Female sex steroid concentrations in the ampullary and isthmic regions of the human fallopian tube and their relationship to plasma concentrations during the menstrual cycle

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The concentrations of estradiol-17β (E_2) and progesterone (P) were measured in the ampullary and isthmic portions of the fallopian tube of nonpregnant menstruating women and the cyclic fluctuations were related to the concentrations of these hormones in plasma. The steroid concentrations were determined by radioimmunoassays. There was no significant difference in the isthmic and ampullary concentrations of either steroid in any of the menstrual phases. The mean value for E_2 was highest in the ovulatory phase and for P during the luteal phase. The tissue (per gm)/plasma (per ml) ratio for the steroid concentrations was above unity in all measurements. The ratio for E_2 was highest (isthmus:12, ampulla:8) in the follicular phase and for P (isthmus:26, ampulla:18) during ovulation. Since these highest ratios were attained when plasma steroid concentrations were relatively low they were interpreted as reflections of a maximal receptor contribution. (AM. J. OBSTET. GYNECOL. 136:986, 1980.)

GENERALLY, it is not known how changes in plasma estrogen and progesterone levels during the menstrual cycle are reflected in their target organs. Recent data on the concentration of these hormones in the endometrium and myometrium indicate that there is an overall poor correlation between plasma and tissue levels.[1-3] There are as yet no data available on the concentrations of these hormones in the fallopian tube of the human or experimental animals. Such data are important not only from the point of view of steroid dynamics in this organ, but also with regard to the fact that estrogen and progesterone govern the motor activity of the tube and thereby probably also govern the transport of the ovum toward the uterus.[4] Steroid-sensitive adrenergic nerves and adrenoceptors in the tube are among the mechanisms which mediate the pattern of tubal motility during the menstrual cycle.[5] In the present study the concentrations of estrogen and progesterone were determined in the ampullary and isthmic regions of the tube during the follicular, ovulatory, and luteal phases of the cycle of menstruating women and were correlated with the steroid concentrations in plasma.

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Material and methods

Material. The fallopian tubes from one or both sides were obtained from 33 menstruating women (31 to 53 years old) subjected to abdominal hysterectomy because of adenomyosis or myoma, or salpingo-oophorectomy for benign ovarian cysts. Material from patients receiving hormone treatment in one form or another was not included.

Morphine chloride (10 mg) and scopolamine bro-

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mide (0.4 mg) were given as preanesthetic medication. Anesthesia was induced by pentothal sodium administrated intravenously, followed by inhalation of a mixture of dinitrous oxide and oxygen together with pethidine chloride intravenously. Succinylcholine iodide was given initially as muscle relaxant, followed by pancuronium bromide.

Immediately after removal of the tubes, surrounding connective tissue was dissected away from the muscle wall. A thin (2 mm outside diameter) probe was introduced into the lumen from the ampullary end in order to localize the ampullary-isthmic junction, where the oviduct was divided. A few millimeters of the adjacent isthmic and ampullary portions, as well as the infundibular part of the tube, were discarded. The remainder was lightly blotted on filter paper, weighed, and stored frozen (−18° C) until analyzed.

Venous blood (approximately 10 ml) was drawn from each woman during the operation into heparinized tubes and centrifuged. The plasma was stored frozen at −18° C.

Tissue digestion and extraction of steroids. The frozen tissues were thawed and then digested in a 0.5 ml mixture containing 5% sodium dodecyl sulfate (SDS) and 0.5N NaOH as described previously.[6] The digested material was extracted three times with 3 volumes of ethyl acetate. The combined extracts were evaporated to dryness under air at +40° C. The dried extract, however, contained a significant amount of SDS, which was removed before radioimmunoassay (RIA) by Sephadex LH-20 chromatography, and 6 ml eluate was collected.[6, 7]

Radioimmunoassay. After evaporation of the collected eluate, the residue was dissolved in 1 ml ethyl acetate and suitable amounts, depending on the predicted concentration, of 17β-estradiol (E_2) and progesterone (P) were taken for the respective RIA.

The procedure for RIA of E_2 was that described by Lindberg and associates.[8] The reliability of this assay for determination of E_2 in tissue extracts has previously been checked.[7] The recovery of authentic unlabeled E_2 added to digested samples and calculated at the end of the completed procedure was 89.8% ± 4.2 (mean ± SEM). The intra-assay coefficient of variation in replicate analysis of E_2 was about 10%. Assays were done in a single batch.

