Maturitas, 10 (1988) 307—316
Elsevier Scientific Publishers Ireland Ltd.

MAT 00522

Serum levels of oestrogens, progesterone, follicle-stimulating hormone and sex-hormone-binding globulin during simultaneous vaginal administration of 17β-oestradiol and progesterone in the pre- and post-menopause

K. Carlström, H. Pschera and N-O. Lunell

Department of Obstetrics and Gynaecology, Karolinska Institutet, Huddinge University Hospital, S-141 86 Huddinge, Sweden

(Received 11 March 1988; revision received 26 May 1988; accepted 26 September 1988)

Serum concentrations of 17β-oestradiol (E_2), unconjugated oestrone (E_1), total oestrone (tE_1), progesterone (P), follicle-stimulating hormone (FSH) and sex-hormone-binding globulin (SHBG) were measured before and after daily intravaginal administration of 250 μg micronized E_2 and 10 mg micronized P for 14 days to 12 post-menopausal and for 1 day only (during cycle days 5—8) to 11 pre-menopausal women. In the post-menopausal women the levels of all steroids increased to maximum values on day 1, 8—10 h after administration and fell thereafter. In the pre-menopausal women the steroid concentrations rose slowly to a plateau level 10—15 h after administration. Significantly higher absorption of E_2 and E_1 (area under the curve increments) was noted in the post-menopausal than in the pre-menopausal women.

In the post-menopausal women the steroid levels measured on days 7 and 14 corresponded to those observed in the very early or late luteal phase. Area under the curve increments were usually smaller on days 7 and 14 than on day 1 and the absorption kinetics altered to a 'pre-menopausal' pattern. FSH levels were significantly reduced as from 12 h after administration on day 1 and onwards. A slight (10%) but significant increase in SHBG levels was noted on day 14. It was concluded that the combined E_2 and P treatment used in this investigation brings about a physiological response with only minimal side effects on the liver as judged from changes in SHBG concentrations.

(Key words: Oestrogens, Progesterone, Vaginal absorption, Pre-menopause, Post-mehopause, Systemic effects)

Introduction

Vaginal administration may offer an attractive alternative route for post-menopausal systemic oestrogen replacement therapy, since it avoids the side effects ascribed to the first passage of the drug through the liver [1,2]. Modern

Correspondence to: Kjell Carlström, Ph.D., Department of Obstetrics and Gynaecology, Karolinska Institutet, Huddinge University Hospital, S-141 86 Huddinge, Sweden.

post-menopausal oestrogen replacement therapy regimens include progestogen supplementation. However, with one exception [3], previous studies on vaginal oestrogen absorption in post-menopausal women were performed with pure oestrogen preparations and yielded partially conflicting results [4—15]. We therefore studied the absorption kinetics and certain systemic effects on the pituitary (follicle-stimulating hormone, FSH) and liver (sex-hormone-binding globulin SHBG) of the simultaneous intravaginal administration of 17β-oestradiol (E_2) and progestrone (P) in post-menopausal women. Measurements were carried out prior to and after 14 days of treatment and the absorption kinetics were compared with those in a group of women of fertile age.

Subjects and methods

Clinical sample and drug delivery system

Twelve (12) women aged 52—70 yr (mean 60.6) who had been post-menopausal for 3—20 yr (mean 10.2 yr) and 11 women of fertile age aged 29—43 yr (mean 37.2 yr) participated in the study. All the subjects were apparently healthy and free from clinical or laboratory signs of hepatic, biliary, renal, intestinal or endocrinological malfunction. None were receiving any medication. Gynaecological examination revealed a highly atrophic vaginal mucosa in all the post-menopausal women.

