The Epithelium of the Rabbit Vagina: A Microtopographical Study by Light, Transmission and Scanning Electron Microscopy*

F. Barberini, S. Correr, F. De Santis and P. M. Motta

Department of Anatomy, Faculty of Medicine, University “La Sapienza”, Rome, Italy

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Summary. In order to obtain a more precise microtopographical surface map of the epithelium of the rabbit vaginal mucosa, investigations by light microscopy (LM), transmission electron microscopy (TEM), and scanning electron microscopy (SEM) have been systematically correlated. The epithelium was examined from the “portio vaginalis cervicis uteri” down to the “vestibulum”. This study shows that the upper 2/3 of the vagina—which, in this species, is very long, measuring 13-14 cm—is lined by a single epithelial layer of tall ciliated and microvillous cells closely resembling the endocervical epithelium with which it is continuous. Moreover, these ciliated and microvillous cells also cover mucosal infoldings in the upper part of the vagina, especially those on the ectocervix and in the fornices, and scattered vaginal crypts. In particular, the ciliated elements decrease in number below the fornices, so that in large areas of the middle part of the vagina only microvillous cells are recognizable. Prior to the squamo-columnar junction, however, the ciliated cells increase again. This study also reveals that in the rabbit the squamo-columnar junction is located at the level of the pubic symphysis and that a squamous pluristratified epithelium covers only the lower 1/3 of the inner surface of the vaginal wall.

In the estrous, i.e., precoital rabbit, the microvillous cells show little sign of secretion, whereas after mating they exhibit remarkable secretory features. These seem to increase progressively with postcoital stages (5 h, 24 h and 10 days) in the form of extensive mucification. These secretions often come in contact with spermatozoa retained in the mucosal infoldings and crypts, and are similar to those occurring in the endometrium, where they clearly depend upon progesterone activity. These epithelial features, different from those of other mammals, including humans, suggest that the greater part of the rabbit vagina accomplishes functions other than serving for copulation and as a fetal passageway. The present findings support the view that the rabbit vagina also plays a role as a reservoir of spermatozoa and in maintaining their viability, like the endometrium and endocervix.

Previous research by scanning electron microscopy (SEM) and/or transmission electron microscopy (TEM) has demonstrated that, in several species of mammals, a cyclical series of changes occurs on the epithelial surface of the vaginal mucosa. These consisted essentially of mucification and/or keratinization, with the appearance of microvilli and/or microridges—also called microplicae or microrugae (Kanagawa et al., 1972; Parakkal, 1973; Rubio, 1976; Centola, 1978; Lamb et al., 1978; Holderegger, 1980; King, 1983; Corbeil et al., 1985; Winterhager and Köhnel, 1985).

In species not possessing a true cyclical sequence of reproductive stages and in which ovulation therefore follows coitus, such as in the rabbit, it is nevertheless possible to detect a succession of morphological modifications of the female reproductive tract mucosa related to ovarian endocrine activity and pregnancy. Animals showing reproductive characteristics of this type may represent a valid model in which to study the specific morphofunctional relationships occurring at a given moment between serum levels of sex hormones and target organs. In fact, if the precoital (i.e., at estrus) animal acts as a control, a more direct comparative evaluation of the structural modifications of the female reproductive tract mucosa related to ovarian endocrine activity and pregnancy.

In previous SEM and TEM studies, we reported changes in the rabbit endometrium both at the precoital stage (estrus phase) and at various time intervals after mating and during pregnancy (Bar-
BERINI et al., 1978). Since only scarce and fragmentary information is available on the topic, we have described in this study the fine structure of the vagina of the same species at pre- and postcoital stages.

We were further intrigued by the possibility of observing contact modalities between spermatozoa and the surface of the vaginal mucosa, either on the convex sides of the mucosal longitudinal folds, or at the bottom of the intervening grooves, since motile sperms tend to migrate mainly in close vicinity to the epithelial lining and are unable to recognize mechanical barriers during their migration (PAKZAD and PAUFLER, 1982). Such observation might prove important in that, at the same level of the above vaginal duct, early phases of some biological phenomena related to fertilization in the rabbit, such as sperm selection processes, probably occur.

