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Studies on the Various Glands of the Tick *Haemaphysalis spinigera* Neumann 1897.*

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Part I. Studies on the Male Accessory Genital Glands.

Introduction.

The histology of the male accessory gland of Ixodid ticks has been described by RUSER 1933, YALVAC 1939, DOUGLAS 1943, and TILL 1961. Granules of secretion have been observed in the cells of the various lobes of the gland, but the nature of these granules is not known. In this paper, histological techniques have been amplified by histochemical techniques to elicit the cytochemical nature of the secretory granules in the various lobes of this gland.

Materials and Methods.

The ticks were received from Dr. H. Trapido in India, and maintained at a temperature of 25±°C and a relative humidity of 100%. They were fed according to the techniques of Varma. Larvae were fed on litters of 3-day old baby rats and nymphs and adults in sleeves or capsules fixed on the backs of rabbits.

For histological studies, ticks were fixed in formol-sublimate, Sanfelice's and Carnoy's fluids as given in CHARLTON & DRURY (1957); embeded in parafin by the Peterfi's methyl-benzoate/celloidin technique and sectioned at 4-7.5 μ. Sections were stained in haematoxylin-eosin, and Mallory's triple stain.

For histochemical studies, ticks were fixed in special fixatives given below:

1. For the fixation of mucopolysaccharides and mucoproteins the method of LILLIE (1954) and the alcoholic lead nitrate method of MOTA et al. (1956) were used.

2. The method of WILLIAMS & JACKSON (1956) was used for the fixation of acid mucopolysaccharides.

3. 70% solution of alcohol was used for the fixation of glycogen in tissues.

4. 10% solution of formalin was used for the fixation of proteins.

5. Formol-sublimate fixative BHARRADWAJ & LOVE (1959) was used as fixative for lipids.

6. Carnoy's fixative was used for the fixation of nucleic acids in tissues. Both Carnoy's fixative and Palade's Osmium were found to be good fixatives for the ground cytoplasm.

The triple stain of HIMES & MORIBER (1956), for D.N.A., polysaccharides, and proteins was modified by the substitution of thionin-SO_2 reagent of DUIJN (1956), in place of Azure-Schiff's reagent, and used as a survey stain. The

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P.A.S. stain of McMANUS (1946) was used to locate polysaccharide grouping. Loss of P.A.S. staining following the action of 1% diastase at 37°C for two hours was taken as an indication of the presence of glycogen. Glycogen was also tested for by the Best's carmine method as given in PEARSE (1961). For the localization of mucoproteins and glucoproteins, the Bismarck brown-Y method of LEACH (1947) and the metachromatic staining method of HESS & HOLLANDER (1947) were used. For the detection of glycolipids slides were treated with methanol/chloroform solution at 60°C for 6-12 hours prior to P.A.S. staining. For the identification of acid mucopolysaccharides, the alcian blue method of STEEDMAN (1950) (see PEARSE 1961) and Müller's colloidal ferric oxide method of MOWRY (1958) as well as the metachromatic staining method of HESS & HOLLANDER (1947) were used. For the identification of basic proteins, the naphthol-yellow-S method of DEITCH (1953, 1955) and the mercuric-bromphenol blue method of BONHAG (1955) were used. BAKER's (1956) modification of the Millons reaction described in GURR (1958) was used for the identification of tyrosine groups. The D.M.A.B.-nitrite method of ADAM (1957) was used to localise tryptophane groups. Arginine was identified by the method of BAKER (1947). The methods of ADAM & SLOPER (1955, 1956) and the alkaline tetrazolium reaction given in PEARSE (1961) were used to identify groups -SS and -SH respectively. Positive results were confirmed by blocking methods as suggested by DANIELLI (1953).

Lipids were identified by the method of BHARRADWAJ & LOVE (1959) and the Sudan black B method (see PEARSE, 1961). D.N.A. was identified by the Feulgen-Schiff technique given in CASSELMAN (1959) and R.N.A., by the methods of KORSON (1951) and KURNICK (1955), positive sites being digested with ribonuclease.

The Male Accessory Genital Gland: Adult Stage.

Morphology.

Lying on the floor of the body cavity and just behind the brain is the accessory genital gland referred to as the white gland by CHRISTOPHERS (1906). It consists of several lobes, the most conspicuous of which is the dorso-median lobe (d.m.l.), which DOUGLAS (1943), divides into antero-dorsal and postero-dorsal lobes (Fig. 1). When the dorso-median lobe is lifted and drawn backwards (Figs. 1 and 2), it is found to run insensibly into a median duct, which, because of the fact that it receives the other lobes of the accessory gland, will be referred to as the collecting duct (c.d. Fig. 1). This finally opens into the ejaculatory duct (ej.d.). Beginning posteriorly, it receives the following lobes along its course to the ejaculatory duct:

1. a pair of postero-ventral lobes (p.v.l.),
2. a pair of lateral lobes which divide into dorso-lateral (d.l.l.), and postero-lateral lobes (p.l.l.),
3. a pair of latero-ventral lobes (l.v.l.),
4. a single medio-ventral lobe (m.v.l.), and
5. a pair of antero-ventral lobes (a.v.l.) (see Figs. 1 and 2).

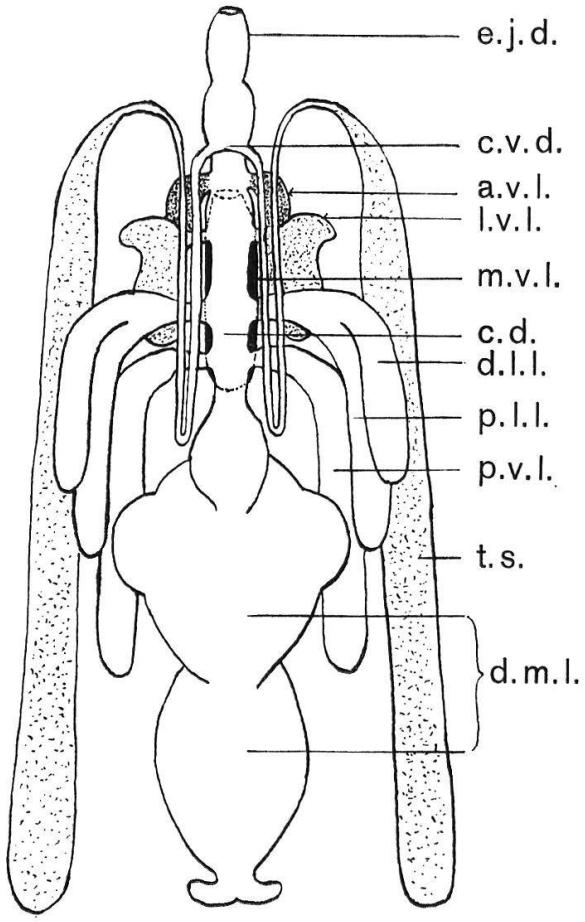

Fig. 1. Unfed male. Drawing of a dorsal view to show the disposition of the genital organs. The dorso-median lobe has been drawn backward. ej.d.: ejaculatory duct, c.v.d.: common vas deferens, a.v.l.: antero-ventral lobe, l.v.l.: latero-ventral lobe, m.v.l.: medio-ventral lobe, c.d.: collecting duct, d.l.l.: dorso-lateral lobe, p.l.l.: postero-lateral lobe, p.v.l.: postero-ventral lobe, t.s.: testis, d.m.l.: dorso-median lobe.

Histology and Histochemistry.

Unfed Males.

In the unfed males, the cells composing the various lobes have a clear staining cytoplasm with few or no secretory granules. The nuclei are closely packed. The lobes are enclosed in a thin connective tissue coat overlain by a single layer flattened of epithelial cells.

Feeding Males.

During engorgement, the accessory gland as a whole increases considerably in size, and the structure of the cells becomes more distinct. The cells of the various lobes are filled with secretory granules of various sizes which stain to varying intensities in

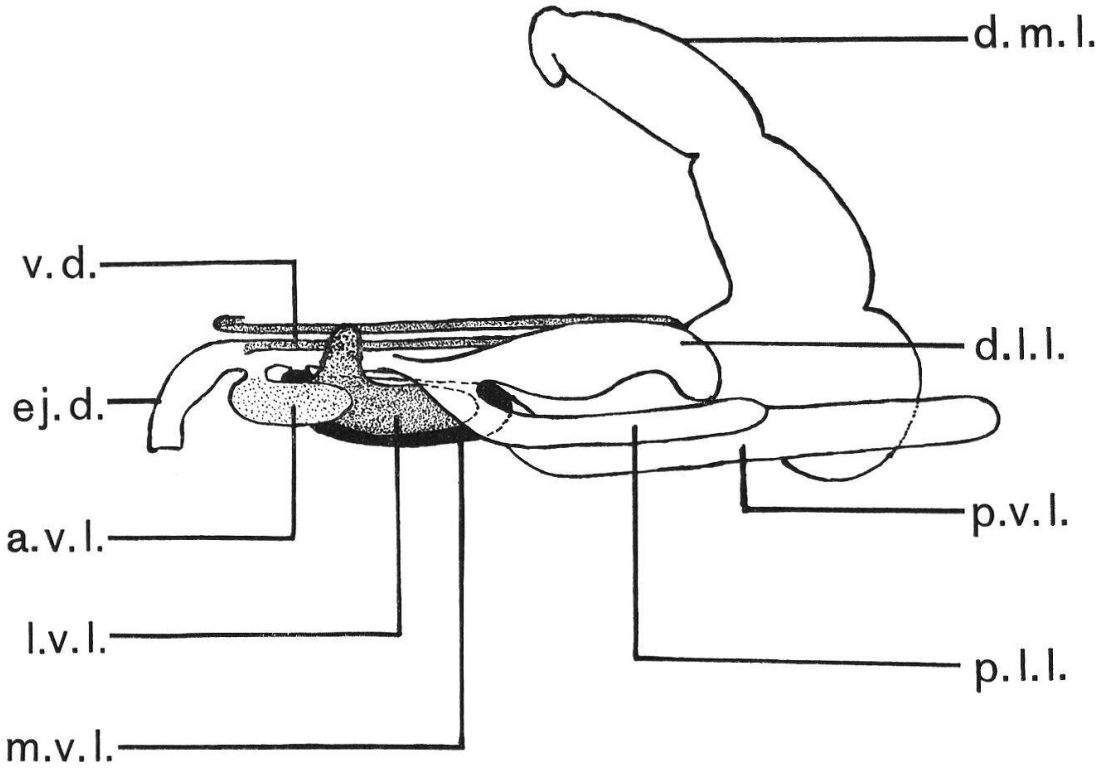

Fig. 2. Unfed male. Drawing of a side view of the accessory genital gland. d.m.l.: dorso-median lobe, d.l.l.: dorso-lateral lobe, p.v.l.: postero-ventral lobe, p.l.l.: postero-lateral lobe, v.d.: vas deferens, ej.d.: ejaculatory duct, a.v.l.: antero ventral lobe, l.v.l.: latero-ventral lobe, m.v.l.: medio-ventral lobe.

haematoxylin-eosin stain. They have been classified as large granules (l.g.), small granules (s.g.), and fine granules (f.g.). To shed further light on the nature of the secretory granules various histochemical techniques were used, the results of which are given in Table 1.

