The Reproductive System of the Planarian Artioposthia triangulata (Dendy).

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With Plates 14–16 and 3 Text-figures.

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INTRODUCTION.

ALTHOUGH Dendy has described for New Zealand a number of species of land planarians which he allotted to the genus Geoplana yet in all his and Moseley's work on these worms no study has been made of the microscopic internal anatomy, mainly because the classification was based on external features.

In examining specimens of Geoplana triangulata Dendy in serial sections I found that they possess muscular gland-organs or adenodactyli (von Graff) which are characteristic of the genus Artioposthia, no representative of which small group has previously been identified as inhabiting New

Zealand. Other unusual features noted were a branching vas deferens and elongated ovaries situated far back and not in the region of the brain as is usual. Moreover, each ovary is a complex organ containing more than one true Germarium associated with specialized nutritive cells.

I wish to express my thanks to Professor Benham, F.R.S., for originally suggesting the work and for his help and criticism throughout the course of the investigation.

Previous Work.

The first record of the occurrence of planarians in New Zealand was made by Hutton (**7**, p. 249) in 1873 in a paper on the geographical relations of the New Zealand fauna. He merely remarked on the occurrence of two or three species of land planarians, 'one or two of which belong to the genus Bipalium'. Nothing further was added to this until 1877 when Moseley (**13**) published a detailed description of Geoplana traversii, a small worm measuring about 3 cm. in length. Two mature specimens of this worm had been presented to Moseley by Mr. W. T. L. Travers during the visit of H.M.S. 'Challenger' to Wellington, and Moseley was able to describe them in detail externally, after dissection and in transverse section; but he made no longitudinal sections owing to lack of material, with the result that the internal structure could be only partly worked out. Two years later Hutton (**8**) published a list of the New Zealand planarians which had so far been described, and gave a brief description of the characteristic external features of each species. This list included three marine forms as well as Geoplana traversii mentioned above, and in the following year Hutton (**9**) added to this list two new species which he himself had discovered—Geoplana moseleyi, a small worm about 27 mm. in length in a preserved specimen, and Rhynchodemus testaceus of the generic identification of which he was then doubtful.

No further work was recorded until 1895 when Dendy (**3**) published the first of a series of notes on the New Zealand Planarians in which he described the external appearance, shape, and colour of several new species including Geoplana triangulata, a large worm from Christchurch with a variety australis especially common in Dunedin. Later, in 1897, Dendy (**4**) referred to this same variety a preserved specimen which had been sent to him from Nelson by Sir James Hector (**6**) who had recorded it in 1893 but had not been able to identify the worm at that time. Dendy's nomenclature was based entirely on external characters.

Von Graff (**5**) included Geoplana triangulata in his monograph but did not add anything further to the description given by Dendy, merely quoting extensively from the original text. Up to the present, occasional papers have been published in the 'Trans. N.Z. Inst.', describing new planarians, but all such classification has been based entirely on external characters and no work has yet been done on the internal anatomy of any planarian occurring in New Zealand. In his monograph von Graff (**5**) erects a new genus, Artioposthia, for such forms of the family Geoplanidae as have muscular gland-organs in connexion with the genital atrium. As I find these present on dissection and on examining serial sections of this worm, I attribute Dendy's species to von Graff's genus Artioposthia.

Ecology.

There seems to be no definite limitation of locality for the two varieties of Artioposthia triangulata as assumed by Dendy. Specimens of the 'type' were collected in Dunedin, not Christchurch, while the variety was found to be equally abundant in both Dunedin and Christchurch, and not confined to Dunedin as stated by Dendy.

While this planarian undoubtedly lives in the soil, it is more readily found under boxes, sacks, tins, or any large object which has been lying undisturbed on the soil for some time. Here the soil must be moist and the worm is usually found in association with slaters, slugs, and centipedes. In moist soil of a clayey nature the worm was found several inches below the surface. In one garden in which this worm was known to occur, none was found after extensive digging in the more dry loamy parts.

On turning over the boxes, &c., I found the planarian on the surface of the soil in a very characteristic coiled position, with the ventral surface pressed firmly against the soil so as to form a flat spiral outlined by the pale lateral bands of the dorsal surface. In captivity the planarian still assumed that position, lying on the surface of the soil with the anterior and posterior ends buried.

Materials and Methods.

For micro-dissection it was difficult to find a method of killing this planarian in an extended position without rendering the worm too soft and friable. Following the recommendation of Ullyot (**15**) I tried Steinmann's (**14**) fluid—a mixture of nitric acid, mercuric chloride, and sodium chloride—which certainly killed the worm with practically no muscular contraction, but the tissues were all rendered very brittle and inclined to absorb water, resulting in distortions which made accurate dissection almost impossible. Very weak alcohol gradually increased in strength proved to be more efficient, but in the alcohol, however weak, the worm in its contortions often ruptured the epidermis, causing a certain amount of internal displacement of some of the organs.

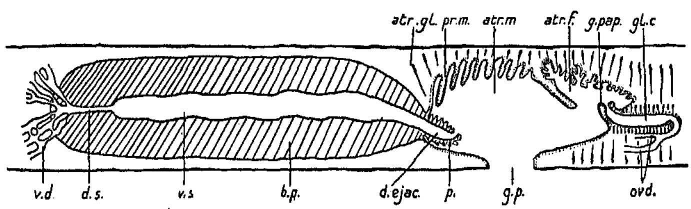
For histological work Carleton's (**1**) method of killing and fixing trematodes was used quite successfully. The worm was placed between two glass plates loosely tied near each end, and left for a short time in that position until it was fully extended. It was then quickly lowered into a dish containing mercuric chloride and acetic acid, heated to 50° C. This resulted in rapid fixation without much apparent shrinkage. After the removal by iodine of any mercury deposit the worms were stored in 70 per cent. alcohol till required. The internal anatomy and histological structure were studied in serial sections which were cut in different planes. As the worm was too large to sectionize completely, horizontal and transverse series of sections were made of the following regions: the anterior end, the pharynx, the genital organs, and the posterior end.

