Oscillation of the velvet worm slime jet by passive hydrodynamic instability

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The rapid squirt of a proteinaceous slime jet endows velvet worms (Onychophora) with a unique mechanism for defence from predators and for capturing prey by entangling them in a disordered web that immobilizes their target. However, to date, neither qualitative nor quantitative descriptions have been provided for this unique adaptation. Here we investigate the fast oscillatory motion of the oral papillae and the exiting liquid jet that oscillates with frequencies \( f \sim 30-60 \text{ Hz} \). Using anatomical images, high-speed videography, theoretical analysis and a physical simulacrum, we show that this fast oscillatory motion is the result of an elastohydrodynamic instability driven by the interplay between the elasticity of oral papillae and the fast unsteady flow during squirting. Our results demonstrate how passive strategies can be cleverly harnessed by organisms, while suggesting future oscillating microfluidic devices, as well as novel ways for micro and nanofibre production using bioinspired strategies.
Rapid motions in nature are seen in a variety of situations associated with escape and predation. Extreme examples include the chameleon tongue that uses an unusual muscle-spring configuration to capture prey\(^5\), and the Venus flytrap that stores elastic energy and uses an instability for the rapid closure of its leaf\(^6\). The velvet worm is an unusual example of how an organism projects itself by squirting a jet of slime in an oscillatory fashion, not only for capturing prey\(^5\), but also for defence\(^4\). Animals use squirting mechanisms for elimination of waste products (for example, urination in vertebrates); reproduction (gamete expulsion); communication (for example, pheromone excretion in mammals); locomotion (for example, squids); defence (for example, horned lizards); and hunting (for example, archer fish)\(^{10,13}\). These jets are normally directed in a straight line. We are aware of only three exceptions: velvet worms (Phylum Onychophora), spitting spiders (Scytodes spp.) and spitting cobras (Naja spp.). Spitting spiders oscillate their fangs\(^{14}\), and spitting cobras actively oscillate their heads\(^{15}\). Despite having been a subject of study for over a century, the mechanism underlying the rapid oscillatory squirting of slime by the velvet worm remains a mystery\(^{4,16,17}\). Indeed, Darwin even hypothesized the creation of the disordered web as a potential origin for the evolution of spider webs\(^{18}\).

In this article, we demonstrate that the fast oscillations of the jet slime and papillae in Onychophora are the result of a syringe-like system that, by means of a geometric amplifier, allows for fast squirt using slow muscular contraction. This fast flow activates an elastohydrodynamical instability that explains the fast papilla oscillations during defence and attack. We present a physical simulacrum that reproduces the instability in the same range of parameters of the natural system opening new venues for self-supported microfluidic devices and applications\(^{19–22}\).

**Results and Discussion**

**Worm attack kinematics.** To capture the dynamics of the squirting process, we filmed several worm attacks (Supplementary Movies 1–3). In Fig. 1a–d, we show a series of snapshots of an attack recorded using high-speed imaging (480 frames per second a.k.a. f.p.s.), with the average duration of a squirt for all specimens being \(\Delta t_{\text{ave}} = 0.064 \pm 0.005\, s\) (Supplementary Table 1; Supplementary Note 2). By tracking the motion of the tip of the jet shown in Fig. 1 and Supplementary Fig. 2, as a function of time, we found that the typical jet speed \(V \approx 3–5\, m\, s^{-1}\). Furthermore, we see that the squirt does not remain oriented but instead sprays an entire region as shown in Fig. 1e. These measurements raise the natural question of the spatio-temporal evolution of the liquid jet and its control by the worm.