Collecting Duct (c.d. Fig. 6a): This part of the gland referred to as the median part of the gland by TILL (1961), is in reality a ventral prolongation of the dorso-median lobe. The cells vary from tall columnar in the posterior region to flattened ones in the anterior region. In the latter region the cells are devoid of secretory granules, whilst in the posterior regions they are filled with small granules. The nuclei are situated at the bases of the cells. The secretory granules are P.A.S. negative staining mainly for basic proteins. Further histochemical tests indicate the presence of disulphide groups (Fig. 6b).

Antero-Ventral Lobe (a.v.l. Fig. 3a): The walls of these lobes are composed of columnar cells enclosing a small lumen. The nuclei are situated at the base of the cells, whilst the rest of the cells are filled with small granules (s.g. Fig. 3b). These granules stain intensely with P.A.S., and mercuric-brom-phenol blue and naphthol-yellow stains indicate the presence of basic proteins. The

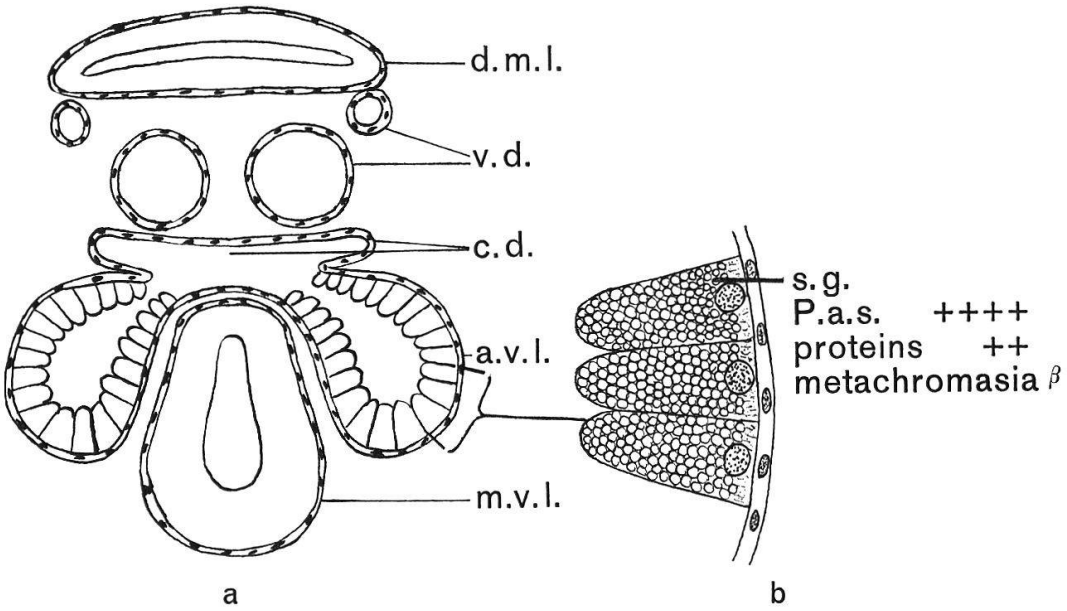

Fig. 3 a. Transverse section through the accessory genital gland of a male attached to its host for 2 weeks. (Drawn from sections stained in haematoxylin-eosin after formol-sublimate fixation.) d.m.l.: dorso-median lobe, v.d.: vas deferens, c.d.: collecting duct, a.v.l.: antero-ventral lobe, m.v.l.: medio-ventral lobe.

Fig. 3 b. Magnified drawing of cells of the antero-ventral lobe to show the histochemical nature of the cells. s.g.: small granules.

granules give β-metachromasia with toluidine blue. They do not stain with alcian blue or colloidal iron, and this, together with the strong P.A.S. staining, suggests that the material may be a muco-protein.

Medio-Ventral Lobe (m.v.l. Fig. 4a): The walls of this lobe are composed of columnar cells enclosing a slit-like lumen which in males which have attached for more than one week becomes much wider and is filled with a colloidal secretion. The nuclei are situated at the base of the cells. The small secretory granules which accumulate in these cells are P.A.S. negative. They contain mainly basic protein rich in tyrosine groups (Fig. 4a, d).

Latero-Ventral Lobe (l.v.l. Fig. 4a): The cells which form the walls of these lobes are small and cubical to columnar in shape with nuclei lying at the bases of the cells. The secretory granules are of two different sizes, and are confined to different portions of the gland. In the distal part of the gland the cells contain fine granules (f.g. Fig. 4b), whilst in the proximal region of the gland the cells contain coarse granules (s.g. Fig. 4c). Histochemical methods used so far did not reveal any difference between the fine and the coarse granules. They were P.A.S. negative, and stained with equal intensity for basic proteins, and further histochemical tests indicated the presence of disulphide groups.

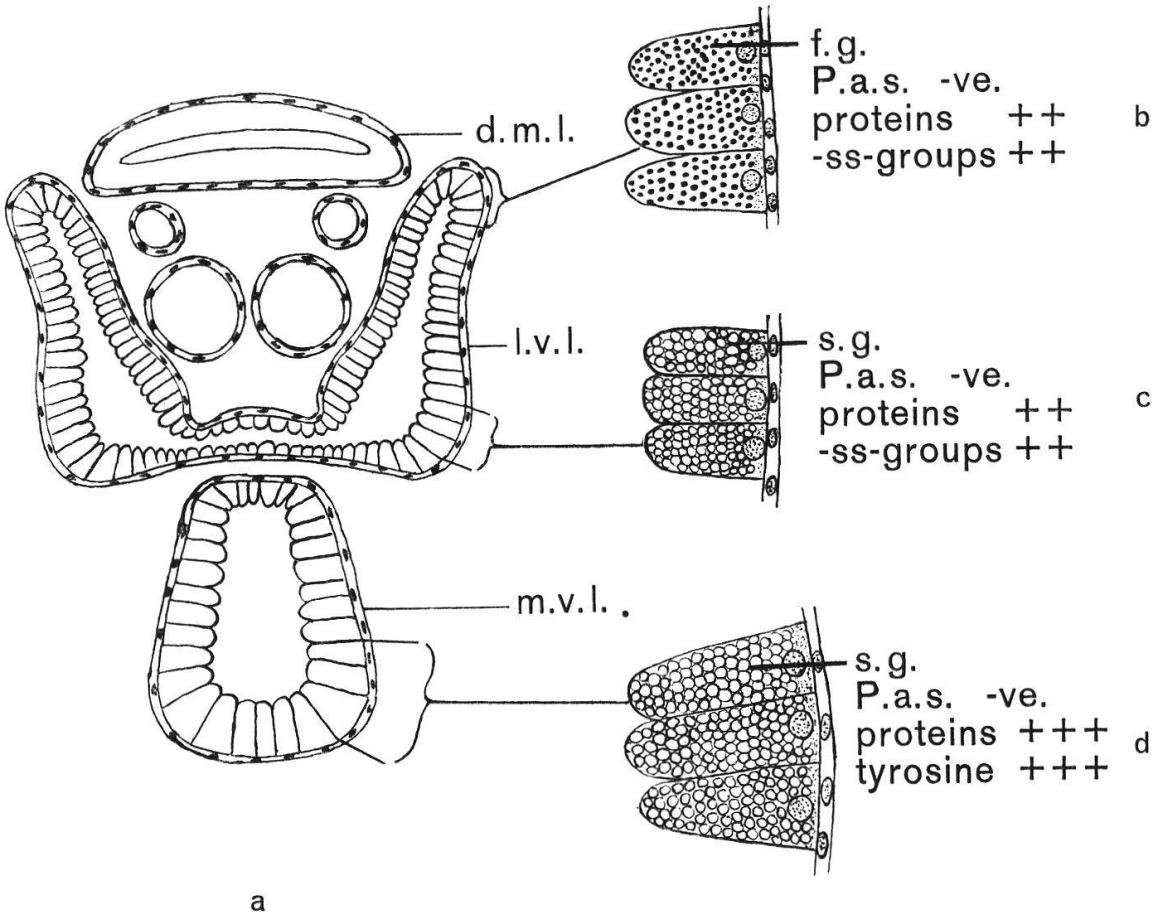

Fig. 4 a. Transverse section through the accessory genital gland of a male attached to its host for 2 weeks. (Drawn from sections stained in haematoxylin-eosin after formol-sublimate fixation.) d.m.l.: dorso-median lobe, l.v.l.: latero-ventral lobe, m.v.l.: median-ventral lobe.

Fig. 4 b, c, d. Magnified drawings of cells of the latero-ventral lobe, *b* and *c*, and medio-ventral lobe *d* to show the histochemical nature of the cells. s.g.: small granules, f.g.: fine granules.

Dorso-Lateral Lobe (d.l.l. Fig. 5a): These lobes are made up of columnar cells with vacuolate cytoplasm containing small granules. The nuclei lie at the bases of the cells. In between these granular cells are a few isolated cells containing a homogeneous cytoplasmic material. The small secretory granules contain mainly proteins, whilst the other isolated cells which contain the colloidal cytoplasmic material (c.cy. Fig. 5b) stain moderately with P.A.S. and also contain a tyrosine-containing protein.

Postero-Lateral Lobes (p.l.l. Fig. 5a): The histological and histochemical appearance of the cells of this lobe are similar to those of the dorso-lateral lobe with which it unites before joining the collecting duct.

Postero-Ventral Lobes (p.v.l. Fig. 6a): The histological appearance of these lobes differs in the different regions of the gland. In

