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Chapter 2

The somatic and autonomic innervation of the clitoris; preliminary evidence of sexual dysfunction after minimal invasive slings

(Submitted)

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The Netherlands

Introduction

Within a decade, the midurethral (vaginal) slings became by far the most popular surgical treatments for stress urinary incontinence (SUI), with more than one million women treated (1). Despite the numerous studies on objective and subjective outcomes of this minimal invasive procedure, very few studies have addressed the impact of vaginal sling procedures on sexuality. Small series evaluating the sexual well-being before and after the tension-free vaginal tape (TVT), the transobturator in-out (TVT-O) and/or the transobturator out-in (TOT) procedures show conflicting results. Of these studies, some suggest deterioration (2-7) of sexual function, some improvement (2-4;8-11), whereas others were equivocal (12-16). A prognostic factor in the improvement of the sexual functioning of these patients is the cure of incontinence during intercourse (17;18). Negative effect on sexuality is hypothesized to be related to the implanted material because of damage to important vascular and/or neural genital structures (2-7).

The clitoris plays an important role in achieving female orgasm by sexual stimuli (19). It is innervated by the dorsal nerves of the clitoris (DNC). These peripheral sensory afferents of the clitoris originate from the pudendal nerve. The clitoris is also innervated with fibres coming from the autonomic pelvic plexus (also known as the inferior hypogastric plexus): the cavernous nerves of the clitoris. Clitoral and labial swelling during sexual arousal, is associated with parasympathetic vasodilator mechanisms, among which nitric oxide (NO) appears to be a primary neurotransmitter contributing to the mediation of this function (20;21). NO control of vasodilatation and neuronal signaling between the cavernous nerves and the dorsal nerve of the clitoris contribute to the engorgement and subsidence of clitoral tissue. This supports the initiation of sexual arousal by tactile stimuli of the clitoris (22). Therefore, in theory, injury to the clitoris and/or its innervating nerves, both somatic and autonomic, could lead to altered sexual function.

To investigate the anatomical relation of vaginal slings to important neural genital structures, basic knowledge about and detailed descriptions of the neuro-anatomy of the clitoris are needed. The clitoris is a structure of which few diagrams and minimal descriptions are provided, potentially impacting its preservation during surgery. Research has demonstrated the integral relationship between the clitoris, and the distal urethra and vagina (23-25). O'Connell et al. provided a major contribution to the research on the anatomy of the clitoris (23;26). They found the clitoris to be intimately related to the distal urethra, which lead them to suggest that the role of the urethra in sexual function is related to the position of the surrounding erectile tissue rather than the urethral sphincter (23). The DNC was described in detail, the autonomic nerves, however, were poorly addressed.

Disruption of the somatic innervation of the clitoris can lead to a diminished sensibility of the clitoris, thereby affecting sexual arousal due to the absence of tactile stimuli. The DNC is located along the medial aspect of the ischiopubic ramus (IPR) where it runs along the pubic bone in a sulcus described as the sulcus nervi dorsalis clitoridis (27). Risk of injury to the DNC along the IPR had been suggested by Delorme with the medial to lateral passage of the needle which is used for placing vaginal slings and may alter postoperative sexual function such as arousal, orgasmic function or pain (28). This possible risk has been illustrated by Lowenstein, showing the topographic relation of mid-urethral sling for stress urinary incontinence to critical female genital structures (29). In a cadaveric study, performed by Ahtari et al., the potential risks of three vaginal slings to the DNC were evaluated. Distances of a TVT, transobturator in-out (TVT-O) and transobturator out-in (Monarc) to the DNC were similar (11-12 mm) (30). Given the outside-in course, the Monarc was claimed to (theoretically) be the safest device. Another cadaveric study found similar results, although they only documented the course of the DNC from the piercing of the perineal membrane to its terminal branching and not its course along the IPR (31).

The cavernous nerves of the clitoris are involved in the neural control of vasocongestion and, consequently, the lubrication-swelling response. Disruption of these nerves could lead to altered vascular function during sexual arousal and possibly disordered orgasm. However, although important for normal sexual function, in afore mentioned studies no attention was paid to possible disruption of the cavernous nerves of the clitoris (29-31).

The aim of this study was to reinvestigate the neuro-anatomy of the clitoris by performing dissection in an adult female pelvis and by using (immuno)histochemical and three-dimensional (3D) reconstruction techniques on a female fetus. In this study we focus on 1) the autonomic innervation of the clitoris, 2) the course of the DNC, 3) to investigate the anatomical sites of potential nerve damage during vaginal sling surgery for SUI.

During dissection, it is difficult to recognize and dissect small nerve fibers such as those from the pelvic plexus. Therefore, there is a high risk of artifacts because tissue strings may be mistaken for small nerves. The study of serially sectioned human fetuses has an advantage over conventional cadaver dissections, because of immunohistochemical staining; structures of interest are easily recognized (32-35).

Dimensions may change but the topographic relationships of tissues persist throughout fetal development which makes the study of fetuses excellent for describing anatomy (32;36). Furthermore, 3D reconstructions of the innervation of the clitoris can be prepared to deduce its anatomical relationship to other pelvic structures and provide an insightful illustration which can be used by pelvic surgeons.

Methods

Fetal 3D-reconstruction

Fetal pelvises from the collections in the departments of Anatomy and Embryology at the Leiden University Medical Centre and at the Amsterdam Medical Centre were studied. Eleven paraffin embedded fetuses (all female), ranging from 10 to 27 weeks of gestation; 6-26 mm crown-rump length (CRL) were serially sectioned. Six were stained with haematoxylin and eosin, three with haematoxylin-azophloxine and two with both haematoxylin and neurofilament (35). The fetal tissue was fixed in 4% paraformaldehyde, embedded in paraffin and transversely sectioned into serial sections of 10 µm. Analysis of the transverse sections was performed from superior part of the pubic arch to just below Alcock's canal. Digital images were taken of the serial sections, photographing every second section. These images were used to prepare three dimensional reconstructions with the Amira software package (v3.0, Visage Imaging GmbH, Fürth, Germany). Unfortunately, in the transversed sections, the perineal membrane (or urogenital diaphragm) was not recognized and thus not reconstructed.

