BRIEF COMMUNICATIONS

No hormonal response in tied fights

Fish androgens may start to surge only when there seems to be a good chance of winning a contest.

Duel action: a cichlid attacks its reflection.

Androgens are the principal sex steroids controlling reproduction and aggression in male fish[1], but their production can also be affected by social interactions[2,3]. Here we show that androgen concentrations are not significantly increased in cichlid fish (*Oreochromis mossambicus*) that are fighting their own image in a mirror, despite their aggressive behaviour towards the virtual intruder. Our results indicate that the hormonal response normally triggered in male contests is not induced under these circumstances by the act of fighting itself, and that it may therefore depend on some indicator of relative fighting ability that cannot be delivered by a mirror-image challenger.

Fish do not recognize their own image in a mirror and so attack it as though it were an intruder[4]. However, there are no pointers (such as differences in rank, strength, injury or fatigue) that can be used to assess the outcome of the attack (winning versus losing) for a fish in this situation. To test whether information on interaction outcome is necessary to trigger an androgen response in fighting fish, we compared androgen concentrations in the urine of fish after mirror-mediated challenge with those in urine from non-fighting controls.

Male cichlids of comparable size and age, raised in similar conditions, were kept in social isolation for seven days before the experiment to minimize inter-individual variation in behavioural and androgen responses that arise from differences in social status[5]. On the day of the experiment, the fish were divided into two groups: one was presented with a mirror at one end of the tank (experimental group; $n = 17$); the other had a sheet of transparent glass instead to control for the presence of a novel object in the aquarium (control group; $n = 14$). We recorded the fighting behaviour of the fish (aggressive displays and attacks) for 20 min after the first interaction with the mirror or glass by using a focal continuous recording method[6]. The principal fish androgens[1], testosterone and 11-keto-testosterone, were measured by specific radioimmunoassay of urine collected 60 min before the test (baseline value) and 30 min, 2 h and 6 h after the end of the test.

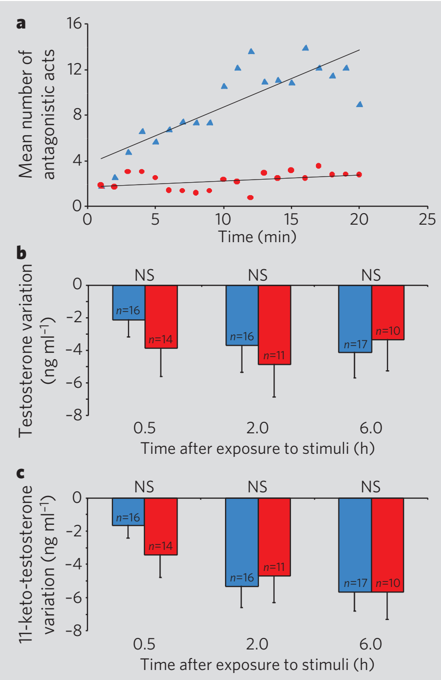

Figure 1 | Mirror-elicited aggression in cichlid male fish fails to trigger an androgen response. a, Escalation in antagonism by fish towards their own image in a mirror, as measured by variation in the frequency of aggressive displays and attacks on the mirror during the period of the trial. Red circles, controls (clear glass in the tank instead of a mirror), $n = 14$; blue triangles, experimental group, $n = 17$. The difference between the two regression lines was tested with analysis of covariance using BIOM software (copyright F. James Rohlf, 1984); slopes: $F = 29.97$, $P < 0.001$; means: $F = 110.26$, $P < 0.001$. **b, c,** Variation in fishes' total urinary concentrations (free + sulphate + glucuronide) of testosterone (**b**) and 11-keto-testosterone (**c**) after exposure to a mirror (blue bars) or to clear glass (red bars) for 20 min (calculated as the difference in concentration before and after stimulation; error bars, s.e.m.). Urine was collected in an Eppendorf tube by applying gentle pressure in the ventro-posterior flank of the fish. Sample sizes differ because urine was not obtained from all individuals at all sampling points. Sample processing and radioimmunoassay were done as described[5,9,10]. NS, not significant in Mann–Whitney tests.

Individuals reacted very aggressively to their own image in the mirror, whereas the presence of non-reflective glass in the tank did not provoke a combative response in fish from the control group. During the course of the attacks, the aggression of subjects in the experimental group escalated towards their mirror images as they apparently tried to resolve the 'contest' (Fig. 1a).

However, despite the marked degree of aggressive behaviour, no significant increase in urine androgens was detected in the experimental relative to the control group (Fig. 1b, c). This contrasts with the increased production of androgens in *O. mossambicus* males in response to territorial intrusions by conspecific males[7], and with the androgen increase in male spectators that are not even directly participating in a fight[8]. Although there is a reduction in androgen synthesis in fish that lose fights and become subordinates[5], this cannot account for the drop in androgens in the experimental group, which is similar to that in the non-fighting control group and is in the range expected for the time of day (morning) at which the measurements were made.

We conclude that information on the likely outcome of a contest is required before the androgen response is triggered in a combatant, and that escalation of the fight occurs independently of circulating androgen levels. This may be an adaptive mechanism that allows individuals to mount an androgen response for the purpose of controlling their social status after they have assessed the relative fighting ability of their challenger.

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DYNAMICAL PHENOMENA

Walking and orbiting droplets

Small drops can bounce indefinitely on a bath of the same liquid if the container is oscillated vertically at a sufficiently high acceleration[1]. Here we show that bouncing droplets can be made to 'walk' at constant horizontal velocity on the liquid surface by increasing this acceleration. This transition yields a new type of localized state[2–5] with particle–wave duality: surface capillary waves emanate from a bouncing drop, which self-propels by interaction with its own wave and becomes a walker. When two walkers come close, they interact through their waves and this 'collision' may cause the two walkers to orbit around each other[6–8].