Various staining methods were tried, such as haematoxylin and eosin, picro-indigo-carmine, methylene blue, but except in the case of haematoxylin mounted in euparal 'vert' which clearly demonstrated the nuclear structure, none of the above stains was so successful as borax-carmine counter-stained with picro-nigrosin which later I adopted as the universal staining method.

External Characters.

The external characters of Artioposthia triangulata correspond with the general description given by Dendy (**3**) for Geoplana triangulata. This is a particularly large worm, a well-grown specimen measuring as much as 8 inches in length, while an average length is 5–6 inches. The worm is narrow and strap-shaped and when crawling elongates itself greatly, the body becoming correspondingly narrower.

The dorsal surface can be distinguished from the ventral by its strong convexity, whereas the ventral surface is flattened; the colour and markings are also quite different on the two surfaces. The dorsal surface of the living worm is a dark purplish-brown colour for the median three-quarters of the width, with marginal bands of pale yellowish dotted with minute specks of the same colour as the median portion. The anterior tip is orange-pink, which colour continues back along the marginal bands for about 1 inch, where there are only a few dark specks. Throughout the length of the dorsal surface there is a narrow mid-dorsal line of a dark purple colour. Dendy (**3**, p. 178) describes this in the posterior portion only, but in all the specimens here examined the line was definite for the whole length. The ventral surface is pale yellowish in colour, thickly peppered with purplish specks similar to those on the marginal bands. There are two apertures on the ventral surface: (1) the pharyngeal aperture, in the mid-ventral line about three-fifths of the length of the worm from the anterior end, and (2) the common genital aperture about half-way between the pharyngeal aperture and the posterior end. Several specimens were found of a variety of this worm which appeared to be much more common than the type and which differed slightly from the type in external markings, such differences being constant for all specimens of this variety. Similarly, differences were found in the internal structure, but so slight were these that without the external differences they could have been regarded as varying stages in the maturity of the worm. This variety differs from the type in the following external features: the marginal bands are more definitely marked, and stand out clearly from the median portion, owing to the dark specks being not nearly so plentifully distributed over the bands as in the type. The specks are, moreover, confined to a narrow strip along the inner edge of the marginal band, leaving an almost uniform yellowish strip along the outer edge. The ventral surface has a definite purplish appearance owing to the dark specks being so numerous as almost to obscure the light background.



TEXT-FIG. 1.

Longitudinal vertical section through the genital pores. For lettering see p. 124.

The small differences in internal structure I shall mention later.

I have seen preserved specimens of this variety in which the marginal specks and those on the ventral surface had lost their colour, leaving only the pale background. Dendy (**3**, p. 180) describes a variety australis in which 'speckles are absent on the marginal bands and on the ventral surface', so that it is quite possible that he is describing this same variety, as I have not seem any living specimen to which this description applies. In any case, since the differences between the variety and the type are so small, it is possible that they are merely due to seasonal changes in the one type of worm.

REPRODUCTIVE ORGANS.

I. General.

The common genital pore occupies the normal position on the ventral surface, between the pharynx and the posterior end, being a little nearer to the pharynx than to the posterior end. The genital pore leads into the atrium masculinum (Text-fig. 1) in the anterior portion of which lies the penis. Posteriorly the atrium masculinum receives the atrium femininum into which the female duct opens.

II. Male Organs.

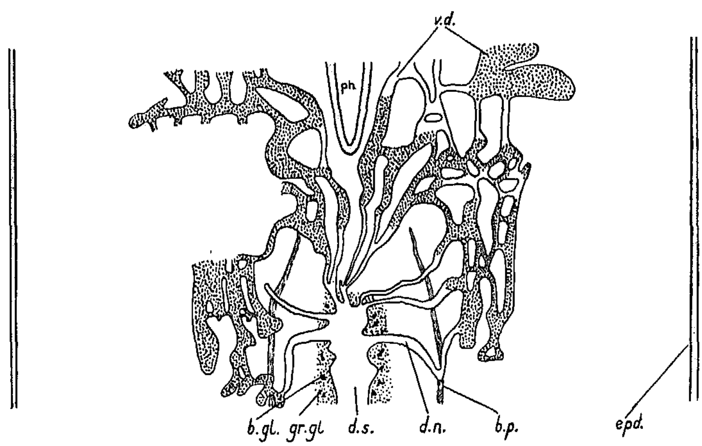
(*a*) Testes.

The numerous testes (fig. 1, Pl. 14) form two longitudinal lateral zones which extend from behind the brain to the region of the glandular canal (fig. 27, Pl. 16) of the female organs. They occupy a ventral position in the body and lie immediately dorsal and lateral to the nerve-cord, but often so close to it that ventral portions of the testicular sacs may be found among the network of nerves. Each testis (fig. 2, Pl. 14) is more or less spherical in shape and is enclosed in a tunica propria of flattened epithelial cells with bulging nuclei. The cavity of the testis is filled with spermatozoa in all stages of development, derived from a germinal epithelium of sperm mother-cells. Dendy (**3**) in the testis of Geoplana spenceri describes a space between the outer sperm mother-cells and the central compact mass of developing spermatozoa. This space is no doubt due to shrinkage of the central mass during fixing and preserving, as I found it in only a few of my sections, while in others the developing spermatozoa extended out to the sperm mother-cells.

The spermatozoa when ripe pass into the vasa efferentia, where their structure may best be seen. The head is small, rounded, and highly refringent and with borax-carmine stains a deeper pink than the tail.

(*b*) Sperm Duct.

The vasa efferentia (figs. 2 and 4, Pl. 14) are extremely slender canals and arise as direct prolongations of the ventral walls of the testes. After a very short course they lead into collecting vessels, the vasa intermedia (figs. 3 and 4, Pl. 14) in which the spermatozoa which were scattered in the vasa efferentia become more densely packed together so that it is fairly easy to follow the course of these vessels on account of their deeply staining contents. The vasa intermedia are slightly convoluted and form a network lying in the upper portion of the circular muscles of the body-wall, just ventral to the nerve. They are of varying length and ultimately open into the vasa deferentia (figs. 1 and 3, Pl. 14). These last (Text-fig. 2) form a very conspicuous part of the worm, and consist, on each side, of a series of wide, convoluted, branching tubes extending from the region of the pharyngeal aperture to the anterior end of the seminal vesicle (fig. 5, Pl. 14, and fig. 20, Pl. 16). They occupy a ventral position



TEXT-FIG. 2.

