Verification of the anatomy and newly discovered histology of the G-spot complex*

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Objectives To expand the anatomical investigations of the G-spot and to assess the G-spot’s characteristic histological and immunohistochemical features.

Design An observational study.

Setting International multicentre.

Population Eight consecutive fresh human female cadavers.

Methods Anterior vaginal wall dissections were executed and G-spot microdissections were performed. All specimens were stained with haematoxylin and eosin (H&E). The tissues of two women were selected at random for immunohistochemical staining.

Main outcome measures The primary outcome measure was to document the anatomy of the G-spot. The secondary outcome measures were to identify the histology of the G-spot and to determine whether histological samples stained with H&E are sufficient to identify the G-spot.

Results The anatomical existence of the G-spot was identified in all women and was in a diagonal plane. In seven (87.5%) and one (12.5%) of the women the G-spot complex was found on the left or right side, respectively. The G-spot was intimately fused with vessels, creating a complex. A large tangled vein-like vascular structure resembled an arteriovenous malformation and there were a few smaller feeding arteries. A band-like structure protruded from the tail of the G-spot. The size of the G-spot varied. Histologically, the G-spot was determined as a neurovascular complex structure. The neural component contained abundant peripheral nerve bundles and a nerve ganglion. The vascular component comprised large vein-like vessels and smaller feeding arteries. Circular and longitudinal muscles covered the G-complex.

Conclusion The anatomy of the G-spot complex was confirmed. The histology of the G-spot presents as neurovascular tissues with a nerve ganglion. H&E staining is sufficient for the identification of the G-spot complex.

Keywords female erectile body, female prostate, female sexual function, Gräfenberg's zone, G-spot, G-spot anatomy, G-spot histology, G-spot immunohistochemistry, vaginal anatomy.

Introduction

An electronic and manual search of publications dating from 1800 to 2013 related to the G-spot, female ejaculation, and adaptive abilities of the anterior vaginal wall zone yielded over 300 peer-reviewed articles. In 1950, Gräfenberg noted ‘An erotic zone could be demonstrated on the anterior wall of the vagina along the course of the urethra’ and also that ‘During orgasm this area is pressed downwards against the finger like a small cystocele protruding into the vaginal canal’.1 Instead of Gräfenberg’s term ‘a small cystocele’, Masters and Johnson described ‘ballooning’ or ‘tenting’ of the anterior vaginal wall.2 In the 1980s, it was suggested by several authors that the G-spot was not an anatomical reality but instead was a sexology concept.3–7 A majority of professionals in this field now believe that a highly sensitive area exists in the vagina.8,9 Using an electr ovaginogram, Shafik et al.10 identified a ‘vaginal pacemaker’: they stated that ‘The vaginal pacemaker seems to
represent the G-spot, which is claimed to be a small area of erotic sensitivity in the vagina. Increasing blood flow within the vaginal wall during the arousal phase of the female sexual response cycle has been well researched and documented. In April 2012, Ostrzenski documented and published historic results relating to the anatomical existence of the G-spot.

Based upon the anatomical existence of the G-spot in the anterior vaginal wall as established by Ostrzenski, we formulated a hypothesis that the microscopic G-spot must be represented by a specific histological tissue composition that distinguishes the G-spot from the surrounding vaginal and urethral walls. To test this hypothesis we selected hae-matoxylin and eosin (H&E) stain and immunohistochemical stains using CD31, S100, D2-40 and smooth muscle actin (SMA). The objectives of this study were: (1) to expand an investigation related to the existence of an anatomical architecture for the G-spot; (2) to establish the microscopic characteristics of the tissue within the G-spot complex structure; (3) to determine whether a routine histological H&E stain can provide enough microscopic information to confirm the specific histological characteristics of the G-spot.

Methods

The protocol for the observational cohort study was developed and approved by the Institutional Review Board of Warsaw Medical University, Poland (no. AKBE 146/12). Subjects were women between 37 and 68 years of age who had died suddenly from acts of violence, suicide or a drug overdose. No medical records were available due to the sudden death of the women and the type of the institution in which the autopsies were performed. The current investigation was conducted in two stages. Stage I was executed in Poland in May and October 2012 where macrodissections and microdissections, H&E staining and ancillary immunohistochemical investigations were performed. From October 2012 to February 2013, Stage II was conducted in the USA to verify the histological and immunohistochemical findings. Additional staining was performed for the identification of the epithelial origin of the G-spot vasculature using CD31.

