Mitchison leaves us with more new questions than answers yet, which is in fact the best possible outcome of a scientific study.

References

1. Loose, M., and Mitchison, T.J. (2014). The bacterial cell division proteins FtsA and FtsZ self-organize into dynamic cytoskeletal patterns. Nat. Cell Biol. *16*, 38–46.
2. Erickson, H.P., Anderson, D.E., and Osawa, M. (2010). FtsZ in bacterial cytokinesis: cytoskeleton and force generator all in one. Microbiol. Mol. Biol. Rev. *74*, 504–528.
3. Li, Y., Hsin, J., Zhao, L., Cheng, Y., Shang, W., Huang, K.C., Wang, H.W., and Ye, S. (2013). FtsZ protofilaments use a hinge-opening mechanism for constrictive force generation. Science *341*, 392–395.
4. Schwille, P. (2011). Bottom-up synthetic biology: engineering in a tinkerer's world. Science *333*, 1252–1254.
5. Mitchison, T., and Kirschner, M. (1984). Dynamic instability of microtubule growth. Nature *312*, 237–242.
6. Howard, J., Hudspeth, A.J., and Vale, R.D. (1989). Movement of microtubules by single kinesin molecules. Nature *342*, 154–158.
7. Loose, M., Fischer-Friedrich, E., Ries, J., Kruse, K., and Schwille, P. (2008). Spatial regulators for bacterial cell division self-organize into surface waves in vitro. Science *320*, 789–792.
8. Zieske, K., and Schwille, P. (2013). Reconstitution of pole-to-pole oscillations of Min proteins in microengineered polydimethylsiloxane compartments. Angew. Chem. Int. Ed. Engl. *52*, 459–462.
9. Osawa, M., Anderson, D.E., and Erickson, H.P. (2008). Reconstitution of contractile FtsZ rings in liposomes. Science *320*, 792–794.
10. Mateos-Gil, P., Paez, A., Hörger, I., Rivas, G., Vicente, M., Tarazona, P., and Vélez, M. (2012). Depolymerization dynamics of individual filaments of bacterial cytoskeletal protein FtsZ. Proc. Natl. Acad. Sci. USA *109*, 8133–8138.
11. Arumugam, S., Chwastek, G., Fischer-Friedrich, E., Ehrig, C., Mönch, I., and Schwille, P. (2012). Surface topology engineering of membranes for the mechanical investigation of the tubulin homologue FtsZ. Angew. Chem. Int. Ed. Engl. *51*, 11858–11862.
12. Thanedar, S., and Margolin, W. (2004). FtsZ exhibits rapid movement and oscillation waves in helix-like patterns in Escherichia coli. Curr. Biol. *14*, 1167–1173.
13. Schaller, V., Weber, C., Semmrich, C., Frey, E., and Bausch, A.R. (2010). Polar patterns of driven filaments. Nature *467*, 73–77.
14. Mendes Pinto, I., Rubinstein, B., Kucharavy, A., Unruh, J., and Li, R. (2012). Actin depolymerization drives actomyosin ring contraction during budding yeast cytokinesis. Dev. Cell *22*, 1247–1260.
15. Murrell, M.P., and Gardel, M.L. (2012). F-actin buckling coordinates contractility and severing in a biomimetic actomyosin cortex. Proc. Natl. Acad. Sci. USA *109*, 20820–20825.
16. Vogel, S.K., Petrasek, Z., Heinemann, F., and Schwille, P. (2013). Myosin motors fragment and compact membrane-bound actin filaments. eLife *2*, e00116.
17. Mendes Pinto, I., Rubinstein, B., and Li, R. (2013). Force to divide: structural and mechanical requirements for actomyosin ring contraction. Biophys. J. *105*, 547–554.
18. Domínguez-Escobar, J., Chastanet, A., Crevenna, A.H., Fromion, V., Wedlich-Söldner, R., and Carballido-López, R. (2011). Processive movement of MreB-associated cell wall biosynthetic complexes in bacteria. Science *333*, 225–228.
19. Garner, E.C., Bernard, R., Wang, W., Zhuang, X., Rudner, D.Z., and Mitchison, T. (2011). Coupled, circumferential motions of the cell wall synthesis machinery and MreB filaments in B. subtilis. Science *333*, 222–225.
20. Fenton, A.K., and Gerdes, K. (2013). Direct interaction of FtsZ and MreB is required for septum synthesis and cell division in Escherichia coli. EMBO J. *32*, 1953–1965.

