Vaginal dryness in primary Sjögren’s syndrome: a histopathological case–control study

Jolien F. van Nimwegen, Karin van der Tuuk, Silvia C. Liefers, Gwenny M. Verstappen, Annie Visser, Robin F. Wijnsma, Arjan Vissink, Harry Hollema, Marian J. E. Mourits, Hendrika Bootsma and Frans G. M. Kroese

Abstract

Objective. The aim was to study clinical, histopathological and immunological changes in the vagina and cervix of women with primary SS, which might explain vaginal dryness.

Methods. We included 10 pre-menopausal female primary SS patients with vaginal dryness and 10 pre-menopausal controls undergoing a laparoscopic procedure. The vaginal health index was recorded. Multiplex immunoassays and flow cytometry were performed on endocervical swab and cervicovaginal lavage samples to evaluate cellular and soluble immune markers. Mid-vaginal and endocervical biopsies were taken and stained for various leucocyte markers, caldesmon (smooth muscle cells), avian V-ets erythroblastosis virus E26 oncogene homologue (ERG; endothelial cells) and anti-podoplanin (lymphatic endothelium). The number of positive pixels per square micrometre was calculated.

Results. One patient was excluded because of Clamydia trachomatis, and two controls were excluded because of endometriosis observed during their laparoscopy. Vaginal health was impaired in primary SS. CD45⁺ cells were increased in vaginal biopsies of women with primary SS compared with controls. Infiltrates were predominantly located in the peri-epithelial region, and mostly consisted of CD3⁺ lymphocytes. In the endocervix, CD45⁺ infiltrates were present in patients and in controls, but a higher number of B lymphocytes was seen in primary SS. Vascular smooth muscle cells were decreased in the vagina of primary SS patients. No differences were found in leucocyte subsets in the vaginal and endocervical lumen. CXCL10 was increased in endocervical swab samples of primary SS patients.

Conclusion. Women with primary SS show impaired vaginal health and increased lymphocytic infiltration in the vagina compared with controls. Vaginal dryness in primary SS might be caused by vascular dysfunction, possibly induced by IFN-mediated pathways.

Introduction

Primary SS is a systemic autoimmune disease with a heterogeneous presentation, including sicca symptoms, systemic symptoms, such as fatigue, and extraglandular involvement [1]. A hallmark of primary SS is lymphocytic infiltration of the salivary and lacrimal glands. Besides sicca symptoms of the eyes and mouth, vaginal dryness is common in women with primary SS, which causes
dyspareunia and sexual dysfunction [2–6]. Although vaginal dryness usually occurs after menopause, in primary SS vaginal dryness often occurs at a younger age [7–9]. Two studies evaluating vaginal health in primary SS reported erythema of the vaginal epithelium [10, 11], whereas others did not find any macroscopic changes of the vagina and cervix [4, 8]. In a previous study, we did not observe changes in the vaginal microbiome in primary SS [12].

The pathophysiology of vaginal dryness in primary SS is still unknown. Normally, the vaginal surface is humidified and lubricated by transudate from the lamina propria, which contains rich venous and lymphatic networks, and by mucus produced by the endocervical glandular epithelium [13]. In pre-menopausal primary SS patients with dyspareunia, lymphocytic infiltrates were found in the stroma underlying the vaginal epithelium [2, 14]. Furthermore, chronic cervicitis was observed in biopsies of 42% of primary SS patients [11]. Local inflammation might influence the production of transudate from blood vessels in the vagina or compromise the function of the mucus-producing glandular epithelium of the endocervix.

In previous studies, few or no healthy controls were included, and no quantitative analyses were performed. Given that leucocytes are present physiologically in the vagina and cervix of healthy women [15–17], quantitative analysis and comparison with a control group are necessary to assess whether the lymphocytic infiltration observed in primary SS is indeed pathological. Furthermore, changes in the vascularization of the vagina were not taken into account as a possible cause of vaginal dryness.