The method for P determination was essentially similar to that described by Youssefnejadian and colleagues.[9] The antiserum (FO 22.5.73), which was a gift from Dr. Kjell Martinsson (Royal College of Veterinary Medicine, Stockholm) and found to be highly specific for P, was used in a dilution of 1/1,500 (vol/vol). Other details have been described previously.[6] There was an

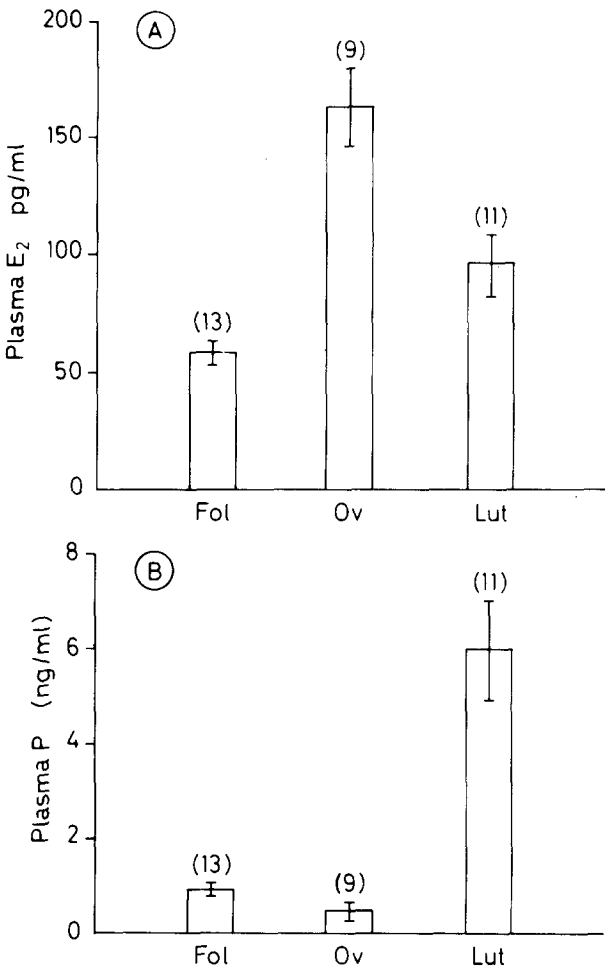

Fig. 1. Plasma concentration of *A*, estradiol-17β and *B*, progesterone during the follicular *(Fol)*, ovulatory *(Ov)*, and luteal *(Lut)* phases. Mean values ± SEM, number of determinations within parenthesis.

almost complete recovery of not only radiolabeled P, but also of unlabeled P (96.5% ± 4.9) added to digested tissue and determined by RIA after extraction.[6]

Protein concentration in the digested tissue was determined in the tissue by Lowry and associates[10] using bovine serum albumin, dissolved in digestion mixture as standard.

Also, E_2 and P concentrations in plasma were determined by these radioimmunoassays.

Definition of cycle stage. The material was divided into three groups with regard to the phase of the menstrual cycle on the basis of plasma steroid levels. The follicular phase included women with plasma $P \leq 2$ ng/ml and $E_2 \leq 100$ pg/ml, the ovulatory phase, women with the same plasma P levels, but $E_2 > 100$ pg/ml, and the luteal phase, those having $P > 2$ ng/ml. The mean values for the plasma steroid levels in the three groups are illustrated in Fig. 1. Whenever mate-

