MORPHOLOGIC ASPECTS OF THE SENESCENCE PROCESSES IN FEMALE GENITAL SYSTEM

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Abstract: Senescence is characterized by a gradual loss of the stem cells activity and an atrophy of all cells and tissues which lead to the deterioration of the functional activity. The senescence processes manifest also in the female genital organs, producing different morphologic alterations. For the evidentiation of these aspects we retrospectively analyze 75 cases, from menopausal and postmenopausal women, investigating the histerectomy specimens, bioptic curettages and cervico-vaginal smears. The specimens were paraffin-embedded and routinely fixed and stained with hematoxylin and eosin (H&E), and the smears were stained using Papanicolau technique. Histologic and cytologic alterations in uterus, ovaries and vagina were observed. There were seen changes of the endometrial glands and stroma, cystic atrophy. Also, it was emphasized the arteriosclerosis of myometrial arteries, with medial hypertrophy. The microscopic aspect of the atrophic endometrium is different in histerectomy specimens than in bioptic curettages. The endometrial cystic atrophy must be differentiated from glandulo-cystic hyperplasia with retrograde atrophy. The cervical squamous epithelium is atrophic, with little or no intracytoplasmic glycogen, and the endocervical glands developed small clusters called endocervical tunnel clusters. These histologic features must not be diagnosed as invasive well differentiated endocervical adenocarcinoma. It has been noticed the absence of primordial and maturing ovarian follicles, the absence of the luteal body (corpus luteum) and atretic follicles and the persistence of corpora albicantia, especially in medulla. In ovarian stroma, different proliferation degrees of the cortical and medular stromal cells were seen, with occasional luteinised stromal cells and cortical cysts. The cervico-vaginal smears presented atrophic features. The described cyto-morphologic aspects are due to insufficient estrogenic stimulation, as a consequence of the lack of ovarian hormone production.

INTRODUCTION

The aging process as well as hormonal insufficiency will ultimately occur in all women. Lifestyle changes often accompany these two processes. As the overall life expectancy of women increases, so does the amount of time that women will spend in the postmenopausal state of their lives (Lobo et al., 2007). Senescence is characterized by a gradual loss of the stem cells activity and an atrophy of all cells and tissues which lead to the deterioration of the functional activity. The senescence processes manifest also in the genital organs, producing different morphologic alterations. Atrophic changes of the vagina and vulva are the most frequent causes of genital complaints in the menopausal woman (Lobo et al., 2007). Estrogen loss from follicular depletion in the menopausal ovary is the major cause of vulvovaginal dysfunction in the older woman, as it accounts for most of the anatomic, cytologic, physiologic, and bacteriologic genital changes that occur. In the vagina, atrophy yields a thin mucosa susceptible especially to infections. There are several morphologic manifestations of atrophic endometrium, but all have in common a thin mucosa that measures about half or less of the thickness of a basal layer of cyclic endometrium (less than 0.5 mm) (Kurman, 2002). The atrophic features of uterine cervix can be cytologic and histologic assessed, on cervico-vaginal smear and on biopsy or histerectomy specimens. Although ovarian stroma typically increases in volume from the fourth to the seventh decades, in postmenopausal women it has a wide spectrum of appearances (Rosai, 2004). Women may start to experience subtle signs of changing estrogen levels early in the perimenopausal years. Although atrophic changes of the female genital organs are unlikely to resolve spontaneously, they are reversed when estrogen therapy is initiated (Lobo et al., 2007).

MATERIAL AND METHODS

For the evidentiation of these atrophic changes we retrospectively analyze 75 cases, from menopausal or postmenopausal women, between 58 and 74 years old. Clinically, the pacients had abnormal uterine bleeding, uterine prolapse, submucous leiomyomata, chronic endometritis, endometrial polyps. After clinical investigation, the pacients underwent total hysterectomy, bioptic curettages or cervico-vaginal smears, according to their symptomatology and diagnostic. The specimens were paraffin-embedded and routinely fixed and stained with hematoxylin and eosin (H&E), and the cervico-vaginal smears were stained using Papanicolaou technique.

