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# The placozoan *Trichoplax*

By Marvin Leria, Magali Requin, André Le Bivic & Andrea Pasini

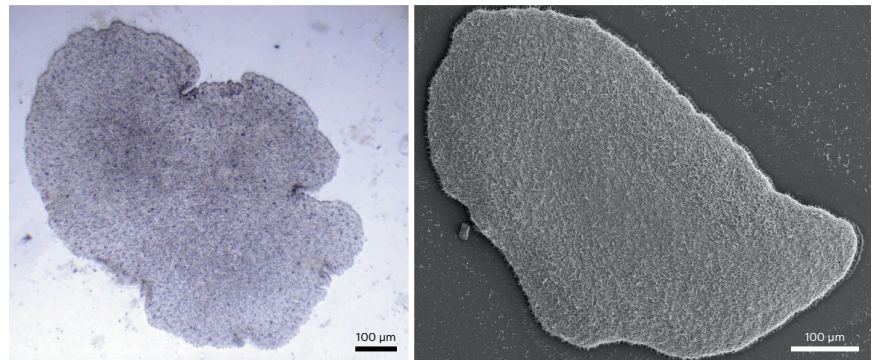


**After having been largely neglected for more than a century, the tiny marine animal *Trichoplax adhaerens* is enjoying a revival and is now poised to become a mainstream model organism for the study of epithelial evolution, diversification and specialization.**

**S**ome animals are so weird that they seem to have come straight out of the fertile imagination of a sci-fi writer. If there were a contest for the strangest-looking animal, *Trichoplax adhaerens* ('sticky hairy platelet') and its fellow placozoans would surely be among the most likely winners.

Since its first description by the German zoologist Franz Eilhard Schulze<sup>1</sup>, who discovered it on the wall of a seawater aquarium at the University of Graz in Austria in 1883, *Trichoplax* appeared to be an unusual organism. Its ameboid aspect and shifting morphology, its lack of apparent body axes and its peculiar anatomy, made of only two ciliated epithelial layers, set it apart from all other known animals. In the absence of a more convincing taxonomic classification, *Trichoplax* was ranged among the Mesozoa, at the time a wastebasket taxon containing a motley crew of morphologically 'simple' and hence supposedly 'primitive', mostly ciliated, organisms. For the next hundred years or so *Trichoplax* was largely forgotten by experimentalists, although it continued to feature in theoretical hypotheses and debates on the origin of multicellular animals.

In the early 1970s *Trichoplax* was rediscovered in samples from the Red Sea by another German zoologist, Karl Gottlieb Grell, who created for it a completely new metazoan phylum, the Placozoa<sup>2</sup>. Recently, this phylum has been expanded to incorporate three newly described species – *Hoilungia hongkongensis*, *Polyplacotoma mediterranea* and *Cladertaria collaboinventa*<sup>3,4</sup> – as well as several *Trichoplax* haplotypes genetically distinguished on the basis of mitochondrial 16S rRNA<sup>4–6</sup>. The phylogenetic position of Placozoa has been controversial: recent evidence points to them being one of the most basal metazoan phyla, together with Porifera and Ctenophora<sup>7</sup>,



**Fig. 1 | *Trichoplax* morphology.** Bright-field and scanning electron microscopy images of *Trichoplax* of the H2 haplotype.

