Cigarette smoking and the degree of maturation of the vaginal squamous epithelium in postmenopausal women

D. Karamanidis[1], D. Tamiolakis[2], Ger. Koutsougeras[2], Ch. Tripsanas[1], K. Retzos[1], S. Karidis[1], B. Liberis[1]

[1]Department of Obstetrics and Gynecology, General Regional Hospital of Alexandroupolis
[2]Department of Cytology, General Regional Hospital of Alexandroupolis, Alexandroupolis (Greece)

Summary

To determine the effects of cigarette smoking on vaginal squamous epithelium in postmenopausal women, we studied the vaginal smear patterns of 199 healthy postmenopausal non-smokers and 41 healthy postmenopausal smokers, with a mean age of 56 years. A statistically significant difference to the hazard of smokers was found in the percentage of smears manifesting absence of maturation of vaginal squamous cells. A high incidence of atrophic-type vaginal smears in the group of smokers was also found independent of postmenopausal age. In the group of non-smokers, there was a statistically significant difference between the cytologic patterns of smears of women who were in the early postmenopausal age (<10 years) and those many years after (≥10 years). Finally our data suggest that smokers had an earlier menopause, on average 2.4 years sooner than non-smokers. Cigarette smoking has an effect on vaginal squamous epithelium, but pathophysiology still remains unclarified.

Key words: Menopause; Vaginal maturation; Vaginal smear; Smoking.

Introduction

Hormonal activity in women begins changing during perimenopausal age due to cessation of the cyclic ovarian function. Clinical and cytologic menopause do not necessarily happen together. As the woman enters clinical menopause she may display a smear pattern manifesting varying degrees of continuing hormonal activity [1], and may reveal cyclic ovarian function.

Three basic cytologic patterns at menopause may be encountered: Estrogenic, intermediate and atrophic pattern.

In a proportion of postmenopausal examinees there is a high level of maturation of squamous cells in their vaginal smears. This fact likely correlates with adrenal cortex function [2, 3] and positively with sexual activity and perhaps even sexual response of postmenopausal women [4]. There is also evidence that a proportion of menopausal women never show complete atrophy due to obesity, hypertension, diabetes, digitalis treatment, tetracycline topical use and hepatic failure [1].

The purpose of this project was to determine if cigarette smoking effects the maturation of the vaginal squamous epithelium in postmenopausal women.

Materials and Methods

At the General District Hospital of Alexandroupolis, 240 cases of postmenopausal healthy women aged 47 to 61 years (mean age 56 years) were examined clinically and cytologically. These females were separated according to smoking habit into 199 non-smokers and 41 smokers, and according to the years in menopause into cases that were from 1-9 years in menopause and cases ≥10 years in menopause. From the group of non-smokers 125 cases were 1-9 years in menopause and 74 cases ≥10 years in menopause. From the group of smokers 26 cases were 1-9 years in menopause and 15 cases ≥10 years in menopause.

The gynecologists obtained smears for Pap tests, and material for the hormonal assessment of squamous epithelium taken from the proximal portion of the lateral wall of the vagina, with attention being paid to avoid contamination with cervical epithelium [5]. All samples were cytospray fixed and then stained using the Papanicolaou method. The slides were examined by means of an Olympus Bx40 C10 ocular lens and 40 objective lens microscope.

To evaluate the hormonal milieu of these smears, the maturation value (MV) of Meisels was used [6]. This is calculated by classifying 100 cells and assigning a value of 1 to superficial cells, 0.5 to intermediate cells and 0 to parabasal cells, then adding these values together (Table 1). This method yielded a single number to compare degrees of squamous maturation. The correspondence between estrogenic, intermediate, atrophic patterns and maturation value was MV ≥ 80, 50 < MV < 80, MV ≤ 50, respectively.

Statistical analysis of the data was performed with the SPSS (Statistical Package for Social Sciences) for Windows. The Student's t-test was applied to compare means between the two groups and the Chi-Square test was used to determine if postmenopausal smokers and non-smokers had the same cytological patterns.

Results

The results for postmenopausal smokers and non-smokers are summarized in Tables 2 and 3. Comparison of MV for smokers and non-smokers showed a markedly lower average value for smokers that was statistically

Revised manuscript accepted for publication May 25, 2001

significant. These data suggest that smokers not only had lower maturation of vaginal squamous cells, but also had an earlier menopause, on average by 2.4 years sooner than non-smokers.

From the cases of non-smokers, 4.52% displayed estrogen-type smear patterns (MV ≥ 80), 31.16% intermediate-type smear patterns (50 < MV < 80) and 64.32% atrophic type smear patterns (MV ≤ 50). From the cases of smokers, 2.44% displayed estrogen-type smear patterns, 14.64% intermediate-type smear patterns and 82.93% atrophic-type smear patterns. Analysis of these results revealed a statistically significant difference between smokers and non-smokers to the disadvantage of smokers who displayed a high percentage of atrophic smears ($p < 0.0001$).