Our purpose therefore was to contribute to a better elucidation of the surface microtopographical anatomy of these structures, particularly concerning both the features of the vaginal mucosa in the estrous phase and mucosal progressive postcoital changes, and the reciprocal relations between male gametes and vaginal epithelial cells.

MATERIALS AND METHODS

The present investigation used eight sexually mature New Zealand female rabbits weighing about 3-4 kg each. All animals were caged separately and fed a standard diet for one week. Two rabbits were then selected as controls (estrous animals), anesthetized with ether, and sacrificed. The others were mated and then divided into three couples. These were progressively killed at various times after copulation (i.e., at 5 and 24 h, and 10 days post coitum). To avoid the occurrence of artifacts resulting from the operative times for specimen removal, in situ fixation was performed in all living rabbits by means of vaginal irrigation with 2.5% glutaraldehyde in sodium cacodylate buffer (0.1M pH 7.35) injected intra-luminally at low pressure, without damaging the epithelial lining. Subsequently, all connections were excised and the vaginas were completely removed.

For light microscopy, whole vaginal walls were cut serially in large fragments along the major axis from the portio to the vestibulum; for SEM and TEM, smaller pieces of the walls were taken serially at various levels of the vaginal canal. All samples were immediately immersed in the same fixative solution for about 24 h at 4°C (MOTTA and PORTER, 1974; MOTTA and VAN BLERKOM, 1975).

For light microscopy, large fragments, about 1 cm in length and 2 mm in thickness, were dehydrated through an ascending graded series of concentrations of ethanol, embedded in paraffin, sectioned by microtome, and then stained with hematoxylin and eosin.

For SEM, some of the smaller blocks were rinsed in the same sodium cacodylate buffer after fixation, dehydrated in an ascending series of acetone concentrations, and then transferred to liquid CO₂ for critical-point drying. The dried samples were supported on aluminum studs by means of conductive silver paint and shadowed with a thin layer of gold in a high-vacuum sputter coater (Edwards). All specimens were observed and photographed with a Cambridge Stereoscan 150 Electron Microscope, operated at 8-20 kV.

For TEM, fragments were cut into smaller parts, approximately 1-2 mm in diameter, postfixed in cold 1.5% osmium tetroxide buffered at pH 7.35, dehydrated through an ascending graded series of ethanol concentrations, and then embedded in Epon (LUFT, 1961). Ultrathin sections, about 90-100 nm in thickness, were obtained with an LKB Ultrotome III ultramicrotome, stained with uranyl acetate (WATSON, 1958) and lead citrate (REYNOLDS, 1963), and finally examined under a Zeiss EM-9A electron microscope. Semithin sections were also obtained and stained with a mixture of methylene blue-Azur II in 1% borax solution.

Fig. 1. General view of the vaginal mucosa arranged in longitudinal folds, separated by deep furrows. SEM, ×150

Fig. 2. Portio vaginalis. Prominent folds from the fornices (f) converge at the os of the cervical canal. SEM, ×30. Inset: The indented outline of the fornix is clearly evident. LM, hematoxylin-eosin stain, ×75

Fig. 3. On the surface of the high, longitudinally-oriented folds, (thick arrows) other, smaller transversely-oriented and anastomosed folds (thin arrows), outlined by shallow grooves, are present. SEM, ×300

Fig. 4. In the lower part of the vagina the wall is raised in several coarse transverse wrinkles (arrows), recalling the "rugae vaginales" of other mammals. SEM, ×110
Figs. 1-4. Legends on the opposite page.
RESULTS

The rabbit vagina is a very long muscular-membranous cylindrical duct, whose longitudinal axis, including the vestibulum, measures about 13-14 cm. Its walls, as revealed by our SEM analysis at low magnification, appear corrugated on their luminal side due to a number of long, coarsely parallel, and variously anastomosed folds, running longitudinally and delineated by the interposition of deep furrows (Fig. 1). Such a configuration was particularly apparent in the upper part of the vaginal duct and in correspondence with the fornices. Here the folds became more prominent and continuous with those taller of the portio, and converged at the os of the cervical canal (Fig. 2). This folding pattern in the cranial regions of the vagina accounted for the indented outline of the mucosa, when it was observed in transverse section by light microscopy (Fig. 2, inset). On the surface of these formations, other, smaller transversely-oriented folds often occurred. These too were anastomosed and divided by less deep grooves in continuity with the major intercolumnar furrows (Fig. 3). In addition, in the lower part of the duct there was another succession of parietal folds arranged perpendicularly to its length. These findings somewhat recalled the “rugae vaginales” present in other species (Fig. 4).