The bouncer transition to walking is continuous and occurs when the vertical acceleration of the bath, γ_m, reaches a critical threshold, γ_m^c. Below γ_m^c, the drops bounce with no horizontal motion. Above γ_m^c, bouncing drops acquire a rectilinear motion along the surface of the bath (Fig. 1a–c). Their velocity V_w is constant (0–20 mm s^{-1}) and increases with γ_m.

Why do the drops start walking? This phenomenon occurs below, but near, the onset of the Faraday instability, a point at which the surface becomes spontaneously wavy. In this regime, the vertical motion of a drop becomes subharmonic, with a period that is double that of the forcing. As a result, it emits a damped Faraday wave. The drop undergoes successive identical parabolic jumps that are locked with its wave. Each jump brings the drop into collision with the side of the central bulge of the wave generated by the previous collision (Fig. 1a). This collision with an inclined surface generates a non-zero horizontal impulse, which can be translated as an equation for the drop's horizontal motion, averaged over a period π/ω_0 of the subharmonic vertical motion

$$m\,\mathrm{d}^2x/\mathrm{d}t^2 = a\sin\{(\pi k/\omega_0)\,\mathrm{d}x/\mathrm{d}t\} - b\,\mathrm{d}x/\mathrm{d}t \quad (1)$$

where m is the drop's mass, a is about 10^{-6} N, k is the wavenumber, and b is about 10^{-6} N m^{-1} s. The left-hand side of equation (1) represents the inertia of the drop; the first term on the right-hand side accounts for the effective force due to the inclined surface, and the second for viscous damping during the collision. Equation (1) predicts the observed continuous transition of the droplet from stationary to walking when $a > b\omega_0/(\pi k)$.

When walkers coexist in a cell, they inevitably collide. These 'collisions' do not involve any contact between the drops but only a deflection of their horizontal trajectories, when the wave generated by a drop affects the horizontal velocity of the other one. The main parameter characterizing this collision is d_c, the minimal distance of approach of the two drops; depending on the value of d_c, the walkers either attract or repel each other. Attraction leads to a twin-star-like orbiting motion of the drops (Fig. 1d, and see movie in supplementary information). The diameters of the orbits take discrete values d_n^{orb}, which self-adapt to the forcing frequency[9,10]. The orbital diameters are slightly smaller than an integer multiple of the Faraday wavelength (λ_F), or $d_n^{\mathrm{orb}} = (n-\varepsilon)\lambda_F$ when the drops bounce in phase. They are $d_n^{\mathrm{orb}\prime} = (n + 1/2 - \varepsilon)\lambda_F$ when the drops bounce in antiphase; the offset, $\varepsilon = 0.2 \pm 0.02$, is such that when a drop collides with the surface, it falls on the inward slope of the wave emitted by the other. This provides the centripetal force needed for the orbital motion. For other values of d_c, each drop falls on the outward slope of the wave of the other, which causes a repulsion.

We have shown that walkers can behave as billiard balls, undergo scattering collisions or form circular orbits, and can even display complex three-body motion (results not shown). The variety of these phenomena can be explained by interaction through waves and by generalizing equation (1) to two or more drops (the resulting equations yield the same quantification of orbits and numerical trajectories, which are very similar to the experimental collisions; S. P. *et al.*, manuscript in preparation). In this system, real particles experience the same non-local interaction as nonlinear waves.

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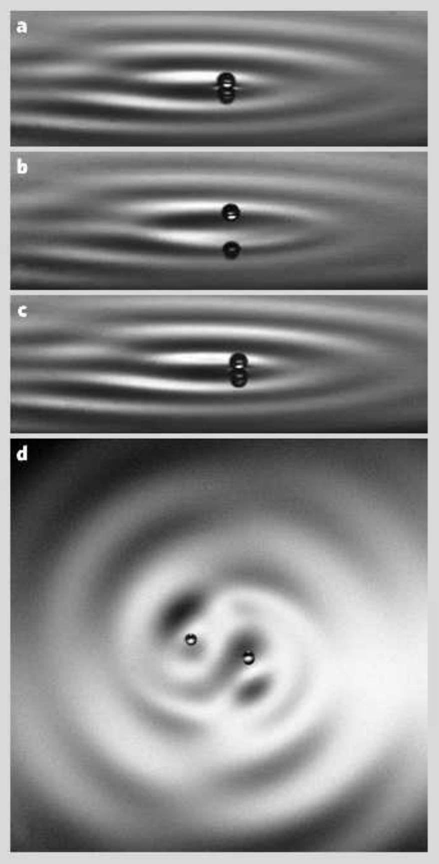

Figure 1 | Behaviour of silicon oil droplets on a bath of silicon oil when it is oscillated vertically. Experimental parameters: oil viscosity, 20×10^{-3} Pa s; forcing frequency, $\omega_0/2\pi = 80$ Hz, diameter of droplets $D\approx0.65$ mm; forcing acceleration, $\gamma_m/g\approx3.9$ (where g is the acceleration due to gravity). **a–c,** Photographs showing the motion of a single drop in interaction with its own localized Faraday wave on the liquid surface. The drop's motion is composed of a series of identical parabolic jumps, each jump bringing the drop into collision with the forward side of the central bulge of the wave generated by the previous collision. **d,** Photograph of two orbiting drops and associated waves. The horizontal motion is in a twin-star-like orbit of diameter $d_n = 5.8$ mm. (For movies, see supplementary information.)

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