**Anatomy of the ejecting system.** Actively controlled muscular action has long been invoked as the natural explanation for the spectacular way in which these worms quickly squirt slime and continues to be the favoured mechanism\(^{16}\). However, papillar oscillations are fast (Fig. 1; Supplementary Table 1 and Supplementary Note 2) in comparison with any other motion of the worm (\(f_{\text{papilla}}/f_{\text{walking}} \approx 30–60\) and with known time scales (\(\sim 0.5\, s\)) for the fastest muscles in the worm\(^{23}\), suggesting a conceptual difficulty with this hypothesis. Therefore, we examined the anatomy of the whole-squirt system and surrounding tissues (Fig. 2; Supplementary Fig. 1). During squirting, the oral papilla extends from its folded shape to its full length (Fig. 2) of up to \(L \sim 6\, mm\) (Supplementary Fig. 2). In Fig. 2a, we see a large reservoir region (re) where slime is stored and a narrow duct that ends at the oral papilla, a syringe-like geometry that facilitates the acceleration of the slime for the fast squirt. In Fig. 2a,b, we show that muscle fibres in the oral papilla are similar to those found in the legs, but fewer in number. Figure 2a,b and Supplementary Fig. 1 also show that muscular fibres found in papilla tissues are consistent with their directional function, with some being annular, typical of sphincter-like systems. The relaxed papilla has an accordion shape (Fig. 2c,d) that is unfolded just before the squirting process, and can thus be easily packed while also having an inhomogeneous bending rigidity, with soft spots that make papillae more pliable and susceptible to bending as slime is squirted.

Our anatomical findings are consistent with earlier evidence that slime papilla are modified limbs\(^{24,25}\) with a nervous system similar to that in their legs\(^{26}\). Detailed descriptions of the Onychophora muscular system\(^{23}\) show that the fastest muscles are located in the jaw with typical twitch time scales \(\sim 0.5\, s\), which, while fast for this primitive worm, are nearly 25 times slower relative to the papillary oscillation time scale \(\sim 20\, ms\). Given that the legs and the papillary muscles are even slower, consistent with the primitive nature of these worms\(^{5,27,28}\), we are left with an obvious question—how are rapid changes in direction that occur over a time scale of a few milliseconds possible without the existence of any specialized rapid muscular actuation or neural control?

**Physical mechanism.** A way around this conundrum is to realize that a physical mechanism can drive the rapid and nearly chaotic oscillations of the papilla just as a garden hose pipe oscillates when water squirts out of it rapidly. Indeed, the inertial effects associated with the exiting fluid jet drive the elastic hose pipe to flutter, a subject that has been well studied experimentally and
Peripatus solorzanoi present in at 15 kV after coating with gold (of a spitting spiders14, and suggests that this geometric amplifier can lead to an increase in the speed during squirting. This is papilla (Supplementary Figs 1 and 3). This geometric amplifier system has a reservoir that contracts slowly driving the slime inertial effects are unimportant. typical sizes are small, unless the velocities are sufficiently large, viscosity and forces characterized by the Reynolds number Re is the characteristic fluid speed, v the fluid kinematic viscosity and R the tube radius. In microfluidic geometries where typical sizes are small, unless the velocities are sufficiently large, inertial effects are unimportant.

Our microscopy studies (see Fig. 2a) show that the squirting system has a reservoir that contracts slowly driving the slime through a small duct that runs close to the centre of the oral papilla (Supplementary Figs 1 and 3). This geometric amplifier can lead to an increase in the speed during squirting. This is functionally similar to structures found in the chelicerae of spitting spiders14, and suggests that this geometric amplifier mechanism could be relevant for a variety of other squirting organisms. Our observations are in contrast with previously reported studies30 that have persisted into the modern literature,31 where no mention of the cross-section reduction has been made. Our micrographs show muscular structures (Fig. 2; Supplementary Fig. 1) around the slime reservoirs, which are functionally consistent with the contraction of this organ. These structures resemble the design of radial tires where a fibre network is used to reinforce the wall32,33, consistent with detailed study of muscular fibres at reservoir level33. Measurements of the squirted volume (Supplementary Notes 1 and 3; Supplementary Fig. 4) and reservoir geometry show that the contraction ratio \( \delta R_{res}/R_{ro} \) is less than 0.03 (with \( R_{ro} \sim 2 \text{ mm} \)) changes in about 0.1 s, enough to produce speeds of \( V \sim 5 \text{ m s}^{-1} \), so that the Reynolds number Re \( \sim 2,700 \) (Supplementary Note 6). Since we do not see perfect synchronization between liquid jets coming from different papilla (Fig. 1a–d; Supplementary Table 1; Supplementary Movies 1 and 2), whole-body contraction as the main driving force in squirting23 is unlikely.

