THE FEEDING MECHANISM AND STRUCTURE OF THE GUT OF *OWENIA FUSIFORMIS* DELLE CHIAJE

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(Text-figs. 1 and 2)

In 1901 Arnold Watson published a paper on the habits of *Owenia fusiformis* delle Chiaje (= *Ammochares filiformis* Watson) in his now classic series of papers on the tube-building habits of various polychaetes. There is little to be added to his description in this respect, but Watson was not concerned with the feeding mechanism, which receives but brief mention. *O. fusiformis* has previously been studied by a number of workers, but none has given much attention to the morphology of the gut or to the method of feeding which are the main considerations of the present paper.

O. fusiformis has been studied from other aspects in some detail, notably by von Drasche (1885), who, while dealing little with the gut, did note that the mid-gut region has a green colour. Claparède (1868, 1873) and Saint-Joseph (1898) also mention this. Some account of other features of the morphology of *Owenia* will be found in the papers of Gilson (1893, 1897), Lo Bianco (1893), Eisig (1887) and McIntosh (1915), while the development and metamorphosis of the mitraria larva belonging to this species is described in the well-known paper of Wilson (1932).

O. fusiformis is not uncommon in clean sand below low water of neap tides on British shores, often in beaches subject to some disturbance and not tenable by polychaetes with a permanently fixed tube or burrow. Though local in occurrence, the species is cosmopolitan in distribution. The worms studied in the present work were collected from Tor Abbey sands, Torquay, Devon, and from near St Mawes in Cornwall. Elwes (1910) first reported *Owenia* as the commonest polychaete in Tor Abbey sands; Wilson collected worms from this beach for his developmental studies (1932), and it is certainly still numerous there. The worms construct long flexible tubes up to 15–20 cm long and 2–5 mm across, consisting of flat grains arranged like roof tiles with the free edge upwards; white or lightly coloured grains seem to be preferred, even when relatively scarce, so that the tubes are often conspicuous, as in the reddish sand of Torquay. The flexibility and probably also the arrangement of the sand grains enables the worm to work the tube up and down in the sand and to rebury itself when necessary. Worms can withdraw the tube completely

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beneath the surface, but when feeding in the laboratory the tube may project 2–3 cm above the sand. Possibly the tubes do not project as much as this in nature. *Owenia* is very hardy, as Watson (1901) found, and will live in the laboratory for long periods without aeration.

STRUCTURE OF THE CROWN AND THE FEEDING MECHANISM

The tip of the tube forms an elastic cone of transparent material which almost closes when the worm retracts; during feeding the crown and the most anterior segments are pushed through the minute opening at the tip of the cone which is then distended, and worms when undisturbed may remain for long periods in this position. On other occasions the crown may be seen within the cone, but it sems unlikely that feeding takes place to any significant extent at this time, though some respiratory exchange may continue. The cone is probably of great protective value when the worm is moving within the tube through the sand. The crown has an elaborate blood supply, and is light red, brown or greenish in colour; while there is no doubt that it is respiratory in function, it also participates in feeding.

The crown is a bilaterally symmetrical, branched structure developed from the prostomium, and has some mobility. Though short it is twice the diameter of the body when expanded (Fig. 1). There are four main branches or units on each side (Fig. 1 A), and each unit (Fig. 1 B) is divided into 4–6 branches, each of which ends in 2–4 small bifid lobes (Fig. 1 C). While there is some variation in arrangement, these lobes are almost always arranged in pairs, strongly ciliated on their oral surface, with the stem of each pair raised at the edges to form a gutter (Fig. 1 C). There are long cilia on these edges which beat inwards towards the axis of the stem, and shorter cilia in the gutter itself which beat towards the mouth. The long cilia on the edges of the gutter cause swirling movements between adjacent branches sweeping particles into the crown. Particles which are too large to be ingested usually do not come to rest in the gutter at all, or, if arrested, collect along the edges and are eventually cleaned off by muscular movements, either by convulsive movements of the whole crown, or by more local movements. Cilia are entirely absent from the aboral surface of the crown, but mucus gland cells are distributed over both oral and aboral surfaces.