To identify an appropriate treatment for vaginal dryness in primary SS, the pathogenesis of this symptom needs to be elucidated. The objective of this study was therefore to assess clinical and histopathological changes in the vagina and cervix of women with primary SS compared with controls, which might explain vaginal dryness. We also explored whether possible inflammatory changes in the vagina and cervix of primary SS patients were reflected by changes in immune cells and effector molecules in the vaginal lumen.

Methods

Study population

In a prospective exploratory case–control study, we included 10 women with primary SS who fulfilled ACR–EULAR criteria and reported vaginal dryness. We also included 10 age-matched controls without systemic autoimmune diseases who were scheduled for a laparoscopic procedure. To eliminate the influence of physiological hormonal changes to the vaginal mucosa, only pre-menopausal patients and controls were included. Other inclusion criteria were age ≥ 18 years and written informed consent. Exclusion criteria were pregnancy or breast-feeding, presence of inflammatory or infectious gynaecological disease, previous chemotherapy, current use of an intra-uterine contraceptive device, hormone replacement therapy or vaginal oestrogen supplementation, and use of systemic CSs or DMARDs ≤ 6 months before inclusion. The study complies with the Declaration of Helsinki and was approved by the Medical Ethics Committee of the University Medical Center Groningen (METC 2015/039).

Study procedures

Participants were instructed not to have sexual intercourse or to use tampons, lubricants or any other vaginal products within 72 h before the study visit. On the day of examination, participants completed a questionnaire that included the female sexual function index and questions about co-morbidities, medication use, smoking status, vaginal symptoms and the presence of vaginal bacterial or fungal infections in the past year. In primary SS patients, the EULAR Sjögren’s syndrome patient reported index and the EULAR Sjögren’s syndrome disease activity index were recorded. Blood samples were obtained.

Gynaecological examination was performed by an experienced gynaecologist. The five domains of the vaginal health index (elasticity, fluid secretion, pH, epithelial mucosa and moisture) were scored on a scale from one to five, resulting in a total score of 5–25 (Supplementary Table S1, available at Rheumatology online) [18]. Cervicovaginal lavage (CVL) samples were collected by flushing 7 ml of PBS over the cervix and vagina, aspirating the PBS and then repeating the procedure [19]. Endocervical swab (ES) samples were collected by rotating eSwabs (Copan diagnostics, Murrieta, CA, USA) in the endocervical canal. The eSwabs were put in 5 ml of PBS. CVL and ES samples were immediately put on ice.

Another eSwab, suspended in eSwab transport medium, was used for PCR to detect Chlamydia trachomatis and Neisseria gonorrhoea. A vaginal secretion sample was collected for fungal culture. ThinPrep Papanicolaou tests (Hologic, Marlborough, MA, USA) were performed on cervical samples collected with a Cervex brush (Rovers Medical Devices, Oss, The Netherlands).

Finally, full-thickness mid-vaginal and endocervical punch biopsies were collected, after administration of local anaesthesia in primary SS patients or general anaesthesia in controls. Vaginal and cervical biopsies were fixed in 4% paraformaldehyde and embedded in paraffin.

Evaluation of vaginal and endocervical biopsies

Vaginal and endocervical tissue sections were stained with Haematoxylin and Eosin (HE), periodic acid–Schiff diastase (PAS-D) and various leucocyte markers (CD45, CD3, CD4, CD8 and CD20). Tissue sections were also stained for blood/lymphatic vessel-associated markers: avian V-ets erythroblastosis virus E26 oncogene homologue (ERG), which is a nuclear stain for endothelial
cells; anti-podoplanin (clone D2-40), which stains lymphatic endothelium; and caldesmon, which stains smooth muscle cells present in the tunica media of arterioles and larger venules. Endocervical tissue sections were additionally stained for CD138, because many plasma cells were seen in HE-stained tissue sections.

