

Development and Structure of the Glandopreputial Sulcus of the Human Clitoris With a Special Reference to Glandopreputial Glands

SEBASTIAN C.J. VAN DER PUTTE* AND DAISY M.D.S. SIE-GO

Department of Pathology, University Medical Center Utrecht, Utrecht, The Netherlands

ABSTRACT

The structure and development of the sulcus between the glans and prepuce of the human clitoris have hardly been investigated. Interest in its structure was raised when in the female, in contrast to the male, glands were found to develop from the solid lamella-like precursor of the glandopreputial sulcus. It prompted a further histological analysis of the sulcus in female fetuses and newborn and an extension of that study to clitorises of adult women. The investigation showed that in the clitoris, in contrast to the penis, the transformation of the glandopreputial lamella into the open sulcus was mostly incomplete and apparently remained so throughout life. As a most striking and probably exclusively female feature, two to eight eccrine glands developed from the base of the lamella in fetuses older than 14.5 weeks gestation. These glands formed secretory coils near and occasionally inside the adjacent distal corpora cavernosa. Some glands showed atresia, cystic dilatation, and squamous metaplasia. A remarkably similar picture was observed in the adult clitorises, in which the secretory coils were often found between the large blood vessels and nerves to the glans and were connected to the sulcus by long excretory ducts. All glands revealed unmistakably eccrine features. It is suggested that their secretion moistens the female glandopreputial sulcus, which is not lubricated by urethral secretion as in the male. The findings may explain the rare clitoral phimosis, cysts, and some pilonidal sinuses. *Anat Rec*, 294:156–164, 2011. © 2010 Wiley-Liss, Inc.

Key words: clitoris; glandopreputial sulcus; glandopreputial gland; development; histology

The sulcus between the glans and the prepuce of the clitoris is an inconspicuous structure, which has received little attention in the literature. As even its existence was mostly ignored (Rhodin, 1974; Moore and Daley, 1996; McLean, 1999; Larsen, 2001; O'Connell et al., 2005; Wilkinson and Hardt, 2007; Standring, 2007), factual knowledge about its development and microscopic anatomy is scarce. The data suggest that, as in the male, it develops on the dorsum of the clitoris as a down-growth of a solid epithelial lamella, which temporarily connects the prepuce to the glans and later splits into the opposed surfaces of the preputial sac lined by cornified stratified squamous epithelium (Glenister, 1956; Cold and Taylor, 1999; Van der Putte, 2005). In line with the results of recent extensive studies of the male prepu-

tial sac, it is considered to be devoid of glands (Barreto et al., 1992; Cold and Taylor, 1999; Velazquez et al., 2007). Moistening of the female sulcus would be the result of fluid transudation as that sulcus is excluded from urethral secretions, which form most of the

*Correspondence to: Dr. S.C.J. van der Putte, Department of Pathology, H04.312, University Medical Center Utrecht, P.O. Box 85500, 3508 GA Utrecht, The Netherlands. Fax: 0031302544990. E-mail: s.c.j.vandeputte@umcutrecht.nl

Received 10 March 2010; Accepted 31 August 2010

DOI 10.1002/ar.21279

Published online 2 November 2010 in Wiley Online Library (wileyonlinelibrary.com).

TABLE 1. Clinical and histological data of 24 specimens of human fetuses

Fetus no.	Gestational age (weeks) ^a	Crown-rump length (mm)	Plane of sections ^b	Mounting of sections
1	10.5	35	Sagittal	Series
2	11	45	Sagittal	Series
3	11.5	55	Sagittal	Series
4	11.5	60	Sagittal	Series
5	12	65	Frontal	Series
6	12	70	Sagittal	Series
7	12	70	Transverse	Series
8	13	85	Sagittal	Series
9	13	85	Sagittal	Series
10	14	93	Sagittal	Series
11	14.5	105	Frontal	Step sections
12	15	110	Sagittal	Series
13	15	110	Frontal	Series
14	16	125	Sagittal	Step sections
15	17.5	140	Frontal	Step sections
16	18	150	Sagittal	Series
17	21	182	Frontal	Series
18	22	190	Sagittal	Step sections
19	26	230	Frontal	Series
20	26	230	Sagittal	Step sections
21	26	230	Transverse	Single section
22	28	260	Frontal	Single section
23	35	320	Frontal	Single section
24	39	360	Frontal	Step sections

^aThe estimated gestational ages are approximations. Values are based on clinicopathological information for fetuses under 12 weeks and on the growth curve for crown-rump lengths by Maroun and Graem (2005) for fetuses of 12 weeks and older.

^bOrientation to the surface of the perineum between the anal and vestibular orifices.

smegma in the male (Parkash et al., 1973; Cold and Taylor, 1999).

Surprisingly, during an investigation into the development of the human perineum, tubular glands were found that developed from the deepest part of the solid lamella (Van der Putte, 2005). It raised the question whether a causal relationship could exist between these glands and poorly understood clitoral cysts, especially because the glands might show segmental atrophy and dilatation during the last trimester before birth. It stimulated further research in both fetal and adult clitorises. The findings were correlated to data in the literature about clitoral cysts and other disorders.

MATERIALS AND METHODS

The investigation was performed on specimens from female fetuses, which formed part of a previous study on the development of the perineum (Van der Putte, 2005), and from adults. The selection comprised 24 normal clitorises from spontaneously born fetuses (Table 1). They ranged in age from 10.5 to 39 weeks gestation and in crown-rump length (CRL) from 35 to 360 mm. The specimens were fixed in 4% formalin, embedded in paraplast, and sectioned at 5–7 μ m. The plane of sectioning was transverse, frontal, or sagittal to the surface of the perineum between anal and vestibular openings. Sections were mounted in complete series, series of step sections (1:5 or 1:10), or as single sections.