We enrolled eight out of ten female cadavers. Permission was requested and granted from the Department of Forensic Medicine Warsaw Medical University to conduct fresh cadaver dissections for scientific research. The cadavers included in this study were of women who had died < 48 hours before the macro- and micro-anatomical dissections were performed. Subjects who demonstrated a disseminated disease such as AIDS, a metastatic neoplastic disease, contagious disease, abnormal configuration or size of the external genitalia, enlarged inguinal lymphatic nodes, intravaginal bruising or haematoma formation were excluded. Those women suspected of having been raped pre- or post-mortem were also excluded, as were cadavers that had been refrigerated for more than 48 hours.

A pelvic surgeon with over 40 years of experience who is also experienced in the anatomical dissection of cadavers (AO) performed the macrodissections and microdissections. The macrodissection was carried out in three planes: the sagittal, oblique and axial. All extirpated G-spot specimens were subjected to microdissection and specimens were preserved in 10% buffered formalin. The Department of Forensic Medicine, Warsaw Medical University prepared the initial paraffin-embedded transverse and longitudinal serial sections of thickness 4 μm and stained them with H&E. The Department of Pathomorphology, Warsaw Medical University took over the paraffin-embedded blocks and H&E-stained slides. Two randomly selected blocks were additionally stained with immunohistochemical markers. The microscopic analyses of the H&E and ancillary immunohistochemical slides were made by a very experienced morphopathologist (A.J.W.). An experienced pathologist (P.G-A.) verified the original histological findings. Photographic images documented the findings. Sections of 4-μm thickness were stained with a standardized immunohistochemical technique using antibodies against SMA (clone 1A4, product code IR611, Dako, Carpenteria, CA, USA) for smooth muscle, CD31 (endothelial cells clone JC70A, product code IR610, Dako) for endothelial cells, D2-40 (anti-human D2-40, clone D2-40, product code IR072, Dako) for endothelial cells of lymphatic origin and S100 (anti-S100, product code IR504, Dako) to highlight the peripheral nerve bundles.

The primary outcome measure was to determine the characteristic anatomical features of the G-spot, building on previous investigations. The secondary outcome measures were to identify the characteristic histological features of the G-spot with H&E stain and ancillary immunohistochemical stains and whether H&E staining provides sufficient information to distinguish the G-spot from the adjacent structures.

The senior author of this article (A.O.) determined the anatomical characteristics of the G-spot by performing macrodissections and microdissections on all cadavers. The characteristic anatomical features of the G-spot were confirmed by P.K., S.T., M.F. and M.N.S. The secondary outcome measures were originally determined by an experienced pathomorphologist (A.J.W.). Verification of the original findings was completed by an experienced pathologist (P.G-A.).

Literature search

A search of the existing literature from 1800 to March 2013 was carried out. Manual and electronic searches were
made using medical subject headings (MeSH), which were selected and used in a search on ISI Web of Science (including conference proceedings), PubMed, ACOG Net, ProQuest, OVID, Cochrane Collection, The Lancet online collection, MD Consult, New England Journal of Medicine, American College of Physician online resources, HighWire, and the Science Citation Index.

Results

The electronic and manual searches failed to identify any histological documentation of the G-spot in the literature. Therefore, this presentation is the first description of the histology of the G-spot in the scientific–clinical literature.

Gross anatomical identification of the characteristic features of the G-spot

In this study group, the G-spot was identified anatomically in all eight women (100%). The G-spot complex was located within the distal anterior vaginal wall, on average 4.5 cm from the urethral meatus (Figure 1D). The G-spot was identified either on the left-lateral or right-lateral urethral borders and rested within the sac wall, the thickness of which was < 2 mm (Figure 1A–C). The anatomical structure of the G-spot itself was surrounded by and intimately fused with vessels, which resemble grape-like clusters (Figure 1A–D). At the tail of the G-spot, a tiny band-like structure grossly resembling a vessel passed into the funnel-shaped cylinder (Figure 1C,D). These vessels were filled with blood to various degrees in each woman (Figure 1A–C). This study documented that the G-spot varies in length, being 7 mm long on average. The G-spot structure and intimately fused vessels combine to create the G-spot complex, which is orientated in a diagonal plane. The angle between the left- or right-lateral urethral boundary and the upper border of the G-spot sac was between 18° and 35° (average 21°). The axial plane (ventral-to-caudal) of the G-spot had three unified parts: a head (the upper pole and widest diameters), a middle part and a tail. The coronal plane (cranial-to-caudal) of the G-spot was cylindrical and fused with vessels. In seven (87.5%) of the women the G-spot complex was found on the left-hand side of the urethral border and in one (12.5%) on the right-hand