RESULTS AND DISCUSSIONS

Histologic and cytologic alterations in uterus, ovaries and vagina were observed. The established modifications are in concordance with those described in the literature. The vaginal mucosa was thinner, with less layers of the epithelium, most of the pacients presenting a chronic

stromal inflammatory infiltrate as a consequence of the atrophy. After menopause, a gradual reduction in the thickness of the vaginal epithelium occurs, first with a loss of superficial cells followed by intermediate cells, such that the mucosa of late menopausal women may be reduced to only six to eight layers of parabasal cells. As a consequence, a normal postmenopausal atrophic pattern may be confused with a high-grade intraepithelial lesion unless care is taken to identify other nuclear abnormalities. In one study, the time required for a vaginal squamous cell to make the transition from progenitor cell through desquamation was about 5 days for both cycling and postmenopausal women (Averette et al., 1970). Because atrophic vaginitis yield similar local symptoms as lichen tuber, psoriasis, eczema, and lichen sclerosus, these dermatologic conditions must always be considered as a differential diagnosis. Infections such as candidosis may be superimposed and should also be considered in this context (Lobo et al., 2007). In one study, it was admitted that estradiol and body mass index (BMI) are associated with vaginal cell maturation and atrophy in postmenopausal women. Vaginal cell atrophy does not depend on the time since menopause (Repse-Fokter et al., 2008). There were seen changes of the endometrial glands and stroma, cystic atrophy. Also, it was emphasized the arteriosclerosis of myometrial arteries, with media hypertrophy (figure 4). The microscopic aspect of the atrophic endometrium was different in histerectomy specimens than in bioptic curettages. The endometrial cystic atrophy must be differentiated from glandulo-cystic hyperplasia with retrograde atrophy. An endometrium that is thick as early- to mid-proliferative phase, but is devoid of morphologic features of either active proliferation or secretion may be consider inactive as far as its response to hormonal stimuli is concerned (Kurman, 2002). The glands and stroma resemble proliferative endometrium, but the glands are usually oriented parallel rather than perpendicular to the surface epithelium. The surface epithelium as well as that lining the glands is columnar to cuboidal and contains pseudostratified nuclei without mitoses, and occasional ciliated cells are seen. The stroma is generally dense throughout, without a clear-cut separation between the basalis and the functionalis layes. Nuclear DNA synthesis and E_2R (estrogen receptors) are maintained in the senescent or inactive but not in the severly atrophic endometrium (Press et al., 1984). This phenomenon explains why exogenous estrogens can ''revitalize'' the inactive endometrium and both glandular and stromal cells acquire receptors for P (progesteron). As a result, unopposed estrogens in the menopause may lead to hyperplasia, whereas P therapy either may convert hyperplasia to secretory endometrium or prevent the development of hyperplasia (Ferenczy, Gelfand, 1989). There are several morphologic manifestations of atrophic endometrium, but all have in common a thin mucosa that measures about half or less of the thickness of a basal layer of cyclic endometrium (less than 0,5 mm). There is a further decrease in the number and volume of glands and stroma. The stroma is often collagenized and resembles the stroma of the isthmus or lower uterine segment in premenopausal women (Kurman, 2002). Endometrial vascular alterations are seldom seen in atrophic endometrium, like arteriosclerosis of the myometrial arteries, include the arcuate and radial arteries, with medial hypertrophy and calcification and narrowing of the lumen. This condition, when associated with cardiovascular collapse may lead to hemorrhagic necrosis of the endometrial mucosa, producing apoplexia uteri (Daly, Balogh, 1968). Atrophic endometrium often has cystically dilated glands, aspect which was found also in our cases (figure 2). This condition is referred to as cystic atrophy. The endometrial mucosa retains the thickness of an otherwise active hyperplasia, but the glandular epithelium is atrophic and the stroma is collagenized. On occasion, both the surface and glandular epithelium is composed of tall columnar to cuboidal cells, including ciliated cells resembling those seen in hyperplasia. Unlike true hyperplasia, this form of atrophy lacks mitotic figures, the mucosa is