Our light and electron microscope investigations have allowed us also to clarify that the rabbit vagina is lined inside by a mucosa that shows some striking differences in structure when compared with that of other mammals, including humans. Indeed, in spite of its morphofunctional changes as an expression of hormonal influence, it nevertheless showed some general features irrespective of the sexual phase of the animal. In particular, the epithelium of the portio vaginalis, fornices and upper 2/3 of the duct was not of the squamous, pluristratified type. In fact, with tall cells arranged in a single layer, it resembled instead the columnar epithelium of the cervical canal in the same species as well as in other mammals and in man (SINGER and JORDAN, 1976; HAPEZ, 1986; FERENCZY, 1987; ODOR and BLANDAU, 1988; ODOR et al., 1989). In particular, the ectocervix and fornices were lined by ciliated and non-ciliated, microvillous cells closely resembling those composing the endocervical epithelium (Fig. 5). Owing to the indented pattern of the mucosa, the inner vaginal lining did not form proper glands, but covered parietal infoldings (Fig. 5, inset). Moreover, in terms of microtopography as observed by SEM and by light microscopy, the amounts of these two cell types changed in relation to their distance from the external os of the cervical canal. The ciliated cells were not found evenly distributed throughout this epithelium. In fact, they were abundant on the surface of the portio and in the fornices.
Figs. 5-12. Vagina, estrous phase. Legends on the opposite page.
but below they became progressively less numerous. As a consequence, few or no cilia were detected in this tract of the vagina (Fig. 6), and the microvillous elements were regularly arranged forming a sort of palisade (Fig. 6, inset). More caudally, near the squamo-columnar junction, however, the vaginal epithelium was composed of both cell types again. Further in this zone, some elements provided with very short microvilli and interspersed among tall and typical microvillous and ciliated cells could be found. Such cells might be likely an expression of squamous metaplasia (Fig. 7). At times, the walls of the upper 2/3 of the duct deepened in irregular crypt-like invaginations. These were continuous with the vaginal cavity and lined inside with an epithelium identical to that covering the parietal luminal surface (Fig. 8). Hence the simple cylindrical epithelium bordering the os of the cervical canal extended uninterrupted onto the ectocervix, into the vaginal fornices and, below these, lined both the upper and middle parts of the vaginal wall. The typical vaginal epithelium, i.e., squamous and pluristratified, was found only in the lower 1/3 of the duct, from the level of the pubic symphysis up to the vestibulum, with the squamo-columnar junction located approximately at the level of the upper edge of the symphysis.

The pluristratified squamous epithelium, consisting of about five cell layers, was provided with surface cells which, when viewed by SEM, showed mosaic features, appearing irregularly polygonal and well defined in contour (Fig. 9). These cells measured 10-12 μm in length and 8 μm in width; they were completely covered by densely packed microvilli. Such microprojections were short (0.2 μm in length) and stubby, ending with a bulbous apical protrusion (“club-tipped” microvilli). At the cell periphery, they formed ridges outlining the intercellular spaces (Fig. 10). Also observed scattered on the surface of some of these cells were irregular and convoluted structures likely originated by fusion of adjacent microvilli (Fig. 11). Furthermore, these structures appeared as typical “microridges” comparable to those occurring in several species, including humans, throughout the vagina. These, in fact, were highly branched, sometimes to the point of covering the entire cell surface. They were about 0.1-0.2 μm wide, 0.2-0.4 μm tall, and varied somewhat in length (generally 1 to 3 μm), owing to their extensive ramifications. Such formations have also been detected along the intercellular spaces, where they sometimes appeared even more developed (Fig. 12).

The gross anatomy of the entire rabbit vagina and the correlated main morphological characteristics showed by its epithelium as topographically analysed by systematic light microscope and SEM observations are reported and summarized in Figure 13a, b).

