I

INTERNAL MORPHOLOGY AND HISTOLOGY OF THE FISH MITE *LARDOGLYPHUS KONOI* (SASA AND ASANUMA) (ACARINA : ACARIDIAE)

1. DIGESTIVE SYSTEM *

By

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Abstract.

The internal morphology and histology of the adult digestive system of *Lardoglyphus konoi* an Acaridiae, are described from serial paraffin sections. The alimentary tract has a typical acarid form, with a well developed stomach armed with a pair of caeca, a distinct colon and a rectum. At least three types ie., flat squamous and unspecialised, tall columnar, and globular cells are present on the walls of the caeca and the stomach. The tall columnar cells are confined to the anterior dorsal wall of the stomach and the anterior two third of the caeca. They contain characteristic spherules which are probably excretory in nature. The globular cells are confined to the hind third of the caeca. The wall of the colon is made mostly of unspecialised cells. The rectal epithelium contains elongated and wedgeshaped cells which give evidences of secretory function. A pair of salivary glands are present but malpighian tubules are not discernible. Differences with the histology of other Acaridiae, mainly the acarids are pointed out.

Introduction

Though the Acaridiae are comparatively small animals, they have become economically important in view of their preference for varied substrata and their profound rate of reproduction. Our knowledge of the internal anatomy of this economically important group of animals is scattered and fragmentary (Nalepa, 1884, 1885 ; Michael, 1901 ; Grandjean, 1937 ; Hughes, 1950 ; Rhode and Oemick, 1967 ; and Kuo and Nesbitt, 1970), and confined mostly to the acarids.

So the internal anatomy and histology of a lardoglyphid which has also the distinction of feeding upon a hitherto not so well reported substratum, namely, dried fish was attempted and the present paper is the first of a series on the histology of the internal organs of the mite.

* Forms part of the thesis of the first author approved for the Doctoral degree by the Kerala University.

Acarologia, t. XVI, fasc. 4, 1974.

Material and Methods.

The mites were cultured in the laboratory on dried anchovy maintained under preferred conditions of temperature and humidity. For sectioning, actively feeding, young adults were isolated, fixed in alcoholic bouin, embedded in paraffin and cut at 5 to 6 microns thickness.

The sections were either stained with iron haematoxylin and eosin, or azan. At least thirty complete sets of serial sections either in the transverse or longitudinal planes were studied for the orientation of the internal organs and detailed histology.

Observations.

The alimentary canal of *Lardoglyphus konoi* (see Fig. 1) starts with the mouth situated in the anterior ventral tip of the body and ends with the anal opening situated mesially in the posterior half of the ventral body wall. The alimentary canal consists of the pharynx, oesophagus, stomach and its caeca, colon and rectum.

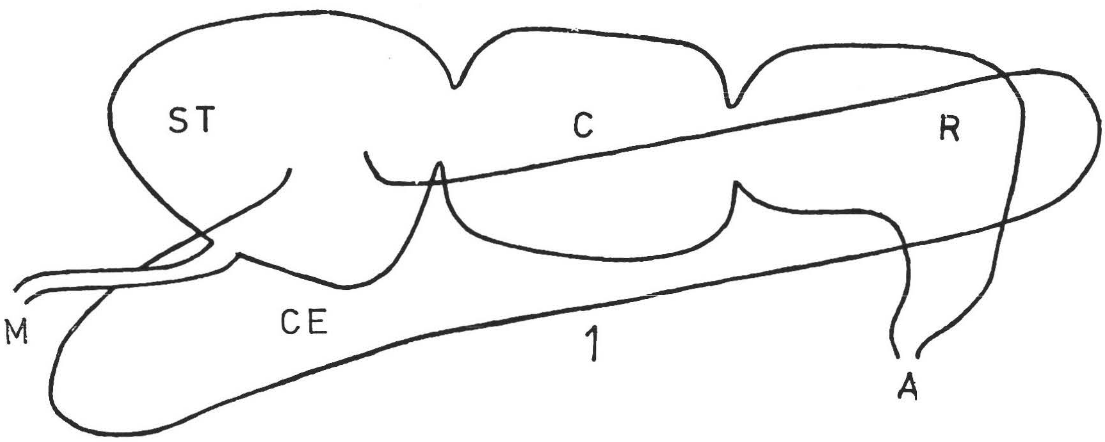


Fig. 1 : Diagramatic representation of the digestive system. ST. stomach ; C. colon ; R. rectum ; CE. Caecum ; M. mouth ; A anus.