One vagitory containing 0.25 mg micronized 17β-oestradiol and 10 mg micronized progesterone (Klimavag, NOVO Industri A/S, Bagsvaerd, Denmark) was deeply inserted intravaginally with a special applicator at 0800 h every day for 14 days in the post-menopausal women and on 1 day only during cycle days 5—8 in the women of fertile age. Venous blood samples were drawn before and then 2, 4, 6, 8, 10, 12, 15 and 24 h after administration on day 1 in all subjects and on days 7 and 14 in the post-menopausal women. Serum was separated by centrifugation and stored at $-20\,^{\circ}C$ until analyzed.

The study was approved by the ethical committee at Huddinge University Hospital and the full, informed consent of all the women participating in the study was obtained.

Hormone and SHBG analysis

Serum concentrations of E_2, P and FSH were determined by direct radioimmunoassay of untreated serum using commercial kits (Diagnostic Products Corporation, Los Angeles, USA). Unconjugated oestrone (E_1) and total oestrone (tE_1, ≥85% oestrone sulphate) were determined in serum by radioimmunoassay after ether extraction as described previously [16]. In the tE_1 assay, extraction was preceded by enzymatic hydrolysis. SHBG concentrations were determined by an immunoradiometric technique using commercial kits (Farmos Diagnostica OY, Turku, Finland).

The detection limits and the intra-assay and interassay coefficients of variation

were as follows: for E_2, 35 pmol/l, 5% and 9% respectively; for P, 0.15 nmol/l, 5.8% and 7.2%; for FSH, 1.2 units/l, 7% and 11; for E_1, 30 pmol/l, 7.0% and 9.8%; for tE_1, 0.3 nmol/l, 7.0% and 8.9%; for SHBG, 0.5 nmol/l, 5% and 11%.

Statistical methods

Either parametric or non-parametric tests were used according to the type of distribution involved. Normally distributed values were expressed as the arithmetic mean ± S.E.M. and otherwise as the mean and range.

The absorption of steroids over 6 and 15 h following insertion of the vaginal suppository was expressed in terms of area under the curve (AUC) increments. For each sampling interval, the area was calculated as the product of (a) the mean concentation during the interval minus the concentration at 0 h, and (b) the duration of the interval (2 h, except for the 12—15-h interval). The AUC value represents the sum of the areas during the sampling intervals spanned by the observation period (0—6 h and 0—15 h).

Results

The steroid levels before and during vaginal administration of E_2 and P are shown in Figs. 1—4. In the post-menopausal women on day 1 peak values for all the steroids were observed 8—10 h after administration, following which their levels fell. A different absorption pattern was observed in the pre-menopausal women, in whom the steroid concentrations rose slowly to a plateau level 10—15 h after application. AUC increments were significantly higher in the post-menopausal women on day 1 than in the pre-menopausal women in the case of E_2 (0—15 h) and E_1 (0—6 h) (Table I).

The mean oestrogen and progesterone levels in the post-menopausal women during prolonged treatment with intravaginal E_2 + P corresponded to those generally observed at the very beginning or end of a luteal phase. Prolonged treatment gradually changed the post-menopausal absorption pattern into a pre-menopausal pattern with delayed or even absent peaks following the insertion of the vaginal suppositories. The AUC increments were usually significantly lower on days 7 and 14 than on day 1 (Table I).

The effects of the treatment on pituitary and liver functions in the post-menopausal women, as indicated by serum concentrations of FSH and SHBG, are shown in Figs. 5. Serum concentrations of FSH were already significantly ($P < 0.05$) lowered in relation to pretreatment levels after 12 h (data not shown) and they remained significantly decreased during the rest of the observation period. The serum concentrations of SHBG were unaffected after 1 week, but a slight (10%) although significant increase was observed after 2 wk of treatment. A minor breakthrough bleeding/discharge of 1 day's duration was observed in 1 of the pre-menopausal and 1 of the post-menopausal women during treatment.