**Postcoital rabbits**

As revealed by light microscopy at 5 h post-coitum, the mucosa of the upper 2/3 of the vaginal wall, forming crypt-like invaginations, showed high epithelial cells with apical secretory protrusions. Spermatozoa resting on the irregular luminal surface of the mucosa were observed often in contact with the cell processes, some of which appeared as round formations detached from the cell body (Fig. 14).

In corresponding SEM pictures at the same time after mating, the non-ciliated cells covering the upper 2/3 of the vaginal wall appeared polygonal in contour and separated by well-defined spaces, so that together they formed an epithelial “carpet” similar to a Roman pavement (“cobblestone-like cells”). These cells possessed numerous microvilli often covered with small droplets. Some of these cells displayed dome-like surfaces hidden by very long microvilli, so that they appeared as a chrysanthemum (“chrysanthemum-like cells”) (Fig. 15). Mucosal intercolumnar crevices were extensively filled by a great number of clustered spermatozoa, whose abundance and high density were in contrast to a more scattered distribution on the top of the longitudinal folds. In general, sperm penetrated with their heads into the grooves of the mucosa and projected their tails toward the vaginal cavity (Fig. 16). At this stage, spermatozoa were in close contact with the epithelial surface, in respect to which they appeared variously oriented, either arranged tangentially and almost parallel to the epithelium, or assuming a bent to nearly perpendicular position to the lining cells. Thus, in the latter case, they could be thrust against the luminal surface so closely that their heads were engulfed by the cilium (Fig. 17).

Mainly the tips of the cilia of the epithelium were covered with a number of mucous droplets, so that groups of cilia might appear stacked and assembled together in bundles. Small amounts of these droplets frequently adhered to the spermatozoa, with mucus apparently spread around by the surrounding beating cilia mainly on the sperm acrosomal region (Fig. 18). In corresponding TEM images, aspects of vaginal epithelial mucification were confirmed. In fact, as the vesicles of mucus formed, they often diffusely filled the apical cell cytoplasm, even to the point of transforming it into mucified masses (Fig. 19). Spermatozoa were observed in close contact with the epithelium, and secretory vesicles, opening on the cell surface, seemed to shed their contents over the sperm heads (Fig. 20).
At 24 h after mating, the lining of the cranial 2/3 of the vaginal duct was largely represented by areas showing "chrysanthemum-like cells" of the type described above. At this time post-coitum, the features of the secretory activity of the mucosa were even more remarkable. In fact, the apical plasmalemma of the epithelial cells often appeared raised into irregular prominences involving wide zones of the luminal surface, and/or in oblong or spheroid, fused "blebs" (Fig. 21). Such plasmalemmal evaginations arose below the microvilli; as they developed, the overlying microvilli flattened out to the point of completely disappearing (Figs. 21, 22). SEM observations also revealed that, at 24 h following coitus, the unequal distribution of the spermatozoa on the free mucosal surface was still maintained, both where they were scarce, and within the crevices, where, on the contrary, they were abundant (Fig. 23). On the heads of the sperm occurring on the vaginal surface, mucus droplets were present either singly or assembled

**Fig. 13.** a. Gross anatomy of the rabbit vagina. In this animal two distinct uterine cervices project into the lumen in the upper part of the duct. The squamo-columnar junction is located approximately between the upper two thirds and the lower one third of the organ. b. The diagram reconstructed on parallel LM, TEM and SEM microtopographical data shows the main morphological characteristics of the mucosal epithelium lining the cervical canal and vagina in the rabbit. uh Uterine horn, f vaginal fornix, s-c j squamo-columnar junction, occ os of the cervical canal, c ciliated cell, m microvillous cell.
together in small amounts at the same time degenerative features consisting of a rupture of the plasmalemma covering the acrosomal region of some gametes were observed (Fig. 24).

At 10 days post-coitum, the cranial part of the mucosa was hypertrophic, consisting of high longitudinal folds separated by deep parallel grooves. The epithelial cells were exuberant, so that their borders were always well delineated (Fig. 25). At higher magnification, it has been noted that these elements project long and densely packed microvilli toward the lumen of the duct and appear as the "chrysanthemum-like" type. These cells, provided with large and irregular protrusions, extensively lined the mucosal surface. Under the SEM, these protrusions appeared flattened and consisted of characteristic smooth and pedunculated "leaf-like" processes often surrounded by microvilli (Fig. 26).

