



## Contribution to the Supplement: 'Lobsters in a Changing Climate' Original Article

# Are juvenile Caribbean spiny lobsters (*Panulirus argus*) becoming less social?

Michael J. Childress\*, Katherine A. Heldt, and Scott D. Miller

Department of Biological Sciences, Clemson University, Clemson, SC 29634-0314, USA

\*Corresponding author: tel: +1 864 985 2384; fax: +1 864 656 0413; e-mail: [mchildr@clemson.edu](mailto:mchildr@clemson.edu)

Childress, M. J., Heldt, K. A., and Miller, S. D. Are juvenile Caribbean spiny lobsters (*Panulirus argus*) becoming less social? – ICES Journal of Marine Science, 72: i170–i176.

Received 7 September 2014; revised 19 January 2015; accepted 24 February 2015; advance access publication 19 March 2015.

Caribbean spiny lobsters are one of the most commercially important fisheries due in large part to their highly gregarious nature that facilitates their harvest by the use of traps or aggregation devices containing conspecifics. Aggregation in this species has been shown to be due to strong attraction to conspecific chemical cues that influence movement rates, discovery of crevice shelters, and den sharing behaviours. Although aggregation has been shown to have many potential benefits (reduction in exposure time and predation risk), it may also have significant costs as well (increase in predator encounters, disease transmission, and fishing mortality). We compared the results of three published and three unpublished Y-maze chemical cue choice experiments from 1996 to 2012 to determine if there has been a decrease in conspecific attraction by early benthic juvenile Caribbean spiny lobsters (15–55 mm carapace length, CL). We found that attraction to conspecific chemical cues decreased since 2010 and was significantly lower in 2012. Lobsters showed individual variation in conspecific attraction but this variation was unrelated to size, sex, or dominance status. We also found localized regional variation in conspecific attraction with lobsters from high shelter/high disease areas showing significantly lower conspecific attraction than those from low shelter/low disease areas. Given that conspecific attraction varies among individuals and potentially increases mortality through either natural (increased disease transmission) or fishery-induced (attraction to traps) mechanisms, we should play close attention to this loss of conspecific attraction in juvenile lobsters. Future studies should investigate both the causation and the ecological significance of changes in conspecific attraction in regions that vary in intensity of disease (PaV1) and fishing pressure.

**Keywords:** behaviour, Caribbean spiny lobster, disease, dominance, fishery-induced behavioural change, gregariousness, habitat loss, odour attraction, *Panulirus argus*, sociality.

## Introduction

Caribbean spiny lobsters, *Panulirus argus*, are one of the most important commercial crustacean fisheries in the Caribbean with estimated landings of 36 938 metric tons per year (SEDAR, 2005). There are multiple life history traits that contribute to their success including a rapid growth rate, enormous fecundity, long-lived larval stage with high dispersal potential, and the ability to use multiple habitat types throughout their development (Butler *et al.*, 2006; Childress and Jury, 2006). Settlement of *P. argus* postlarvae (puerulus stage) occurs in shallow water seagrass, hardbottom, or mangrove habitats after a 5- to 7-month larval period (Butler *et al.*, 2006). Newly settled benthic juveniles are solitary and widely dispersed in seagrass or macroalgae (Marx and Herrnkind, 1985) where they avoid predators primarily by crypsis (Butler *et al.*, 1997; Anderson *et al.*, 2013). They

transition from macroalgae to natural crevice shelters at ~15 mm carapace length (CL; Butler and Herrnkind, 1997; Childress and Herrnkind, 1997) and begin orienting toward conspecific chemical cues at this size (Childress and Herrnkind, 1996; Ratchford and Eggleston, 1998).

Another important characteristic that *P. argus* and most other commercially important Palinurids share is a gregarious temperament mediated by attraction to chemosensory cues from conspecifics (Childress, 2007). Conspecific attraction plays a key role in several ontogenetic habitat transitions of *P. argus*. The attractive chemical is released in the urine (Horner *et al.*, 2006) and detected by the antennules (Horner *et al.*, 2008). Chemosensory cues potentially influence the coastal orientation of postlarvae (Goldstein and Butler, 2009), the choice of settlement habitat by postlarvae

(Livingston-Zito and Childress, 2009), hasten the transition between macroalgal, seagrass, and hardbottom habitats (Childress and Herrnkind, 2001a), facilitates the choice of crevice shelters (Nevitt *et al.*, 2000), and decreases predation risk by reducing search time to find shelters (Childress and Herrnkind, 2001b). Shelter sharing may further decrease predation risk through dilution or group defence of shelters (Mintz *et al.*, 1994; Briones-Fourzán and Lozano-Álvarez, 2008) and coordination of mass migrations (Herrnkind *et al.*, 2001).

Despite the wealth of potential benefits for conspecific attraction, there are also potential costs to aggregation. Juvenile lobsters may choose to defend shelters rather than sharing them, showing significant aggression against intruding conspecifics (Berrill, 1975; Heldt *et al.*, 2015). Lobsters avoid the chemical cues of injured conspecifics presumably to reduce the risk of predator encounters (Parsons and Eggleston, 2005; Briones-Fourzán *et al.*, 2008). Conspecific attraction can bring small lobsters to shelters co-occupied by large lobsters as well as lobster predators such as snappers, groupers, and nurse sharks. While the large lobsters are not at risk from these predators, small lobsters attracted to large shelters may be at increased risk of predation (Schrattwieser, 1999). Conspecific attraction may also increase exposure to diseases, such as *Panulirus argus* virus 1 (PaV1), a lethal virus transmitted by close contact with infected individuals (Shields and Behringer, 2004; Butler *et al.*, 2008), although this may be mediated by the ability to healthy lobsters to detect and avoid infected individuals (Behringer *et al.*, 2006; Anderson and Behringer, 2013). Furthermore, fishers exploit conspecific attraction by using sublegal lobsters (called “shorts” by the fishers) as bait in commercial traps to attract legal-sized adults (Heatwole *et al.*, 1988), or by the addition of large aggregation devices (i.e. casitas) that draw sublegal and legal-sized lobsters together for easier harvest (Eggleston *et al.*, 1990; Briones-Fourzán *et al.*, 2000).