Longitudinal horizontal section, showing network of vasa differentia opening by narrow ducts into the seminal duct, *d.s.* For lettering see p. 124.

in the body lying dorsally to the nerve-cord, and in the ripened condition the branches extend for almost the whole width of the worm. The vasa deferentia are capable of great distension and form a reservoir for storing the spermatozoa.

As far as I can find in the literature available to me, no such branching vas deferens has been described for any land-planarian. In this group the posterior end of the vas deferens is normally convoluted and swollen, often to a great extent, but nowhere is there any description of branches. This branching system was quite apparent in worms under a dissecting microscope (fig. 5, Pl. 14). Three separate series of sections of this region were cut, and reconstruction drawings made of the complete system. Of the variety one series was cut horizontally (Text-fig. 2) and one transversely to the long axis of the worm. In these the branches were not greatly distended, so that there was ample evidence of the branching, with wide spaces between the branches. The third series was a transverse one of the type, and here the worm was obviously more mature in that the vasa deferentia were so swollen and distended as to occupy almost the whole width of the worm with very narrow spaces between the branches.

The testicular sacs are much more numerous and closely aggregated in front of the vasa deferentia, but those scattered sacs which lie adjacent to the vasa deferentia open directly into them without either vasa efferentia or vasa intermedia.

The vasa deferentia of each side converge posteriorly and give origin to four to six narrow ducts (*d.n.*, Text-fig. 2; figs. 20 and 21, Pl. 16) which are straight tubes lying almost in one plane. As a rule they do not store spermatozoa. These narrow ducts, which I consider to be still part of the vasa deferentia, enter a median canal surrounded by the musculature of the penis-bulb (*d.s.*, Text-figs. 1 and 2; fig. 21, Pl. 16). This median canal I call the seminal duct as it leads from the vas deferens into the seminal vesicle. There seems to be no definite symmetry or regular arrangement of the narrow ducts as they enter the seminal duct, some joining together before entering, others entering separately. The seminal duct continues back as a narrow tube for a short distance and then widens suddenly to form the seminal vesicle (*v.s.*, Text-fig. 1; fig. 5, Pl. 14) surrounded by the characteristic loose basket-work of muscle (*b.k.m.*, figs. 20 and 22, Pl. 16) which extends from the thick muscular walls of the seminal vesicle to the outer wall of the penis-bulb. The width of the seminal vesicle and the amount of folding of its walls depend on the degree of maturity of the specimen; but in a fully mature worm the seminal vesicle may extend in width almost to the wall of the penis-bulb.

Von Graff (**5**, p. 163) describes the posterior part of the vas deferens which becomes swollen with stored spermatozoa as forming an outer seminal vesicle. This he calls 'false' to distinguish it from the 'true' seminal vesicle which has its own strong musculature and therefore preserves a certain constant form. This is readily understood in those species with only one pair of long convoluted vasa deferentia in the posterior ends of which the spermatozoa are stored. But in Artioposthia triangulata, in which the whole of the vas deferens on each side forms a branching network, such a term as 'false' seminal vesicle is not applicable. I reserve, therefore, the term seminal vesicle for the single median tube surrounded by a definite musculature of its own which leads on from the seminal duct.

The seminal vesicle has an approximate length of 9·6 mm. and passes gradually into the ductus ejaculatorius (Text-fig. 1, and fig. 20, Pl. 16) which traverses the penis. The dorsal wall of the ductus now becomes thick and deeply indented, while the ventral wall loses its musculature and flattens out as the duct enters the penis which projects into the anterior end of the atrium musculinum.

Histology of the above Organs.

The wall of the vas efferens (fig. 2, Pl. 14) has the same simple structure as the testicular sac of which it is a direct prolongation. It is formed of flattened epithelial cells with elongated bulging nuclei. In a stained section there was no evidence of cilia, though von Graff (**5**, p. 162) describes them as being present. Dendy (**2**, p. 88) evidently does not find them in Geoplana spenceri, and of the more recent writers the few who describe the structure of the vasa efferentia do not find cilia. If they are present in this worm they would be recognizable, as I found cilia elsewhere in preserved material.

The vasa intermedia have slightly thicker walls than the vasa efferentia, with larger nuclei and no cilia. No muscular layer was found in either the vasa efferentia or the vasa intermedia.

In the vas deferens (fig. 6, Pl. 14) the wall is still thicker, but the thickness varies with the distension of the tubes by the contained spermatozoa. Surrounding the epithelium is a thin layer of circular but no longitudinal muscles. Von Graff (**5**, p. 163) describes the epithelium of the vas deferens as being scarcely half as high as that of the oviduct with round nuclei not so thickly pressed together as in the oviduct. To quote the exact words: 'In der That ist das Epithel derselben kaum halb so hoch als jenes der Oviducte, die Kerne sind rund und nicht so dicht gedrängt wie in diesen.' This description does not apply here as the epithelium in both the distended and non-distended portions is more than half the height of that of the oviduct (fig. 10, Pl. 15), while the nuclei are elongated not rounded. Very long cilia were easily seen in the parts with no spermatozoa, but were sometimes obscured in those parts which contained the massed spermatozoa, still we may assume that they are a constant feature of the whole of the vasa deferentia.

According to von Graff (**5**, p. 163) the vas deferens in the front part of the body has no 'muscularis', but muscles appear in it in the region of the pharynx and increase towards the penis. This corresponds with the condition found in Artioposthia triangulata in which the anterior ducts (vasa efferentia and intermedia) have no muscular wall, whereas the vasa deferentia which begin in the region of the pharynx have a definite circular muscle-layer which becomes thicker in narrow ducts at the posterior end.

The narrow ducts are lined with cubical cells with round nuclei and very long cilia, and as they enter the seminal duct the epithelial cells become much longer and narrower until in the seminal duct itself (fig. 9, Pl. 15) they become columnar and are almost thread-like with long compressed nuclei. This characteristic shape may, perhaps, be related to the pressure of the secretion of the male accessory glands which I shall describe later.

