MORPHOLOGICAL LIPID PATTERNS IN VAGINAL EXFOLIATED CELLS IN POSTMENOPAUSAL PATIENTS

by

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Introduction

Post menopausal women show variable degrees of estrogenic activity (Meisels, 1966). A significant number retain some amount of estrogenic activity far into old age (Dewaard and Oettle, 1965; Stone *et al* 1967; and Pundel, 1968). The estrogenic activity is usually evaluated by colpocytology.

It has been demonstrated that human exfoliated vaginal and cervical cells contain lipid granules (Wheeler and Danziger, 1955; Masin and Masin, 1964 and Maillet *et al* 1978). The morphological patterns of the lipid granules is distinctive in the two phases of the menstrual cycle and is related to the differential hormonal stimuli (Masin and Masin, 1964; Maillet *et al* 1978, Pati *et al* 1983). Few studies are available on the lipid patterns in vaginal cells of post menopausal patients. Lipid granules were seen to vary with the estrogenic activity and were absent from atrophic smears (Masin and Masin, 1964, and Maillet *et al* 1978). These observations need to be confirmed.

The present study was planed to see if lipid patterns show a good correlation with colpocytology and whether the lipid pattern study can be used as an effective technique to evaluate estrogenic activity in postmenopausal patients.

Material and Methods

The studies were done in 14 post menopausal women varying in age from 43 to 84 years attending the gynecology out patient of All India Institute of Medical Sciences Hospital. The patients were menopausal for at least 12 months and were not taking any hormones.

Collection of Vaginal Smears: Vaginal material was obtained by lightly scraping the upper 1/3rd of lateral vaginal wall with the help of a wooden spatula and smeared on two slides. One smear was wetfixed in 95% ethyl alcohol and the other smear in Baker's fixative (Baker's fixative is prepared with 10 ml. of formalin, 1 gm. of calcium chloride and 90 ml. of distilled water).

The smear was fixed in 95% ethanol for a minimum period of 30 minutes and was stained by modified Papanicolaou stain (Gill 1969).

Staining of Smears for Lipid Granules: The smears were fixed in Baker's fixative

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for a minimum of 48 hours. They were stained with Oil red 0 employing the following steps. The slide was washed in running tap water for 2-5 minutes to wash away formalin and then rinsed in 50% isopropylalcohol. The slides were stained with 1% oil red 0 solution for 30 minutes. The smear was rinsed again with 50% Isoprophylalcohol and then washed in running water. It was counter stained with 1% light green solution for 10 minutes. The slides were again washed in running water and mounted in glycerine.

Interpretation of Slides: Slides were screened and cases showing nonspecific inflammation, Trichomonas vaginalis or Candida infection were excluded from the study.

In the Papanicolaou stained slides, 300 squamous cells were counted and expressed as Maturation Index (M.I.).

In the smears stained for lipid, 100 cells were examined. Number of cells showing lipid granules were counted and expressed as percentage. The lipid granules were classified as small, medium or large using a high power objective (400 x). Small granules were less than 1-u and appeared as pin point granules, medium were about 3-4 times the size of small granules and large were about 6-8 times or more than the size of small granules. The number of cells showing small, medium or large granules was expressed as percentage.

Observations

Maturation Index (M.I.): The patients were divided into 3 groups based on M.I. (Table I). In 6 patients, the smears showed large number of parabasal cells varying from 25-85% (mean value 45.5%) (group A). In another 6 cases(intermediate cells were predominantly observed (88-100%, mean value 94.84%). (group B). In 2 patients the superficial cells were increased and comprised more than 10% of total cells (group C).

TABLE

Lipid Patterns and Maturation Index in Post Menopausal Patients

Group	Age in years	Years Menopausal	Maturation Index	Lipid Status	% cells		
				Positive for lipid	with small granules	with medium granules	with large granules
A	60	6	85/13/2	58	100	0	0
	60	12	50/50/0	86	86	14	0
	47	2	43/56/1	81	100	0	0
	55	12	40/60/0	36	83.33	16.67	0
	55	11	30/70/0	64	84.38	15.63	0
	65	12	25/75/0	65	100	0	0
B	45	1	1/99/0	73	90.41	9.59	0
	60	12	12/88/0	74	75.68	22.97	1.35
	84	30	0/94/6	80	87.5	12.5	0
	54	6	0/100/0	46	86.96	13.04	0
	55	4	0/98/2	75	69.44	26.66	4
	46	1	0/90/10	85	84.71	11.77	3.53
C	43	1	2/80/18	62	90.32	9.68	—
	57	6	0/50/50	92	94.57	5.43	—

Lipid Stain: The lipid patterns observed were also quite variable. The percentage of lipid positive squamous cells varied from 36-92%. In group A cases with parabasal cell predominance, the percentage of squamous cells with lipid varied from 36-86% (mean value 65%) and most of the lipid globules were of small size (83.33-100%-mean value 92.29%). The medium and large size granules were 0-16.67% (mean value 9.72%) and 0% respectively (Table I).

In group B cases with predominance of intermediate cells, the percentage of lipid positive cells varied from 46-85% (mean value 72.17%) and the percenatge of cells with small, medium and large sized granules varied from 69.44 to 90.41% (mean value 82.45%), 9.59-26.66% (mean value 16.09%) and 0-4% (mean value 1.48% respectively (Table I).

In group C patients with estrogenic activity, the percentage of lipid positive cells were 62% and 92%. These cells predominantly contained small size lipid granules (mean value 92.43%). Medium sized granules were seen in a few cells and large granules in none (Table I).

Discussion

Post menopausal women show variable cytohormonal patterns. Meisels (1966) studied 5920 manopausal patients and showed progressive decrease of the estrogenic effects with passing of years in about half of the cases. Twenty-one per cent of patients had complete atrophy, 40% had moderate estrogenic activity and 10% cases had high estrogenic activity far into old age. Appreciable number of post menopausal women retaining estrogenic activity far into old age has been described by many other workers also (Dewaard and Oettle, 1965; Stone *et al* 1967; and Pundel, 1968). Complete atrophy may be absent even after the age of 70 years (Pundel, 1968).

Similarly, we observed variable amount of proliferation and atrophic smear unrelated to the years of menopause in the group of 14 patients studied by us.

Maillet *et al* (1978) observed that the amount of lipid granules varied in proportion to the persistence of estrogenic activity. Masin and Masin (1964) found absence or scarcity of lipid granules in atrophic smears. In the present study, results with lipid staining are different from the above workers. Lipid was observed in 36 to 86 per cent of squamous cells in the group of women having significant numbers of parabasal cells in vaginal smears and these granules were mostly small in size. The smears showing intermediate cell predominance showed more medium and large size lipid granules than the cases with atrophic or estrogenic smears. The smears from 2 patients with more superficial cells have an increased number of lipid containing cells and predominantly small granules. Thus we found lipid granules in significant number of cells in menopausal patients and a pattern in the size of the granules was observed in patients showing complete atrophy to proliferative activity.

However, our results demonstrate that lipid patterns cannot be used to evaluate estrogenic activity in post menopausal patients as similarity in lipid patterns is being observed between groups showing atrophy and proliferative activity.

Summary

Morphological lipid patterns in vaginal cells of 14 post menopausal women were studied. Lipid granules were seen in significant number of cells with different patterns in smear with atrophy, inter-

mediate cell predominance and smears showing estrogenic activity. However, this technique is not found useful to evaluate estrogenic activity in these patients.

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