thin and the stroma is relatively rich in collagen fibers. It is possible that the changes reflect the estrogenic response of otherwise atrophic endometrial epithelium that has been under either endogenous or exogenous estrogenic stimulation (Rosai, 2004). There were 6 cases of extreme endometrial atrophy, with endometrial stromal fibrosis (figure 1), and only the surface epithelium and rare glands remain lined by low cuboidal cells. In response to long-standing irritation by the chronic inflammatory exudates, the surface epithelium may undergo squamous metaplasia, which in extreme cases lines the entire endometrial cavity resulting in the condition reffered to as ichthyosis or psoriasis uteri (Kurman, 2002). In a prospective nonrandomized study on 752 postmenopausal women with abnormal uterine bleeding (AUB), it was emphasized that hysteroscopy is a significantly more accurate diagnostic method for the detection of focal abnormalities of the endometrium than transvaginal ultrasonography (TVS), for all patients with an endometrial thickness less than 4 mm (Tinelly et al., 2008). Another study revealed that endometrial microcalcifications are histologically heterogeneous and are associated with older patient age, postmenopausal state, atrophic endometrium, and endometrial polyps. Those found incidentally by means of pelvic ultrasonography, in author's experience, did not portend malignancy (Truskynovsky et al., 2008). The cervical squamous epithelium was atrophic, with reduced or no intracytoplasmic glycogen. Surface epithelial maturation and stromal papillae were absent. These cellular alterations should not be confused with cervical intraepithelial neoplasia. The atrophic epithelial covering does not adequately protect the subepithelial vasculature against trauma, a situation that frequently leads to bleeding and inflammation (Rosai, 2004). The endocervical glands appear in histologic sections as distinct clusters of as many as 50 small glands. Connections between these glands and the endocervical surface may not be apparent. The glands in these clusters are often distended by inssipated mucus. Because of the destension of the glands by mucus, the lining columnar epithelium is frequently flattened, Fluhmann naming them as endocervical tunnel clusters, aspect which was often described in our cases (figure 3) (Kurman, 2002). These histologic features must not be diagnosed as invasive well differentiated endocervical adenocarcinoma. In older and postmenopausal women, the functional squamocolumnar junction is nearly always located above the external os, because it moves continue throughout the reproductive years (Kurman, 2002). Regarding ovarian changes, our results are coherent with the reports of other researchers. It has been noticed the absence of primordial and maturing ovarian follicles, the absence of the luteal body (corpus luteum) and atretic follicles and the persistence of corpora albicantia (figure 5), especially in medulla. In ovarian stroma, different proliferation degrees of the cortical and medular stromal cells were seen, with occasional luteinised stromal cells, cortical cysts. Occasional primordial follicles, however, may persist for several years after cessation of menses, accounting for sporadic ovulation and follicle cyst formation accompanied by postmenopausal bleeding (Kurman, 2002). After this period, the only follicle-derived structures tipically encountered are occasional unresorbed corpora fibrosa and corpora albicantia, the latter typically within the medulla. Although ovarian stroma usually increases in volume from the fourth to the seventh decades (Snowden et al., 1989), the ovarian stroma in postmenopausal women has a wide spectrum of appearances (Loubet et al., 1984; Snowden et al., 1989). At one extreme there is stromal atrophy manifested by a thin cortex and minimal amounts of medullary stroma, which becomes less cellular because of an increase in intercellular collagen, and its cells have smaller, darker, more inactive appearing nuclei. At the other extreme, there is marked stromal proliferation warranting the designation "stromal hyperplasia" to connote a pathologic process. Usually, there are varying degrees of nodular or diffuse proliferation of cortical and medullary stromal cells, that lie

