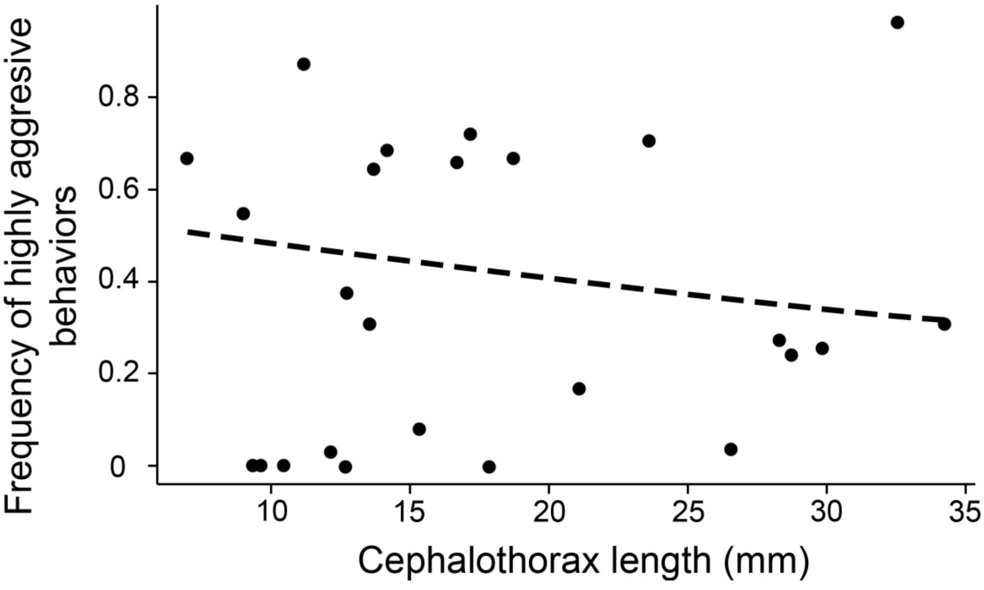


Figure 5. Relation between the cephalothorax length and the frequency of high aggressive behaviors in *P. brasiliensis* ($\chi^2_{1,24}$ = 24.767, p = 0.344).

ARTIGO 02

Does ecology affects decision-making during crayfish contests?

Juliana Resende Costa ; Alexandre Varaschin Palaoro , Thomas Heinrich Breithaupt & Sandro Santos

Núcleo de Estudo em Biodiversidade Aquática, Programa de Pós-Graduação em Biodiversidade Animal, Universidade Federal de Santa Maria, Avenida Roraima 1000, Camobi, Santa Maria, RS, Brazil, Postal address 97105-900.

Department of Biological Sciences, University of Hull, Cottingham Road, Kingston upon Hull, Yorkshire, England, Postal address HU6 7RX.

Abstract

Asymmetries between predictor traits of resource holding potential (RHP) and contest parameters are usually employed to distinguish among assessment strategies employed by rivals in agonistic interactions. As RHP is not only influenced by intrinsic factors but also by the context individuals are in, assessment strategies may change as life habit also changes. We aim to demonstrate how life habit can lead to different assessment strategies, using crayfish species as models. Thus, we tested whether individuals of each species decide based solely on their own information (self-assessment, SA), or use information about the opponent (sequential mutual assessment, SAM). Due to a close relation between RHP and life context, we hypothesize that the aggressive and solitary open water species will rely on SAM while the subsocial burrowing species will rely on SA. We had three experimental setups: random pairs; RHP-matched pairs that could not communicate; and RHP-matched pairs that could communicate. To differ between SA and SAM/Cumulative Assessment (CAM) models, we regressed RHP proxies with contest duration of the random pairs, and the RHP-matched pairs that could not communicate. When SAM/CAM strategies were the best fit, we tested the difference in contest duration between RHP-matched pairs that could not communicate with those that could communicate, thus testing role of communication in decision-making. Species differed in their type of assessment: the open water species relied on a mixed strategy of SAM and CAM, and the burrowing species relies on a SA strategy. Therefore, the habitat could be influencing information reliability and exchange, selecting the most suitable strategy for each species.

Keywords: Assessment strategies; Resource holding potential; Life habit; Crayfish.

Introduction

Fighting against conspecifics is often used to obtain resources, such as food, mates, shelter and territory (MOORE et al., 2008). During a contest, individuals may endure costs until one contestant is compelled to stop fighting (BRIFFA & ELWOOD, 2009). Fighting strategies can vary from noncontact displays (e.g. butterflies; KEMP, 2013) to deadly fights (e.g. ants; HARDY et al., 2013), and this escalation depends on decision-making (ARNOTT & ELWOOD, 2008). To decide when to withdrawal, the individual often relies on the internal motivational state to fight, its fighting ability (i.e. energy reserves), and, sometimes, on the opponent's fighting ability. Fighting ability is often referred as resource holding potential (RHP *sensu* PARKER, 1974) and is usually correlated with body size and weaponry (VIEIRA & PEIXOTO, 2013). The asymmetries between contestants' RHPs and parameters from contests (i.e. contest duration) present an accurate method to distinguish between assessment strategies during fights. Nevertheless, as RHP is not a stated value (BRIFFA et al., 2013), life context (e.g. resource value, evolutionary background) can affect the reliability on the available information and, further, the assessment strategy (BRIFFA & ELWOOD, 2009). In a review, ARNOTT and ELWOOD (2009) provided evidence for two assessment strategies based on assumptions regarding decision-making: self-assessment (SA), sequential assessment (SAM).

The SA strategy is assumed in the war of attrition models (WOA) and the cumulative assessment model (CAM). The WOA model assumes that individuals rely only on their own energy expenditures during contests, and when they reach a certain cost threshold, they give up (MESTERTON-GIBBONS et al., 1996; PAYNE & PAGEL, 1996, 1997). The CAM also assumes a cost-threshold to give up. However, it adds the costs inflicted by the opponent to the decision-making process. According to this model, higher RHP individuals are better at inflicting costs than low RHP individuals (PAYNE, 1998). Even though both SA models use only the individual's own RHP as source of information, they can be distinguished by the predictions of each model (Table I). Differing from the SA models, the SAM strategy assumes that individuals will rely on information exchange between opponents (ENQUIST & LEIMAR, 1983). Lastly, the opponent only assessment (OOA) strategy assumes that the decision-making will rely only on the opponent's information. Although unlikely to occur, evidence for OOA strategy has been found (REDDON et al., 2011), and thus, testing it also becomes relevant.

Investigating which traits are related to the RHP within a group is essential. The environmental context in which individuals are found can influence RHP (ARCHER, 1988). Specifically, the availability of vital resources may lead to distinct strategies, since different groups can respond distinctly to the same condition (e.g. mole rats, HERBST & BENNETT, 2006; orb-weaver spiders, HODGE & UETZ, 1995). Even though RHP is connected with the intrinsic fighting ability of the contestants (e.g. body size, strength), resource availability is capable of altering resource value and hence the individual's motivation to fight. Thus, if the circumstances (e.g. resource quality and quantity, contestants' age and reproductive state) increase the resource value, individuals are likely to invest more energy in contests despite having lower RHP (STOCKERMANS & HARDY, 2013). Since environmental conditions provides resources and may impose constrains towards the evaluation of resource value (DANKS, 2007), differential life habits in closely related species could influence assessment strategies during contests.