The HE- and PAS-D-stained sections were examined by a dedicated gynaecopathologist to check for gynaecological morbidity and fungal infections. Immunohistologically stained sections were analysed quantitatively by counting the number of diaminobenzidine-stained pixels per square micrometre of parenchyma, using the Positive Pixel Count algorithm (v.9.1) in ImageScope v12.1 (Leica biosystems, Wetzlar, Germany). For CD4, only strongly positive pixels were counted, to exclude non-specific staining. The epithelial layer was excluded from analysis of endothelial markers and CD138, because no blood or lymphatic vessels are present in the epithelium, and CD138 is expressed by stratified squamous epithelium. To quantify vaginal atrophy, the epithelial thickness and number of cell layers were counted at ×40 magnification, in three areas of the biopsy in which the epithelium was thinnest and no dermal papillae were present. The mean epithelial thickness and number of cell layers were calculated.

Evaluation of cellular and soluble immune markers
Serum was frozen at −80°C. EDTA whole blood was lysed with ammonium chloride and centrifuged. The supernatant was discarded, and cells were washed and suspended in FACS buffer at a concentration of 10^6 cells/ml. To collect endocervical material, the swabs containing ES samples were gently scraped on the edge of the Falcon tubes in which they were kept after collection. The ES and CVL samples were then resuspended and centrifuged, after which the supernatant was frozen at −80°C, and cells were resuspended in FACS buffer at a concentration of 10^6 cells/ml.

Flow cytometry analysis of leucocyte subsets in cells from whole blood, ES and CVL was performed on the day of collection of the samples. Cells were washed and stained with antibodies directed against leukocyte markers (Supplementary Table S2, available at Rheumatology online), after which they were washed and resuspended in FACS buffer. Shortly before analysis, cells were stained with propidium iodide (eBioscience, San Diego, California, USA) and passed through a 35 μm nylon mesh. Antibody panel optimization and titrations were performed in cells from whole blood and confirmed in ES and CVL cells. Fluorescence-minus-one controls were included to determine the background fluorescence. Data were acquired using a LSRII flow cytometer (BD Biosciences, Franklin Lakes, NJ, USA). Data were analysed using FlowJo (Tree Star, Ashland, Oregon, USA). The gating strategy is described in Supplementary Fig. S1, available at Rheumatology online.

Serum samples and supernatants of the CVL and ES samples were thawed and analysed for levels of APRIL (a proliferation-inducing ligand), BAFF (B-cell activation factor), IFN-γ, RANK-ligand, TNF-α, CCL2, CCL4, CX3CL, CXCL9, CXCL10, CXCL11, CXCL13, IL-6, IL-7, IL-8 and IL-17A, using a human magnetic Luminex pre-mixed 16-plex assay (R&D Systems, Minneapolis, MN, USA), according to the manufacturer’s protocol. Data were acquired on a Luminex 200 system.

Statistical analysis
Statistical analyses were executed using SPSS Statistics v.23 (SPSS, Chicago, IL, USA). The Mann–Whitney U-test, χ² test or Fisher’s exact test was used as appropriate to compare differences between groups. Spearman’s correlation coefficients were used to evaluate correlations. P-values of <0.05 were considered to indicate statistical significance.

Results
Clinical characteristics
One primary SS patient was excluded owing to the presence of Chlamydia trachomatis. Two controls were excluded owing to detection of endometriosis during laparoscopy, because the pathogenesis of endometriosis comprises immunological changes [20], and an association between endometriosis and primary SS has been described [21, 22]. Characteristics of remaining participants are shown in Supplementary Table S3, available at Rheumatology online. The median age was 36 (interquartile range (IQR) 33–46) years for primary SS patients (n = 9) and 41 (IQR 36–44) years for controls (n = 8). All primary SS patients had a positive salivary gland biopsy (focus score ≥1), and seven (78%) were anti-SSA antibody positive. Median ACR–EULAR score was 9 (interquartile range; IQR 5–9) and median EULAR Sjögren’s syndrome disease activity index 6 (IQR 3–9).