This leads to the conclusion that the instability arises due to a competition between fluid inertia and elastic resistance. When a liquid moves steadily through a flexible pipe at small \( V \), flow-induced damping prevents any oscillations from growing. For large enough \( V \), centrifugal and Coriolis forces make the pipe unstable for fluid speeds \( V > V_c \). In the limit when the effects of gravity can be neglected (Supplementary Notes 5–7; Supplementary Fig. 8), a simple scaling argument allows us to estimate the frequency of oscillations \( \omega \) by balancing the stabilizing elastic bending resistance with the destabilizing inertial forces, that is,

\[
\frac{EI}{\lambda^4} \sim \frac{MVf}{\lambda} \sim \frac{MV^2}{\lambda^2},
\]

where \( EI \) is the bending stiffness, \( \lambda \sim 2L \) is the approximate oscillation wavelength for the cantilevered papilla and \( M \) is the mass density per unit length of the fluid in the pipe. This yields

\[
\omega \sim \left( \frac{EI}{M} \right)^{1/2} \frac{1}{4L^2}
\]

Similarly, a typical speed scale can be estimated as \( u_0 \sim \omega \frac{\lambda}{\lambda^2} = (EI/M)^{1/2} (1/L) \). For the specimen shown in Supplementary Fig. 2, the oscillation frequency \( \omega \sim 58 \text{ Hz} \), papilla length \( L = 6.0 \text{ mm} \), papilla outer diameter \( D = 1.0 \text{ mm} \) and papilla inner diameter \( d = 0.5 \text{ mm} \), from where \( \beta \sim 0.25 \). The measured frequency allows to estimate the effective Young’s modulus of the papilla to be \( E \sim 20 \text{ kPa} \) consistent with measurements using small magnets to pull the papilla that yields \( E \sim 40 \text{ kPa} \) (Supplementary Table 2), and hence the typical speed \( u_0 \sim 0.5 \text{ m s}^{-1} \). The precise critical speed \( V_c \) depends on boundary conditions, which for the cantilever case gives \( V_c \sim 2\pi u_0 = 3 \text{ m s}^{-1} \). At this critical speed, stability is lost via a Hopf type bifurcation29 (see Supplementary Note 5, 6 for further information).
For unsteady flows, such as when the jet is being accelerated inside the flexible papilla, the fluid acceleration $\frac{dv}{dt}$ can destabilize the system at even lower jet speeds (see Eqs. 3, 4 in Methods and Supplementary Fig. 8). Our measurements show that $V \approx 3.2$–$5.0$ m s$^{-1}$. Therefore, even without muscular action the papilla will become unstable due to a simple physical instability.

**Synthetic simulacrum.** To show that it is indeed possible to drive these oscillatory motions on small scales, we made a synthetic papilla out of a soft elastomer (see Methods) in the form of a flexible micropipe with a rectangular cross-section that defines an oscillation axis for its softest bending mode. Our pipe was moulded out of polydimethylsiloxane (PDMS) with a Young's modulus $E = 288$ kPa (Supplementary Figs 5–7; Supplementary Note 4), with thickness $h = 1.42$ mm, width $w = 1.60$ mm, inner diameter $d = 0.81$ mm and length $L = 9.5$ mm. Our model system is simpler than the natural one in at least two aspects: there is no roughness along the inner part of the duct or an external accordion-like geometry. Our experimental results (Fig. 3; Supplementary Fig. 5; Supplementary Note 5; Supplementary Movie 4) show that synthetic papilla becomes unstable and oscillates when the liquid reaches a speed of $V_c = 8.6$ m s$^{-1}$. This occurs in the same range of dimensionless parameters that for the natural organ in agreement with theoretical predictions and shows that fast muscular action at the papillary level is unnecessary for oscillations. From measurements of external diameters (Fig. 2d), we found that $B = EI$ can locally change up to 1/10 of the stiffness corresponding to a uniform soft papilla of diameter $D_0$, showing that the accordion-like microstructure will lower $V_c$. Microtubes with accordion-like shapes made out of the same polymer were made varying the degree of external roughness. Our data shows that $V_c$ lowers as we increase the amplitude of external roughness (Supplementary Note 6; Supplementary Fig. 9). The flow of liquids in pipes with inner roughness has been extensively studied. The effect of inner pipe imperfections is to decrease the effective cross-section of the tube, as well as to anticipate the transition to turbulence. All these effects together lower $V_c$, being the most important for the fluid regime described, the accordion-like modulations of the tube. Naturally, this experiment also suggests a prototype of a fluid driven micromechanical actuator.