Cadaver study

The pelvis of a female cadaver without signs of any pelvic surgery was used for this study. Usually, the age of death of these women is older than 70 years of age. Preservation of the cadaver was performed by injection of the embalming fluid AnubiFiX™ into the femoral artery. Due to this fixation process, the cadaver remains flexible which enables a natural dissection and allows surgical procedures. The cadaver was donated to the university for medical research and hence does not require separate ethics approval to be dissected. A trained urologist (H.W.E.) performed parts of both procedures (TVT and TVT-O) on one cadaver, one sling placement on each side. Both procedures were performed exactly similar as they are normally performed on patients.

The pelvis was sectioned through the midline from the pubic symphysis anteriorly to the sacrum posteriorly. The urethra was sectioned along its full length in the midline. The clitoris and its somatic and autonomic nerves were dissected and the shortest distance between the needle/sling and the nerves were measured and various stages of the dissection were recorded photographically.

An interactive three-dimensional reconstruction is attached to this article, available as a supplementary file showing the pelvis, the pelvic organs, the clitoris, its autonomic (cavernous) nerves and the branches of the pudendal nerve, the obturator nerve and the levator ani muscle of a female foetus (12 wk of gestation; same foetus as shown in Figures 1A-H).

Results

Three-dimensional reconstruction

Anatomy of the clitoris

The clitoris is a multiplanar structure positioned deep to the labia minora, labial fat and vasculature, bulbospongiosus and ischiocavernosus muscles, inferior to the pubic arch and symphysis. It has a broad attachment to the pubic arch, and via extensive supporting tissue to the mons pubis (the adipose tissue lying above the pubic bone of adult females, anterior to the symphysis pubis) and labia. The clitoris consists of a tip, also known as the glans clitoridis, the erectile body and the crura (or corpora cavernosa) (Fig. 1A). The clitoris has a close relationship to the distal urethra and vagina (Fig. 1B). Because this study involved human fetuses, aged 10-27 weeks of gestation, these two structures are still partly merged.

Dorsal nerve of the clitoris

The three-dimensional reconstruction illustrates the course of the dorsal clitoral nerves. They originate from the pudendal nerve in the Alcock's canal immediate medial to the pubic bone and lateral to the rectum, forming a bundle that fannes out laterally, passing the levator ani muscle and ascending to the clitoral bodies (Fig. 1C, D, E and F). The 3-D images also revealed that both dorsal nerves of the clitoris run medial and close to the puboischial ramus (Fig. 1E). Furthermore, the close relationship of the clitoral crura to the clitoral dorsal nerve was notable. They further traverse distally along the clitoral crura and run posterior to the body of the clitoris before hooking over it in anterolateral direction. There, they medially cross the cavernous nerves and pass over the clitoral body (Fig. 1E and G). As they pass over the body, they merge with the autonomic nerves. After passing over the body, the merged somatic and autonomic nerves pass further caudal and anterior over the clitoral body to the glans clitoridis (Fig. 1E and H).

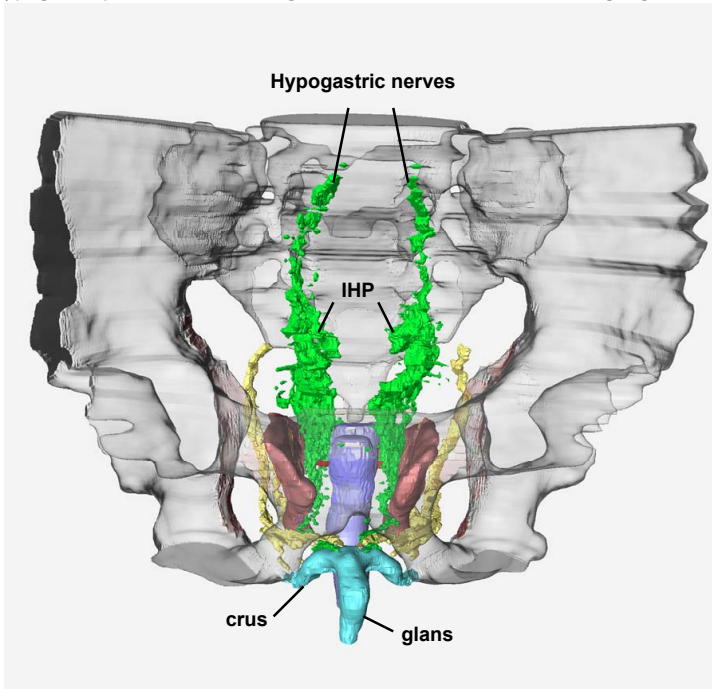
Autonomic nerves of the pelvic plexus

From the superior hypogastric plexuses, two nerves (the hypogastric nerves) run bilaterally into the small pelvis, to be joined by the pelvic splanchnic nerves coming from sacral roots S2 to S4, to form the pelvic plexus, also known as the inferior hypogastric plexus (IHP) on both sides of pelvic organs (Fig. 1A). The IHP showed to be a triangularly shaped plexus in a sagittally plane. It is in close contact with its target organs as a flat meshed plaque of nerve tissue, stretching from anterolateral to the rectum, passing the cervix and vagina laterally and extending from the lateral vaginal wall to the base of the bladder and lateral to the urethra with branches to the clitoris.