The surgical pathology files of the University Medical Centre Utrecht were searched for vulvectomies, that is, complete or partial surgically removed vulvas. Pathology reports were reviewed for the indication of vulvectomy, age of the patient, and the presence of a clitoris, which was not involved in the pathological process. The 30 vulvectomies were performed for squamous cell carcinoma (N = 26), malignant melanoma (N = 2), and lichen sclerosus (N = 2). The patients were between 43 and 92 years old (mean age: 70 years). The pathological specimens consisted of transverse slices removed from clitorises as part of a standard pathological procedure. They could contain cranial, middle, and/or caudal segments of a glandopreputial sulcus. In most cases, a single slice had been removed. Multiple slices allowing a picture of the whole clitoris were available in five cases. The specimens were fixed in 4% formalin and embedded in paraplast. Eleven single slices and five multiple slices from one clitoris were serially sectioned at 5 μ m and mounted as step sections (1:5 or 1:10) and as complete series at selected levels. Sections were stained with hematoxylin and eosin. Histochemical tests comprised periodic acid-Schiff (PAS) staining with or without diastase predigestion for the detection of glycogen and mucoproteins and Alcian blue (pH 2.5) for mucoproteins.

Immunohistochemical stains comprised the use of antibodies detecting the low-molecular-weight keratin (CK7) and smooth muscle actin (SMA) for the discrimination between secretory and excretory segments of cutaneous glands, and epithelial membrane antigen (EMA) for the identification of intercellular secretory canaliculi, which are characteristic of eccrine glands. The tests were performed on rehydrated 4- μ m paraffin-embedded sections using an automatic Bond-Max immunohistochemistry system (Leica, Newcastle). Slides were heated for 10 min at 56°C. CK7 was brought in a pH 6.0 citrate buffer for 20 min at 99°C, EMA was similarly treated by ethylene diaminetetraacetic acid, pH 9, and no antigen retrieval was needed for SMA. The anti-CK7 clone OVTL 12/30 (Biogenex, San Ramon, USA) was used at a dilution of 1:800, the anti-EMA clone E29 (Dako, Glostrup, Denmark) at a dilution of 1:400, and the anti-alpha-SMA clone 1A4 (Sigma, St. Louis, USA) at a dilution of 1:16,000. The primary antibody incubation was 15 min. It was followed by postprimary incubation for 8 min, polymer-peroxidase for 8 min, and diaminobenzidine for 10 min (Bond polymer refine detection system) at room temperature. Sections were counterstained with hematoxylin. Tissues that were previously shown to demonstrate reactivity with the different antibodies were used as positive controls.

This human research was performed according to the guidelines of the University Medical Centre Utrecht.

RESULTS

Development of the Glandopreputial Lamella and Sulcus in Female Fetuses

The first indication of the formation of the glandopreputial lamella was an increase in the concentration of basal cells with dark nuclei in the epithelium covering the peripheral extension of dense stroma of the glans at the early fetal stage between ~10.5 and 11.5 weeks gestation (35–60 mm CRL) (Fig. 1). An initial depression filled by stratified squamous epithelium (Fig. 1a) was

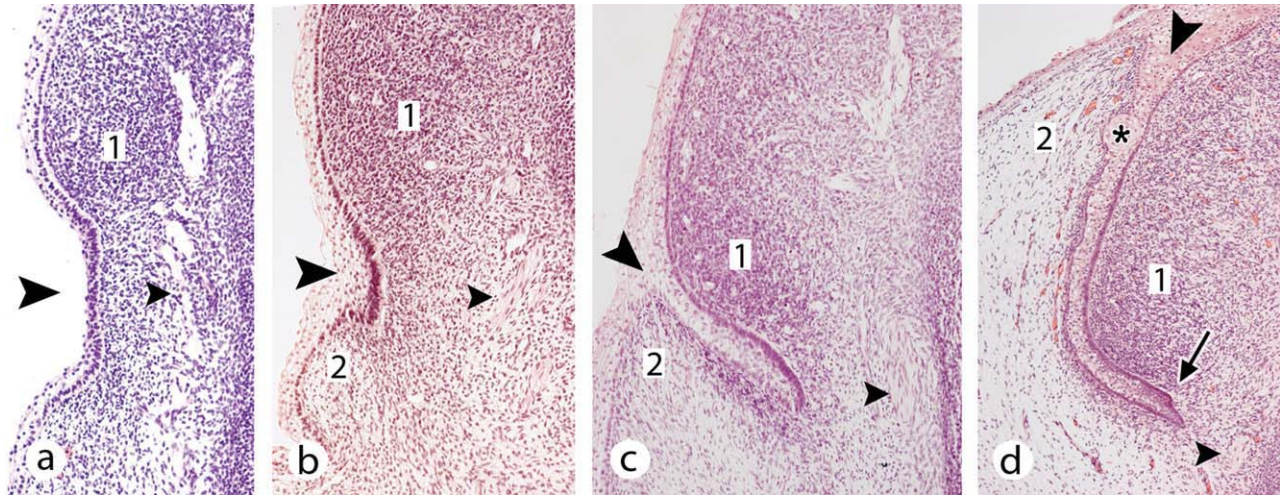


Fig. 1. Early development of the glandopreputial lamella of the clitoris in human fetuses of 11.5 (a), 11.5 (b), 12 (c), and 14.5 weeks (d) gestational age [55, 60, 65, and 105 mm crown-rump length (CRL)]. The figures demonstrate the transformation of a shallow sulcus lined by epidermis with strikingly crowded nuclei into a solid lamella (large arrowheads) while preserving its basic structure and

intimate relationship to the expanding corona of the glans (1), distalward growing prepuce (2), and underlying nerves to the glans (small arrowheads). This indicates that the lamella grows outward in association with the corona and prepuce. Asterisk, whorl; arrow, earliest primordium glandopreputial gland. Magnifications: a, $\times 80$; b and c, $\times 100$; d, $\times 65$.

later transformed into an outward growing solid glandopreputial lamella in concert with the increase in volume of the flanking corona of the glans medially and the prepuce laterally (Fig. 1b). The prominent dense basal cell layer remained at the deepest part of the lamella and gradually decreased distalward over the glans. It also extended for a short distance over the inner layer of the prepuce derived from the originally most peripheral glans stroma, before passing into stratified squamous epithelium with paler basal cells changing into epidermis near the edge of the prepuce (Fig. 1c,d).

The lamella slightly broadened in fetuses older than ~ 14.5 weeks gestation (105 mm CRL) and revealed the appearance of variable numbers of whorls at the side of the prepuce causing an undulating epithelial outline. Most whorls had disappeared at the end of pregnancy but some had been transformed into small isolated cavities with central cornification inside an otherwise still solid lamella.

