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To breathe or fight? Siamese fighting fish differ when facing a real opponent or mirror image.

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Abstract
Displays are a feature of animal contest behaviour and have been interpreted as a means of gathering information on opponent fighting ability, as well as signalling aggressive motivation. In fish, contest displays often include frontal and lateral elements, which in the latter involves contestants showing their flanks to an opponent. Previous work in a range of fish species has demonstrated population-level lateralization of these displays, preferentially showing one side to their opponent. Mirrors are commonly used in place of a real opponent to study aggression in fish, yet they may disrupt the normal pattern of display behaviour. Here, using Siamese fighting fish, *Betta splendens*, we compare the aggressive behaviour of males to a mirror image and real opponent behind a transparent barrier. As this species is a facultative air-breather, we also quantify surface breathing, providing insights into underlying fight motivation. Consistent with previous work, we found evidence of population-level lateralization, with a bias to present the left side and use the left eye when facing a real opponent. Contrary to expectations, there were no differences in the aggressive displays to a mirror and real opponent, with positive correlations between the behaviour in the two scenarios. However, there were important differences in surface breathing, which was more frequent and of longer duration in the mirror treatment. The reasons for these differences are discussed in relation to the repertoire of contest behaviour and motivation when facing a real opponent.

Keywords: aggression, contests, lateralization, mirrors, surface breathing.
1. Introduction

Although there is considerable taxonomic variation in contest behaviour (Arnott and Elwood 2009a; Hardy and Briffa 2013), displays typically precede and intersperse with escalated fighting behaviour. These displays are usually interpreted as providing a means of gathering information on the size and fighting ability, termed resource holding potential (RHP), of an opponent (Parker 1974). However, evidence to support this interpretation is often lacking (Taylor and Elwood 2003; Arnott and Elwood 2009a; Elwood and Arnott 2012, 2013; Fawcett and Mowles 2013). An alternative view is that they serve to signal information about the aggressive motivation of each contestant (Camerlink et al. 2015).

The prefight display phases are typically dynamic processes involving the contestants interacting in a particular pattern. There are numerous examples across diverse taxa, and for invertebrates these include; the prefight cheliped displays of hermit crabs (Elwood et al. 2006), the waving of the enlarged claw during fiddler crab contests (Backwell et al. 2000) and of legs in spiders (Elwood and Prenter 2013). Commonly cited vertebrate examples include; the stereotyped displays of lizards (Van Dyk and Evans 2008) and frogs (Reichert and Gerhardt 2014), the mutual vocal displays occurring between male red deer (Clutton-Brock and Albon 1979) and fallow deer (Jennings et al. 2012), as well as the conspicuous lateral visual displays termed parallel walks occurring in these ungulates (Jennings and Gammell 2013).

Aggressive displays have been studied extensively in fish species, which lend themselves to laboratory and field studies (e.g. Enquist et al. 1990). The contest displays of many fish comprise frontal displays and lateral displays, which in the latter involves contestants showing their flanks to an opponent. During lateral displays, fish can align in two ways, with their heads either facing in the same direction (head to head) or in opposite
directions (head to tail). Moreover, a range of fish species has been shown to exhibit
population-level lateralization, preferentially showing one side to their opponent (Bisazza and
de Santi 2003; Reddon and Balshine 2010). For example, competing convict cichlids,
*Amatitlania nigrofasciata*, more commonly show their right than their left flank (Arnott et al.
2011; Elwood et al. 2014). When both contestants show their right side the head to tail
configuration results and this is more common than the head to head configuration (Arnott et
al. 2011). Such population-level lateralization of displays thus provides some predictability
and enables coordination of these agonistic interactions (Ghirlanda et al. 2009), potentially
facilitating a mutual assessment process (Arnott et al. 2011). Additionally, it may also act to
reduce the likelihood of injury should one fish escalate the contest (Rogers 1989; Bisazza et
al. 2000; Arnott et al. 2011). The importance of coordinating displays in fish, such as head to
tail positioning, can be examined by comparing mirror images to live opponents behind a
transparent partition (Elwood et al. 2014), because with a mirror the fish can never align in
the head to tail configuration and this key feature of the mutual display is lost.