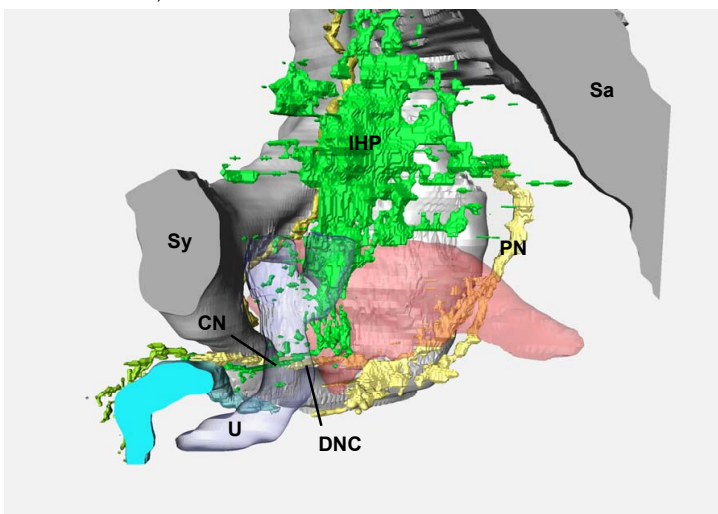
Its nerves extend onto the lateral walls of the proximal and mid-vagina, where they form a dense network. These nerves travel superior to cover the proximal anterior vaginal wall, where

Figure 1

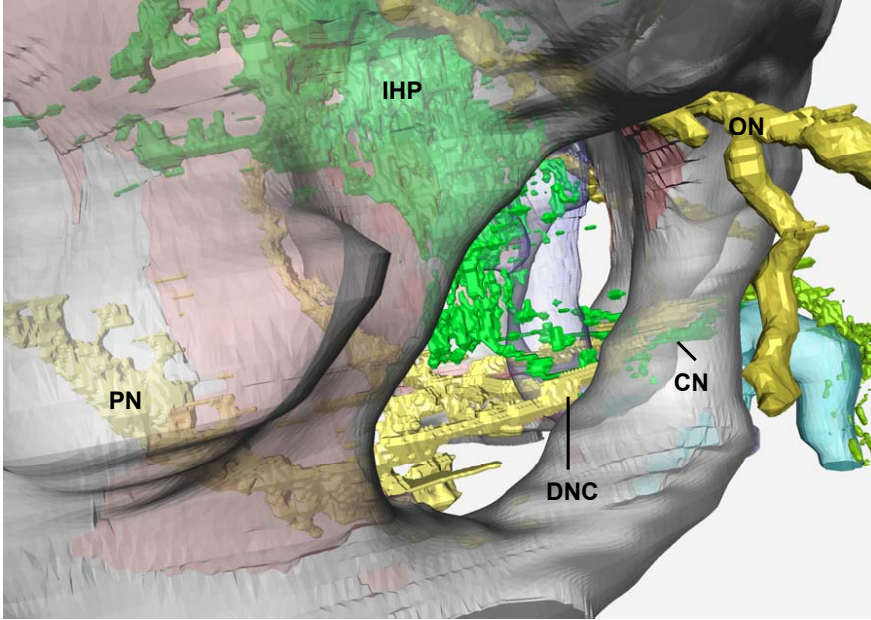
- A. Anterior view (3D) of the clitoris (blue), the pelvis (grey), the urethra/vagina (U, purple), the pudendal nerves (yellow), the hypogastric (autonomic) nerves (green) travelling into the pelvis and forming the inferior hypogastric plexus (IHP), the merged autonomic and dorsal nerves (light green).



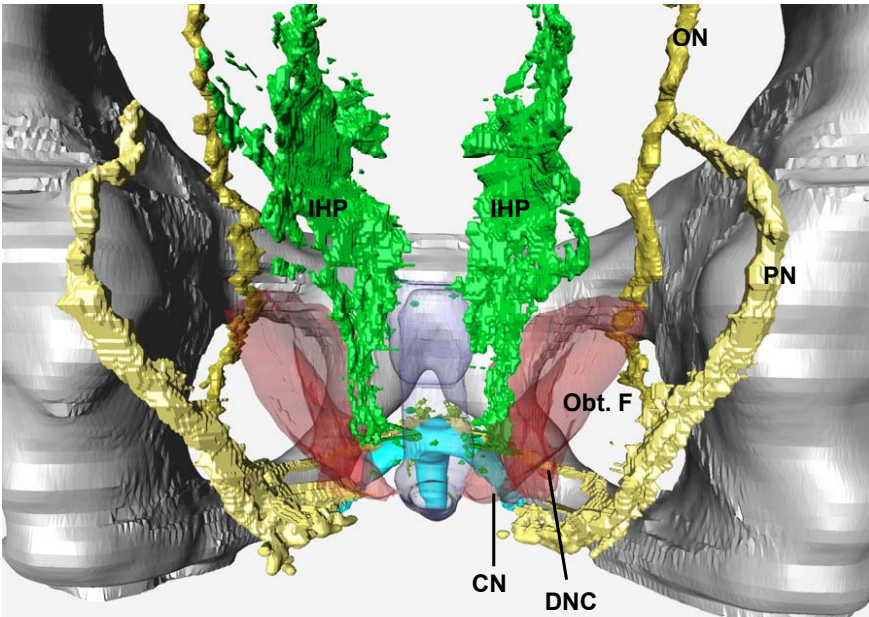
- B. Lateral view (3D) of the midsagittal cut pelvis with the symphysis (Sy) centered (grey), the levator ani muscle (red) the clitoris (blue), the autonomic (green) and somatic (yellow) nerves and the urethra/vagina (U, purple). IHP: inferior hypogastric plexus, PN: pudendal nerve, CN: cavernous nerve, DNC: dorsal nerve of the clitoris, Sa: sacrum.