DISCUSSION

This study has shown that the rabbit vagina is both macroscopically and microscopically very different in comparison to that of other mammals, including humans. Both its length — if considered in relation to the body size of the animal — and especially the structure of its epithelium suggest that it accomplishes functions other than copulation.

Previous TEM and SEM reports on the lower parts of the female reproductive apparatus in mice and rats have revealed the fine structural characteristics of the vaginal mucosa. Many of these studies have satisfactorily described the peculiar features of its squamous epithelium, which is furnished with a luminal plasmalemma raised up to form either microridges or microvilli. The former, which are also called microplicae, are supposed to be capable of maintaining a protective layer of mucus on the free epithelial surface, thus reducing friction damage during mating (Parakkal, 1973; Centola, 1978; Lamb et al., 1978; Corbeil et al., 1985). The latter are short and stubby formations, ending with an apical expansion ("club-tipped microvilli") often associated with small circular holes, which serve as supplementary anchorage devices between overlapping cells of desquamating epithelia (Rubio, 1976; Centola, 1978).

Several papers also have emphasized morphological changes in the vaginal epithelium in various mammals according to the phases of the ovarian cycle, especially concerning its superficial cell layer. Specifically, this alternately undergoes keratinization, with the appearance of microridges, induced by estrogens, and then mucification, with the presence of microvilli, dependent upon progesterone, respectively (Parakkal, 1973; Rubio, 1976; Centola, 1978; Lamb et al., 1978; King, 1983; Corbeil et al., 1985; Winter-Hager and Kuhn, 1985; Ohnishi et al., 1986). Some of such features seem to be in agreement with others experimentally induced in the vaginal epithelium of various mammals by means of sex hormones administrated pre- and postnatally. These findings have generally been ascribed to the maturative estrogen action effects on the squamous epithelium, leading to keratinization (Eddy and Walker, 1969; Parakkal, 1973; Rubio, 1976; Cunha et al., 1978; Holderegger, 1978).

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**Fig. 14.** The epithelial cells lining the vaginal wall project their apex towards the lumen (L) of the duct. Spermatozoa (s-→) resting on the mucosal surface and in the crypt-like invaginations come in contact with the cells. LM, methylene blue Azur II stain, ×1,100

**Fig. 15.** Microvillous polygonal cells ("cobblestone-like cells") are separated by well-defined spaces. Note the various lengths of the microvilli and a number of small droplets interspersed on their tips. The cells covered by longer microvilli show dome-like surfaces, assuming a "chrysanthemum-like" appearance (arrows). SEM, ×2,000

**Fig. 16.** The majority of sperm heads fill the bottom of the longitudinal furrows of the mucosa and project their tails towards the lumen (arrows). In contrast to such an arrangement, note the relative scantiness of spermatozoa on the mucosal folds. SEM, ×500

**Fig. 17.** A sperm head almost perpendicularly oriented to the mucosal surface is engulfed by cilia (ci-→). Numerous microvilli (mi) cover adjacent cells. SEM, ×15,000

**Fig. 18.** Bundles of densely packed cilia support a quantity of mucous droplets on their tips (arrows). Small amounts of these adhering to the spermatozoa seem to be spread by the surrounding cilia mainly on the acrosomal region (arrowheads). SEM, ×7,500

**Fig. 19.** The cytoplasm of microvillous cells (m-→) contains a number of gathered mucous secretory vesicles (v), tending to form apical masses. c Ciliated cell, L vaginal lumen. TEM, ×4,400

**Fig. 20.** Mucous secretory vesicles (v) opening onto the epithelial surface occasionally shed their contents over a sperm head (arrow) in close contact with two microvillous secretory cells. mi Microvilli. TEM, ×9,600
Figs. 21-24. Vagina, upper part, 24 h post-coitum.