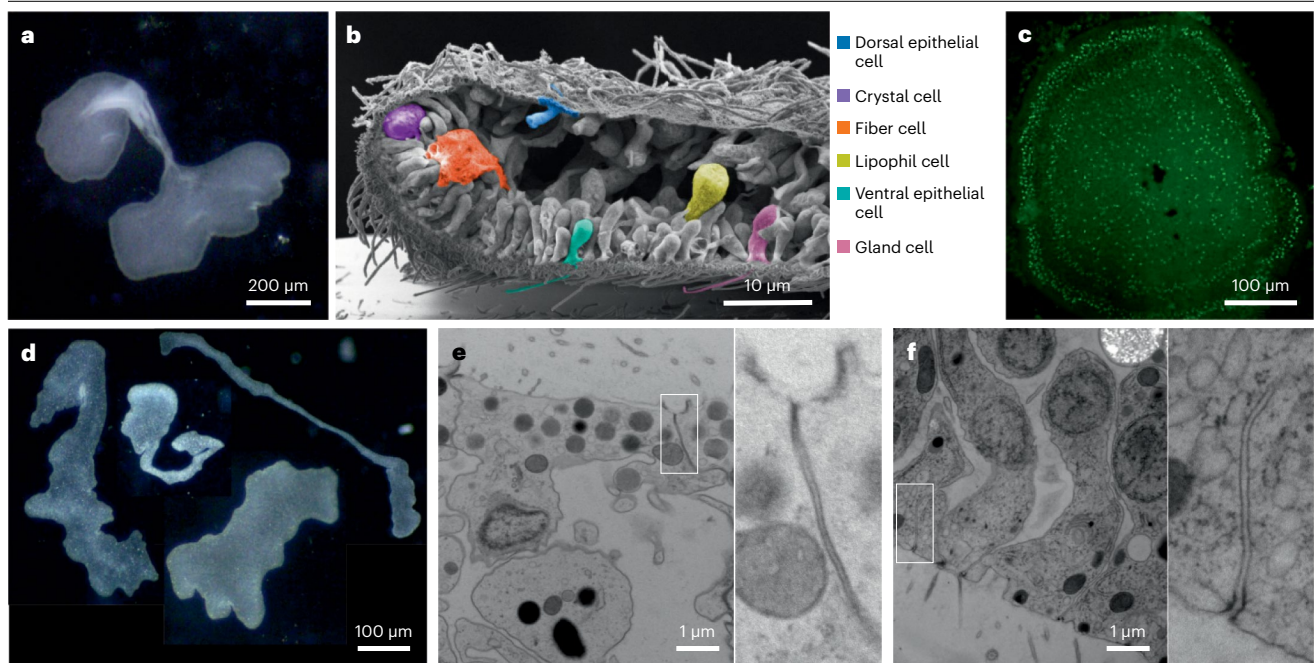
although an alternative explanation is that they might constitute a very derived sister group to Cnidaria. Grell's rediscovery and the phylogenetic debate sparked a renewed interest in placozoans in general and *Trichoplax* in particular, and over the last 50 years knowledge has been slowly but steadily accumulating on this unusual organism, which is now poised to become an exciting model for studies in evolutionary cell and organismal biology.

*Trichoplax* is a small and flat (around 1–2 mm in diameter and 20–30 µm thick) benthic marine animal mainly found in shallow subtropical and tropical waters (Fig. 1). It lives on rocks, corals and other substrates and feeds on biofilms of algae, bacteria and other microorganisms by means of external digestion. As with all placozoans, its life cycle remains enigmatic. Reproduction occurs mostly asexually, generally by binary fission (Fig. 2a), although budding of juveniles ('swarmers') from the adults has been described<sup>8,9</sup>. Sexual reproduction and embryonic development have never been observed in the laboratory, but there are hints that they might occur in nature<sup>8</sup>.

At 98 Mb, the *Trichoplax* genome is one of the smallest among all free-living animals, but it possesses a rich gene repertoire, with many of the gene families found in morphologically more complex metazoans<sup>7</sup>. In contrast, its mitochondrial genome is one of the largest in the animal kingdom<sup>10</sup>. Remarkably, the regulation of gene expression in *Trichoplax* appears to rely essentially on short proximal promoter

regulatory elements, without the need for more distal enhancer elements, a feature that sets *Trichoplax* apart from other metazoans and closer to unicellular eukaryotes<sup>11</sup>. A pioneering series of in situ hybridization experiments has revealed several instances of regionalized gene expression pattern<sup>12</sup>. Some genes are expressed in concentric patterns, some in a 'salt and pepper' fashion across both upper and lower epithelia or restricted to only one of the two, while others reveal unexpected asymmetric patterns.