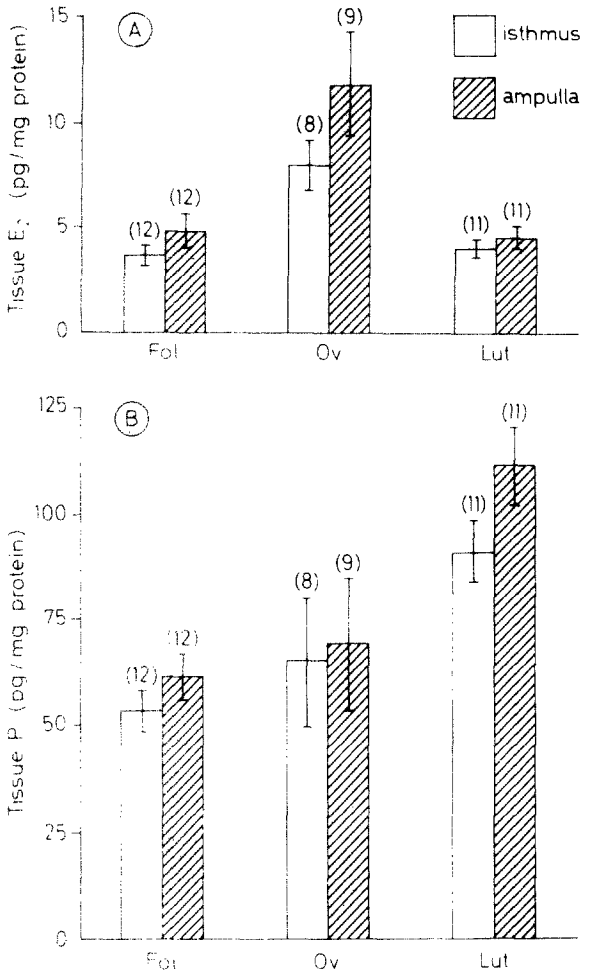

Fig. 2. Tissue concentrations of *A*, estradiol-17β and *B*, progesterone during the follicular *(Fol)*, ovulatory *(Ov)*, and luteal *(Lut)* phases. Mean values ± SEM, number of determinations within parenthesis.

rial was available the cyclic stage was confirmed on hematoxylin-eosin–stained endometrial biopsies. The mean ages of the women in the groups thus defined showed no statistically significant differences from one another.

Chemicals. SDS was purchased from Sigma Chemical Co. E_2 and P were purchased from Ikapharm, Israel. Radioactive [2,4,6,7-H^3] estradiol (102 Ci/mmol) and [1,2,6,7-H^3] progesterone (105 Ci/mmol) were obtained from New England Nuclear Corporation, and Sephadex LH-20 from Pharmacia Fine Chemicals, Sweden.

Statistics. Mean values for the steroid and protein concentrations in the various groups were compared using the unpaired Student t test, and the levels in the ampulla and isthmus and left and right tubes, respectively, compared in the paired t test. Linear regression analysis was done with the least square method.

Table I. Protein content per gram of tissue in the human fallopian tube at different phases of the menstrual cycle

	Protein (mg/gm wet weight)	
Phase	*Isthmus*	*Ampulla*
Follicular	188.4 ± 16.9 (13)	95.3 ± 5.3 (13)
Ovulatory	143.1 ± 12.8 (8)	91.7 ± 7.2 (9)
Luteal	167.5 ± 12.8 (11)	97.0 ± 8.0 (11)

Mean values ± SEM: number of determinations in parenthesis.

Table II. E_2 and P concentrations (pg/mg protein) in right and left fallopian tubes from 5 women

Isthmus E_2		*Ampulla E_2*		*Isthmus P*		*Ampulla P*	
Right	*Left*	*Right*	*Left*	*Right*	*Left*	*Right*	*Left*
3.9	2.5	1.6	1.1	39	38	40	38
2.5	1.9	1.9	2.1	54	50	54	54
4.6	3.7	2.7	3.0	61	43	43	55
8.1	8.0	10.1	12.5	68	64	63	74
3.0	2.0	3.0	3.0	48	37	38	38

Paired t test showed no statistically significant difference between the concentrations in the right and left tube.

Results

There was a marked difference in the protein content per unit of weight in the isthmic compared to the ampullary region of the fallopian tube (Table I), although the difference between the various cyclic stages was not statistically significant. The values for the concentration of E_2 and P are therefore expressed on the basis of tissue protein. There was, however, in both regions an overall good correlation between steroid concentrations expressed on weight basis and on the basis of tissue protein content ($r = 0.76$ to 0.96 for the two regions during the various phases). The steroid concentrations (amount per protein content) in the two regions are shown in Fig. 2. There was no significant difference in the concentrations between the ampulla and isthmus (paired t test) at any of the menstrual phases. The mean values for the tissue concentration of E_2 were highest both in the ampulla and the isthmus around the time of ovulation. On the other hand, P showed a slight insignificant rise from the follicular to the ovulation period, whereas in the luteal phase a pronounced and significant increase was found (Fig. 2). There was a high degree of linear correlation between the ampulla and isthmus with regard to the concentration of E_2 and P ($r = 0.89$ and 0.97, respectively) during the ovulation phase in contrast to the follicular ($r = 0.43$ and 0.54) and luteal ($r = 0.25$ and 0.04) phases. In five cases the steroid concentration was determined separately in the two tubes from individual