Nevertheless, mirrors are frequently used instead of a live opponent in studies on
aggression in fish (Cantalupo et al. 1996; Earley et al. 2000; Wilson et al. 2011; Balzarini et
al. 2014). Mirror images might be a good choice of stimuli when repeated measures design
require several bouts and opponent variability can be avoided. However, the validity of using
mirrors for studying contest behaviour has been questioned. For example, mirror images fail
to elicit the same brain gene expression (Desjardins and Fernald 2010) or the same hormonal
responses (Oliveira et al. 2005) as live opponents. Furthermore, Elwood et al. (2014) recently
compared the displays of convict cichlids to a mirror and a real opponent, finding a lower
frequency of displays to a mirror but with individual displays of greater duration. This slower
pace of the interaction to a mirror suggested that social responses from opponents are a key
component necessary to elicit the normal repertoire of contest behaviour, as has also been suggested for lizards (Ord and Evans 2002). The presence of an appropriately responding opponent during aggressive displays thus appears to be a key driver necessary for the interaction to progress.

Although only focusing on displays in their study, Elwood et al. (2014) predicted that the disruption of normal display behaviour caused by the mirror may lead fish to abandon displays in favour of other activities, including other forms of aggression such as biting. This is one focus of the current study. In addition, Elwood et al. (2014) confirmed previous findings of population-level lateralization in the convict cichlid (Arnott et al. 2011), with a right-sided lateral display bias that was evident to both a real opponent and when facing a mirror. With Siamese fighting fish, *Betta splendens*, interacting with real opponents and when facing a mirror image there was evidence of population-level right side bias (Bisazza and de Santi 2003), but this was not found in other studies with a mirror (Cantalupo et al. 1996; Takeuchi et al. 2010). A recent study demonstrated a population-level right side preference to real opponents during early reproductive stages but not at late stages (Forsatkar et al. 2015).

Indeed, some individuals switched from a right side bias to a left side bias after spawning thus illustrating the confusion about laterality in this species. Here we use *B. splendens* in mirror and real opponent tests to examine population-level lateralization. Further, we test the predictions of Elwood et al. (2014) concerning the utility of a mirror image in eliciting normal contest behaviour and, in particular, if the motivation to compete is the same to a mirror as to a real opponent. *B. splendens* offers a widely used model for studies on aggression (e.g. Simpson 1968). Their pre-fight displays consist of raised opercula and spread fins during frontal displays and lateral displays (Castro et al. 2006). The frontal displays impair respiration and are physiologically demanding such that contestants will interrupt the display.
sequence at intervals to engage in surface breathing (Regan et al. 2015).

We recorded the frequency, total duration and the median duration of left lateral displays, right lateral displays, frontal displays, and surface breathing, as well as attempted bite frequency. The frequency and median duration should relate positively and negatively to vigour whereas total duration is the product of frequency and median duration and is the more commonly used measure of displays. Using this information we examine four key predictions. First, consistent with the findings of Elwood et al. (2014), we predict there will be evidence of population-level lateralization when displaying to both a mirror and real opponent. Second, we predict the mirror will elicit a decreased frequency of displays and longer duration of individual displays than when facing a real opponent. Third, we predict if the mirror impairs the normal sequence of displays, individuals may switch to more escalated aggression revealed by increased levels of attempted biting in the mirror treatment. Fourth, we examine the frequency and duration of surface breathing to gain insights regarding the effectiveness of mirrors compared to real opponents for eliciting agonistic displays. Because breathing rate has been linked to the vigour of displaying (Regan et al. 2015), we predict that should the display vigour differ between real and mirror opponents, there will be more surface breathing in the condition with higher vigour. However, there are two other aspects that might influence surface breathing. First, if the mirror offers a substandard stimulus resulting in a lower level of aggressive motivation then this might cause the fish to switch to breathing more frequently in the way that male newts breathe more readily when courting an unresponsive female (Halliday and Sweatman 1976). Note that the vigour of the displays might not necessarily differ with motivation because animals might not signal future intentions about persistence in contests (Dawkins and Krebs 1978). Another way that surface breathing might differ between real and mirror opponents is that fish might take their cues to breathe by the breathing
attempts of the opponent. With a mirror the apparent opponent will not be the first to go to the surface and will not be the first to resume displaying following surface breathing. This might disrupt the pattern of breathing when compared to a real opponent. Using the information on levels of surface breathing, coupled with information on contest behaviour, should enable us to disentangle which of the above scenarios is correct.