Except the firm attachment at the oral and anal ends, the alimentary canal lies somewhat loosely in the body cavity, held in position by five pairs of connective tissue strands emerging from the hypodermis and investing themselves on the outer surface of its wall. The anteriormost pair among these support the alimentary canal at the junction between the stomach and the colon, the next three pairs the colon at its lateral walls, and the hindmost pair, the rectum at its posterior half.

Pharynx.

The pharynx starts with a crescentic mouth opening situated at the distal end of the gnathosoma, and runs longitudinally backwards and upwards along its median line.

As in the typical acarines the pharynx in *L. konoi* is formed of two chitinous, crescentic half tubes, each joined together along the lateral margins. The two halves enclose a furrow like lumen within, the convexity of which is directed downwards.

The floor of the pharynx is more chitinised than the roof. It is also strengthened by longitudinal folds running the entire length of the pharynx. The roof of the pharynx also is chitinised, but flexible. Two sets of muscles, the dilators and the occlusors are inserted into the dorsal surface of the roof. The dilators are long muscles, and five pairs in number, with the members of the pair arranged in lateral rows on either sides of the median line. Their lower ends are attached to the roof of the pharynx and the other ends to the roof of the rostrum. The occlusors are short muscles, running transversely above the pharyngeal roof, between the two horns of the crescent. They are several in number and arranged one behind the other, along the entire length of the pharynx.

Oesophagus.

About the level of the base of the chelicera the pharynx takes a loop like curve upwards, and opens into the oesophagus. The oesophagus immediately after, almost forms an angle with the pharynx at their junction, and runs upwards and backwards through a median foramen in the brain. Consequent to the formation of an angle with the pharynx the lumen is very nrrow at the junction. The poste rior tip of the oesophagus reaches beyond the wall of the stomach and projects slightly into the lumen as in other acarines.

The wall of the oesophagus is roughly triangular in cross section and completely fills the foramen within the brain even when empty. It is formed of an outer tunic and an inner chitinous tube. The wall appears to be slightly thickened for the entire length towards the median point of all the three sides of the triangle. This is because of the muscle fibres closely apposed to the outer surface of the oesophagus and which run between the ventral muscle mass and the posterior end of the gnathosoma.

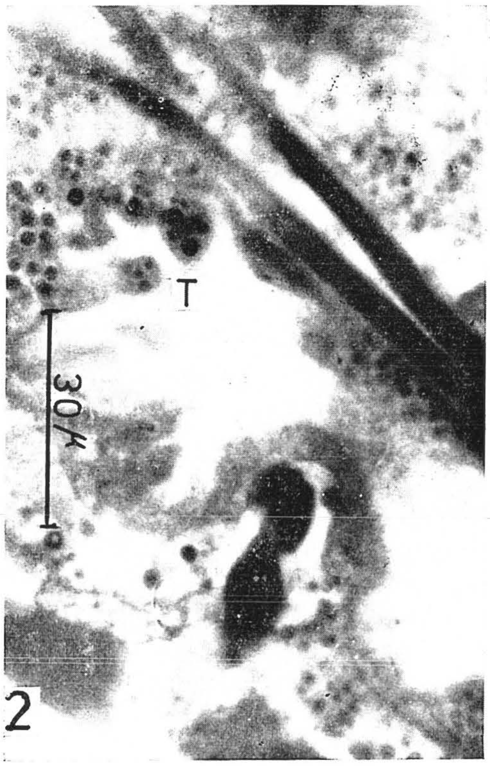
Stomach and its caeca.