In this paper, we compare three experiments of conspecific attraction of juvenile Caribbean spiny lobsters from Florida Bay, FL (USA) to three previous studies on conspecific attraction in Florida (Anderson and Behringer, 2013), Mexico (Briones-Fourzán *et al.*, 2008), and the Bahamas (Ratchford and Eggleston, 1998). We also examine how covariates such as sex, size, dominance status, and regional quality (low and high disease) influence conspecific attraction. Specially, we hypothesize that individuals differ in their degree of conspecific attraction and these differences correlate with differences in size, sex, or dominance status. Furthermore, we hypothesize that conspecific attraction has decreased, and is lowest in regions with high disease prevalence.

## Methods

### Collection and housing conditions

Juvenile (15–55 mm CL) Caribbean spiny lobsters were collected by divers using hand nets from Florida Bay, FL (USA). Collections of juvenile lobsters in 2004, 2010, and 2011 were made on the bayside of Long Key. Collections in 2012 were made at one region with low disease (0–1% PaV1 visibly infected individuals) bayside of Lower Matecumbe Key and one region with high disease (5–10% PaV1 visibly infected individuals) bayside of Grassy Key. We recorded the sex, CL in mm, number of missing limbs, and the visible PaV1 status (disease present, disease absent, following the methods of Shields and Behringer, 2004) of every individual. Any animal showing visible signs of PaV1 infection or having been in contact with a PaV1 visibly infected individual while in captivity were excluded from further analysis. To accurately track

individuals, we attached a uniquely color-coded antennae band. Lobsters were shipped to Clemson University in Clemson, SC (USA) via overnight express service and were assigned to 120 l aquaria in size-matched pairs (max size difference 2.4 mm CL). Each housing tank included a single crevice shelter made from stacked patio pavers, a submersible aquarium filter, and a 10-cm air stone. Artificial seawater (Instant Ocean<sup>®</sup>) conditions were monitored daily and 25% water changes were performed weekly. Tanks were placed in a research greenhouse under naturally varying photoperiod from 10L:14D to 14L:10D depending on the season. Temperature varied from 22 to 28°C, salinity varied from 32 to 38 psu, and pH varied from 7.8 to 8.3. Each individual was fed *ad libitum* frozen pink shrimp daily and all uneaten food was removed the following day. Newly molted individuals were re-measured, re-tagged with an individually color-coded antenna band, and excluded from behavioural or chemical attraction observations for 72 h. There was no evidence of visible PaV1 infection for any of the individuals analysed in this study during the entire period of captivity (2–6 months).

### Dominance behaviour observations

We used direct behavioural observations to determine an individual's dominance status to examine if dominance increases or decreases conspecific attraction. Each pair of size-matched ( $\pm 2.4$  mm CL) lobsters housed together were observed for a minimum of 14 focal animal observations of 10 min duration, half shortly after sunrise (0700–1000 h) and half shortly after sunset (1900–2200 h) under low level (25 W) red light illumination. All acts of aggression were summed across all observations for each individual, including antenna whips, antenna pushes, and body pushes. The individual within each pair having initiated the most aggressive acts was considered the dominant (D) while its partner was considered the subordinate (S). Observations conducted in consecutive months found dominance status remained highly stable (<3% reversals) for up to 6 months in captivity (Heldt, 2013).

### Conspecific attraction experiments

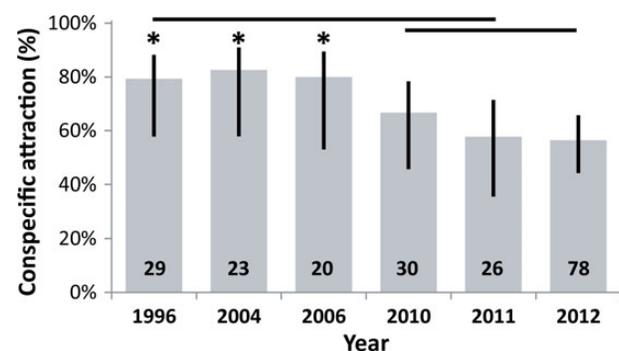
Conspecific chemical cue choice tests were conducted using a Y-maze wet table similar to the one depicted in Briones-Fourzán *et al.* (2008) (Figure 1) with the following differences. The dimensions of the Y-maze were 38 cm width  $\times$  18 cm depth  $\times$  126 cm length with a 100-cm long central divider and a volume of  $\sim 40$  l. The Y-maze contained an overflow drain pipe at the release end that emptied into a 600-l reservoir tank. Seawater from this tank was pumped through a filter and returned to the opposite end of the Y-maze via a PVC spray bar with valves to control the flow of water to each arm of the maze. Flow rates were set to  $\sim 2$  l per minute for each arm of the Y-maze. Each arm of the Y-maze contained a single crevice shelter made up of patio pavers. Behind each crevice shelter was an upstream compartment separated by a perforated opaque divider where a single conspecific could be held while releasing urine-born chemical cues.

