Mate competition during pseudocopulation in shipworms

Authors & Affiliations

J. Reuben Shipway[1,2*], Nancy C. Treneman[3] & Daniel L. Distel[4]

[1]Institute of Marine Sciences, University of Portsmouth, Portsmouth, UK

[2]Department of Microbiology, University of Massachusetts, Amherst, 01003, USA

[3]Oregon Institute of Marine Biology, PO Box 5389, Charleston, OR, 97420, USA

[4]Ocean Genome Legacy, Marine Science Center, Northeastern University, Nahant, MA, 02151, USA

E-mail: reuben.shipway@port.ac.uk

*Corresponding author

Abstract

Shipworms are predominantly wood-eating bivalves that play fundamental roles in biodegradation, niche creation and nutrient cycling across a range of marine ecosystems. Shipworms remain confined to the wood they colonise as larvae; however, continual feeding and rapid growth to large sizes degrades both food source and habitat. This unique lifestyle has led to the evolution of a stunning diversity of reproductive strategies, from broadcast spawning to spermcasting, larval brooding, and extreme sexual size dimorphism with male dwarfism. Some species also engage in pseudocopulation, a form of direct fertilization where groups of neighbouring individuals simultaneously inseminate one another via their siphons - the only part of the animal extending beyond the burrow. Among the Bivalvia, this exceptionally rare behaviour is unique to shipworms and remains infrequently observed and poorly understood. Herein, we document pseudocopulation with video footage in the giant feathery shipworm (*Bankia setacea*), and novel competitive behaviours, including siphon wrestling, mate guarding, and the removal of a rival's spermatozoa from the siphons of a recipient. As successful sperm transfer is likely greater for larger individuals with longer siphons, we suggest that these competitive behaviours are a factor selecting for rapid growth and large size in species that engage in pseudocopulation.

Key Words: Teredinidae; Wood-Boring Bivalves; Reproduction; Mate Competition, Sperm-Transfer; Ephemeral Niches

Introduction

Most sessile marine invertebrates are broadcast spawners; fertilization success is predominantly determined by proximity to other reproductive individuals and success diminishes with increased distance due to sperm dilution (Levitan & McGovern, 2005). To enhance fertilization success, marine invertebrates deploy a variety of sperm delivery strategies, including sperm packing into clumps or bags, synchronous spawning in large groups, and pseudocopulation with direct sperm transfer (Levitan, 1998). For example, barnacles use excessively long penises to deposit sperm inside their neighbour's shell (Hoch et al. 2016). Direct sperm transfer is rare among the Bivalvia (Huber, 2015) and only one family of predominantly wood-boring clams, commonly known to as shipworms, are reported to engage in this reproductive behaviour.

Shipworms (Teredinidae) are the principal degraders of wood in marine ecosystems and play a fundamental role in nutrient cycling, biodegradation and niche creation (Cragg *et al.,* 2015). Several adaptations facilitate their xlyotrepetic (wood-boring) and xyloptrophic (wood-eating) lifestyle, including; highly adapted valves lined with rows of drilling teeth; a long, vermiform body and a calcareous tube which lines the inside of the burrow offering further protection to the animal; siphons (incurrent and excurrent) which extend beyond the burrow providing the only contact to the external environment; and, calcareous pallets - paddle-like structures which seal the burrow entrance upon retraction of the siphons (Voight, 2015).

Adult shipworms spend their entire lives burrowing into and feeding on the wood they colonised as larvae. Continual growth drives degradation and fragmentation of this habitat, leading to an existence that is both unpredictable and ephemeral (MacIntosh *et al*., 2014). This, coupled with the sporadic distribution of wood in marine environments, has selected for multiple adaptations maximising survival. These include rapid development to reproductive maturity (Calloway & Turner, 1983) at sizes as small as 2mm (MacIntosh *et al*., 2014), potential growth to extremely large sizes (1.8 m, Muller & Lana, 1986), and prolific reproductive output, with some broadcast spawning species releasing over 100 million eggs in a single spawning (Sigerfoos, 1908).

Shipworms have evolved a complex range of life history and reproductive strategies, which are among the most diverse within the Bivalvia (Shipway *et al*., 2016). Newly settled larvae first develop as males before transitioning to females (protandry). But, shipworms can also exhibit simultaneous, consecutive and rhythmical-consecutive hermaphroditism (Nair & Saraswathy, 1971), with rapid transition between sexual phases (Coe, 1936). Broadcast spawning is the most common reproductive strategy in this family, although some species internally fertilize through spermcast mating, and retain larvae to either D-stage veligers or pediveligers (Turner, 1966). The seagrass inhabiting *Zachsia* *zenkewitschia* also internally fertilizes through spermcast mating and retains D-stage veliger larvae, but sperm is provided by a harem of dwarf males inhabiting a specialised pouch on the female mantle next to the siphons (Shipway *et al*., 2016).