Development of Glandopreputial Glands in Female Fetuses

A most striking feature of the female lamella was the development of multiple gland primordia, which developed from its basal layer in a restricted area at the glanular side of its deepest part in female fetuses older than ~ 14.5 weeks gestation (105 mm CRL) (Figs. 1d, 2a,c). These primordia numbered five to eight in total for one lamella. They were most numerous in the middle part of the sulcus and could be absent at the cranial and caudal sides. A variable proportion of these primordia, mostly near the base of the lamella, formed tubular glandular structures with a distinct two-layered epithelium which ended in a broader, at first solid and later cribriform "bud" with developing canaliculi characteristic of eccrine differentiation (Fig. 2a-d). These glands extended deep

into the clitoris where they were found in tissues very close to the corpora cavernosa, often between nerves and large blood vessels related to the glans, and occasionally inside the corpora (Fig. 2e). One or two glands consistently occurred at each side but often glands also developed from the cranial part of the lamella into tissues at the dorsum side of the distal corpus cavernosum (Fig. 2d). Some primordia appeared to become abortive elements by revealing atresia, squamous metaplasia, or cystic dilatation (Fig. 2a-c,f). Immunohistochemistry of the secretory coils demonstrated reactivity for the low-molecular-weight keratin CK7. In the newborn, EMA was variably present inside the cytoplasm of the epithelial cells. It was concentrated on the luminal surface, thereby outlining intercellular canaliculi characteristic of eccrine differentiation (Fig. 2e). SMA-positive myoepithelium was positioned at the periphery. Excretory ducts showed a concentration of EMA in the lumen and a gradual disappearance of CK7 and SMA in the segment following the secretory coil. Histochemistry showed glycogen granules in some secretory cells and a faint reactivity for mucin in alcian blue and diastase-resistant PAS staining in most cells.

Structure of the Glandopreputial Sulcus of the Adult Clitoris

The glandopreputial sulcus of the clitorises in the predominantly old age group investigated revealed a considerable variation in structure from completely solid lamellas to fully open sulcuses. The most common variant consisted of an open sulcus superficially and a variably long tapering solid lamella at a deeper level (Fig. 3a-c). The open part showed a lining of markedly cornifying stratified squamous epithelium at the side of the glans. Strong cornification was also observed at the side of the prepuce, but in half the number of specimens, a

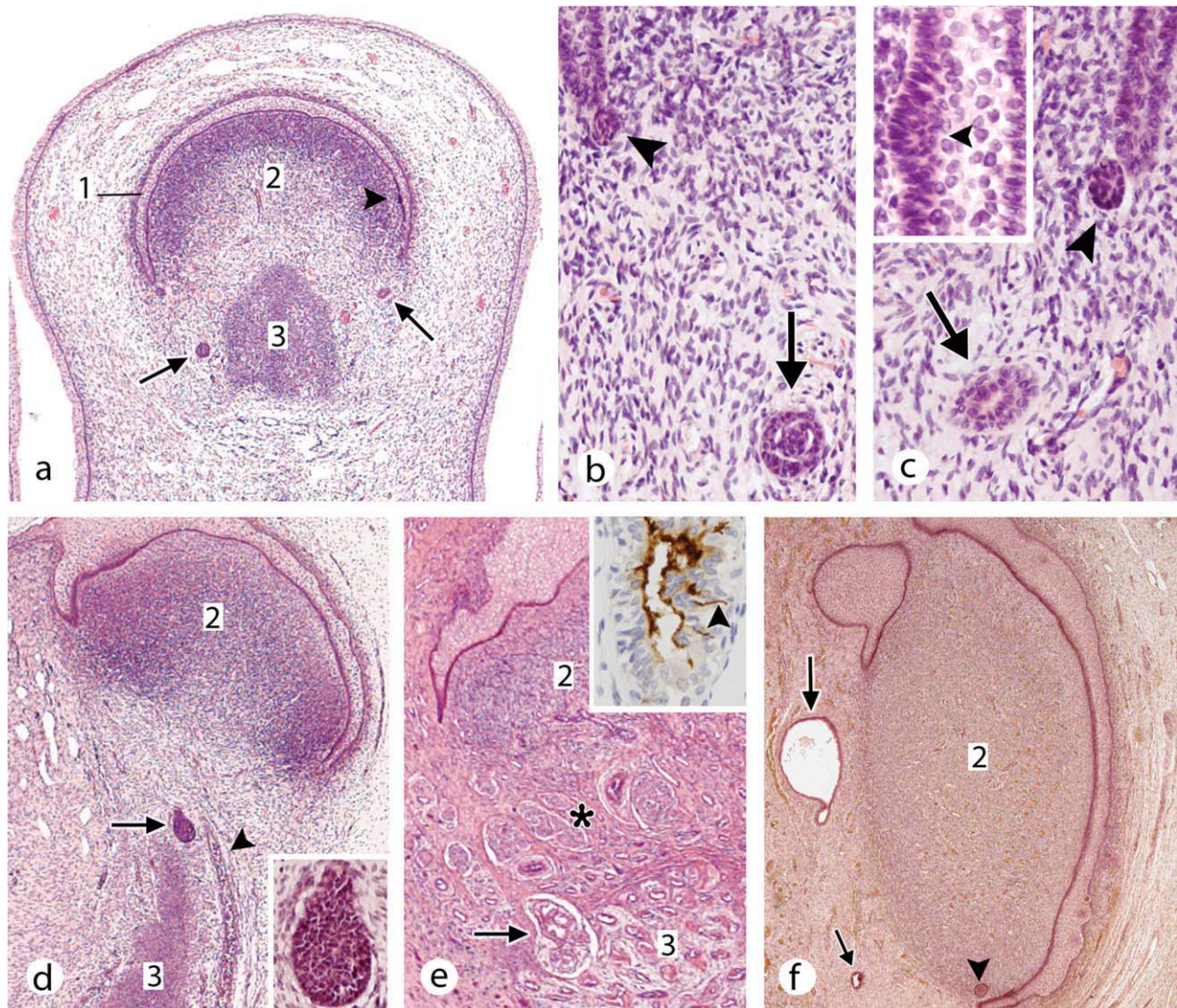


Fig. 2. Development of glandopreputial glands in female human fetuses of 21 (a–c), 18 (d), 39 (e), and 35 weeks (f) gestational age (182, 150, 360, and 320 mm CRL). (a–c) A transverse section through the clitoris at low magnification (a) shows the topography of a very early gland primordium (arrowhead) and early glands (arrows) which grew from the glandopreputial lamella (1) at the base of the glans (2) into the tissues around the distal corpus cavernosum (3). Details demonstrate in b a still cribriform secretory segment (arrow) and an abortive primordium (arrowhead) and in c an excretory duct (arrow) and another abortive primordium (arrowhead). Inset, early primordium (small arrowhead). (d) A parasagittal section illustrates an early and

