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Oral versus Vaginal Absorption in Oestradiol in Postmenopausal Women. Effects of Different Particles Sizes

D. E. Englund and E. D. B. Johansson

Department of Obstetrics and Gynaecology, University Hospital, Uppsala, Sweden

ABSTRACT

Crystalline oestradiol-17β is poorly absorbed from the gastrointestinal tract. Three different fractions and a standard fraction containing oestradiol-17β of a known particle size and surface area, were administered orally, to postmenopausal women, to test if changes in particle size will influence the absorption. The bioavailability of each fraction was determined by measurements of peripheral plasma oestrogens. Two different dosages of the standard fraction were given vaginally to compare the bioavailability after oral and vaginal administration. The gastrointestinal absorption was dependent of the particle size of oestradiol. The smaller particle the more rapid and effective absorption as reflected by increasing area under the plasma concentration curve of oestrone and oestradiol. The smallest particle, however, resulted in a pronounced initial oestradiol peak. The coarser particles were more slowly absorbed with more even plasma oestrogen elevation for a sustained period of time. The vaginal absorption of oestradiol was more effective than the gastrointestinal. When the same amount of an equal preparation according to particle size, was given vaginally the maximal plasma concentration was almost 40 times higher than when given orally.

INTRODUCTION

Oestrogen therapy to oestrogen deficient women is well established. However, the management of this treatment is far from solved. The dose and the choice of oestrogen as well as the route of administration is still a matter of research.

The choice between synthetic and natural oestrogens is under debate. There is no significant preference for the use of natural oestrogens in the treatment of climacteric disorder (5, 12, 26, 30). However, the fertile women secrete

mainly oestradiol from the ovaries and when this secretion ceases in the menopause the only source of oestrogen available is oestrone derived from peripheral conversion of androstenedione (4, 13, 21, 27). Therefore oestradiol and oestrone are considered the most physiologic oestrogens to use in replacement therapy.

Crystalline 17β-oestradiol is poorly absorbed from the gastrointestinal tract (1, 6). In 1975 Yen et al. (29) reported that orally administered micronized oestradiol was absorbed, as reflected by raised oestrogen levels and some depression of the gonadotrophins in plasma. In 1977 Helles et al. (15) reported that 2 mg oestradiol given orally was absorbed equally well as 2 mg oestradiol valerianate. Clinically, oral micronized oestradiol is a highly successful therapy for menopausal women with oestrogen deficiency symptoms as hot flushes and genital atrophy (7).

According to Lebech (17, 18) the crystal size of oestradiol is an important factor for the absorption from the gastrointestinal tract. Oestradiol is a drug of very low water solubility, less than 5 μg/ml. If therefore the dissolution rate is the rate limiting step in the absorption process, changes in particle size of the drug will influence the absorption. It is well established that a reduction of particle size and thereby an increase in the surface are by micronization for insoluble drugs often results in enhanced dissolution rates and thereby an increase in the absorption (11).

The present study was performed in order to test this theory with regard to oestradiol. Three different fractions and a standard fraction containing oestradiol of a known particle size and surface area, were administered orally in suspension. The bioavailability of each fraction was determined by measurement of peripheral plasma oestrogens. It has recently been shown (10, 22, 24) that oestradiol is well absorbed from the vagina. Therefore in a following investigation, the same patients treated orally with oestradiol were given the standard fraction in two different dosages of oestradiol vaginally to compare the bioavailability of the two different routes of administration.

MATERIAL AND METHODS

Five healthy postmenopausal women volunteered for the study. All were menopausal for at least two years. They had not been on oestrogen therapy the last three months. The women were told not to eat nor drink eight hours before and four hours after administration of oestradiol. The study was double blind. The subjects were their own controls.

Two mg oestradiol-17β was administered orally in 10 ml water solution containing 0.1% kollidon K 25 and 0.1% sodiumbenzoat followed by ingestion of 200 ml of water. The particle size distribution of the four fractions administered are measured on a Coulter Counter TA II (See Table 1). From the size distribution data it is possible to calculate the surface area value. These measurements were made by I-L Kvorning and M Strid Christensen, Novo AG, Copenhagen, Denmark.