Perhaps the most unusual reproductive strategy in this family involves pseudocopulation, where the excurrent siphon from one or more individual penetrates the incurrent siphon of its neighbour, with a direct transfer of spermatozoa resulting in internal fertilization (Rhoades, 1951; Turner, 1966; Hiroki *et al.,* 1994). This behaviour has been reported in four species - *Bankia gouldi* (Rhoades, 1951), *B. setacea* (Townsley *et al.,* 1966), *B. martensi* (Velasquez *et al.,* 2011) and *Nausitora fusticula* (Hiroki *et al.,* 1994) - which typically broadcast spawn gametes but may pseudocopulate in populations with high settlement density where individuals are in close proximity. Among the Bivalvia, this rare form of direct fertilization is unique to shipworms, yet remains poorly documented and rarely observed (Huber, 2015).

Herein, we document pseudocopulation in the giant feathery shipworm (*Bankia setacea*), a species endemic to the northern Pacific Ocean, providing the first video footage of this form of reproduction. Further, we observe novel behaviours during pseudocopulation that are suggestive of rival mate competition, and highlight several key questions required to address gaps in our understanding of this unique behaviour in the wild.

Methodology

Pine panels (25 cm x 18.5 cm x 2 cm) were deployed on 28/11/2016 in Charleston, (Oregon, USA, 44.345576 - 124.322706) at a depth of 1 m and retrieved on 23/5/2017. Panels containing live shipworms were then maintained in the aquarium at 12 °C with a salinity of 30 PSU. Pseudocopulation and spawning were induced during water changes that elevate the temperature of seawater above 14 °C. Photographs and video footage of pseudocopulation and associated behaviours were recorded using the GoPro Hero 3+. Siphon length and distance between boreholes were measured with ImageJ, using the 2 cm wood panel width for calibration. Specimens were extracted for species identification following the key provided by Turner (1971).

Results

Specimens were identified as the giant feathery shipworm (Fig. 1A), *Bankia setacea*, based on pallet morphology. A panel containing 79 individuals was maintained in the aquarium and 2.75 hours of footage of pseudocopulation was recorded. The average length of extended excurrent siphons measured 44 mm (ranging from 19 to 69 mm, n=27). The distance between copulating individuals ranged from 4 - 49 mm with an average of 21 mm (n=30). Whilst most individuals engaged in pseudocopulation (n=74), a small number (n=5) were isolated and did not pseudocopulate; their closest neighbour being beyond the range of their extended siphons. These individuals were located an average distance of 31 mm from their closest neighbour and had an average excurrent siphon length of 9 mm. Isolated individuals spawned gametes into the water column simultaneously when pseudocopulation occurred between those capable of it.

Five distinct stages of pseudocopulation were identified. First, the inhalant siphons of receptive individuals become inactive and flare open (Fig. 1B), and the sensory papillae stand erect along the entire length of the excurrent siphons of donor individuals (Fig. 1C). In the second stage the excurrent siphon probes across the surface of the wood until it encounters receptive incurrent siphons of neighbouring individuals. Once located, the excurrent siphon entwines the incurrent siphon (Fig. 1D). Then, the excurrent siphon penetrates the incurrent siphon (Fig. 1E). Lastly, spermatozoa are transferred to the recipient siphon. Up to six individuals were recorded pseudocopulating with one another and group pseudocopulation was typically observed in regions with the densest settlement (Fig. 1F). Individuals were observed simultaneously giving and receiving spermatozoa, with the excurrent siphon penetrating a neighbour’s incurrent siphon, whilst its own inhalant siphon is penetrated by another individual. After pseudocopulation, eggs (presumably fertilized) were spawned into the water column by the recipient shipworm (Fig. 1G). On a small number of occasions, individuals were seen rejecting spermatozoa and expelling it from their incurrent siphon.

