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On mitosis in embryos and larvae of polyclads (Platyhelminthes)

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The epidermis of flatworms is thought to be maintained by cells arising in the parenchyma (1-3). The taxon Catenulida is the only platyhelminth taxon with documented intraepidermal mitoses (4). On the other hand, in the gastrodermis, undifferentiated or proliferating cells are observed in many taxa of the flatworms (5-12), although they are not found in the Tricladida (13) and have not been studied in a number of other taxa of the Platyhelminthes.

There are only fragmentary data on the spatial arrangement of cell proliferation in the epidermis and gastrodermis in the early ontogenetic stages of flatworms. The question of particular interest is at which ontogenetic stage the final localization of stem cells is determined. Comparative data on mitotic activity during ontogeny are required for better understanding of tissue evolution in the flatworms.

We studied cell division in intact embryos and larvae of two polyclad species, *Notoplana humilis* (Stimpson, 1857) (development with an adult-like larva) and *Cycloporus japonicus* Kato, 1927 (development with a Müller's larva). Embryos at the stage of organogenesis (16 and 11 specimens, respectively) and larvae (9 and 15 specimens, respectively) were examined in series of stained paraffin sections.

In the embryos of *N. humilis* during early organogenesis, ectodermal mitoses were rare (~0.5 mitoses/embryo); in late organogenesis and in the larvae no mitotic figures were found in the ectoderm, with the exception of one dubious case. In *C. japonicus* ectodermal mitoses were observed both in the embryos of all stages of organogenesis and in the Müller's larvae (~1 mitosis/specimen). Both in the embryos and the larvae of each species mitotic figures were absent in the intestine, but occurred in mesodermal (mesenchymal) cells.

It has been shown for adult and juvenile polyclads that cell proliferation takes place in the parenchyma and gastrodermis but is absent in the epidermis (14, 15, 11, 12). The above mentioned and present studies support the hypothesis that in polyclads the formation of definitive epidermis is associated with the disappearance of mitotic cells in the integument, which occurs at different stages of morphogenesis. The time of this disappearance seems to depend on the type of development of the polyclad. On the other hand, formation of the definitive gastrodermis is linked with the appearance of mitotic cells in the intestine upon isolation from the parenchyma.

During organogenesis, ectodermal mitoses are known for only two turbellarian species, *Macrostomum appendiculatum* (Macrostomida) and *Minona trigonopora* (Proseriata) (16, 17). The current study has shown that a polyclad with a Müller's larva maintains mitotic cells in the ectoderm not only during embryogenesis, but also in the larva. Thus, *C. japonicus* presents the first example of a turbellarian with ectodermal mitotic activity in the late stages of morphogenesis. Since the existence of intraepidermal mitotic cells, which has been observed only for the Catenulida, may be considered as a plesiomorphic feature for the plathelminths (4), the ectodermal mitosis in Müller's larvae may indicate that the Müller's larva is plesiomorphic for this character.

Our conclusions can be supported by (*a*) analysis of the accumulation of mitotic figures resulting from blocking of mitoses in embryos/larvae and (*b*) more precise identification of cells as being "mitotic" using a suitable label.

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REFERENCES

1. EHLERS, U. (1985). *Das phylogenetische System der Plathelminthes*. G. Fischer, Stuttgart, N.Y. (317 pp).
2. SMITH, J.P.S., S. TYLER & R.M. RIEGER (1986). Is the Turbellaria polyphyletic? *Hydrobiologia*, 132: 13-21.
3. DROBYSHEVA, I.M. (1991). **C**ambiality of epidermis in the turbellaria. *Proceedings of the Zoological Institute of the USSR AS*, 241: 53-87.
4. EHLERS, U. (1992). No mitosis of differentiated epidermal cells in the Plathelminthes: mitosis of intraepidermal stem cells in *Rhynchoscolex simplex* Leidy,1851 (Catenulida). *Microfauna Marina*, 7: 311-321.
5. PULLEN, E.W. (1957). A histological study of *Stenostomum virginianum*. *J. Morph.*, 101: 579-621.
6. BORKOTT, H. (1970). Geschlechtliche Organisation, Fortpflanzungsverhalten und Ursachen der sexuellen Vermehrung von *Stenostomum sthenum* nov. spec. (Turbellaria, Catenulida). Mit Beschreibung von 3 neuen *Stenostomum*-Arten. *Z. Morph. Tiere*, 67: 183-262.

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7. Bennet, C.E. (1975). *Fasciola hepatica*: development of caecal epithelium during migration in the mouse. *Experimental Parasitology*, 37: 426-441.
8. Reuter, M. & I. Palmberg (1987). An ultrastructural and immunocytochemical study of gastrodermal cell types in *Microstomum lineare* (Turbellaria, Macrostomida). *Acta Zoologica* (Stockholm), 68 (3): 153-163.
9. Palmberg, I. (1991). Differentiation during asexual reproduction and regeneration in a microturbellarian. *Hydrobiologia*, 227: 1-10.
10. Bogitsh, B.J. (1993). A comparative review of the flatworm gut with emphasis on the Rhabdocoela and Neodermata. *Transactions of the American Microscopical Society*, 112 (1): 1-9.
11. Drobysheva, I.M. (1997). Cell proliferation in *Notoplana humilis* (Polycladida, Plathelminthes). *Doklady Biological Sciences*, 353 (1): 128-130.
12. Drobysheva, I.M. (1999). Cell proliferation in the ontogeny of polyclads (Platyhelminthes). *Proceedings of the Zoological Institute RAS*, 281: 79-85.
13. Hori, I. (1986). Ultrastructural observations on gastrodermal regeneration in the planarian *Dugesia japonica* (Turbellaria). *Hydrobiologia*, 132: 217-222.
14. Drobysheva, I.M. (1987). A study of the physiological regeneration of epidermis in the polyclad Notoplana humilis using the technique of thymidine autoradiography. *Proceedings of the Zoological Institute of the USSR AS*, 167: 90-96.
15. Drobysheva, I.M. (1988). An autoradiographic study of the replacement of epidermis in polyclad turbellarians. *Fortschritte der Zoologie*, 36: 97-101.
16. Sailern-Aspang, F. (1957). Die Entwicklung von *Macrostomum appendiculatum* (Fabricius). *Zoologische Jahrbücher. Abteilung für Anatomie und Ontogenie der Tiere*, 76: 311-330.
17. Reisinger, E., I. Cichocki, R. Erlach & T. Szyskowitz (1974). Ontogenetische Studien an Turbellarien: ein Beitrag zur Evolution der Dotterverarbeitung im ektolecitalen Ei. I. Teil. II. Teil. *Zeitschrift für zoologische Systematik und Evolutionsforschung*, 12: 161-195; 241-278.