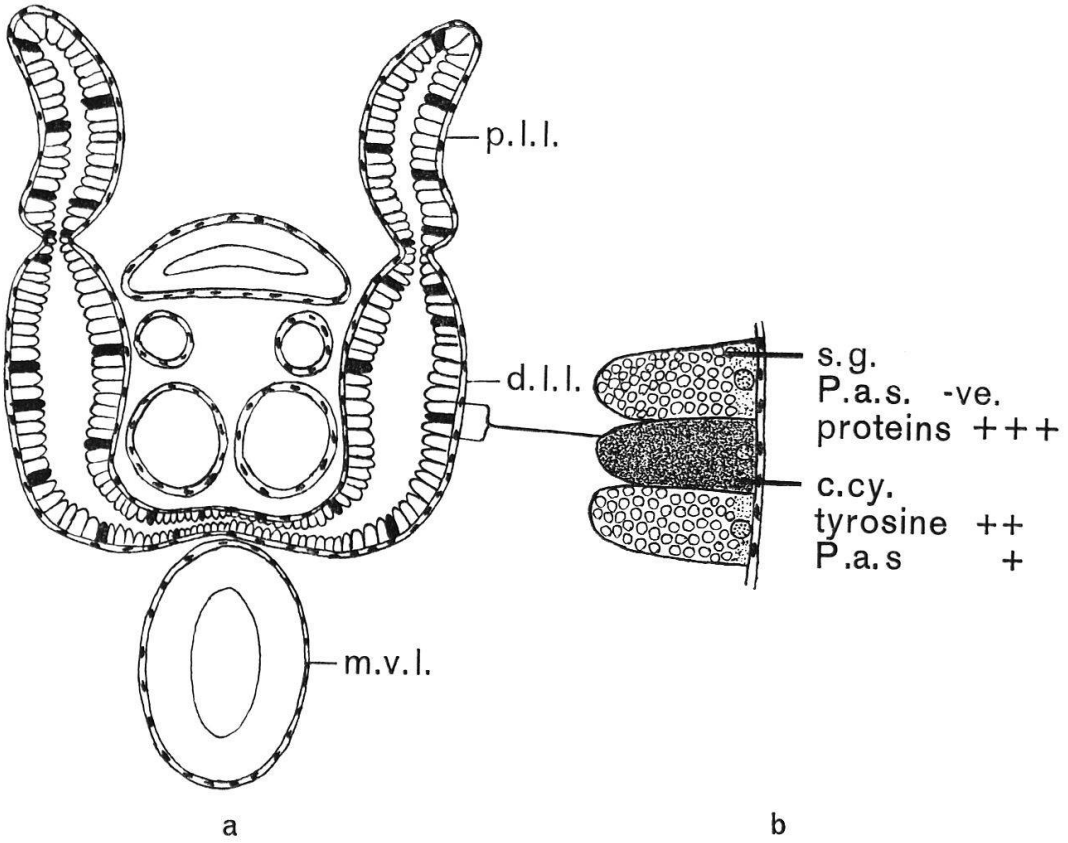

Fig. 5 a. Transverse section through the accessory genital gland of a male attached to its host for 2 weeks. (Drawn from sections stained in haematoxylin-eosin after formol-sublimate fixation.) p.l.l.: postero-lateral lobe, d.l.l.: dorso-lateral lobe, m.v.l.: median-ventral lobe.
Fig. 5 b. Magnified drawing of cells of the dorso-lateral and postero-lateral lobes to show the histochemical nature of the cells. c.cy.: colloidal cytoplasm, s.g.: small granules.

the proximal region of this gland there are two types of cells. In the region of the gland adjoining the collecting duct the cells are columnar with small nuclei situated in the basal region of the cells. These contain closely packed small secretory granules. These granules contain mainly basic protein apparently linked to a moderately P.A.S. positive material. They also contain moderate amounts of lipids and disulphide groups (Fig. 6c). Next to these cells in the posterior direction are large cubical to dome-shaped cells with homogeneous finely granular cytoplasmic material. The nuclei are large and are situated at the bases of the cells. The finely granular cytoplasmic material in these cells is P.A.S. negative; it contains mainly basic protein associated with lipids and disulphide groups (Fig. 6d). This material is released into the lumen of the gland in the form of large globules which coalesce to form a gelatinous secretion in the lumen of these lobes. Scattered among these dome-shaped cells are slender club-shaped cells

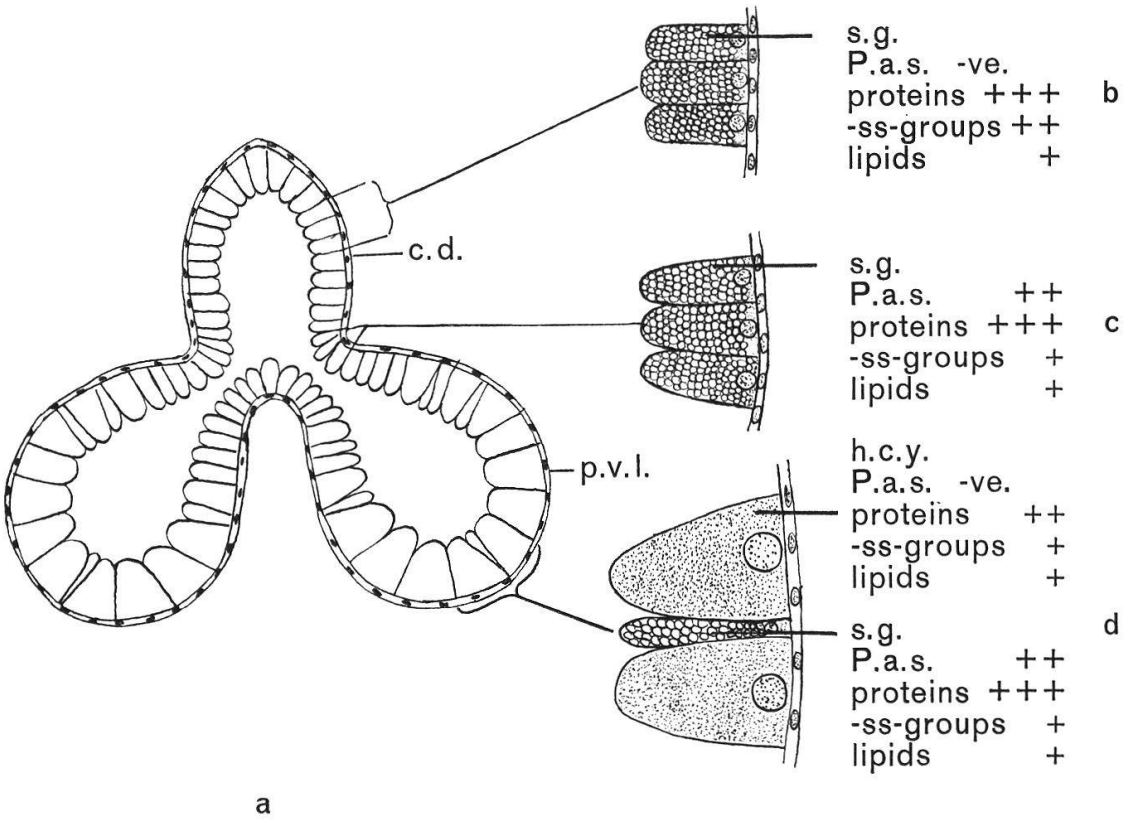

Fig. 6 a. Transverse section through the accessory genital gland of a male attached to its host for 2 weeks. (Drawn from sections stained in haematoxylin-eosin after formol-sublimate fixation.) c.d.: collecting duct, p.v.l.: postero-ventral lobe.

Fig. 6 b, c, d. Magnified drawings of cells of the collecting duct (c.d.) and postero-ventral lobe (p.v.l.) to show the histochemical nature of the cells. h.cy.: homogeneous cytoplasm, s.g.: small granules.

filled with small secretory granules. These granules contain mainly basic proteins and stain with moderate intensity in P.A.S. They also contain some lipids and disulphide groups (Fig. 6d s.g.). The distal portion of the gland is occupied by tall columnar cells with large nuclei located in the mid portion of the cells. Above the nuclei the cells are packed with small secretory granules which contain basic proteins apparently linked to a moderately P.A.S. positive substance. They also contain moderate amounts of lipids and disulphide groups. Below the nuclei, the cytoplasm is finely granular and is mainly protein in nature, but contains traces of lipids and disulphide groups (Fig. 7b).

Dorso-Median Lobe (d.m.l. Fig. 8). The sizes of the secretory granules in the cells of this lobe varied a great deal. This, together with the variation of the histochemical nature of the granules in the different parts of the gland, made the analysis of histochemical results on this gland difficult. This can be seen after careful

TABLE 1.

Histochemical Results on Accessory Genital Glands of Feeding Males.

Methods		A.V.L.	M.V.L.	L.V.L.		C.D.	D.L.L.	P.D.L.	P.V.L.
		s.g.	s.g.	f.g.	s.g.	s.g.	s.g.	s.g.	s.g.
A *Polysaccharides*									
P·A·S·		++++	–	–	–	–	–	–	++
Glycogen	Diastase / P·A·S·	–	–	–	–	–	–	–	–
	Best's Carmine	–	–	–	–	–	–	–	–
Mucoproteins Glycoproteins	Pepsin / P·A·S·	–	–	–	–	–	–	–	–
	Bismarck Brown - y	–	–	–	–	–	–	–	–
	Metachromasia	β	–	–	–	–	–	–	–
Glycolipids	- Methanol: Chloroform								
	P·A·S·	–	–	–	–	–	–	–	–
Acid Mucopolysaccharides	Colloidal Iron	–	–	–	–	–	–	–	–
	Alcian Blue	–	–	–	–	–	–	–	–
B *Proteins*									
Basic Proteins	Mercuric Brom-Phenol-Blue	++	+++	++	++	+++	+++	++	+
	Naphthol - Yellow - S	++	+++	++	+++	+++	+++	++	++
Tryptophane	- D·M·A·B - Nitrite	–	–	–	–	–	–	–	±
Tyrosine	- Millon's Reagent	–	+++	–	–	±	–	–	±
Arginine	- Sakagukyi's Reaction	–	–	–	–	–	–	–	–
SS - SH Gps.	- Alkaline Tetrazolium	–	–	–	–	–	–	–	–
- SS - Gps.	- P·F·A·A·B Reaction	+	–	++	++	+	–	–	++
α Amino Acids	- Ninhydrin - Schiff	–	–	–	–	–	–	–	–
C *Lipids*									
Sudan Black B		–	–	–	–	–	–	–	–
Regaud's Haematoxylin		–	–	–	–	+	–	–	+

± indicates a faint positive reaction. The number of plus signs indicates the relative staining intensity. A.V.L.: antero-ventral lobe, M.V.L.: medio-ventral lobe, L.V.L.: latero-ventral lobe, C.D.: collecting duct, D.L.L.: dorso-lateral lobe, P.D.L.: postero-dorsal lobe, P.V.L.: postero-ventral lobe, s.g.: small granules, f.g.: fine granules.

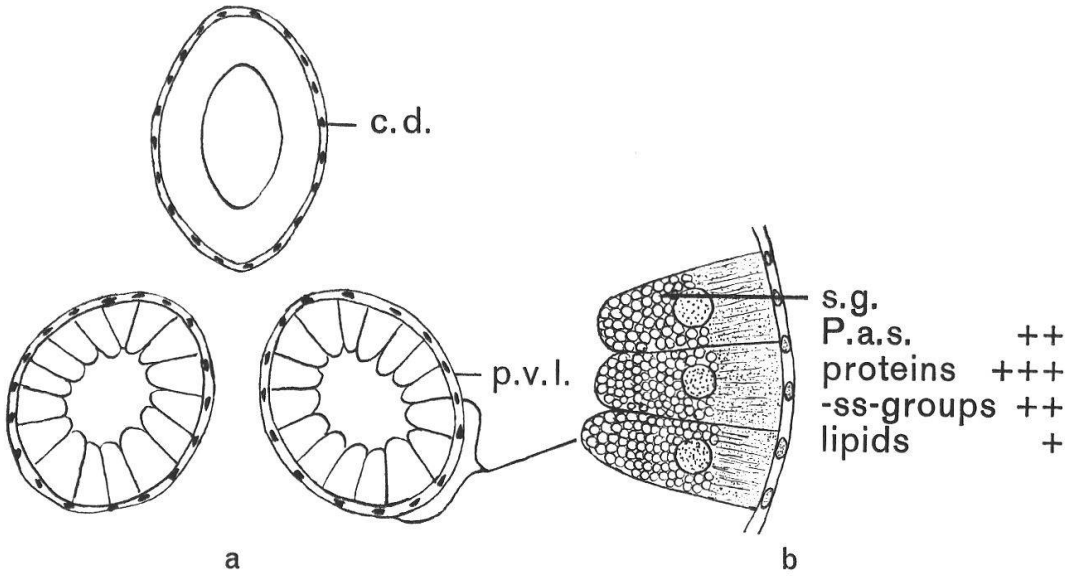

Fig. 7 a. Transverse section through the accessory genital gland of the male attached to its host for 2 weeks. (Drawn from sections stained in haematoxylin-eosin after formol-sublimate fixation.) c.d.: collecting duct, p.v.l.: postero-ventral lobe.