Chemical cue trials were initiated at  $\sim 1000$ – $1400$  h and were run for 24 h. For each trial, a single test subject was chosen and single source lobster was selected from the same housing tank. The source lobster was placed in either the right or left compartment behind the shelters as determined by a flip of a coin, while the other compartment remained empty. Thus one shelter contained conspecific chemical cues while the other shelter contained a seawater control. A single test subject was transferred from its housing tank and placed at the choice end of the Y-maze opposite the shelters.

A perforated partition was positioned such that the test subject was blocked from entering the arms of the Y-maze but could sense chemical cues within the water coming down each arm of the maze. The test subject was allowed a 10-min acclimation period in this choice end of the Y-maze before the partition was removed allowing access to arms of the Y-maze. After 12 h and again after 24 h, the position of the test subject was recorded as either in the chemical cue shelter, in the control shelter, or in the no-choice end of the Y-maze. Very few trials resulted in no choice (<4%) and those individuals were retested at a later date up to three times until a choice preference was recorded. Conspecific attraction data were graphed as the percent of individuals that chose the chemical cue side of the Y-maze and the 95% confidence interval was calculated using the modified Wald method (Agresti and Coull, 1998).

### Meta-analysis of annual variation in conspecific attraction

Chemical cue preference test results from three previous studies (Ratchford and Eggleston, 1998; Briones-Fourzán *et al.*, 2008; Anderson and Behringer, 2013) were compared with three chemical cue preference tests reported here (Table 1). Although the methodologies and dimensions of their Y-maze choice tanks differed slightly, their designs were qualitatively the same (see Supplementary materials for Y-maze dimensions and specific differences in the methods of these previous studies). In all these studies, juvenile lobsters were



**Figure 1.** Conspecific attraction (mean  $\pm$  95% CI) measured as percent preference for conspecific odour over control odour in a Y-maze choice test vs. year the study was conducted. Numbers inside of bars represents the number of lobsters tested. Each bar represents a different conspecific attraction study; 1996 is from Ratchford and Eggleston (1998), 2004 is from this study, 2006 is from Briones-Fourzán *et al.* (2008), 2010 is from Anderson and Behringer (2013), 2011 is from this study, and 2012 is from this study. Asterisks above each bar indicate a significant preference ( $p < 0.05$ ) for conspecific chemical cues. Solid lines above the bars indicate results that are statistically similar using contingency table *post hoc* tests. Conspecific attraction significantly decreased between 2006 and 2010.

allowed the opportunity to choose among two arms of a Y-maze with either control water lacking conspecific chemical cues or water containing conspecific chemical cues. Choice was indicated by the occupation of a crevice shelter on either the conspecific or control sides of the Y-maze. To assure independence of observations, only the results of a single choice test for each individual were analysed.

The raw data from each of these three previous studies (number choosing control and number choosing conspecific cues) and the raw data from our trials conducted in 2004, 2011, and 2012 were analysed individually by year using a log-linear contingency test against the expected random distribution. Then the ratios of individuals attracted to conspecifics vs. control were compared with each other by log-linear contingency test. *Post hoc* comparisons were made by pairwise contingency tests to specifically identify those years that statistically differed from each other.

### Influence of dominance status on conspecific attraction

This experiment was specifically designed to test whether individuals differ in their attraction to conspecific chemical cues and whether these differences can be explained by sex, size, or dominance status. Sixteen juvenile lobsters (31.3–54.1 mm CL) from Long Key, FL were held in captivity for 6 months in winter of 2010 and ten juvenile lobsters (32.8–38.3 mm CL) from Long Key were held in captivity for 6 months in autumn 2011 while being tested for conspecific chemical cue attraction. Testing began immediately and was completed within the first 3 months of captivity. We found no change in conspecific attraction with time in captivity over this testing period (Heldt, 2013).

Conspecific chemical cue choice tests were then conducted using a Y-maze wet table as described above, but with the following modifications. Every individual was tested a total of six times with source lobsters from one of three dominance status classifications: familiar, unfamiliar dominant, and unfamiliar subordinate. Familiar source lobsters were individuals from the test subjects' housing tank. Unfamiliar dominant source lobsters were dominant individuals from a different housing tank while unfamiliar subordinate source lobsters were subordinate individuals from a different housing tank. Both unfamiliar dominant and unfamiliar subordinate source lobsters were selected to be close in size to the test subject lobster ( $\pm 7.8$  mm CL). Each test subject was tested twice with each of these three types of conspecific cues for a total of six choice tests. Since the dominance status of the source lobster was not found to influence the choice of an individual, we calculated the proportion of the six trials where an individual chose the conspecific odour as a measure of an individual's level of conspecific attraction. Analysis of covariance was used to analyse the % conspecific attraction after arcsine square-root transformation. Size was the covariate with sex and dominance status as fixed effects.

**Table 1.** Comparison of six conspecific attraction experiments.

Authors (year published)	Journal	Year tested	Collection location	Sample size (n)	Size min	Size max	Size mean
Ratchford and Eggleston (1998)	Animal Behaviour	1996	Bahamas	29	49.2	97.7	69.5
This study	ICES JMS	2004	Florida Keys	23	19.5	42.6	33.4
Briones-Fourzán <i>et al.</i> (2008)	Biological Bulletin	2006	Mexico	20	38.8	75.9	
Anderson and Behringer (2013)	MEPS	2010	Florida Keys	30	25.0	50.0	34.1
This study	ICES JMS	2010–11	Florida Keys	26	31.3	54.1	38.4
This study	ICES JMS	2012	Florida Keys	78	15.5	42.4	35.1

### Influence of region on conspecific attraction

This experiment was specifically designed to test whether individuals differ in their attraction to conspecific chemical cues and whether these differences can be explained by the local prevalence of disease from where they were collected. Seventy-eight juvenile lobsters (15.5–42.4 mm CL) from two regions of Florida Bay were held in captivity for 4 weeks in May and June 2012 while being tested for conspecific attraction. Thirty-six individuals came from the region near Lower Matecumbe Key. This region is characterized by a recent mass mortality of sponges, lower juvenile density (2–20 lobsters/625 m<sup>2</sup>), and low levels of PaV1 infection (0–1% of individuals visibly infected). Forty-two individuals came from the region near Grassy Key. This region is characterized by an intact sponge community, higher juvenile density (15–50 lobsters/625 m<sup>2</sup>), and high levels of PaV1 infection (5–10% of individuals visibly infected).