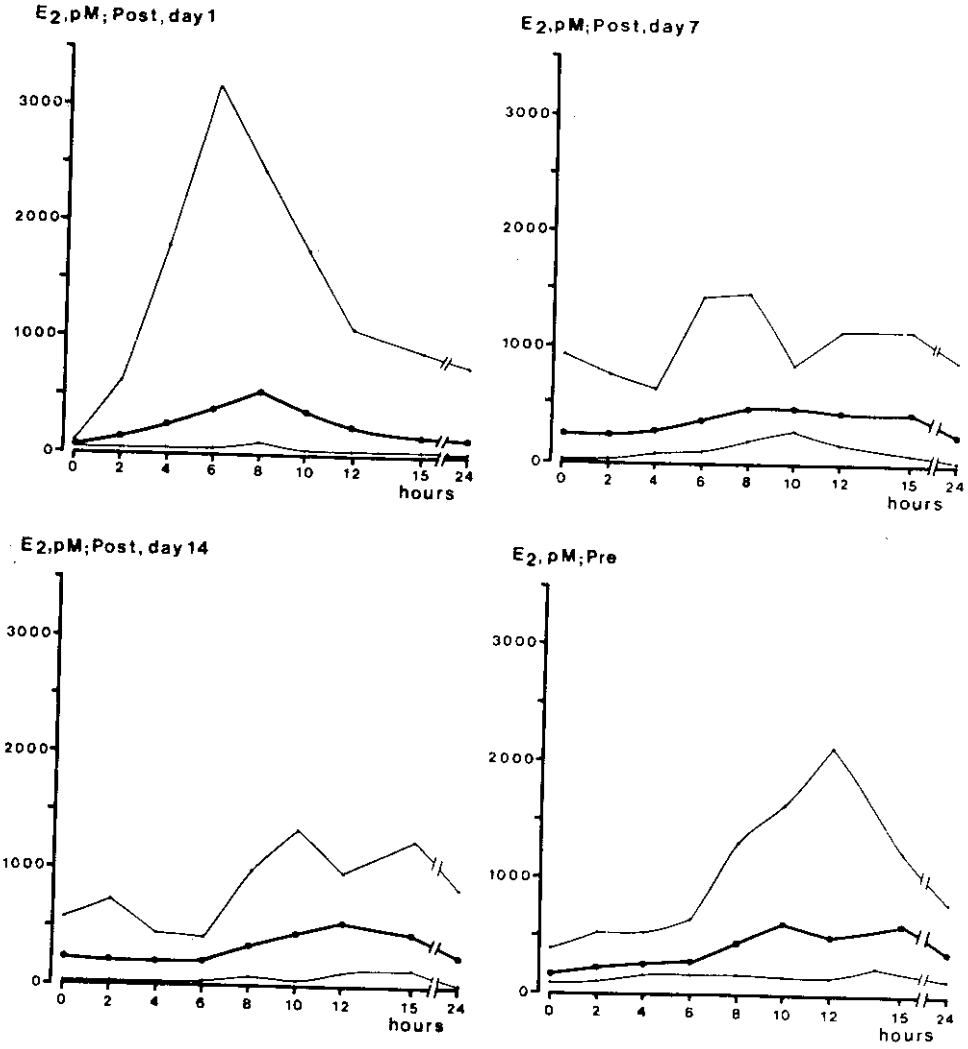

Fig. 1. Peripheral serum levels of 17β-oestradiol (E_2) before and during daily intravaginal administration of 250 μg E_2 and 10 mg progesterone (P) to 12 post-menopausal women (Post) and to 11 women of fertile age (Pre), showing geometric mean and range.

Discussion

The present results are in accordance with those obtained with the same preparation by Kâlund-Jensen and Myrén in a limited study in post-menopausal women [3] and confirm previous reports of efficient vaginal absorption of steroids in post-menopausal women producing systemic effects. The results further