Fig. 21. Blebs bulging from the apical surface of microvillous cells (arrows). At this level microvilli become shorter and reduced in number. SEM, ×10,500

Fig. 22. Luminal side of the vaginal epithelium. The microvillous projections (mi) over the secretory mucified masses (asterisk) flatten and tend to disappear. L vaginal lumen. TEM, ×9,600

Fig. 23. Mucosal crevices holding numerous spermatozoa (arrows) outline epithelial surface areas, where many spermatozoa are still present. SEM, ×1,000

Fig. 24. Small secretory droplets (arrows) from adjacent ciliated cells occur on the sperm heads. In correspondence with these areas, a rupturing of the plasmalemma overlying the acrosome is noted (asterisk). SEM, ×9,000
1980; EROSCHENKO and OSMAN, 1986), whereas mucification or reduced cornification were related to progesterone (EDDY and WALKER, 1969; PARAKKAL, 1973; HOLDEREGGER, 1980).

Our correlated SEM and TEM investigations in the rabbit showed that the upper and middle parts of the vaginal duct are lined by a columnar monostratified epithelium, similar to that of the uterus including the endocervix and, in some zones, even of the oviduct (MOTTA and ANDREWS, 1976; BARBERINI et al., 1978; VAN BLERKOM and MOTTA, 1979; ODOR and BLANDAU, 1988). On the other hand, typically squamous epithelium (mechanical-type proper) extends only from the level of the pubic symphysis towards the vestibulum. In that part of the vaginal epithelium — similar in morphology to the endometrium and covering about the upper 10 cm of the duct — we noted that contact between the secretory droplets of microvillous cells and spermatozoa may occur. This sperm-epithelium relationship resembled that referred in the uterus of the same species (MOTTA and VAN BLERKOM, 1975; MOTTA et al., 1977; BARBERINI et al., 1978). It seems, therefore, that this long part of the duct may also contribute, as do the upper portions of the female reproductive tract, to sperm viability.

Further, with the purpose of clarifying the distribution of ciliated and microvillous cells in the cranio-caudal direction, our study allowed a more complete microtopographical surface map. This integrates former and still fragmentary observations on the exact location of these cell types in the vagina of this species (HAFEZ, 1972; KANAGAWA et al., 1972; MOTTA and VAN BLERKOM, 1975; PAKZAD and PAUFLER, 1982; BARBERINI et al., 1989a, b). In particular, our results demonstrated that the ciliated elements are abundant on the ectocervix and in the fornices, but progressively diminish in number to finally disappear from large areas of the middle part of the vagina. Near the squamo-columnar junction, however, these are numerous again (see diagram Fig. 13b). The presence of a highly ciliated vaginal epithelium is consistent with the assumption that spermatozoa might be moved by coordinated ciliary beating. Furthermore, their ascent is favored by rhythmical contractions of the parietal musculature (PAKZAD and PAUFLER, 1982), and by their own movements. The vaginal cilia might act also to facilitate the release and spread of secretions from microvillous cells over the sperm plasmalemma, in a way similar to that reported in the uterus and oviduct (HAFEZ, 1972; KANAGAWA et al., 1972; MOTTA and VAN BLERKOM, 1975; BARBERINI et al., 1978; FLÉCHON and HUNTER, 1981).

Our ultrastructural findings also confirm various interactions that occur between the epithelial surface and spermatozoa. Sperm heads often overlay the tips of microvilli and/or cilia, appearing to be supported by these structures, or else they are hidden among the cilia in such a manner that their tails project into the vaginal lumen.
The occurrence of spermatozoa in the rabbit vaginal crevices has also been reported by PHILLIPS and MAHLER (1978) and PAKZAD and PAUFLER (1982). Actually, at various times after copulation, the number of spermatozoa on the epithelial surface appeared to progressively diminish; on the contrary, they tended to assemble on the slopes, at the bottom of the grooves, and in the crypts from which their tails frequently stood out. It has been reported (BEDFORD, 1971) that the normal fertilization rate in the rabbit remains unaffected after the elimination of sperm from the vagina a few minutes post coitum. Therefore, the rabbit vaginal furrows and crypts, at least those occurring on the ectocervix and in the fornices, and because of their depth, may act in accordance to functions proposed for the cervical crypts of other species such as the ewe (MATTNER, 1963a, b) and humans (MOGHISI, 1973): namely, as sperm reservoirs or “parking areas” where spermatozoa might remain alive nestled within the cell secretions. Furthermore, the occurrence in the rabbit cervical crypts of motile sperm, which contact ciliated cells and are hardly removable by flushing, has been reported (OVERSTREET and COOPER, 1978). From such grooves they could migrate at established intervals of time in the direction of the uterus and oviduct. Likewise, the role of storage sites with the possible survival of spermatozoa has also been suggested for the deep infoldings of the uterine tube in the pig (DU MESSNIL DU BUISSON and DAUZIER, 1955; RIGBY, 1966; FLÉCHON and HUNTER, 1981; HUNTER, 1981). In this regard, it has been recently demonstrated that spermatozoa stored in crypts of the oviductal isthmus of the hamster maintain their motility and survive in percentages markedly higher than that of sperm free in the lumen, until 8 h after mating (SMITH and YANAGIMACHI, 1990). Thus, in spite of the more caudal location of the furrows and crypts of the vagina in comparison with those of the tube, the result would be in both cases an increased possibility that sperm would reach the ovum.