The epithelium of the seminal vesicle (fig. 7, Pl. 14) is secretory in function, and the cells are large and very striking in appearance. They are bluntly conical with the distal portions of the cells not in contact with one another. The nucleus and cytoplasm are confined to the proximal part of each cell while the distal region stores the secretion which may be poured into the cavity of the seminal vesicle through the notched apex of the cell. This secretion consists of mucin, as on testing with mucicarmine it took the characteristic violet shade. The wall of the seminal vesicle is very muscular (fig. 7, Pl. 14, and fig. 22, Pl. 16) with a layer of circular muscles just outside the epithelium followed by alternate layers of longitudinal and circular muscles. The whole forms a very firm muscular wall to the seminal vesicle which in its turn is surrounded by the basket-work of muscle connected with the outer wall of the penis-bulb.

In the ductus ejaculatorius (fig. 8, Pl. 14) the muscular wall is not so clearly marked off, and the secretory cells of the seminal vesicle are replaced by ciliated cells which at the free end of the penis merge into the ciliated epithelium of its outer surface.

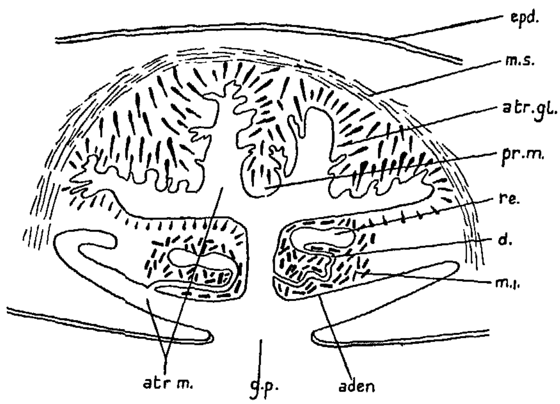
(*c*) Penis.

The penis (fig. 5, Pl. 14) is a very small, inconspicuous, muscular organ the wall of which is transversely folded and is no doubt capable of great extension. It is traversed by the ductus ejaculatorius, the external opening of which is in the form of a wide transverse slit lying nearer the ventral than the dorsal surface, and discharges the male products into the atrium masculinum.

(*d*) Atrium Masculinum and Adenodactyli.

The atrium masculinum presents some peculiar features which I have not found described in such literature as is available to me. Viewed from the ventral surface (fig. 5, Pl. 14) the atrium is triangular in shape with the base of the triangle toward the seminal vesicle. The dorso-lateral walls on each side bear three thick, rounded, muscular masses or adenodactyli (von Graff) which project into the atrial cavity leaving between them only a long narrow vertical space. Ventrally and ventro-laterally the adenodactyli are not attached to the wall of the atrium (Text-fig. 3; fig. 24, Pl. 16) which in this region is consequently quite wide—about half the width of the worm. Anteriorly the adenodactyli of the two sides are joined by a firm muscular archway (fig. 5, Pl. 14) which curves over the penis, while posteriorly they converge and join to form a kind of shallow muscular scoop (*sc.m.*) which projects freely backwards and marks the posterior limit of the atrium masculinum.

Each of the adenodactyli is made up of strong wide bands of muscle which are loosely woven together and enclose a glandular reservoir (Text-fig. 3; fig. 24, Pl. 16). This is a wide, elongated cavity slightly constricted in the middle and leading at one end into a winding duct which opens into the atrial cavity. The reservoir is lined with curious elongated secretory cells (fig. 11, Pl. 15) arranged in groups of varying heights. These cells have round nuclei near the free end below the terminal secretion which is discharged into the cavity of the reservoir. The wall of



TEXT-FIG. 3.

Transverse section through the genital pore. For lettering see p. 124.

the duct is formed of cubical cells with short cilia and rounded nuclei. These cubical cells take a bright orange stain with picro-nigrosin and appear also to contain secretory granules.

This description of the adenodactyli and their glands agrees in all essential points with that of von Graff (p. 179) for the adenodactyli of Artioposthia, with this important exception that von Graff does not describe a duct leading from the reservoir to the atrial cavity, nor does he figure such a duct in his plates. He describes a flask-shaped or pear-shaped gland which is both a reservoir and a duct and yet the secretion seems to remain in the reservoir, or perhaps is distributed to the surrounding muscular mass. I will quote von Graff's (**5**, p. 179) description of the gland and its cell structure:

"Es handelt sich nämlich bei allen diesen Organen um

> flaschen- oder birnförmige Drüsen, die von einem kräftigen Muskelmantel umgeben sind. Bei den Artioposthia-Arten besteht die Drüse stets aus zweierlei Zellen: Kleinen cubischen (meist) Cilien tragenden Epithelzellen, welche das centrale, zugleich als Reservoir und Ausführungsgang dienende Lumen der Drüse bekleiden, und birnförmigen secretorischen Zellen. Letztere finden sich rings um das Drüsenlumen und besonders massenhaft um den Fundus desselben angehäuft und convergiren mit ihren Ausführungsgängen, um zwischen dessen Epithelzellen einzudringen und das körnige Secret in das centrale Reservoir zu entleeren."

I find this description very vague and difficult to picture. If the secretion is discharged by the cells into the reservoir it would seem to require a duct to carry the secretion from the reservoir to the exterior, and yet I can find no such duct described or figured. In Artioposthia triangulata the reservoir is lined with secretory and the duct with ciliated cells. Lucy M. Wood (**16**, p. 612) in describing the male adenodactyli of Artioposthia harrisoni says, 'In transverse section they show a central cavity surrounded by gland-cells which are enclosed in a muscular sheath.' Again there is no duct, and one may wonder what ultimately becomes of the glandular secretion which is poured into this central cavity by the surrounding gland-cells. It can serve no definite purpose unless it reaches the outside of the adenodactyli, and that can only be effected by means of a duct from the central cavity to the atrium.

Apart from this difference the adenodactyli of Artioposthia triangulata with their enclosed glands agree essentially with von Graff's description. Although in the normal condition of this worm the adenodactyli appear as rounded masses, and do not project through the genital aperture as seems to be the case with the majority of male adenodactyli, yet they are quite extensible and have been observed to be thrust out into the atrial cavity as elongated processes enclosing the reservoir and duct. In this respect they resemble more the female adenodactyli of Artioposthia fletcheri (**5**, p. 227) which do not project so far into the atrial cavity.