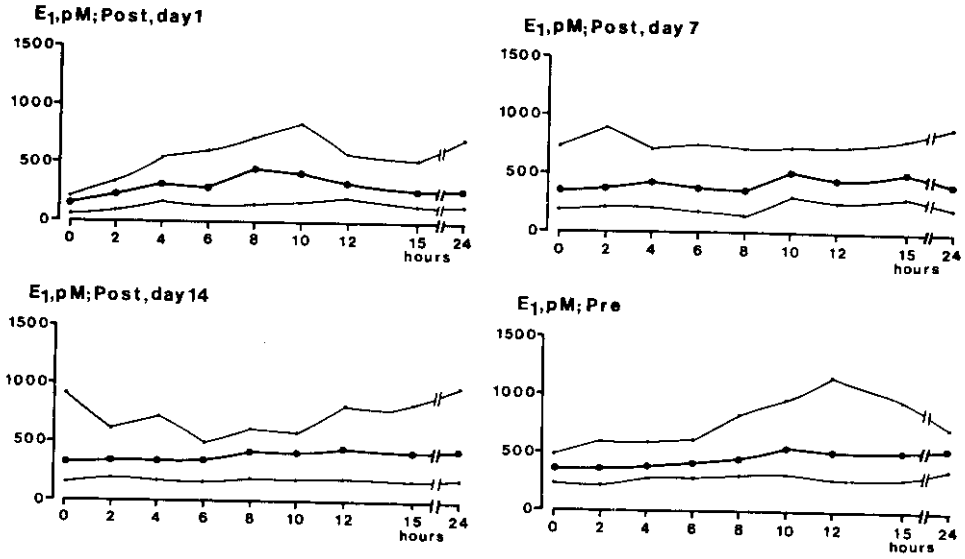

Fig. 2. Peripheral serum levels of unconjugated oestrone (E_1) before and during daily intravaginal administration of 250 μg E_2 and 10 mg P to 12 post-menopausal women (Post) and to 11 women of fertile age (Pre), showing geometric mean and range.

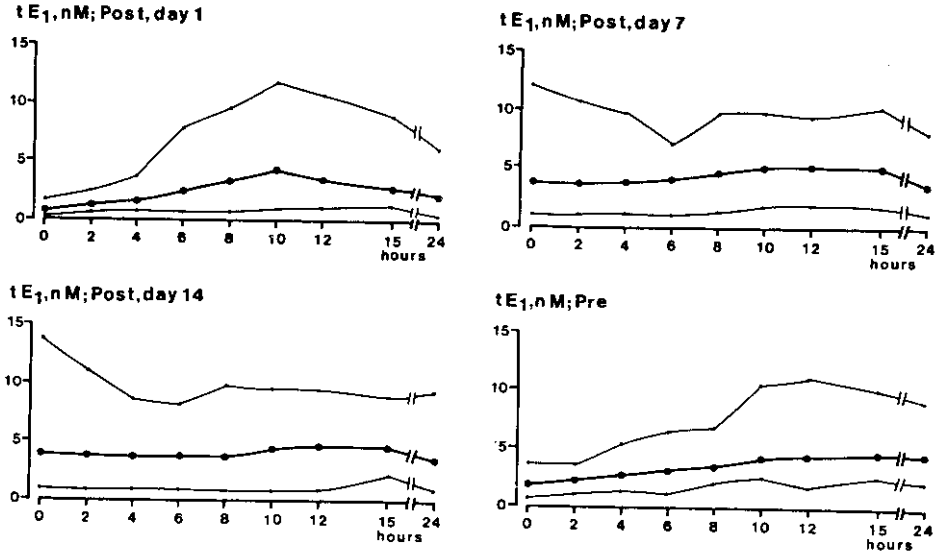

Fig. 3. Peripheral serum levels of total oestrone (tE_1) before and during daily intravaginal administration of 250 μg E_2 and 10 mg P to 12 post-menopausal women (Post) and to 11 women of fertile age (Pre) showing geometric mean and range.