TABLE 1. Particle size analyses

		OESTRADIOL			
		Standard	Fraction I	Fraction II	Fraction III
Coulter	90% >	1.92 /um	0.56 /um	3.48 /um	18.5 /um
Counter	60% >	4.95 /um	0.86 /um	6.30 /um	28.0 /um
Vol. %	10% >	10.40 /um	1.55 /um	9.43 /um	39.1 /um
Hatch	d_w	4.80 /um	0.88 /um	6.00 /um	27.7 /um
Choate	d_s	3.96 /um	0.80 /um	5.60 /um	26.6 /um
	α	1.86	1.55	1.45	1.33
	S	1.23 m^2/g	6.10 m^2/g	0.87 m^2/g	0.18 m^2/g

d_w: weight mean diameter (50% size) α: the standard deviation
d_s: surface mean diameter S: the specific surface area

Two mg and 2.5 mg oestradiol of the standard fraction were administered intravaginally in a water-soluble base. Two ml of this base containing oestradiol-17β was applied in the posterior fornix of the vagina. The subjects stayed recumbent for at least half an hour after the application. The period between each oestradiol administration was at least one week. All oestradiol preparations were supplied by Novo Industri A/S, Novo Allé, DK-2880 Bagsvaerd, Denmark.

One single blood sample was drawn immediately before and 7½, 15, 30, 45, 60 and 90 minutes, 2, 4, 8, 12, 24 (48 and 72) hours after the administration of oestradiol. The blood was drawn to heparinized tubes, centrifuged and the plasma was withdrawn and stored at -20° C until analysed. The plasma levels of oestradiol were measured by the radioimmunoassay (RIA) described by Edqvist and Johansson (9). An antiserum against oestradiol-17β-6(O-carboxymethyl)-oxime-bovine serum albumin was used. Oestrone will crossreact to 11 per cent with this antibody (19). The oestradiol levels presented are corrected for crossreaction with oestrone. For the estimation of oestrone the RIA described by Axelsson et al. (2) was used. The per cent crossreaction with oestradiol is 0.44 in the oestrone assay.

The plasma levels of oestrone and oestradiol measured after oral administration of 2 mg oestradiol (Standard, Fraction I, II and III) are presented in Fig. 1. The results are presented as mean of five subjects. ($\bar{X}$ pg/ml $\pm$ SD are presented in Table 2).

The pretreatment oestrone level was 32 pg/ml ($\pm$ SD 9) and oestradiol was 32 pg/ml ($\pm$ SD 10.4). The areas under the oestrone and oestradiol plasma concentration curves (pg/ml hours) are presented in Table 3.

The bioavailability of fraction I, II and III and the standard fraction was significantly different. The calculations were made on the following areas, both under the oestrone and oestradiol plasma concentration curves:

0 - 1 hours $p < 0.01^{**}$

1 - 8 hours $p < 0.001^{***}$

0 - 72 hours $p < 0.05^{*}$

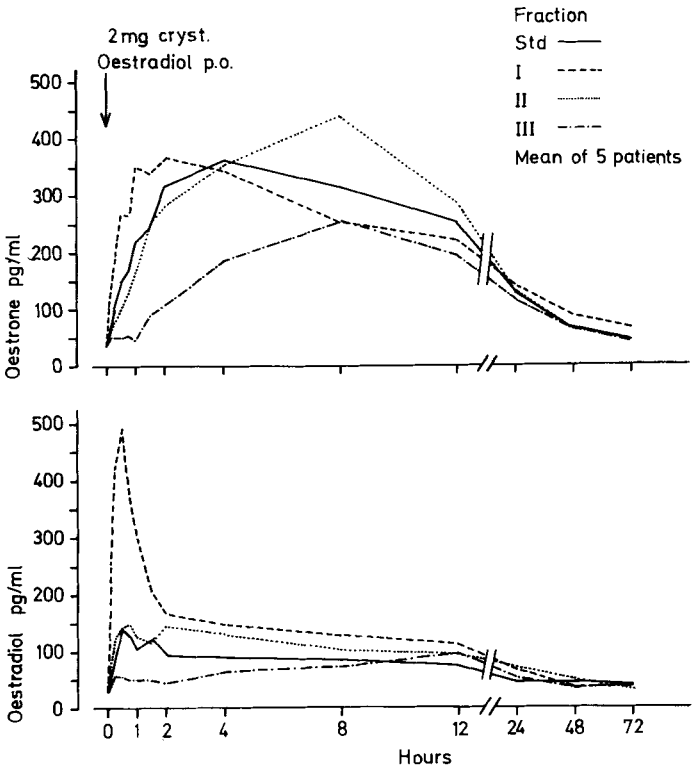

Fig. 1. Plasma levels of oestrone and oestradiol after oral administration of four different fractions of crystalline oestradiol-17β to five postmenopausal women. All values are expressed in pg/ml and as mean of five.