Gynaecological symptoms and examination
Compared with controls, patients with primary SS showed lower female sexual function index scores (indicating sexual dysfunction), used lubricants more often and had an increased prevalence of superficial dyspareunia (Table 1). The vaginal health index score was significantly lower in primary SS patients, indicating impaired vaginal health (Table 1; Fig. 1). Of vaginal health index subdomains, the mucosa score was significantly decreased in primary SS, indicating frailty and a higher tendency of the epithelium to bleed. Upon inspection of the vulva, vagina and cervix, no major abnormalities were found. Some redness of the vulva was noted in three primary SS patients. One patient with active cutaneous vasculitis on her legs showed petechiae on the labia majora. Superficial vulvar rashes were seen in three patients and one control. Vaginal pH did not differ significantly between groups,
and none of the participants showed signs of vaginal atrophy.

**Histological findings**

No major abnormalities or fungal infections were found in vaginal or cervical HE- and PAS-D-stained tissue sections. One vaginal biopsy from a control was excluded from further analysis, because it was very superficial, consisting of 98% epithelium. Three primary SS patients and two controls were excluded from analysis of endocervical biopsies, because only ectocervical tissue or mucus was collected owing to difficulties in reaching the endocervical tissue through the external cervical ostium.

No significant differences were found in the number of cell layers (patients: median 25, IQR 21–33; controls: median 25, IQR 20–26) or thickness (patients: median 251 μm, IQR 197–271 μm; controls: median 243, IQR 142–252 μm) of the vaginal epithelium.

**Lymphocytic infiltration in vagina and endocervix**

Compared with controls, vaginal tissue from primary SS patients contained significantly higher numbers of CD45+ cells (Table 2; Fig. 2). Lymphocytic infiltrates in primary SS patients were mainly located in the lamina propria immediately below the epithelium (peri-epithelial layer), with a peri-epithelial localization and aggregates in dermal papillae (Figs 3 and 4). Of all leucocyte subsets, only CD3+ lymphocytes were significantly increased in the vagina. In endocervical tissue sections, there was no significant difference in the total number of CD45+ cells, although the number of CD20+ B lymphocytes was significantly higher in primary SS patients (Table 2; Fig. 2). Lymphocytic infiltration in the endocervix was also located mostly in the peri-epithelial layer (Figs 3 and 4).

**Endothelial changes in vagina and cervix**

To explore whether blood vessels and lymphatic vessels in the vagina and endocervix are affected in primary SS, we stained for endothelial markers (Supplementary Fig. S2, available at Rheumatology online). The number of caldesmon+ cells was significantly lower in vaginal biopsies of women with primary SS, indicating a decrease in vascular smooth muscle cells (Table 2; Fig. 2).
seemed to be a tendency towards an increase in the number of lymphatic endothelial cells (D2-40) in primary SS. No significant differences were found in other endothelial markers in the vagina or endocervix.

Immune markers in blood, CVL and ES

Next, we explored whether the histological changes in the vagina and endocervix are reflected by cellular and soluble immune markers in the lumen. No differences

