Lesions of the subepithelial stromal zone of the lower female genital tract. An immunopathological study

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Summary

The stroma of ten fibroepithelial and ten adenomatous cervical polyps, 12 fibro-epithelial vaginal polyps, 20 non neoplastic cervices, 15 cone specimens with CIN changes and five specimens with infiltrative squamous cell carcinoma was investigated. Vimentin, desmin, actin, collagen III, CD34, ER and PgR were studied using the Ventana Automatic Immunostaining System. A subepithelial stromal layer was detected in 17/20 normal specimens, 16/20 cervical polyps, 10/12 vaginal polyps and in 6/15 cone biopsies but not in neoplastic specimens. Vimentin staining was positive in all fibroepithelial polyps, normal specimens and in 6/10 adenomatous polyps. Actin was negative in the loose subepithelial zone but positive in the underlying stroma. Desmin, alpha-l-antitrypsin and CD34 were detected in isolated cells in normal specimens. ER and PgR were observed in isolated subepithelial cells, with greater density in the cells of the fibromuscular wall. In conclusion, loose mesenchymal stroma in the lower female genital tract of a fibrohistiocytic nature was observed with minimal smooth muscle participation - which is the matrix of vaginal and cervical polyps.

Key words: Vagina; Cervix; Histology; Immunohistochemistry; Polyps; Vagina.

Introduction

The stroma of the lower female genital tract is typically described as fibromuscular, consisting mainly of smooth muscle fibres and connective tissue. The study of Elliott and Elliott [1], however, has drawn attention to the presence of a 0.5-5 cm wide subepithelial stromal zone which runs from the endocervix to the vulva in mature females. There is evidence that this stromal zone may be the origin of fibroepithelial cervical and vaginal polyps, known as pseudo sarcomas because of the commonly reported presence of isolated large atypical cells [2, 3].

The purpose of this study was to investigate the morphology, histochemical and immunohistochemical properties of normal cervical stroma as well those of cervical and vaginal polyps and neoplastic processes of the cervix.

Materials and Methods

Cases for this study were obtained from the files of the Pathology Laboratory of Aretaieion University Hospital during the last decade. The following specimens were included in the study: ten fibroepithelial and ten adenomatous cervical polyps, 12 fibroepithelial vaginal polyps, 15 cone biopsies performed for cervical intraepithelial neoplasias, five cervices with squamous cell carcinoma and 20 cervices from total hysterectomies due to uterine leiomyomata. All tissues for light microscopy were formalin-fixed and paraffin-embedded and processed for hematoxylin and eosin stains and elastic van Gieson's stain. Immunohistochemical procedures were performed by the Ventana Automatic Immunostaining System and the following antibodies were used: anti-vimentin (V9mab Eurodiagnostics) anti-desmin (D33mab Novosan) anti-smooth muscle actin (IA4 mabBiogenex, a-1-antitrypsin (pAb, Thermo), anti-collagen III (III-53mab, MediCorp), anti-CD34 (QBend mab, Novokasta,) ER (6F11Novokastra mab), PgR (IA6, Novocasta mab).

Internal controls, which included normal epithelium, fibrous tissue, smooch muscle and other specific tissues were assessed for immunostaining.

Results

Clinical data. The ages of the patients ranged from 30 to 70 years (mean 48 years) for the group of 25 total hysterectomies and from 24 to 47 years (mean 36 years) for the remaining patients with polyps, and dysplastic and neoplastic lesions of the cervix. Nine patients from the first group and one patient from the second were postmenopausal. None of the patients was pregnant, or had a history of hormonal therapy. All vaginal polyps and seven of the cervical polyps were discovered due to postcoital bleeding. The rest were discovered during routine examination. The size of the polyps varied from 6 to 35 mm.

Three of the 15 patients with dysplastic epithelial changes, in whom cone biopsies showed foci of microinvasive carcinomas, underwent more extensive surgical therapy. Three to five years follow-up was obtained in these patients and no recurrences were observed.

Hysterectomy specimens showed uterine leiomyomata and the cervices were grossly normal. All cases with infiltrating squamous cell carcinomas were Stage 1 and appropriate therapeutic procedures were followed.