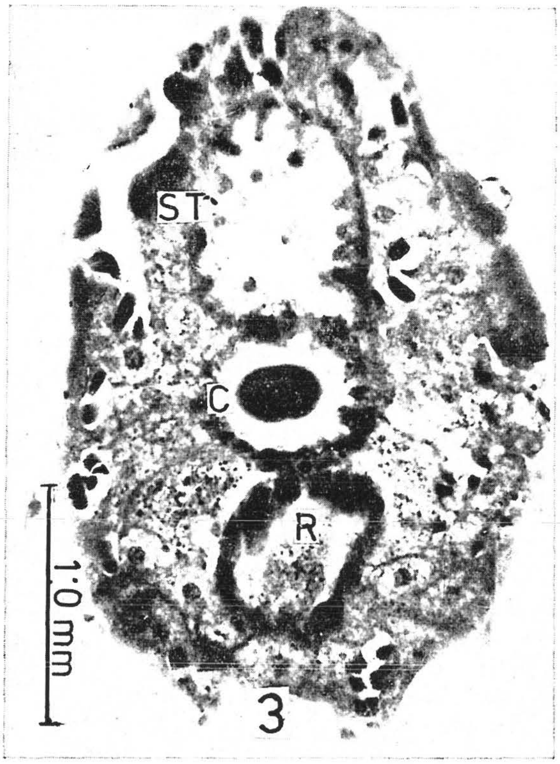
The oesophagus opens posteriorly at the level of the second pair of legs on the ventral side of the anterior half of the stomach (see Fig. 1). The stomach is a pear shaped sac with the narrow end directed forwards. On either sides and slightly in front of the opening of the stomach into the colon, arises a caecum which is blunt at either ends. Anteriorly the caecum is directed obliquely downwards and extends up to the front margin of the propodosoma. Posteriorly it runs obliquely upwards and extends beyond the posterior limit of the rectum. (see Fig. 1).

Outermost on the walls of these structures there is a thin, deeply staining basement membrane. Inner to it is the epithelium. In both the stomach and caeca the epithelium is simple and formed of a single layer of cells. Though only one layer of cells goes into the formation of the epithelium all the cells in the layer are not of uniform characters.

Most of the cells in the anterior two third of the caeca and few cells in the anterior dorsal wall of the stomach (see Fig. 3) are of the columnar type. (T. Fig. 2). They are tall with attenuated bases and with convex distal ends projecting well into the lumen. The cytoplasm though does not show much affinity towards stains is granular. The nuclei are spherical or spindle shaped and basally situated. These cells are characteristically loaded with a large number of deeply stained spherules, (see Fig. 2) which are abundantly present in the body cavity around the caeca also. (see Fig. 2). These spherules appear to be held within a central vesicle within each of the cells. Due to the presence of the vesicle, the protoplasm in each cell is confined to the basal region and to the outer shell of the vesicle. In many cases fragments of these cells loaded with the spherules can be seen fioating in the lumen of the caeca and stomach (see Fig. 3).

The posterior two third of the caeca is formed mostly of globular cells (see Fig. 5). As the very name indicates, these cells project as globules into the lumen. The cytoplasm within these





Figs. 2-3 : Vertical longitudinal section through the anterior region of the caecum. T. tall columnar cells with the contained spherules ; 3) Frontal section through a more ventral plane. ST. stomach with the distal ends of columnar cells floating in the lumen ; C. colon with the contained food ; R. rectum.

cells is very sparse and reduced to a narrow rim around one or two large vacuoles present in the cells. The cytoplasm has not much affinity for stains. Nuclei are basally situated, spherical and prominent. In some cases the distal ends of these cells are disrupted, and protoplasmic threads can be seen projecting into the lumen. At times the distal halves of these cells with the contained vacuoles can also be seen floating in the lumen. (see Fig. 5).

For the rest, the epithelial matrix of the stomach and the caecal walls (see Fig. 3,5) is made up of flattened squamous cells. The cytoplasm of these cells has no inclusions and the nuclei are small, rounded and centrally located. It appears that the flat squamous cells are the unspecialised cells of the epithelium, from which the specialised types ie the columnar and the globular cells take their origin.

The colon.

The stomach is continued into the colon which is a globular sac (see Fig. 1). Anteriorly it opens by a rather wide and circular opening into the posterodorsal wall of the stomach and posteriorly by a similar opening into the rectum. The colon usually contains a digested or partly digested food bolus (see Fig. 3, 4) which has a solid appearance and which is covered over by a peritrophic membrane.