2. Methods

2.1. Animals

Thirty male, *B. splendens* were obtained from a local supplier (Grosvenor Tropicals, Belfast, U.K.) in batches of six size-matched subjects and housed in individual glass tanks measuring 30 x 20 cm and 20 cm high, with approximately 2 cm depth of gravel. A controlled artificial 12:12 h light:dark cycle was in place, tanks filled with 8 litres of treated tap water, aerated for 30 minutes per day, and maintained at a temperature of 21-23°C. Fish were fed daily with flake food, and on the day of experimental trials were fed after observations had concluded.

2.2. Experimental protocol

Tanks were aligned end to end with opaque partitions visually isolating the fish outside of test sessions (as per Elwood et al. 2014). For at least 7 days prior to the onset of experimental trials the fish were maintained in these individual tanks, thus ameliorating any behavioural effects of prior winning or losing experiences (reviewed in Hsu et al. 2006). Each fish was tested twice, once displaying against a mirror and once against a real opponent, in a random order and with a gap of 10-15 min between tests. When observations against the mirror were conducted the opaque partition between the two tanks was removed and immediately replaced with a 20 x 20 cm mirror and the focal fish was filmed for 15 minutes. When a real opponent
was used the opaque partition was removed from between the two tanks and the focal fish was filmed for 15 minutes. During filming, the laboratory was isolated from disturbance. Each focal fish was exposed to the stimulus fish in the tank to the right, except for the last in the row of tanks, which was moved so that it could see the first fish as the stimulus 24 h after moving. Thus all focal fish/stimulus fish combinations were novel and pseudoreplication was avoided.

2.3. Behavioural Measures

Of the 30 tested male subjects, 10 were omitted from further analyses due to a lack of display behaviour by the focal or stimulus fish in one or both situations. Results are therefore based on recordings from 20 focal fish. The films were observed and behavioural displays recorded using Observer v. 3.0 software (Noldus Technology, Wageningen, The Netherlands). The following activities were recorded: left lateral display, right lateral display, frontal display, bite attempt and surface breathing. A left lateral display was recorded if the fish was displaying its left lateral side at 45 degrees or less to the glass at the end of the tank closest to the mirror/opponent, while a right lateral display if the right side was shown at 45 degrees or less, and a frontal display was recorded if the fish was head on to the glass (>45 degrees) (as per Elwood et al. 2014). An attempted bite was recorded when a subject made open-mouthed contact with the glass/mirror, directed towards the real or perceived opponent. Surface breathing was recorded when a subject ceased displaying and began gulping air at the surface.

2.4. Ethical note

This study was carried out in accordance with the guidelines for the treatment of animals in behavioural research (ASAB 2012). Moreover, our experimental set-up prevented physical
contact between fish, eliminating the risk of injury as a result of aggression, and staged
interactions were brief (as recommended by Huntingford 1984). Following discussions with
the local Home Office veterinary inspector it was deemed that there was no likelihood of fish
being harmed by the procedure and thus no licence was required. Further, the maintenance of
fish in individual tanks without transferring for each test ensured there was minimal handling,
disturbance, or opportunity to succumb to harm from conspecifics. Following the
experimental phase fish were returned unharmed to the local supplier.