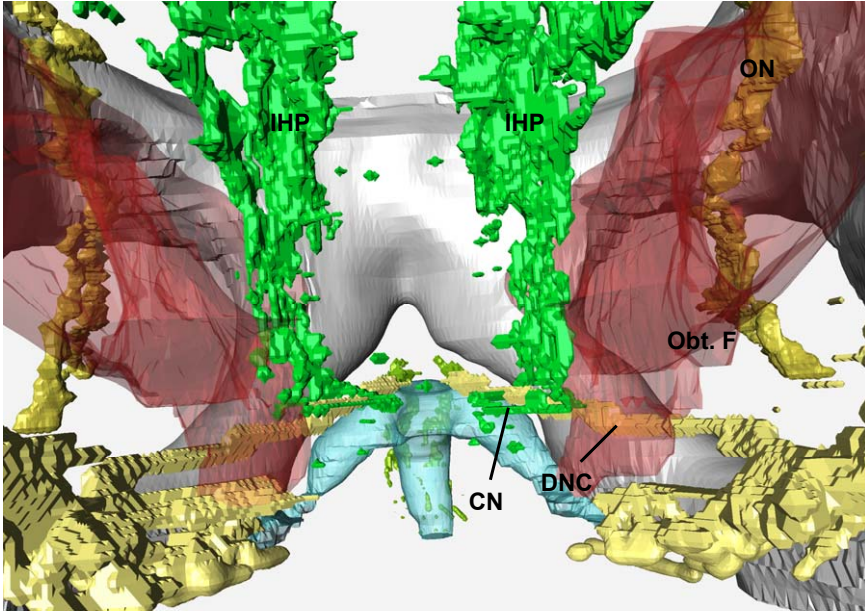
- C. Lateral view (3D) of the pelvis (grey) on the obturator foramen, the clitoris (blue), the autonomic (green) and somatic (yellow) nerves and the urethra/vagina (purple). IHP; inferior hypogastric plexus, PN; pudendal nerve, CN; cavernous nerve, DNC; dorsal nerve of the clitoris.



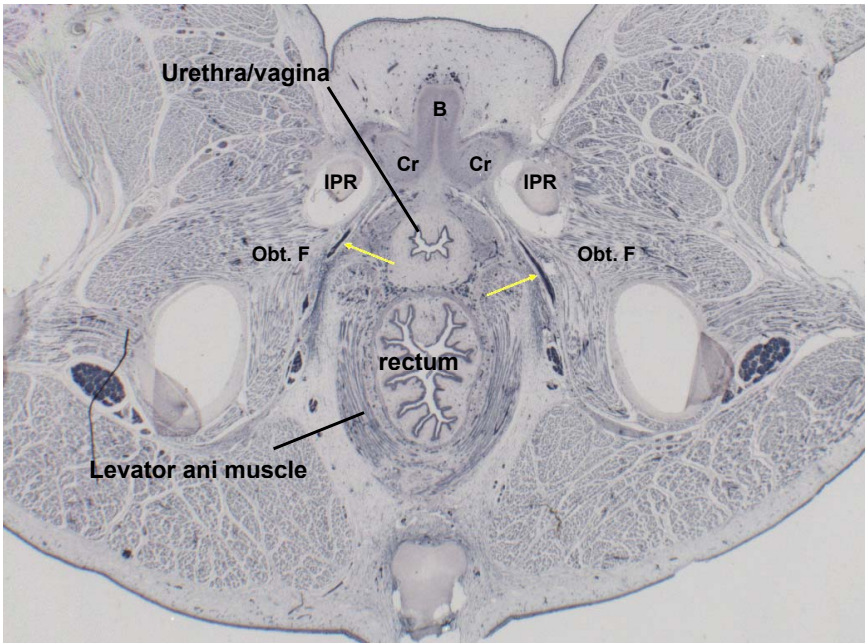
- D. Posterior view (3D) of the pelvis with the pubic bone centered (grey), the clitoris (blue), the autonomic (green) and somatic (yellow) nerves and the urethra/vagina (purple). IHP; inferior hypogastric plexus, ON; obturator nerve, PN; pudendal nerve, Obt. F; obturator foramen, CN; cavernous nerve, DNC; dorsal nerve of the clitoris



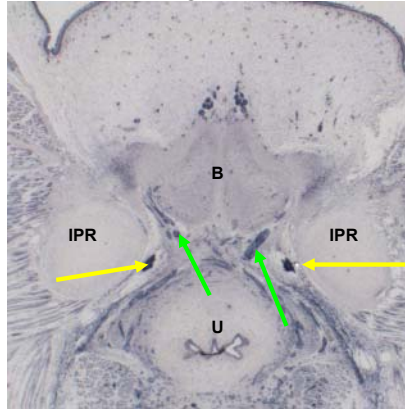
- E. Anterior and slightly lateral view (3D), of the pelvis (grey) the obturator foramen (Obt. F), the obturator nerve (ON), the clitoris (purple), the dorsal nerve of the clitoris (DNC, yellow) and the cavernous nerves (green, CN) coming from the inferior hypogastric plexus (IHP). ON; obturator nerve,



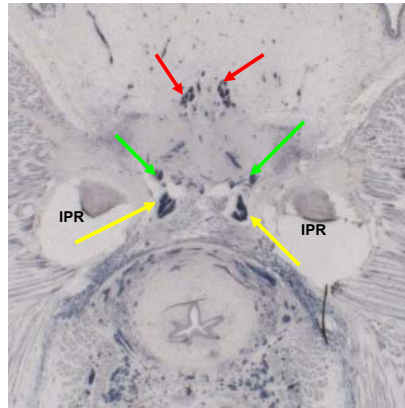
- F. Stained section, showing the body of the clitoris (B) with its crura (Cr) close to the ramus inferior of the pubic bone (IPR), and the dorsal clitoral nerves (yellow arrows) passing along the obturator foramen (Obt. F.).



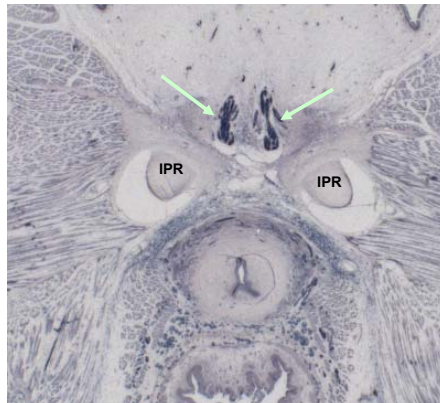
- G. Close-up of a stained section. The close relationship of the ramus inferior of the pubic bone (IPR) to the clitoral dorsal nerve (yellow arrows) is notable and it shows that the branches of the cavernous nerves of the clitoris pass medial to the dorsal nerves (green arrows). B: clitoral body, U: urethra/vagina.