These chemical cue choice tests were conducted at the Keys Marine Laboratory, Long Key, FL (USA). Individuals were housed in 50 l tanks with flow-through filtered seawater from Florida Bay under natural photoperiod (13L : 11D), salinity (36–40 psu), and temperature (26–30°C). Conspecific attraction choice tests were conducted using a Y-maze wet table as described above with the following modifications. Trials were only 1 h duration beginning at 1900–2300 hours and ending at 2000–0000 hours. While this was a considerably shorter duration than our previous studies, it was necessary to complete the number of trials needed within a 1-week period for each cohort of lobsters. Analysis of video-taped trials in the laboratory have shown that Y-maze side association changes very little from the first to the last hour over a 24-h period (see Supplementary materials).

The number of individuals choosing control and number choosing conspecific cues were analysed by region to determine if conspecific attraction was random using a log-linear contingency test. Then the ratios of individuals attracted to conspecifics vs. control for the low and high disease regions were compared by log-linear contingency test.

## Results

### Meta-analysis of annual variation in conspecific attraction

Juvenile spiny lobster conspecific attraction has changed over the years ( $n = 206$ ,  $d.f. = 5$ ,  $G = 12.64$ ,  $p = 0.0270$ , Figure 1). Juvenile spiny lobsters showed a significant preference for conspecific chemical cues in 1996 ( $n = 29$ ,  $G = 5.665$ ,  $p = 0.0173$ , Ratchford and Eggleston 1998), 2004 ( $n = 23$ ,  $G = 5.53$ ,  $p = 0.0186$ , this study), and 2006 ( $n = 20$ ,  $G = 4.051$ ,  $p = 0.0441$ , Briones-Fourzan *et al.*, 2008). However, studies conducted since 2010 have not found a significant preference for conspecific chemical cues. Studies in 2010 ( $n = 30$ ,  $G = 1.724$ ,  $p = 0.1892$ , Anderson and Behringer, 2013), 2011 ( $n = 26$ ,  $G = 0.310$ ,  $p = 0.5778$ , this study), and 2012 ( $n = 78$ ,  $G = 0.644$ ,  $p = 0.4222$ , this study) suggest a gradual decrease in the attractiveness of conspecific chemical cues (Figure 1). An analysis of other possible variables that differed across these experiments including location, maze dimensions, flow rates, housing conditions, and experimental methods were no better explaining these differences than the year the study was conducted (see Supplementary Table S1).

*Post hoc* comparisons (Figure 1) found that our conspecific attraction results from 2004 were not significantly different from those measured by Ratchford and Eggleston (1998) or Briones-Fourzan *et al.* (2008). Similarly, our conspecific attraction results from 2011 were not significantly different from those measured by

Anderson and Behringer (2013). However, the combined results from 1996 to 2006 (58 of 72 choosing conspecific chemical cues) vs. the combined results from 2010 to 2012 (79 of 134 choosing conspecific chemical cues) were significantly different ( $n = 206$ ,  $G = 10.33$ ,  $p = 0.0013$ ).

### Influence of dominance status on conspecific attraction

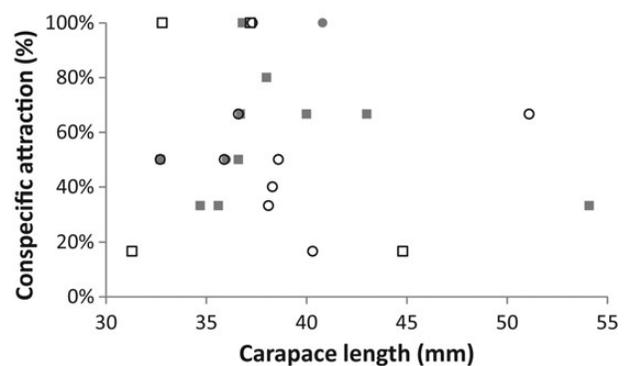
Juvenile lobsters establish dominance ranks with similar size conspecifics with one individual usually showing 10–50% more aggressive acts than its partner (Heldt *et al.*, 2015). But an individual's dominance status did not influence its preference for conspecific chemical cues ( $d.f. = 1$ , 22,  $F = 0.3613$ ,  $p = 0.5539$ ). Nor did an individual's sex ( $d.f. = 1$ , 22,  $F = 0.0340$ ,  $p = 0.8554$ ) or CL ( $d.f. = 1$ , 22,  $F = 0.2801$ ,  $p = 0.6019$ ) influence its conspecific attraction (Figure 2). Furthermore, there was no significant effect of the dominance status of the individual used as the chemical cue source on conspecific attraction ( $d.f. = 1$ , 22,  $F = 1.160$ ,  $p = 0.2922$ ). There were, however, individual differences in conspecific attraction with some juveniles choosing conspecific chemical cues consistently more than other individuals.