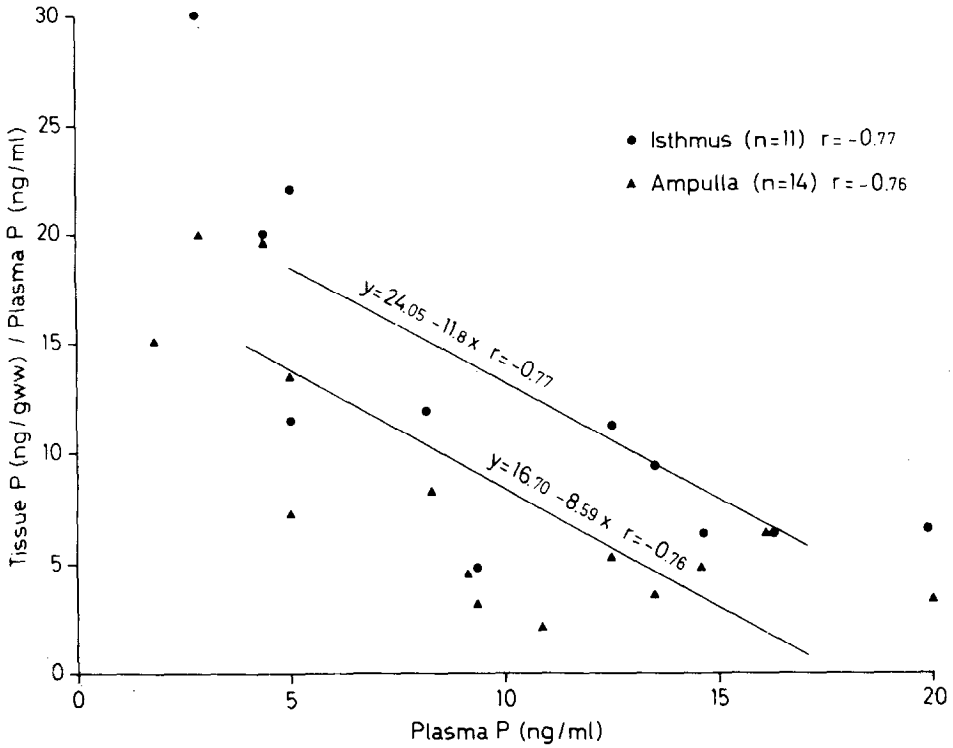

Fig. 3. Correlation between plasma progesterone and tissue/plasma progesterone ratio in follicular phase for isthmus (•) and ampulla (▲) in the linear regression analysis. The equations for the lines and the correlation coefficient (r) are given. The number of women from which the material was obtained is given in parenthesis.

Table III. Ratios between the tissue and plasma concentrations of E_2 and P in the fallopian tube (mean ± SEM) and the correlation coefficient (r) in the linear regression analysis of the tissue versus plasma concentrations of the steroids

		Isthmus		*Ampulla*		*Isthmus*		*Ampulla*	
Phase	*N*	*E_2 ratio*	*r*	*E_2 ratio*	*r*	*P ratio*	*r*	*P ratio*	*r*
Follicular	12	12.6 ± 2.0	0.03	7.8 ± 1.1	0.57	14.4 ± 2.7	0.50	8.9 ± 1.7	0.71
Ovulatory	8	6.7 ± 1.2	0.40	6.2 ± 1.0	0.39	25.7 ± 4.6	0.91	17.9 ± 4.0	0.81
Luteal	11	8.2 ± 1.4	0.21	4.8 ± 0.7	0.31	3.4 ± 0.5	0.53	2.3 ± 0.4	0.16

N = number of tissues analyzed.

patients (Table II). No significant side-to-side difference was found irrespective of cyclic stage.