Fig. 7 b. Magnified drawing of cells of the postero-ventral (p.v.l.) lobe to show the histochemical nature of these cells. s.g.: small granules.

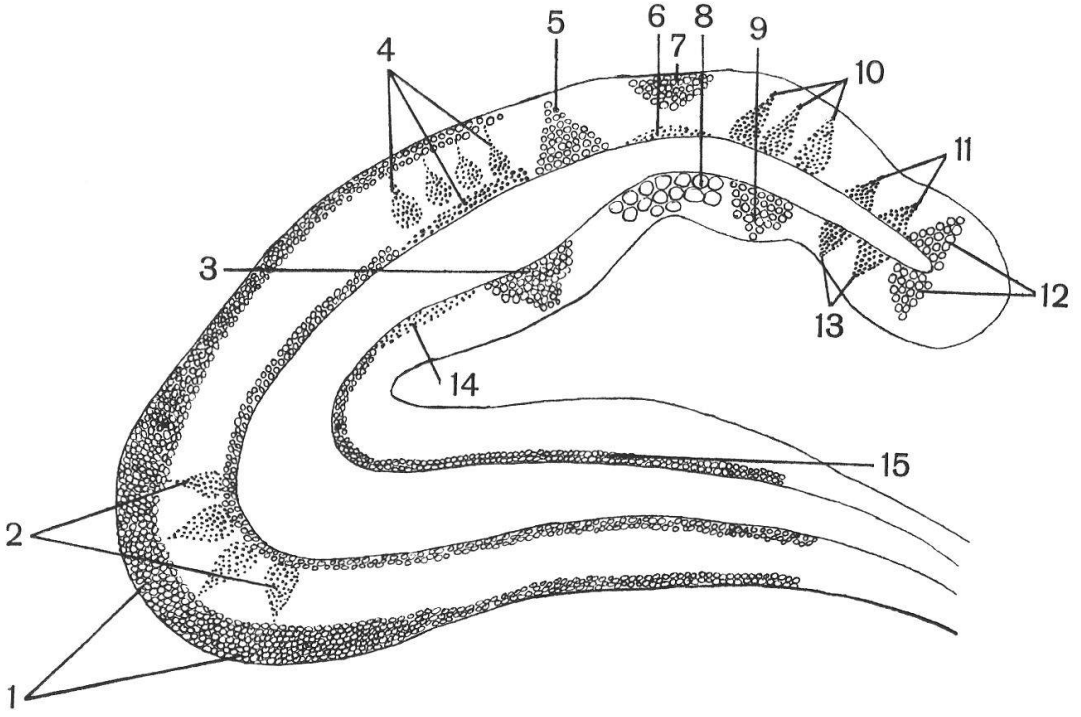

Fig. 8. Drawing of a longitudinal section through the dorso-median lobe to show the histochemical nature of the different kinds of granules. l.g.: large granules, s.g.: small granules, f.g.: fine granules.

1. s.g., proteins +++, tyrosine ++, SS groups ++, lipids +
2. f.g., P.A.S. +++, proteins ++
3. s.g., acid mucopolysaccharides ++, metachromasia γ
4. f.g., P.A.S. ++++

examination of Fig. 8. Suffice it to say that the products of this lobe includes glycogen (in the small granules s.g.) mucoproteins or glycoproteins (in the small granules), acid mucopolysaccharides (in the small granules), other carbohydrate-protein complexes (in the small and fine granules), disulphide groups and lipid-protein complexes (in the small granules).

Discussion.

The morphology of the accessory genital gland of *Haemaphysalis spinigera* is similar to that of other Ixodid ticks so far studied, RUSER (1933), DOUGLAS (1943), TILL (1961), the writer (unpublished observation on *Amblyomma variegatum, Hyalomma anatolicum, Hyalomma dromedarii, Rhipicephalus sanguineus, Rhipicephalus evertsi, Dermacentor marginatum,* and *Hyalomma leachi*), but differs from those of Argasidae, see ROBINSON & DAVIDSON (1914, Fig. 3), WAGNER-JEVSEENKO (1958, Figs. 2, 3, 4), and ROSHDY (1962, Figs. 5, 6, 7).

ROBINSON & DAVIDSON (1914), who worked on the histology of the gland in *Argas persicus,* differentiated two types of tissue. The spongy tissue of the spongy lobes consisted of connective tissue stroma, the lacunae of which are filled with a clear, pale secretion, whilst the granular lobes are formed of cylindrical cells filled with granules of irregular form. RUSER (1933) and DOUGLAS (1943) described spongy and granular tissues in the accessory genital gland of *Hyalomma aegyptium,* and *Dermacentor andersoni,* respectively, but as TILL (1961) has shown "Douglas and Ruser are not in complete agreement as to which of the lobes in the Ixodidae are spongy and which granular". The findings of the writer in *Haemaphysalis spinigera* are in agreement with those of TILL (1961), who found that all the lobes were granular, although "they do not acquire this granular appearance until sometime after they have started to feed". However, the reticulate appear-

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5. s.g., P.A.S. ++, proteins +++
6. f.g., P.A.S. +++, proteins ++
7. s.g., proteins +++, trypophane ++, tyrosine +, SS groups +
8. l.g., P.A.S. +, proteins +
9. s.g., P.A.S. +++, glycogen ++
10. f.g., P.A.S. +++, proteins +++, α amino acid +
11. f.g., P.A.S. ++, proteins ++
12. s.g., P.A.S. +++, proteins ++
13. f.g., P.A.S. ++, proteins ++
14. f.g., P.A.S. +++
15. s.g., P.A.S. -ve., proteins +++, lipids +, SS groups +

ance of the dorso-lateral and postero-lateral lobes would in the absence of granules resemble the spongy tissue in *Argas persicus*. It is therefore likely that as already concluded by TILL in *Rhipicephalus appendiculatus,* the spongy nature of the two lobes in *Haemaphysalis spinigera* represents a phase in the secretory cycle of the gland.

Histochemical methods have yielded some useful data on the nature of the granules in the different lobes. However, failure to obtain sections through a spermatophore has made it difficult to ascribe to the different lobes their part in the formation of the spermatophore. However, the products identified in the lobes of the accessory genital gland are similar to those identified by TATCHELL (1962) in the accessory genital gland of *Argas persicus,* hence it is possible that the structure of the spermatophores of both ticks might be similar. In addition, the writer was able to identify some glycogen in the median dorsal lobes. This might probably serve as a store of carbohydrate for the synthesis of N-acetyl-glucosamine for the formation of a chitinous material (ROGERS, 1961). The presence of disulphide groups also suggests keratinization.

Résumé.

1° Les glandes accessoires se composent d'un lobe médio-dorsal qui, postérieurement, se poursuit en un canal collecteur médian s'ouvrant sur le canal éjaculateur, d'un unique lobe médio-ventral, d'une paire de lobes antéro-latéraux, d'une paire de lobes ventro-latéraux, d'une paire de lobes dorso-latéraux, d'une paire de lobes postéro-latéraux et enfin d'une paire de lobes ventro-latéraux.

2° Dans une tique non nourrie, les cellules des lobes ne contiennent que peu de granules sécrétés.

3° Dans les tiques se nourrissant, des granules de sécrétion sont formés dans les cellules de tous les lobes ; ils se colorent en tons variés de rouge avec l'hématoxyline-éosine.

4° Des réactions histochimiques montrent que le lobe médio-dorsal sécrète du glycogène, des mucoprotéines, un complexe de protéine-lipide et des acides de mucopolysaccharides. Le lobe médio-ventral produit une substance contenant principalement une protéine de base riche en tyrosine. La sécrétion des lobes antéro-ventraux consiste principalement en protéine de base associée à des groupes disulfides. Les lobes postéro- et dorso-latéraux sécrètent une substance riche en protéine de base. La sécrétion des lobes postéro-ventraux contient des protéines et des phospholipides. La sécrétion des lobes antéro-ventraux est probablement une mucoprotéine.

5° On pense que les sécrétions des différents lobes contribuent à la formation du spermatophore.

Zusammenfassung.

1. Die männlichen genitalen Anhangdrüsen bestehen aus einem dorsomedianen Lappen, der nach rückwärts in einen Verbindungsgang führt, welcher

sich seinerseits in den Ductus ejaculatorius öffnet; aus einem einzelnen medioventralen Lappen; aus einem Paar von antero-lateralen, einem Paar von dorsolateralen, einem Paar von postero-lateralen und endlich aus einem Paar von postero-ventralen Lappen.

2. Bei hungernden Zecken enthalten die Zellen dieser verschiedenen Drüsenlappen nur wenige oder gar keine Sekretionsgranula.

3. Bei saugenden Zecken werden in allen Zellen der Anhangdrüsen Sekretionsgranula gebildet. Diese lassen sich mit Hämatoxylin-Eosin in verschiedenen Rottönen färben.

4. Histochemische Untersuchungen haben ergeben, daß der dorso-mediane Lappen Glycogen, Mucoprotein, einen Lipid-Protein-Komplex und saure Mucopolysaccharide ausscheidet. Der median-ventrale Lappen scheidet eine Substanz aus, welche hauptsächlich basische Proteine reich an Tyrosin enthält. Die Sekretion der antero-ventralen Lappen besteht hauptsächlich aus basischen Proteinen mit Disulfidgruppen. Die postero-lateralen und dorso-lateralen Lappen scheiden eine Substanz aus, die reich an basischen Proteinen ist. Die Sekretion der postero-ventralen Lappen enthält Proteine und Phospholipide, während diejenige der antero-ventralen Lappen wahrscheinlich ein Mucoprotein ist.

5. Man vermutet, daß die Sekrete der verschiedenen Drüsenlappen zur Bildung des Spermatophors beitragen.

Part II. Studies on the Ovary, Oviduct, Tubular Accessory Genital Gland and Gene's Organ.

Introduction.

Studies on the ovaries of some ticks of medical importance is of interest and importance since some viruses, rickettsia, and protozoa are known to be transovarially transmitted to the larva, and then transstadially to the adult, thus maintaining to some extent these organisms in nature. The Ixodid tick *Haemaphysalis spinigera* in particular has gained some importance in the past decade being incriminated as the chief vector of K.F.D. virus discovered in India in 1957 (see WORK & TRAPIDO, 1957) and WORK (1958). The isolation of this virus from *Haemaphysalis spinigera* indicates that this species might play some part in virus maintenance, TRAPIDO et al. (1959). Since histological methods alone have not revealed any difference between normal and virus-infected invertebrate tissues, LA MOTTE (1961), the introduction of other techniques such as electronmicroscopy in the study of virus-infected cells is worthwhile. In this study histochemical techniques employed have been of some value in amplifying histological methods as well as eliciting the nature of cell components, since such knowledge will facilitate the identification of cell components at the electronmicroscope level when this is taken up at a future date. Since the only natural method of infecting ticks with arbor viruses is through blood feeding, the changes that occur in the tissues consequent upon a blood meal have been observed by light microscopy from the newly emerged adult stage up to the ovipositing female. During this study, interesting observations were made on other organs associated with the ovaries and

these have therefore been incorporated into this paper. They include, the oviducts, the tubular accessory genital glands, and the Gene's organ and its associated gland.