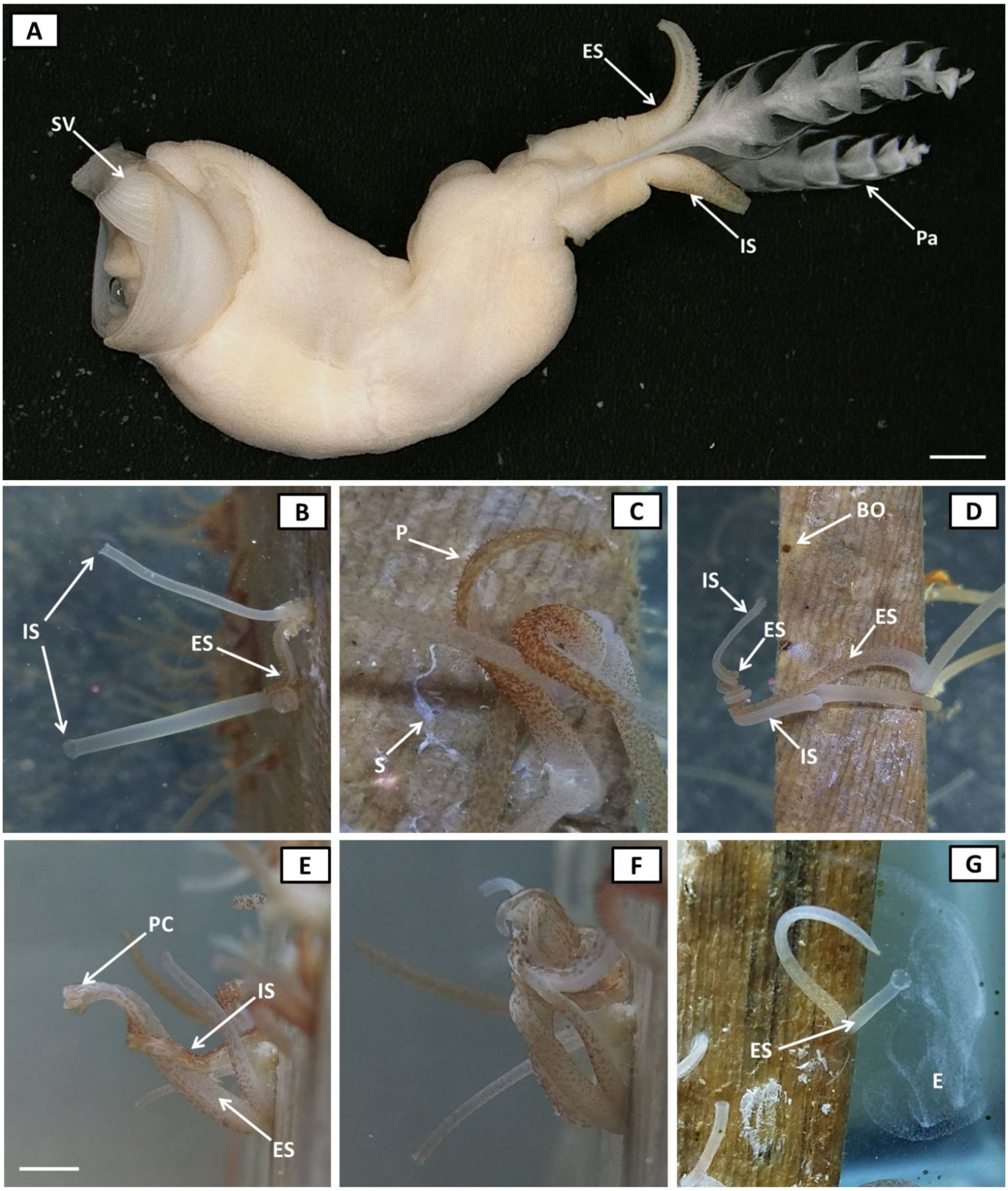

Figure 1. Pseudocopulation in the giant feathery shipworm: A) specimen removed from wood; B) flaring of the incurrent siphonal opening; C) sensory papillae on excurrent siphons become erect; D) an excurrent siphon entwines the incurrent siphon of neighbouring individual; E) an excurrent siphon penetrates the incurrent siphon of a neighbour and transfers spermatozoa; F) multiple individuals simultaneously display pseudocopulatory behaviour; G) eggs are spawned into the water column. BO, burrow opening; E, eggs; ES, excurrent siphon; PC, pseudocopulation; IS, incurrent siphon; P, papillae; Pa, pallet; S, sperm; SV shell valve . Scale bars = 1 mm (a) and 1 cm (b-g).

A number of previously unreported competitive mating behaviours were observed in smaller groups of three or four individuals. Excurrent siphons from neighbouring individuals wrestled (Fig. 2A), pushed rival excurrent siphon away (Fig. 2B), or pulled receptive incurrent siphons beyond the range of a competitors' excurrent siphon (Fig. 2C). The 'winning' individual then inserted its excurrent siphon into the incurrent siphon of its quiescent neighbour and transferred spermatozoa (Fig. 2D). Fig. 2E documents a case of unsuccessful pseudocopulation, where the rival failed to penetrate the incurrent siphon sufficiently, leaving its spermatozoa covering the surface of the recipient's incurrent siphon (Fig. 2E). Immediately afterwards, the donor removed its rivals spermatozoa from the recipients siphon with its excurrent siphon (Fig. 2F). All behaviours outlined in Figures 1 and 2 are shown in the video provided in Supplemental Figure 1 (https://figshare.com/s/46dd983f1d0e09c8d8c4).