still solid eccrine secretory bud (arrow) (detail in inset) in close proximity to the corpus cavernosum (3) and dorsal artery of the clitoris (arrowhead). (e) A frontal section shows a secretory coil (arrow) which is partially positioned inside the corpus cavernosum (3). Inset, EMA shows intercellular canaliculi (small arrowhead) characteristic of eccrine differentiation. 2, glans; asterisk, area of large nerves and vessels of the glans. (f) A parasagittal section reveals a normal (small arrow) and a dilated excretory duct (large arrow) and an abortive primordium (arrowhead) beneath the glans (2). Magnifications: a, $\times 30$; b and c, $\times 185$, inset $\times 280$; d, $\times 48$, inset, $\times 140$; e $\times 145$, inset, $\times 225$; f, $\times 25$.

variably long deep part is lined by a noncornifying para-keratotic stratified squamous epithelium (Fig. 3d). The often strikingly thin solid part may have formed irregular long and thin rete ridges and may contain a few whorls and small cavities (Fig. 3a). This combination of solid and open sulcus was observed in caudal and middle segments of the sulcus in particular.

A completely open sulcus lined by a markedly cornified stratified squamous epithelium occurred in five specimens in the middle and cranial parts of the sulcus.

A solid lamella was seen in sections of caudal segments of seven clitorises. In six of these cases, informa-

tion about other parts of the lamella was not available. In the seventh case, the whole lamella was proven solid.

A hair shaft was found inside the lumen of two sulcuses in which, like in the other sulcuses, neither hair follicles or sebaceous glands were observed.

Glandopreputial Glands of Adult Clitorises

Glandopreputial glands were found in 27 of 30 clitorises. They were absent in three clitorises represented by single slices from caudal or cranial parts of the sulcus, which could not be considered representative for the

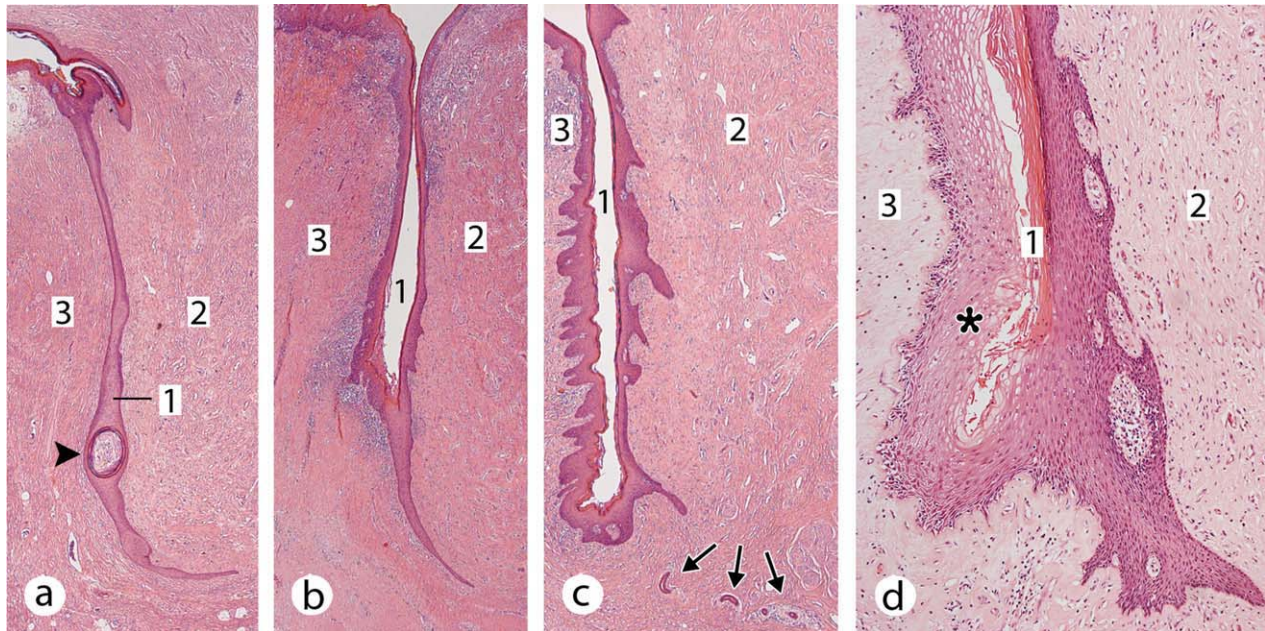


Fig. 3. Glandopreputial lamella and sulcuses in clitorises of adult women. Transverse sections demonstrate a range from a completely solid lamella (1) in **a** to the most common variant of a partially open and partially solid variant (1) in **b**, and a completely open sulcus (1) in **c**. The epithelium is stratified

squamous and may form a complete horny layer in the sulcuses but may also differentiate into a noncornifying epithelium at the deep preputial side (asterisk in **d**). 2, glands; 3, prepuce; arrowhead, cornified cavity; arrows, glandopreputial gland. Magnifications: **a**, $\times 17$; **b**, $\times 19$; **c**, $\times 22$; **d**, $\times 57$.

sulcus as a whole. The limited number of completely analyzed clitorises did not allow a clear overall assessment of the number and distribution of glands in a certain clitoris. However, the collected data could suggest that, similar to the pattern in the fetus, one or two glands occurred on each side and none to three glands at the cranial side (Fig. 4).