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Life-History Evolution: At the Origins of Metamorphosis

Metamorphosis is a widespread life history strategy of animals but apart from some model organisms it is poorly characterized. A recent study of moon jellies highlights the similarities and differences between the various types of metamorphosis and illuminates its molecular determinants.

Thomas W. Holstein[1] and Vincent Laudet[2]

Because we humans are living on land surrounded by mammals or birds that exhibit life cycles similar to our own, we often do not appreciate that most animals have much more complex life histories. In fact, extant metazoans exhibit a wide variety of life cycles — sometimes incredibly complex ones, especially in parasitic species. The most common strategy is a biphasic cycle with a larva emerging from the egg and a metamorphosis that allows the transformation of the larva into a juvenile that will experience sexual maturation and become an adult. This strategy often involves dramatic morphological, physiological, behavioral and ecological transformations between the larvae and the juvenile [1,2]. Even in some vertebrates, such as teleost fishes, the difference between a larva and a juvenile can be so big that the two forms have sometimes been mistaken as different species [3]. The advantage of such biphasic life history strategies are numerous [2]: for example, larval stages allow for dispersal, as in most marine animals larvae are pelagic forms that take advantage of marine currents to travel far from their site of release. Moreover, this system allows the larva and the juvenile to exploit different ecological niches — most tadpoles, for example, are aquatic herbivores while most frogs are terrestrial carnivores. Finally, biphasic life history strategies allow the larva and the adult to become specialized in different activities: the fly larva (the maggot) is a specialized form to exploit short-term food sources and to grow rapidly. In contrast the adult's main function is to find a mate and to reproduce; in some insects such as mayflies the adult is even unable to feed. The diversity of life history strategies based on this relatively simple system — larva, metamorphosis and juvenile — is enormous, but very little is known about how it originated. A study on the moon jelly *Aurelia aurita* by Fuchs *et al.* [4] in *Current Biology* provides a first and fascinating insight into the evolutionary origin of metamorphosis.

All cnidarians have free-swimming planula larvae that settle and develop into sessile polyps. In anthozoans (corals and sea anemones) these polyps can propagate asexually, but never form medusae. The other groups of cnidarians (called Medusozoa; Figure 1) have sessile polyps, which can either propagate asexually or undergo a transition into a pelagic, free-swimming medusa (jelly) that differentiates gonads and carries out sexual reproduction. The morphology of a medusa is closely related to that of a polyp. In essence, both have a gastrula-shaped body plan with two germ layers (ectoderm and endoderm), an intermediate extracellular matrix (mesoglea) and one opening of the gastric cavity at the oral side. In the medusa, the mesoglea forms an enlarged jelly-like umbrella at the aboral side, which ensures free floating