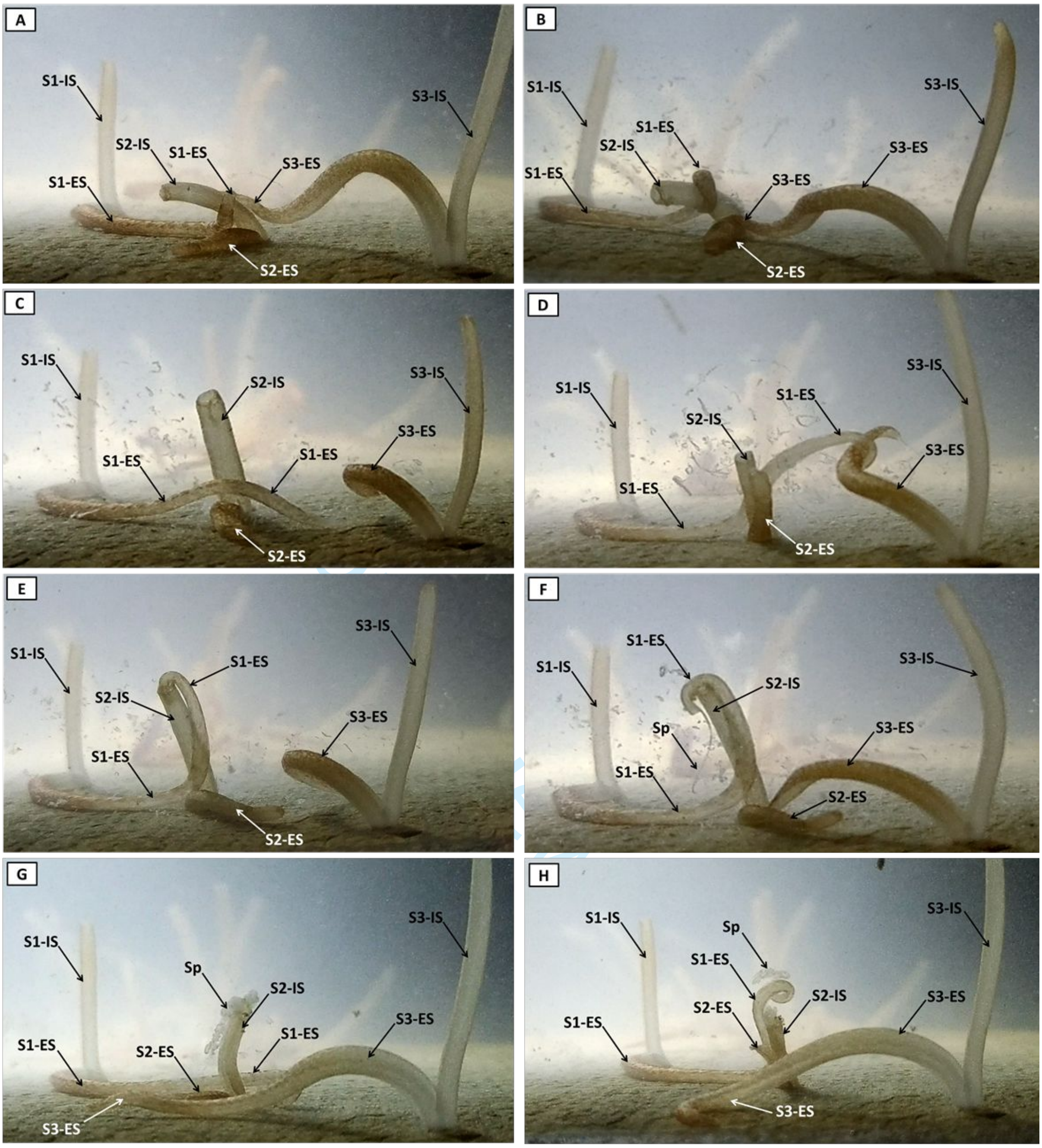

Figure 2. Mate competition during pseudocopulation in the giant feathery shipworm: a) two rival shipworms (S1, S3) compete for access to a recipient incurrent siphon; **b)** S1 uses its excurrent siphon to pull the recipient incurrent siphon (S2) towards itself and away from its competitor (S3); **c-d)** S1 pushes the excurrent siphon of its competitor (S3) away from the incurrent siphon of the middle specimen (S2); **e)** S1 inserts its excurrent siphon into the incurrent siphon of S2 and transfers spermatozoa; **f-g)** after an unsuccessful attempt to transfer spermatozoa, S3 leaves its spermatozoa on the surface of the incurrent siphon of S2; **h)** S1 then removes its rivals spermatozoa (S3) from the incurrent siphon of the recipient (S2). ES, excurrent siphon; IS, incurrent siphon; Sp, spermatozoa.

Discussion

Herein, we provide the first video footage of pseudocopulation and sperm transfer in shipworms - an extremely rare and unusual form of reproductive behaviour among the Bivalvia involving direct fertilization between neighbouring individuals. We also document rival mate competition during pseudocopulation, including: sparring of excurrent siphons between neighbouring individuals; mate guarding, where individuals will pull a receptive neighbours incurrent siphon towards itself and beyond the range of rivals; and, the removal of a rivals spermatozoa from the incurrent siphon of a potential mate. Whilst many taxa exhibit sophisticated plastic responses when encountering rival mates (Bretman et al., 2011; Wedell et al., 2002), this is the first record of putatively competitive mating observed in shipworms, and to our knowledge, the first among the Bivalvia.

It has been previously hypothesised that the rapid growth, swift development to maturity and large sizes of shipworms are traits associated with the ephemeral existence of life in wood (Turner, 1966; MacIntosh *et al*., 2014), but are also broadly characteristic of other taxa inhabiting temporary and unpredictable niches (Stearns, 1992). However, these traits combined with the xylotrepetic and xylophagous lifestyle of shipworms lead to rapid degradation of both the habitat and primary food source of these sessile animals. Indeed, the giant feathery shipworm can consume up to 82 % of the total internal volume of wood in a twelve-month period, has one of the fastest growth rates among the family reaching a total body length of 97.6 cm in just nine months (Haderlie & Mellor, 1972), with growth rates accelerating in larger animals (Kofoid & Miller, 1927). This is in stark contrast to other wood-boring marine invertebrate taxa (crustaceans from the family Cheluridae and Limnoriidae and the bivalve family Xylophagaidae), which display rapid growth to maturity but then remain at a relatively constant body size throughout their life span (Kühne & Becker, 1964; Cookson, 1991; Voight, 2015).