### Influence of region on conspecific attraction

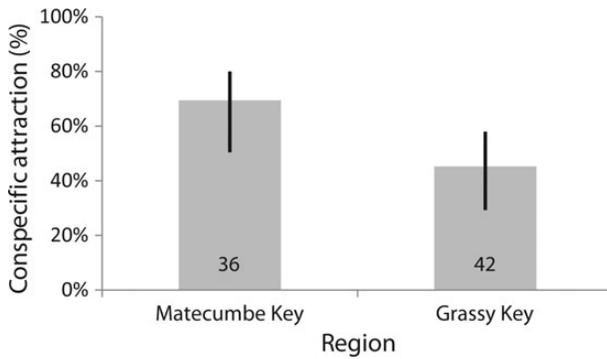
Lobsters collected from the Grassy Key region of Florida Bay (sponges present, high juvenile lobster densities, high prevalence of PaV1 visibly infection) had significantly lower conspecific attraction than those from the Lower Matecumbe Key region of Florida Bay (sponges absent, low juvenile lobster density, and low prevalence of PaV1 visibly infection). This regional effect was statistically significant ( $n = 78$ ,  $d.f. = 1$ ,  $G = 4.687$ ,  $p = 0.0304$ , Figure 3). However, since these regions differ in multiple factors, it is unclear if this difference is influenced by the prevalence of the disease, the availability of sponge crevice shelters, and/or the density of juvenile lobsters.

## Discussion

Juvenile Caribbean spiny lobsters historically have demonstrated a significant preference for conspecific chemical cues when tested in Y-maze choice tests (Ratchford and Eggleston, 1998; Briones-Fourzan *et al.*, 2008). However, in our conspecific attraction



**Figure 2.** Individual conspecific attraction measured as per cent preference for conspecific odour over control odour in six Y-maze choice tests vs. the size (mm CL) of the individual. Squares are males and circles are females. Individuals with shaded symbols are dominant and individuals with open symbols are subordinate. Although conspecific attraction varied among individuals, this variation was not related to size, sex, or dominance status.



**Figure 3.** Conspecific attraction (mean  $\pm$  95% CI) measured as per cent preference for conspecific odour over control odour in a Y-maze choice test vs. Florida Bay region where the lobsters were collected. Numbers inside of bars represent the number of lobsters tested. Matecumbe Key had lower lobster density, lower disease prevalence, and no sponge shelters. Grassy Key had higher lobster density, higher disease prevalence, and large sponge shelters. Neither of these groups of lobsters showed a statistically significant preference for conspecific chemical cues, but they did differ significantly in their ratio of individuals choosing conspecific vs. control sides of the Y-maze.

experiments, we demonstrate that juvenile Caribbean spiny lobster attraction to conspecific chemical cues has decreased through time. Individuals collected in 2004, significantly preferred familiar conspecific chemical cues in 82.6% of trials. However, individuals collected in 2011 and 2012 showed no significant preference for conspecific chemical cues (57.7 and 66.5% preferences, respectively). Since these experiments were conducted using the same Y-maze, and similar laboratory conditions, it cannot be dismissed as differences due to the experimental methodologies (see Supplemental materials). Furthermore, our observations corroborate the findings of Anderson and Behringer (2013) that also found no significant conspecific attraction for Florida Bay lobsters collected in 2010.

There are several possible explanations that we investigated to potentially explain this loss of conspecific attraction since 2010. First, we found that when individuals were tested multiple times, there were differences between individuals. These differences were not due to the individual's dominance status, sex, or size. Nor were these differences due to the identity or dominance status of the individual used as the source. However, individual variation indicates that a juvenile's attraction to conspecifics shows a degree of repeatability. This is what one would expect if the response to conspecific chemical cues has some level of heritability (Dingemans and Dochtermann, 2013). Heritable behavioural traits may respond to natural or artificial selection leading to changes in the mean and variance of the trait in future generations (Bell et al., 2009).

Previous studies have found that larger juvenile *P. argus* tend to be more aggressive and dominant over smaller juveniles (Berrill, 1975; Heldt et al., 2015) and that dominance status is also communicated through the release and detection of urine-borne cues (Shabani et al., 2008, 2009). Therefore, it is interesting that conspecific attraction did not differ based on the familiarity or the dominance status of the source individual. Perhaps this is because the source lobster was separated from the choosing lobster in the Y-maze and thus, was not in direct physical contact. Previous experiments have not repeatedly measured the response of the same individual multiple times, so the degree in which individual

repeatability in chemical cue preference has changed is unknown and should be investigated.

Second, variation in the conspecific chemical cue attraction across years and regions may reflect decreases in crevice shelter availability or increases in shelter competition. Historically, large sponges (>20 cm diameter) were the primary natural shelter occupied by juvenile spiny lobsters (Butler and Herrnkind, 1997; Forcucci et al., 1994; Herrnkind et al., 1997; Childress and Herrnkind, 1997). Beginning in 1991 and again in 2007, large regions of Florida Bay experienced wide-spread cyanobacteria blooms that killed nearly all sponges >20 cm diameter (Butler et al., 1995). Our 2004 collections of juvenile lobsters from the bayside hardbottom of Long Key, FL, occurred before the mass sponge mortality of fall 2007. However, after the mass sponge mortality, juvenile lobsters were largely restricted to areas with natural solution holes, artificial structures, or boat channels with exposed rock ledges and rubble piles like our Long Key collecting location. Therefore, lobsters collected after 2007 may have experienced fewer natural crevice shelters and elevated shelter competition. The loss of natural crevice shelters may have increased competition for those few remaining shelters. Although juvenile spiny lobsters are known to share shelters, there are only a finite number of lobsters that can receive protection from any crevice shelter. Decreased shelter number and increased shelter competition might explain why lobsters are less attracted to conspecific chemical cues, but our results did not support this prediction. Lobsters from regions hard hit by the mass sponge mortality actually showed higher attraction than lobsters from regions with an intact sponge community. They also show higher den fidelity (Heldt et al., 2015). Thus, where shelters were most abundant, lobsters were least attracted to the chemical cues of conspecifics. While habitat loss might have influenced the decrease in conspecific attraction in Florida Bay juveniles, it should be rigorously examined for other regions of the Caribbean where conspecific attraction has also decreased.