Materials and Methods

see Part I. p. 235-236.

The Female Genital System.

Morphology.

The female genital system of *Haemaphysalis spinigera* consists of a single tubular ovary (ov.), a pair of oviducts (ov.d.), which fuse into a common oviduct (c.d.), a vagina (va.), a pair of tubular accessory glands (a.g.), discharging into the distal portion of the vagina dorsally, a receptaculum seminis (s.r.), and a connecting tube (c.t), which joins the receptaculum with the vagina (Figs. 1 and 2).

Histology and Histochemistry.

Unfed Female.

Ovary: In transverse section the ovary is roughly tubular consisting of a single layer of cubical cells enclosing a narrow lumen. The cytoplasm of the cells is faintly eosinophilic in haematoxylin-eosin and histochemical tests indicate that it contains mainly basic proteins. The cells contain two types of nuclei. The smaller and densely staining nuclei lie nearer the lumen and are the true epithelial cell nuclei, whilst the nuclei of the future ova are much larger, stain less intensely, and lie nearer the body cavity.

Oviducts: Each oviduct is a relatively thick-walled tube, consisting of three layers, namely an inner epithelial layer, a fibrous middle layer and an outer layer of flattened epithelial cells. The wall of the proximal portion of the common oviduct which is shiny white in appearance is thick and is composed of columnar epithelial cells (Fig. 3). These cells are overlaid by a thin layer of highly folded cuticle (c Fig. 3). Externally, the epithelial layer is surrounded by three or more layers of circular muscles. These probably cause a wave-like contraction to force the eggs into the vagina. The distal portion of the common oviduct (d.c.o. Fig. 4), which is transparent, is a much wider dorso-ventrally flattened tube, lined with columnar cells, with nuclei situated at the bases of the cells. The thin cuticular lining (c) is not folded, and externally, there is no muscular coat, but only a single layer of flattened epithelial cells (Fig. 4).

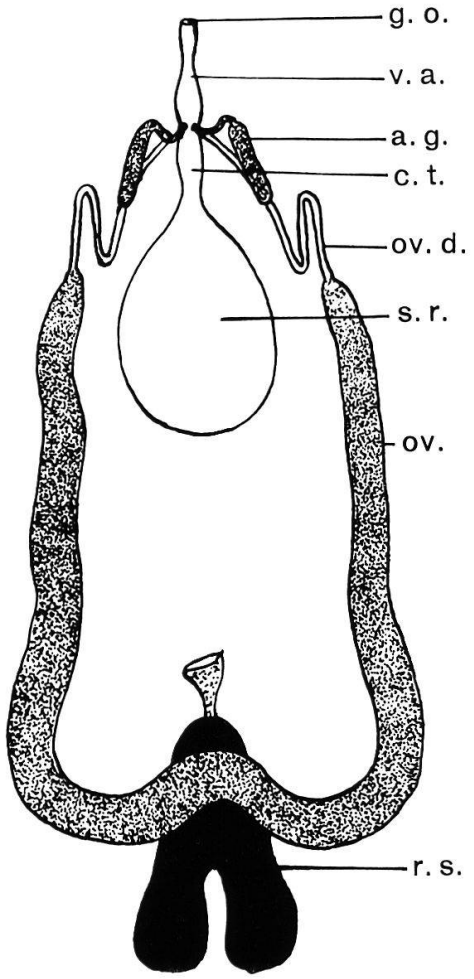

Fig. 1. Drawing of dorsal view of the genital organs of an unfed female. g.o.: genital opening, v.a.: vagina, a.g.: accessory genital gland, c.t.: connecting tube, ov.d.: oviduct, s.r.: receptaculum seminis, ov.: ovary, r.s.: rectal sac.

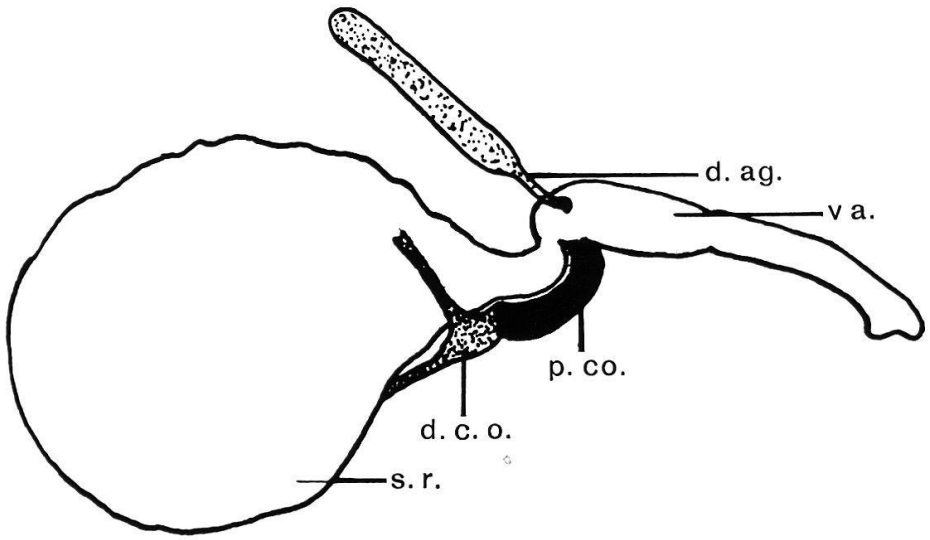

Fig. 2. Drawing of a side view of the s.r.: receptaculum seminis, c.t.: connecting tube, a.g.: tubular accessory genital gland, va.: vagina and the common oviduct to show their relationship to one another. p.c.o. proximal part of common oviduct, d.c.o.: distal part of common oviduct, d.a.g.: duct of tubular accessory genital gland.

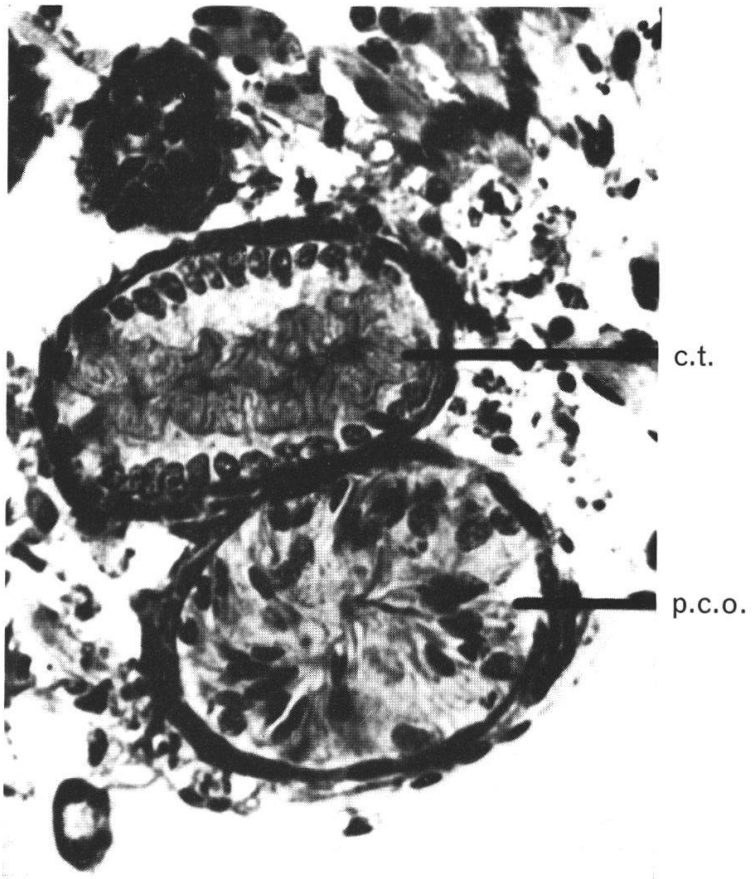

Fig. 3. Transverse section through the p.c.o.: proximal part of the common duct, and the c.t.: connecting tube. × 600. (Formol-sublimate; Haematoxylin-eosin.)

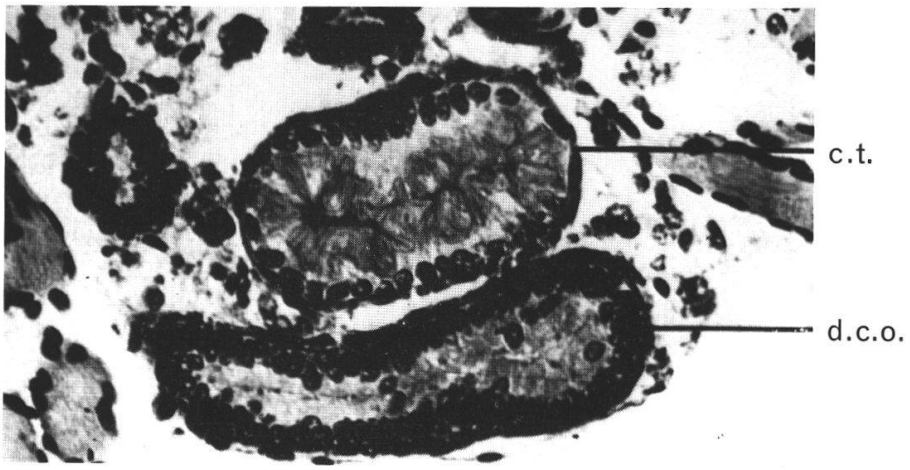

Fig. 4. Transverse section through the d.c.o.: distal part of common oviduct, and the c.t.: connecting tube. × 600. (Formol-sublimate; Haematoxylin-eosin.)

Tubular Accessory Genital Gland: A transverse section through this gland shows that it is composed of a single layer of wedge-shaped cells surrounding a duct-like lumen. The nuclei are medially disposed and in the newly emerged adults, the apical portions of the cells contain some finely granular eosinophilic material, which is predominantly basic protein, whilst the basal portions of the cells is fibrillar and basophilic.

Feeding Females.