Figure 1. Gross anatomical views of the G-spot and surrounding distended vessels. (A) The G-spot sac is opened and G-spot (the white arrow) with surrounding vessels resembling a blue grape-like cluster structure above the instrument is depicted. (B) The vascular bundle migrates into the distal part of the G-spot. The wall of the G-spot sac is visible under the arrowhead. (C) A sagittal close-up view of the G-spot complex is presented. The tail of the G-spot is depicted (the arrow). The rope-like vascular structure protrudes from the G-spot tail (the arrow). The pink colour of the G-spot (arrow) distinguishes this structure from the surrounding vessels, which appear dark blue and are filled with retained blood (the white circle). (D) The left side of the diagram is an illustration of the location of the G-spot complex, which is embedded within the vaginal wall (the white circle) (U, uterus; V, vagina; B, bladder; UM, urethral meatus; R, rectum). On the right, the G-spot complex is presented. The arrow indicates the tail of the G-spot and the white circle encompasses the fused vessels with the G-spot structure.
side. The G-spot complex had the ability to expand to an average of five times its original size when removed from the G-spot sac.

**Histological identifications of the characteristic features of the G-spot**

Histologically, the G-spot tissue architecture consisted of a neurovascular complex embedded within a fibroadipose tissue bed, which housed a large number of peripheral nerve bundles and a neuroganglion. The overlying circular and longitudinal muscles covered the neurovascular complex. The vascular component comprised a large tangled vein-like vascular structure, with some thrombosed and some collapsed vessels with empty lumens, resembling arteriovenous malformations. There were a few smaller feeding arteries in the adjacent adipose tissue. The vascular component did not resemble erectile tissue microscopically Figures 2–4 and see Supporting information, Figures S1–S7).

A panoramic view of a cross-section of the G-spot is shown in Figure 2. There is a large irregularly shaped and partially thrombosed vein-like structure on the right (A) and a smaller round artery at the upper centre (B). The prominent nerve bundles (C) are stained brown (immunohistochemical stain for S100). Figure 3 shows the thick-walled feeding artery (B) and adjacent collapsed vein-like structures (A). The complex vascular structures are embedded in fibroadipose tissue (D). Figure 4 shows a few cross-sections of the thrombosed vein-like tangled vessels (A) and adjacent peripheral nerve bundles (C) within the fibroadipose tissue. The walls of the vascular structures, both thrombosed and collapsed (A in the figures), were diffusely stained by SMA, a smooth muscle marker, confirming the predominantly muscular nature of the wall (Figure 2). Both thrombosed and collapsed vascular structures and the smaller arteries were lined by a thin endothelial cell layer, which is visually enhanced by CD31 immunohistochemical staining (Figures 2–4 and see Supporting information, Figures S1, S2). We used D2-40, a specific immunohistochemical marker for endothelial cells of lymphatic origin, to establish the type of these large tangled vessels seen grossly (Figure 1A–C). Microscopically, Figure 2 reveals negative staining of endothelial cells, confirming a non-lymphatic origin for these vascular structures. The abundant peripheral nerve bundles of the G-spot are demonstrated in Figure 2, and a large nerve ganglion resting within the adipose tissue bed is depicted in Figures S6 and S7. The overlying circular and longitudinal muscles covered the G-spot complex. Using ancillary techniques we were able to identify the composition of the vessel walls, which contained predominantly smooth muscle, depicted as stained brown with SMA (Figure 2 and see Supporting information, Figures S1–S7).