Outermost on the wall of the colon is a thick lamella which forms the base to the epithelial cells. The epithelium is of the simple type and formed of a single layer of cells. Unlike the stomach and the caeca, the epithelium of the colon is mostly pavement like (see Fig. 4) and formed of flattened, squamous cells. The cytoplasm has no inclusions but is localised along dense tracts across the bases of the cells. Because of the presence of the dense tracts of protoplasm, sections will give the appearance of wavy bands towards the base of the epithelium. The nuclei of the cells are small, spherical or oval, and situated towards the free border.

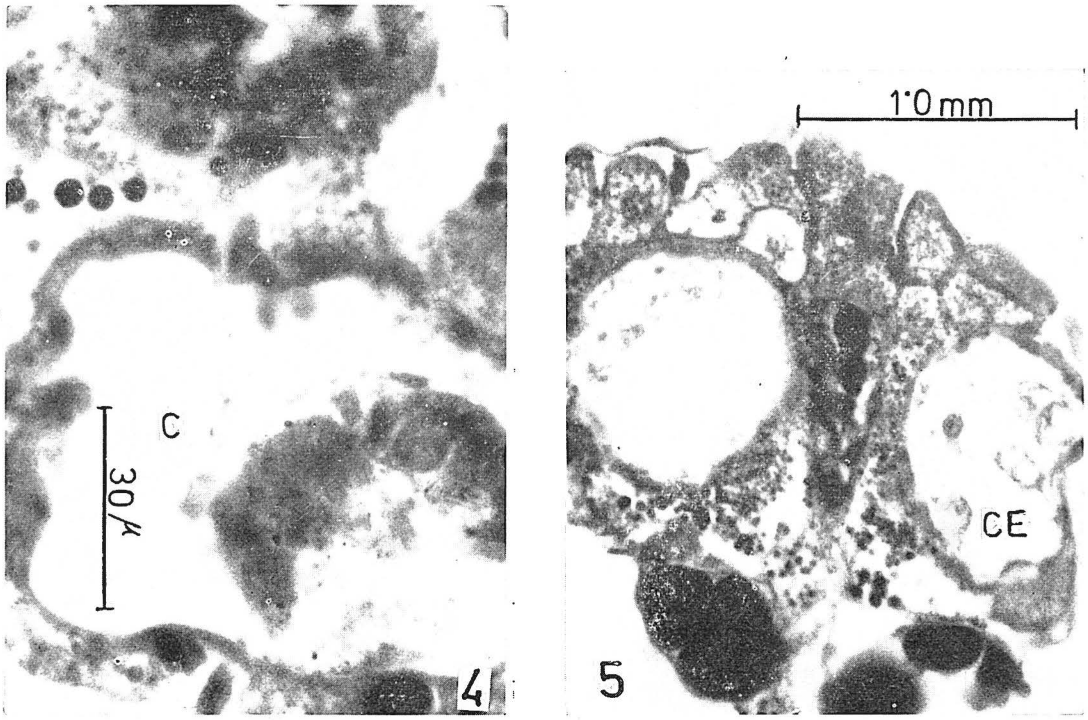


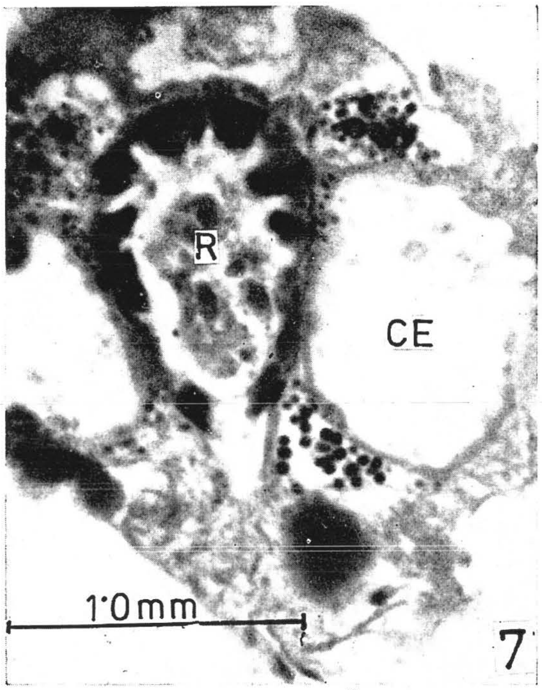
FIG. 4-5 : Transverse section of the colon. C. colon ; 5). Transverse section through the posterior region of the body. CE. caecum showing the globular cells of the epithelium.