**TABLE 2** Quantitative analysis of leucocyte and endothelial markers in the vagina and endocervix

| Marker | Vagina | Endocervix |
|--------|--------|------------|
|        | Primary SS | Control | P-value | Primary SS | Control | P-value |
| CD45   | 0.34 (0.26–0.53) | 0.26 (0.12–0.27) | 0.012* | 1.12 (0.45–1.82) | 0.60 (0.32–2.97) | 1.000 |
| CD3    | 0.49 (0.28–0.56) | 0.19 (0.12–0.27) | 0.008* | 0.66 (0.38–1.28) | 0.44 (0.20–1.57) | 0.485 |
| CD4    | 0.23 (0.14–0.34) | 0.13 (0.12–0.32) | 0.470 | 0.66 (0.25–1.21) | 0.34 (0.24–1.25) | 1.000 |
| CD8    | 0.48 (0.32–0.99) | 0.34 (0.22–0.51) | 0.210 | 1.00 (0.73–1.49) | 0.64 (0.28–2.05) | 0.485 |
| CD20   | 0.22 (0.17–0.47) | 0.20 (0.14–0.40) | 0.837 | 0.53 (0.44–2.45) | 0.32 (0.25–0.55) | 0.041* |
| ERG    | 0.23 (0.17–0.26) | 0.26 (0.18–0.28) | 0.470 | 0.50 (0.41–0.78) | 0.67 (0.23–0.87) | 0.818 |
| Caldesmon | 0.06 (0.03–0.07 | 0.11 (0.07–0.21) | 0.031* | 0.15 (0.06–0.57) | 0.14 (0.05–0.30) | 0.818 |
| D2-40  | 0.11 (0.06–0.26) | 0.06 (0.04–0.09) | 0.210 | 0.30 (0.12–0.41) | 0.20 (0.09–0.27) | 0.240 |
| CD138a | ND | ND | ND | 1.03 (0.17–2.01) | 0.22 (0.11–2.87) | 0.792 |

Values are the median (interquartile range) number of positive pixels per square micrometre.

CD138 was analysed in six patients and five controls, because one control did not show representative endocervical tissue in the CD138-stained tissue section.

*P<0.05.

D2-40: anti-podoplanin (clone D2-40); ERG: avian V-ets erythroblastosis virus E26 oncogene homologue; ND: Not done.

Low scores correspond to low vaginal health. pSS: primary SS.
were found in the proportion of leucocyte subsets in CVL or ES (Supplementary Table S4, available at Rheumatology online).

A significantly higher level of CXCL10 was found in ES samples of patients with primary SS (Supplementary Table S5, available at Rheumatology online). No other
significant differences in chemokine or cytokine levels of patients and controls were found in ES or CVL samples. In serum, CXCL10 and CXCL11 were significantly increased in primary SS patients. Within the group of primary SS patients, a strong correlation was seen between CXCL10 in ES and CXCL10 in serum ($p = 0.717, P = 0.03$) and between CXCL10 in serum and the number of CD45$^+$ cells in the vagina ($p = 0.667, P = 0.05$). Levels of IFN-$\gamma$, IL-17A, CCL4, CX3CL and CXCL9 were below detection limits in serum, CVL and ES in most patients.

**Discussion**

Women with primary SS and vaginal sicca symptoms often experience sexual dysfunction and dyspareunia. We observed that women with primary SS have impaired vaginal health and an increased tendency of the vaginal epithelium to bleed. Furthermore, we found a peri-epithelial infiltration and decreased number of vascular smooth muscle cells in the vaginal wall of primary SS patients, which are likely to contribute to vaginal dryness. In contrast to post-menopausal women, the vaginal dryness in women with primary SS cannot be explained by atrophic vaginitis, because no signs of atrophy or increased pH were found.

Our study provides the first in-depth, quantitative evaluation of immunological and histopathological markers in the cervicovaginal mucosa of a well-defined group of primary SS patients, compared with healthy controls. By including only pre-menopausal patients, matching patients for age and screening for infections, we minimized the influence of confounders. We found higher numbers of infiltrating CD45$^+$ cells in vaginal biopsies of primary SS patients, with a peri-epithelial localization and aggregates in dermal papillae. This difference in CD45$^+$ cells seems to be largely attributable to CD3$^+$ T cells. Although both CD4$^+$ and CD8$^+$ T cells were present in peri-epithelial infiltrates, neither were significantly overrepresented in primary SS patients. The exact phenotype of the infiltrating CD3$^+$ T cells in the vagina of primary SS patients remains to be established. In the endocervix, CD45$^+$ infiltrates were present in patients and controls, but with a higher number of B lymphocytes in primary SS patients. The vaginal and endocervical epithelium remained intact in primary SS. Lymphocytes did not seem to migrate through the

![Fig. 3 Haematoxylin and Eosin and CD45 stains in the vagina and endocervix of a primary SS patient and a control](https://academic.oup.com/rheumatology)
epithelial layer, because no differences were found in the composition of leucocyte subsets in the vaginal and endocervical lumen using flow cytometry.