Among crustaceans, crayfish species can be found in a wide array of environments (CRANDALL & BUHAY, 2008). For example, the signal crayfish, *Pacifastacus leniusculus*, has a solitary life-style in streams and lakes (henceforth open water environments) (TIERNEY et al., 2000). This species is known as highly aggressive and individuals readily engage in escalated contests of highly aggressive behaviors (SÖDERBÄCK, 1992). Studies on agonistic encounters have reported the importance of chemical communication in *P. leniusculus* and the essential role of urine release in dominance hierarchy formation (BREITHAUPT & EGER, 2002; BERRY & BREITHAUPT 2008; 2010). Using the fanning behavior (i.e. create water flow by pumping mouthpart appendages), crayfish are able to exchange chemicals in stagnant environments (BREITHAPT & EGER, 2002). Even though fanning is primarily used in searching for food, this behavior has been observed during agonistic encounters (BREITHAUT, 2001; BERGMAN et al., 2005). This condition points out the possible exchange of information between contestants and leads to particular assumptions regarding assessment strategies in *P. leniusculus*.

On the other hand, burrowing crayfish species face different selective pressures. The semiterrestrial habitat diminishes the impacts of living kilometers away from water bodies, but imposes restrict living areas and hampers dispersion (RICHARDSON, 2007). Although being burrow-dependent, burrows are capable of offering almost all resources, which may favor social adaptations. Indeed, aggregations and burrow sharing have been observed in this group (BUCKUP & ROSSI, 1980; NORROCKY, 1991; PUZALAN et al., 2001), as well as a markedly lower aggression towards conspecifics (DALOSTO et al., 2013). In addition to the low aggression, DALOSTO et al. (2013) highlighted that species of the burrowing genus *Parastacus* exhibited contests that do not follow a clear escalation. The absence of a clear escalation could be evidence that contestants are not gathering information as the contest progresses. This, together with the low aggressiveness, may indicate restrictions of the semiterrestrial life in assessing information and could have selected a self-assessment strategy during agonistic encounters.

Herein, we aim to elucidate the influence of the life habit in the assessment strategies during agonistic encounters of the open water species *P. leniusculus* and the burrowing species *Parastacus brasiliensis*. Since environmental pressures modified the life habits of these crayfish species (i.e. the agonistic behavior; MOORE, 2007), and RHP is deeply connected with the context of agonistic interactions (ARNOTT & ELWOOD, 2009), we hypothesized that *P. leniusculus* and *P. brasiliensis* will show different assessment strategies during contests. As *P. leniusculus* inhabits an open water environment which enables chemical cues to be exchanged, we predict that this species will rely on the SAM strategy (ELWOOD & ARNOTT, 2012). Meanwhile, *P. brasiliensis* inhabits a semiterrestrial environment and live mostly inside burrows (RICHARDSON, 2007). Thus, given the constraints of the habitat, we predict that this species will rely on a SA strategy.

Materials and Methods

Organisms manipulation

We used 30 adults of *P. brasiliensis* collected in the municipality of Silveira Martins, RS, Brazil (29°39’25.14” S; 53°37’33.53” W); and 50 adults of *P. leniusculus* bought in the city of Kingston upon Hull, Yorkshire, England (53º46’19.3”N; 0º22’06.5”O). We performed the experiments with *P. brasiliensis* at the Núcleo de Estudo em Biodiversidade Aquática, in the Universidade Federal de Santa Maria, Brazil, during 2013 and 2014. Meanwhile, the experiments performed with *P. leniusculus* took place in the Department of Biological Sciences, in the University of Hull, England, during 2014. Before performing the experiments, we kept each crayfish in individual opaque aquaria with constant aeration, under controlled temperature (20 ± 3 °C) and under natural photoperiod. We fed *P. brasiliensis* with decaying leaves taken from the sampling site, while *P. leniusculus* were fed with shrimp and pees. We ceased feeding five days before the beginning of the experiments to standardize fighting motivation (MOORE, 2007). Although we performed the experiments separately, we used the same experimental protocol with minor adjustments for both species because of the different life histories. All adjustments are explained in detail below.

Experimental setup and observations

For the contests, we used an aquarium (*P. leniusculus*: 44 x 24 x 24 cm; *P. brasiliensis*: 21 x 13 x 15 cm), henceforth called arena, divided in two compartments by one removable divider. We recorded all experiments using a digital video camera (*P. leniusculus*: Panasonic® HC-V210 camcorder; *P. brasiliensis*: Sony® Handycam HDR-CX560 camcorder) placed perpendicularly above the arena. We recorded *P. brasiliensis*' contests between 8 pm and 5 am, since they are preferably nocturnal (PALAORO et al., 2013). We used red incandescent light bulbs (40W) placed equidistantly from the arena since crustaceans are less sensitive to this wavelength (ZIMMERMANN et al., 2009). For *P. leniusculus*, we recorded the contests between 8 am and 7 pm since they were blindfolded and the activity would not change between night and daytime. We blindfolded all *P. leniusculus* following the method described in BERRY and BREITHAUPT (2010) to ensure both species were in similar experimental conditions (i.e. low visibility in *P. brasiliensis*).

During the experiments, each member within a pair was placed in one of the ends of the arena. The individuals had 20 min to acclimate to the new conditions and, once the divider was removed, another 20 min to interact. We marked one individual of each pair to allow later identification of the contestants. We paired the individuals according to the experimental set-up each pair would be submitted. We used three experimental set-ups differing on the arena's divider and pairing type:

1) Non-perforated divider and random pairs: crayfish could not communicate during acclimation and were randomly paired (*P. leniusculus*, n = 7; *P. brasiliensis*, n = 9);
2) Non-perforated divider and RHP-matched pairs: crayfish could not communicate during acclimation and were paired to a maximum of 10% of variation between the RHP traits (*P. leniusculus*, n = 10; *P. brasiliensis*, n = 9);
3) Perforated divider: crayfish could communicate tactile/chemically during acclimation and were paired to a maximum of 10% of variation between the RHP traits (*P. leniusculus*, n = 8; *P. brasiliensis*, n = 9).

We quantified the contest duration and determined the dominance within a pair. Since individuals fight more when confined to a limited space (BERGMAN & MOORE, 2003), and dominance was always established in the first three bouts, we quantified the contest duration by summing the duration of the first three bouts. We considered a bout initiated when one of the contestants exhibited an aggressive behavior followed by its opponent response (aggressive or submissive behavior) and the interaction lasted at least 15 s. We deemed the bout finished when one of the contestants exhibited a submissive behavior and did not engage in a new bout within 10 s. We determined the dominance by deeming the loser the individual that fled and its opponent the winner.

Organisms' measurements and RHP traits

To determine the traits that influence the RHP, we measured the cephalothorax length (CL), both right and left claw length (CwL) and height (CwH) of all individuals using a caliper rule. We used two claw measurements since they are both related to claw strength and may influence the probability of winning a contest (DENNENNMOSER & CHRISTY, 2013; PALAORO et al., 2014). To investigate if individuals exhibited dimorphism between claws, we performed two ANCOVAs for each species testing the relation between CL and each of the claw measurements. We used each claw measurements (CwL and CwH) as the response variable, CL as the explanatory variable and the claw sizes (right and left) as a factor. We did not find differences between claw sizes for both species or interactions between CL and claw position (Table SI; Figure 1a, b and 2a, b). Thus, since individuals exhibited no dimorphism between claws and no functional (i.e. feeding, fighting) differences can be regarded, we choose to use only measurements from one claw (the right claw) to our further analysis. In addition, we found a positive allometry between claw measurements (i.e. length and height) and body size (Table SI; Figure 1a, b and 2a, b respectively). Therefore, to obtain claw measurements independent of body size, we performed two linear regressions using the CL as the explanatory variable and CwL and CwH as response variables and used the standardized residuals of each model in the remainder of the analyses.