**Figure 2.** A panoramic microscopic view of the neurovascular complex (immunohistochemical stain for S100, Dako; scanning magnification × 40). The view shows bluish grape-like vascular structures microscopically composed of cross-sections of a complex large vascular structures (A, right) that contain fibrin and blood cells (partially thrombosed). A feeding thick-walled artery (B) is also shown in Figure 3. There are abundant peripheral nerve bundles stained brown by S100 immunostain (C). Adjacent to the nerve bundles on the left side of the picture there are a few collapsed vein-like structures are shown (A, left).

**Figure 3.** Thick-walled and thin-walled blood vessels (H&E stain; original magnification × 200). A thick-walled feeding artery (B) and adjacent thinner-walled collapsed vein-like structures (A) are embedded in a fibroadipose tissue bed (D).

**Figure 4.** Thrombosed complex large vessels (H&E original stain; magnification × 200). (A) The thrombosed complex large vascular structures. (C) The fibroadipose tissue bed which is rich in nerve bundles.
information, Figure S3). The endothelial cells were negative for D2-40 (Figure 2 and see Supporting information, Figures S4 and S5). The peripheral nerve bundles within the fibroadipose bed stained for S100 (C in Figure 2 and see Supporting information, Figures S6 and S7). Figure S7 shows a large nerve ganglion (E) resting within the adipose tissue bed (S7D).

Discussion

Main findings

The findings of this investigation confirmed Ostrzenski’s previous anatomical description of the G-spot. The present study is the first to demonstrate the microscopic architecture of the G-spot. We found characteristic histological features that distinguish the G-spot from the surrounding vaginal and urethral walls. The histology of the vagina and the urethra has been presented elsewhere. The presence of a nerve ganglion within the G-spot complex was the paramount discovery, because a nerve ganglion has never before been documented within the vaginal wall (see Supporting information, Figure S3). The vascular component comprised large vein-like vessels and smaller feeding arteries which were tangled together. There were occasional arteriovenous interconnections (Figures 2–4 and see Supporting information, Figures S1 and S2). We also established that the G-spot does not contain erectile tissue (corpus cavernosum) (the histology of erectile tissue can be found elsewhere). H&E and immunohistochemical methods were used to stain the G-spot tissue to enhance microscopic analysis. The microscopic examination established that the G-spot is a complex neurovascular structure (Figure 2 and see Supporting information, Figures S6 and S7). The simplicity, low cost and accessibility of the H&E staining technique and staining materials in general make the histological evaluation of the G-spot complex an easy process in every histological laboratory.

The neurocomponent contains abundant peripheral nerve fibres and nerve bundles (Figure 2 and see Supporting information, Figure S6) as well as a nerve ganglion (see Supporting information, Figure S7). The presence of a nerve ganglion within the G-spot structure was a surprising discovery, because a nerve ganglion has never been identified within the vaginal wall. The morphological structure of autonomic nerve ganglia and their networks can be found elsewhere and these analyses are beyond the scope of this paper.

Strengths and limitations

The strengths of this investigation are verifications of the anatomical existence of the G-spot complex and documentation of its characteristic histological features. Through the centuries, numerous attempts have failed to show the anatomical existence of the G-spot, but we have been able to document its anatomy, confirming Ostrzenski’s work. Also, the present study has established the characteristic histology of the G-spot complex. These discoveries have significant potential for clinical and scientific research, although at the moment potential clinical applications are strictly speculative and require the development of a new study protocol(s) to confirm our suppositions.

Limitations of our study were that our work was conducted on fresh adult human female cadavers. Inherent weaknesses exist in such an anatomical study because the results are subject to the interpretation of the researcher. Moreover, a shortcoming of any study on fresh cadavers is the presence of post-mortem topographic distortions. Also, the absence of accepted terms can affect how the findings are described. There may be variations in the anatomy of the G-spot complex in different ethnic and racial groups, which could not be established in this study because the only cadavers available were those of white women.

Interpretations

Histological similarities of the G-spot cannot be presented here because as far as we know no similar description has ever been presented in the literature. The G-spot has been presented as a glandular tissue. Our microscopic analysis did not identify any glandular tissue within the structure of the G-spot complex. Likewise, we did not find any histological evidence of erectile tissue as previously suggested in the gross anatomical description by Ostrzenski. A detailed description of the structure of erectile tissue has been published elsewhere. The current histological investigation demonstrated the presence of a neurovascular structure with a nerve ganglion within the G-spot tissue architecture (see Supporting information, Figures S2, S6 and S7). The presence of a nerve ganglion within the G-spot distinguishes this anatomical structure from the adjacent anterior vaginal wall, because a nerve ganglion has not been identified within the vaginal wall. Also, arteriovenous entanglements and interconnections were characteristic findings for the G-spot (Figures 2–4, see Supporting information, Figures S1 and S2).

