Regenerating the Model of Regeneration with Ctenophores

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Ramon-Mateu, J., Ellison, S. T., Angelini, T. E., and Martindale, M. Q. 2019. Regeneration in the ctenophore *Mnemiopsis leidyi* occurs in the absence of a blastema, requires cell division, and is temporally separable from wound healing. *BMC Biology*. 17:80. https://doi.org/10.1186/s12915-019-0695-8

Regeneration is common throughout the animal kingdom. There are four modes of regeneration: stem cell-mediated regeneration, where stem cells regrow lost structures or tissues; morphallaxis, where cells transdifferentiate into a different cell fate; compensatory regeneration, where differentiated cells divide to create new cells; and epimorphosis, where dedifferentiation forms an undifferentiated stem cell mass - a blastema (Barresi & Gilbert, 2020). Blastema formation is common for the regrowth of amputated structures (Ramon-Mateu et al., 2019). Another common aspect of regeneration is the reliance on cell proliferation (Barresi & Gilbert, 2020; Ramon-Mateu et al., 2019). Most animals use both tissue remodelling and cell proliferation for regeneration (Ramon-Mateu et al., 2019). Despite these common features of animal regeneration, mechanisms of regeneration differ among animal phyla.

Organisms from the phylum Ctenophora can regenerate their whole body; however, the molecular mechanisms driving this regeneration remain unknown (Ramon-Mateu et al., 2019). Ctenophores are crucial model organisms for investigating the evolution of animal regeneration based on their early phylogenetic branching position (Martindale, 1986). Ctenophore cydippids are emerging as new model organisms due to their small size and ease of visualization, while retaining the same regenerative abilities as adult ctenophores (Martindale, 1986). The study by Ramon-Mateu et al. (2019) explores wound healing and regeneration of ctenophores using cydippids of *Mnemiopsis leidyi*. Specifically, the study investigates four main inquiries regarding *M. leidy* regeneration: if a blastema-like structure forms for regeneration, whether cell proliferation is required for regeneration, what the source and nature of the contributing regenerative cells is, and what the role of wound healing in regeneration is. This study aims to discover molecular mechanisms of ctenophore regeneration in order to comprehend the global evolution of animal regeneration processes.

To observe the course of *M. leidyi* regeneration and determine if a blastema is formed during early regeneration, Ramon-Mateu et al. (2019) bisected the cydippids and labeled proliferating cells with ethynyl deoxyuridine (EdU). Surprisingly, they found that EdU+ cells are scarcely spread throughout the wound site early after amputation, at the time a blastema would typically form in other self-regenerating animals. Instead of a blastema forming, cells accumulated at the injury site and completed regeneration 48 hours post-injury (Focus Figure 1). Similarly, when the apical organ was amputated, cells gathered around the amputation site to differentiate into the new organ, without forming a blastema-like structure (Focus Figure 1).

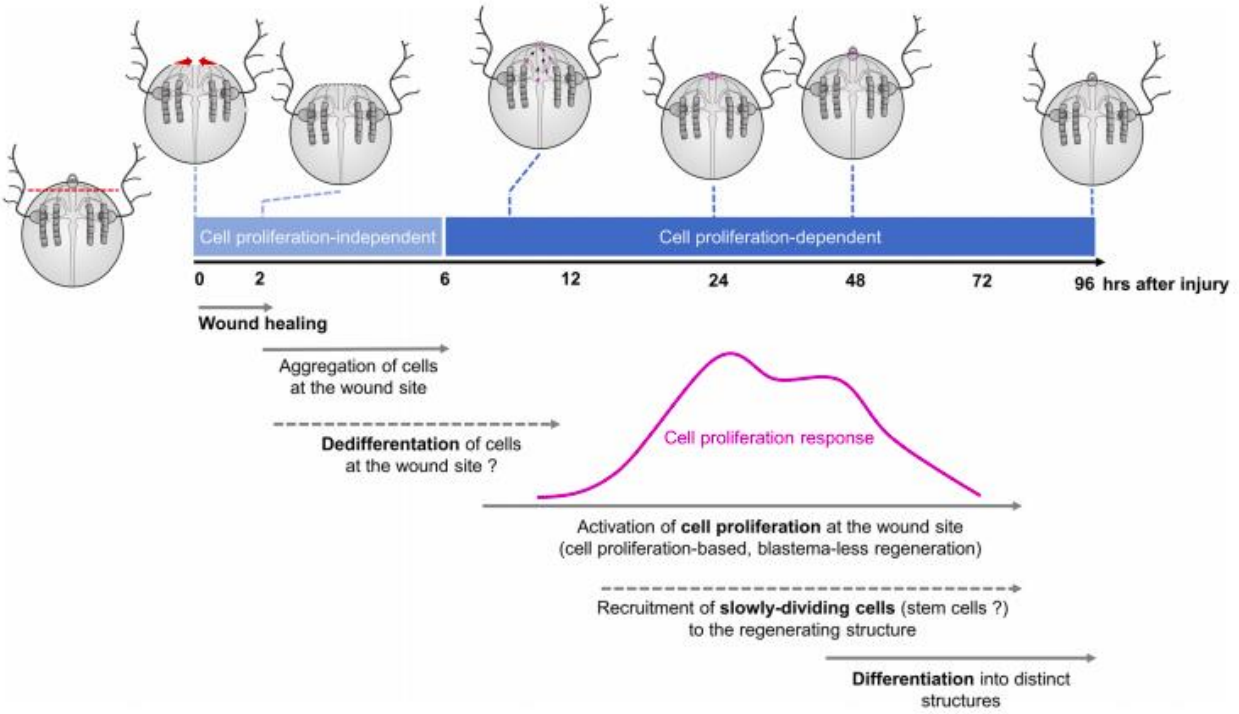
From these results, Ramon-Mateu et al. (2019) concluded that blastema formation is not needed for ctenophore regeneration.