Whilst detrimental to longevity and survivability of life in wood, rapid development and growth to large size in shipworms may confer significant reproductive advantages through pseudocopulation. Shipworms that grow rapidly and reach larger sizes gain several advantages, including: increased fecundity and production of gametes for pseudocopulation and internal fertilization; longer siphons capable of inseminating mates further away; and, the effect of reducing both habitat and food availability for other cohabitants within the wood. Individuals that grow to large sizes are likely to fertilize more eggs via pseudocopulation, increasing the probability that larvae sharing their genes will survive in the following generation. This is broadly similar to observations of pseudocopulation in the acorn barnacle *Semibalanus balanoides,* where fertilization success positively correlates with body size, penis length and negatively with neighbour distance (Hoch & Yuen, 2009). As such, we suggest that the competitive mating behaviour during pseudocopulation may provide an alternative explanation to the hypothesis that large sizes are traits associated with the ephemeral niche of marine wood. To test this, future research should measure the ratio between siphon length and total body size in *Bankia setacea* to determine whether pseudocopulation selects for individuals with longer siphons, or individuals with both long siphons and large body size.

Pseudocopulation among shipworms has been only been observed on limited occasions and several gaps in our knowledge and understanding of this rare and unusual reproductive behaviour exist. Presently, pseudocopulation is recorded in 4/81 shipworm species (Rhoades, 1951; Townsley *et* al., 1966; Hiroki *et* al., 1994; Velasquez *et* al., 2011), and it is unknown how widespread this behaviour is within the family. Pseudocopulation is highly unlikely in the giant chemoautotrophic shipworm *Kuphus polythalamius*, and the deepsea species *Nivanteredo coronata*. For *K. polythalamius*, the siphons are separated by the architecture of the thick calcareous tube (Shipway *et al*., 2018), and in *N. coronata*, both the incurrent and excurrent siphons are joined along their entire length, prohibiting pseudocopulation (Velásquez & Shipway, 2018). We note that the incurrent siphons of *B. setacea* lack papillae, but the excurrent siphon is lined with rows of external papillae that become turgid during pseudocopulation (Fig. 1A, C, Supplemental Video). This is in contrast to other species with well-documental siphonal characters, which feature papillae located either internally or at the tip of the siphon, and are found on both the incurrent and excurrent siphons (Shipway *et al*., 2019a; Shipway *et al*., 2019b). It is possible that external papillae, particularly on the excurrent siphon, are a key morphological indicator for species that perform pseudocopulation and further research should assess the role these papillae play as chemical or mechanical sensory apparatus.

The sex ratios of individuals performing pseudocopulation is unknown. Shipworms exhibit simultaneous, consecutive and rhythmical-consecutive hermaphroditism (Nair & Saraswathy, 1971), and transformation between the sex phases can occur abruptly during the breeding season (Coe, 1936). Histological analysis on the shipworm *Nausitora fusticula* revealed that animals introducing their excurrent siphons to recipient individuals during pseudocopulation were identified as male and female respectively (Hiroki *et al*., 1994). However, previous studies have revealed the complexities of distinguishing ripe from partially spawned and spent gonads in histological preparation (McKoy, 1980) and several individuals observed in this study were both recipients and donors of spermatozoa.

Further investigation should determine if donor individuals are choosing recipients based on their sex expression and reciprocity of sperm transfer.

To date, all observations of shipworm pseudocopulation were carried out in aquariums with wood occupied by a single species. In nature, competition for wood can be high and sympatry is common, with up to 11 different species from multiple genera recorded occupying the same piece of wood (Cragg, 2007). The natural environmental conditions which facilitate pseudocopulation, whether this behaviour occurs in wood with high levels of sympatry, and the chemical or sensory mechanisms which detect these environmental conditions and con-specific mates in mixed species habitats are important questions in understanding how pseudocopulation contributes to reproductive success of these keystone species in the wild.

Acknowledgements

We thank the Oregon Institute of Marine Biology for access to resources, Dr Simon Cragg (University of Portsmouth, UK) and Dr Janet Voight (Chicago Field Museum, USA) for their helpful comments and suggestions, and Sacha Shipway for video editing.

Authors Contributions

JRS designed and performed the experiments and analysed the data. All authors provided critical feedback and helped shape the research, analysis and manuscript.

Ethical & Data Accessibility Statement

No potential conflict of interest was reported by the authors in this manuscript (RSBL-2020-0626.R1). The authors confirm that the data supporting the findings of this study are available within the article and its supplementary materials.

References

1. Levitan, D. R., & McGovern, T. M. (2005). The Allee effect in the sea. Marine conservation biology: the science of maintaining the sea’s biodiversity. Island Press, Washington DC, 47-57.
2. Levitan, D. R. (1998). Sperm limitation, sperm competition and sexual selection in external fertilizers. Pp. 173–215inT. Birkhead & A. Møller, eds. Sperm competition and sexual selection.
3. Hoch, J. M., & Yuen, B. (2009). An individual barnacle, *Semibalanus balanoides*, with two penises. Journal of Crustacean Biology, 29(1), 135-136.
4. Huber, M. (2015). Compendium of Bivalves 2. A full-color guide to the remaining seven families. A systematic listing of 8'500 bivalve species and 10'500 synonyms. ConchBooks.