Using an electrovaginogram, Shafik et al. documented the presence of a generator (a ‘pacemaker’) for vaginal electrical activity located in the distal anterior vagina. Indeed, this location corresponds to our findings in this study and to Ostrzenski’s original results. Our histological findings strongly support the pacemaker theory of the G-spot (see Supporting information, Figures S6 and S7).

We reviewed previously published magnetic resonance images and concluded that some of these were highly suggestive of the existence of a G-spot complex that was
best delineated during the phase of sexual arousal. This review cannot be considered as conclusive evidence and a new magnetic resonance imaging study protocol must be developed to provide definitive evidence.

There may be significant potential for future clinical and scientific research applications of the discovery of the anatomy and histology of the G-spot. A finding of G-spot insufficiency may help in the diagnosis and eventual therapy of sexual dysfunction. 20-23 It has been postulated in the literature that the existence of a G-spot could lead to vaginally activated orgasms; therefore vaginal anorgasemia may exist when function of the G-spot is compromised. 19 Since we now know that the G-spot complex is a neurovascular anatomical structure, there is the potential for diagnosis and therapy of insufficiency of the G-spot complex. Hypothetically, based upon our current findings, insufficiency of the G-spot complex could result from neurogenic or vasculogenic impairment or a combination of both. The aetiology of vasculogenic and neurogenic origins of sexual dysfunction has already been presented in the scientific–clinical literature.20-23 The results of our investigation could therefore lead to the development of clinical diagnostic and therapeutic tools.

The findings of our study strongly suggest that injections into the anterior vaginal wall could result in inadvertent injury to the G-spot complex. Analysis of the anecdotal marketing literature (because our search of the clinical-injury to the G-spot complex. Analysis of the anecdotal marketing literature failed to identify any reports) determined that collagen injections in the anterior vaginal wall were promoted in a G-spot Amplification® procedure. Any injection in the anterior vaginal wall should be withheld until such time that the results of well-designed and well-executed scientific/clinical studies indicate otherwise. Additionally, a scientific–clinical investigation should be conducted to document the rationale for anecdotal reports of hormonal injections (estrogens, androgens or oxytocin) into the anterior vaginal wall to improve or treat human female sexual function.

In the current study we have documented the existence of a G-spot complex in each woman studied regardless of the woman’s reproductive or postmenopausal age. However, the protocol of our study did not call for evaluation of G-spot hormonal receptors. Such a determination will require an additional study.

Thabet’s findings,18 analysed in Ostrzenski’s article,12 related to the anatomical discovery of the G-spot. We agree with these authors’ suggestions that precautions should be undertaken during an operation on the anterior vaginal wall to avoid ligating vasculature or injuring the G-spot24 complex. Based upon our findings, Ostrzenski changed the point of surgical incision from the midline to a lateral location during an anterior vaginal reconstruction to avoid inadvertent injury to the G-spot complex.

Conclusion

The anatomical existence of the G-spot complex was confirmed, and its characteristic anatomical features correspond to previous presentations. Histologically, the G-spot is a neurovascular complex structure with a nerve ganglion. Staining with H&E is sufficient for the diagnosis of a G-spot complex structure.

The findings of this study can be incorporated into clinical practice in the diagnosis and treatment modalities for G-spot insufficiency. Also, the results of this investigation set the groundwork for potential clinical scientific research into the vaginally activated orgasm and, more generally, the role of the G-spot in female sexual function and dysfunction.

Disclosure of interest

None.

Contribution to authorship

AO designed the study, performed macro- and micro-dissections, coordinated the study, reviewed the literature and wrote the manuscript. PK selected clinical materials for the study and supervised the H&E staining. AJW performed the original immunohistochemical staining and described histological and immunohistochemical findings. PG-A verified the histological and immunohistochemical findings, performed additional immunohistochemical tissue staining and wrote the histological part of the manuscript. MNS prepared and positioned women for dissections, secured video and digital image documentations and obtained copies of the pertinent scientific articles. ST assisted in anatomical dissections. MF assisted in anatomical dissections and conducted the literature search.