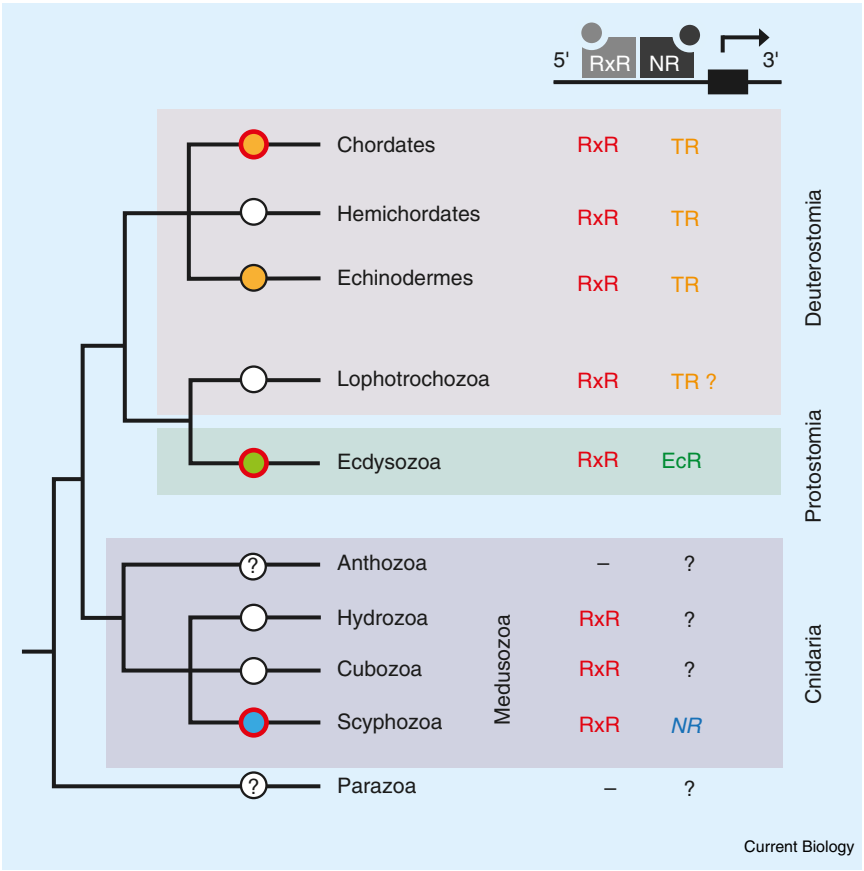

Figure 1. Comparative molecular aspects of metazoan indirect development.

In all major metazoan clades indirect development (circle) has been described. The proposed core model (grey) consists of a nuclear retinoid acid receptor (RxR), which is based on genome models present in most metazoans, and an interacting nuclear receptor (NR), which induce downstream target genes of metamorphosis. NRs are the thyroid hormone receptor (TR, yellow) from vertebrates (chordates), the ecdysone receptor (EcR) of the ecdysozoans, or unknown NRs (blue). In scyphozoans (Cnidarian) there is evidence for a peptide interacting NR (filled blue circle). Evidence for thyrosine and ecdysone are symbolized by filled yellow and green circles, respectively. Red circles indicate a proven function of RxR in indirect development.

and swimming of the animal. The polyp–medusa transition in scyphozoans (e.g. in the moon jelly *Aurelia aurita*) is called strobilation. During strobilation, transverse fissions transform the entire polyp into multiple disc-like young medusae. In the related box jellyfish (cubozoans), the polyps metamorphose into a single medusa [5], while in hydrozoans, the polyps generate medusae by lateral budding.

Apart from amphibians and insects, our understanding of the molecular cascade controlling metamorphosis is very poor (Figure 1). In both systems, hormones (thyroid hormone in amphibians, ecdysone and juvenile hormone in insects), whose production is controlled by the nervous system, as well as growth and environmental signals, control a complex tissue-specific gene regulatory network that drives metamorphosis [1,2]. Core regulators of this process on the transcriptional level are nuclear hormone receptors, i.e. the ecdysone receptor (EcR) in insects and the thyroid hormone receptor (TR) in amphibians. Each of these receptors can interact with a nuclear receptor (RxR) that is activated by 9-*cis* retinoic acid [6]. However, we have no clear idea if the process of metamorphosis in diverse metazoans derives from a common mechanism, or whether each case is the product of independent evolution.

During strobilation, the polyp's body is transformed into multiple disc-like young medusae called *ephyrae* that can actively swim and develop into mature moon jellies. To unravel the polyp–medusa transition, Fuchs *et al.* [4] performed a transcriptome analysis using normal and strobilating polyps, as well as freshly detached ephyrae. By comparing these samples the authors found strobila-specific transcripts encoding key mediators in the retinoic acid (RA) signaling pathway. The RXR nuclear hormone receptor has been previously identified in the box jelly *Tripedalia cystophora* [7], but its function was unclear. Now, Fuchs *et al.* [4] could convincingly demonstrate that RA signaling is essential for the induction of metamorphosis in *Aurelia aurita*. Expression of the RXR nuclear hormone receptor and retinol dehydrogenases RDH 2 were strongly up regulated at the onset of strobilation. Furthermore, treatment of polyps with retinol and its metabolite retinoic acid induced strobilation, while antagonists of the RXR receptor (UV13003) had an antagonistic effect.