Next, we randomly chose a focal individual of each pair, and used only this individual in RHP traits' analyses to avoid pseudo-replication. Then, we calculated a proportion between the trait measure of the focal individual and its opponent (1 + (focal individual/opponent); BRIFFA et al., 2013). If the measured traits affect the winning chances, we expected that winners would have higher values than losers. As RHP can be composed by more than one trait (BRIFFA et al., 2013), we used logistic models to test the influence of different traits in contest outcome (i.e. winning probability). We used the RHP traits as explanatory variables, and the focal individual dominance (winner = 1; loser = 0) as the response variable within a logistic regression. The models were composed by each RHP trait alone and the following combinations: CL + CwL; CL + CwH; CwL + CwH; CL + CwL + CwH. Since we have seven possible models of how RHP traits influence the winning probability, we used the Akaike's Information Criterion corrected for small samples (AICc) to select the most parsimonious models. We considered the models with $\Delta \leq 2$ the most parsimonious (BURNHAM & ANDERSON, 2002), and we used the traits contained in these AICc models as proxies for the RHP in remainder analyses.

Distinguishing between assessment models

To distinguish between WOA, OOA and SAM/CAM, we performed a multiple linear regression using the data of random pairs (set-up 1), following TAYLOR and ELWOOD's (2003) framework. We used contest duration as the response variable and the RHP traits selected in prior analysis as the explanatory variables. We then compared the results to the predictions of the assessment models (Table I). If required, we used two further analyses to distinguish between SAM and CAM. First, we performed another multiple linear regression using the data from the RHP-matched pairs of the non-perforated divider treatment (set-up 2). We also used contest duration as the response variable and the RHP traits as the explanatory variables. Second, we tested if the contest duration was different according to the divider type using a t-test. In this analysis, we considered contest duration of RHP-matched pairs as the response variable and the divider type as factor (two levels: non-perforated divider and perforated divider). We also used a Levene test to compare variances between treatments with the same data. Afterwards, we compared the results to the predictions of the assessment models (Table I). We performed all analyses using the R environment (R DEVELOPMENT CORE TEAM, 2013). We used the 'bbmle' package (BOLKER & R DEVELOPMENT CORE TEAM, 2013) for the AIC analysis and the 'car' package (FOX et al., 2011) for the Levene test.

Results

Pacifastacus leniusculus

From the AICc results, we selected CL and CwL as the most parsimonious RHP traits (Table II) and performed all further analyses using these traits as RHP proxies. For random pairs, we found a significant negative relation between contest duration and the CL of winners, and a significant positive relation between contest duration and the CL of losers (Figure 3; Table III). No significant relation was found between contest duration and the CwL of winners or losers (Table III). For RHP-matched pairs of the non-perforated treatment, contest duration had no relation with both CL and CwL of winners and losers (Figure 4; Table IV). No significant difference was found between the two divider types ($t_{14} = 0.79$; $P = 0.44$; Figure 5), and the variances were also not significantly different ($F = 1.64$; $p = 0.22$; Figure 5).

Parastacus brasiliensis

The model containing the combination of CL and CwL was indicated as the most parsimonious (Table II). Consequently, we performed all further analyses using CL and CwL as RHP predictors. In the multiple linear regression in random pairs we found no relation between contest duration and the CL of winners; and a significant positive relation between contest duration and the CL of losers (Figure 3; Table III). No significant relation was found between contest duration and the CwL of winners or losers (Table III).

Discussion

Our hypothesis that *P. leniusculus* and *P. brasiliensis* would have distinct assessment strategies for contests was corroborated. Indeed, the distinct life histories of each species could have selected adaptations that resulted in distinct assessment strategies: *P. leniusculus* appears to rely on a mixed strategy of the CAM and SAM models, using a mix of costs and information on the opponent to flee, whereas *P. brasiliensis* relies on the WOA model strategy during contests. Below, we discuss why species uses that particular assessment model, and discuss how life history may affect decision making during contest.

Pacifastacus leniusculus: solitary and aggressive

Our results support a cognitively more complex assessment strategy in which individuals of *P. leniusculus* rely not only on their own RHP but also on the opponent's RHP. The random pairs experiment predicted either CAM or SAM models, and our subsequent results were unable to distinguish between those two models. The RHP-matched pairs from the non-perforated divider treatment corroborated the SAM model as the best model.

However, this evidence is supported on a non-significance, which does not corroborate an argument (BRIFFA & ELWOOD, 2009). Thus, we also tested for the SAM model based on significant results – if the individuals were using information from the opponent to give up, contests with perforated dividers would have lower duration than contests in non-perforated divider. Nevertheless, we found no significant results, suggesting that individuals were not communicating, at least at that stage of the contest.

The assessment strategy in *P. leniusculus* may be a consequence of its life history (i.e. ability to create a water flow during contests and exchange urine cues, BREITHAUPT, 2001; BREITHAUPT & EGER, 2002; high motivation to fight and energy expenditure in contests, SÖDERBÄCK, 1992; TIERNEY et al., 2000). In crustaceans, urine release is capable of altering contests dynamics as it is related to fighting motivation (BREITHAUPT et al., 1999). From that point of view, the contestants may rely on urinary cues and use that information in decision-making, as predicted by the SAM model. A physiological cue such as urine may be considered reliable. However, as symmetry between contestants' increases, more information is needed to make a decision (HACK, 1997). Previous studies on crustaceans provided evidence of a mixed assessment strategy. Even though the exchange of information through urine suggested lower fighting motivation in losers, individuals apparently also relied on costs inflicted by the opponents' claws (PALAORO et al., 2014). Therefore, the exchange of information provided by chemical and tactile information lead to the combination of the two assessment models SAM and CAM, respectively, as the contest escalates.

Likewise, *P. leniusculus* may be using a mixed strategy to acquire a more reliable set of information in contests. Studies on assessment strategies have been increasingly suggesting that individuals switch between strategies depending on asymmetries between contestants and contest escalation (e.g. HSU et al., 2008; MESTERTON-GIBBONS & HEAP, 2014; PALAORO et al., 2014). MESTERTON-GIBBONS and HEAP (2014) suggested a mixed model that switches between mutual and self-assessment strategies according to resource value. Although we had no conspicuous resource in the arena, individuals could have considered the arena a low value resource. The model of MESTERTON-GIBBONS and HEAP (2014) predicted that, when resource value is low, individuals would switch from CAM to SAM as the contests escalates. Our data fits that prediction since we found mixed results. Furthermore, we only tested for information exchange during the moments before the contests, and individuals may be exchanging information later on the contest. Investigating contest escalation between contestants of matched and unmatched RHPs, and acquisition of clear and valuable resources could provide answers on how strategies switch in our studied species.

Parastacus brasiliensis: the burrow-dwelling

Fighting motivation is related to both RHP and resource value (ARNOTT & ELWOOD, 2008). If resources are highly available, fighting motivation may decrease and lead to less costly contests. The majority of resources used by *P. brasiliensis* are available inside a burrow, and even though the burrow itself can be considered a resource, we would not expect intense disputes for burrows in this species. *Parastacus brasiliensis* dig burrows in riverbanks, often below the water level (BUCKUP, 2003), which makes the soil loose and moist. These soils are easier to dig and consume less energy than hard and dry soils (LOVEGROVE, 1989). Thus, although being a vital resource for the individual, it is easily acquired by adults. Indeed, the easiness of obtaining resources has been discussed to favor a more social behavior toward burrowing crayfish conspecifics (RICHARDSON, 2007).

In addition to behavioral modifications, groups well adapted to terrestrial life also exhibit physiological changes such as recognition systems through chemical cues (LINSENMAIR, 2007). Among crayfish species, water flow has an important role in the exchange of information. However, since *P. brasiliensis* has no data regarding a communication system adapted to a more terrestrial life, it is possible that this burrowing species struggles with acquiring reliable information inside burrows. Therefore, where water flow is absent, the amount of urine released by a potential opponent may not be a reliable cue. In terms of decision-making, the burrower life context could have selected a less complex and cognitively simpler assessment strategy such as self-assessment (TAYLOR & ELWOOD, 2003). If information from urine release is not reliable in the semiterrestrial environment, burrowing crayfish may have adapted their assessment strategy relying on more trustworthy information sources such as the individual's own RHP. Therefore, the high availability of resources together with the more social behavior in a semiterrestrial environment may have favored the development of the WOA strategy among burrowing crayfish.

