Mouth and Blastopore

By the Haeckelian view of early development, gastrulation by invagination reflects the evolutionary history of primitive Metazoa. It is a corollary of this theory that gastrulation by ingression is a derived form of emboly. Nevertheless ingression, the method of endoderm formation among the lower cnidarians, is probably the more primitive mode of gastrulation, as observed by Metschnikoff[1,2], Hyman[3,4] and others. The diploblastic level of organization probably arose in early Metazoa by a process resembling ingression, or some other type of delamination, giving rise to solid planula-like forms. It is therefore probable that invagination is merely a developmental expedient, which has arisen secondarily and independently in the more advanced Cnidaria and other metazoan groups.

The blastopore results from invagination, narrowing as gastrulation progresses. In keeping with Haeckel's interpretation, it is generally thought to represent the primitive mouth. It may nevertheless be merely a product of the mechanics of development, without especial phylogenetic significance. The Cnidaria in which gastrulation occurs by ingression, while possessing a mouth and cœlenteron, have no blastopore or archenteron during development. It appears from the embryology of these cœlenterates that mouth and blastopore were independently acquired in early Metazoa. Furthermore, a mouth may have formed before the origination of embolic gastrulation and a blastopore. Following the evolution of embolic gastrulation, the blastopore preceded the mouth in embryonic development, for it relates to the fundamental process of germ-layer formation. Similarly, it appears that enteron and archenteron were independently evolved. Terms such as protostoma, Protostomia and archenteron are therefore strictly inapt.

The mouth in some non-cœlomate animals and Protostomia does not coincide with the blastopore or its point of closure. Also, in many higher cœlenterates, non-cœlomate groups and Protostomia the blastopore does not persist as a mouth, but closes before the mouth forms. This possibly signifies the evolutionary independence of the two structures.

It is generally agreed that the embryonic axis corresponds with the polar axis of ancient Metazoa, the animal pole representing the primitive perceptive and dominant region, directed forward in locomotion. Biologists who support the ingression theory, but identify the blastopore with the primitive mouth, believe that the mouth arose at the more inert end in a solid, diploblastic ancestral metazoan. This entails the puzzling conception of a structure for the selection and ingestion of food separated from the dominant, sensitive region of the body. It seems more probable that the mouth originally occupied an antero-ventral position.

In cœlenterates the mouth typically forms at the posterior end of the swimming larva, in the blastoporal region when gastrulation is by emboly, and does not change position during development. This may be a special feature correlated with the sessile habit of the polyp.

Typically, in cases of gastrulation by invagination, the gastrula presents two centres of high metabolic activity of different kinds: the animal pole, corresponding with the controlling and perceptive region of original Metazoa and larval forms; and the blastopore, a zone of rapid cell division, and a focus of developmental activity and organization. Among the Protostomia, the mouth arises in the blastoporal field, later coming to occupy an antero-ventral position. The mouth thus develops in a region of high formative activity, and may form from the blastopore, a ready-made aperture. This may be merely a developmental convenience. (By a similar short cut in development, in some Protostomia and Deuterostomia a persistent blastopore forms the anal opening.) In the embryonic development of many annelids, the blastopore elongates and constricts into two. One of the apertures so formed, the mouth, separates to its definitive ventral anterior position. The second aperture, remaining at the vegetal pole, soon closes. The anus later arises at this point. The division of the blastopore has been interpreted as separation of the mouth from a transitory anal pore. It more probably represents dissociation of the mouth from the blastopore.

Among the Deuterostomia, the mouth typically originates at an antero-ventral point. This is believed to indicate that the mouth has evolved anew and assumed the function of the primitive mouth, homologous with the blastopore. It is more probable that the mouth in the Deuterostomia occupies the original position, and the blastopore at no time functioned as mouth.

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[3] Hyman, L. H., "The Invertebrates". 1. "Protozoa through Ctenophora" (New York and London, 1940).
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Softening Chitin for Histology

The chitinous integument of many invertebrates consists not of chitin alone but also incorporates much structural protein. In the more highly sclerotinized arthropod shells, indeed, this may far exceed the true chitin. The methods frequently used for softening 'chitin' for histological purposes make use of this fact, and none of them actually alters the chitin itself, acting instead on the protein moiety of the integument. 'Diaphanol', for example, the most widely used agent for softening 'chitin', acts by breaking benzene rings in the aromatic amino-acid residues of the proteins[1]. Inevitably such an action is accompanied by extensive damage to the tissues of the specimen, for not only is the protein of the skeleton digested but also the proteins of the tissues which we wish to examine. The use of diaphanol, or of other agents which digest the protein moiety of the chitinous integument, is therefore incompatible with precise histological or cytological investigation. If the integument consists mainly of structural protein, then we have no way out of the dilemma except to dissect the tissues free from all traces of the offending shell. If, however, as in the less-sclerotinized shells, it consists chiefly of true chitin, then it is possible to attack the chitin enzymically, leaving the tissues untouched.

Possibly the best source of a suitable chitinase is the unripe puff-ball, *Lycoperdon* spp. Tracey[2] advocates this source of the enzyme for biochemical analysis. He suggests that the puff-balls should be torn up and pressed to yield a juice which, when buffered at *p*H 5 with acetate buffer, will readily digest chitin. This juice may be stored for a year or more in a refrigerator under toluene. I have used