TABLE 2. Two mg oestradiol orally to five postmenopausal women.

Plasma levels of E_1 and E_2 at different times after oral administration of 2 mg oestradiol	Standard				Fraction I				Fraction II				Fraction III			
	$\bar{X}E_1$ pg/ml	± SD	$\bar{X}E_2$ pg/ml	± SD	$\bar{X}E_1$ pg/ml	± SD	$\bar{X}E_2$ pg/ml	± SD	$\bar{X}E_1$ pg/ml	± SD	$\bar{X}E_2$ pg/ml	± SD	$\bar{X}E_1$ pg/ml	± SD	$\bar{X}E_2$ pg/ml	± SD
0	32	9	32	10												
7½ minutes	69	28	92	53	103	48	261	139	60	33	86	7	50	22	44	10
15	102	42	119	30	177	74	435	234	76	18	126	27	50	22	59	13
30	149	47	154	47	267	45	519	350	101	27	152	45	48	20	59	17
45	167	54	146	75	264	41	404	249	130	46	161	44	51	25	51	10
60	219	99	129	60	350	75	344	184	166	69	145	54	44	19	51	8
90	242	105	148	53	339	104	241	69	248	128	141	56	84	16	60	26
2 hours	314	104	127	35	363	88	208	83	282	111	177	74	105	29	56	25
4	360	184	130	53	342	136	185	59	349	108	167	37	183	61	86	23
8	313	107	119	23	250	69	155	60	435	192	150	31	253	132	100	43
12	249	115	103	29	219	73	133	34	286	105	120	23	188	106	118	92
24	129	45	59	26	138	48	80	24	131	47	86	33	109	32	64	25
48	65	28	49	14	83	47	42	24	66	23	57	19	56	19	43	12
72	45	15	45	23	63	31	43	30	46	21	36	12	35	19	44	17

TABLE 3. Two mg oestradiol orally to five postmenopausal women.

The area under the plasma concentration curve, pg/ml x hours	Standard E_2	Standard E_1	Fraction I E_2	Fraction I E_1	Fraction II E_2	Fraction II E_1	Fraction III E_2	Fraction III E_1
Time 0 - 1 hours	125	135	393	225	134	102	55	48
1 - 8 hours	893	2274	1331	2237	1132	2428	572	1239
8 -72 hours	3840	7032	4338	7484	4642	7654	3854	5988
Total area 0 -72 hours	4858	9441	6062	9946	5908	10184	4481	7275

The quoitent oestradiol/oestrone calculated from the area under the plasma concentration curves, during the first hour after administration of oestradiol was 1.7 for the smallest particle and 1.1 for the coarsest particle. During time intervals 1 - 8 hours, 8 - 72 hours and 0 - 72 hours after oral administration of oestradiol the quoitent oestradiol/oestrone was 0.4 - 0.6 for each preparation (See Table 4).

TABLE 4. Two mg oestradiol orally to five postmenopausal women.

The E_2/E_1 ratio under the plasma concentration curve, pg/ml x hours	Standard E_2/E_1	Fraction I E_2/E_1	Fraction II E_2/E_1	Fraction III E_2/E_1
Time 0 - 1 hours	0.9	1.7	1.3	1.1
1 - 8 hours	0.4	0.6	0.5	0.5
8 -72 hours	0.5	0.6	0.6	0.6
Total area 0 -72 hours	0.5	0.6	0.6	0.6

The plasma concentrations of oestradiol and oestrone after vaginal administration of 2 mg and 0.25 mg oestradiol (standard preparation) are presented in Fig. 2. The results are presented as mean of five subjects. ($\bar{X}$ pg/ml $\pm$ SD are presented in Table 5).

When 0.25 mg oestradiol was given vaginally the maximal concentration of oestradiol was 675 pg/ml, or almost 5 times higher than when 2 mg of an equal preparation, according to particle size, was given orally. When 2 mg oestradiol was given vaginally the maximal concentration of oestradiol was almost 40 times higher than when an equal amount was given orally.

The absorption of oestradiol from the vagina was very rapid. Maximal plasma levels of oestradiol were measured within 1 hour and of oestrone within 2 hours

TABLE 5. 0.2 mg and 2 mg oestradiol vaginally to five postmenopausal women.