- H. Stained section, showing both cavernous nerves (green arrows) and the dorsal nerves of the clitoris (yellow arrows) hooking over the clitoral body and travelling further caudally alongside and into the clitoral body and glans (red arrows).



- I. Stained section, the autonomic nerves merge with the branches of the dorsal nerves as they pass over the clitoral body (light green arrows).



they form the cavernous nerves at the 2 and 10 o'clock positions along the urethra. (Fig. 1B and F)

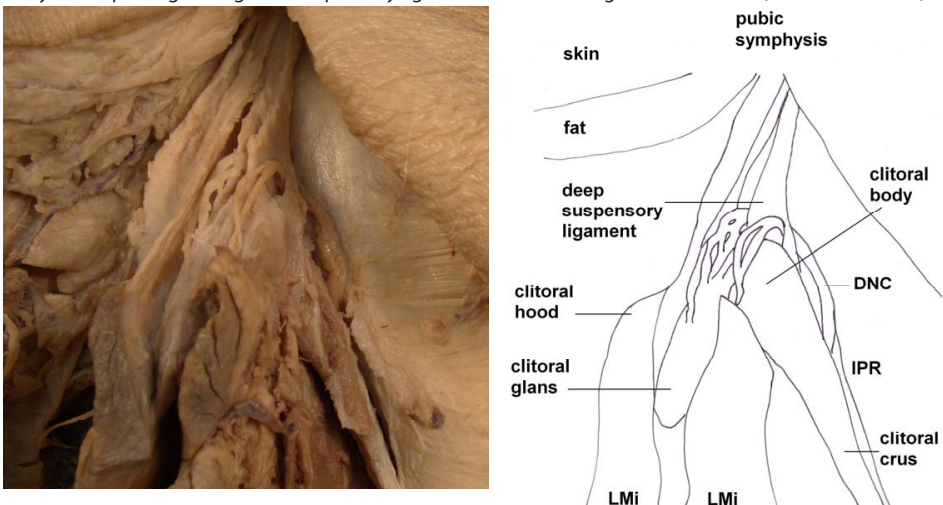
There, they travel further caudal to the clitoral bodies crossing the dorsal clitoral nerve medially (Fig. 1C). The nerve bundles then travel alongside the branches of the dorsal nerve passing over the clitoral body. After passing over the clitoral body, these autonomic nerves merge with the branches of the dorsal nerve and travel further caudally alongside and into the clitoral body and glans (Fig. 1E, H and I).

Dissection

Anatomy of the clitoris

The initially almost straight clitoral crura commence proximally running along the puboischial ramus and join distally under the pubic symphysis as a single clitoral body that projects anteriorly into the glans. There it projects into the fat of the mons pubis. They are situated between the clitoral crura and form a midline core in the triangular shaped clitoral structure. Dissection shows that the apex of this triangular structure is the most superior point of the clitoral body, where it attaches to the under surface of the pubic symphysis by the deep suspensory ligament (Fig. 2). As the clitoral body projects from the bone into the mons pubic fat, it descends and folds back on itself in a boomerang-like shape in a dorsalcaudalward direction forming the glans clitoridis. The glans of the clitoris is a relatively small nodular structure that becomes partially covered by the glando-preputial lamella and prepuce (or clitoral hood).

Figure 2. Anterior view of the dissection clitoris. The mons pubis has been opened to show the deep suspensory ligament. Furthermore, the clitoral crus, body and glans are shown with the dorsal nerve of the clitoris (DNC) ascending along the inferior ramus of the pubic bone (IPR), hooking over the clitoral body whilst passing through the suspensory ligament and branching into the clitoris. (LMi: labia minora)



Dorsal nerve of the clitoris

The course of the pudendal nerve (PN) around the ischial spine was approached posteriorly by removal of the skin and superficial fascia between the anterior inferior iliac spine, the ischial tuberosity and the posterior superior iliac spine. The gluteus maximus muscle was dissected from its origin to expose underlying structures. The sacrotuberous ligament was identified and transected to identify the PN subjacent to the sacrotuberous ligament and around the ischial spine of the pelvis. The entrance of PN into Alcock's canal was identified. Alcock's canal was then unroofed which revealed the three main branches of the PN, namely, the inferior rectal nerve, the perineal nerve and the dorsal nerve of the clitoris (DNC). The DNC was then followed until its termination in the clitoris. The DNC travels along the perineal membrane (or urogenital diaphragm) and runs inferior to the inferior pubic ramus. By opening the perineal membrane the TVT-O tape was exposed. (Figure 3 A-B) The distance of the TVT-O to the DNC was 2 mm and they were separated by the perineal membrane. (Figure 3)

Figure 4 is a schematic lateral view on a midsagittal sectioned right pelvis showing the course of the DNC from the PN to the clitoris lateral from the levator ani muscle.

Figure 3 A. Frontal view of the right female genital and perineal area. In order to expose the DNC the skin was opened between the right labia majora and minora. To show the route of the TVT-O sling, the perineal membrane was opened subsequently. DNC; dorsal nerve of the clitoris, TVT-O; the tensionfree vaginal tape obturator; IPR; inferior pubic ramus, LMa; labia majora, LMi; labia minora.