As for the function of the adenodactyli, one cannot make a definite statement until one has made a careful study of the habits of this worm. One can only conjecture from the small size of the penis that the adenodactyli with their great muscularity do in some way supplement the function of the penis.

The dorsal wall of the atrium masculinum, i.e. the tract between the dorsal portions of the adenodactyli, bears along its length an irregular series of long, narrow, muscular processes (*pr.m.*, Text-fig. 3; fig. 5, Pl. 14; fig. 24, Pl. 16) which are smaller than the adenodactyli and have not their complex glandular structure.

The atrium masculinum is lined with a single-layered epithelium of cubical ciliated cells continuous with the outer epithelium of the penis. There appears to be no definite muscle-layer lying immediately underneath the epithelium as is generally the case, but closely interlacing muscle-fibres are found in the parenchyma outside the epithelium, and these no doubt serve the same purpose.

(*e*) Accessory Glands.

Two glands are found in association with the seminal duct and seminal vesicle. These lie in the parenchyma surrounding the duct, and may be differentiated according to the form of the secretion and the colour which they assume on staining.

(1) A 'diffuse' gland (*b.gl.*, fig. 21, Pl. 16) extending from the epithelium of the duct right out to the wall of the penis-bulb. It occurs especially round the seminal duct and to a much less extent round the seminal vesicle. With borax-carmine the secretion takes the same bright red colour as that of the skin glands.

(2) A finely 'granular' gland (*gr.gl.*, fig. 21, Pl. 16) which is stained a bright pink with borax-carmine. This gland surrounds only the seminal duct and extends throughout its length. As it passes back along the duct the gland becomes more concentrated until towards the posterior end it forms a deep pink band round the seminal duct (fig. 23, Pl. 16), by which time the 'diffuse' gland has become greatly reduced, being found only at irregular intervals behind this region.

The ducts of these glands (fig. 9, Pl. 15) as of the skin glands are merely extensions of the body of the gland-cells which force their way between the epithelial cells of the cavities which they surround, and there swell up continuously with the pressure of the secretion from behind. The epithelial cells are consequently pressed together so that ultimately they assume a thread-like form such as we find in the seminal duct. In the seminal vesicle there is no such modification of the epithelium, as the accessory glands are very scattered and the main secretion takes place from the epithelial cells themselves. Stained masses of the secretion can be seen in the cavities of the seminal duct and of the seminal vesicle where the secretion performs the usual function of mixing with the spermatozoa on their way to the genital atrium.

A diffuse atrial gland similar to that surrounding the seminal duct and seminal vesicle occurs throughout the parenchyma of both the dorsal muscular processes and the muscular masses. The gland-cells take a deep crimson stain with borax-carmine and prolongations of these cells force their way between the outer epithelial cells and so empty their products into the atrium masculinum.

III. Female Organs.

(*a*) Ovaries.

The ovaries of Artioposthia triangulata are quite unusual and, in the literature available to me, I can find no description which is applicable to the condition here found.

There is a single pair of ovaries (fig. 5, Pl. 14; fig. 20, Pl. 16) which are situated laterally or ventro-laterally in the region of the seminal vesicle, and lie just outside the posterior limb of the gut. Each ovary (fig. 5, Pl. 14) is a long fusiform body more or less regularly constricted on its ventral surface. This fusiform shape seems to be peculiar to this worm, as von Graff (**5**, p. 151) describes the ovary as being 'usually spherical with occasional oval forms'; Dendy (**3**, p. 82) finds the ovary of Geoplana spenceri 'pear-shaped', while Kaburaki (**11**, p. 145) finds it 'nearly oval' in Geoplana whartoni. It will be shown later, however, that this so-called ovary of Artioposthia triangulata is really a complex structure enclosing more than one true ovary or germarium, as well as specialized parovarian and amoeboid cells.

The position of the ovary in the body is also unusual. As a general rule, in land planarians the ovary is situated far forward, in the region of the brain, and von Graff (**5**, p. 151) quotes a variety of examples to illustrate this, adding that the posterior boundary of the ovary does not extend beyond the anterior third of the body. In Artioposthia triangulata the ovary is much farther back, its anterior end being 77 mm. from the anterior end of a worm of 152 mm. length, i.e. half-way back. This unusual position of the ovary does not seem to have any bearing upon its relationship with the other genital organs.

The ovary (fig. 12, Pl. 15; fig. 25, Pl. 16) is enclosed in an epithelium of pavement cells with long flattened nuclei, surrounding which is a layer of loosely arranged circular muscle-fibres (*c.m.*). The main portion of the ovary is made up of parenchyma which, at intervals along the ventral surface, extends from the ovary towards the adjacent wall of the seminal vesicle (fig. 20, Pl. 16). The ovarian epithelium is interrupted at these points of extension, and the circular muscle-fibres follow the course of the parenchymatous band towards the seminal vesicle. In a transverse section (fig. 12, Pl. 15, and fig. 25, Pl. 16) the ovary is seen to consist of an outer area of parenchyma enclosing a more or less definite inner region in which are found the germaria (*gm.*) and groups of specialized cells connected with the nutrition of the ova (*am.* and *par.*).

The germaria (fig. 5, Pl. 14) are situated on the dorsal or dorso-lateral margin of the inner area of the ovary. In two specimens of the variety I found two germaria in each ovary, and in one specimen of the type I found four (as illustrated). Without examining a great many worms in all stages of maturity I am not able to state definitely whether the number of germaria is constant and definite for each variety, or whether it varies according to the maturity of the worm. I am inclined to think that the latter is the true explanation, as in the worm in which four germaria were present all the genital organs were in a more advanced stage of development.

When four germaria are present the anterior one is situated at about one-quarter of the length of the ovary from the anterior end, with the other three arranged at regular intervals behind. The anterior germarium is large and mature and lies directly opposite the internal opening of the oviduct (fig. 12, Pl. 15) which enters the ovary on the ventral surface. The remaining three germaria become successively smaller and more immature until the posterior one is very small and contains only a very few ova. The same relationship is seen in the variety with only two germaria present. The anterior one is large and mature and has the same relative position as in the type, while the second germarium which is quite small lies about half-way along the ovary.