Influence of the life habit on decision making

The species herein studied show markedly differences in agonistic behavior as results from adaptations to distinct life habits. While *P. leniusculus* readily engage in contests

(TIERNEY et al., 2000), *P. brasiliensis* appears to be more tolerating towards conspecifics (DALOSTO et al., 2013). The adaptations suffered by these two crayfish species show how important life habit can be within a group. Since life context influences RHP and hence, fighting ability (ARCHER, 1988), it is possible that the assessment strategies were also influenced by behavioral adaptations. From this point of view, natural selection may have favored assessment strategies that evolved along with habitat pressures regarding aggressive interactions as well as resource value and acquisition.

The intrinsic aspects of the contestants' abilities (i.e. RHP) present objective components to determine the outcome of a contest (STOCKERMANS & HARDY, 2013). In this case, the strategy in contests would be modulated directly by the ability of contestants in acquiring resources through contests. Nevertheless, subjective factors are of a great importance in defining the context in which contestants are found. The resource value may alter fighting motivation and result in an unexpected result at first sight (STOCKERMANS & HARDY, 2013). Herein, we used RHP and contest duration as tools to designate the assessment strategies of two crayfish species. We were also able to present differences in the assessment strategies of two crayfish species and suggest the importance of life habit in assessing information during agonistic interactions. Even though our results did not show a clear strategy in assessment during contests, we were able to suggest the models that are most likely to be used in both studied species. While *P. leniusculus* may be using a mixed strategy (CAM and SAM) due to escalated contests and urinary cues, *P. brasiliensis* would rely on a WOA strategy concerning the semiterrestrial life constraints. Analysis on the level of aggressiveness of the studied species could help enlightening the assessment strategies as fighting escalation may be related to information gathering. Therefore, differences in the aggression towards a conspecific may also be a reliable measurement of asymmetries in RHP evaluation.

Acknowledgements

We would like to thank CNPq for the scholarship to J.R.C., CAPES for the scholarship to A.V.P. and productivity grant (process 308598/2011-3) to S.S. Thank you to Mr. Ângelo Baggio, Mr. Alderi Baggio and Mrs. Lúcia Baggio for kindly allow us to collect the animals in their property. Thank you to the Biological Sciences laboratory in the University of Hull, UK, for providing equipment and space. We would also like to thank

Sonia Jennings for taking good care of the crayfish and Vic Swetez for technological assistance.

References

ARCHER, J. **The Behavioural Biology of Aggression**. Cambridge: Cambridge University Press, 1988.

ARNOTT, G. & ELWOOD, R. W. Information gathering and decision making about resource. **Animal Behaviour** 76: 529-542, 2008.

ARNOTT, G. & ELWOOD, R. W. Assessment of fighting ability in animal contests. **Animal Behaviour** 77: 991-1004, 2009.

BERGMAN, D. A. & MOORE, P. A. Field observations of intraspecific agonistic behavior of two crayfish species, *Orconectes rusticus* and *Orconectes virilis*, in different habitats. **Biological Bulletin** 205: 26-35, 2003.

BERGMAN, D. A.; MARTIN, A. L. & MOORE, P. A. Control of information flow through the influence of mechanical and chemical signals during agonistic encounters by the crayfish, *Orconectes rusticus*. **Animal Behaviour** 70 : 485-496, 2005.

BERRY F. C. & BREITHAUPT T. Development of behavioural and physiological assays to assess discrimination of male and female odours in crayfish, *Pacifastacus leniusculus*. **Behaviour** 145: 1427-1446, 2008.

BERRY F. C. & BREITHAUPT T. To signal or not to signal? Chemical communication by urine-borne signals mirrors sexual conflict in crayfish. **BMC Biology** 8: 25, 2010.

BOLKER, B. & R Development Core Team. bbmle: Tools for general maximum likelihood estimation. R package version 1.0.13. Vienna, Austria: R Foundation for Statistical Computing, 2013 http://cran.r-project.org/package-bbmle.

BREITHAUPT, T. & EGER, P. Urine makes the difference: chemical communication infighting crayfish made visible. **Journal of Experimental Biology** 205: 1221-1231, 2002.

BREITHAUPT, T.; LINDSTROM, D. P. & ATEMA, J. Urine release in freely moving catheterised lobsters (*Homarus americanus*) with reference to feeding and social activities. **Journal of Experimental Biology** 202: 837–844, 1999.

BREITHAUT, T. Fan organs of crayfish enhance chemical information flow. **The Biological Bulletin** 200: 150-154, 2001.

BRIFFA, M. & ELWOOD, R. W. Difficulties remain in distinguishing between mutual and self-assessment in animal contests. **Animal Behaviour** 77: 759-762, 2009.

BRIFFA, M.; HARDY, I. C. W.; GAMMELL, M. P.; JENNINGS, D. J.; CLARKE, D. D. & GOUBAULT, M. Analysis of animal contest data. *In*: HARDY I. C. W. & BRIFFA M. eds. **Animal Contests**: 47-85. Cambridge, U.K.: Cambridge University Press, 2013.

BUCKUP, L. & ROSSI, A. O gênero *Parastacus* no Brasil. **Revista Brasileira de Biologia** 40: 663-381, 1980.

BUCKUP, L. Família Parastacidae. *In*: G. A. S. MELO (ed.), **Manual de identificação dos Crustacea Decapoda de água doce do Brasil**: 117-141. São Paulo: Editora Loyola, 2003.

BURNHAM, K. P. & ANDERSON, D. R. **Model Selection and Multimodel Inference: a Practical Information-theoretic Approach**. New York: Springer, 2002.

CRANDALL, K. A. & BUHAY, J. E. Global diversity of crayfish (Astacidae, Cambaridae, and Parastacidae-Decapoda) in freshwater. **Hydrobiologia** 595:295-301, 2008.

DALOSTO, M. M.; PALAORO, A. V.; COSTA, J. R. & SANTOS, S. Aggressiveness and life underground: the case of burrowing crayfish. **Behaviour** 150: 3-22, 2013.

DANKS, H. V. The elements of seasonal adaptations in insects. **Canadian Entomologist** 139: 1-44, 2007.

DENNENNMOSER, S. & CHRISTY, J. The design of a beautiful weapon: compensation for opposing sexual selection on a trait with two functions. **Evolution** 67:1181-1188, 2013.

ELWOOD, R. W. & ARNOTT, G. Understanding how animals fight with Lloyd Morgan's canon. **Animal Behaviour** 84: 1095-1102, 2012.

ENQUIST, M. & LEIMAR, O. Evolution of fighting behaviour: decision rules and assessment of relative strength. **Journal of Theoretical Biology** 102: 387-410, 1983.

FOX, J.; WEISBERG, S.; R DEVELOPMENT CORE TEAM. car: An R companion to applied regression. R package version 1.0.13. Vienna, Austria: R Foundation for Statistical Computing, 2011 http:// http://cran.r-project.org/web/packages/car/index.html.

HACK, M. A. Assessment strategies in the contests of male crickets, *Acheta domesticus* (L.). **Animal Behaviour** 53: 733-747, 1997.

HARDY I. C. W., GOUBAULT M. & BATCHELOR T. P. Hymenopteran contests and agonistic behaviour. *In*: HARDY I. C. W. & BRIFFA M. eds. **Animal Contests**: 147-177. Cambridge, U.K.: Cambridge University Press, 2013.