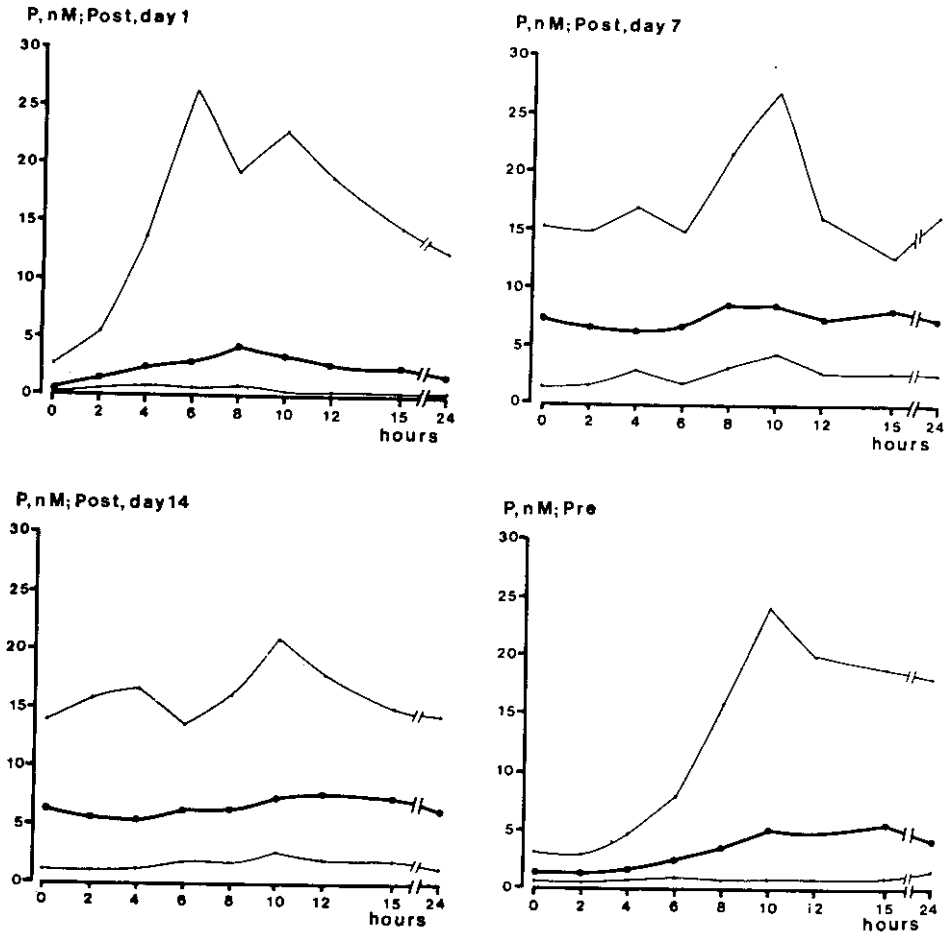

Fig. 4. Peripheral serum levels of progesterone (P) before and during daily intravaginal administration of 250 μg E_2 and 10 mg P to 12 post-menopausal women (Post) and to 11 women of fertile age (Pre), showing geometric mean and range.

confirm our previous finding that vaginal absorption of oestrogens is less efficient in women of fertile age than in post-menopausal women [11] and that prolonged oestrogen treatment changes the vaginal absorption pattern in post-menopausal women [12]. Reduced vaginal absorption of oestrogens, in varying degrees, after long-term oestrogen treatment has been reported by most other groups [3,5,6,9,10,13,14]. In a careful study, Haspels et al. [5] showed that this occurs even when low oestrogen doses producing no systemic effects are used.

It may be argued that a declining 'base level' due to the metabolism of previously administered oestrogen possibly counter-balances the potential increase in oestrogen levels following administration on days 7 and 14, thus leading to

TABLE I

AREA UNDER THE CURVE (AUC) INCREMENTS IN SERUM STEROIDS IN POST-MENOPAUSAL (POST) AND PRE-MENOPAUSAL (PRE) WOMEN FOLLOWING INTRAVAGINAL APPLICATION OF 250 μg 17β-OESTRADIOL (E_2) AND 10 mg PROGESTERONE (P). THE E_2 AND UNCONJUGATED OESTRONE (E_1) AUCs ARE EXPRESSED AS pmol/l × h AND THE TOTAL OESTRONE (tE_1) AND P AUCs AS nmol/l × h. MEAN AND RANGE ARE INDICATED