5. Cragg SM, Beckham GT, Bruce NC, Bugg TD, Distel DL, Dupree P, Etxabe AG, Goodell BS, Jellison J, McGeehan JE, McQueen-Mason SJ. Lignocellulose degradation mechanisms across the Tree of Life. Current Opinion in Chemical Biology. 2015 Dec 1;29:108-19.
6. Voight, J. R. (2015). Xylotrophic bivalves: aspects of their biology and the impacts of humans. Journal of Molluscan Studies, 81(2), 175-186.
7. MacIntosh, H., de Nys, R., & Whalan, S. (2014). Contrasting life histories in shipworms: Growth, reproductive development and fecundity. Journal of experimental marine biology and ecology, 459, 80-86.
8. Calloway, C. B., & Turner, R. D. (1983). Documentation and implications of rapid successive gametogenic cycles and broods in the shipworm *Lyrodus floridanus* (Bartsch)(Bivalvia, Teredinidae). Journal of Shellfish Research, 3, 65-69.
9. de Paula Müller, A. C., & da Cunha Lana, P. (1986). TEREDINIDAE (MOLLUSCA, BIVALVIA) DO LITORAL DO PARANÁ, BRASIL. Revista Nerítica, 1(3).
10. Sigerfoos, C. P. (1908). Natural history, organization, and late development of the Teredinidae, or ship-worms (No. 639). US Government Printing Office.
11. Shipway, J. R., O’Connor, R., Stein, D., Cragg, S. M., Korshunova, T., Martynov, A., Haga, T., & Distel, D. L. (2016). *Zachsia zenkewitschi* (Teredinidae), a rare and unusual seagrass boring bivalve revisited and redescribed. PLoS One, 11(5), e0155269.
12. Nair, N. B., & Saraswathy, M. (1971). The biology of wood-boring teredinid molluscs. In Advances in marine biology (Vol. 9, pp. 335-509). Academic Press.
13. Coe, W. R. (1941). Sexual phases in wood-boring mollusks. The Biological Bulletin, 81(2), 168-176.
14. Turner, R. D. (1966). A survey and illustrated catalogue of the Teredinidae (Mollusca: Bivalvia). A survey and illustrated catalogue of the Teredinidae (Mollusca: Bivalvia).
15. Rhoades, A. S. (1951). Fourth progress report on marine borer activity in test boards operated during 1950. In, Observations on living Teredinidae. Fourth Progress Report [Rept. No. 7550], William F. Clapp Laboratories Inc.
16. Hiroki, K., Leonel, R. M. V., & Lopes, S. G. B. C. (1994). Reproductive events of Nausitora fusticula (Jeffreys, 1860)(Mollusca, Bivalvia, Teredinidae). Invertebrate Reproduction & Development, 26(3), 247-250.
17. Townsley, P. M., Richy, R. A., & Trussell, P. C. (1966). The laboratory rearing of the shipworm, *Bankia setacea* (Tryon). National Shellfisheries Association, Proceedings, 56, 49-52.

18. Velásquez, M., Gallardo, C. S., & Lira, C. Fecundación interna en *Bankia martensi* (Stempell, 1830)(Bivalvia: Teredinidae) del sur de Chile (Bivalvia: Teredinidae) del sur de Chile. AMICI MOLLUSCARUM, 33.
19. Turner, R. D. (1971). Identification of marine wood-boring molluscs. Organisation for economic co-operation and development.
20. Bretman, A., Gage, M. J., & Chapman, T. (2011). Quick-change artists: male plastic behavioural responses to rivals. Trends in ecology & evolution, 26(9), 467-473.
21. Wedell, N., Gage, M. J., & Parker, G. A. (2002). Sperm competition, male prudence and sperm-limited females. Trends in Ecology & Evolution, 17(7), 313-320.
22. Stearns, S. C. (1992). The evolution of life histories (No. 575 S81).
23. Haderlie, E. C., & Mellor, J. C. (1973). Settlement, growth rates and depth preference of the shipworm *Bankia setacea* (Tryon) in Monterey Bay. Veliger, 15, 265-286.
24. Kofoid, C. A., & Miller, R. C. (1927). Biological selection. Pp. 183-343. *In:* Marine borers and their relation to marine construction on the Pacific Coast. Final report. San Francisco Bay Marine Piling Committee.
25. Kühne, H., & Becker, G. (1964). Der Holz-Flohkrebs *Chelura terebrans* Philippi (Amphipoda, Cheluridae): Morphologie, Verbreitung, Lebensweise, Verhalten, Entwicklung und Umweltabhängigkeit. Duncker & Humblot.
26. Cookson, L. J. (1991). Australasian species of Limnoriidae (Crustacea: Isopoda). Memoirs of the Museum of Victoria, 52(2), 137-262.
27. Shipway, J. R., Altamia, M. A., Haga, T., Velásquez, M., Albano, J., Dechavez, R., Concepcion, G. P., Haygood, M. G., & Distel, D. L. (2018). Observations on the life history and geographic range of the giant chemosymbiotic shipworm *Kuphus polythalamius* (Bivalvia: Teredinidae). The Biological Bulletin, 235(3), 167-177.
28. Velásquez, M., & Shipway, J. R. (2018). A new genus and species of deep-sea wood-boring shipworm (Bivalvia: Teredinidae) *Nivanteredo coronata* n. sp. from the Southwest Pacific. Marine Biology Research, 14(8), 806-815.
29. Shipway, J. R., Altamia, M. A., Rosenberg, G., Concepcion, G. P., Haygood, M. G., & Distel, D. L. (2019). *Tamilokus mabinia*, a new, anatomically divergent genus and species of wood-boring bivalve from the Philippines. PeerJ, 7, e6256.
30. Shipway, J. R., Altamia, M. A., Rosenberg, G., Concepcion, G. P., Haygood, M. G., & Distel, D. L. (2019). A rock-boring and rock-ingesting freshwater bivalve (shipworm) from the Philippines. Proceedings of the Royal Society B, 286(1905), 20190434.