HERBST, M. & BENNETT, N. C. Burrow architecture and burrowing dynamics of the endangered Namaqua dune mole rat (*Bathyergus janetta*) (Rodentia: Bathyergidae). **Journal of Zoology** 270: 420-428, 2006.

HODGE, M. A. & UETZ, G. W. A comparison of agonistic behavior of colonial web-building spiders from desert and tropical habitats. **Animal Behaviour** 50: 963-972, 1995.

HSU, Y.; LEE, S.; CHEN, M.; YANG, S. & CHENG, K. Switching assessment strategy during a contest: fighting in killifish *Kryptolebias marmoratus*. **Animal Behaviour** 75: 1641-1649, 2008.

JOHNSEN, S. I. & TAUGBØL, T. NOBANIS – Invasive Alien Species Fact Sheet – *Pacifastacus leniusculus*. **Online Database of the European Network on Invasive Alien Species**. NOBANIS, 2010 www.nobanis.org, Data of access 21/01/2015

KEMP, D. J. Contests behaviour in butterflies: fighting without weapons. *In*: HARDY I. C. W. & BRIFFA M. eds. **Animal Contests**: 134-146. Cambridge, U.K.: Cambridge University Press, 2013.

LINSENMAIR, K. E. Social biology of terrestrial Isopods. *In*: DUFFY J. E. & THIEL M. eds. **Evolutionary Ecology of Social and Sexual System - Crustaceans as Model Organisms**: 339-364. New York: Oxford University Press, 2007.

LOVEGROVE, B.G. The cost of borrowing by the social mole-rate (Bathyergidae) *Cryptomys damarensis* and *Heterocephalus glaber*: the role of soil moisture. **Physiological and Biochemical Zoology** 62: 449-469, 1989.

MESTERTON-GIBBONS, M. & HEAP, S. M. Variation between self- and mutual assessment in animal contests. **American Naturalist** 183: 199-213, 2014.

MESTERTON-GIBBONS, M.; MARDEN, J. H. & DUGATKIN, L. A. On wars of attrition without assessment. **Journal of Theoretical Biology** 181: 65-83, 1996.

MOORE, J. C.; OBBARD, D. J.; REUTER, C.; WEST, S. A. & COOK, J. M. Fighting strategies in two species of fig wasp. **Animal Behaviour** 76: 315–322, 2008.

MOORE, P. A. Agonistic behavior in freshwater crayfish: the influence of intrinsic and extrinsic factors on aggressive encounters and dominance. *In*: DUFFY, J.E. & THIEL, M. eds. **Evolutionary Ecology of Social and Sexual Systems – Crustaceans as Model Organisms**: 90-114. New York: Oxford University Press, 2007.

NORROCKY, M. J. Observations on the ecology, reproduction and growth of the burrowing crayfish *Fallicambarus* (*Creaserinus*) *fodiens* (Decapoda, Cambaridae) in north-central Ohio. **The American Midland Naturalist** 125: 75-86, 1991.

PALAORO, A. V.; DALOSTO, M. M.; COUTINHO, C. & SANTOS, S. Assessing the importance of burrows through behavioral observations of *Parastacus brasiliensis*, a Neotropical burrowing crayfish (Crustacea), in laboratory conditions. **Zoological Studies** 52: 2-9, 2013.

PALAORO, A. V.; DALOSTO, M. M.; COSTA, J. R. & SANTOS, S. Freshwater decapod (*Aegla longirostri*) uses a mixed assessment strategy to resolve constests. **Animal Behaviour** 95: 71-79, 2014.

PARKER, G. A. Assessment strategy and the evolution of fighting behaviour. **Journal of Theoretical Biology** 47: 223-243, 1974.

PAYNE, R. J. H. Gradually escalating fights and displays: the cumulative assessment model. **Animal Behaviour** 56: 651-662, 1998.

PAYNE, R. J. H. & PAGEL, M. Escalation and time costs in displays of endurance. **Journal of Theoretical Biology** 183: 185-193, 1996.

PAYNE, R. J. H. & PAGEL, M. Why do animals repeat displays? **Animal Behaviour** 54: 109-119, 1997.

PUNZALAN, D.; GUIAŞU, R.C.; BELCHIOR, D. & DUNHAM, D.W. Discrimination of conspecific-built chimneys from human-built ones by the burrowing crayfish, *Fallicambarus fodiens* (Decapoda, Cambaridae). **Invertebrate Biology** 120: 58-66, 2001.

R DEVELOPMENT CORE TEAM. R: A language and environment for statistical computing. Vienna, Austria: R Foundation for Statistical Computing. 2013.

REDDON, A. R.; VOISIN, M. R.; MENON, N.; MARSH-ROLLO, S. E.; WONG, M. Y. L. & BALSHINE, S. Rules of engagement for resource contests in a social fish. **Animal Behaviour** 82: 93-99, 2011.

RICHARDSON, A. M. M. Behavioral Ecology of Semiterrestrial Crayfish. *In*: DUFFY J. E. & THIEL M. eds. **Evolutionary Ecology of Social and Sexual System-Crustaceans as Model Organisms**: 319-338. New York: Oxford University Press, 2007.

SÖDERBÄCK, B. Predator avoidance and vulnerability of two co-occurring crayfish species, *Astacus astacus* (L.) and *Pacifastacus leniusculus* (Dana). **Annales Zoologici Fennici** 29: 253-259, 1992.

STOCKERMANS, B. C. & HARDY, C. W. Subjective and objective components of resource value additively increase aggression in parasitoid contests. **Biology Letters** 9: 20130391, 2013. http://dx.doi.org/10.1098/rsbl.2013.0391.

TAYLOR, P. W. & ELWOOD, R. W. The mismeasure of animal contests. **Animal Behaviour** 65: 1195-1202, 2003.

TIERNEY, A. J.; GODLESKI, M. S. & MASSANARI, J. R. Comparative analysis of agonistic behavior in four crayfish species. **Journal of Crustacean Biology** 20: 54-66, 2000.

VIEIRA, M. C. & PEIXOTO, P. E. C. Winners and losers: a meta-analysis of functional determinants of fighting ability in arthropod contests. **Functional Ecology** 27: 305-313, 2013. doi: 10.1111/1365-2435.12051

ZIMMERMANN, B.L.; AUED, A.W.; MACHADO, S.; MANFIO, D.; SCARTON, L.P. & SANTOS, S. Behavioural repertory of *Trichodactylus panoplus* (Crustacea: Trichodactylidae) under laboratory conditions. **Revista Brasileira de Zoologia**. 26: 5-11, 2009.