Third, concomitant with decreasing water quality and declining shelter availability, juvenile lobsters from Florida Bay are exposed to PaV1, a lethal viral pathogen that is transmitted by close contact with infected individuals. Although the origin of PaV1 in the Florida Keys is unknown (Shields and Behringer, 2004), and its clinical prevalence in the population (5–9%) has remained relatively stable since its discovery (Behringer et al., 2011), exposure to the pathogen may still be having a significant effect on variability between individuals in conspecific attraction. Behringer et al. (2006) have demonstrated that healthy lobsters can detect and avoid the chemical cues of visibly infected individuals but infected lobsters cannot. Therefore, one possible explanation for the decline in conspecific attraction may be an increased number of individuals with subclinical PaV1 infection within the populations of juvenile lobsters we tested. Moss et al. (2012) found that locations in the Florida Keys vary widely in the prevalence of individuals that test positive for PaV1 and that individuals may indeed be positive for PaV1 virus via PCR assay but not clinically infected (Behringer et al., 2012). However, this explanation is unlikely to explain the variation we observed in attraction to conspecific chemical cues because first, visibly infected lobsters were excluded from study; second, lobsters were held in captivity for 3–6 months after collection; and third, variation in attraction was not found to be related to the source individual.

Fourth, disease may have played a more subtle role in explaining our results. If individuals have variation in their attraction to conspecifics as demonstrated in dominance status experiment and

PaV1 eliminates those individuals that are most attracted to conspecific chemical cues, then those that survive may be less attracted to conspecifics. This explanation might be supported by our data since the prevalence of visibly infected individuals near Grassy Key (4.64%) was higher than the prevalence of visibly infected individuals near Lower Matecumbe Key (1.64%). Thus, where disease was relatively high, conspecific attraction was relatively low (45.2%) whereas where disease was relatively low, conspecific attraction was significantly higher (69.4%). Clearly, more research is needed on the localized effect of disease prevalence and its effect on the conspecific attraction of those that survive.

So what can explain a decrease in conspecific attraction? Certainly, the presence of PaV1 is widespread and could be causing a wide-spread decline in conspecific attraction (Behringer *et al.*, 2011). Researchers from Mexico also report that levels of juvenile conspecific attraction from recent Y-maze trials are showing similar declines (P. Briones-Fourzán, pers. comm.). This suggests that this pattern is not unique to juvenile lobsters from Florida. Alternatively, since spiny lobsters are fished very heavily throughout the Caribbean using baited traps or artificial shelters where those individuals most attracted to conspecifics are those most likely removed by the fishery. Is it possible that the decrease in conspecific attraction is due to fishery-induced selection on attraction behaviour? Other life history traits have been significantly impacted by intensive fishing including size at first reproduction, sperm limitation, and sex ratios (Kuparinen and Merilä, 2007). Like these life history traits, behaviours are also subject to the intensive selection the fishery exerts on the population (Uusi-Heikkilä *et al.*, 2008). Given the importance of conspecific attraction to growth, survival, and reproduction in Caribbean spiny lobsters, it would make sense that fishery-induced selection against gregariousness may be having a significant impact on lobster behaviour. This hypothesis will be very hard to test given that fishing occurs across the entire geographic range of this species and the dispersal of the larvae leads to genetic panmixis. Furthermore, why is the decline in conspecific attraction showing up now, when fishing practices have been removing the most gregarious individuals for several decades? These are the important questions that we should focus on in the future to better understand the impact of human activities on this commercially important species.

### Supplementary data

Supplementary material is available at the ICESJMS online version of the manuscript.

### Acknowledgements

This study was conducted in the Florida Keys National Marine Sanctuary under research perm its (FKNMS-2005-038, FKNMS-2008-035, and FKNMS-2009-118) and the collections of juvenile lobsters were permitted by the Florida Fish and Wildlife Conservation Commission (SAL-05-1071-SR, SAL-08-1071-SR, and SAL-11-1071-SR). Experiment D was conducted at the Keys Marine Laboratory. We thank Lonny Anderson, Cindy Lewis, Kylie Smith, Frank Kolencik, Kelsey McClellan, and Ken Sercy for assistance in the collection of animals and the collection of behavioural data. We thank J. Antonio Baeza, Donald Behringer, Rodney Bertelsen, Margaret Ptacek, Kylie Smith, and the Conservation of Marine Resources creative inquiry team for helpful suggestions on earlier versions of the manuscript. Funding was provided by the Clemson University SC Life and Creative Inquiry Programs (MJC), Sigma Xi Grant in Aid of Research, (KAH), Lerner Gray Fund for Marine

Research (KAH), Slocum-Lunz Foundation (KAH), and Clemson University Calhoun Honors Grant (SDM).