The cytologic pattern of vaginal smears changed from estrogen-type to atrophic type in the late postmenopausal age of non-smokers (Table 4). Postmenopausal smokers displayed a high percentage of atrophic smears independent of the duration of menopause (Table 5).

Table 1. — *Maturation value.*

MV	MI
95	01/10/90
50	05/90/5
5	90/10/0

MV: Parabasal x 0 + Intermediate x 0.5 + Superficial x 1. MI: Maturation Index.

Table 2. — *Description of clinical and cytological characteristics in the two groups.*

	Non-smokers (n=199)	Smokers (n=41)
Mean age	56.2 (47-61)	55.02 (47-60)+
Mean age at onset of menopause	50.2 (46-57)	47.8 (46-52)°
Mean years in menopause	6.38 (1-15)	6 (1-13)+
Mean MV	47.54 (16-98)	38.22 (10-89)°

Numbers in () are ranges; *p<0.001; +not significant.

Table 3. — *Postmenopausal non-smokers and smokers and maturation value.*

	MV≥80	50<MV<80	MV≤50	Total
Nonsmokers	9 (4.52%)	62 (31.16%)	128 (64.32%)	199
Smokers	1 (2.44%)	6 (14.64%)	34 (82.93%)	41

Chi square = 123.82; p<0.0001.

Table 4. — *Postmenopausal non-smokers. Maturation value and duration of menopause.*

Years in menopause	MV≥80	50<MV<80	MV≤50	Total
<10	8 (6.4%)	40 (32%)	77 (61.6%)	125
≥10	1 (1.35%)	22 (29.73%)	51 (68.92%)	74

Chi square = 23.004; p<0.0001.

Table 5. — *Postmenopausal smokers. Maturation value and duration of menopause.*

Years in menopause	MV≥80	50<MV<80	MV≤50	Total
<10	1 (3.85%)	4 (15.38%)	21 (80.77%)	26
≥10	0 (0%)	2 (13.33%)	13 (86.67%)	15

Chi square = 5.048; p=0.08 (not statistically significant).

Discussion

Menopause is – strictly speaking – the arrest of menstruation. Most women however do not become amenstrual rapidly between one month and the next, but rather the changes occur slowly and may continue over several years. Cytology is performed to evaluate the endocrine activity in menopausal women. The degree of maturation of the vaginal squamous epithelium is hormone-dependent. Expectedly, at a time when hormone supply stops there is no uniformity in smear patterns displayed from different women, moreover even from the same woman in different months [7]. The MV system gives a single figure from 0 to 100. As a rule, baseline smears at menopause show complete atrophy (MV<50).

Several women have at first scarce, and later, more frequent unovulatory cycles characterized by abnormal estrogen supply and absence of secretory changes. These factors are cytologically manifested by an irregular caryotypic index, curves and absence of progesterone effects [8]. As we observed in this study, healthy females who have entered clinical menopause display varying menopausal smear patterns. We believe that cigarette smoking is one of the factors responsible for these differentiating degrees of maturity.

When menstrual arrest is established, the most significant hormonal alteration is decreased estradiol supply. Androstenedione supply is less significantly decreased due to the adrenal cortex function. This hormone converses to estrone. The decrease of estradiol supply and the less significant decrease of estrone result in an alteration of the ratio of estradiol: estrone > 1 to < 1. Similarly the relation of androgens: estrogens is altered [9]. With increasing age, the adrenal contribution of precursors for estrogen production proves inadequate. In this final stage of estrogen availability, levels are insufficient to sustain secondary sex tissues and atrophy of the vaginal mucosal surfaces takes place many years after initiation of menopause.

There is evidence that postmenopausal female smokers have higher serum levels of triglycerides, LDL cholesterol and apolipoprotein B, higher ratios of LDL:HDL cholesterol and apolipoprotein B: A-I but lower serum levels of HDL cholesterol and apolipoprotein A1 [10]. The more atrophic smear patterns in smokers could be explained by the suggestion that cigarette smoking induces increased androgen serum levels in early postmenopausal women [11]. Higher serum levels of 11-hydroxyandrosterone were reported in healthy postmenopausal smokers and this finding confirms previous evidence that smoking does not affect serum estradiol in pre- or postmenopausal women but gives no support to preceding data of increased serum levels of the same adrenal steroids in postmenopausal female smokers [12]. Serum androstendione and DHEAS concentrations were higher in female smokers than in non-smokers, the difference being statistically significant only in postmenopausal examinees. One possible mechanism is an enzymatic block by components of cigarette smoke. Acute adrenocorticotropin stimulation of postmenopausal smokers did

not trigger abnormal elevations of precursor steroids [13, 14]. In a more recent study, there is evidence linking cigarette smoking with a generalized incompatibility in adrenal cortical hormone levels. However there is no data for a partial block in the cortisol synthesis pathway to explain the increased adrenal androgen levels in smokers [15]. Another observation of this study, confirming previous reports, supports the assumption that smoking increases the risk of early menstrual arrest [16-18].