In some cases very few cells in the epithelium of the colon are columnar (see Fig. 3) with a club shaped distal end projecting into the lumen. Cytoplasm of these cells may contain one to few vacuoles. The nuclei are small, spherical and basal in position.

Rectum.

The rectum is an elongated, funnel shaped structure, (see Fig. 1) with its broad mouth opening into the posteroventral wall of the colon. In the posterior half, the rectum descends almost vertically downwards, and opens to the outside by a median slit, the anal opening, in the ventral body wall.

As in the preceding regions the outer surface of the wall of the rectum is invested with a basement membrane. Inner to the basement membrane there is the epithelium which consists of a single layer of cells. Most of the cells in the epithelium are tall and wedge-shaped. (see Fig. 7). These cells are wider at the bases and appear triangular in sections. Though bathed by body fluid containing characteristic spherules, (see Fig. 7) in contrast to the tall cells present at the distal end of the caeca, none of these spherules are present in the tall cells of the rectum. The cytoplasm within these cells shows the characteristic presence of a basophilic zone which is rather extensive towards the centre of the cells. This zone of basophilic cytoplasm is enveloped on all sides by a light staining rim of homogeneous cytoplasm. The nuclei are comparatively large and vesicular and located towards the bases of the cells. Distributed between the columnar cells there are a few short squamous cells also. Their cytoplasm is homogeneous and contains a centrally located, small, spherical nucleus.



FIGS. 6-7 : Magnified view of the salivary gland. SG. salivary gland ;
7) Transverse section through the rectum and posterior caeca. R. rectum ; CE. caecum.

For a short distance from the anal opening the structure of the rectal wall is slightly different from the preceding region of the rectum. Here the epithelium is formed of flat squamous cells and their border towards the lumen is lined by chitin. In view of this difference, the distal portion with chitinous lining may be termed as « anal atrium ».

Salivary glands.

They consist of a pair situated ventrolaterally in the anterior end of the body cavity, on either sides of the narrow longitudinal neck of the stomach. The gland is closely lodged between the side of the stomach and the lateral wall of the anterior half of the caeca. The proximal end which is blunt and wider, is directed into the body cavity and reaches up to the point where the caecum arises from the stomach. The anterior end of the gland which is tapering, runs gently outwards and downwards and reaches up to the base of the chelicera. The gland is bounded by a limiting membrane formed by connective tissue sheath. The gland proper consists of large cells which are highly vacuolated and contain prominent, deeply staining nuclei. Because of the vacuolation, the cytoplasm is very sparse within the cells and is reduced to a network, holding the nuclei in position. (see Fig. 6).

Malpighian tubules.

Malpighian tubules were first described in *Tyroglyphus farinae* by BERLESE (1897). MICHAEL (1901) has observed that in Tyroglyphidae, the malpighian tubules may be comparatively small and sometimes even apparently absent altogether. VITZTHUM (1943) says that the malpighian tubules of such tyroglyphid that possesses them are always devoid of contents, thereby implying that at least in some tyroglyphids the malpighian tubules are absent. Though in *T. farinae* Hughes. (1950), noticed a pair of malpighian tubules, according to her, the tubules play little part in the excretion of guanine. In *L. konoi* several animals which were sectioned have failed to reveal the malpighian tubules or any comparable structures opening into the region of the junction between the colon and the rectum, or into any of the hinder segments of the alimentary canal.

DISCUSSION.

According to Hughe's (1950) nomenclature, the stomach and its caeca of the present description will correspond to the anterior stomach and caeca, the colon to the intermediate colon, and the rectum to the post colon, and, these will comprise the mesenteron of her description. The region up to the oesophagus will correspond to the foregut, and the anal atrium to the hindgut of her description.