Plasma levels of E_1 and E_2 at different times after vaginal application of 0.2 mag and 2 mg oestradiol	0.2 mg $\bar{X}E_1$ pg/ml	±SD	$\bar{X}E_2$ pg/ml	±SD	2 mg $\bar{X}E_1$ pg/ml	±SD	$\bar{X}E_2$ pg/ml	±SD
0	28	4	25	0				
7½ minutes	62	56	99	81	66	29	319	412
15	59	20	331	99	151	50	3207	2326
30	84	19	480	215	348	70	4880	2780
45	95	26	645	284	487	171	5632	2780
60	99	31	619	383	502	138	4848	2214
90	111	48	675	770	514	190	4812	3796
2 hours	94	46	128	109	522	325	3243	3918
4	71	28	46	14	456	325	1373	1971
8	48	19	35	12	264	157	296	169
12	39	9	31	11	190	121	178	71
24					107	52	64	13
48					55	25	42	13

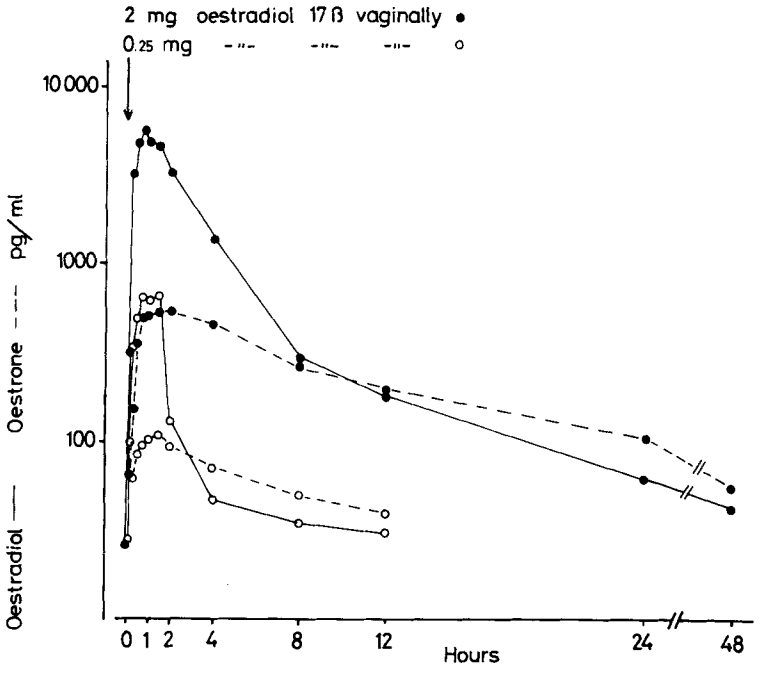

Fig. 2. Plasma levels of oestrone and oestradiol after vaginal administration of 2 mg and 0.25 mg oestradiol-17β to five postmenopausal women. All values are expressed in pg/ml and as mean of five.

When 0.25 mg oestradiol was given vaginally the maximal concentration of oestradiol was 675 pg/ml, or almost 5 times higher than when 2 mg of an equal preparation, according to particle size, was given orally. When 2 mg oestradiol was given vaginally the maximal concentration of oestradiol was almost 40 times higher than when an equal amount was given orally.

The absorption of oestradiol from the vagina was very rapid. Maximal plasma levels of oestradiol were measured within 1 hour and of oestrone within 2 hours after application. The plasma levels of oestradiol, however, declined rapidly after the initial peak. There was no initial oestrone peak and the oestrone levels were lower than the oestradiol levels during the first hours after application. The oestrone levels, however, decreased slowly and after the initial oestradiol peak the oestrogens were in the same range.

DISCUSSION

Our results indicate that the absorption of oestradiol from the gastrointestinal tract is dependent of the particle size of oestradiol. The smaller particle size the greater bioavailability, as reflected by increasing area under the plasma concentration curves of oestrone and oestradiol. The smallest particle size, however, resulted in a very pronounced initial plasma oestradiol elevation during the first two hours after administration. When the standard fraction and medium size fraction were administered an even plasma oestradiol elevation during almost 24 hours was found and there was no initial peak. This pattern seems to be more physiological since there is no significant diurnal change in peripheral plasma oestradiol or oestrone levels in menstruating women (3, 14). In postmenopausal women Vermeulen (27) has reported a nycto-hemeral rhythm for oestrone but not for oestradiol.