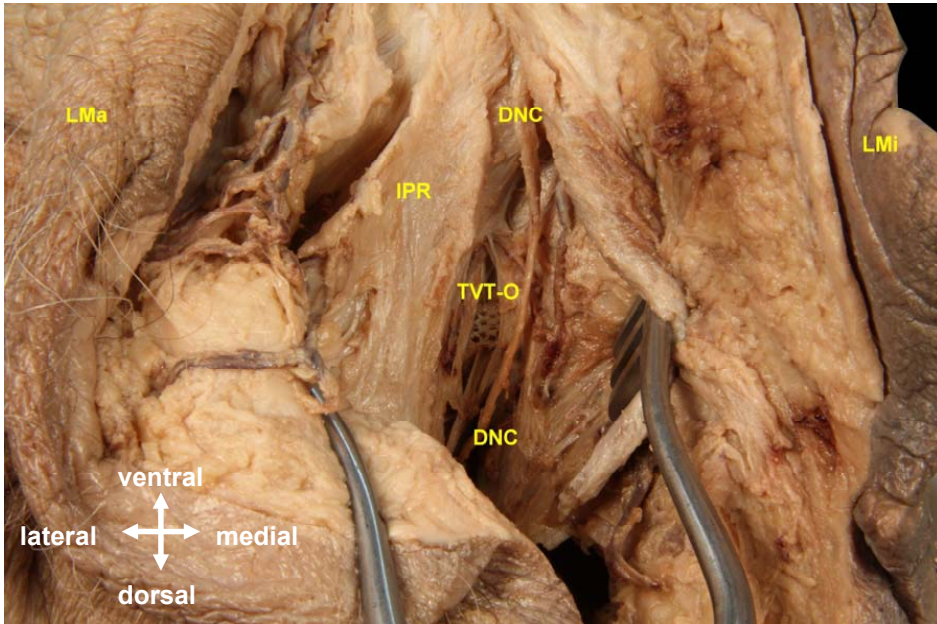


Figure 3 B. Close-up of figure 3A. The bulbospongiosus muscle and clitoral crus were moved medially to show the course of the DNC. To show the route of the TVT-O sling, the perineal membrane was opened. DNC; dorsal nerve of the clitoris, TVT-O; the tensionfree vaginal tape obturator; IPR; inferior pubic ramus.

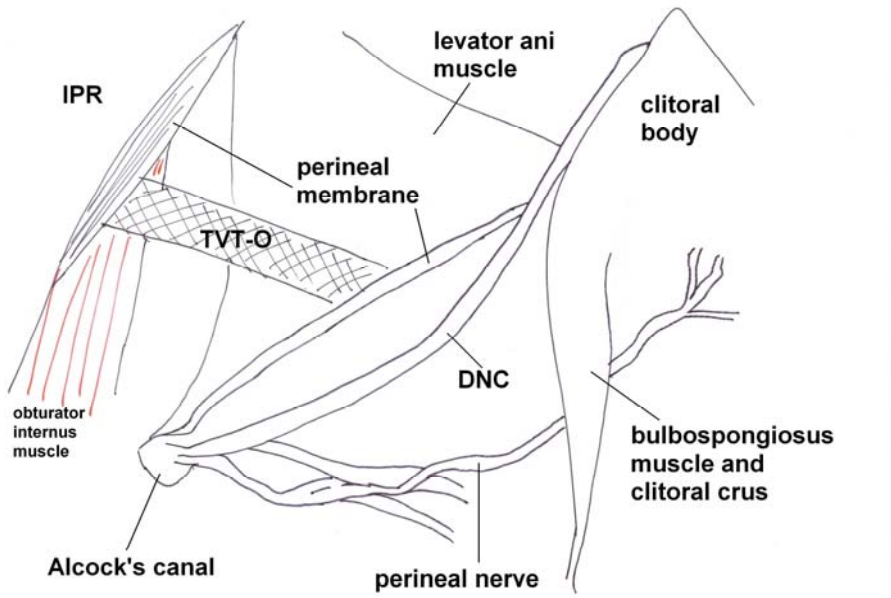
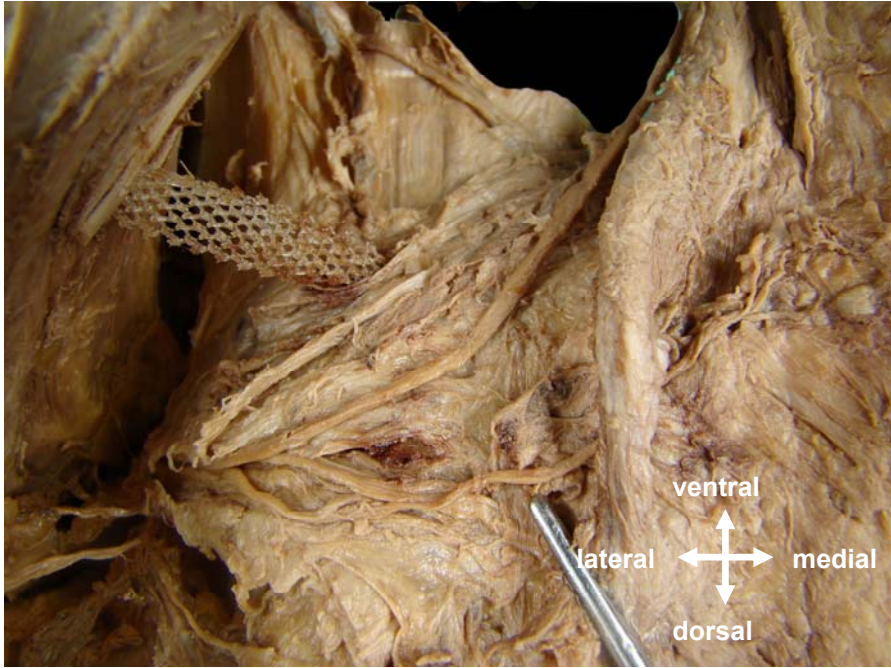
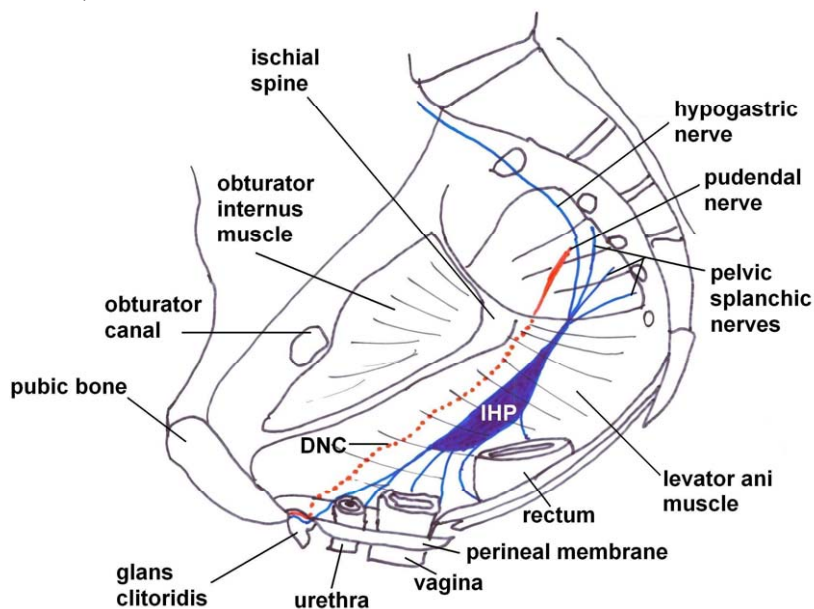