**Methods**

**Sample collection.** Specimens of *Peripatus solorzanoi* ($n = 3$) were collected in Guayacan, Siquirres, Costa Rica (10°3′21.38″ N, 83°32′44.04″ W, 500 m a.s.l.). The specimen of *Epperipatus acacioi* ($n = 1$) was collected in Minas Gerais, Brazil (20°22′44″ S, 43°32′55″ W, 1220 m a.s.l.). The worms were housed in plastic polystyrene (PS) terraria (370 × 240 × 280 mm) with a window covered with a metallic net as air vent. The temperature varied between 18 and 23 °C. The terraria received artificial illumination with standard daylight fluorescent lamps and had a 3–4 cm layer of leaf litter extracted from the original habitat of the
The boundary conditions in this problem are \( z(0,t) = 0, z'(0,t) = 0, z(L,t) = 0 \) and \( z''(L,t) = 0 \). The relevant physical parameters needed to specify the parameter space for this problem can be found in equation (3) in a dimensionless form. This is easily achieved by using the dimensionless time and coordinates \( \eta = x/L, \xi = x/L, \mu = \nu t/L, \quad \tau = \nu t/L \). Where \( \nu t \) is the bending scale, and \( \nu u \) is a characteristic speed. The dimensionless equation reads:

\[
\begin{align*}
\frac{\partial^2 u}{\partial \xi^2} + \left[ u \left( \frac{1}{2} u' + (d - \xi)(1 - \xi) \right) \frac{\partial^2 u}{\partial \eta^2} \right] \\
+ 2\nu \frac{\partial u}{\partial \eta} - \frac{\partial^2 u}{\partial \xi^2} = 0
\end{align*}
\]  

(4)

**Numerical simulation.** We have solved equation (4) using different numerical approaches. We did compare different methods as the coefficients of the PDE are not constant and may abruptly change as a function of time. The results we show were obtained using the Garlekin method. We use as basis the set of functions provided by the problem:

\[
\Psi = 0
\]

(5)

with the boundary conditions (BC) \( \psi(0) = 0, \psi'(0) = 0, \psi'(1) = 0 \) and \( \psi'(3) = 0 \).

Where:

\[
\sigma = \frac{\partial \Psi}{\partial \xi} + \frac{\partial^2 \Psi}{\partial \eta^2}
\]

(6)

This problem endows us with a good expansion basis that fulfils the BC. After using the anzats,

\[
\eta_{\text{max}}(\xi, \tau) = \max_{j=1}^{\text{max}} \Psi_{j}(\xi)\Psi_{j}(\tau)
\]

and using the inner product properties of \( \langle \psi_j(\xi) \rangle_{\text{max}} \) over the domain \( [0,1] \), we reduced equation (4) into a linear system using the first \( n_{\text{max}} \) basis functions. We emphasize that equation (4) captures well the onset of the instability. However, the oscillation amplitude does not saturate without including nonlinear terms in curvature\(^2\). To keep the model simple, we augmented equation (4) by a confining term \( \eta_{\text{lim}} \), and included the experimentally found damping \( s \) (Supplementary Fig. 8).

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