Our findings are in line with previous observations showing the presence of inflammatory infiltrates in the vagina and cervix of women with primary SS in HE-stained sections [2, 11, 14]. Why lymphocytes migrate to these sites is not yet known, but it is likely that CXCL10 is involved. This IFN-induced chemokine plays a dominant role in the pathogenesis of primary SS, and increased levels are reported in saliva, tear fluid, serum and now also in ES samples [23, 24]. The origin of CXCL10 in the ES samples is not yet known. Given the correlation with serum levels, a part of CXCL10 in the ES samples might be derived from serum by transudation, but it might also be produced locally. Salivary gland ductal epithelial cells produce CXCL10, which subsequently results in formation of periductal infiltrates [25]. Likewise, vaginal and endocervical epithelial cells might produce this chemokine, explaining the characteristic peri-epithelial vaginal infiltrate in the lamina propria.

The formation of transudate from the lamina propria, which is rich in capillaries and post-capillary venules, is important for humidification of the vagina. The lymphocytic infiltrate might either damage capillaries/post-capillary venules at these sites or otherwise interfere with generation of the transudate. Importantly, we observed that numbers of vascular smooth muscle cells are significantly decreased in the vagina of primary SS patients. Whether this decrease reflects destruction of vascular smooth muscle cells or a decrease in total number of arterioles remains to be elucidated. Either way, a decrease in smooth muscle cells might disturb the production of transudate, considering the important role of smooth muscle cells in the regulation of the blood flow in the vaginal vascular network during sexual arousal [26].

Although the reason for the decrease in smooth muscle cells is not clear, there are several studies showing that blood vessel homeostasis is disturbed in primary SS. First, numbers of circulating endothelial precursor cells are increased in primary SS, indicating endothelial damage [27]. Second, there is an increase in numbers of circulating angiogenic T cells, which contribute to endothelial repair but may also have cytotoxic and pro-inflammatory effects [28]. Third, soluble intracellular adhesion molecule 1 and soluble vascular cell adhesion molecule 1 are elevated in serum of primary SS
patients, which are associated with endothelial cell activation and dysfunction [29]. Fourth, functional impairment of the arterial wall and vascular smooth muscle cells has been described in primary SS [29, 30]. Taken together, we hypothesize that vaginal dryness is impaired in primary SS patients as a result of vascular dysfunction. Endothelial damage might also explain the increased bleeding tendency of the vaginal epithelium in primary SS patients. The development of vascular dysfunction might be mediated by the IFN pathway, in a similar manner to that in SLE, in which IFN alters the balance between endothelial cell apoptosis and vascular repair mediated by endothelial cell progenitors and myeloid angiogenic cells [31, 32].

This study focused on the vaginal and cervical epithelium, because these are the main sources of vaginal lubrication. Whether the vestibular glands (Bartholin’s and Skene’s glands) are affected by primary SS remains unknown. However, Bartholin’s glands provide only a small contribution to lubrication of the vestibule of the vagina of healthy individuals [33] and whether the paraurethral glands (Skene’s glands) contribute to lubrication of the vulva is still under debate [34]. Skene’s glands most probably produce some fluid only during orgasm, if ever.

Limitations of our study are the small sample size and subjective measurement of vaginal dryness. Furthermore, given that we did not include primary SS patients without vaginal dryness or non-primary SS controls with vaginal dryness, it still has to be evaluated whether the cervicovaginal changes that we found in women with primary SS are the cause or a consequence of vaginal dryness, and whether they are specific for primary SS patients. Future studies should quantify vaginal lubrication objectively in a larger group of patients and evaluate the relationship of vaginal dryness to our findings. Lastly, although we aimed to include all patients during the follicular phase of the menstrual cycle, two controls were included in the luteal phase, because their laparoscopic procedures could not be planned in the follicular phase. Menstrual cycle phase might influence soluble immune markers in the vagina and cervix, but probably does not influence cellular markers [15, 19, 35].