Figure 4 A schematic lateral view on a midsagittal sectioned right pelvis showing the course of the autonomic nerves from the hypogastric nerve to the target organs and the course of the DNC from the pudendal nerve, lateral from the levator ani muscle to the clitoris.



Autonomic nerves

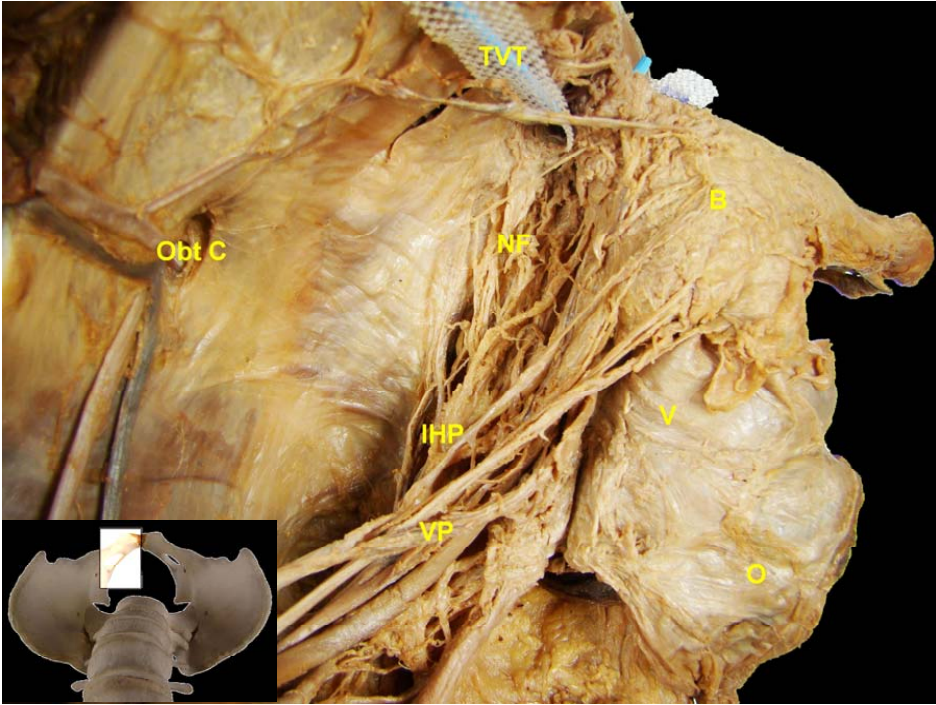
The superior hypogastric plexus was identified inferior to the bifurcation of the aorta. The proximal hypogastric nerves were identified in the subperitoneal layer (between the peritoneum and the endopelvic fascia) and followed alongside the ureter into the small pelvis to the inferior hypogastric plexus (IHP). Figure 4 is a schematic lateral view on a midsagittal sectioned right pelvis showing the course of the autonomic nerves from the hypogastric nerve to the target organs.

The IHP, a flat meshed plaque of nerves was dissected. Its branches, which follow the connective tissue plane within the small pelvis which supports the uterine cervix, vagina and bladder, were identified and dissected into their target organs. Special attention was paid to the branches passing along the urethra and innervating the clitoris. The autonomic nerves, running from the IHP, were found to be pierced by the TVT-needle. (Figure 5)

Discussion

This study describes the neuro-anatomy of the clitoris; its somatic and autonomic pathways. Previous studies on the innervation of the clitoris were mainly focused on the dorsal nerve of the clitoris, paying no attention to the cavernous nerves coming from the pelvic plexus, which play an important role in female sexual function (23;25-27;31;37-39). The cavernous nerves are

Figure 5: View from above (abdominal view) into left female half pelvis. In order to expose the area lateral to the vagina, the uterus was removed from the level of the cervix and the bladder and vagina had been retracted medially and anteriorly. The peritoneum and part of the fascia have been removed. IHP; inferior hypogastric plexus; B; bladder; V; vagina; O; Ovarian, TVT; the TVT tape; NF; nerve fibres from the IHP to the vagina and clitoris, Obt C; obturator canal.



involved in the neural control of vasocongestion and, consequently, the lubrication-swelling response. Disruption of these nerves could lead to altered vascular function during sexual arousal and possibly disordered orgasm.

In 1982, Walsh and Donker described the anatomic location of the pelvic plexus (or IHP) in men and the nerves innervating the corpora cavernosa (40). These pioneering observations and descriptions of the anatomical basis for radical prostatectomy fostered resurgence in the use of surgery as treatment for localized prostate cancer and led urologic surgeons to refine a nerve-sparing technique within the following two decades. Only in the recent years, attention is paid to the IHP in females and nerve-sparing techniques are being developed in surgery for cervical cancer (41-43).