31. McKoy, J. L. (1980). Sexual phases in *Bankia neztalia* (Bivalvia: Teredinidae) from Wellington Harbour. New Zealand Journal of Marine and Freshwater Research, 14(3), 277-282.
32. Cragg, S. M. (2007). In, The Ecology of Papua.

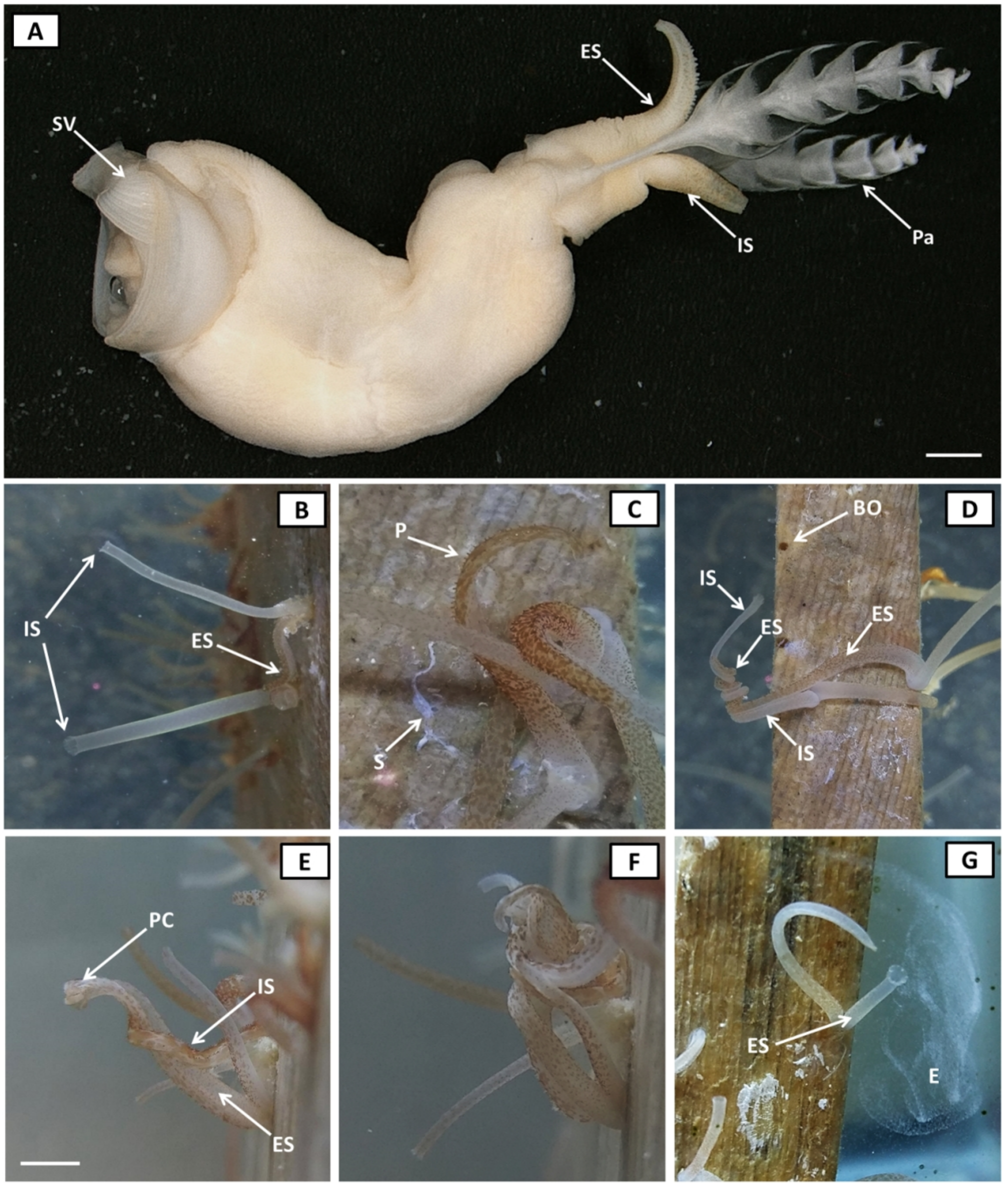

Figure 1. Pseudocopulation in the giant feathery shipworm: A) specimen removed from wood; B) flaring of the incurrent siphonal opening; C) sensory papillae on excurrent siphons become erect; D) an excurrent siphon entwines the incurrent siphon of neighbouring individual; E) an excurrent siphon penetrates the incurrent siphon of a neighbour and transfers spermatozoa; F) multiple individuals simultaneously display pseudocopulatory behaviour; G) eggs are spawned into the water column. BO, burrow opening; E, eggs; ES, excurrent siphon; PC, pseudocopulation; IS, incurrent siphon; P, papillae; Pa, pallet; S, sperm; SV shell valve . Scale bars = 1 mm (a) and 1 cm (b-g).