In conclusion, our study shows that women with primary SS and vaginal dryness have sexual dysfunction, impaired vaginal health and increased lymphocytic infiltration in the vaginal lamina propria. We postulate that vaginal dryness in women with primary SS is caused by vascular dysfunction, possibly induced by IFN-mediated pathways.

Acknowledgements

We thank Ellen Klinkert for her support during data collection.

Funding: This work was supported by a grant from the Dutch Arthritis Society (ReumaNederland) [14-1-301 to H.B.].

Disclosure statement: The authors have declared no conflicts of interest.

Supplementary data

Supplementary data are available at Rheumatology online.

References

1 Brito-Zerón P, Baldini C, Bootsma H et al. Sjögren syndrome. Nat Rev Dis Primers 2016;2:16047.
2 Mulherin DM, Sheenan TP, Kumararatne DS et al. Sjögren’s syndrome in women presenting with chronic dyspareunia. Br J Obstet Gynaecol 1997;104:1019–23.
3 van Nimwegen JF, Arends S, van Zuiden GS et al. The impact of primary Sjögren’s syndrome on female sexual function. Rheumatology 2015;54:1286–93.
4 Priori R, Minniti A, Derme M et al. Quality of sexual life in women with primary Sjögren syndrome. J Rheumatol 2015;42:1427–31.
5 Isik H, Isik M, Aynioglu O et al. Are the women with Sjögren’s syndrome satisfied with their sexual activity? Rev Bras Reumatol Engl Ed 2017;57:210–6.
6 Ugurlu GK, Erten S, Ugurlu M, Caykoylu A, Altunoğlu A. Sexual dysfunction in female patients with primary Sjögren’s syndrome and effects of depression: cross-sectional study. Sex Disabil 2014;32:197–204.
7 Haga HJ, Gram Gjesdal C, Irgens LM, Østensen M. Reproduction and gynaecological manifestations in women with primary Sjögren’s syndrome: a case-control study. Scand J Rheumatol 2005;34:45–8.
8 Marchesoni D, Mozzanega B, Sandre PD et al. Gynaecological aspects of primary Sjögren’s syndrome. Eur J Obstet Gynecol Reprod Biol 1995;63:49–53.
9 Maddali Bongi S, Del Rosso A, Orlandi M, Matuscicerin M. Gynaecological symptoms and sexual disability in women with primary Sjögren’s syndrome and sicca syndrome. Clin Exp Rheumatol 2013;31:683–90.
10 Bloch K, Buchanan W, Wohl M, Bunim J. Sjogren’s syndrome. A clinical, pathological and serological study of sixty-two cases. Medicine 1965;44:187–231.
11 Capriello P, Barale E, Cappelli N, Lupo S, Teti G. Sjögren’s syndrome: clinical, cytological, histological and colposcopic aspects in women. Clin Exp Obstet Gynecol 1988;15:9–12.
12 van der Meulen TA, van Nimwegen JF, Harmsen HJ et al. Normal vaginal microbiome in women with primary Sjögren’s syndrome-associated vaginal dryness. Ann Rheum Dis 2019;78:707–9.
13 Robboy SJ, Mutter GL, Prat J et al., eds. Robboy’s pathology of the female reproductive tract. Edinburgh: Churchill Livingstone/Elsevier, 2009.
14 Skopouli FN, Papanikolaou S, Malamou-mitsi V, Papanikolaou N, Moutsopoulos HM. Obstetric and gynaecological profile in patients with primary Sjögren’s syndrome. Ann Rheum Dis 1994;53:569–73.
15 Lee SK, Kim CJ, Kim D-J, Kang J. Immune cells in the female reproductive tract. Immune Netw 2015;15:16–26.
16 Scurry J, Day T. Sjogren syndrome and non-Sjogren sicca syndrome: do they affect the vulva? Int J Gynecol Pathol 2018;37:152–3.