	AUC				Significance (*P* value)		
	Post Day 1	Post Day 7	Post Day 14	Pre (fertile age)	Day 1 vs. day 7[a]	Day 1 vs. day 14[a]	Day 1 vs. Pre[b]
E_2 0—6 h	2367 (0—8172)	535 (0—2742)	100 (0—324)	462 (114—942)	< 0.05	< 0.05	NS
E_2 0—15 h	8967 (690—17425)	2700 (0—6390)	2103 (0—4215)	4314 (405—9240)	< 0.01	< 0.01	< 0.05
E_1 0—6 h	714 (12—1644)	354 (0—1044)	246 (0—984)	71.5 (0—246)	NS	NS	< 0.01
E_1 0—15 h	2625 (0—5535)	1095 (0—2880)	990 (0—3195)	1145 (0—4380)	NS	< 0.05	NS
tE_1 0—6 h	6.2 (0.8—17.7)	2.2 (0—7.3)	2.5 (0—17.9)	3.8 (0—11.0)	< 0.05	NS	NS
tE_1 0—15 h	34.1 (1.5—78.5)	12.0 (0—45.6)	10.1 (0—59.3)	22.5 (0—66.8)	< 0.05	< 0.05	NS
P 0—6 h	17.0 (0—57.6)	5.6 (0—29.4)	4.0 (0—41.4)	4.5 (0—13.8)	NS	< 0.05	NS
P 0—15 h	59.1 (3.0—138.0)	24.5 (0—102.0)	26.4 (0—136.0)	49.2 (0—106.5)	NS	< 0.05	NS

[a]*t*-test for paired observations.
[b]Mann-Whitney *U* test.

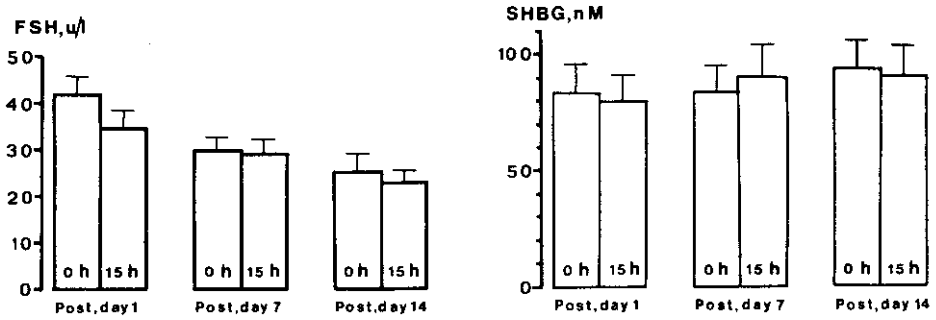

Fig. 5. Peripheral serum levels of follicle-stimulating hormone (units of human pituitary FSH 68/39) and sex-hormone-binding globulin (SHBG) before and during daily intravaginal administration of 250 μg E_2 and 10 mg P to 12 post-menopausal women, showing arithmetic mean and S.E.M. FSH concentrations were significantly ($P < 0.001$) lower than pretreatment levels on day 1 after 15 h as well as on days 7 and 14 (0 and 15 h). SHBG concentrations were significantly ($P < 0.05$) higher than pretreatment levels on day 14 (0 and 15 h).

unchanged serum levels. Such a mechanism might contribute to a certain degree, but the significantly lower absorption of E_2 and E_1 in women of fertile age indicates that the degree of maturation of the vaginal epithelium could be more decisive in this respect. However, a mature vaginal epithelium does not obstruct the absorption of steroids.

