The secretion from the mammalian oviduct has long been a matter of dispute. Thus, the presence of secretory granules has sometimes even been denied, and different opinions are found regarding the details of the secretory process.

Like other parts of the differentiated Müllerian duct, the epithelium of the oviduct is influenced by the hormonal factors regulating the sexual cycle. Thus, during the period preceding oestrus nonciliated cells have been found to accumulate secretory material, which is released after ovulation. The knowledge obtained by light microscopy has now been widened by electron microscope studies of rabbit and rat (Borell et al. 1956, 1959; Nilsson 1958). The fine structure of the epithelium of the bovine oviduct has not been investigated. There exists only a study of the epithelial surface (Björkman and Fredricsson 1960), where a correlation of the microvilli of the ciliated cells with alkaline phosphatase activity was suggested. The secretory granules of the bovine oviduct were reported to contain glycogen (Weeth and Herman 1952), which is in contrast with the conditions in rat, mouse, rabbit, sheep and man (Fredricsson 1957, Hadek 1955).

The present study was designed to gather further information for the understanding of the secretory process of the Fallopian tube.

The material and its preparation

a) Material, fixation. The oviducts were obtained at the Slaughter House of Stockholm. Material for electron microscopy was taken from 13 apparently normal cows in different sexual phases (follicular 5, postovulatory 2, non-pregnant luteal 4, and pregnant luteal 1). The phase was determined by inspection of the ovaries and records of the reproductive history of the animal.

To secure rapid fixation, the slaughtering schedule was modified to obtain the oviducts immediately after the animals had been shot and bled. Small pieces of tissue from the ampullary part were fixed in Palade's fluid for 2 hours. After dehydration the tissue blocks were embedded in n-butylmethacrylate, which was polymerized at 60°C. Sections were made with a Porter-Blum ultramicrotome and examined in a Siemens Elmiskop I at initial magnifications of 1,400 to 21,000 diameters and an acceleration voltage of 80 kV.

Material for histochemistry was obtained from 20 cows, killed at various stages of the oestrus cycle. The oviducts of 12 cows were fixed in cold acetone and embedded in paraffin
The bovine oviduct epithelium 501

m.p. 52° C. The other eight specimens were freeze-dried and embedded in polyethylene glycol. Details and biochemical controls of this technique have been published previously (FREDRICSSON 1958). Post-fixation of the sections from the freeze-dried material was obtained by immersion in ethanol for 10 min or in gaseous formaldehyde produced by paraformaldehyde for one hour when not otherwise stated (cf. FREDRICSSON 1959 b).

b) Histochemical tests. 1. Basophilia was demonstrated by staining with toluidine blue including controls digested with ribonuclease (BRACI-IET 1953). The concentration of the enzyme was 0.1 mg/ml, and the sections were incubated at pH 6 for one hour at 37° C (the preparation was obtained from the Sigma Chemical Co., St. Louis, U.S.A.). Extraction of other sections with water at the same pH and temperature was also included in the test. The sections used for the demonstration of basophilia were postfixed over paraformaldehyde for 60 min, followed by SIERA'S solution (1946) for 15 min.

2. Polysaccharides. The periodic acid Schiff reaction (PAS) was performed using an alcoholic Schiff reagent (Mowrey et al. 1952).

The diastase test was found to be dependent on the kind of fixation used, and the present material was especially sensitive to variations of this kind. In order to find the best conditions, sections of freeze-dried oviduct epithelium were subjected to different fixatives before the staining. Sections of freeze-dried rat liver were placed on the same slides to serve as controls of the diastase effect. The following post-fixations were tried:

1. Absolute ethanol, 10 min.
2. Gaseous formaldehyde, 60 min.
3. Gaseous formaldehyde, 60 min followed by 10% acetid acid, 10 min.
4. Gaseous formaldehyde, 60 min followed by 2% trichloroacetic acid, 10 min.
5. Gaseous formaldehyde, 60 min followed by 4.5% mercuric chloride, 10 min.
6. Gaseous formaldehyde, 60 min followed by Susa fixative, 10 min.

Material pretreated in this way was then stained with the PAS-technique. Controls were obtained by digestion in one per cent diastase solution for one or two hours at room temperature as recommended by GRAUANN (1959). The solvent consisted of phosphate buffer of pH 6.5 (LILLIE and GRECO 1947). Controls were also extracted with the solvent alone. The diastase preparation was obtained from Merck AG, Darmstadt, Germany.

3. Lipids were demonstrated after postfixation over paraformaldehyde for 60 min, using a saturated solution of Sudan black B in 70% methanol as recommended by MEIER (1959). No signs of non-specific staining were observed in this material.

4. Acid phosphatase. Fresh frozen section were used and an ordinary azo coupling method was tried, using α-naphthyl phosphate and diazotized o-aminooazotoluene (Fast Garnet GBC Salt, I.C.I. Ltd) (PEARSE 1960).

5. Non-specific esterase was demonstrated by using α-naphthyl acetate as substrate in the presence of tetrazotized diorthoanisidine (Naphthol Diazo Blue B, Du Pont de Nemours & Co, Boston) (PEARSE 1960). The incubation period was 15 min, and the sections were postfixed over paraformaldehyde for 30 min. Attempts were also made to demonstrate this enzyme with 5-brom-indoxyl-0-acetate according to HOLT (1958), but no staining was obtained even after 2 hours' incubation.

Observations

a) Ultrastructural organization. The epithelium is simple columnar. The ciliated and secretory cells are of about the same size and outer shape (Fig. 1). They rest on an even basement membrane, which is double lined and has a thickness of about 30 μμ. The ciliated and secretory cells are mingled and cells of the same kind occur singly or in small groups. Occasionally rounded basal cells are also found, which seem not to be attached to the basement membrane. In the basal part of the epithelium narrow slits between the cells are found during the non-oestrogenic phase (Fig. 1), whereas during the oestrogenic phase large spaces are sometimes seen.