2.5. Statistical Analyses

From behavioural observations, we obtained the frequency, total duration and median duration
of each of the following activities; left lateral display, right lateral display, frontal display,
surface breathing. In addition, we obtained the frequency of attempted bites. The data were
not normally distributed and the nonparametric Wilcoxon matched-pairs signed ranks test for
non-independent data was used to compare responses to real opponents and mirror images.
The same test was used to compare the frequency, total duration, and median durations of left
and right lateral displays. Spearman rank correlations were used to examine relations between
displays to mirrors and real opponents for each display component, as well as relations
between surface breathing and agonistic activities. Although we used multiple tests,
Bonferroni corrections were not applied given they have been criticised (Nakagawa 2004) for
exacerbating the problem of low statistical power for behavioural studies, where the risk of
Type II errors is generally greater than the risk of Type I errors. Moreover, we were interested
in comparing overall patterns of behaviour between the two scenarios, therefore necessitating
the need for multiple comparisons. Finally, any significant results we reported are also
biologically plausible in light of previous findings (e.g. Elwood et al. 2014). All analyses were

3. Results

3.1. Laterization when displaying to a mirror and real opponent.

With a real opponent there was a greater total duration of left side display compared to the right side and a greater median duration of individual left lateral displays but not for the frequency (Table 1). By contrast there was no lateral bias when displaying to a mirror (Table 1).

3.2. Comparison of displays and surface breathing to a mirror and real opponent.

There were no differences between a mirror image and live opponent in the frequencies, median durations and total durations of aggressive displays or frequency of biting (Table 2). However, surface breathing was greater to a mirror in terms of frequency, median duration and total duration (Figure 1, Table 2).

3.3. Correlations between surface breathing and agonistic activities.

When facing a real opponent the frequency of surface breaths was positively related to various indicators of display vigour, in terms of the frequency of bite attempts, left lateral displays, right lateral displays and frontal displays (Table 3). Further, the frequency of breaths was negatively related to both the median duration of left lateral displays and the median duration of frontal displays but did not relate to the total duration of left and right lateral displays or frontal displays (Table 3).

When facing a mirror the frequency of breathing events was positively related only to the frequency of biting events and negatively with the median duration of frontal displays but
3.4. Correlations for display components between mirror images and real opponents

The frequency of bites were positively correlated between the two conditions (Table 4). There was a non-significant tendency for positive correlations between conditions for the frequency of left lateral displays and right lateral displays but not for frontal displays. There were positive correlations between the two conditions for the total duration of left lateral display and for the right lateral display with the median durations of these displays also being positively correlated between conditions. Other measures were not significantly related (Table 4).

4. Discussion

We found evidence of population-level lateralization for lateral displays when facing a real opponent, with a bias to present the left side and use the left eye, in terms of total duration and the median duration of the individual aggressive displays but not for frequency. Preferential use of the left side or left eye during aggressive encounters has been reported for many vertebrates (Vallortigara and Rogers 2005). However, variable results have been reported for different fish species, with some reporting a left side / eye use bias (Sovrano et al. 1999; Ariyomo and Watt 2013) and others reporting a right side bias (Bisazza and de Santi 2003; Arnott et al. 2011; Elwood et al. 2014). Further, the Siamese fighting fish of the present study only showed a significant population bias when facing real opponents and not with a mirror image. Similarly, Ariyomo and Watt (2013) reported greater left-eye preference in male zebrafish when viewing a real opponent compared to a mirror image. However, previous findings in Siamese fighting fish are conflicting, with Bisazza and de Santi (2003) finding a
population-level bias with a mirror, whereas Cantalupo et al. (1996) and Takeuchi et al. (2010) did not. Moreover, Elwood et al. (2014) found evidence of population-level lateralization of convict cichlids in both the mirror and real opponent scenarios. This led to the suggestion that the side-bias was a feature of the individual, rather than supporting a previous suggestion (Arnott et al. 2011) that laterality provides a means for individuals to coordinate their actions and cooperate in information exchange by aligning in a predictable posture. The present findings add to recent and mounting evidence identifying influences of lateralization on animal contest behaviour in a range of species (domestic cows, Phillips et al. 2015; fallow deer, Jennings 2012, 2014a, b; flies, Benelli et al. 2015a, b; Romano et al. 2015; mosquitoes, Benelli et al. 2015c; Przewalski horses, Austin and Rogers 2014).