Figures and Tables

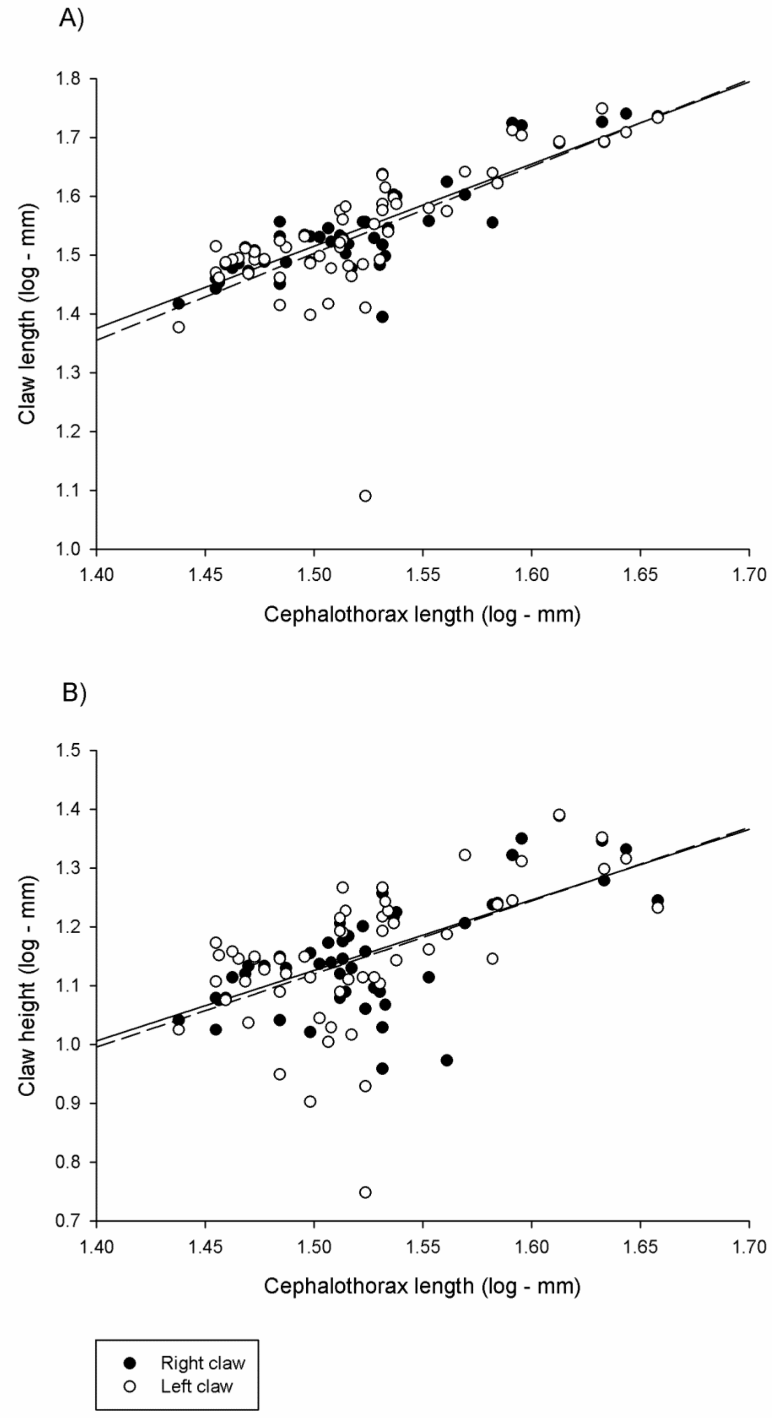


Figure 1. *Pacifastacus leniusculus*. A) Relation between cephalothorax length and the left and right claw height. The straight line represents the regression for the right claw length and the dashed line represents the regression for the left claw length. B) Relation between cephalothorax length and both claws height. The straight line represents the regression for the right claw height and the dashed line represents the regression for the left claw height.

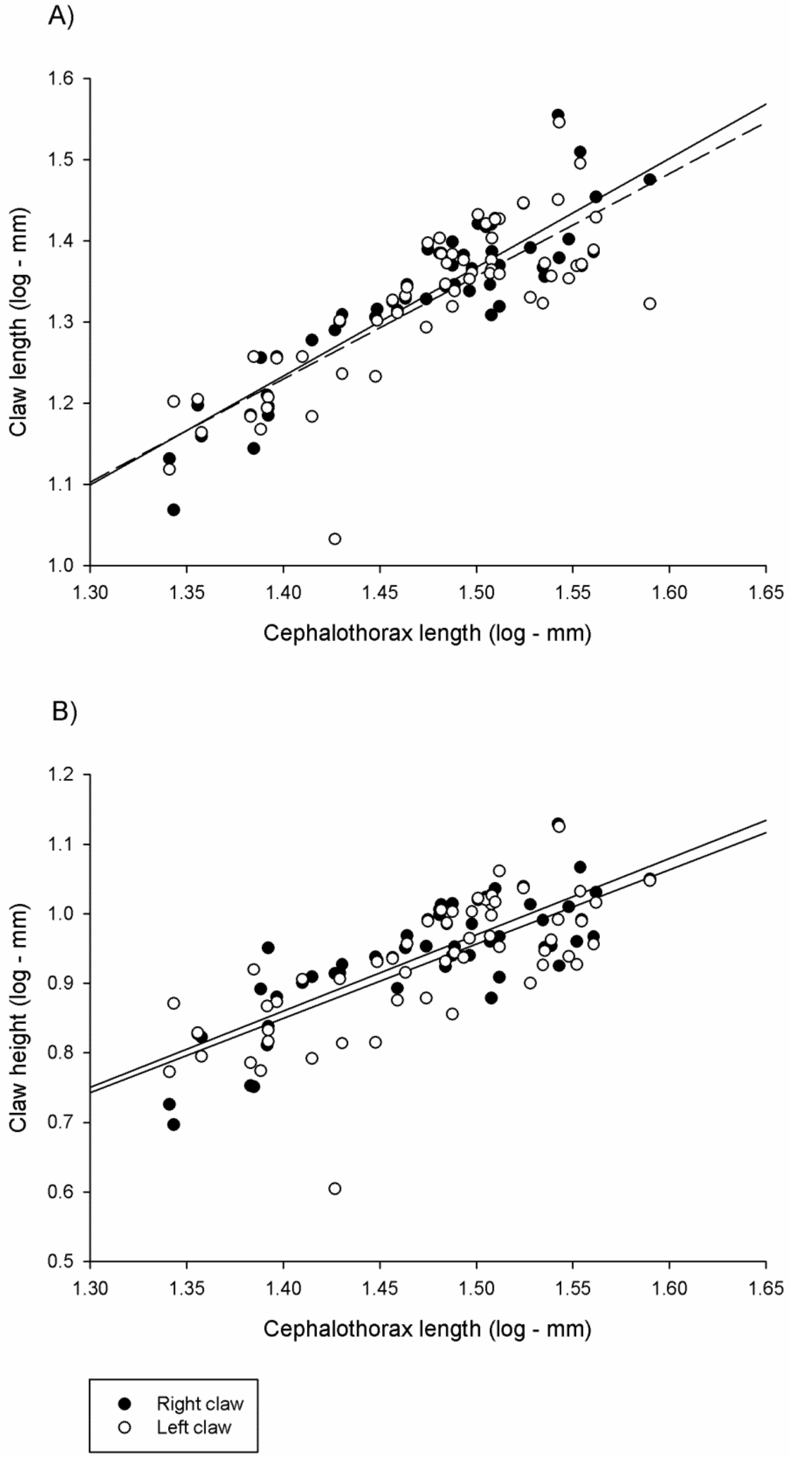


Figure 2. *Parastacus brasiliensis*. A) Relation between cephalothorax length and the left and right claw height. The straight line represents the regression for the right claw length and the dashed line represents the regression for the left claw length. B) Relation between cephalothorax length and both claws height. The straight line represents the regression for the right claw height and the dashed line represents the regression for the left claw height.

Table I. Predictions for the assessment models adapted from ARNOTT & ELWOOD, 2009.

	WOA	**CAM**	**SAM**	**OOA**
Pairs with different RHPs without prior information exchange (set-up 1)	Loser: contest duration increases as RHP traits increases Winner: contest duration increases as RHP traits increases or should not vary in relation to RHP traits	Loser: contest duration increases as RHP traits increases Winner: contest duration decreases as RHP traits increases	Loser: contest duration increases as RHP traits increases Winner: contest duration decreases as RHP traits increases	Loser: no relation Winner: contest decreases as RHP traits increases
Pairs with similar RHPs without prior information exchange (set-up 2)	Contest duration increases as mean RHP of the pair increases	Contest duration increases as mean RHP of the pair increases	No relation	Contest duration decreases as mean RHP of the pair increases
Comparison of pairs with similar RHPs with and without prior information exchange (set-up 3)	No variation in contest duration in relation to RHP proxies	No variation in contest duration in relation to RHP proxies	Contest duration decreases with prior information exchange	Contest duration decreases with prior information exchange

Table II. Results from the AICc to determine the best traits to predict RHP. AICc: Akaike's Information Criterion corrected for small samples; Δ_i: difference in the AICc value between the most parsimonious combinations and combination *i*.; w_i: Akaike weight of model *I*; df: degrees of freedom.