Since a chitinous lining for the oesophagus had been reported among the Acari only in the Trombidiformes (SIG THOR, 1904), the Halacaridae (THOMAE, 1925), and the Analgesidae (LONNFORS, 1930), HUGHES (1950) has doubted the soundness of the homology of the acarine oesophagus, with that of the other arachnids where it forms a part of the oesophagus. As reported by HUGUES herself in *T. farinae* (1950) and later in *Caloglyphus mycophagus* (Rhode and Oemick, 1967 ; Kuo and Nesbitt, 1970), the present study reveals the presence of a chitinous lining for the oesophagus and thus strengthens the homology of the acarine oesophagus with that of other arachnids.

According to LONNFORS (1930) the sarcoptiform type of stomach can be derived by the loss of all but the posterior pair of caeca and the gradual loss seems to be represented by the present day species. For example the anterior pair has been reduced to the proventricular gland in Oribatidae as described by MICHAEL (1883). The Analgesidae represent still further reductions ; first the proventricular glands occur as a pair of patch of cells on the anterior wall of the stomach then as a single median patch above the opening of the oesophagus formed by the fusion of the originally paired rudiments. In HUGUES' (1950) view the condition in *T. farinae* with the presence of large cells anterodorsally in the stomach wall represents the last mentioned condition of reduction and the tall vacuolated cells at the posterior end of the caeca are homologous to the postventricular glands. But KUO and NESBITT (1970) have doubted this homology because in the species they have studied, the tall cells are not limited to the anterior regions of the gut or the posterior end of the caeca. In *Lardoglyphus* the tall type of cells are confined to the anterior regions of the stomach and caeca alone, and the stomach is the sarcoptiform type, thereby strengthening the postulate of reduction said to have taken place in the present day species (LONNFORS, 1930).

It would appear that the number of the dilator muscles of the pharynx may vary between the acarids and the lardoglyphids. In *Glycyphagus platygaster* (Michael 1901) and *T. farinae* (Hughes 1950) they are five pairs ; in the present species only four pairs are present.

MICHAEL (1901) shows the presence of ring muscles as in the oribatids around the pharynx ot tyroglyphids also. Such muscles are however absent in the present species. It is noteworthy in this connection that later authors of the internal anatomy of Acaridiae (HUGHES, 1950 ; RHODE and OEMICK, 1967 ; and KUO and NESBITT, 1970), do not speak of the ring muscles on the pharynx.

In *C. mycophagus* (Acaridae) the ventral wall of the pharynx according to RHODE and OEMICK (1967) is held rigid by an invagination of the integument. Such an arrangement is not present in the lardoglyphid.

Accounts of *Tyroglyphus longior* and *Carpoglyphus anonymus* (Nalepa, 1884.), Tyroglyphidae (Michael, 1901.), and *C. mycophagus* (Rhode and Oemick, 1967 ; Kuo and Nesbitt, 1970.) are suggestive that the oesophageal wall of the Acaridiae has no epithelium. But as evidenced by the present study, there is an epithelial layer though narrow, and formed of short, flattened cells, beneath the chitinous intima.

MICHAEL (1901) has observed that probably because they feed on solid food, facility is provided both in the Tyroglyphidae and Oribatidae to sufficiently dilate the oesophagus, and its wall does not fill completely the foramen in the brain through which it passes. Such a conjecture based on the nature of food is not borne out by the present species, because, even though it also feeds solely on solid food, the oesophageal wall even when empty, completely fills the cavity within the brain.

Among the Acaridiae so far investigated, three types of cells within the stomach and caecal epithelium, as noticed in the present species, have been described only in the species *C. mycophagus* (Kuo and Nesbitt, 1970). In the same species RHODE and OEMICK (1967) unambiguously mention the presence of only two types, namely the squamous type of cells, and vacuolated cells, in these regions. But if the " glands " which they mention as present in the posterior lateral caecum correspond to the " globular cells " of KUO and NESBITTS' description, the two descriptions concur in the presence of three types of cells in the epithelium of the stomach and caecaum of the species.