Details of ethics approval

Warsaw Medical University Bioethics Committee approved the study protocol (AKBE 146/12).

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Supporting Information

Additional Supporting Information may be found in the online version of this article.
Figure S1. Thrombosed large vascular structures and the smaller feeding artery.

Figure S2. Collapsed vessels are casted (H&E stain, original magnification × 200).

Figure S3. The vascular smooth mussels (immunohistochemical stain for smooth muscle actin [SMA] clone1A4-DAKO; original magnification × 200).

Figure S4. Endothelial cells are displayed (immunohistochemical stain for CD31, endothelial Cells, Clone JC70A-DAKO; original magnification × 200).

Figure S5. The documentation of nonlymphatic origin of the large vascular structures is depicted (immunohistochemical stain for D2-40-DAKO); original magnification × 200).

Figure S6. Peripheral nerve bundles are revealed (immunohistochemical stain for S-100, original magnification × 100).

Figure S7. The G-spot’s nerve-ganglion is revealed (haematoxylin & eosin stain; original magnification × 200).

References

G-spot: the facts to the fantasy

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Mini commentary on ‘Verification of the anatomy and newly discovered histology of the G-spot complex’

Dating as far back as the eleventh century, ancient Indian texts, the Kamaśastra and Jayamangala scripts, describe an erogenous area of heightened sensitivity, able to induce sexual pleasure in the vagina. Subsequently, Regnier de Graaf, the seventeenth century Dutch physician known for his scientific contribution on ovarian (Graafian) follicles, referred to an erogenous zone in the vagina.

Publishing in the International Journal of Sexology in 1950, German gynecologist Ernst Gräfenberg stated ‘An erotic zone always could be demonstrated on the anterior wall of the vagina along the course of the urethra’ (Gräfenberg. Int J Sexol 1950;3;145–8). This area was noted to be 1–2 cm wide.


Since this time, much research has been dedicated to the fact or fiction of the G-spot. An ambitious review, aimed at providing an overview of the evidence both supporting and refuting the existence of an anatomically distinct G-spot, was undertaken by Amichai Kilchevsky and colleagues and published in Journal of Sexual Medicine in 2012 (Kilchevsky et al. J Sex Med 2012;9;1355–9). The authors noted that dozens of trials have been published in literature attempting to confirm the existence of a G-spot, using surveys, pathological specimens, various imaging modalities and biochemical markers. Interestingly, the majority of women surveyed believed in the G-spot, even if they were unable to locate it. Although the authors noted studies characterising vaginal intervention that show differences in nerve distribution across the vagina, they are not reproducible. Moreover, radiological imaging fails to demonstrate a unique anatomic entity. As a result, the authors conclude that objective measures fail to provide strong and consistent evidence for the existence of a G-spot anatomical site. The authors ultimately conclude that ‘reliable reports and anecdotal testimonials of the existence of a highly sensitive area in the distal anterior vaginal wall raise the question of whether enough investigative modalities have been implemented in search of the G-spot’.

The current study by Ostrzenski et al. (BJOG 2014; DOI: 10.1111/1471-0528.12707) which is a follow-up article to a single cadaveric dissection of the G-spot by the lead author (J Sex Med 2012;9;1355–9), now provides reliable evidence for the G-spot’s existence in the anterior vaginal walls of eight consecutive fresh human female cadavers that were dissected; G-spot microdissection was also performed. Interestingly, the authors are able to identify the G-spot intimately fused with vessels, creating a complex in all women studied. The investigators note a ‘large tangled vein-like vascular structure’ resembling arteriovenous malformations. They also found a band-like structure protruding from the G-spot tail. Histological evaluation of the G-spot tissue yields findings of a neurovascular complex structure, including peripheral nerve bundles, nerve-ganglia, large vein-like vessels and smaller feeding arteries. This G-spot complex is covered by circular and longitudinal muscles.

After reading this article and witnessing the reproducibility of the findings as noted by Ostrzenski et al., I believe for the first time that we can agree that the G-spot truly is a distinct anatomical entity. No longer should we debate G-spot, fact or fantasy; rather there are facts to the fantasy.

Disclosure of interests
The author has no commercial or other conflicting interest regarding the above commentary.