Microscopic features. A loose sub epithelial stromal layer, 2 to 6 mm in width, was detectable in both the endocervix and the ectocervix (Figure 1) in 17/20 normal cervical specimens, 16/20 cervical polyps, 10/12 vaginal polyps and in 6/15 cone biopsies, but not in neoplastic

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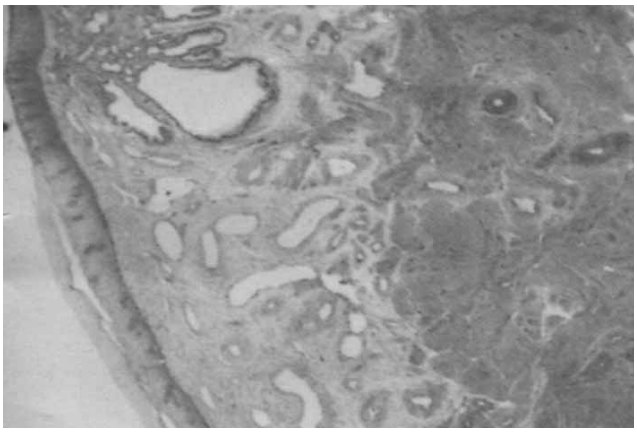

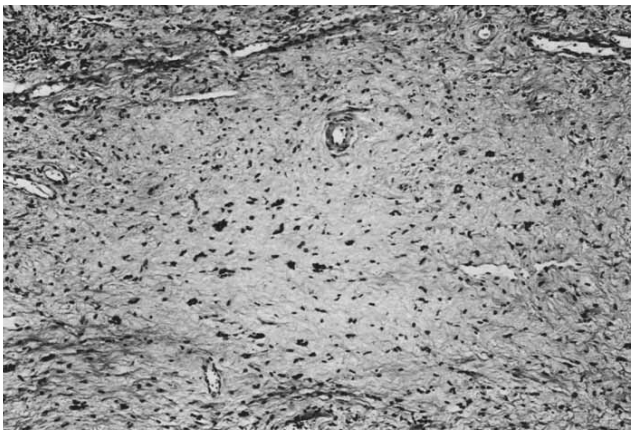

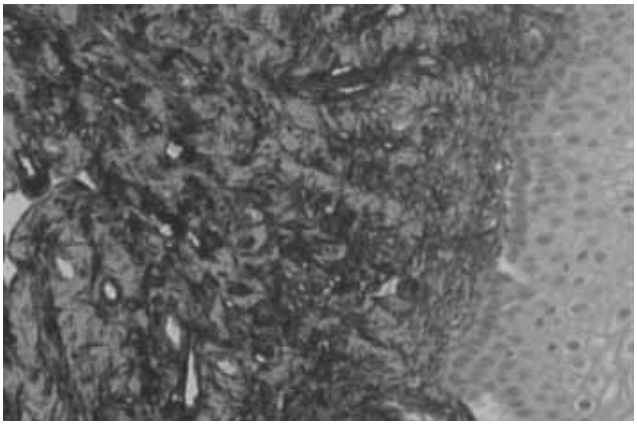

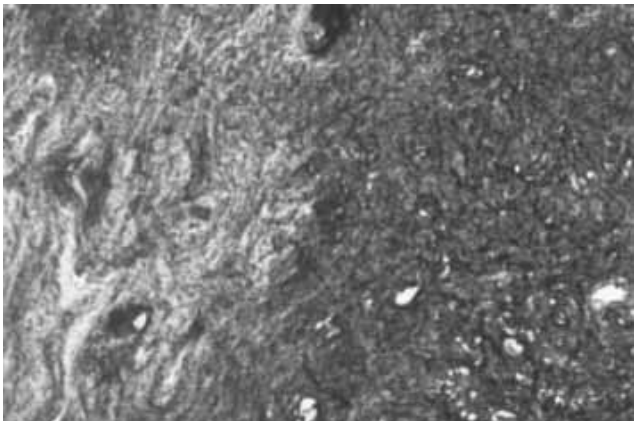

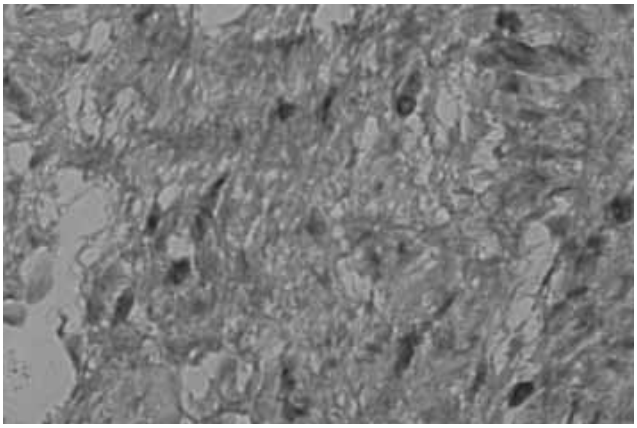

Figure 1. — Histological section of cervix showing loose subepithelial stroma over the fibromuscular wall (hematoxylin-eosin, x 25).