As there is only one oviduct on each side of the worm, with only one opening leading into the ovary opposite the anterior germarium, we may presume either that the germaria as they develop and ripen move forward in turn until they lie opposite the opening of the oviduct, or that after the escape of the ripe ova from the anterior germarium the oviduct withers anteriorly and forms a new connexion with the second germarium, and so on with the others as they in turn ripen.

The germaria (fig. 14, Pl. 15, and figs. 25 and 26, Pl. 16) are pear-shaped sacs which lie with their broad ends along the outer margin of the inner area of the ovary and their pointed ends towards the centre. There is a definite tunica propria of flattened epithelial cells (fig. 14, Pl. 15), surrounding which are a few irregularly placed muscle-fibres. This description of the germarium may be compared with that of Moseley (**12**, p. 136), for the ovary of Bipalium, which is pear-shaped with a 'distinct but delicate membranous capsule' and 'externally to the capsule a wide space occupied by an irregular mesh-work of connective tissue'—this latter corresponding with the outer area of parenchyma in the ovary of Artioposthia triangulata. In Artioposthia the germarium is lined with a germinal epithelium (fig. 14, Pl. 15) continuous with an inner irregular mesh-work of branched cells forming a stroma in which are embedded the ova. The nuclei of the stroma cells are exactly similar to those of the germinal epithelium, being oval in shape with deeply staining chromatin, which bears out von

Graff's statement (**5**, p. 151) that the stroma cells are differentiated germinal epithelial cells and not parenchyma.

In the young oocytes next to the germinal epithelium one can see quite clearly the various phases of meiosis through which the nucleus is passing—definite chromosomes, bouquet stage, &c. As the ova grow and increase in size, the nucleus becomes larger and vesicular with one deeply staining nucleolus in the chromatin network. Yolk-bodies are formed in the cytoplasm, at first only one or two which increase to as many as ten in a mature ovum. These yolk-bodies are not composed of fat or oil but take a protein stain.

The immature germarium is similar to the mature in general structure, but has only a few large ova occupying the central part of the mass. In a mature germarium spaces may sometimes be observed around the ova or to one side of them. These spaces are probably what Moseley (**12**, p. 137) describes as 'egg-capsules'; but are no doubt here, as in Moseley's specimens, due to the shrinkage of the cytoplasm of the ova, as they are present in some of my preparations and not in others.

Von Graff states that at the end of the functioning period the epithelial cells and stroma cells disappear, the latter being used to form yolk-bodies in the ripe eggs. This condition does not exist in Artioposthia triangulata, as in the mature germarium (fig. 14, Pl. 15) the germinal epithelium and stroma cells are quite as large and numerous as in a less advanced germarium, unless such a reduction takes place at the very end of the functioning period just before the ova leave the germarium.

Dendy (**2**, p. 82) describes in Geoplana spenceri spindle-shaped cells which appear after the formation of the young ovum and adhere closely to its surface. These spindle cells later on disappear when the ovum becomes mature. I think that there is no doubt that these spindle cells correspond to the stroma cells of the germarium of Artioposthia triangulata which are present in all the early stages of development, and which, according to von Graff, are gradually absorbed and worked up into the yolk bodies of the ripe ova.

The inner portion of the ovary (fig. 12, Pl. 15, and fig. 25, Pl. 16) is roughly outlined by a layer of deeply staining cells which are connected with rows and groups of similar cells in the inner parenchyma. These deeply staining cells I shall call parovarian cells for reasons which I shall give later. The inner parenchyma is loosely arranged to form a number of irregular cavities in which are found isolated cells or small clumps of two or three cells which I shall call amoeboid cells on account of their characteristic shape. These two types of cells constitute the specialized cells of the inner area of the ovary and I shall now describe each type in detail.

The parovarian cells (figs. 17 and 18, Pl. 15) present a variety of shapes from cubical cells with large round nuclei to elongated ones with long narrow nuclei. They are immediately noticeable in any preparation stained with Borax-carmine on account of the brilliant red colour taken by the cell inclusions. What these inclusions are I cannot say, but sometimes they have the form of fine granules, at others they appear as a tangled mass of very finely looped threads. The nucleus is in some cases small and round at the base of the cell, in others, large and vesicular and in the centre. These parovarian cells seem to be constantly dividing amitotically as Woodworth (**17**, p. 33) describes for Phagocata gracilis, and one can see the elongated dumb-bell-shape of the nucleus so characteristic of amitotic division. The parovarian cells ultimately become continuous with similar cells arranged as a wall to a tube (fig. 12, Pl. 15, and figs. 25 and 26, Pl. 16) which leaves the germarium at its inner pointed end and leads towards the internal opening of the oviduct. This parovarian tube arises on the inner side of the germarium as a small papilla outlined by stroma cells which may be compared with the papilla described by Moseley (**12**, p. 138) for Rhynchodemus as follows: 'In Rhynchodemus the oviduct was found to take origin from a papilla on the upper and inner side of the ovary and projecting into its cavity. The papilla is formed of spindle-cells and a number of similar cells are to be found scattered in the loose tissue around its base.'

As the parovarian cells which lead from the germarium have especially deeply staining inclusions, it may be assumed that these inclusions are for the nutrition of the ova and will be discharged into the oviduct. I have repeatedly observed in the first portion of the oviduct small deeply staining granules which may be presumed to have come from the parovarian cells of this region.

Von Graff (**5**, p. 152) describes two types of parovarium: (1) an appendage which opens into the oviduct immediately after it leaves the ovary, and which is really a mass of yolk. This may also extend outwards and give rise to the main yolk-glands; and (2) an appendage which is exactly similar to the ovary in structure and may be considered to be merely an abnormal diverticulum of the ovary. The parovarian cells of Artioposthia triangulata belong to type (1) though they are not massed together to form a solid gland, but are arranged in rows of cells.