The crown forms a funnel at the base closed by three lips, a single dorsal lip, and two ventro-lateral in position. These lips are all expansions of the prostomium, and when dilated close the mouth by coming together at the centre (Fig. 1 A). They are hollow and extremely mobile; dilation is caused by the inflow of coelomic fluid, but each lip has a complicated musculature and is capable of a variety of rippling movements. The crown contains an extensive coelomic cavity. Closure of the crown is brought about by the circular crown muscles acting against the hydrostatic pressure of the body fluid; when

the crown is expanded these crown muscles are relaxed (Fig. 1B). These circular muscles extend into all the branches except into the bifid tips, so that each gutter is capable of a certain amount of inrolling which can assist both in directing the strings of particles towards the mouth, and in freeing the crown of unwanted matter. While convulsive closure of the whole crown takes place
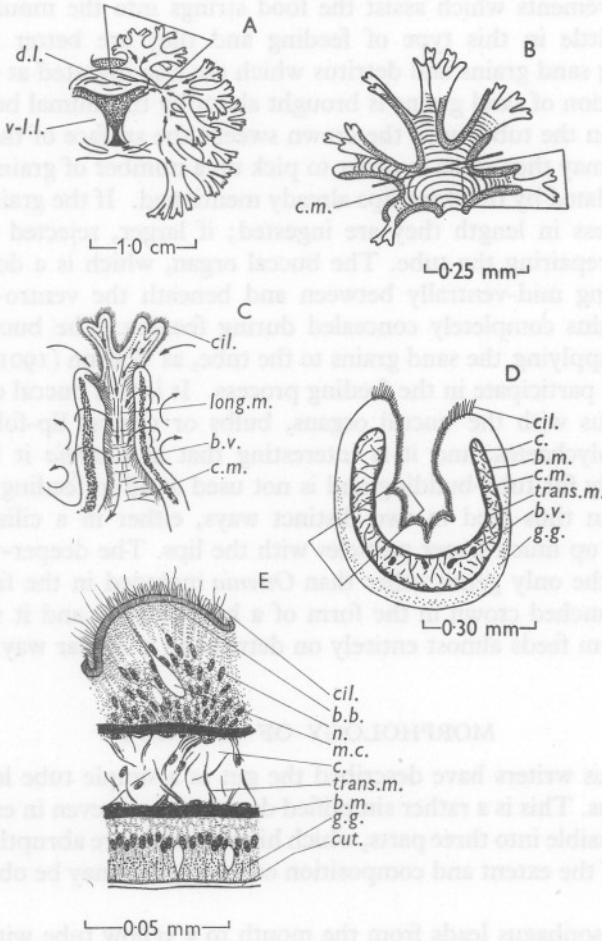

Fig. 1. The crown and feeding mechanism. A, the left half of the crown seen from above; arrows indicate direction of ciliary currents; the area marked is shown in B. B, a single unit showing the gutter and circular muscles; the area marked is shown in C. C, a single lobe showing arrangement of cilia and direction of ciliary currents. D, section through a unit corresponding to the plane of the line shown in A; the area marked is shown in E. E, section through the axis of a unit. *b.b.* brush-border; *b.m.* basement membrane; *b.v.* blood vessel; *c.* coelom; *c.m.* circular muscle; *cil.* cilia; *cut.* cuticle; *d.l.* dorsal lip; *g.g.* granules of green pigment; *long.m.* longitudinal muscle; *m.c.* mucus gland cell; *n.* nuclei; *trans.m.* transverse muscle; *v-l.l.* ventro-lateral lip.

at intervals during feeding, different parts of the crown are capable of inde-
pendent movement. There are also muscle fibres across the coelomic space
between aboral and oral surfaces (Fig. 1 D, E) and longitudinal fibres in the
bifid tips (Fig. 1 C).