The topography had not altered either when compared with the pattern in the fetuses. The glands were embedded in dense parallel and longitudinal oriented connective tissue directly related to the clitoris (Figs. 3c, 4, 5a,b). Nineteen secretory coils and 33 excretory ducts were found. The coils were still in the direct vicinity of the main neurovascular structures related to the glands (Fig. 5a).

The secretory coils occurred in two types. A minority (7 of a total of 19 secretory coils) revealed the characteristic morphology of typical eccrine glands with their dark mucoid cells and clear glycogen-containing cells and a peripheral layer of SMA-reactive myoepithelium. Between the cells were EMA-positive canaliculi characteristic of eccrine secretory epithelium (Fig. 5c). A majority of the coils were lined by a less distinctive epithelium consisting of a single layer of monotonous columnar cells with a weakly staining eosinophilic or amphophilic cytoplasm. This epithelium could also show intercellular canaliculi, but these were often less numerous than in the characteristic variant (Fig. 5d). The CK7-positive secretory epithelium passed gradually into the conspicuous two-layered cuboidal epithelium at the transition to the CK7- and SMA-negative excretory ducts.

As the result of a considerably increased distance between these structures and the glandopreputial lamella after birth, the excretory ducts had lengthened considerably. All 33 excretory ducts revealed the eosino-

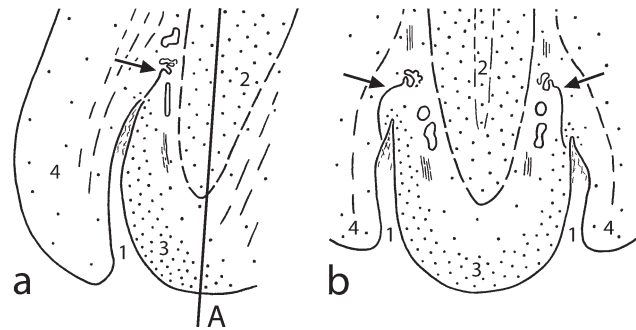


Fig. 4. Diagrams of a midsagittal and a transverse section through the adult clitoris to illustrate the topography of glandopreputial glands and the direction of sectioning. **(a)** The midsagittal section shows a glandopreputial gland (arrow) extending from the cranial part of the glandopreputial sulcus (1) into the vicinity of the corpus cavernosum (2) where its secretory coil is situated amid blood vessels and nerves of the glans (3). 4, prepuce. **(b)** A transverse section (plane of section A indicated in **a**) illustrates bilateral glandopreputial glands (arrows) opening into the deep glandopreputial sulcus. Magnification: $\times 10$.

philic cuticle-like luminal border typical of the eccrine excretory duct and lacked myoepithelium (Fig. 5d). The number of small spinal duct cells increased before the duct merged with the epithelium of the wall of the glandopreputial sulcus or lamella. In three specimens, a segment of the duct was atrophic (Fig. 5e) before it formed a small cystic dilatation and ended blindly near the lamella. A few small cysts were completely isolated. Unlike the ducts in the fetuses, those in the adults opened at various positions into the glandopreputial lamella or sulcus. Orifices were found at the glandar side, at the base, or at the side of the prepuce. The