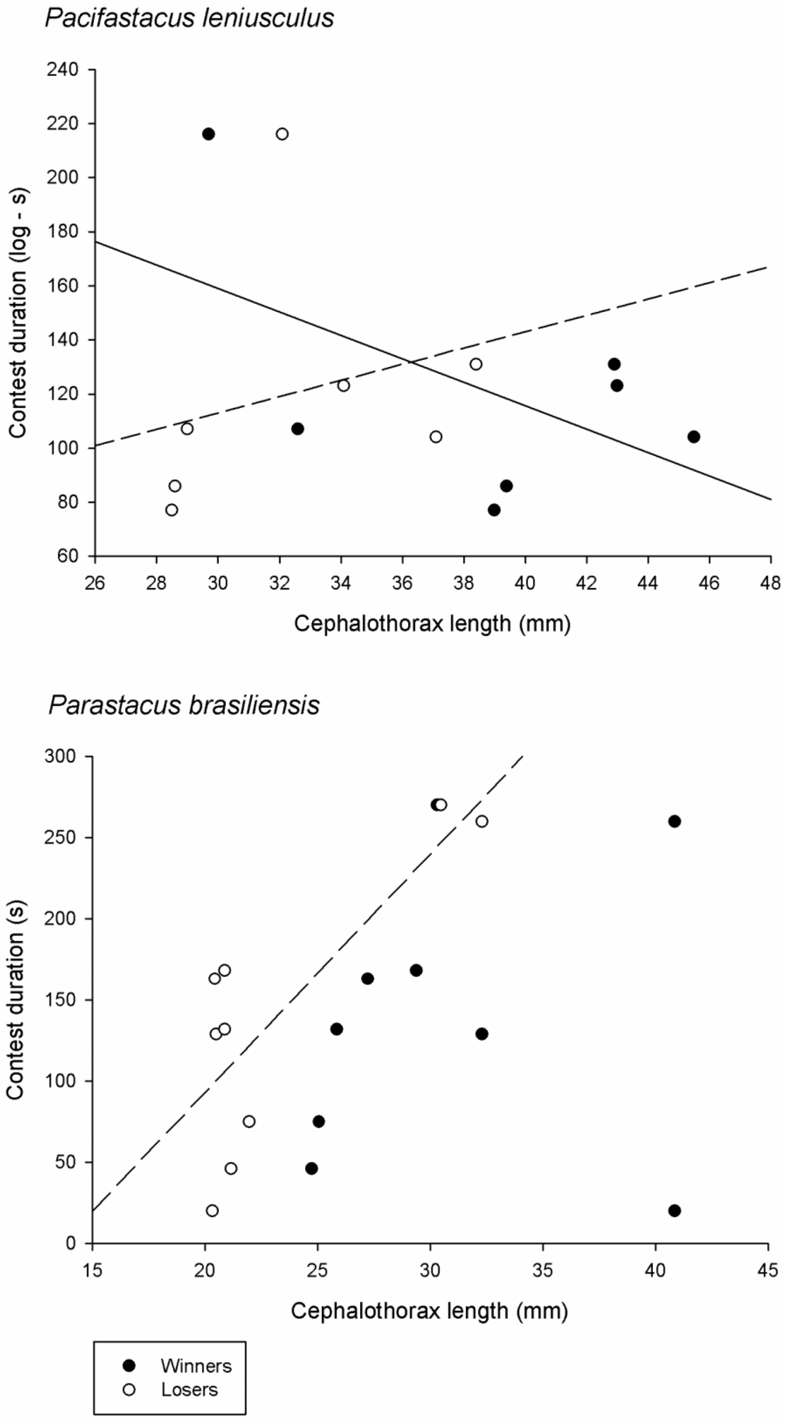
Combinations	**AICc**	**Δ_i**	**df**	**w_i**
Pacifastacus leniusculus				
1. Cephalothorax length	34.4	0.0	2	0.5162
4. Cephalothorax length + claw length	36.1	1.7	3	0.2243
5. Cephalothorax length + claw height	36.6	2.2	3	0.1690
6. Cephalothorax length + claw length + claw height	38.3	3.9	4	0.0752
3. Claw height	43.0	8.6	2	0.0071
2. Claw length	43.3	8.9	2	0.0061
7. Claw height + claw length	45.5	11.1	3	0.0020
Parastacus brasiliensis				
4. Cephalothorax length + claw length	29.1	0.0	3	0.6341
6. Cephalothorax length + claw length + claw height	31.8	2.7	4	0.1642
1. Cephalothorax length	32.1	3.0	2	0.1439
5. Cephalothorax length + claw height	34.6	5.5	3	0.0409
2. Claw length	37.6	8.5	2	0.0090
7. Claw height + claw length	38.4	9.3	3	0.0060
3. Claw height	40.8	11.7	2	0.0018

Table III. Non-perforated divider and random pairs' treatment: results from the multiple linear regression using contest duration as the response variable and cephalothorax length (CL) and claw length (CwL) of winners and losers as the explanatory covariables.

	Estimate	**F**	**P**
Pacifastacus leniusculus			
Intercept	5.15351	77.951	0.0126
Winner CL	-0.08544	24.910	0.0379
Loser CL	0.08518	19.501	0.0476
Winner CwL	0.02209	1.177	0.3913
Loser CwL	-0.08263	4.190	0.1773
Parastacus brasiliensis			
Intercept	-275.537	4.502	0.1011
Winner CL	-2.521	0.583	0.4877
Loser CL	21.608	15.444	0.0171
Winner CwL	-1.454	0.020	0.8935
Loser CwL	-27.099	3.104	0.1528

Table IV. *Pacifastacus leniusculus*. Non-perforated divider and RHP-matched pairs' treatment: results from the multiple linear regression using contest duration as the response variable and cephalothorax length (CL) and claw length (CwL) of winners and losers as the explanatory covariables.

	Estimate	**F**	**P**
Intercept	8.11086	1.557	0.267
Winner CL	0.25576	0.225	0.655
Loser CL	-0.36381	0.385	0.562
Winner CwL	-0.01764	0.003	0.954
Loser CwL	-0.01991	0.003	0.954



Pacifastacus leniusculus

Parastacus brasiliensis

Figure 3. Relation between cephalothorax length and contest duration in non-perforated divider and random pairs' treatment. The straight line represents the regression for winners and the dashed line represents the regression for losers.