Ciliary feeding, which is continued with the worm in a more or less erect
posture may continue for many hours. During this time the lips may undergo
rippling movements which assist the food strings into the mouth, but they
participate little in this type of feeding and they are better adapted for
manipulating sand grains and detritus which are also ingested at other times.

The ingestion of sand grains is brought about by the animal bending over,
usually within the tube, until the crown sweeps the surface of the sand. The
crown itself may then close in order to pick up a number of grains, which are
then manipulated by the three lips already mentioned. If the grains are about
0·2 mm or less in length they are ingested; if larger, rejected or used for
building or repairing the tube. The buccal organ, which is a double-lipped
structure lying mid-ventrally between and beneath the ventro-lateral lips,
usually remains completely concealed during feeding. The buccal organ is
adapted for applying the sand grains to the tube, as Watson (1901) described,
and does not participate in the feeding process. It is this buccal organ which
is homologous with the buccal organs, bulbs or ventral lip-folds in other
sedentary polychaetes, and it is interesting that in *Owenia* it has become
adapted solely for tube-building and is not used at all in feeding.

Owenia can thus feed in two distinct ways, either in a ciliary manner,
or by taking up much larger particles with the lips. The deeper-water genus
Myriochele, the only genus other than *Owenia* included in the family, has a
simple unbranched crown in the form of a hollow cone, and it seems likely
that this worm feeds almost entirely on detritus in a similar way.

MORPHOLOGY OF THE GUT

Most previous writers have described the gut as a simple tube leading from
mouth to anus. This is a rather simplified description, as even in external view
the gut is divisible into three parts, which histologically are abruptly separated.
Some idea of the extent and composition of these parts may be obtained from
Fig. 2 A–D.

A short oesophagus leads from the mouth to a yellow tube with thick and
much involuted walls. Farther back the gut assumes a deep green colour, and
this region has somewhat thinner walls and is less infolded; between the level
of the 5th and 6th pairs of tori the green colour diminishes, and the hind gut
is very thin-walled and capable of great distension.

The floor of the crown has ciliated tracts leading down into the mouth as
already described. The whole of this buccal region is ciliated as is the oeso-
phagus, the tall epithelial cells of which are characterized by a brush-border,

long cilia, finely granular cytoplasm and elongate nuclei (Fig. 2E). Basal granules beneath the brush-border and converging fibres characteristic of cells of this kind may be seen leading from the granules towards the middle of the cell where the nucleus lies. Gland cells, staining blue with Azan, occur in the epithelium of the buccal region and in the oesophagus, between the
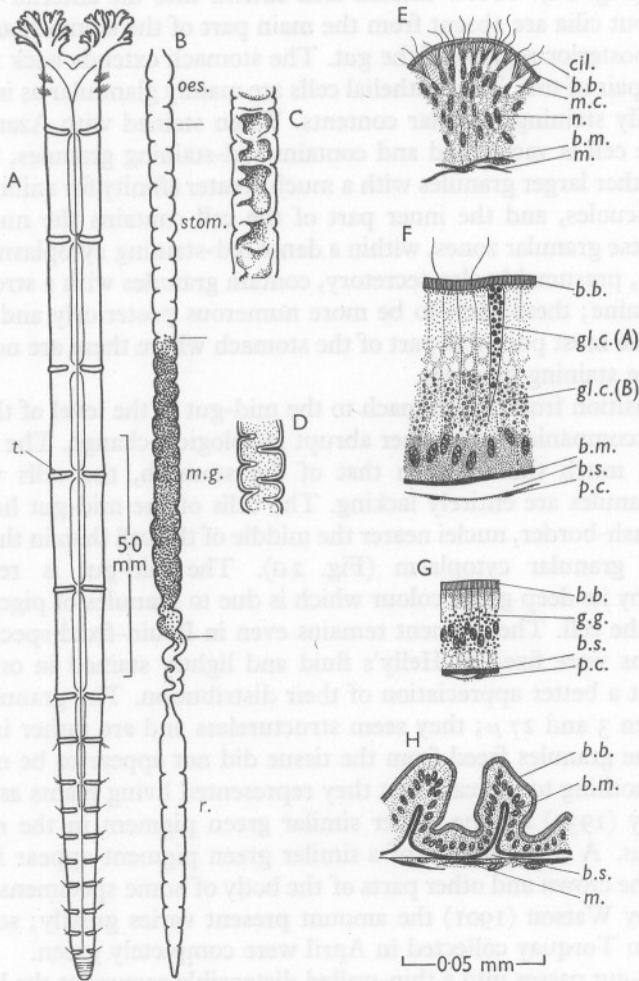