Conclusively our results suggest that smoking is linked with cytologic menopause (atrophic smear patterns), independent of clinical menopause, and with early menstrual arrest. Perhaps this effect is due to elevated levels of androstendione and DHEAS in female smokers and a possible theory to explain this is an enzymatic block due to cigarette smoke components. Still, more research is needed to assess the influence of cigarette smoking in the vaginal squamous epithelium of postmenopausal females.

References

[1] Koss L. G.: "Diagnostic Cytology and its Histopathologic Bases". 4th ed., Philadelphia, J.B. Lippincott Company, 1992, 276.

[2] De Waard F., Baanders-van Halewijn E. A.: "Cross-sectional data on estrogenic smears in post-menopausal population". *Acta Cytol.*, 1969, *13*, 675.

[3] De Waard F., Pot H., Tonokens-Nanninga N. E., Baanders-van Halewijn E. A., Thijssen J. H.: "Longitudinal studies on the phenomenon of postmenopausal oestrogen production". *Acta Cytol.*, 1972, *16*, 273.

[4] Leiblum S. *et al.*: "Vaginal atrophy in the postmenopausal woman. The importance of sexual activity and hormones". *JAMA*, 1989, *249*, 2195.

[5] Soost H.: "Comparative studies on the degree of proliferation of the vaginal and ectocervical epithelium in the hormonal evaluation of a patient by means of exfoliative cytology". *Acta Cytol.*, 1960, *4*, 199.

[6] Meisels A.: "The maturation value". *Acta Cytol.*, 1967, *11*, 249.

[7] Kalogeraki A. *et al.*: "Cigarette smoking and vaginal atrophy in postmenopausal women". *In Vivo*, 1990, *10*, 597.

[8] Wachtel G. E.: "Exfoliative Cytology in Gynecological Practice". (2nd ed.), London, Butterworth, 1969, 139.

[9] Chryssikopoulos A.: "Pharmacodynamics and pharmakokinetics of estrogens which are used in the hormonal replacement therapy (HRT) of postmenopausal women in accordance to the way and route of administration". *Adol. Gynec. Reprod. Menop.*, 1998, *1003*, 208.

[10] Haarbo J., Hassager C., Schlemmer *et al.*: "Influence of smoking, body fat distribution and alcohol consumption on serum lipids, lipoproteins and apolipoproteins in early postmenopausal women". *Atherosclerosis*, 1990, *84*(2-3), 239.

[11] Schlemmer A., Jensen J., Riis B. J. *et al.*: "Smoking induces increased androgen levels in early postmenopausal women". *Maturitas*, 1990, *12*(2), 99.

[12] Key T. J., Rike M. C., Baron J. A., Moore J. W. *et al.*: "Cigarette smoking and steroid hormones in women". *J. Steroid. Biochem. Mol. Biol.*, 1991, *39*(4A), 529.

[13] Ruiz E., Osorio E., Ortega E.: "Androgenic status in cyclic and postmenopausal women: a comparison between smokers and non-smokers". *Biochem. Int.*, 1992, *27*(5), 841.

[14] Lobel S. M., Barbieri R. L., Walsh B. W., Yeh J.: "Acute adrenocorticotropin stimulation of postmenopausal cigarette smokers and non-smokers: effects on steroidogenesis". *Fertil. Steril.*, 1993, *59*(1), 229.

[15] Baron J. A., Comi R. J., Cryns V., Brinck-Johngen T., Mercer N. G.: "The effect of cigarette smoking on adrenal cortical hormones". *J. Pharmacol. Exp. Ther.*, 1995, *272*(1), 151.

[16] Midgette A. S., Baron J. A.: "Cigarette smoking and the risk of natural menopause". *Epidemiology*, 1990, *1*(6), 474.

[17] Tajtakova M. *et al.*: "The effect of smoking on menopause". *Unit Lek*, 1990, *36*(7), 649.

[18] Torgerson M., Avenell A., Russel L. T., Reid D. M.: "Factors associated with onset of menopause in women aged 45-49". *Maturitas*, 1994, *19*(2), 83.

Address reprint requests to:
D. KARAMANIDIS, M.D.
Opsikiou 43
68100 Alexandroupolis (Greece)