The ratios between tissue and plasma steroids in the two regions at the various menstrual stages are presented in Table III. It can be seen for both the isthmus and ampulla that, whereas the capacity to accumulate E_2 was most prominent during the follicular phase, it was highest in the ovulation phase for P. As also shown in Table III, this agrees with a high degree of linear correlation between the plasma and tissue values for P during the ovulation phase.

It was found that there was a tendency for an inverse relationship between the tissue/plasma ratio for P and the plasma level of the hormone in the individual patients. Assuming the tissue/plasma ratio of P reflected the amount of available P receptors, this ratio was plotted against the plasma P concentration in order to test the possibility of receptor inactivation. Linear regression analysis showed a fairly good correlation between the two parameters for both isthmus and ampulla during the follicular phase. This analysis was not performed for the ovulation phase because of the interference by the high E_2 level, and it was not considered feasible during the luteal period when the levels of P are too high for detection of any variability in P receptors (Fig. 3).

Comment

The tissue concentrations of E_2 and P were measured in the ampullary and isthmic regions of the human oviduct during the follicular, ovulatory, and luteal phases of the menstrual cycle as defined on the basis of plasma steroid levels and confirmed by histologic examination of the endometrium in cases where the operation also included hysterectomy. There are no previous data available on the content of E_2 or P concentration in the human tube. The concentration of either steroid in the tube reported here was largely similar to that reported for the human nonpregnant myometrium by Runnenbaum and colleagues.[3]

However, the P value for tubal tissue during the proliferative phase obtained in this study was about ten times higher than the corresponding myometrial value reported by Runnenbaum and associates,[3] but in the same order of magnitude as that recently found by us (to be published). The discrepancy may be due to an inadequate extraction of P from the myometrial tissue.[6] The cyclic fluctuations of the hormones in the tubal tissue followed the same pattern as that reported for the myometrium presented by Runnenbaum and associates[3] in the myometrium. Our results show that in general the correlation between the tissue and plasma levels is poor, and that there was always a considerably higher steroid level in the tissue than in plasma, although the tissue/plasma ratio for the steroid concentrations varied during various phases. This suggests that the fallopian tube can be considered as a target organ for female sex steroids. Indeed, the existence of estrogen and progesterone receptors has recently been demonstrated in the fallopian tube.[11, 12]

A comparison of the hormone concentrations in the isthmus and ampulla is best made on the basis of protein content, since this was different per unit of wet weight in the two regions. The protein concentration was considerably higher in the isthmus which is not surprising in view of the fact that this is a more muscular tissue compared to the ampulla. In fact, the protein content in the isthmus compares well with that recently found for the myometrium.[13] However, there was no significant difference in the protein concentration of either the ampulla or the isthmus in the various cyclic stages, which justifies the comparison of the steroid levels between different phases in the same tissue. According to this there was no statistically significant difference in either the E_2 or P level of the ampulla and isthmus in any of the phases, although the mean values for the ampulla were always slightly higher than for the isthmus.

The changes in the hormonal concentration in both tissues from one phase to another are best compared in relation to plasma levels, since the concentration in the blood is also changing during the menstrual cycle. We have therefore calculated the tissue/plasma ratio for such a comparison. This ratio was highest for both isthmus and ampulla in the follicular phase for E_2 and in the ovulatory phase for P.

The tissue/plasma ratio of the steroid concentrations can be considered to reflect the steroid accumulating ability of the tissue, which should be influenced to a large degree by the concentration of the receptors for each hormone. This should particularly be the case when the plasma concentrations are relatively low. When plasma hormone concentration is high so would be the concentration in the tissues, but the contribution from binding of hormone to nonspecific sites (not receptors) in the tissue might be considerable under these conditions. Accordingly, in our data the receptor ability would be best expressed during the follicular phase for E_2 and during ovulation for P. In the luteal phase, however, besides the changes in the plasma concentration that would influence the tissue concentration, there might be additional effects of P on E_2 and P receptors. P has been shown to significantly reduce receptors for both E_2 and P in the uterus.[14–16]

The general conclusion from the present data is that although there was a similarity in the patterns of the cyclic fluctuations for the mean steroid concentrations in the tube and plasma, the correlation between the individual plasma and tissue values, as established in the linear regression analysis, was poor.

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