But what is the interaction partner of RXR in *Aurelia*? Here, the answer is less clear, because so far no interaction partner of RXR has been described. However, there is indirect evidence for the existence of such a factor. Fuchs *et al.* [4] made the interesting discovery that a gene encoding methyltransferase 1 (DNMT1) was among the strobilation specific genes. DNMT1 has a crucial role in tissue-specific DNA methylation during development and cancer [8]. Strobilation was blocked when polyps were treated with the DNA methyltransferase inhibitor 5-azacytidine [9]. A further transcriptome analysis using strobilating and 5-azacytidine-treated polyps revealed three strobilation-specific genes encoding novel secreted proteins. One protein (CL390) was a peptide precursor containing a WSRRRWL peptide. When this peptide was added to the culture medium it could also effectively induce strobilation. These data suggest that CL390 is the putative strobilation hormone.

The mechanism of interaction between 9-*cis* RA and *CL390* remains unclear. One possibility could be that CL390 is a ligand of RXR itself. Unfortunately, there are no data addressing this interaction directly. As 9-*cis* RA can stimulate both the *RxR* and *CL390* genes,

Fuchs *et al.* [4] conclude that similar to the metamorphosis insects and amphibians, the RXR receptor in *Aurelia* belongs to a core molecular module, which is interacting with an unknown nuclear receptor. This nuclear receptor is supposed to bind CL390 and in turn to interact with the activated RXR, which then is inducing the genes required for the polyp–medusa transition.

As is often the case, this discovery poses more questions than it answers. RXR is known to be a heterodimeric partner with other nuclear receptors, for example TR, the receptor for thyroid hormones (acting during amphibian metamorphosis), or EcR, the receptor for ecdysone (acting during insect metamorphosis). Although no orthologue of either EcR or TR can be found in the genome of *Aurelia*, strikingly, there are data suggesting that iodine and even thyroxin can induce metamorphosis in *Aurelia* [10,11]. It will therefore be very interesting to identify the heterodimeric partner of RXR, as it may provide a link to another hormonal system that could play an important role in this process.

Another question that will be interesting to follow up is the precise role of 9-*cis* RA. It is clear from the data presented in Fuchs *et al.* [4] that this molecule can trigger strobilation, but whether this molecule is actually present in *Aurelia* is still unknown. This is not a trivial question, as in mammals, even if 9-*cis* RA is a potent RXR ligand (there are drugs developed for human disease based on its activity), its presence *in vivo* and its possible role as an endogenous ligand is still under debate [6]. Some authors have proposed that fatty acids, such as docohexaenoic acid (DHA), may be the real endogenous ligands for RXR [12]. DHA has been shown to activate RXR from invertebrate species [13] and it is therefore possible that DHA or another derivative could be the compound that controls strobilation in *Aurelia*. This could provide a link between the control of metamorphosis and the physiology of the animal.

Another issue is what happens during the other life-cycle transition of cnidarians, the planula–polyp transition, which is also considered a metamorphosis [14]. This transition is initiated after settlement of the planula larva to an appropriate substrate and includes the formation of a functional gut and tentacles at the oral side of the primary polyp. In the hydrozoan *Hydractinia echinata*, treatment with 9-*cis* RA during the planula–polyp transition had no effects on the transition, besides an increase in the number of tentacles in primary polyps [15]. This is in line with the results of Fuchs *et al.* [4], arguing against a function of 9-*cis* RA or RXR in the planula–polyp transition. Based on these observations, one should consider the possibility that the planula–polyp transition is simply a form of direct development.

The data of Fuchs *et al.* [4] have important implications for the evolutionary origin of metamorphosis and the evolution of metazoan life cycles. Indirect development is a quite common phenomenon in many basal marine metazoans. Jägersten [16] postulated an ancient planktotrophic larval stage, a hypothesis that was further developed in the trochaea-theory [17]. Indirect development as a primitive condition was questioned by Wolpert [18], because it contradicts the principle of gradualism. Instead, he favored the view that metamorphosis evolved by modification of direct development. According to Wolpert's view, metamorphosis gradually evolved several times and independently in various lineages of the metazoan tree. By comparison, Davidson *et al.* [19] proposed as a cellular basis for indirect development the existence and developmental use of yet "undifferentiated set aside cells", which retain indefinite division potential for the morphogenesis of large structures.