### References

- Agresti, A., and Coull, B. A. 1998. Approximate is better than exact for interval estimation of binomial proportions. *American Journal of Statistics*, 52: 119–126.
- Anderson, J. R., and Behringer, D. C. 2013. Spatial dynamics in the social lobster *Panulirus argus* in response to diseased conspecifics. *Marine Ecology Progress Series*, 474: 191–200.
- Anderson, J. R., Spadaro, A. J., Baeza, J. A., and Behringer, D. C. 2013. Ontogenetic shifts in resource allocation: colour change and allometric growth of defensive and reproductive structures in the Caribbean spiny lobster *Panulirus argus*. *Biological Journal of the Linnean Society*, 108: 87–98.
- Behringer, D. C., and Butler, M. J., IV. 2010. Disease avoidance influences shelter use and predation in Caribbean spiny lobster. *Behavioural Ecology Sociobiology*, 64: 747–755.
- Behringer, D. C., Butler, M. J., IV, Moss, J., and Shields, J. D. 2012. PaV1 infection in the Florida spiny lobster (*Panulirus argus*) fishery and its effects on trap function and disease transmission. *Canadian Journal of Fisheries Aquatic Sciences*, 69: 136–144.
- Behringer, D. C., Butler, M. J., IV, and Shields, J. D. 2006. Avoidance of disease by social lobsters. *Nature*, 441: 421.
- Behringer, D. C., Butler, M. J., IV, Shields, J. D., and Moss, J. 2011. Review of *Panulirus argus* virus 1—a decade after its discovery. *Diseases of Aquatic Organisms*, 94: 153–160.
- Bell, A. M., Hankison, S. J., and Laskowski, K. L. 2009. The repeatability of behaviour: a meta-analysis. *Animal Behaviour*, 77: 771–783.
- Berrill, M. 1975. Gregarious behaviour of juveniles of the spiny lobster, *Panulirus argus* (Crustacea: Decapoda). *Bulletin of Marine Science*, 25: 515–522.
- Briones-Fourzán, P., and Lozano-Álvarez, E. 2008. Coexistence of congeneric spiny lobsters on coral reefs: differences in conspecific aggregation patterns and their potential antipredator benefits. *Coral Reefs*, 27:275–287.
- Briones-Fourzán, P., Lozano-Álvarez, E., and Eggleston, D. B. 2000. The use of artificial shelters (casitas) in research and harvesting of Caribbean spiny lobsters in Mexico. *In Spiny Lobsters: Fisheries and Culture*, 2nd edn, pp. 420–446. Ed. by B. F. Phillips, and J. Kittaka. Blackwell Science, Abingdon.
- Briones-Fourzán, P., Ramírez-Zaldívar, E., and Lozano-Álvarez, E. 2008. Influence of conspecific and heterospecific aggregation cues and alarm odors on shelter choice by syntopic spiny lobsters. *Biological Bulletin*, 215: 182–190.
- Butler, M. J., IV, Behringer, D. C., and Shields, J. D. 2008. Transmission of *Panulirus argus* virus 1 (PaV1) and its effect on the survival of juvenile Caribbean spiny lobster. *Disease Aquatic Organisms*, 79: 173–182.
- Butler, M. J., IV, and Herrnkind, W. F. 1997. A test of recruitment limitation and the potential or artificial enhancement of spiny lobster populations in Florida. *Canadian Journal of Fisheries and Aquatic Sciences*, 54: 452–463.
- Butler, M. J., IV, Herrnkind, W. F., and Hunt, J. H. 1997. Factors affecting the recruitment of juvenile Caribbean spiny lobsters dwelling in macroalgae. *Bulletin of Marine Science*, 61: 3–19.
- Butler, M. J., IV, Hunt, J. H., Herrnkind, W. F., Matthews, T., Childress, M., Bertelsen, R., Sharp, W., *et al.* 1995. Cascading disturbances in Florida Bay, USA: cyanobacteria blooms, sponge mortality, and implications for juvenile spiny lobster *Panulirus argus*. *Marine Ecology Progress Series*, 129: 119–125.
- Butler, M. J., IV, Steneck, R. S., and Herrnkind, W. F. 2006. Juvenile and adult ecology. *In Lobster Biology, Management, Aquaculture and Fisheries*, pp. 263–309. Ed. by B. F. Phillips. Blackwell Publishing, London, UK.
- Childress, M. J. 2007. Comparative sociobiology of spiny lobsters. *In Evolutionary Ecology of Social and Sexual Systems: Crustaceans as*