According to the same authority the parovarium has two connexions with the oviduct; one, directly, as the oviduct leaves the ovary, and the other, indirectly, through the main yolk-glands which arise from the parovarium and which open into the oviduct at intervals along its length. I can find no connexion between the parovarian cells of Artioposthia triangulata and the main yolk-glands. In all the specimens which I examined, the yolk-glands were well developed, and if they had arisen from the parovarian cells, as von Graff suggests frequently happens, there would I think have been some evidence of this connexion here.

Woodworth (**17**, p. 33) also describes for Phagocata gracilis the chains of yolk-cells leading from the parovaria to form rudimentary yolk-glands which branch to form 'a dendritic system of rapidly dividing cells which ramify through the tissues'. He considers that this is the normal origin of the yolk-glands which is so clearly shown in Phagocata, but which may not be so evident in another genus where the cells of the yolk-gland and of the parovarium are more differentiated and therefore are not so obviously related. But even then one should be able to find some evidence of a connexion between the two groups of cells, and I entirely failed to do so after careful examination of stained serial sections.

Von Graff, although he does not appear to have looked into this matter himself, in reviewing the evidence of research workers to date, is not so definite as Woodworth in his statements, and in all the specific examples he quotes there is no mention of a direct connexion between the parovarium and the yolk-gland. I cannot but believe that, in Artioposthia triangulata at least, the parovarium and the yolk-gland develop separately and each forms for itself its own connexion with the oviduct.

The amoeboid cells are to be found in irregular cavities in the inner parenchyma (fig. 12, Pl. 15) either isolated, or in clumps, or lining the walls of these cavities, so that it would appear as if the cells developed from the parenchymatous tissue and dropped into the cavities when mature. A single cell (figs. 15 and 16, Pl. 15) has the characteristic irregular shape with a large nucleus, and in the cytoplasm a clear non-staining vesicle about the same size as the nucleus. The vesicle probably contains a reserve product of an oily nature which has been dissolved out in the process of fixing and staining, and which may help in the nutrition of the egg as these cells pass into the oviduct with the ova.

The oviduct on entering the ovary on its ventral surface passes through the outer parenchyma and widens to form the internal opening (fig. 12, Pl. 15) just inside the inner parenchyma. The germarium lies almost opposite this on the dorsal wall and the connexion between the germarium and the internal opening of the oviduct is as follows. The parovarian tube which leads away from the germarium ends abruptly in a cavity of amoeboid cells which does not seem to be definitely outlined and which is connected with other similar cavities in the central portion of the ovary. From here a short tube containing amoeboid cells enters the internal opening of the oviduct from which it can be distinguished by the fact that the oviduct is ciliated, and almost encloses the tube of amoeboid cells (fig. 13, Pl. 15). The fact that the oviduct does not leave the germarium itself gives both the parovarian and the amoeboid cells an opportunity either of discharging their nutritive products into the oviduct (as in the case of the former) or of themselves entering the oviduct (as in the case of the latter).

(*b*) Oviducts.

The oviduct on each side arises ventrally from the ovary about one-quarter of its length from the anterior end, and continues as a narrow tube straight back towards the posterior end of the worm lying just dorsally to the longitudinal nerve (fig. 5, Pl. 14). After passing the atrium femininum the oviducts converge slightly towards each other, and then turn abruptly at right angles to meet in the middle line (Text-fig. 1). This common duct continues back for a very short distance, and then turns forward to enter the posterior end of the glandular canal (fig. 5, Pl. 14). The oviduct is round in section and is lined with short columnar cells with long cilia (fig. 10, Pl. 15). Surrounding the epithelium is a layer of circular and longitudinal muscles.

(*c*) Glandular Canal.

The glandular canal runs forward from the single oviduct to the posterior end of the atrium femininum which it enters on a small papilla on the left (Text-fig. 1, and fig. 5, Pl. 14). The canal (fig. 27, Pl. 16) is twice the width of the oviduct in section, but the epithelium is folded so as to be capable of much greater extension. The epithelium is formed of ciliated columnar cells, surrounding which is a layer of circular and longitudinal muscles.

In a section stained with borax-carmine (fig. 27, Pl. 16) the whole of the wall of the canal as well as the parenchyma for some distance round it appear as a bright red mass. This is due to a glandular secretion which is present in such large quantities as to obscure the tissues through which it passes. The glands themselves are scattered throughout the width of the worm and open into the glandular canal along its whole length, the secretion no doubt being used in the formation of a cocoon.

(*d*) Yolk-glands.

The yolk-glands (figs. 20 and 21, Pl. 16) are found throughout the length of the worm occupying the spaces between the diverticula of the alimentary tract. The gland on each side consists of irregularly branched groups of cells (fig. 5, Pl. 14), each group constituting a follicle (fig. 19, Pl. 15) which, however, has no enclosing membrane. The cells are large and roughly hexagonal from mutual pressure, and each cell has a thick firm cell-wall which encloses a mass of highly refringent spherical deutoplasmic bodies—the yolk spheres. The nucleus is large, oval, and granular and is situated at one side of the cell.

As described by von Graff (**5**, p. 155) the glands at first appear as single cells scattered throughout the parenchyma, these cells by division forming smaller and then larger groups of cells until the complete gland is formed. The young cells (fig. 19, Pl. 15) are situated on the periphery of the follicle and are smaller than the mature cells and of a variety of shapes. With haematoxylin and eosin the granular cytoplasm takes a uniform deep violet colour and encloses a large granular nucleus with a deeply staining karyosome.

Since the yolk-glands extend from one end of the worm to the other, and since the oviduct in Artioposthia triangulata is shorter than in other worms of this group where the ovary is far forward, it remains a still greater problem how the yolk-granules from the anterior and posterior regions of the worm find their way into the oviduct. At regular intervals along the oviduct (fig. 5, Pl. 14) appear short stumpy branches, each of which connects with an adjacent yolk follicle. The enclosed cells break down when ripe and their contents are directed into the oviduct by the ciliated epithelium of the branch. Neighbouring masses are able to dispose of their products through the cavity left by the yolk follicle which has already discharged its products into the oviduct. Since the yolk-masses are connected throughout the worm, and since these are more mature in the region of the oviduct than at the ends of the worm, it may reasonably be assumed that the unripe yolk-masses move up to take the place of the ripened ones which have already discharged their contents into the short branches of the oviduct lying alongside. Such a movement of yolk could be caused by the constant contraction and expansion of the gut diverticula during the passage of food through the worm and by the muscles of the body generally.