Fig. 2. The morphology and histology of the gut. A, the whole worm in ventral view. B, external appearance of the whole gut; anterior part of rectum is empty, the hinder part filled. C, part of the stomach showing thick walls and involutions. D, part of the mid-gut showing the thin walls and simpler infoldings. E–H, transverse sections of the oesophagus (E), stomach (F), mid-gut (G) and rectum (H), all drawn to the same scale. Abbreviations not used in Fig. 1: *b.s.* blood sinus; *gl.c.(A)*, gland-cell with granules with azocarmine affinity; *gl.c.(B)*, with granules with aniline-blue affinity; *m*, muscle; *m.g.* mid-gut; *oes.* oesophagus; *p.c.* peritoneal cell; *r.* rectum; *stom.* stomach; *t.* torus.

ciliated cells. These mucus cells are scattered throughout the length of the oesophagus which extends back to the level of the 1st pair of tori; externally the oesophagus is distinguishable by its lighter colour from the yellow stomach, but internally there is a sharp histological transition.

The stomach has much thicker walls than the oesophagus and is much involuted (Fig. 2C). A few ciliated cells extend into the anterior part of the stomach, but cilia are absent from the main part of the stomach and from all the more posterior regions of the gut. The stomach extends back to the level of the 3rd pair of tori. The epithelial cells are mainly glandular as indicated by their heavily staining granular contents. When stained with Azan the outer part of the cell is vacuolated and contains red-staining granules, the middle part has rather larger granules with a much greater affinity for aniline blue, but without vacuoles, and the inner part of the cell contains the nucleus lying beneath these granular zones, within a dense red-staining cytoplasm (Fig. 1F). Other cells, presumably also secretory, contain granules with a strong affinity for azocarmine; these seem to be more numerous posteriorly and are represented in the most posterior part of the stomach where there are no cells with aniline-blue staining granules.

The transition from the stomach to the mid-gut at the level of the 3rd pair of tori is accompanied by another abrupt histological change. The wall of the mid-gut is much thinner than that of the stomach, the cells with blue-staining granules are entirely lacking. The cells of the mid-gut have a well-marked brush-border, nuclei nearer the middle of the cell than in the stomach, and a fine granular cytoplasm (Fig. 2G). The mid-gut is recognizable externally by its deep green colour which is due to granules of pigment in the middle of the cell. The pigment remains even in Bouin-fixed specimens, but some worms were fixed in Helly's fluid and lightly stained in orange G in order to get a better appreciation of their distribution. The granules vary in size between 3 and 27 μ; they seem structureless and are rather irregular in outline. The granules freed from the tissue did not appear to be motile, and there was nothing to indicate that they represented living plants as suggested by Berkeley (1930) for the rather similar green pigment in the mid-gut of *Chaetopterus*. A few granules of a similar green pigment appear in the epidermis of the crown and other parts of the body of some specimens (Fig. 1D). As noted by Watson (1901) the amount present varies greatly; some of the worms from Torquay collected in April were completely green.