Previously, we reported distinct differences in the displays of convict cichlids to a mirror compared to a real opponent (Elwood et al. 2014). In particular, the displays of cichlids to a mirror appeared to be slower, with lower frequencies and longer individual displays to the mirror image, explained by the apparent opponent in the mirror never making the first move and thus failing to cause the focal fish to switch display (Elwood et al. 2014). In the present study, however, we found no difference between aggressive displays to mirrors and to real opponents. Further, we had speculated that Siamese fighting fish might respond to a mirror image by escalating to biting more frequently or for longer because the image would not cooperate as might a real opponent, but that was not the case. Thus, Siamese fighting fish appear to display to mirror images in a similar manner to that of real opponents despite the mirror image never making the first move or lining up in a head to tail configuration. It seems that the displays of these fish are organised more in line with the endogenous motivation of the focal fish rather than with the specific actions of the “opponent”. This idea of the endogenous motivation being a major factor in controlling the display actions is supported by
our finding of positive correlations between aggressive activities to mirror and real opponents. Such correlations have also been noted by others (Dore et al. 1978; Balzarini et al. 2014; Elwood et al. 2014). Cichlids also fight mirrors and live rivals according to their own endogenous motivation under normal conditions, but switch tactics to fight according to opponent size when injected with isotocin (Reddon et al. 2012).

Despite the lack of differences in aggressive displays between the mirror and real opponent conditions, there were distinct differences in surface breathing. Fish observing a mirror image went to the surface substantially more often and each breathing event was of longer duration resulting in a greater total duration spent surface breathing compared to when confronting a real opponent.

Surface breathing is a means of getting additional oxygen that cannot be supplied in the normal manner from the water via the gills. The number of breaths taken has been shown here to be related to the vigour of displays in terms of positive relations with frequency, particularly the biting frequency (see also Alton et al. 2013; Regan et al. 2015), and negatively with the median duration, so we agree that breathing is typically affected, at least in part, by oxygen requirement. However, we found no difference in the frequency or vigour of displays between the mirror and real opponents and thus display vigour cannot account for the marked difference in surface breathing frequency between these situations. The duration of each surface breathing event was also greater to a mirror than to a real opponent. However, when we examined correlations between median duration of breathing and other behavioural measures no significant relations were found. Note that a longer time at the surface might not necessarily mean that more oxygen is taken up at each visit. One possibility allowing for differences in metabolic demand is that mirrors might induce a higher degree of fear (Desjardins and Ferdinand 2010) and that fear might increase metabolic demand. However,
this hypothesis requires experimental investigation.

If the increased breathing when confronted by a mirror is not due to a higher oxygen requirement, it might be due to a lower motivation to display to the mirror. That is the mirror image might provide an inappropriate, lower value stimulus for agonistic behaviour, which then allows for the behaviour to be interrupted more frequently and for longer by a subordinate activity (*sensu* McFarland and Sibly 1975; Halliday and Sweatman 1976).

Moreover, other researchers have previously suggested that air breathing can take the form of a `displacement’ activity (Dore et al. 1978). Fight motivation might be reduced in the mirror treatment but not result in reduced contest vigour compared to when facing a real opponent because animals should not disclose future intentions in contests (Dawkins and Krebs 1978).

One way to clearly establish if this is the case would be to probe fight motivation using a novel startling stimulus during the interaction. This causes an animal to temporarily break off from the fight, with the time taken to resume the contest providing a measure of fight motivation, with the technique having now been successfully used for a number of species (e.g. hermit crabs; Elwood et al. 1998; fish, Arnott et al. 2009b, c, 2010; cuttlefish, Schnell et al. 2015).

Another explanation for the increased frequency and duration of surface breathing events in the mirror treatment is that breathing of the opponents under normal circumstances is typically coordinated, with the fish engaging in near simultaneous air breathing (Simpson 1968). The mirror image cannot be the first to restart display, as might happen with a real opponent causing the focal fish to respond. This might account for the increased duration of surface breathing bouts in the mirror treatment. However, it is more difficult to use a similar argument for the increased frequency of breathing. The mirror image cannot be the first to initiate breathing and thus fewer breathing events should occur, the opposite to our findings.
Conversely, in real contests should a focal animal show intention of going to the surface this might be inhibited because the opponent continues to display. A mirror image will not continue to display and thus might enable more frequent breathing.