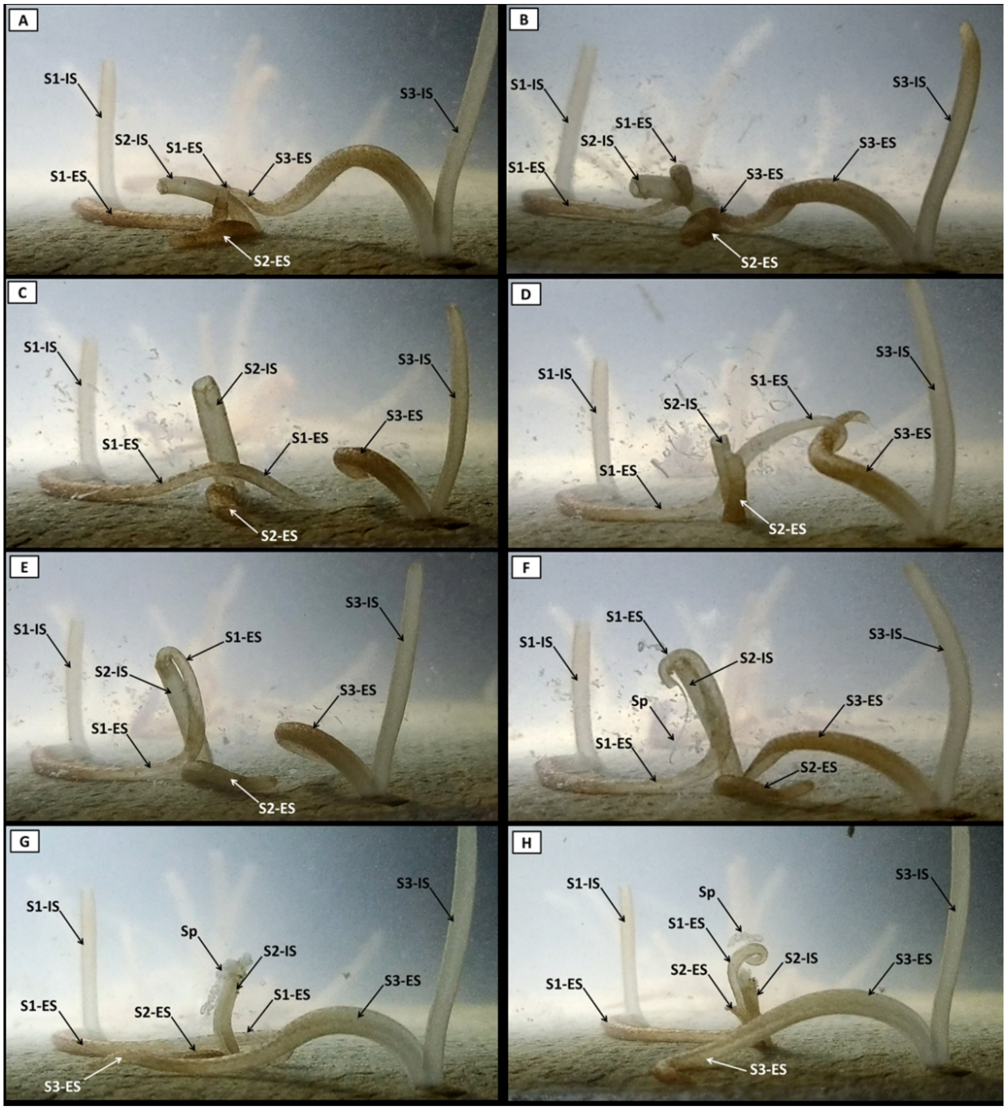

Figure 2. Mate competition during pseudocopulation in the giant feathery shipworm: a) two rival shipworms (S1, S3) compete for access to a recipient incurrent siphon; b) S1 uses its excurrent siphon to pull the recipient incurrent siphon (S2) towards itself and away from its competitor (S3); c-d) S1 pushes the excurrent siphon of its competitor (S3) away from the incurrent siphon of the middle specimen (S2); e) S1 inserts its excurrent siphon into the incurrent siphon of S2 and transfers spermatozoa; f-g) after an unsuccessful attempt to transfer spermatozoa, S3 leaves its spermatozoa on the surface of the incurrent siphon of S2; h) S1 then removes its rivals spermatozoa (S3) from the incurrent siphon of the recipient (S2). ES, excurrent siphon; IS, incurrent siphon; Sp, spermatozoa.