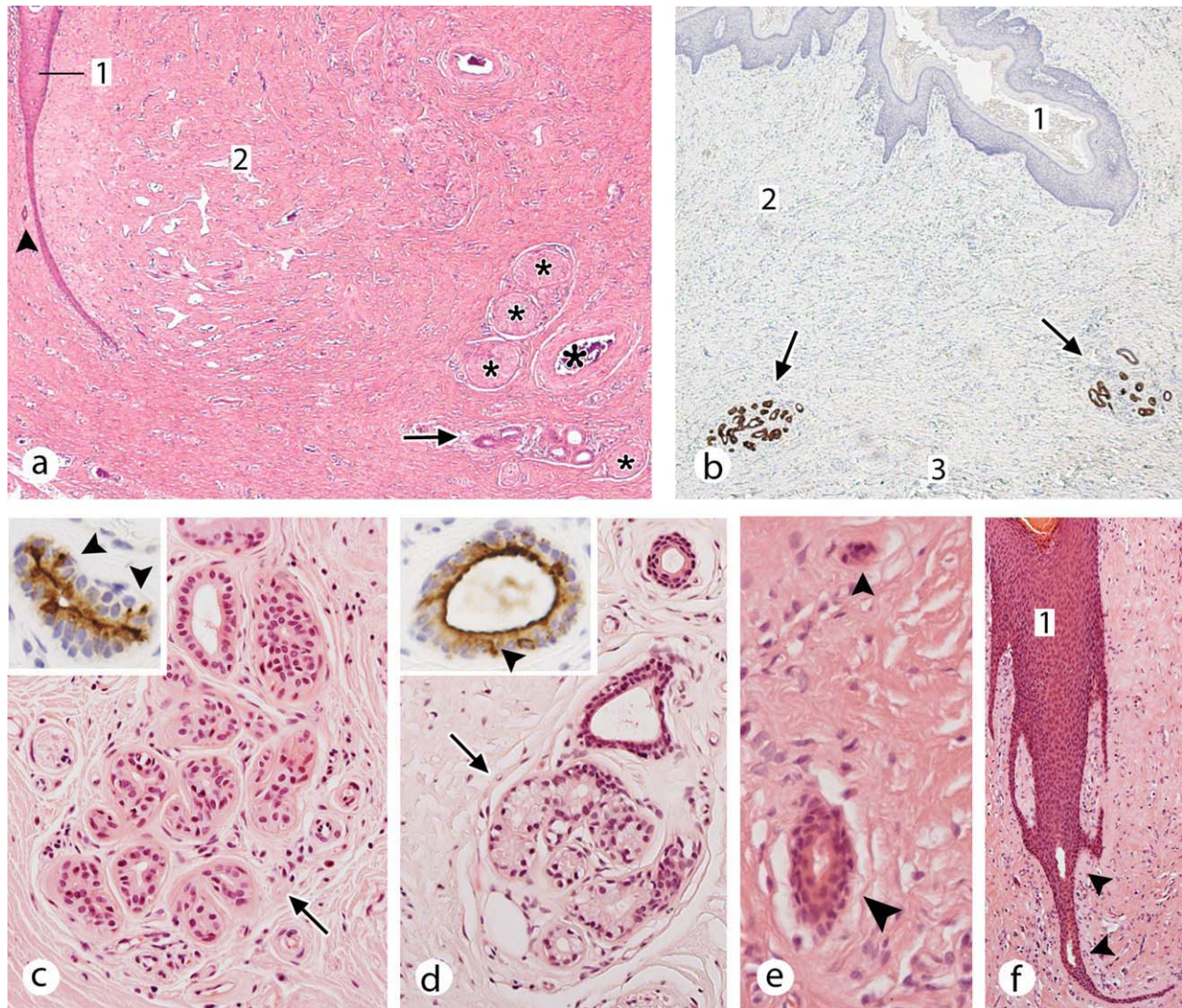


Fig. 5. Glandopreputial glands in clitorises of adult women. (a) The typical configuration of a glandopreputial gland opening into the lateral part of the glandopreputial sulcus (1) shows a secretory coil situated between a large artery (large asterisk) and nerves (small asterisks) of the glans (2). A typical eccrine excretory duct (arrowhead) courses lateral to the solid part of the sulcus before opening into its superficial open part. (b) Anti-cytokeratin 7 highlights the secretory coils (arrows) of two glands, which are situated anterior to the tip of the corpus cavernosum (3) and open into the cranial part of a glandopreputial sulcus (1). 2, glans. (c and

d) Secretory coils (arrows) demonstrate the variegated histology of a typical eccrine gland (c) and the more monotonous epithelium of a variant eccrine gland (d). The insets show examples of EMA-stained eccrine canaliculi (arrowheads) in similar glands. (e) A detail of an excretory duct demonstrates the usual eccrine structure (large arrowhead) and in this specimen also an atrophic segment (small arrowhead). (f) A terminal segment of an excretory duct (arrowheads) traverses the whole length of a solid lamella-like part of a glandopreputial sulcus (1). Magnifications: a, $\times 35$; b, $\times 12$; c and d, $\times 160$, inset, $\times 260$; e, $\times 275$; f, $\times 60$.

position of the orifices at the right side could differ from those at the left side in the same fetus. Almost all ducts entered the open part of a sulcus superficial to the solid part. One duct showed a protracted intraepithelial course through the whole length of a solid part to the open segment (Fig. 5f). In a specimen with a completely solid lamella, a duct at the left side ended in a small isolated cornified intralamellar cavity and two ducts at the right side ended blindly in small dilatations outside the lamella.

The glands were often surrounded by loosely arranged finely fibrillar connective tissue and intercellular mucin.

DISCUSSION

Terminology

The inconspicuous anatomical structure between the glans and the prepuce of the clitoris has, to the best of our knowledge, yet not been named. The term "preputial sac" used for the male counterpart seems inappropriate for the very small female sulcus, which is also partially solid in most instances. The frequently used term "glandular lamella" for the preceding solid plate (Johnson, 1920; Glenister, 1956; O'Rahilly and Müller, 2001) ignores the essential participation of the prepuce. The term "glando-preputial lamella" used for the initial solid plate by Hamilton and Mossman (1972) in their textbook

on embryology seems more appropriate. The present introduction of the term "glandopreputial sulcus" for the succeeding open sulcus is in line with this terminology.

Development of the Glandopreputial Lamella and Sulcus in Female Fetuses

This investigation demonstrated that the neglected glandopreputial lamella and sulcus of the clitoris are actually interesting structures. The scarce data reported (Saalfeld, 1899; Glenister, 1956) supported the general idea that there was a basic similarity in formation and morphology of the sulcus between male and female with the exception of the vestibular groove which keeps the lateral margins of prepuce and lamella separated in the female. With respect to the formation of the sulcus and prepuce, more extensive research in the male (Schweigger-Seidel, 1866; Herzog, 1904; Hart, 1908; Johnson, 1920; Hunter, 1935) eventually resulted in the now generally accepted theory of a combination of preputial folding over the glans penis and simultaneous ingrowth of an epithelial lamella (Glenister, 1956).

In a recent study, however, it was noted that during the whole developmental process, the deep margin of the lamella did not change its original position close to large underlying nerves passing toward the glans (Van der Putte, 2005) as had also been observed by Johnson in 1920. This is inconsistent with an inward growing lamella. As there were no indications of a folding of the prepuce over the glans either, it was suggested that the lamella grows outward together with the flanking and intimately connected prepuce and corona of the glans.

No information was found about the transformation of the solid glandopreputial lamella of the clitoris into the (partially) open sulcus. Rare studies of the process in the male suggested a confluence of whorl-derived small cornified and initially isolated cavities which develop inside the solid lamella (Gairdner, 1949; Van der Putte, 2005) or a central keratinization under influence of androgen hormones (Oster, 1968).

This study could not reconstruct this process in the female because it takes place after birth and specimens from children and young adults were not available. However, it did reveal that the number of cornified cavities in the still solid lamella in third trimester female fetuses was far smaller than the number in their male counterparts. In a fetus of 39 weeks gestation, a central split bordered by parakeratotic noncornified stratified squamous epithelium extending from the opening between the labia minora into a solid lamella without cornified cavities suggested that such splitting may play an important role in at least a proportion of the sulcuses. This process may be influenced by sex hormones as prenatal treatment of male rhesus monkeys caused a delay in separation (Thompson et al., 1981).

Development of Glandopreputial Glands in Female Fetuses

The development of tubular glands from the deepest part of the glandopreputial lamella in every fetus studied has, to the best of our knowledge, not been reported before. Their development in such a "remote" spot at the base of the glans is most remarkable. It is true that the lamella derives from the early epidermis that covers

the genital tuberculum and may therefore be considered capable of forming epidermal appendages. But their origin from glans epithelium at such an isolated place far from the nearest cutaneous appendages, which develop many months later at the outside of the prepuce, may explain why they were not observed before.

The histiogenesis of the glands revealing a large terminal bud differentiating into a secretory coil with intercellular canaliculi strongly suggested that they were eccrine glands.