The mid-gut passes into a thin-walled distensible rectum at the level of the 5th–6th pairs of tori (Fig. 2H). It is here that sand grains and other matter collect before being defaecated in discrete pellets. Defaecation seems to be an intermittent process, and a great deal of matter may collect in the extensive rectum before the worm reverses in its tube to void the contents. The faecal pellets are surrounded by a thin membrane which stains blue with Azan. There appears to be no mid-ventral gutter so commonly present in this region

in other polychaetes, and the rather short cells of the hind-gut appear to have a brush-border without cilia, and with a fine granular cytoplasm. Transmission of the gut contents is presumably mainly muscular, both by the action of the gut musculature itself, though this is very poorly developed in all regions, and by the squeezing action of the body-wall muscles acting through the coelomic fluid. Strong peristaltic waves of contraction may often be seen in the hind part of the body in worms removed from their tubes.

Worms continued feeding when iron saccharate was added to the water, and these were fixed in Bouin's fluid 3 h after the addition. The whole gut of several worms was sectioned, and the sections submitted to the Prussian blue reaction and then lightly counterstained with orange G. Iron particles were found only in the mid-gut cells. It is interesting to note that the region in which the iron had been absorbed coincided exactly with that occupied by the green pigment. No tests have been made for enzymes, but the histology suggests that the stomach is the main, if not the only, secretory part of the gut, the mid-gut the absorptive part, and the rectum serves to store and elaborate the faecal pellets.

ANALYSIS OF THE GREEN PIGMENT

The mid-gut region of over fifty worms was carefully dissected out and extracted in ether:acetic acid, 5:1 mixture, after grinding in a small Griffeth-pattern tissue homogenizer. The dark green solution had a brilliant deep red fluorescence in ultra-violet light. The acetic acid was washed out by shaking repeatedly with distilled water (the first three washings buffered with sodium acetate) until the washings were no longer acid to blue litmus paper. The ethereal solution was then roughly dried by passing through ether-soaked filter paper, and concentrated *in vacuo*. The filtrate was then examined by long-paper chromatography (Kennedy, 1953) using 2:6 lutidine-water system. Two spots were obtained, one with an R_F value of 0·53, suggesting a tetra-carboxyl porphyrin such as coproporphyrin, the other with an R_F value of 0·89, suggesting a mono-carboxyl porphyrin compound such as phaeophorbide.

The ethereal solution was further concentrated *in vacuo* in the cold and extracted with 0·1 % w/v HCl, when a red-fluorescent extract was obtained; this further suggested coproporphyrin which has an HCl value of 0·08. The porphyrin was driven back into ether, neutralized with saturated potassium acetate, and the ether solution washed carefully with distilled water, three times. It was then roughly dried by passing through ether-soaked filter paper as before, and evaporated to dryness *in vacuo*. The pigment was then esterified by dissolving the residue in MeOH:H_2SO_4, 19:1 mixture. This was left for 48 h at room temperature, after which time the solution was diluted with water and the esterified porphyrin extracted with chloroform. The chloroform extract was washed free from acid with water, roughly dried by filtering

through chloroform-soaked paper and redissolved in dry chloroform. The solution was examined with the Hartridge reversion spectroscope and also by the chromatography method of Chu, Green & Chu (1951) when the pigment was found to consist of coproporphyrin III only (Kennedy & Vevers, 1954; Kennedy & Dales, unpublished). The spectrum showed the following maxima (in mμ):

I	II	III	IV
622	566·6	533	500·1

The epiphase from the extract of the ethereal solution with HCl was washed repeatedly with water until free from acid. The solution was examined in the Hartridge reversion spectroscope and also in the 'Unicam' S.P. 600 quartz spectrophotometer when the following maxima characteristic of phaeo-phorbide-b were shown (in mμ):

	I	II	III	IV	Sorêt
Hartridge reversion	652	602	537·5	504·5	
'Unicam'	655	603	537	505	408

The ethereal solution was then extracted with 0·2 % w/v sodium bicarbonate (Willstätter & Stoll, 1913). The hypophases were red-fluorescent, the epiphases were not; this indicates the presence of phaeophorbide-b only. There was also a residual yellow pigment in the epiphase after extraction with sodium bicarbonate, which when concentrated had a very slight yellowish fluorescence and which was possibly a carotenoid.