To investigate whether cell proliferation is active during ctenophore regeneration, Ramon-Mateu et al. (2019) labeled actively dividing cells with EdU post-surgery. They found that, after amputation and bisection, cell proliferation is activated at the wound site (Focus Figure 1). They then analyzed whether this proliferation is necessary for regeneration by blocking proliferation with an inhibitor, hydroxyurea (HU), to halt cell proliferation during after surgery. Ramon-Mateu et al. (2019) found that with proliferation blocked, regenerative ability of cydippids was lost. Hence, they concluded that active cell proliferation is necessary for ctenophore regeneration. However, wound healing still occurred, suggesting both that wound healing and regeneration are independent events, and that wound healing is cell proliferation-independent (Focus Figure 1). They then washed the HU treatment off to restore proliferation and determine if this change is sufficient to regenerate the structures. Interestingly, regenerative ability was recovered, although not fully, when regeneration was unblocked - 38% of bisected cydippids regenerated fully, and 100% of apical organ-amputated cydippids regenerated the organ (Ramon-Mateu et al., 2019). Ramon-Mateu et al. (2019) concluded that cell proliferation is active during ctenophore regeneration, ctenophore regeneration is cell proliferation-dependent, and that the wound epidermis does not have a significant impact on the regulation of regeneration.

The source of regenerative cells varies across species, but often are supplied by discrete stem cell pools (Ramon-Mateu et al., 2019). To locate stem cell niches in the ctenophore body, Ramon-Mateu et al. (2019) labeled cydippids with EdU to track slowly dividing cells and found that EdU+ cells are localized in the tentacle bulbs, the apical organ, under comb rows, and in the pharynx. After amputation of the apical organ, EdU+ cells from these populations were found at the wound site (Focus Figure 1). Ramon-Mateu et al. (2019) concluded that these multipotent stem cells populations throughout the ctenophore body may be recruited to assist regeneration.

Ramon-Mateu et al. (2019) surveyed the cellular events during ctenophore wound healing and regeneration, and compared them with other previously studied organisms. They developed a model of ctenophore regeneration: Non-blastemal cell proliferation-dependent regeneration that recruits slowly dividing cells from throughout the body to the wound site. Non-blastemal cell proliferation-dependent regeneration is uncommon, but has been reported elsewhere, such as transdifferentiation in the lens of newts and compensatory proliferation of human livers (Ramon-Mateu et al., 2019). Further studies could examine whether the proliferating cells at the wound site are a result of dedifferentiation events, and examine the molecular signaling of the wound epidermis to investigate if it truly does not regulate regeneration. Interestingly, the ctenophore's lack of blastema formation contrasts with the majority of other known animal phyla with regenerative capabilities (Ramon-Mateu et al., 2019)

Further studies tracking cells and analyzing gene expression data during regeneration could investigate this difference and determine the conservation of blastema formation in evolution. Ramon-Mateu et al. (2019) offer an interesting and unique model of regeneration with their study, and demonstrate how vital invertebrates are to understanding the evolution of animal adaptation.



Focus Figure 1: Summary of the non-blastemal, cell proliferation-dependent model of ctenophore regeneration proposed by Ramon-Mateu et al. (2019). When a ctenophore is injured, cell proliferation-independent wound healing immediately commences and cells aggregate at the wound site to form the wound epidermis. Once regeneration begins, cells actively proliferate at the wound site and slowly-dividing cells from throughout the body are recruited to the wound site to reconstruct the structures, but this does not form a blastema-like mass. Cells are then able to differentiate and regenerate the structures.

Additional Literature Cited

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