The absorption pattern of the preparation studied was similar to that displayed by pure oestrogens, indicating that the P supplementation did not exert a significant influence. Villanueva et al. [17] reported enhanced absorption of P in post-menopausal women treated with oral oestrogen in comparison with untreated women. This is at variance with the findings of the present study, in which unchanged or decreased absorption of P followed oestrogen treatment. Circulating P is rather strongly bound to corticosteroid-binding globulin (CGB) [18] and it has been suggested that the higher CBG levels induced by oral oestrogen could be a possible mechanism underlying the increased absorption of P during oral oestrogen treatment [17]. In contrast to oral oestrogens, parenterally-administered oestrogens have little or no effect on 'steroid sensitive' carrier proteins such as CBG and SHBG [1,2]. This may explain the divergent results as regards P absorption.

The decreased FSH levels during treatment are a further indication of the systemic effects of the E_2 + P preparation. As regards its effects on the liver, a slight but significant increase in SHBG concentrations was observed on day 14. With the exception of the study by Goebelsmann et al. [19], in which very high doses of the strong synthetic oestrogen ethinyl oestradiol were used, previous studies on the effects of intravaginal oestrogens on SHBG in post-menopausal women have shown these to be non-existent or minor, despite considerably increased serum oestrogen concentrations [3,7,10,15,20]. The very modest change in SHBG seen in the present study is of the same order as the changes reported in

recent controlled studies on the menstrual cycle [21—23]. We have in fact previously suggested that sex steroids may not be the main physiological regulators of SHBG and other 'steroid sensitive' liver proteins [1].

It may be concluded that vaginal absorption patterns differ in pre-menopausal and post-menopausal women. Combined E_2 and P treatment changes the post-menopausal absorption kinetics picture to a pre-menopausal pattern. Steroid concentrations during this type of replacement therapy remain within the physiological limits for women of fertile age and the liver side effects are minimal as judged on the basis of SHBG concentrations.

References

1 Stege R, Fröhlander N, Carlström K, Pousette Å, von Schoultz B. Steroid-sensitive proteins, growth hormone and somatomedin C in prostatic cancer: Effects of parenteral and oral estrogen therapy. Prostate 1987; 10: 333—338.

2 Cedars MI, Judd HL: Non-oral routes of estrogen administration. Obstet Gynecol N Am 1987; 14: 269—298.

3 Kåhlund-Jensen H, Myrén CJ. Vaginal absorption of oestradiol and progesterone. Maturitas 1984; 6: 359—367.

4 Dickerson J, Bressler RNR, Christian CD, Hermann HW. Efficacy of estradiol vaginal cream in postmenopausal women. Clin Pharmacol Ther 1979; 26: 502—507.

5 Haspels AA, Luisi M, Kicovic PM. Endocrinological and clinical investigations in post-menopausal women following administration of vaginal cream containing oestraiol. Maturitas 1981; 3: 321—327.

6 Stumpf PG, Maruca J, Santen RJ, Demers LM. Development of a vaginal ring for achieving physiologic levels of 17beta-estradiol in hypoestrogenic women. J Clin Endocrinol Metab 1982; 54: 208—210.

7 Mattsson L-Å, Cullberg G: A clinical evaluation of treatment with estriol vaginal cream versus suppository in postmenopausal women. Acta Obstet Gynecol Scand 1983; 62: 397—401.

8 Mandel FP, Geola FC, Meldrum DR, Lu JHK, Eggena P, Sambhi MP, Hershman JM, Judd HC. Biological effects of various doses of vaginally administered conjugated equine estrogen in postmenopausal women. J Clin Endocrinol Metab 1983; 57: 133—139.

9 Heimer G, Englund D. Estriol: Absorption after long-term vaginal treatment and gastrointestinal absorption as influenced by a meal. Acta Obstet Gynecol Scand 1984; 63: 563—567.

10 Veldhuis JD, Samojlik E, Evans WS, Rogol AD, Ridgeway CE, Crowleg WF, Klop L, Chechinska E, Kirschner MA, Thorner MD, Stumpf P. Endocrine impact of pure estradiol replacement in postmenopausal women: Alterations in anterior pituitary hormone release and circulating sex steroid hormone concentrations. Am J Obstet Gynecol 1986; 155: 334—339.