Figure 2. — Histological section of a fibroepithelial vaginal polyp showing atypical and giant cells in loose stroma (hematoxylin-eosin, x 40).

Figure 3. — Positive vimentin immunostain reaction in the subepithelial cervical stroma (immunostain, x 120).

Figure 4. — Positive desmin immunoreaction, focal in the subepithelial stroma and dense in the fibromuscular wall (immunostain x 120).

Figure 5. — Histological section of cervix showing positive immunostain reaction to ER in isolated stromal cells (immunostain x 250).

specimens. This zone was more evident in the ectocervix and thinner in the endocervix. In three fibroepithelial cervical polyps, rare spindle-shaped cells with atypical nuclear forms were observed. Stromal mitoses were absent. A deeper zone consisting of fibromuscular tissue was also present in the cervical wall. In three normal cervices the existing chronic inflammatory reaction covered the sub epithelial stroma.

The same loose structure was focally observed in the stroma of 6/10 adenomatous cervical polyps and focally in the stroma of six cone biopsies where a co-existing inflammation covered the stroma. The specimens with microinvasive and the infiltrative carcinomas showed an inflammatory and/or focal fibrous stromal reaction which obscured the subepithelial stromal layer.

The vaginal polyps were well circumscribed or grossly papillary and covered by stratified squamous epithelium, with slight hyperplasia, sometimes with hyperkeratosis and parakeratosis, but no dysplasia. The core of these polyps was composed of a loose edematous matrix, quite similar to the subepithelial stromal layer previously described, with varying numbers of spindle and stellate-shaped cells and occasional giant and multinucleated-cells (Figure 2).

Histochemical findings. Histochemical staining revealed loose fibro-connective tissue and collagen fibers

running parallel to the epithelial surface. Fine elastic fibers were also observed running perpendicular to the epithelial surface in the subepithelial zone, and parallel to the surface in the deeper layers.

Immunohistocbemical findings. Vimentin staining was positive in normal cervices and the stromal cells of all fibro epithelial polyps (Figure 3) and in 6/10 adenomatous polyps. In the vaginal polyps, in particular, the stromal spindle or stellate cells were strongly positive for vimentin and focal condensation of these cells under the surface epithelium was evident. Collagen III staining was detected only in scanty fibrils but a diffuse reaction of the stroma was observed in all cases.

Immunostaining for actin was negative but desmin was positive in the loose sub epithelial zone, observed in scanty spindle-like cells. Both markers were markedly positive in the fibromuscular portion of the cervix (Figure 4). Alpha-1-antitrypsin was detected in a few isolated stromal cells and CD34 was positive in stromal cells around vessels. Estrogen receptors were observed only sparsely in a few stromal cells (Figure 5) and markedly in the fibromuscular wall. Similar results were focally observed in specimens with neoplastic changes, but the inflammatory and fibrous reaction obscured the characteristic distribution described above.

Discussion

It has been postulated that two different kinds of stroma exist in the lower female genital tract – a fibromuscular stroma, which is the main substrate of the cervical and vaginal wall, and a loose subepithelial myxoid stroma, first described by Elliott and Elliott [1]. The latter runs from the endocervix to the vulva in mature females, and according to our findings was usually thicker in the ectocervix. It is composed of loose fibroconnective tissue with fibroblasts, elastic fibers, type III collagen, and reticulin fibers.

The physiologic significance of the subepithelial zone is not very clear. Mucitelli *et al*. [4] suggest that the function of the zone may be to help the immense expansion of the vulvovaginal area during labor and its rapid reconstitution during puerperium. Ultrastructurally, large stromal cells have the distinctive features of an active contractile apparatus and may provide the necessary shrinkage of cervicovaginal tissue during puerperium [4].