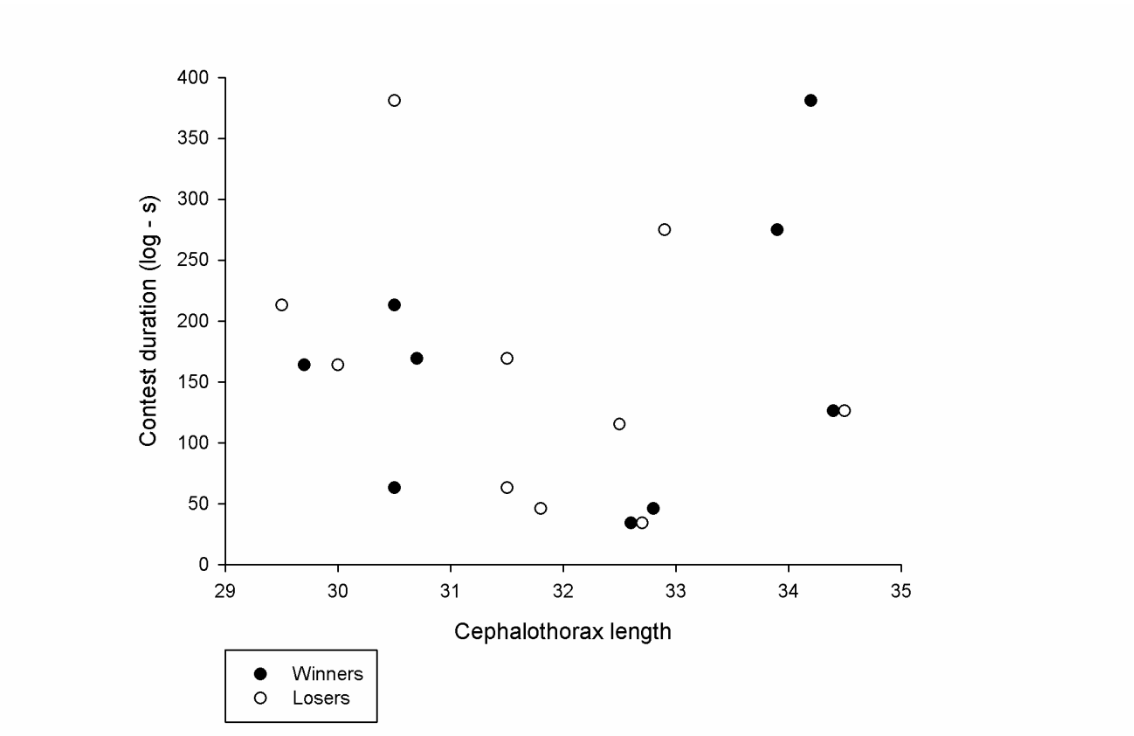


Figure 4. *Pacifastacus leniusculus*. Relation between cephalothorax length and contest duration in non-perforated divider and RHP-matched pairs' treatment.

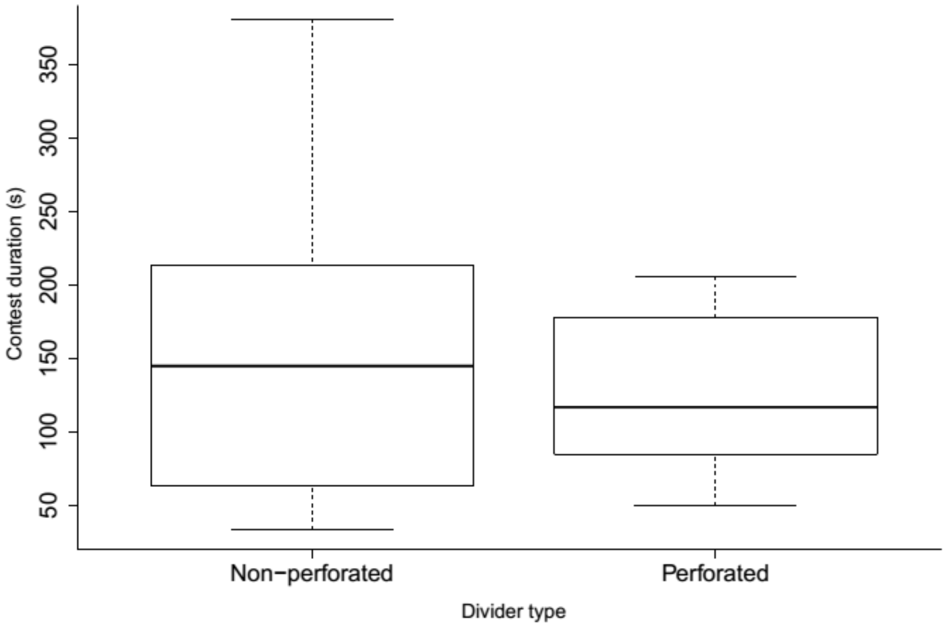


Figure 5. *Pacifastacus leniusculus*. Relation between RHP-matched non-perforated and perforated divider pairs' treatment two treatments based on the divider type and contest duration (two-tailed t test: $t_{14} = 0.7888$; $P = 0.4431$; Levene test: $F = 1.6419$; $P = 0.2183$).

Table V. Relation between cephalothorax length and the claws (right and left) length and height: results from the ANCOVA analyzes using each claw measurement (length and height) as the response variable, cephalothorax length as the explanatory co-variable and the claw sizes (right and left) as a factor.

		F	**dl**	**P**
Pacifastacus leniusculus				
Claw length	Claw side	0.737	1	0.392
	Cephalothorax length	150.058	1	<0.001
	Claw side * cephalothorax length	0.110	1	0.740
Claw height	Claw side	0.092	1	0.762
	Cephalothorax length	57.426	1	<0.001
	Claw side * cephalothorax length	0.019	1	0.890
Parastacus brasiliensis				
Claw length	Claw side	0.949	1	0.332
	Cephalothorax length	280.923	1	<0.001
	Claw side * cephalothorax length	0.231	1	0.632
Claw height	Claw side	1.349	1	0.248
	Cephalothorax length	151.880	1	<0.001
	Claw side * cephalothorax length	0.026	1	0.872

4 CONCLUSÕES

Neste estudo, foi observada uma diferença significativa entre os confrontos ao longo dos estágios de vida dos lagostins. Juvenis escavadores apresentaram confrontos mais curtos em relação aos adultos; sendo que não houve diferença na frequência de atos altamente agressivos e no número de confrontos. A duração do confronto foi diretamente afetada pela frequência de atos poucos agressivos, sendo que adultos apresentaram uma frequência desses atos maior que os juvenis. Esses resultados indicam que adultos e juvenis possuem motivações distintas ao engajar em um confronto e que podem utilizar estratégias de luta diferentes. Uma vez que o limiar de tolerância deve acompanhar o tamanho, juvenis podem atingir seu limiar de tolerância mais rapidamente e apresentar confrontos mais curtos. Além disso, os quelípodos podem ser determinantes na decisão de persistir ou desistir de um confronto. Enquanto juvenis possuem quelípodo pequenos e sofrem mudas com alta frequência, adultos possuem quelípodos grandes e de alto custo. Perder um quelípodo durante um confronto é altamente dispendioso para um adulto, e por isso, indivíduos adultos podem investir mais tempo de seus confrontos em atos pouco agressivos e atingir altos níveis de agressão somente quando necessário. Nossos resultados também mostraram que as estratégias de confronto diferem em lagostins adultos de hábitos de vida distintos. As análises de troca de informação durante os confrontos apontam para uma estratégia de avaliação baseada puramente na auto-avaliação em *P. brasiliensis*, enquanto *P. leniusculus* deve usar uma combinação de estratégias baseadas na avaliação mútua e nos custos infligidos pelo oponente. Essas estratégias divergentes podem ser consequência de pressões do meio em que vivem. Isto porque *P. leniusculus* pode contar com a troca de pistas químicas, originadas da liberação de urina durante os confrontos, e com o escalonamento dos confrontos. Por sua vez, o habito de vida semiterrestre dificulta a troca química e conduz *P. brasiliensis* a uma estratégia baseada somente na informação do próprio indivíduo.

Estudos acerca de lagostins escavadores no Brasil tem ganhado enfoque nos últimos anos. Entretanto, devido ao hábito de vida especializado, pouco se sabe sobre esses animais. O presente estudo dedicou dois capítulos a esclarecer as influências do hábito escavador no comportamento agonítico de *P. brasiliensis* através de mudanças ao longo dos estágios de vida e da comparação da dinâmica de confronto com a espécie *P. leniusculus*. Vista como uma espécie possivelmente sub-social, *P. brasiliensis* é ideal para estudos acerca de mudanças na agressão em prol da sobrevivência dentro das tocas e oferece oportunidades de comparação com lagostins não escavadores na influência do hábito de vida em adaptações comportamentais.