Ovary: When the tick begins to feed, the nuclei of the supporting epithelium begin to multiply, whilst the nuclei of the ova begin to enlarge, staining less and less intensely with basic dyes. The cytoplasm of the supporting epithelium is fibrillar and finely granular staining with moderate intensity in P.A.S., but more intensely for basic proteins. Up to this stage there are no individual cell walls in the supporting epithelium, but as development proceeds, they become more visible (Fig. 5). The cytoplasm of the oogonium is uniformly granular and highly basophilic in haematoxylin-eosin, and histochemical test shows that the basophilia is due mainly to R.N.A. associated with basic proteins which are not removable by ribonuclease extraction. The cytoplasm of the oogonia does not stain with P.A.S. at this stage. As the tick continues to engorge the oogonia proceed to enlarge and develop but as TILL (1961) has already observed in *Rhipicephalus appendiculatus,* the oogonia develop at different rates so that various stages of development can be seen in one section through the ovary (Fig. 5). As the oogonia enlarge, they protrude into the haemocoele pushing the connective tissue sheath before them. The smallest oogonium is highly basophilic; basophilia of the cytoplasm decreasing in intensity as the oocytes enlarge until in the largest oocyte the cytoplasm does not show any basophilia. As already indicated, the basophilia is due mainly to R.N.A. associated with basic proteins. This association of R.N.A. with protein synthesis was first observed by CASPERSSON (1941). Since then a great number of investigators who have studied a wide variety of material have confirmed this observation; see BRACHET (1942), BRANDT (1941), HYDEN (1943), HAMBERGER & HYDEN (1945), THORREL (1944) and CASPERSSON (1950). However, in the fully developed egg, R.N.A. is not demonstrable cytochemically in the yolk globules. Development of the oogonia is accompanied by a marked enlargement of the nucleus, the chromatin material of which is visible as lightly staining large clumps peripherally disposed in the nucleus. Later

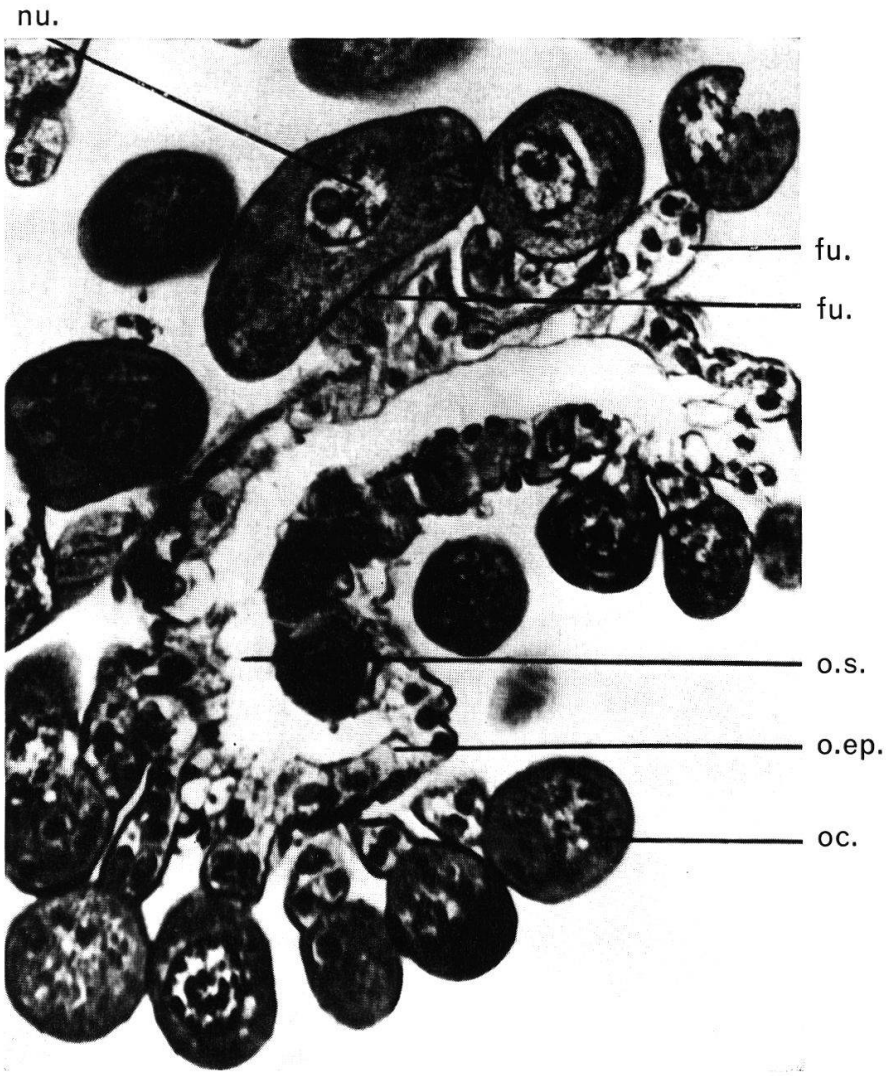

Fig. 5. Transverse section through the ovary of a partly fed female. fu.: funicle, o.s.: ovarian space, o.ep.: ovarian epithelium, oc.: oocyte, nu.: nucleus. × 600. (Formol-sublimate; Haematoxylin-eosin.)

on the chromatin material becomes thread-like and less stainable. As the oocyte develops, one or two nucleolei appear and the chromatin material begins to be less and less stainable. This is consonant with the findings of WAGNER-JEVSEENKO (1958) in *Ornithodoros moubata*. At the same time as the chromatin becomes invisible, a Feulgen positive material in the form of clumps of different sizes begin to appear all over the nucleolus and nucleus, a finding, similar to that of WAGNER-JEVSEENKO (1958) for *Ornithodoros moubata*. As growth proceeds, this Feulgen positive material begins to disappear from the nucleus and nucleolus, but appears in the cytoplasm, at first, scattered all over the latter in the form of irregular clumps. These Feulgen positive granules, which WAGNER-JEVSEENKO (1958) considered as chromosomal material derived from the nuclei of the oocytes, are believed to be

probably similar to the intracellular microorganisms previously described in the oocytes of *Ornithodoros moubata* by MUDROW (1932), JASCHKE (1933), and ROSHDY (1961). The nucleus shows fine network of Feulgen negative threads. As a result of rapid growth of the oocyte, it is pushed into the haemocoele, attached to the ovarian wall by stalks or funicles formed by proliferating cells of the supporting epithelium (fu. Fig. 5). As in the case of *Rhipicephalus appendiculatus* TILL (1961), and *Ornithodoros moubata* WAGNER-JEVSEENKO (1958), the nucleus comes to lie near the funicular end of the oocyte. The deposition of yolk material begins from the periphery as small more or less spherical granules, which stain bright pink in haematoxylin-eosin, and they grow larger as the tick continues to feed (see Fig. 6). Later on they become large globules which fill the whole of the oocyte, in most cases obscuring the nuclei. The latter stage was observed in fully engorged adults, the oocyte attaining their maximum size about two to three days after the tick has dropped off the host. At the same time as yolk formation takes place, capsule formation proceeds. According to LEES & BEAMENT (1948), it is produced by the oocytes themselves. In this way it differs from the chorion of insects which is produced by the folicular epithelium (see IMMS, 1957, and WIGGLESWORTH, 1953). The yolk bodies stain intensely with P.A.S. and further histochemical tests indicate that they contain large amounts of basic proteins apparently linked to the carbohydrate material in the form of a complex. BONHAG (1958) is of the opinion that the carbohydrate protein complex of insect yolk is either a muco- or glycoprotein. The basic protein component of the carbohydrate-protein complex was rich in tyrosine, which incidentally has been found in appreciable amounts in the gut lumen and cells. The yolk bodies also contain traces of tryptophane, and moderate amounts of lipid material as well as sulphydryl groups. The capsule on the other hand is P.A.S. negative in contrast to the intense P.A.S. staining of the tunica propria. It contains mainly proteins and some disulphide groups. Like TILL (1961) and others, I failed to observe the process of ovulation. According to LEES & BEAMENT (1948) it has apparently never been observed. A large number of spermiophores were observed in the oviducts of partly-fed and fully-fed females (Fig. 6), and in some few cases, a small number of spermiophores were observed in the epithelial cells of the oviducts.

Oviducts: During engorgement, the cubical epithelial cells of the oviduct begin to proliferate and to increase in size. They appear as small columnar cells, with nuclei situated in the basal halves of the cells. The cytoplasm of these cells is highly fibrillar especially in the basal portions of the cells, and uniformly finely granular.

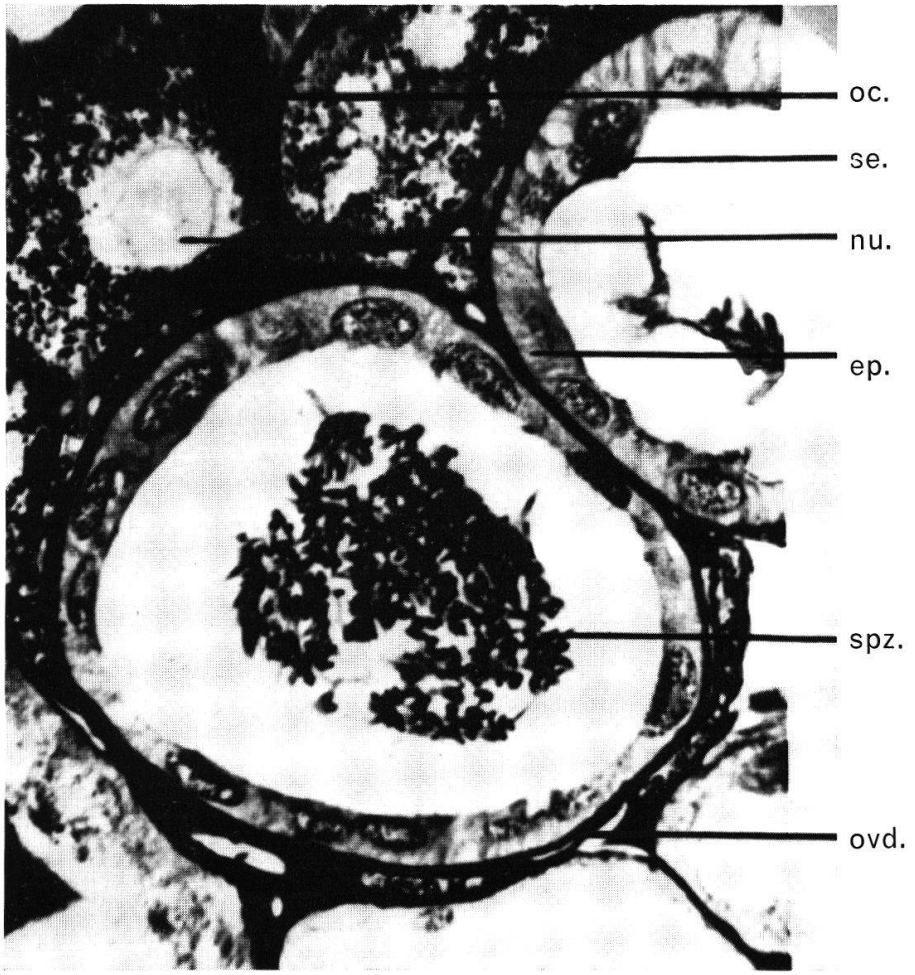

Fig. 6. Transverse section through the oocytes and oviduct of a partly fed female. oc.: oocyte, se.: secretion, nu.: nucleus, ep.: epithelial cell, spz.: spermatozoa, ovd.: oviduct. × 600. (Formol-sublimate; histochemical triple strain.)