*Trichoplax* are generally comprised of tens of thousands to hundreds of thousands of cells but contain a limited number of morphologically distinct cell types (Fig. 2b), organized in two epithelial sheets that delimit a narrow inner cavity<sup>13</sup>. The upper epithelium is almost exclusively constituted by mushroom-shaped, monociliated dorsal epithelial cells, with a very flat and thin apical aspect. In some *Trichoplax* haplotypes, a few dorsal cells also contain large vesicles, called 'shiny spheres', thought to be involved in defense against predators. The lower epithelium mostly consists of columnar, monociliated ventral epithelial cells, with interspersed secretory lipophil cells, gland cells and peptidergic cells. The ciliary beating of the ventral epithelial cells is largely responsible for the gliding movements of the animal on the substrate, while the large lipophil cells secrete digestive enzymes and the gland cells produce mucus<sup>13</sup>. Single-cell transcriptomics recently showed that the peptidergic cells are a heterogeneous



**Fig. 2 | Features of *Trichoplax* epithelia.** **a**, Stereomicroscope backlit view of a live, dividing *Trichoplax* H2. **b**, Scanning electron microscopy image of a fractured *Trichoplax* H2, colored to highlight the different cell types. **c**, Confocal microscopy image of a *Trichoplax* H2 stained with an antibody against a peptide expressed by gland cells, showing their distribution in the lower epithelium. **d**, Stereomicroscope composite of four backlit live *Trichoplax* H2 to highlight

their morphological plasticity. **e**, Transmission electron microscopy showing cells of the upper epithelium. Note the lack of basal lamina and the small, apical intercellular junctions (white box, magnified at right). **f**, Transmission electron microscopy showing cells of the lower epithelium. Note the lack of basal lamina and the small, apical intercellular junctions (white box, magnified at right).

population, comprising at least 14 subpopulations that express different combinations of neuropeptides – short soluble peptides (5 to >60 amino acids) that act as chemical messengers to modulate the activity of target cells<sup>11</sup>. Staining with several lectins, lipid stains or antibodies to neuropeptide revealed that some cell populations of the lower epithelium are distributed in concentric rings (Fig. 2c). All around the rim of the animal are gravity-sensing crystal cells, while the inner body cavity contains branching fiber cells, whose function is currently unclear, but that show contractile and phagocytic activities.

Despite its morphological and histological simplicity and, notably, its lack of an organized, interconnected neuron-based nervous system, *Trichoplax* shows positive and negative taxis in response to several chemical and physical stimuli (food availability, temperature, light, pH, gravity), defensive behaviors against predators and fast, complex differential responses to several neuropeptides, including behavioral and morphological changes. Indeed, a striking feature of *Trichoplax*, and one that we are investigating in our lab, is its morphological plasticity. Single

individuals are able to transition from irregularly rounded, pancake-like shapes to highly branched forms or even to extremely elongated filaments, in a matter of a few minutes (Fig. 2d). The physiological meaning of these shape changes and the environmental stimuli that trigger them are still unknown, but they might be under the control of neuropeptides<sup>14</sup>.

In addition, *Trichoplax* is efficient at repairing wounds, which allows any spontaneous or inflicted tear in the epithelial bilayer to close within minutes, presumably by cell rearrangements within the epithelial sheets, without cell proliferation<sup>15</sup>. Morphological remodeling and wound-closure ability are also likely to underlie the capacity of *Trichoplax* to immediately give rise to two independent and seemingly functional individuals upon experimental bisection.

The cellular and molecular bases of such an extraordinary plasticity remain by and large unexplored. A possible involvement of fiber cell contractility has been proposed, but the peculiar features of *Trichoplax* epithelia are also likely to play a major role. Both upper and lower epithelia lack basal lamina, and their constituent cells are only connected by small

apical adherens-like intercellular junctions (Fig. 2e,f). In addition, the monociliated epithelial cells show dynamic behaviors, such as actomyosin-based ultra-fast apical constriction and shape changes of the apical surface, which might help to simultaneously endow *Trichoplax* epithelia with plasticity and resilience to rupturing<sup>16</sup>.