Although the IHP has been described in females, little attention has been paid to the cavernous nerves coming from this IHP and their anatomical relation to other pelvic structures. Yucel et al reported that the cavernous nerve supply the female urethral sphincter complex and clitoris (25). The branches of the cavernous nerve were described and, as in our study, noted to join the clitoral dorsal nerves. The cavernous nerves have also been described in mice, using immunostaining to show communicating nerves between the cavernous nerve and the dorsal

nerve of the clitoris which supports the initiation of sexual arousal by tactile stimuli and following clitoral swelling (22). This study underlines the importance of both somatic and autonomic innervation of the clitoris in normal female sexual function.

In the study performed by O'Connell et al. the course of the dorsal nerve was described but the cavernous nerves were poorly addressed (26). Other important studies on the neuro-anatomy of the clitoris focused mainly on the DNC (26;27;39;44). Similar to our findings, the DNC is described to originate from the pudendal nerve and to ascend along the ischiopubic rami.

Vaginal sling procedures for stress urinary incontinence have been developed in the nineties by Ulmsten (45). After research showed the procedure to be safe and effective, the TVT and derived procedures became a well established surgical procedure for the treatment of female urinary stress incontinence. Especially in these early years, no attention was paid to the topographic relation to important genital structures. Only in recent years, the possible risk of nerve damage during vaginal sling procedures, especially the obturator procedures, has been suggested (29).

The aim of this study was not only to describe the neuro-anatomy of the clitoris, but also its relation to surrounding structures which are anatomical landmarks in vaginal tape procedures for SUI. When performing vaginal sling surgeries, a sagittal incision is made within the anterior vaginal wall mucosa about 1 centimeter from the urethral meatus and the vaginal mucosa is dissected of the mid-urethra. When performing the TVT procedure, a tape is placed (blindly) behind the pubic symphysis using trocars attached to the tape when performing the TVT procedure (46).

When performing the TOT, the 'outside-in' procedure, a similar midline incision is made in the anterior vaginal wall between the mid-urethra and bladder neck, enough to introduce the index finger. Dissection is carried out laterally to the level of the vaginal sulcus without penetration of endopelvic fascia. The IPR of the pubic bone and the obturator foramen are located manually, and the medial edge of the ramus is pinched between thumb and index finger. The skin puncture is made at the level of the clitoris right above the pinching thumb. The curved sling passer is guided from the thumb to the index finger and then rotated and delivered to the vaginal incision with the tip on the index finger. The arm of the tape is hooked to the tip of the passer and brought out to the skin (46).

When performing the TVT-O, the 'outside-in', a similar midline incision is made and peri-urethral tunnels are developed bilaterally. Unlike the TOT, where the dissection stops at the IPR, with the TVT-O the obturator membrane is perforated with the tip of the scissors. A winged metal trocar, which is designed to help guide the tape around the IPR, is inserted into the peri-urethral tunnel and the tip is pushed just beyond the perforated membrane. The trocar is then rotated around the IPR to exit out the skin through stab incisions located 2 cm above a horizontal line at the level of the urethral meatus and 2 cm outside the thigh folds. The same procedure is performed on the other side and the sling is tensioned and the procedure completed (46).

As described, during these procedures, the part of the mid-urethra along the anterior vaginal wall is an important surgical site; here the first incision is made. This study illustrates that the urethra is surrounded by autonomic nerves coming from the IHP. Not only do they travel to and innervate the urethra, the cavernous nerves travel from the vaginal nervous plexus occupying the 2 and 10 o'clock positions on the anterolateral vagina and they travel at the 5 and 7 o'clock positions along the urethra. It is therefore, possible that during the mid-urethral incision in all vaginal procedures and the para-urethral tunneling during the obturator procedures, the cavernous nerves are disrupted. The results of the cadaveric part of this study showed that indeed the cavernous nerves are being pierced during the TVT procedure.

The IPR plays an important role in the obturator procedures; the tapes are placed around this bony structure. Furthermore, dissection is performed paraurethral to the IPR, where during the inside-out technique the obturator membrane is perforated with scissors. Because the DNC travels along the medial side of the IPR, especially at the level of the urethral meatus, it is at risk to be damaged during obturator procedures, both outside-in and inside-out. The results of the dissections confirmed that the DNC is at risk for nerve damage during the TVT-O procedure.

Achtari et al. have also dissected female cadavers and measured the distance of the DNC to the TVT, TVT-O and Monarc. The results showed a distance varying from 19-40 mm, the TVT-O being the closest to the DNC (30). A similar study in fresh cadavers measured a distance between the DNC and the TOT of 3-14 mm. Despite this small distance, they concluded the TOT to be a safe procedure. This study was however biased because straight needles were used to mimic the course of the TOT, instead of curving trocars (31). As the results of this study shows, it is possible that during dissection to the foramen and during placement of the tape, the DNC is injured.

This study is the first to illustrate, in detail, both somatic and autonomic pathways of the clitoris and thereby, significant progress has been made in the field of female sexual anatomy and its representation. This may facilitate further research in the related fields of female sexual health and education and can be used by surgeons in the field of urogynecology. Furthermore, the topographic relation of vaginal slings to the important critical female genital structure, the clitoris, has been illustrated and described for the first time. Future (clinical) research should be performed to confirm these results and to investigate the consequences of injury to the clitoral nerves on the clitoral sexual response and female sexual functioning.

Conclusion

This study shows and described the somatic and autonomic innervation of the clitoris in detail including their relation to important surrounding structures.

Furthermore, the relation of vaginal sling procedures for SUI to the clitoris and its innervation has been evaluated. Given the course of the dorsal clitoral nerve; inferior to the inferior pubic

ramus, it is at risk for iatrogenic injury after placement of transobturator tape. Furthermore, the autonomic innervation of the vaginal wall is disrupted by the tensionfree vaginal tape procedure, which could lead to altered lubrication-swelling response. When the “inside-out” technique is used, the introducer can come into contact with the dorsal nerve of the clitoris because the introducer passes through the obturator foramen close to the ischio-pubic ramus.

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