The early development of the glands at a stage that the base of the glandopreputial lamella is very close to the major blood vessels and nerves of the glans and to the ill-defined extremity of the corpus cavernosum explains the intimate relationship between the structures which will apparently last for life.

The change in position of a number of orifices from the medial to the lateral sides of the glandopreputial sulcus is probably due to alterations in shape and structure of the lamella.

Structure of the Glandopreputial Sulcus of the Adult Clitoris

Information about the microscopic anatomy of the adult female glandopreputial sulcus appears practically nonexistent. This investigation demonstrates that the female sulcus is not merely a small variant of the male sulcus but has some specific features. Apart from the posterior interruption by the vestibular opening, the study shows that most sulcuses of the clitoris had not split completely as is the normal process in the male, but had preserved the solid lamella-like character of the fetal stage to a variable degree. In addition, this distinctive female sulcus proved to have its own specific moistening system of glandopreputial glands, whereas the moistening of the male preputial sac depends on fluid produced by the urethra and its glands (Cold and Taylor, 1999).

Glandopreputial Glands of the Adult Clitoris

No information was found in the literature about fluid-producing glands opening into the glandopreputial sulcus of the female clitoris. In the only report dealing with the question whether such glands occur in the female, their existence was denied (Saalfeld, 1899). Long-lasting and sometimes fierce discussions about their presence in the male had recently resulted in the conclusion that the concept of preputial glands discharging their products in the preputial sac was in fact a myth (Baretto et al., 1992; Taylor et al., 1996; Cold and Taylor, 1999). However, these disputes were about sebaceous glands (Hyman and Brownstein, 1969). The absence or presence of other types of glands opening in the preputial sac was not disputed (Saalfeld, 1899; Parkash et al., 1973; Taylor et al., 1996).

In a previous study, it was demonstrated that, indeed, no glands developed inside the preputial sac of male fetuses apart from a single primordium in an 18-week-old (150-mm CRL) fetus (Van der Putte, 2005). This study showed that the situation is distinctly different in the female clitoris by revealing that from the early fetal stage of 14.5 weeks gestation to old age, tubular glands

probably form a normal constituent of the glandopreputial sulcus of the clitoris.

The striking correspondence in the number of glands and in the intimate relationship of the secretory coils to the central structures of the clitoris between fetuses and aged women is in accordance with the notion that eccrine glands serve the individual for life (Folk and Semken, 1991). This close relationship itself is best explained by their original proximity at an early developmental stage.

The glandopreputial glands proved to be eccrine glands because all excretory ducts had the small diameter and typical luminal cuticle of the eccrine duct. In addition, a large proportion of the secretory coils showed the light and dark cells with intercellular canaliculi of the typical eccrine gland. The glands that showed a less characteristic and more monotonous secretory epithelium most likely represented a variant of the typical eccrine gland because they revealed canaliculi in some specimens and differed markedly in morphology from apocrine, apoeccrine, and mammary-like glands (Van der Putte, 1991, 1994).

The human glandopreputial glands differ essentially from the "preputial glands" observed in mammals such as rats (Reznik and Ward, 1981) and mice (own unpublished observations), which originate from the epidermis at the outside of the prepuce and are modified sebaceous glands.

The consistency in presence and configuration of the glands precludes an ectopic character and indicates that the glands are an essential part of the clitoris, with the moistening of the glandopreputial sulcus as a most likely main function.

Clinical Aspects

Congenital malformations that can be related to the glandopreputial sulcus or its glands appear to be very rare. Phimosis as illustrated by Goldstein (2007) may well be the clinical interpretation of a partially or completely solid lamella, which is shown to be a normal variation in this study.

Congenital clitoral cysts are the most prevalent anomaly, although only three reports were found (Merlob et al., 1978; Teague and Anglo, 1996; Abudaia et al., 1999). Detailed histological information is missing but localization and epithelial lining do fit in well with a deviation caused by the remarkable developmental history of the glandopreputial sulcus. It is conceivable that the clinically irrelevant cystic dilatation of isolated cavities in the solid parts of the sulcus and in adjacent excretory ducts of glandopreputial glands noticed in some older fetuses during this investigation may occasionally expand to a true congenital cyst and may even form the substrate for the rare clitoral cysts, which became manifest during puberty or later in life (Concetti, 1940; Schmidt et al., 1999; Guelinckx and Sinsel, 2002; Linck and Hayes, 2002).

The equally rare pilonidal sinus of the clitoris is considered to be an acquired condition, which is caused by hair shafts "sucked" into the trough-like sulcus which may result in inflammation and destruction of the epithelium progressing into the formation of a fistula (Betson et al., 1962; Patey and Curry, 1962; Radman and Bhagavan, 1972; Werker and Kon, 1990).

The presence of hair shafts in two normal glandopreputial sulci of adult clitorises as observed in this investigation may support this theory. However, such mechanism does not preclude that, theoretically, the glandopreputial sinus epithelium from which apparently eccrine glands originate may rarely give rise to one or more hair follicles. Such a deviation may explain the presence of hair-producing follicles in a "sinus pilonidalis" (Palmer, 1957) or a "dermoid cyst" (Abudaia et al., 1999). No information was found about benign and malignant tumors, which could be related to the glandopreputial glands.

ACKNOWLEDGMENTS

The authors are grateful to Mr. JL Hof and JAS van Ginkel of the Laboratory of Histopathology, Mr. K van der Ven of the Laboratory of Molecular and Immunopathology, and their colleagues for their technical assistance.