When oestradiol is given by mouth it is rapidly metabolized to oestrone. This metabolism occurs at least partly in the intestinal wall (8) and the result will be peripheral plasma levels of oestrone higher than oestradiol (16, 29). Our results show that this conversion is not established the first hour after administration of the smallest oestradiol particle. The plasma oestradiol levels are higher than the oestrone levels. One explanation to this phenomenon may be that at least a part of the smallest oestradiol particles are absorbed via the lymphatic system and escape immediate metabolism in the intestinal mucosa and the liver. This mechanism has been postulated for oral oily solution of oestradiol decanoate which maintains the plasma oestrone/oestradiol ratio between 0.5 - 0.6 as long as it is administered (20).

The plasma ratio of oestradiol/oestrone, however, does not reveal the tissue concentrations of the hormones, e.g. the affinity of uterine receptors is higher for oestradiol than for oestrone (25). Therefore oestradiol, even though lower in peripheral plasma, may be the oestrogen that predominates at the receptor. Further, oestrone is metabolized to oestradiol at receptor level (28). Accordingly the peripheral plasma oestrogen levels are an incomplete measurement of the oestrogenicity of a drug.

The vaginal route of administration of oestradiol has been studied in order to circumvent the conversion of oestradiol to oestrone in the mucosa of the small intestine. However, due to endogenous conversion there will still be some metabolism of oestradiol to oestrone. Our data on vaginal application support earlier reports that the intravaginal route is far more complete the the oral route (10, 22, 23, 24). In our study the plasma peak levels of oestradiol following vaginal application of 2 mg oestradiol (standard particle size preparation) were about 40-fold higher than those achieved following oral administration of the same dose. With 0.25 mg given vaginally the oestradiol peak level was about 5-fold higher than when 2 mg was given orally. The oestradiol levels, however, declined rapidly. The oestrone elevation was considerably less pronounced the first hours after the vaginal application of oestradiol, but the elevation was sustained and after a few hours at the same level as oestradiol. This is quite different from results reported by Rigg et al. (22, 23). They administered oestradiol in a cream base and suspended in normal saline intravaginally and found relatively smaller increments of oestrone levels as compared to oestradiol levels. This pattern with oestradiol levels higher than the oestrone levels was sustained for 24 hours. Further, the cream vehicle retarded the vaginal absorption of oestradiol. They found that the oestrone and oestradiol levels obtained with 2 mg oestradiol cream were achieved with 0.5 mg oestradiol suspended in normal saline. Schiff et al. (24) administered oestradiol suspended in normal saline vaginally and reported results in agreement with Rigg et al. (22).

The route of administration obviously is important. Also the vehicular for oestradiol when given vaginally is important.

In this study oestradiol was applied vaginally in a water-soluble base. The absorption was very rapid and effective. However, the plasma oestrogen levels decreased rather rapidly indicating that most of the oestradiol was absorbed shortly after application. In a study using intravaginal silastic rings releasing 0.2 mg oestradiol a day, we found oestrone and oestradiol concentrations higher than in this study with 0.25 mg given as a single dose. Further the plasma levels of oestrone and oestradiol were almost equal and in the same range throughout a 21-day treatment period (10).

In conclusion micronized oestradiol-17β is absorbed when given orally. The absorption is dependent of the particle size of oestradiol. The smaller particle the more rapid and effective the absorption. The most rapid absorption (Fraction I), however, is no advantage since the plasma oestrogen levels are very high the first hours after administration and then decrease rapidly. The medium fractions (Standard + fraction II) are more slowly absorbed, but the plasma oestrogens are evenly elevated for a sustained period of time. The coursest fraction (Fraction III) shows a significant smaller absorption with the peak concentration at 12 hours compared to the other preparations, which peak at 0 - 2 hours.

The vaginal administration of oestradiol has the advantage over the oral route that the intestinal conversion of oestradiol to oestrone is circumvented. The results also point out that the vaginal route of administration is more effective than the oral route. The vehicle for vaginal administration of oestradiol, however, is an important factor. A more retarded vehicle than the water-soluble base used in this study is preferable.

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Address for reprints:

D. Englund, M.D.

Departments of Obstetrics and Gynecology
University Hospital
S-750 14 Uppsala 14

Sweden