The fibrillar material is highly basophilic, and histochemical tests indicate that this is due mainly to R.N.A. The finely granular material is faintly eosinophilic and stains mainly for basic proteins. In the late stages of feeding, the granules stain quite intensely with P.A.S., and later this P.A.S. material becomes concentrated at the apices of the cells (se. Fig. 6). A similar P.A.S. positive material was observed in the lumen of the oviducts. This material is a carbohydrate-protein complex since it also stains for basic proteins which is fast to pepsin digestion. This material is non-metachromatic and non-orthochromatic with toluidine blue. It might be a mucosubstance (see STACEY & BARKER, 1962). The nuclei enlarge considerably, consequent upon feeding, becoming vesicular with prominent evenly distributed chromatin material as well as one or two prominent nucleoli. As the tick continues to engorge, the lumen of the oviduct widens, and as a result, the epi-

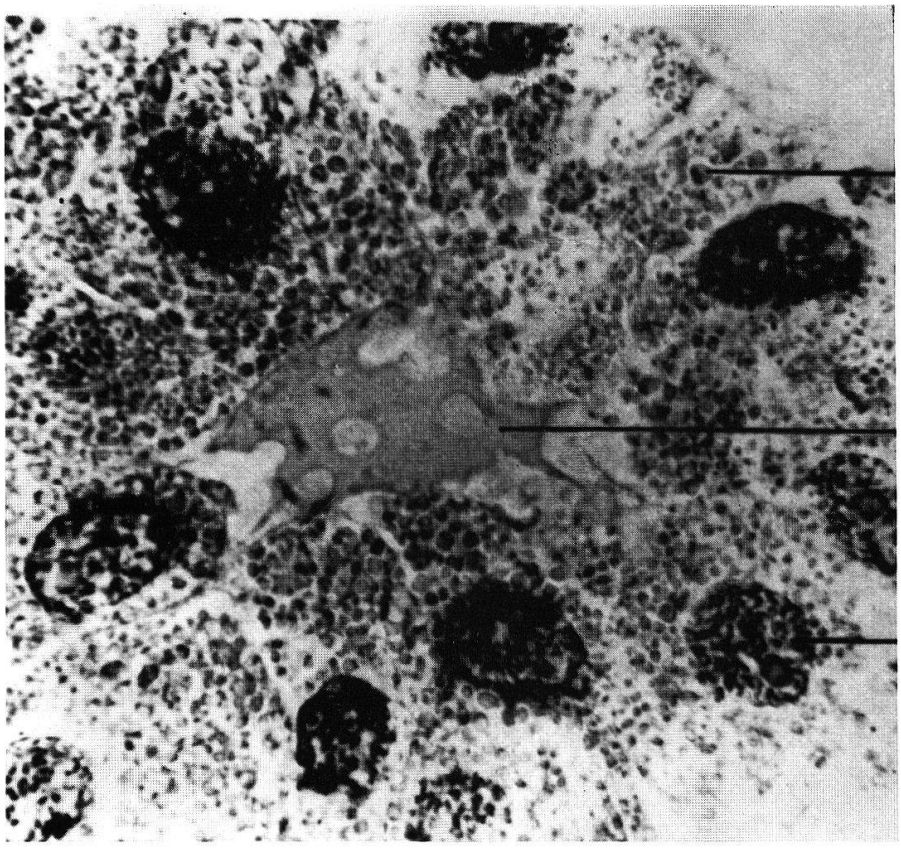

Fig. 7. Transverse section through the tubular accessory genital gland of an ovipositing female. s.g.: secretory granules, se.: secretion, nu.: nucleus. × 1500. (Formol-sublimate; Haematoxylin-eosin.)

thelial cells become stretched, so that they become cubical and later rectangular in shape. In the fully engorged tick, the lumen of the oviduct is much wider and the epithelial cells very much flattened. The vesicular nuclei become flattened due to alteration in shape of the cells. In ovipositing ticks the epithelial cells show extreme flattening, so that some of them look like pavement epithelial cells. In ovipositing ticks, the highly folded cuticular lining of the proximal region of the common oviduct becomes flattened due to stretching, and the epithelial cells as well as the cells of the muscle coat become hypertrophied.

Tubular Accessory Genital Gland: Histological and histochemical changes in the epithelial cells of this gland become evident in ticks which have attached for about 36 hours. The gland starts to increase in size, and the lumen which in the unfed tick is like a duct becomes wider. The wedge-shaped epithelial cells show increase in size, becoming columnar or dome-shaped. The cytoplasm of the cells at this stage is fibrillar, and finely granular, and basophilic in haematoxylin-eosin. Histochemical tests indicate that the basophilia is due mainly to R.N.A. As the tick continues to engorge,

TABLE 1.

Histochemical results on tubular accessory genital gland and Gene's organ and gland of fully fed female *.

Method	Tubular Accessory Genital Gland	Gene's Organ and Gland
	Coarse Granules	Fine Granules
P.A.S. Technique	–	x
Pepsin/P.A.S., Trypsin/P.A.S.	–	x
Metachromasia—Toluidine Blue	–	–
Basic Proteins { Naphthol Yellow -S	xxx	xxx
Basic Proteins { Mercuric Bromophenol Blue	xxx	xxx
Tyrosine—Millon's Reagent	xxx	$\bar{x}$
Tryptophane—D.M.A.B.-nitrite	$\bar{x}$	$\bar{x}$
Arginine—Sakagukyi's Reagent	–	$\bar{x}$
α-Amino Acids—Ninhydrin-Schiff	–	x
-SS- -SH- Groups—Alkaline tetrazolium	–	xx
-SS- Groups—P.F.A.A.B. Reaction	xx	–
Lipids—Regaud's haematoxylin	xx	xx

* $\bar{x}$ sign indicates a faint positive reaction. The number of x signs indicates the relative intensity of staining.

more of the granular material continues to accumulate in the enlarging cells. The nuclei increase in size becoming vesicular, and localized in the mid-portions of the cells. In fully engorged ticks the gland is very much enlarged and the cells are filled with coarse granules (Fig. 7). These granules stain bright pink in haematoxylin-eosin and contain mainly basic proteins rich in tyrosine groups. The granules are P.A.S. negative and do not stain with toluidine blue. They contain some lipid material as well as disulphide groups (see Table 1). In ticks which are about to oviposit this granular secretory material is liberated into the lumen of the gland in the form of a colloidal material (Fig. 7).

Gene's Organ.

Morphology.

The Gene's organ consists of an eversible cuticular sac (c.s.) enclosed in a hypodermal sac (h.s. Fig. 8). Posteriorly this sac divides into two horns (hn. Fig. 8) as TILL (1961) also observed in *Rhipicephalus appendiculatus* although *Ixodes* are reported to possess four horns (see ARTHUR, 1953). In *Dermacentor ander-*

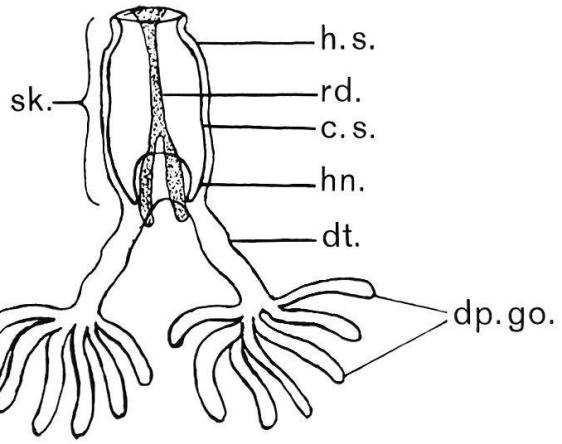

Fig. 8. Drawing of Gene's organ of a fully fed female. h.s.: hypodermal sac, rd.: rod, c.s. cuticular sac, hn.: horn, dt.: duct, dp.go.: digitate process of Gene's organ, sk.: stalk.

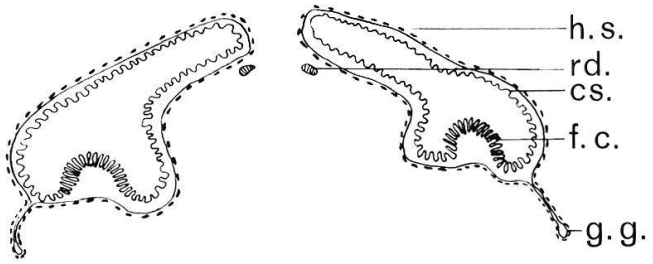

Fig. 9. Drawing of a transverse section through the posterior region of Gene's organ. h.s.: hypodermal sac, rd.: rod, cs.: cuticular sac, f.c.: folded cuticle, g.g.: gland of Gene's organ.

soni DOUGLAS (1943), *Ornithodoros moubata* LEES & BEAMENT (1948), and *Argas persicus* ROBINSON & DAVIDSON (1914), the Gene's organ has a pair of horns. A pair of glands (g.g. Fig. 9) are associated with the Gene's organ. In the unfed female, these are inconspicuous consisting of a pair of small sacs. Extending from the posterior border of the basis capituli and lying below the hypodermal sac is a rod-like structure (r.d. Fig. 8). Posteriorly it divides into two. This ARTHUR (1953) considered as probably playing an indirect role in the eversion and retraction of the sac of Gene's organ. As already observed in *Ixodes hexagonus, Ixodes canisuga, Ixodes ricinus* ARTHUR (1953) the Gene's organ is without any musculature in *Haemaphysalis spinigera.*

Histology and Histochemistry.

Unfed Female.

In the unfed female, the epithelial cells of the hypodermal sac are flattened, and individual cell boundaries are not evident (Fig. 9). The cells have a clear staining cytoplasm. The epithelial

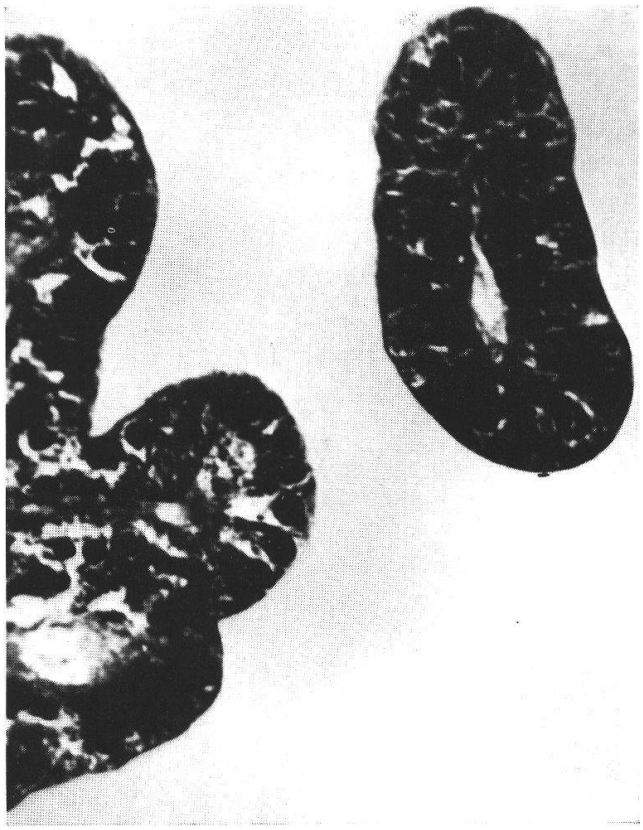

Fig. 10. Transverse section through the gland of Gene's organ of a female which has engorged for two days. × 600. (Formol-sublimate; Haematoxylin-eosin.)

cells of the gland itself are slightly deeper than those of the hypodermal sac.