The data of Fuchs *et al.* [4] indicate that RXR receptors and RA signaling were already present in the common ancestors of cnidarians and bilaterians. Probably they are the molecular basis for Davidson's cells, which was utilized in different branches of the metazoan tree of life (cnidarians, insects and vertebrates) by newly evolved taxon-specific nuclear receptors interacting with this ancient RA signaling module.

References

1. Laudet, V. (2011). The origins and evolution of vertebrate metamorphosis. Curr. Biol. *21*, R726–R737.
2. Shi, Y.-B. (Ed) (2013). Animal metamorphosis. Curr. Topics Dev. Biol. *103*, 2–440.
3. Johnson, G.D., Paxton, J.R., Sutton, T.T., Satoh, T.P., Sado, T., Nishida, M., and Miya, M. (2009). Deep-sea mystery solved: astonishing larval transformations and extreme sexual dimorphism unite three fish families. Biol. Lett. *5*, 235–239.
4. Fuchs, B., Wang, W., Graspeuntner, S., Li, Y., Insua, S., Herbst, E.M., Dirksen, P., Böhm, A.M., Hemmrich, G., Sommer, F., *et al*. (2014). Regulation of polyp-to-jellyfish transition in Aurelia aurita. Curr. Biol. *24*, 263–273.
5. Werner, B., Cutress, C.E., and Studebaker, J.P. (1971). Life cycle of *Tripedalia cystophora* Conant (Cubomedusae). Nature *232*, 582–583.
6. Kane, M.A. (2012). Analysis, occurrence, and function of 9-cis-retinoic acid. Biochim. Biophys. Acta *1821*, 10–20.
7. Kostrouch, Z., Kostrouchova, M., Love, W., Jannini, E., Piatigorsky, J., and Rall, J.E. (1998). Retinoic acid X receptor in the diploblast, Tripedalia cystophora. Proc. Natl. Acad. Sci. USA *95*, 13442–13447.
8. Law, J.A., and Jacobsen, S.E. (2010). Establishing, maintaining and modifying DNA methylation patterns in plants and animals. Nat. Rev. Genet. *11*, 204–220.
9. Goffin, J., and Eisenhauer, E. (2002). DNA methyltransferase inhibitors-state of the art. Ann. Oncol. *13*, 1699–1716.
10. Spangenberg, D.B. (1967). Iodine induction of metamorphosis in Aurelia. J. Exp. Zool. *165*, 441–449.
11. Spangenberg, D.B. (1971). Thyroxine induced metamorphosis in Aurelia. J. Exp. Zool. *178*, 183–194.
12. de Urquiza, A.M., Liu, S., Sjoberg, M., Zetterstrom, R.H., Griffiths, W., Sjovall, J., and Perlmann, T. (2000). Docosahexaenoic acid, a ligand for the retinoid X receptor in mouse brain. Science *290*, 2140–2144.
13. Bouton, D., Escriva, H., de Mendonça, R.L., Glineur, C., Bertin, B., Noël, C., Robinson-Rechavi, M., de Groot, A., Cornette, J., Laudet, V., *et al*. (2005). A conserved retinoid X receptor (RXR) from the mollusk Biomphalaria glabrata transactivates transcription in the presence of retinoids. J. Mol. Endocrinol. *34*, 567–582.
14. Grasso, L.C., Negri, A.P., Fôret, S., Saint, R., Hayward, D.C., Miller, D.J., and Ball, E.E. (2011). The biology of coral metamorphosis: molecular responses of larvae to inducers of settlement and metamorphosis. Dev. Biol. *353*, 411–419.
15. Müller, W.A. (1984). Retinoids and pattern formation in a hydroid. J. Embryol. Exp. Morph. *81*, 253–271.
16. Jägersten, G. (1972). Evolution of the Metazoan Life Cycle (London: Academic Press).
17. Nielsen, C. (1985). Animal phylogeny in the light of the trochaea theory. Biol. J. Linnean Soc. *25*, 243–299.
18. Wolpert, L. (1999). From egg to adult to larva. Evol. Dev. *1*, 3–4.
19. Davidson, E.H., Peterson, K.J., and Cameron, R.A. (1995). Origin of bilaterian body plans: evolution of developmental regulatory mechanisms. Science *270*, 1319–1325.

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