LITERATURE CITED

- Abudaia J, Habib Z, Ahmed S. 1999. Dermoid cyst: a rare cause of clitorimegaly. *Pediatr Surg Int* 15:521-522.
- Barreto J, Caballero C, Cubilla A. 1992. Penis. In: Sternberg SS, editor. *Histology for pathologists*. New York: Raven Press. p 721-730.
- Betson JR, Chivelle TL, George RP. 1962. Pilonidal sinus involving the clitoris. *Am J Obstet Gynecol* 84:543-545.
- Cold CJ, Taylor JR. 1999. The prepuce. *Br J Urol* 83 (Suppl 1):34-44.
- Concetti F. 1940. Sulla natura e sull'etiopatogenesi dell cisti del prepuzio clitorideo e del clitoride. *Ann Ostet Ginec* 62:1533-1557.
- Folk EG, Semken HA. 1991. The evolution of sweat glands. *Int J Biometereol* 35:180-186.
- Gairdner D. 1949. The fate of the foreskin. *Br Med J* 2:1433-1437.
- Glenister TW. 1956. A consideration of the processes involved in the development of the prepuce of man. *Br J Urol* 28:243-249.
- Goldstein I. 2007. Urologic management of women with sexual health concerns. In: Wein AJ, editor. *Campbell-Walsh urology*. Philadelphia: WB Saunders Company. p 863-889.
- Guelinckx PJ, Sinsel NK. 2002. An unusual case of clitoral enlargement: its differential diagnosis and surgical management. *Acta Chir Belg* 102:192-195.
- Hamilton WJ, Mossman HW. 1972. *Embryology. Prenatal development of form and function*. Cambridge: The Williams and Wilkins Company, W Heffer and Sons. p 417-418.
- Hart DB. 1908. On the role of the developing epidermis in forming sheaths and lumina to organs, illustrated specially in the development of the prepuce and urethra. *J Anat (Lond)* 42:50-56.
- Herzog F. 1904. Beiträge zur Entwicklungsgeschichte und Histologie der männlichen Harnröhre. *Arch f Mikrosk Anat* 63:710-714.
- Hunter RH. 1935. Notes on the development of the prepuce. *J Anat (Lond)* 70:68-75.
- Hyman AB, Brownstein MH. 1969. Tyson's "glands". Ectopic sebaceous glands and papillomatosis penis. *Arch Dermatol* 99:31-36.
- Johnson FP. 1920. The later development of the urethra in the male. *J Urol* 4:447-501.
- Larsen WJ. 1997. *Human embryology*. New York: Churchill Livingstone. p 256-274.
- Linck D, Hayes MF. 2002. Clitoral cyst as a cause of ambiguous genitalia. *Obstet Gynecol* 99:963-966.
- Maroun LL, Graem N. 2005. Autopsy standards of body parameters and fresh organ weights in nonmacerated and macerated human fetuses. *Pediatr Dev Pathol* 8:204-217.
- McLean JM. 1999. Anatomy and physiology of the vulva. In: Ridley CM, McLean JM, editors. *The vulva*. 2nd ed. Oxford: Blackwell Science. p 37-63.

- Merlob P, Bahari C, Liban E, Reisner H. 1978. Cysts of the female external genitalia in the newborn infant. *Am J Obstet Gynecol* 132:607–610.
- Moore KL, Dalley AF, II. 1996. Clinically oriented anatomy. Philadelphia: Lippincott, Williams and Wilkins. p 389–415.
- O'Connell HE, Sanjeevan KV, Hutson JM. 2005. Anatomy of the clitoris. *J Urol* 174:1189–1195.
- O'Rahilly R, Müller F. 2001. Human embryology and teratology. 3rd ed. New York: Wiley-Liss. p 336–337.
- Oster J. 1968. Further fate of the foreskin. Incidence of preputial adhesions, phimosis, and smegma among Danish schoolboys. *Arch Dis Childh* 43:200–203.
- Palmer E. 1957. Pilonidal cyst of the clitoris. *Am J Surg* 93:133–136.
- Parkash S, Jeyakumar S, Subramanyan K, Chaudhuri S. 1973. Human subpreputial collection: its nature and formation. *J Urol* 110:211–212.
- Patey DH, Curry RC. 1962. Pilonidal sinus presenting in the suprapubic region of a woman. *Lancet* 1:620–621.
- Radman HM, Bhagavan BS. 1972. Pilonidal disease of the female genitals. *Am J Obstet Gynecol* 114:271–272.
- Reznik G, Ward JM. 1981. Morphology of neoplastic lesions in the clitoral and preputial gland of the F334 Rat. *Cancer Res Clin Oncol* 101:249–263.
- Rhodin JAG. 1974. Histology. A text and atlas. London, Toronto: Oxford University Press. p 703–749.
- Saalfeld E. 1899. Ueber die Tyson'schen Drüsen. *Arch Mikr Anat* 53:212–218.
- Schmidt A, Lang U, Kiess W. 1999. Epidermal cyst of the clitoris: a rare cause of clitorimegaly. *Eur J Obstet Gynecol Reprod Biol* 87:163–165.
- Schweigger-Seidel F. 1866. Zur Entwicklung des Preputium. *Arch Pathol Anat Physiol Klin Med (Virchows Archiv)* 37:219–228.
- Standring S. 2007. Gray's anatomy. The anatomical base of clinical practice. London: Churchill Livingstone. p 1280.
- Taylor JR, Lockwood AP, Taylor AJ. 1996. The prepuce: specialized mucosa of the penis and its loss to circumcision. *Br J Urol* 77:291–295.
- Teague JL, Anglo L. 1996. Clitoral cyst: an unusual cause of clitorimegaly. *J Urol* 156:2057.
- Thompson RS, Hess DL, Binkerd PE, Hendrickx AG. 1981. The effects of prenatal diethylstilbestrol exposure on the genitalia of puberal *Macaca mulatta*. II. Male offspring. *J Reprod Med* 26:309–316.
- Van der Putte SCJ. 1991. Anogenital "sweat" glands. Histology and pathology of a gland that may mimic mammary glands. *Am J Dermatopathol* 13:557–567.
- Van der Putte SCJ. 1994. Apoeccrine glands in nevus sebaceous. *Am J Dermatopathol* 16:23–30.
- Van der Putte SCJ. 2005. The development of the perineum in the human. *Adv Anat Embryol Cell Biol* 177:1–135.
- Velazquez EF, Gold CJ, Baretto JE, Cubilla AL. 2007. Penis. In: Mills SE, editor. Histology for pathologists. Philadelphia: Lippincott, Williams and Wilkins. p 965–980.
- Werker PMN, Kon M. 1990. A pilonidal sinus of the clitoris? *Ann Plast Surg* 25:63–64.
- Wilkinson EJ, Hardt NS. 2007. Vulva. In: Mills SE, editor. Histology for pathologists. Philadelphia: Lippincott, Williams and Wilkins. p 983–997.