between these two extremes. Ovarian stromal changes that can be considered normal aging phenomena include occasional luteinized stromal cells, broad irregular areas of cortical fibrosis of fibromatous nodules, cortical granulomas, spherical hyaline scars and surface papillary stromal proliferations (Lobo et al., 2007). Less commonly, focal decidual transformation of the ovarian stroma may be seen in otherwise normal ovaries. There were also described surface epithelial inclusion glands and cysts within the cortex and mild degrees of hilus cell hyperplasia. The medullary blood vessels exhibit a greater tortuosity, appearing more numerous and closely packed as a result of parenchymal atrophy, and should not be mistaken for a hemangioma on microscopic examination (Kurman, 2002). Many of these vessels may be calcified or have thickened walls and narrowed lumens as a result of mural deposition of a hyaline, amyloid-like material. Despite age related atrophic changes, the postmenopausal ovary is not devoid of hormonal activity. In one study it was revealed that postmenopausal ovary produces trace quantities of steroid hormones, mainly androgens, confirming the presence of steroid receptors and activity of main enzymes involved in steroidogenesis process (Laszczynska et al., 2008). Concordant with the literature, the cervico-vaginal smears presented atrophic features, like flat, monolayer sheets of parabasal-like cells with preserved nuclear polarity, generalized nuclear enlargement, up to three to five times the area of an intermediate cell nucleus, sometimes with a slight increase in nuclear/cytoplasmic ratio. The chromatin was uniformly distributed; there were naked nuclei due to autolysis. An abundant inflammatory exudate and basophilic granular background that resembles tumor diathesis was present in 8 cases (figure 6). Globular collections of basophilic amorphous material (blue blobs) reflect either degenerated parabasal cells or inspissated mucus. Atrophy is a normal aging phenomenon with a wide spectrum of cellular changes and variable amounts of inflammation (Lobo et al., 2007). Reporting of atrophic changes is variable and nonreproducible. However, atypical cellular changes associated with atrophy or atrophy with inflammation may warrant an interpretation of atypical squamous cells (ASC). Multinucleated histiocytes are frequently observed in specimens from postmenopausal women in the absence of inflammation (Solomon, Nayar, 2004). To determine whether postmenopausal asymptomatic women with normal endometrial cells in their smear are at higher risk for endometrial pathology compared with women without these cells, the researchers determine the histologic and otherwise cytologic follow-up outcome of 29,144 asymptomatic postmenopausal women. It was revealed that neither age nor hormone use or elevated maturation index showed significant impact on the outcome. Asymptomatic postmenopausal women with normal endometrial cells in their smear are at significant higher risk for (pre)cancerous endometrial lesion than women without these cells. These cases should be reported to the physician with an explicit comment that normal endometrial cells in a smear of a postmenopausal woman is an abnormal finding, possibly associated with significant endometrial pathology. It raises the question whether further gynecological examination would be more appropriate (Siebers et al., 2006).
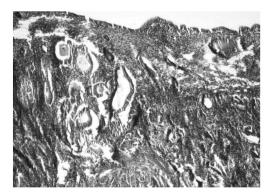
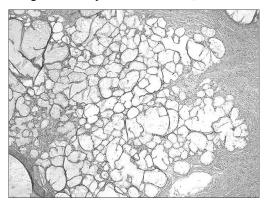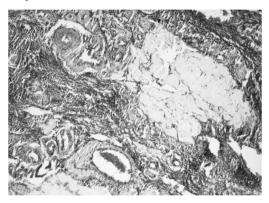Figure 1. Atrophic endometrium, HE X 10

Figure 2. Dilated endometrial gland in atrophic endometrium, HE X 10

Figure 3. Endocervical tunnel cluster, HE X 5

Figure 4. Medial hypertrophy of myometrial artery, HE X 10

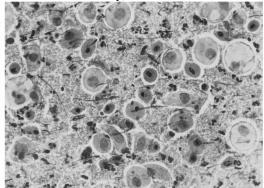Figure 5. Corpus albicans in ovarian medulla, HE X 10

Figure 6. Atrophic vaginitis, parabasal cells, Pap X 40

CONCLUSIONS

The described cyto-morphologic aspects are due to insufficient estrogenic stimulation, as a consequence of the lack of ovarian hormone production.

An accurate understanding of the endocrine changes with the morhpologic implications occurring during this phase of the reproductive life cycle has significant therapeutic and diagnostic implications.

Because these atrophic changes respond favorably to estrogen therapy, hormonal and nonhormonal treatments can provide patients with relief and a return to their previous level of function.

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