11 Furuhjelm M, Karlgren E, Carlström K. Intravaginal administration of conjugated estrogens in premenopausal and postmenopausal women. Int J Gynaecol Obstet 1980; 17: 335—339.

12 Carlström K, Karlgren E, Furuhjelm M, Ryd-Kjellén E. Effects of intravaginal oestrogen treatment upon the vaginal absorption of conjugated equine oestrogens. Maturitas 1982; 4: 277—281.

13 Kicovic PM, Cortes-Prieto J, Milojevic S, Haspels AA, Aljinovic A. The treatment of postmenopausal vaginal atrophy with ovestin vaginal cream or suppositories: Clinical, endocrinological and safety aspects. Maturitas 1980; 2: 275—282.

14 Widholm O, Vartiainen E. The absorption of conjugated oestrogen and sodium oestrone sulphate from the vagina. Ann Chir Gynaecol Fenn 1974; 63: 186—190.

15 Luisi M, Franchi F, Kicovic PM. A group-comparative study of effects of Ovestin cream versus Premarin cream in post-menopausal women with vaginal atrophy. Maturitas 1980; 2: 311—319.

16 Brody S, Carlström K, Lagrelius A, Lunell N-O, Rosenborg L. Adrenocortical steroids, bone mineral content and endometrial condition in post-menopausal women. Maturitas 1982; 4: 113—122.

17 Villanueva B, Casper RF, Yen SSC. Intravaginal administration of progesterone: Enhanced absorption after estrogen treatment. Fertil Steril 1981; 35: 433—437.
18 Dunn JF, Nisula BC, Rodbard D. Transport of steroid hormones: Binding of 21 endogenous steroids to both testosterone-binding globulin and corticosteroid-binding globulin in human plasma. J Clin Endocrinol Metab 1981; 53: 58—68.
19 Goebelsmann U, Maschak A, Mishell DR. Comparison of hepatic impact of oral and vaginal administration of ethinyl estradiol. Am J Obstet Gynaecol 1955; 151: 868—877.
20 Stumpf PG, Lloyd TA. Response of a liver protein, sex hormone binding globulin (SHBG), to modulations of circulating oestradiol. Maturitas 1984; 6(2): 195—196.
21 Dowsett M, Attree SC, Virdee SS, Jeffcoate SL. Oestrogen-related changes in sex hormone binding globulin levels during normal and gonadotrophin-stimulated menstrual cycles. Clin Endocrinol (Oxf.) 1985; 23: 303—312.
22 Apter D, Bolton NJ, Hammond GL, Vihko R. Serum sex hormone-binding globulin during puberty in girls and in different types of adolescent menstrual cycles. Acta Endocrinol (Copenh) 1984; 107: 413—419.
23 Plymate SR, Moore DE, Cheng CY, Bardin CW, Southworth MB, Levinsky MJ. Sex hormone-binding globulin changes during the menstrual cycle. J Clin Endocrinol Metab 1985; 61: 993—996.

Maturitas, 10 (1988) 317—332
Elsevier Scientific Publishers Ir

MAT 00484

Cultural cc
syndi

Margaret Lo

[a]*Faculty of Medicine, McGill U*
[b]*Faculty of Medicine, U*

(Received 27 Novembe

Europe and North America symptoms. In this study, in the asked to describe the menopau health at midlife was then dist replies received from 1141 nor group the symptoms these wom based on the factor scores, one- symptom experience and the epi status. Symptom experience was

(Key words: Symptoms, Japan,

Introduction

Information on the mainly from research in I widening this data bas complexities of doing c Researchers customarily concepts of the body an thought and involve ide which are particular to doing research in societi and their bodies, or the i

As illustration, this pa 1983—1984. An earlier p and social context of the the collection and analy sources: first, interviews in which they were aske associated with this peri