The loose subepithelial zone, often populated by cells with bizarre nuclear features, may be the origin of the so-called fibroepithelial polyps of the vagina [1-4] and possibly of the cervix [5].

A striking similarity, both histological and immunohistochemical, of the stroma of vaginal polyps to that of the subepithelial zone found in normal vaginas has been reported [6].

Our study confirms the expression of vimentin, type III collagen and desmin in both vaginal polyps and the loose subepithelial zone.

The belief that polyps are the expression of a hyperplastic process arising in this zone is in agreement with our results.

The possible role of pregnancy and steroid hormones in this process has been argued by some investigators because 33% of fibroepithelial polyps of the vagina reported in the literature occurred in pregnant women and also because of steroid receptors detected in the atypical stromal cells.

None of our patients was pregnant or receiving any hormone therapy. It is interesting that we detected a loose subepithelial stromal zone not only in younger but also in elderly patients, and that the microscopic and immunohistochemical features of the zone were similar independently of age.

The histogenesis of the atypical and multinucleated giant cells in fibroepithelial polyps remains unclear. Some investigators have proposed an origin either from smooth muscle cells or from fibroblasts-histiocytes. A possible differentiation of multifunctional mesenchymal elements along two cell lines (divergent histogenesis) has also been proposed [4].

Based on immunohistochemical studies, Ostor *et al*. [3] concluded that the stromal cells were of fibrohistiocytic origin. On the contrary, Muticelli *et al*. [4] supported the mesenchymal nature of those cells.

Rollason *et al*. [8], using electron microscopic examination, showed that fibroepithelial polyps had the ultrastructural features usually associated with myofibroblasts and concluded that these tumors would be better designated polypoid myofibroblastomas. Other investigators [9] proposed that adenomatous polyps may develop as a result of a granulation tissue reaction after some local injury of the vaginal mucosa. According to the same authors, delayed differentiation of myofibroblasric cells may explain why granulation tissue sometimes does not shrink but becomes a polyp.

In conclusion, the results of our study indicate that in the lower female genital tract there exists a loose subepithelial layer rich in fibroblasts, elastic and reticulin fibers characteristically arranged, which may be the origin of fibroepithelial vaginal polyps and possibly of commonplace cervical polyps.

References

[1] Elliott G.B., Elliott J.D.A.: "Superficial stromal reaction of the lower female genital tract". *Arch. Pathol.,* 1973, *95*, 100.
[2] Chirayil S.J., Tobon H.: "Polyps of the vagina: a clinicopathological study of 18 cases". *Cancer,* 1981, *47*, 2904.
[3] Ostor A.G., Fortune D.W., Riley C.B.: "Fibroepithelial polyps with atypical stromal cells (Pseudosarcoma botryoides) of vulva and vagina: a report of 13 cases". *Int. J. Gynecol. Pathol.,* 1988, *7*, 351.
[4] Mucitelli D.R., Charles E.Z., Kraus F.T.: "Vulvovaginal polyps: histologic appearance, ultrastructure, immunohistochemical characteristics and clinicopathologic correlations". *Int. J. Gynecol. Pathol.,* 1990, *9*, 20.
[5] Lement P.B.: "Miltinucleated stromal giant cells of the uterine cervix". *Arch. Pathol. Lab. Med.,* 1985, *109*, 200.
[6] Al-Nafussi A.I., Rebello G., Hughes D., Blessing K.: "Benign vaginal polyps: a histological, histochemical and immunohistochemical study of 20 polyps with comparison to normal vaginal subepithelial layer". *Histopathology,* 1992, *20*, 145.

[7] Hartmann C.A., Sperling M., Stein H.: "So-called fibroepithelial polyps of the vagina, exhibiting an unusual but uniform antigen profile characterized by expression of desmin and steroid hormone receptors but no muscle-specific action on macrophage markers". *Am. J. Clin. Pathol*., 1990, *93,* 604.
[8] Rollason T.P., Byrne P., Williams A.: "Immunohistochemical and electron microscopic findings in benign fibroepithelial vaginal polyps". *J. Clin. Pathol*., 1990, *43*, 224.
[9] Halvorsen T.B., Johannesen E.: "Fibroepithetial polyps of the vagina: are they old granulation tissue polyps?". *J. Clin. Pathol*., 1992, *45*, 235.

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