Feeding Females

In feeding females, the epithelial cells of the gland proper begin to proliferate and increase in size, giving rise to a digitate structure (dg. Fig. 8), on either side of the Gene's organ. Each finger-like process is tubular and those of one side discharge into a duct (dt. Fig. 8) leading into the space between the hypodermal sac and the cuticular sac. In ticks which have attached for five days, the epithelial cells of the gland proper are small and columnar with spherical nuclei situated at the base of the cells or in the middle part. The cytoplasm is finely granular and is basophilic. Histochemical tests showed that the basophilia is due mainly to R.N.A. As the tick continues to engorge the cells of the gland, increase in size and the lumen of the gland widens considerably. The finely granular cytoplasmic material continues to increase in quantity (Fig. 10) and histochemical tests indicate that it contains

Fig. 11. Transverse section through the gland of Gene's organ of an ovipositing female. × 600. (Formol-sublimate, Haematoxylin-eosin.)

mainly basic proteins although it gives a slight positive reaction with P.A.S. (see Table 1). In fully engorged females, the proliferation of cells continues, and this is very marked in ovipositing females in which cells at different stages of secretory activity could be observed in any one digitate process of the gland (see Fig. 11). Some of the cells are cubical, and others are slender and columnar whilst others are dome-shaped with vacuolate and reticular cytoplasm. The latter are probably spent cells which have already discharged their secretion. The smaller cells still contain a homogeneous finely granular secretory material which in some parts of the gland are discharged into the enlarged lumen of the gland in the form of a colloidal material. The secretory material consists mainly of protein associated with some low concentration of P.A.S. positive substance. It also contains traces of tyrosine and tryptophane and moderate amounts of lipid material, and sulphydryl groups (see Table 1). The presence of sulphydryl groups might suggest the stabilization of structural proteins by the formation of disulphide links. However, the negative performic

acid alcian blue test for -SS-groups indicates that this reaction has not occurred.

Discussion.

The general course of development of eggs in *Haemaphysalis spinigera* is similar to that described by NORDENSKIÖLD (1910) for *Ixodes ricinus*, TILL (1961) for *Rhipicephalus appendiculatus*, LEES & BEAMENT (1948), and WAGNER-JEVSEENKO (1958) in *Ornithodoros moubata*. The last author, by the use of some histochemical methods, was able to observe some Feulgen positive granules in the cytoplasm and nucleus of the oocytes. She figured these granules from the time of their appearance in the young oocytes up to the fully developed egg and considered them as D.N.A. positive granules derived from the chromatin of the nucleus, basing her premise on the fact that the appearance of these granules, coincides with the failure of the nuclear chromatin to stain with Feulgen stain. The author's findings are similar to that of WAGNER-JEVSEENKO (loc. cit.) but is of the opinion that the Feulgen positive granules are in all probability the rickettsia-like organisms ROSHDY (1961) observed in the oocytes of *Argas persicus* and later in other Ixodid ticks (ROSHDY, pers. comm.). Histochemical findings suggest that the yolk of the eggs is a carbohydrate-protein complex in which the protein is rich in tyrosine groups. The yolk granules of insect egg is a carbohydrate-protein complex and this is believed to be a mucoprotein (see BONHAG, 1958). The author is uncertain whether the yolk bodies in the oocytes is a mucoprotein especially when they were non-metachromatic with toluidine blue and were negative with Bismark Brown Y. The yolk bodies also contain moderate amount of lipid material, probably due to phospholipids especially when the yolk bodies of some arachnids have been shown to contain phospholipids (NATH, 1960). It is not known whether the formation of yolk globules occurs by aggregation of smaller granules. The relatively small numbers of yolk globules in the fully developed eggs as compared with the large number of small yolk granules in the developing oocytes is in support of such a process of yolk globule formation. In this connection it is of interest to note that electronmicroscopic studies have indicated that the formation of yolk globules of some species of *Mollusca* and *Echinodermata* occurs by the aggregation of protein particles (BOLOGNARI, 1961a, 1961b).

The cells of the oviducts secrete a P.A.S. positive colloidal material rich in basic proteins but non-metachromatic with

toluidine blue. This substance might possibly function as a lubricant for the eggs. It might as well be similar to the viscid material which WAGNER-JEVSEENKO (loc. cit.) observed in the uterus and oviducts of *Ornithodoros moubata* and considered to be an attractant for the spermatozoa.

The similarity in the histochemical nature of the cells of the accessory genital gland and the gland of Gene's organ (see Table 1) might suggest some functional relationship. The accessory genital gland might possibly play a supplementary role to the gland of Gene's organ. LEES & BEAMENT (1948) described a lobed accessory genital gland in *Ixodes ricinus,* and regarded this gland as a possible source of an incomplete layer of wax observed on the surface of the egg in the vagina; the final process of waxing being completed by the Gene's organ. In view of the absence of the lobed accessory genital gland in *Haemaphysalis spinigera,* the supposition that the tubular accessory genital gland might be a possible source of an incomplete waxing layer is not unjustifiable. ROBINSON & DAVIDSON (1914), DOUGLAS (1943), and VITZTHUM (1943) believe that the secretion of the tubular accessory genital gland acts as a lubricant. However, the writer believes that secretion found in the cells of the connecting tube (a structure which connects the receptaculum seminis with the vagina [see Fig. 2]) acts as a lubricant since histochemical tests carried out by the author indicated that this mucous secretion was a mucoprotein. It is of interest to note that the mucous secretion in the alimentary tract of man believed to act as a lubricant, FLOREY (1954) is believed to be a mucoprotein LATNER et al. (1954).

The histological appearance of the gland of Gene's organ is similar to those of other ticks so far described; see ROBINSON & DAVIDSON (1914), LEES & BEAMENT (1948), ARTHUR (1953) and TILL (1961). Histochemical findings suggest that the secretion of the gland is mainly protein in character. This is consonant with the findings of LEES & BEAMENT (1948) in *Ornithodoros moubata.* In the following study, the cells of this gland was found to contain appreciable amounts of a P.A.S. positive substance. However, contrary to expectation only traces of arginine was observed by the histochemical method employed.

Acknowledgement.

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Résumé.

1° La morphologie de l'organe génital femelle de la tique *Haemaphysalis spinigera* est brièvement décrite.

2° Une section transversale de l'ovaire montre qu'il s'agit d'un tube consistant en une seule assise de cellules enfermant une lumière étroite. Les noyaux sont destinés à devenir les noyaux des cellules de l'épithèle ovarien.

3° Pendant la nutrition, l'ovaire grandit et les cellules de l'épithèle ovarien prolifèrent.

4° Alors que l'ovaire grossit, les granules vitellins apparaissent dans le cytoplasme, et leur taille s'agrandissant, ils deviennent de larges sphérules dans les œufs complètement développés. Ces sphérules consistent en un complexe de protéines-carbohydrates et de lipides.

5° Au cours du développement des ovocystes, des granules donnant une réaction de Feulgen positive apparaissent dans le cytoplasme et le nucleus des ovocytes.

6° Dans les femelles gorgées, les cellules épithéliales des oviductes grossissent considérablement et une substance (PAS positive) s'accumule au sommet des cellules. Une substance colloïdale identique qui, elle aussi, est PAS positive, se retrouve dans la lumière des oviductes.

7° Les femelles se nourrissant montrent une accumulation de sécrétion dans les cellules de la glande génitale accessoire.

8° Pendant l'oviposition, les granules de sécrétion sont déchargés dans la lumière de la glande sous forme d'une substance colloïdale onctueuse. Ces sécrétions contiennent des protéines de bases riches en tyrosine et associées à des lipides et des groupes disulphides.

9° L'organe de Géné est un sac dévaginable inclus dans un sac hypodermique qui, postérieurement, se divise en deux cornes.

10° Au cours de la nutrition de la femelle, une paire d'organes consistant en deux projections digitales se développent en relation avec les cornes.

11° Chaque « doigt » est tubulaire et se compose d'une couche monocellulaire dans laquelle s'accumulent de fines sécrétions granulaires.

12° Ces sécrétions sont colloïdales et contiennent principalement des protéines de base associées à quelque lipide et des groupes sulfidryles.

13° La nature des granules se trouvant dans les oocytes en développement (positives à la réaction Feulgen) et les fonctions probables des sécrétions de l'oviducte et des glandes génitales accessoires sont brièvement discutées.

Zusammenfassung.

1. Die Morphologie des weiblichen Genitalapparates der Zecke *Haemaphysalis spinigera* wird kurz beschrieben.

2. Im Querschnitt ist das Ovar röhrenförmig und besteht aus einer einzigen Lage von Zellen, welche ein schmales Lumen umschließen. Die Nuclei sind dazu bestimmt, die Zellkerne des Eiepithels zu werden.

3. Während des Saugaktes vergrößert sich das Ovar, und das Eiepithel schwillt an.

4. Sobald die Eier größer werden, erscheint Dottermaterial im Cytoplasma. Das Dottermaterial vermehrt sich und bildet große kugelige Gebilde im vollentwickelten Ei. Diese kugeligen Gebilde bestehen aus einem Kohlenhydrat-Protein-Komplex und aus Lipiden.

5. Während der Entwicklung der Oocyten erscheint im Cytoplasma und in den Nuclei ein granulöses Material, das Feulgen-positiv reagiert.

6. Im vollgesogenen Weibchen werden die Epithelzellen der Ovidukte beträchtlich größer, und eine PAS-positive Substanz sammelt sich am oberen Rand der Zellen an. Eine ähnliche kolloide Substanz, ebenfalls PAS-positiv, ist auch im Lumen der Ovidukte vorhanden.

7. Im saugenden Weibchen sammeln sich auch in den Zellen der akzessorischen Genitaldrüsen Sekretionsgranula an.

8. Bei der Eiablage werden die Sekretionsgranula in das Lumen der Drüsen ausgestoßen in Form einer weichen kolloiden Substanz. Die Sekretion enthält basisches Protein, reich an Tyrosin, Lipide und Disulfidgruppen.

9. Die Genesch'schen Organe haben die Form eines umstülpbaren Sackes, der seinerseits in einem hypodermatischen zweihörnigen Sack liegt.

10. In der saugenden Zecke entwickelt sich am Genesch'schen Organ ein Paar fingerähnliche Fortsätze in Verbindung mit den Hörnern des hypodermatischen Sackes.

11. Diese fingerförmigen Fortsätze sind röhrenförmig und bestehen aus einer einzelnen Schicht von Zellen, in denen feines granulöses Sekretionsmaterial angehäuft ist.

12. Diese kolloidale Sekretion enthält basische Proteine, einige Lipide und Sulfidrylgruppen.

13. Zum Schluß werden die Beschaffenheit der Granula (Feulgen-positiv) in den reifenden Oocyten und die möglichen Funktionen der Sekretion der Ovidukte und der akzessorischen Genitaldrüsen kurz besprochen.