But, however, these three types of cells described in the acarid, do not wholly correspond with the three types of cells of *Lardoglyphus* histologically ; only the squamous cells and the globular cells seem to tally with the a and c types respectively of *C. mycophagus*. It needs mention however that the a type or the squamous cells of *C. mycophagus* has numerous fine crystalline granules within them. In *L. konoi* the squamous cells are unspecialised; their cytoplasm is homogenous. The b type or the cuboidal type of cells of *C. mycophagus* is greatly different from the tall columnar cells of *L. konoi*. The only points of resemblances between the two types of cells is the presence of crystalline deposits (spherules or granules) within the cells, and also in the process of their distal tips pinching off, and freely floating with the contained deposits, within the lumen of the gut. But, many vacuoles are present in the cuboidal cells of *C. mycophagus*, reducing the cytoplasm within them to a reticulum. In the place of the vacuoles of these cells, a large number of spherules only, are present in the columnar cells of *L. konoi*. Morphologically also, the two types of cells are different ; the bases of the columnar cells are attenuated, whereas those of the cuboidal cells are broad. The cuboidal cells are more suppressed, and not so tall as the columnar cells of *L. konoi*.

The columnar cells of *L. konoi* resemble to a large extent in appearance and position, the « clavate cells » described by MICHAEL (1901) in the anterior wall of the stomach and caeca of the tyroglyphids.

In the feature of containing crystalline concretions within the cells, and in the free floating of the cell remnants with the contained concretions within the lumen, shown by the tall columnar cells of the present species, near equal condition is obtained in addition to the cuboidal cells of *C. mycophagus*, by the " clavate cells " described by MICHAEL (1901) in the ventriculus and the anterior caecum of Tyroglyphidae and the " taller cells " described by HUGHES (1950) at the anterodorsal region of the caecum of *T. farinae*. It may however be mentioned here, that these latter types of cells have their own differences from the columnar cells of *L. konoi*.

In the present species it was indicated, that the distribution of the columnar type of cells is restricted to the anterior portions of the stomach and caeca. Such localisation of a type of cell to a particular region of the gut does not seem to occur in the acarids, because, in *C. mycophagus* the cuboidal cells, though more abundant in the anterior caeca extend to the lateral sides of the posterior caeca also (Kuo and Nesbitt, 1970), and in *T. farinae* also the tall cells which are more abundantly present in the anterior caeca, extend to the posterior caeca also (Hughes, 1950).

The wall of the colon seems to be different in the acarids and *Lardoglyphus*. In *C. mycophagus*, the wall is formed of tall celiated (Rhode and Oemick, 1967.), or conical columnar (Kuo and Nesbitt, 1970.) cells and in *G. platygaster*, the cells at the two ends of the organ differ ; those at the anterior end are flattened while those at the posterior end are smaller and thicker (Michael, 1901). In *L. konoi* the wall of the colon is almost wholly formed of squamous cells and only a few elongated cells are present and they are distributed at random on the wall.

The rectal cells of the acarids and lardoglyphid seem to concur in the general shape and form, but the presence of extensive basophilic zones in the cells of *L. konoi* is significant.

A brush border for the rectal cells of *C. mycophagus* (Kuo and Nesbitt, 1970.), a striated border on the flat and vacuolated cells of the stomach and caeca, and celiated border for the tall cells of the colon in the same species (Rhode, and Oemick, 1967.), and a striated border for the rectal cells of *T. farinae* (Hughes, 1950), have been noticed. In *L. konoi* a striated border or celiated edge could not be found in any cell in any part of the gut. What is present is few protoplastic threads projecting from the free edges of a few apparently active cells only.

All except Kuo and Nesbitt (1970) found that the salivary glands in acarids open by a pair of ducts into the buccal cavity, and Lonnfors (1930) and Rhode and Oemick (1967) found them open above the coxa of leg one. In the lardoglyphid the salivary glands open at the posterior aspect of the buccal cavity.

Even though extensive musculature has been cited by Michael on the colon and stomach of Oribatidae, and Vitzthum (1943) speaks in a general way of acarids as having muscle coat posterior to the stomach, the present study confirms the absence of any intrinsic muscle on the alimentary canal of the Acaridiae in conformity with the finding of Michael (1901) in the Tyroglyphidae and Hughes (1950) in *T. farinae*.

Acknowledgement.

The senior author is grateful to Dr. N. Krishna Pillai, Head of the Department of Aquatic Biology and Fisheries for giving all the facilities and to the University of Kerala for financial support through a Senior fellowship.

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