Extracts of green specimens from which the gut had been removed were similarly treated, and the green pigment in the epidermis was also found to be phaeophorbide-b alone.

This work was mostly done at Plymouth, and I am indebted to the Director and staff of the Laboratory for their help, and to Dr G. Y. Kennedy of the Department of Cancer Research, Sheffield, in particular, for analysing the green pigment.

SUMMARY

Owenia may feed either by ciliary means or by swallowing sand and detritus. The gut consists of three main parts, a fore-gut or stomach which is secretory, a non-secretory mid-gut region which is absorptive, and a hind-gut which is non-secretory, non-absorptive and serves to elaborate and store the faecal pellets. The mid-gut contains coproporphyrin III and also phaeophorbide-b, to which the green colour is due. The green pigment in the epidermis is also phaeophorbide-b, but phaeophorbide-a is apparently absent from the body.

REFERENCES

BERKELEY, C., 1930. The green bodies of the intestinal wall of certain Chaetopteridae. *Quart. J. micr. Sci.*, Vol. 73, pp. 465–76.

CHU, J. C., GREEN, A. A. & CHU, E. J., 1951. Microchromatography of porphyrin esters. *J. biol. Chem.*, Vol. 190, pp. 643–6.

CLAPARÈDE, É., 1868. Les Annélides Chétopodes du Golfe de Naples. *Mém. Soc. Phys. Genève*, T. 19, pp. 313–584.

—— 1873. *Recherches sur la structure des Annélides Sédentaires.* 199 pp. Geneva: Georg.

DRASCHE, R. VON, 1885. *Beiträge zur feineren Anatomie der Polychaeten. Zweites Heft: Anatomie von* Owenia filiformis *delle Chiaje.* 22 pp. Wien: C. Gerold's Sohn.

EISIG, H., 1887. Monographie der Capitelliden des Golfes von Neapel. *Fauna u. Flora Neapel*, Monogr. 16, 906 pp. Berlin: Friedländer.

ELWES, E. V., 1910. Notes on the littoral Polychaeta of Torquay. Part 3. Other families. *J. mar. biol. Ass. U.K.*, Vol. 9, pp. 59–81.

GILSON, G., 1893. Recherches sur les cellules sécrétantes. II. Les glandes filières de l'*Owenia fusiformis* delle Chiaje. *Cellule*, T. 10, pp. 299–325.

—— 1897. Les valves septales de l'*Owenia*. *Cellule*, T. 12, pp. 377–416.

KENNEDY, G. Y., 1953. Partition paper chromatography of the prophyrins. *Scand. J. clin. Lab. Invest.*, Vol. 5, pp. 281–4.

KENNEDY, G. Y. & VEVERS, H. G., 1954. The occurrence of porphyrins in certain marine invertebrates. *J. mar. biol. Ass. U.K.*, Vol. 33, pp. 663–76.

LO BIANCO, S., 1893. Gli annellidi tubicoli trovati nel Golfo di Napoli. *Atti Accad. Sci. fis. mat. Napoli*, Ser. 2, Vol. 5, pp. 1–97.

MCINTOSH, W. C., 1915. *A Monograph of the British Marine Annelids*, Vol. 3, Part 1, 368 pp. London: Ray Society.

SAINT-JOSEPH, A. de, 1898. Les Annélides Polychètes des côtes de France (Manche et Océan). *Ann. Sci. nat.*, Sér. 8, T. 5, pp. 209–464.

WATSON, A. T., 1901. On the structure and habits of the Polychaeta of the family Ammocharidae. *J. Linn. Soc. (Zool.)*, Vol. 28, pp. 230–60.

WILLSTÄTTER, R. & STOLL, A., 1913. *Untersuchungen über Chlorophyll.* 424 pp. Berlin: Springer.

WILSON, D. P., 1932. On the Mitraria larva of *Owenia fusiformis* delle Chiaje. *Phil. Trans.*, B, Vol. 221, pp. 231–334.