Some intriguing morphodynamics have also been revealed concerning the columnar vaginal epithelium. These are very likely related to the levels of sex hormones circulating pre and post coitum. In fact, copulation in the rabbit immediately induces LH release, with a serum peak of this hormone occurring in 1–2 h (SCARAMUZZI et al., 1972), and ovulation occurring at approximately 10 h thereafter (WALTON and HAMMOND, 1928). The ultrastructural aspects of the apocrine and microapocrine secretions of the vaginal microvillous epithelial cells closely resemble those found in the rabbit uterine mucosa under similar postcoital conditions (i.e., 15, 24, 36, 58 h and 10 days) (BARBERINI et al., 1978). Similar data were also reported in the endocervical epithelium of this species after the administration of HCG (ODOR and BLANDAU, 1988). These secretory features of the vagina, though they also occurred slightly in the estrous phase, may thus be interpreted as prevailingy progesterational-type modifications. Similarly, the “leaf-like processes” visualized by SEM in our investigation might correspond to the so-called “pinopods”. These structures, considered to be sites of endocytosis, were mainly found in the rat uterus in the early days of pregnancy, where they have been also interpreted as indicating decidualization (ENDERS and NELSON, 1973; PARR, 1980). In this respect, the uptake of proteins from the uterine lumen by the apical surface of the epithelial cells has been suggested to be relevant to immunological sensitization against foreign materials in preparation for embryo attachment, since the uterine cavity can be exposed to significant amounts of exogenous substances in consequence of mating (TUNG et al., 1988).

A final note of interest concerns the nature and possible embryonic derivation of this duct. On the basis of the different histological findings in the upper 2/3 and in the lower 1/3 of the rabbit vagina, the hypothesis of the double origin of this organ (KOFF, 1933; CUNHA, 1975; O’RAHILLY, 1977) seems to be corroborated, in opposition to the unitarian theory of its formation (MEYER, 1934; BULMER, 1957; MINH et al., 1984). In fact, on account of the similar morphological features of the upper and middle parts of the vaginal mucosa to those found in the oviduct and in the uterus, the upper 2/3 of the vagina might derive from the paramesonephric (Müller’s) ducts. On the other hand, from the urogenital sinus, which is endodermal in origin, there might arise only that portion covered by a pluristratified squamous epithelium in continuity with that of the vulva. Indeed such a deduction about the origin of the vagina, based simply upon mere morphological observations of the adult rabbit mucosa, confirms some embryological studies in this animal. Specifically, it has been reported that in this species the Müllerian epithelium lines the majority of the vaginal walls (the so-called “Müllerian vagina”), and that only the lower part of the duct originates from the urogenital sinus (the “sinus vagina”), when this undergoes a frontal partition in the urethra ventrally and vagina dorsally (FORSBERG, 1973, 1976).

In conclusion, both the noteworthy length and the unique structure of the rabbit vagina suggest that the actual coital organ is only its inferior portion, which is provided with a squamous stratified epithelium. In the upper parts, owing to the endometrium-like fea-
tures of its mucosa, the biological processes facilitating sperm viability might begin earlier in this species. The rabbit vagina, therefore, may represent not only a duct simply related to copulation, collection and/or conservation of spermatozoa and operation as a fetal passageway, but also might perform further particular roles in the complex reproductive events of this species.

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Prof. F. BARBERINI
Department of Anatomy
University of Rome "La Sapienza"
Faculty of Medicine
Via A. Borelli, 50
00161 Rome, Italy