Understanding how such a basally divergent, but possibly very derived, metazoan as *Trichoplax* is able to reconcile the need for tissue integrity maintenance with its extreme epithelial plasticity will shed light both on conserved ancestral features of the common metazoan epithelial design and on the emergence of morphological and physiological novelties. A related issue is to understand how complex collective cell behaviors can be elicited and coordinated in *Trichoplax* in the absence of a connected neural network, and presumably only or mostly by means of neuropeptide-dependent paracrine signaling<sup>11,14</sup>. The answers to such questions will result in a better comprehension of the evolutionary history of epithelia and their interaction with the neural signals. In addition, potential long-term outcomes in the field of

tissue engineering and biomaterials might be possible.

These are of course challenging tasks, which will likely require an interdisciplinary research strategy, exploiting both classical ‘wet biology’ techniques and more quantitative approaches, such as advanced image analysis and cell tracking, as well as in silico modeling. *Trichoplax* is relatively easy to maintain in a research laboratory, with no other requirements than a controlled temperature, weekly changes of water and feeding with unicellular algae. While its small size and thin, flat body make *Trichoplax* almost ideally suited for imaging and modeling, the limited availability of reliable tools to interfere with gene function constitutes a major hurdle. The fact that under laboratory conditions *Trichoplax* reproduces mostly asexually means not only that forward genetics approaches are impossible, but also that zygotes or very early developmental stages in which to inject or electroporate foreign genetic material are not available. Possible alternative strategies are the (almost inevitably mosaic) transfection of whole animals or the genetic manipulation of as yet unidentified stem cells.

Some results already suggest that whole-body transfection is feasible, and it is on improving these approaches that the small but growing community of *Trichoplax* researchers should focus in the near future. Only then will this long-neglected, shape-shifting animal make yet another surprising change to become a mainstream model organism.

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## References

- Schulze, F. E. *Zool. Anz.* **6**, 92–97 (1883).
- Grell, K. & Benwitz, G. *Cytobiologie* **4**, 216–240 (1971).
- Eitel, M. et al. *PLoS Biol.* **16**, e2005359 (2018).
- Osigus, H.-J., Rolfes, S., Herzog, R., Kamm, K. & Schierwater, B. *Curr. Biol.* **29**, R148–R149 (2019).
- Eitel, M., Osigus, H.-J., DeSalle, R. & Schierwater, B. *PLoS One* **8**, e57131 (2013).
- Miyazawa, H. et al. *Genome Biol. Evol.* **13**, evaa213 (2021).
- Srivastava, M. et al. *Nature* **454**, 955–960 (2008).
- Eitel, M., Guidi, L., Hadrys, H., Balsamo, M. & Schierwater, B. *PLoS One* **6**, e19639 (2011).
- Zuccolotto-Arellano, J. & Cuervo-González, R. *Mech. Dev.* **162**, 103608 (2020).
- Dellaporta, S. L. et al. *Proc. Natl. Acad. Sci. USA* **103**, 8751–8756 (2006).
- Najle, S. R. et al. *Cell* **186**, 4676–4693.e29 (2023).
- DuBuc, T. Q., Ryan, J. F. & Martindale, M. Q. *Mol. Biol. Evol.* **36**, 966–973 (2019).
- Smith, C. L. et al. *Curr. Biol.* **24**, 1565–1572 (2014).
- Varoqueaux, F. et al. *Curr. Biol.* **28**, 3495–3501.e2 (2018).
- Mayorova, T. D. et al. *Sci. Rep.* **11**, 23343 (2021).
- Armon, S., Bull, M. S., Aranda-Díaz, A. & Prakash, M. *Proc. Natl. Acad. Sci. USA* **115**, E10333–E10341 (2018).

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## Author contributions

M.L., M.R. and A.P. designed, performed and analyzed the experiments. M.L., M.R. and A.P. prepared the figures. A.L.B. provided support and funding. A.P. provided funding. A.P. wrote and edited the manuscript.

## Competing interests

The authors declare no competing interests.