DEVELOPMENTAL BIOLOGY

Inducing concentric worm holes

Robert K. Herman

INDUCTION is the process in development in which the fate of one cell mass is determined by another. A simple example occurs during vulval development in the nematode *Caenorhabditis elegans*: a gonadal cell called the anchor cell induces three neighbouring cells to embark on a programme of cell division and morphogenesis, which culminates, in a few hours, in the formation of a vulva[1–5]. On page 470 of this issue, Hill and Sternberg[6] report strong evidence that they have identified the anchor-cell signalling molecule, which they find is a member of the EGF (epidermal growth factor) group of growth factors.

The vulva of *C. elegans* is formed during the late larval stages of hermaphrodite development. Each hermaphrodite produces oocytes which can be fertilized either by sperm manufactured internally or by sperm injected by a copulating male. The function of the vulva, then, is twofold: it provides an entrance for male sperm and an exit for fertilized eggs.

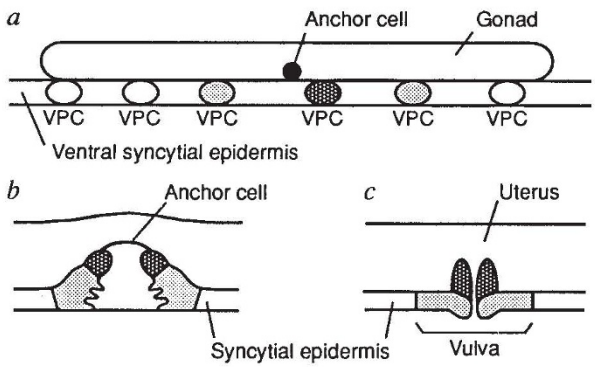
Each of six cells situated in a longitudinal row on the ventral side of the hermaphrodite is capable of generating descendants that contribute to the vulva. These six cells are called the vulval precursor cells (VPCs). The name, though, is slightly misleading because normally only three VPCs make a vulva, the remaining three taking an alternative developmental path: they divide once and then fuse with a large multinuclear epidermis that surrounds them and the developing vulva, and covers most of the worm's body except for the head and tail. The three VPCs selected for vulval development are those closest to the anchor cell; together they generate 22 descendants that undergo cell fusions and intricate morphogenetic movements to produce a vulva (see figure). The evidence that the anchor cell induces vulval development comes from experiments in which it has been removed, either by laser ablation or by mutation. All six VPCs then fuse with the surrounding syncytial epidermis and no vulva is formed.

Normally the same three VPCs are chosen for vulval development because they are invariably closest to the anchor cell. However, the ability of the other three to contribute to vulval development has been demonstrated by experiments in which a normal vulval-producing VPC has been destroyed by a laser microbeam or the relative positions of the VPCs and the anchor cell have been changed by mutation; in these cases, a vulva may be formed from a different set of three VPCs. In addition to producing the inductive signal, the anchor cell becomes the focus of vulval morphogenesis and ensures that the vulval and uterine openings are concentric.

Hill and Sternberg[6] propose that the anchor-cell signal is the product of the *lin-3* gene, mutations in which lead to animals with VPC cell lineage (hence *lin*) defects: all six VPCs behave as if no anchor cell signal were sent, and no vulva is formed. The authors present five lines of evidence for their proposal. First, *lin-3* was cloned and shown to encode an EGF-like growth factor; this would be an appropriate ligand for what had previously been proposed as the receptor for the anchor-cell signal, an EGF-receptor tyrosine kinase produced by the gene *let-23* (ref. 7). Second, a multicopy array of wild-type *lin-3* genes in transgenic animals was shown to induce vulval development by all six VPCs, presumably because of overexpression of the inducing signal by the anchor cell. Third, the vulval overinduction caused by the multiple copies of *lin-3* was suppressed by mutation in *let-23*. Fourth, the overinduction was also suppressed by destruction of the gonad, including the anchor cell. Finally, a *lin-3–lacZ* multicopy transgenic array produced its fusion protein almost exclusively in the anchor cell — and not in the VPCs or surrounding epidermis. These results constitute a firm case for the authors' supposition that *lin-3* produces the signalling molecule in the anchor cell.

Here, as in much recent work, it is exhilarating to see vertebrate protein motifs turn up in a modest creature where questions of function can be readily addressed. Oncogenic motifs were discovered earlier in the worm from studies of *let-60*, which encodes a Ras protein[4,8], and *sem-5*, which has *src* homology regions[9]; both of these genes act downstream of *let-23* in the vulval developmental pathway. Indeed, the worm also uses the products of *lin-3*, *let-23*, *let-60* and *sem-5* in other developmental pathways. Because null alleles of each of these genes result in early larval lethality, the elucidation of the roles of these genes in vulval development depended on the ingenious use of leaky mutations that preferentially affect vulva formation. The use of other alleles and other approaches, perhaps including genetic mosaics, should promote our understanding of what other roles these genes have in development; these would presumably also involve intercellular communication.

The story of vulval development will soon become even richer in molecular detail. Mutations in more than 30 genes result in aberrant vulval development; these genes and their mutations will continue to make wonderful grist for the cloner's mill. Some of the genes have been implicated in the transmission or reception of additional intercellular signals[3–5], one of which is a lateral inhibitory signal between adjacent VPCs; its receptor is probably the product of the well-studied gene *lin-12* (refs 3,5,10), but the signal itself is unknown. Another great unknown is the molecular basis of the beautiful vulval morphogenesis that ultimately results from the sending and receiving of the anchor-cell signal. □



The three vulval precursor cells nearest the gonadal anchor cell are induced to generate 22 descendants that undergo cell fusions and morphogenesis to produce a vulva. *a*, Third larval stage; *b*, fourth larval stage; *c*, adult. Patterns of shading in *b* and *c* correspond to regions contributed by ancestral VPCs shown in *a*. (Adapted from refs 3 and 4.)

Robert K. Herman is in the Department of Genetics and Cell Biology, University of Minnesota, St Paul, Minnesota 55108, USA.

1. White, J. in *The Nematode* Caenorhabditis elegans (ed. Wood, W. B.) 81–122 (Cold Spring Harbor Laboratory, New York, 1988).
2. Sulston, J. in *The Nematode* Caenorhabditis elegans (ed. Wood, W. B.) 123–155 (Cold Spring Harbor Laboratory, New York, 1988).
3. Horvitz, H. R. & Sternberg, P. W. *Nature* **351**, 535–541 (1991).
4. Sternberg, P. W. & Horvitz, H. R. *Trends Genet.* **7**, 366–371 (1991).
5. Lambie, E. J. & Kimble, J. *A. Rev. Genet.* **25**, 411–436 (1991).
6. Hill, R. J. & Sternberg, P. W. *Nature* **358**, 470–476 (1992).
7. Aroian, R. V., Koga, M., Mendel, J. E., Ohshima, Y. & Sternberg, P.W. *Nature* **348**, 693–699 (1990).
8. Han, M. & Sternberg, P. W. *Genes Dev.* **5**, 2188–2198 (1991).
9. Clark, S. G., Stern, M. J. & Horvitz, H. R. *Nature* **356**, 340–344 (1992).
10. Greenwald, I. & Seydoux, G. *Nature* **346**, 197–199 (1990).