No receptaculum seminis and no uterus were found.

(*e*) Atrium Femininum.

The atrium femininum (Text-fig. 1 and fig. 5, Pl. 14) is much smaller than the atrium masculinum, being less deep dorso-ventrally and only about one-quarter of its length. It tapers posteriorly where it receives on the left side a small papilla on which opens the glandular canal. The atrial cavity is lined by ciliated epithelium and the dorsal wall bears irregular muscular processes which, however, have none of the characteristic features of adenodactyli.

Summary.

The main subject of this paper is a detailed description of the reproductive organs of a planarian initially described by Dendy as Geoplana triangulata. Five unusual features are observed in the reproductive system:

1. The vas deferens consists of a series of wide convoluted branching tubes extending from the region of the mouth to the anterior end of the seminal vesicle.
2. The penis is very small and inconspicuous.
3. The atrium masculinum is provided with three pairs of muscular gland-organs or adenodactyli.
4. The paired ovaries are situated one on each side of the seminal vesicle, not in the region of the brain as is usual.
5. Each ovary is a long fusiform body enclosing more than one true ovary or germarium, as well as specialized parovarian and amoeboid cells which are probably nutritive, and are associated with the internal opening of the oviduct.

The writer refers Geoplana triangulata Dendy to the genus Artioposthia owing to the presence of adenodactyli in the atrium masculinum. Each adenodactylus encloses a glandular reservoir from which a ciliated duct leads to the atrial cavity. The actual function of the adenodactyli is obscure, but the very small size of the penis and the fact that the adenodactyli are extrusible suggests the possibility of these latter performing the function of a penis.

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Explanation of Lettering.

aden., adenodactylus; *al.c.*, alimentary canal; *am.*, amoeboid cell; *am.t.*, tube of amoeboid cells; *atr.f.*, atrium femininum; *atr.gl.*, atrial gland; *atr.m.*, atrium masculinum; *b.gl.*, diffuse brightly staining gland of sperm duct; *bk.m.*, basket-work of muscle; *b.p.*, penis-bulb; *c.*, cilia; *c.ep.*, ciliated epithelium; *chs.*, chromosomes; *c.i.*, cell inclusions of parovarian cells; *c.m.*, circular muscles; *d.*, duct leading from glandular reservoir to atrial cavity; *d.ejac.*, ductus ejaculatorius; *d.b.gl.*, duct of brightly staining gland; *d.gr.gl.*, duct of granular gland; *d.n.*, narrow duct; *d.s.*, seminal duct; *ep.*, epithelium; *epd.*, epidermis; *epd.gl.*, epidermal gland; *fol.*, yolk follicle; *ger.ep.*, germinal epithelium; *gl.c.*, glandular canal; *gm.*, germarium; *g.p.*, genital pore; *g.pap.*, genital papilla; *gr.gl.*, granular gland of seminal duct; *l.m.*, longitudinal muscles; *m.i.*, interwoven muscle of adenodactylus; *m.s.*, muscular sheath surrounding genital atrium; *n.c.*, nerve-cord; *nu.*, nucleus; *nuc.*, nucleolus; *o.c.m.*, outer circular muscle; *om.*, ovum; *ooc.*, oocyte; *ov.*, ovary; *ovd.*, oviduct; *p.*, penis; *pa.i.*, inner parenchyma of ovary; *pa.o.*, outer parenchyma of ovary; *pap.*, papilla; *par.*, parovarian cells; *par.t.*, tube of parovarian cells; *ph.*, pharynx; *ph.ap.*, pharyngeal aperture; *pr.m.*, muscular processes of atrium masculinum; *re.*, glandular reservoir; *sc.m.*, muscular scoop; *se.*, secretion; *se.c.*, secretory cells; *sp.*, spermatozoa; *sp.c.*, spindle cells; *str.*, stroma; *t.*, testis; *t.pr.*, tunica propria; *tr.m.*, transverse muscle; *vac.*, vacuole; *v.d.*, vas deferens; *v.ef.*, vas efferens; *v.i.*, vasa intermedia; *v.s.*, seminal vesicle; *y.b.*, yolk-body; *y.br.*, branch of oviduct to yolk follicle; *y.c.*, yolk-cell; *y'.c'.*, young yolk-cell; *yk.gl.*, yolk-gland; *y.s.*, yolk-sphere.

DESCRIPTION OF PLATES 14–16.

Plate 14.

Fig. 1.—Horizontal section of one side of Artioposthia triangulata showing ventral portion of the testes, represented as clear spaces among the branches of the nerve. × 18.

Fig. 2.—Section of a single testis with vas efferens. × 200.

Fig. 3.—Horiz. sect. of one side slightly more ventral than fig. 4, to show the course of the vasa intermedia entering the vasa deferentia. × 18.

Fig. 4.—Semi-diagrammatic transverse section of ventral portion showing connexion between vasa efferentia and vasa intermedia. × 40.

Fig. 5.—Drawing of dissection from ventral surface to show main genital organs (semi-diagrammatic). × 5.

Fig. 6.—Transverse section of wall of typical vas deferens. × 550.

Fig. 7.—Transv. sect. of wall of seminal vesicle. Secretion stained with picro-nigrosin. × 550.

Fig. 8.—Transv. sect. of wall of ductus ejaculatorius.' × 550.

Fig. 9.—Transv. sect. of wall of seminal duct stained with borax-carmine to show two kinds of glands. × 550.

Plate 15.

Fig. 10.—Transv. sect. of wall of oviduct. × 550.

Fig. 11.—Transv. sect. of lining of reservoir. Glandular secretion stained with picro-nigrosin. × 550.

Fig. 12.—Transv. sect. of ovary showing the entrance of the oviduct—slightly diagrammatic. × 85.

Fig. 13.—Transv. sect. of inner end of oviduct showing entrance of tube of amoeboid cells.

Fig. 14.—Transv. sect. of mature germarium. × 400.