The idea that the focal animal should attempt to match the display behaviour of the opponent is predicted by some contest theory models (e.g. ‘war of attrition without assessment, Mesterton-Gibbons et al. 1996; ‘energetic war of attrition’, Payne and Pagel 1996, 1997) such that surface breathing is constrained to times of absolute need by both opponents. Indeed, it is possible that with real opponents each attempts to prevent the other from taking sufficient breaths and thus inflicts costs in terms of anaerobic respiration and build-up of lactate. However, anaerobic capacity appears not to affect display vigour whereas enhanced capacity for aerobic metabolism is linked to vigour and outcome (Regan et al. 2015). The idea of opponents influencing each other’s surface breathing is further supported by breathing being better correlated with fight behaviour when facing the real opponent compared to the mirror treatment.

5. Conclusions

The lateralization of displays showed a left side bias but only when facing a real opponent and not with a mirror image. Despite this difference in lateralization, fish did not differ in their levels of aggressive displays to mirror images and real opponents. They did, however, differ in surface breathing, with far more and longer breathing acts when facing a mirror image. This could not be due to increased oxygen requirement when facing a mirror and the increased frequency may be explained by a lower motivation to display to a mirror. The increased duration might be due to the mirror image never being the first to resume displaying and thus the image does not induce the focal fish to stop breathing and return to aggressive
display. This study adds to mounting evidence of differences in behaviour when fish face a mirror image compared to real opponent beyond a transparent barrier, therefore questioning the utility of using mirrors. We accept, however, that even a live fish beyond a barrier is not the same as interactions between unrestrained opponents.

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Figure captions

Figure 1. Comparison of the frequency (a), total duration (b) and median duration (c) of surface breathing when the focal fish (n=20) displayed to a real opponent or mirror image.
Table 1. Summary of results from Wilcoxon matched-pairs signed ranks tests, examining lateralization of lateral displays to a mirror image and real opponent (n=20 focal fish). Median values presented, and durations presented in seconds. Significant P values (P<0.05) are in bold.

<table>
<thead>
<tr>
<th>Display measure</th>
<th>Left side</th>
<th>Right side</th>
<th>Z statistic</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Real opponent</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total duration</td>
<td>198.55</td>
<td>184.35</td>
<td>-2.43</td>
<td><strong>0.015</strong></td>
</tr>
<tr>
<td>Median duration</td>
<td>3.69</td>
<td>3.01</td>
<td>-2.60</td>
<td><strong>0.010</strong></td>
</tr>
<tr>
<td>Frequency</td>
<td>62.50</td>
<td>61.50</td>
<td>-0.02</td>
<td>0.983</td>
</tr>
<tr>
<td><strong>Mirror image</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total duration</td>
<td>235.10</td>
<td>220.35</td>
<td>1.57</td>
<td>0.117</td>
</tr>
<tr>
<td>Median duration</td>
<td>3.31</td>
<td>3.19</td>
<td>-0.49</td>
<td>0.627</td>
</tr>
<tr>
<td>Frequency</td>
<td>67.50</td>
<td>60.00</td>
<td>-1.46</td>
<td>0.144</td>
</tr>
</tbody>
</table>
Table 2. Summary of results from Wilcoxon matched-pairs signed ranks tests, comparing aggressive displays and surface breathing to a mirror and real opponent (n=20 focal fish). Median values presented, and durations presented in seconds. Significant P values (P<0.05) are in bold.