17 Pudney J, Quayle AJ, Anderson DJ. Immunological microenvironments in the human vagina and cervix: mediators of cellular immunity are concentrated in the cervical transformation zone. Biol Reprod 2005;73:1253–63.
18 Bachmann G. Urogenital ageing: an old problem newly recognized. Maturitas 1995;22(Suppl):S1–5.
19 Kyongo JK, Jespers V, Goovaerts O et al. Searching for lower female genital tract soluble and cellular biomarkers: defining levels and predictors in a cohort of healthy Caucasian women. PLoS One 2012;7:e43951.
20 Ahn SH, Monsanto SP, Miller C et al. Pathophysiology and immune dysfunction in endometriosis. Biomed Res Int 2015;2015:795976.
21 Sinaii N. High rates of autoimmune and endocrine disorders, fibromyalgia, chronic fatigue syndrome and atopic diseases among women with endometriosis: a survey analysis. Hum Reprod 2002;17:2715–24.
22 Shigesi N, Kvasikoff M, Kirtley S et al. The association between endometriosis and autoimmune diseases: a systematic review and meta-analysis. Hum Reprod Update 2019;25:486–503.
23 Hernández-Molina G, Michel-Peregrina M, Hernández-Ramirez DF, Sánchez-Guerrero J, Llorente L. Chemokine saliva levels in patients with primary Sjögren’s syndrome, associated Sjögren’s syndrome, pre-clinical Sjögren’s syndrome and systemic autoimmune diseases. Rheumatology 2011;50:1288–92.
24 Lee YJ, Scofield RH, Hyon JY et al. Salivary chemokine levels in patients with primary Sjögren’s syndrome. Rheumatology 2010;49:1747–52.
25 Aota K, Kani K, Yamanoi T et al. Distinct regulation of CXCL10 production by cytokines in human salivary gland ductal and acinar cells. Inflammation 2018;41:1172–81.
26 Traish AM, Botchevar E, Kim NN. Biochemical factors modulating female genital sexual arousal physiology. J Sex Med 2010;7:2925–46.
27 Bartoloni E, Alunno A, Bistoni O et al. Characterization of circulating endothelial microparticles and endothelial progenitor cells in primary Sjögren’s syndrome: new markers of chronic endothelial damage? Rheumatology 2015;54:536–44.
28 Alunno A, Ibba-Manneschi L, Bistoni O et al. Angiogenic T cells in primary Sjögren’s syndrome: a double-edged sword? Clin Exp Rheumatol 2019;37(Suppl 118):S36–41.
29 Gerli R, Vaudo G, Becci EB et al. Functional impairment of the arterial wall in primary Sjögren’s syndrome: combined action of immunologic and inflammatory factors. Arthritis Care Res 2010;62:712–8.
30 Priddar T, Tikz C, Özkaya S et al. Endothelial dysfunction in patients with primary Sjögren’s syndrome. Rheumatol Int 2005;25:536–9.
31 Thacker SG, Berthier CC, Mattinzoli D et al. The detrimental effects of IFN-α on vasculogenesis in lupus are mediated by repression of IL-1 pathways: potential role in atherosclerosis and renal vascular rarefaction. J Immunol 2010;185:4457–69.
32 Buie JJ, Renaud LL, Muise-Helmericks R, Oates JC. IFN-α negatively regulates the expression of endothelial nitric oxide synthase and nitric oxide production: implications for systemic lupus erythematosus. J Immunol 2017;199:1979–88.
33 Levin RJ. The ins and outs of vaginal lubrication. Sex Relationship Ther 2003;18:509–13.
34 Dwyer PL. Skene’s gland revisited: function, dysfunction and the G spot. Int Urogynecol J 2012;23:135–7.
35 Patton DL, Thwin SS, Meier A et al. Epithelial cell layer thickness and immune cell populations in the normal human vagina at different stages of the menstrual cycle. Am J Obstet Gynecol 2000;183:967–73.