- Model Organisms, pp. 271–293. Ed. by E. Duffy, and M. Thiel. Oxford University Press, Oxford, UK.
- Childress, M. J., and Herrnkind, W. F. 1996. The ontogeny of social behaviour among juvenile Caribbean spiny lobsters. *Animal Behaviour*, 51: 675–687.
- Childress, M. J., and Herrnkind, W. F. 1997. Den sharing by juvenile Caribbean spiny lobsters (*Panulirus argus*) in nursery habitat: co-operation or coincidence? *Marine and Freshwater Research*, 48: 751–758.
- Childress, M. J., and Herrnkind, W. F. 2001a. Influence of conspecifics on the ontogenetic habitat shift of juvenile Caribbean spiny lobsters. *Marine Freshwater Research*, 52: 1077–1084.
- Childress, M. J., and Herrnkind, W. F. 2001b. The guide effect influence on the gregariousness of juvenile Caribbean spiny lobsters. *Animal Behaviour*, 62: 465–472.
- Childress, M. J., and Jury, S. H. 2006. Behaviour. In *Lobsters: Biology, Management, Aquaculture and Fisheries*, pp. 78–112. Ed. by B. F. Phillips. Blackwell Publishing, Oxford, UK.
- Dinglemanse, N. J., and Dochtermann, N. A. 2013. Quantifying individual variation in behaviour: mixed effect modelling approaches. *Journal of Animal Ecology*, 82: 39–54.
- Eggleston, D. B., Lipcius, R. N., Miller, D. L., and Coba-Cetina, L. 1990. Shelter scaling regulates survival of juvenile Caribbean spiny lobster *Panulirus argus*. *Marine Ecology Progress Series*, 62: 79–88.
- Forcucci, D., Butler, M. J., IV, and Hunt, J. H. 1994. Growth and population dynamics of juvenile Caribbean spiny lobster, *Panulirus argus*, in Florida Bay, FL (USA). *Bulletin of Marine Science*, 54: 805–818.
- Goldstein, J. S., and Butler, M. J., IV. 2009. Behavioral enhancement of onshore transport by postlarval Caribbean spiny lobster (*Panulirus argus*). *Limnology and Oceanography*, 54: 1669–1678.
- Heatwole, D. W., Hunt, J. H., and Kennedy, F. S., Jr. 1988. Catch efficiencies of live lobster decoys and other attractants in the Florida spiny lobster fishery. *Florida Marine Research Publications*, 44: 1–15.
- Heldt, K. A. 2013. Individual behavioural variation of juvenile spiny lobster (*Panulirus argus*) denning behaviours and the role it plays in shelter competition during habitat loss. MS thesis, Clemson University, Clemson, SC, USA, 126 pp.
- Heldt, K. A., Bridges, W. C., Jr, and Childress, M. J. 2015. Behavioral response to habitat loss in juvenile spiny lobsters. *Marine Ecology Progress Series*, 521: 117–128.
- Herrnkind, W. F., Butler, M. J., IV, Hunt, J. H., and Childress, M. J. 1997. Role of physical refugia: implications from a mass sponge die-off in a lobster nursery in Florida. *Marine and Freshwater Research*, 48: 759–769.
- Horner, A. J., Nickles, S. P., Weissburg, M. J., and Derby, C. D. 2006. Source and specificity of chemical cues mediating shelter preference of Caribbean spiny lobsters (*Panulirus argus*). *Biological Bulletin*, 211: 128–139.
- Horner, A. J., Weissburg, M. J., and Derby, C. D. 2008. The olfactory pathway mediates sheltering behaviour of Caribbean spiny lobsters, *Panulirus argus*, to conspecific urine signals. *Journal of Comparative Physiology A: Neuroethology Sensory Neural and Behavioural Physiology*, 194: 243–253.
- Kuparinen, A., and Merilä, J. 2009. Determining and managing fisheries-induced evolution. *Trends in Ecology and Evolution*, 22: 652–659.
- Livingston-Zito, A. N., and Childress, M. J. 2009. Does conspecific density influence the settlement of Caribbean spiny lobster *Panulirus argus* postlarvae? *New Zealand Journal of Marine and Freshwater Research*, 43: 313–325.
- Marx, J. M., and Herrnkind, W. F. 1985. Macroalgae (*Rhodophyta*, *Laurencia* spp.) as habitat for young juvenile spiny lobsters, *Panulirus argus*. *Bulletin of Marine Science*, 36: 423–431.
- Mintz, J. D., Lipcius, R. N., Eggleston, D. B., and Seebo, M. S. 1994. Survival of juvenile Caribbean spiny lobster: effects of shelter size, geographic location and conspecific abundance. *Marine Ecology Progress Series*, 112: 255–266.
- Moss, J., Butler, M. J., IV, Behringer, D. C., and Shields, J. D. 2012. Genetic diversity of the Caribbean spiny lobster virus, *Panulirus argus* virus 1, and the discovery of PaV1 in lobster post-larvae. *Aquatic Biology*, 14: 223–232.
- Nevitt, G., Pentcheff, N. D., Lohmann, K. J., and Zimmer, R. K. 2000. Den selection by the spiny lobster *Panulirus argus*: testing attraction to conspecific odors in the field. *Marine Ecology Progress Series*, 203: 225–231.
- Parsons, M. D., and Eggleston, D. B. 2005. Indirect effects of recreational fishing on behaviour of the spiny lobster *Panulirus argus*. *Marine Ecology Progress Series*, 303: 235–244.
- Ratchford, S. G., and Eggleston, D. B. 1998. Size- and scale-dependent chemical attraction contribute to an ontogenetic shift in sociality. *Animal Behaviour*, 56: 1027–1034.
- Schratwieser, J. 1999. The impact of resident and transient predators on the population dynamics of juvenile Caribbean spiny lobster in Florida. MS thesis, Old Dominion University, Norfolk, VA, USA.
- SEDAR. 2005. SEDAR 08, Stock Assessment Report 3, Southeastern US Spiny Lobster. NOAA-SEFSC, 319 pp.
- Shabani, S., Kamio, M., and Derby, C. D. 2008. Spiny lobsters detect conspecific blood-borne alarm cues exclusively through olfactory sensilla. *Journal of Experimental Biology*, 211: 2600–2608.
- Shabani, S., Kamio, M., and Derby, C. D. 2009. Spiny lobsters use urine-borne olfactory signaling and physical aggressive behaviours to influence social status of conspecifics. *Journal of Experimental Biology*, 212: 2464–2474.
- Shields, J. D., and Behringer, D. C. 2004. A new pathogenic virus in the Caribbean spiny lobster *Panulirus argus* from the Florida Keys. *Diseases of Aquatic Organisms*, 59: 109–118.
- Uusi-Heikkilä, S., Wolter, C., Klefoth, T., and Arlinghaus, R. 2008. A behavioural perspective on fishing-induced evolution. *Trends in Ecology and Evolution*, 23: 419–421.

Handling editor: Jonathan Grabowski