

### Hæmal System of Regular Echinoids

EXAMINATION of accounts of the echinoid hæmal system shows that the term 'hæmal' has never been really justified: its function has not been conclusively demonstrated, and the precise nature of the movement of fluid within the system as a whole has never been satisfactorily established.

Uncertainty even as to the gross anatomy of the system is well illustrated in the instance of the radial hæmal sinuses. Thus Perrier<sup>1</sup> did not describe them, Chadwick<sup>2</sup> could not demonstrate them and doubted their existence. Cuénot<sup>3</sup> affirmed their existence, but admitted that they could not be injected.

Tiedemann<sup>4</sup> first noted the contractile nature of the hæmal vessels. Perrier<sup>1</sup> noted that some vessels of *Echinus esculentus* showed non-rhythmical contraction and thought that the direction of the resultant hæmal flow was not constant. Farmanfarmaian and Phillips<sup>5</sup> could not detect any fluid movement in the hæmal system of *Strongylocentrotus purpuratus*, although a rhythmic beat was observed in the outer sinus (external marginal vessel) and in the collateral sinus of the stomach. Stott<sup>6</sup> considered that, in the absence of a pumping mechanism, distribution of nutrients was effected by migration of amœbocytes within the hæmal system. Boolootian and Campbell<sup>7</sup> have described a contractile chamber and pulsating vessel associated with the axial gland of *S. purpuratus*. They consider that fluid can enter the contractile chamber by two routes, the inner hæmal sinus and also by microscopic ostia. The abstract of the paper states that a pulsating vessel and a compartmented contractile chamber were found to move cœlomic fluid from the perivisceral cavity into and throughout the hæmal system of the urchin. This statement is not fully justified in the account. The authors also state that the rhythmic contraction of the pulsating vessel together with the pulsations of the stone canal may be important in moving fluids through the entire water vascular system.

I have examined the so-called hæmal system of *Echinus esculentus* and *Psammechinus miliaris*. Sea-water contamination was minimized as this causes rapid deterioration of the internal tissues. I can confirm that the collateral and internal marginal vessels of the stomach of both species are contractile, rates of 10 contractions per 60–90 sec being recorded for the internal marginal vessel and 10 contractions per 45–180 sec for the collateral vessel in *P. miliaris*. Particulate matter, principally clumped amœbocytes, was observed through the walls of the vessels. The particles are small and difficult to observe in fresh preparations.

Despite evidence of contractility its significance is not yet clear. First, although some vessels showed a patent lumen (for example, collateral, internal and external marginal vessels of the stomach region), some, such as the peri-œsophageal hæmal ring and the œsophageal vessel, did not, being largely occluded by spongy tissue. Secondly, the particulate contents of the vessels showed ebb-and-flow rather than uni-directional movement, although a net transfer in their position was sometimes achieved. The most usual pathway was as follows: material passed from the œsophageal to the intestinal end of the internal marginal vessel at a rate of about 15 mm/min, and passed along the collateral vessel towards the œsophagus at a similar rate. Net transfer of material from the intestinal to the œsophageal end of the internal marginal vessel simultaneous with collateral flow towards the intestine was also observed but simultaneous flow in both vessels towards the œsophagus or the intestine was not observed.

Pathways available to injected dyes were investigated: it was not possible to inject the œsophageal vessel or the peri-œsophageal hæmal ring as a lumen could not be detected. Neither was it possible to inject the collateral vessel due to its delicate suspension and extreme con-

tractility. Injection into the internal marginal vessel showed that dye passes readily into the collateral vessel when injected from either end of the vessel towards the other, but there is resistance to injection towards the œsophagus. Material injected towards the œsophagus may pass into the origin of the œsophageal vessel, and, similarly, material injected towards the intestine may pass a short way along the external margin of the intestine.

It is concluded that it is unlikely that in these species the so-called hæmal system functions as a true circulatory system. However, there is undoubtedly active movement, and possibly a more restricted circulation, within the vessels of the stomach region. Recent work<sup>5,6</sup> has suggested that absorbed nutrients pass rapidly to the cœlomic fluid. Preliminary experiments, in which the presence of radioactive material was sought in the hæmal vessels after oral injection of isotope-labelled substances in *P. miliaris*, suggest that the hæmal vessels around the stomach may function in absorption, as previously surmised<sup>1,9</sup>. Perhaps they may assist passage of extracted nutrients to the cœlomic fluid, both 'extraction' and diffusion being assisted by the oscillation of fluid within these vessels.

This investigation was conducted, under the supervision of Prof. N. Millott, during the tenure of a Department of Scientific and Industrial Research studentship.

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<sup>1</sup> Perrier, E., *Arch. Zool. exp. gén.*, **4**, 605 (1875).

<sup>2</sup> Chadwick, H. C., *Liverpool Marine Biol. Comm.*, **Mem. III** (1900).

<sup>3</sup> Cuénot, L., in *Traité de Zoologie*, edit. by Grassé, P. P., **2** (Paris: Masson, 1948).

<sup>4</sup> Tiedemann, F., *Anatomie der Röhren-Holothurie des pomeranzfarbigen Seesterns und Stein-seegels*. (Landshut: Joseph Thomannschen Buchdruckerei, 1816).

<sup>5</sup> Farmanfarmaian, A., and Phillips, J. H., *Biol. Bull., Woods Hole*, **123**, 105 (1962).

<sup>6</sup> Stott, F. C., *Proc. Zool. Soc., Lond.*, **125**, 63 (1955).

<sup>7</sup> Boolootian, R. A., and Campbell, J. L., *Science*, **145**, 173 (1964).

<sup>8</sup> Lasker, R., and Glese, A. C., *Biol. Bull., Woods Hole*, **106**, 328 (1954).

<sup>9</sup> Bonnet, A., *Ann. Inst. océano., Monaco*, Sér. **2**, 209 (1925).

### Effects of Histones on Embryonic Development

I HAVE recently examined in detail the effects of actinomycin D, an inhibitor of RNA polymerase and of messenger RNA synthesis, on the development of amphibian eggs<sup>1,2</sup>. It is now known that histones also inhibit RNA polymerase<sup>3,4</sup> and it has been suggested that they could act as the natural regulators of genetic activity<sup>5,6</sup>; by combining with specific segments of DNA molecules, they would block the production of messenger RNAs by these segments. It has been suggested<sup>7-9</sup> that such a mechanism could account for the 'switching on and off' of gene activity during embryonic development. It was of interest to compare the effects of exogenous, abnormal histones on embryonic development with those of actinomycin D. The latter are quite different from those of another basic protein, ribonuclease, which quickly stops cleavage in amphibian eggs<sup>10</sup>, while actinomycin D has no effect prior to gastrulation.

The lysine-rich fraction of thymus histones, prepared by the method of Allfrey *et al.*<sup>4</sup>, was used in the present work. Three types of experiments were performed on eggs of *Pleurodeles* at various stages of their development: addition of histones to the medium, replacement of the blastocoele fluid by a histone-containing solution and micro-injection of histones in just fertilized eggs.

(1) *Addition of histones (3 to 100 µg/ml.) to external medium.* No immediate effect on cleavage could be detected, even at the highest concentrations: when early morulae or late blastulae are treated with histones (10–90 µg), development stops at the gastrula stage. A very conspicuous effect of histones (10–30 µg/ml.) is