<table>
<thead>
<tr>
<th>Display measure</th>
<th>Mirror image</th>
<th>Real opponent</th>
<th>Z statistic</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Frequency</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Left lateral</td>
<td>67.50</td>
<td>62.50</td>
<td>-1.35</td>
<td>0.179</td>
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<tr>
<td>Right lateral</td>
<td>60.00</td>
<td>61.50</td>
<td>-0.50</td>
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<td>Frontal</td>
<td>91.50</td>
<td>88.00</td>
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<td>0.624</td>
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<tr>
<td>Bites</td>
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<td>2.50</td>
<td>-1.07</td>
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<tr>
<td>Surface breaths</td>
<td>18.00</td>
<td>10.50</td>
<td>-3.14</td>
<td><strong>0.002</strong></td>
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<tr>
<td><strong>Total duration</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<td>Left lateral</td>
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<td>Right lateral</td>
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<td><strong>Median duration</strong></td>
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<tr>
<td>Left lateral</td>
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<td>Right lateral</td>
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<td>Frontal</td>
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<td>3.77</td>
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<td>0.332</td>
</tr>
<tr>
<td>Surface breaths</td>
<td>2.29</td>
<td>2.07</td>
<td>-2.093</td>
<td><strong>0.036</strong></td>
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</tbody>
</table>
Table 3. Spearman Rank correlations are shown for the frequency of surface breaths and
display components to a real opponent and a mirror image (n=20 focal fish). Correlations for
the median duration of breathing events and other display components are also shown for the
two conditions. Significant P values (P<0.05) are in bold.

<table>
<thead>
<tr>
<th>Display measure</th>
<th>Frequency of surface breaths</th>
<th>Median duration of surface breaths</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Real opponent</td>
<td>Mirror image</td>
</tr>
<tr>
<td></td>
<td>$r_s$</td>
<td>$P$</td>
</tr>
<tr>
<td>Frequency</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bites</td>
<td>0.556</td>
<td>0.018</td>
</tr>
<tr>
<td>Left lateral</td>
<td>0.706</td>
<td>0.002</td>
</tr>
<tr>
<td>Right lateral</td>
<td>0.874</td>
<td>0.003</td>
</tr>
<tr>
<td>Frontal</td>
<td>0.648</td>
<td>0.005</td>
</tr>
<tr>
<td>Total duration</td>
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<td></td>
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<tr>
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<td>0.56</td>
</tr>
<tr>
<td>Right lateral</td>
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<td>0.229</td>
</tr>
<tr>
<td>Frontal</td>
<td>0.143</td>
<td>0.54</td>
</tr>
<tr>
<td>Median duration</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Left lateral</td>
<td>-0.531</td>
<td>0.019</td>
</tr>
<tr>
<td>Right lateral</td>
<td>-0.247</td>
<td>0.273</td>
</tr>
<tr>
<td>Frontal</td>
<td>-0.502</td>
<td>0.027</td>
</tr>
</tbody>
</table>
Table 4. Correlations between specific display components to a mirror and those to real opponents (n=20 focal fish). Significant P values (P<0.05) are in bold.

<table>
<thead>
<tr>
<th>Display measure</th>
<th>Spearman $r_s$</th>
<th>$P$ value</th>
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<tbody>
<tr>
<td><strong>Frequency</strong></td>
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<tr>
<td>Bites</td>
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<td><strong>0.003</strong></td>
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<td>Left lateral</td>
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<tr>
<td>Right lateral</td>
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<td>0.075</td>
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<tr>
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<td>0.290</td>
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<tr>
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<td>0.10</td>
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<tr>
<td><strong>Total duration</strong></td>
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<td></td>
</tr>
<tr>
<td>Left lateral</td>
<td>0.507</td>
<td><strong>0.027</strong></td>
</tr>
<tr>
<td>Right lateral</td>
<td>0.531</td>
<td><strong>0.021</strong></td>
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<tr>
<td>Frontal</td>
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<td>0.084</td>
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<tr>
<td><strong>Median duration</strong></td>
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<td>Left lateral</td>
<td>0.605</td>
<td><strong>0.008</strong></td>
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<tr>
<td>Right lateral</td>
<td>0.770</td>
<td><strong>0.001</strong></td>
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<tr>
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<td>0.820</td>
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<tr>
<td>Surface breaths</td>
<td>0.170</td>
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</tbody>
</table>
Figure 1.
Real opponent vs. Mirror image

Median duration (s) of surface breaths