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Metabolic Clearance Rate of Progesterone During Lactation in the Rhesus Monkey[1] (38215)

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In a prior communication (1), we reported that the concentration of progesterone in the ovarian venous effluent of lactating monkeys (*Macaca mulatta*) on day 25 postpartum was not significantly different from that observed during the luteal phase of the menstrual cycle. The concentration of progesterone in peripheral serum, however, was approximately twenty times higher in the luteal phase than during lactation. This study was performed to determine if the low peripheral concentrations of progesterone during lactation might be due to an increased metabolic clearance rate of progesterone.

Materials and Methods. Thirteen experiments, using 6 normally cycling and 5 lactating rhesus monkeys, were performed. Two of the animals were studied in both the follicular and luteal phases of the cycle. All of the nonlactating and 3 of the lactating animals were from the Primate Research Laboratory of the University of Pittsburgh; the remaining 2 lactating animals were studied at the Wisconsin Regional Primate Center using the same reagents, equipment, and protocol. The housing, feeding regimen, and blood sampling techniques have been described previously (2).

Metabolic clearance rates (MCR) were determined by a continuous infusion method similar to that described by Little *et al.* (3). Each animal was acutely restrained in a primate chair; all experiments were started between 0900 and 1300 hr. Progesterone-1, 2, 6, 7-^{3}H (sp act = 96 Ci/m*M*, New England Nuclear, Boston, Massachusetts, 2.5 μCi/ml absolute ethanol) was diluted 1:4 with normal saline, and was infused into the saphenous vein via a catheter (Bard 1917R, 24 in.) connected to a pump (Harvard Apparatus Company, Harvard, Massachusetts) by medical grade Teflon connecting tubing. Constant output of radioactivity at the catheter tip was reached in 30 min, and was maintained for the remainder of each experiment. The reproducibility of the pump flow rate was checked after every third experiment. The amount of radioactive steroid infused was determined at the end of each experiment by collecting 3 consecutive 1 min samples from the catheter tip.

Starting at 90 min after the initiation of the infusion, 3–5 blood samples were taken at 4–27 min intervals by femoral venipuncture from the side opposite the saphenous catheter. The plasma was separated and used for the determination of the progesterone MCR. Less than 2% of the radioactive progesterone was found to be associated with the erythrocytes.

The triplicate 1 min pump output samples were diluted to 25 ml in absolute ethanol. 20 μl (5 ng) progesterone-4-^{14}C, (sp act =

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52.8 mCi/m*M*, New England Nuclear, Boston, Massachusetts) was added to 1 ml aliquots of the diluted pump output samples, and to the steady-state blood samples from each experimental animal in order to correct for procedural losses incurred during the isolation of authentic progesterone. The plasma samples were then extracted with 3 × 7 ml freshly opened anesthetic grade diethyl ether (Squibb) and the supernatants were combined and evaporated to dryness at 45° under a stream of air. The residue of the ether extracts of plasma and the dried residue of the pump output samples were applied to 0.5 × 13.5 cm Sephadex LH-20 columns and eluted with heptane:methanol:ethyl acetate:water (900:75:50:0.1). Progesterone emerged in the 3.5–7.5 ml fraction while its major metabolites, 20α-hydroxypregn-4-en-3-one (4) and 17α-hydroxyprogesterone (5), emerged in the 7.5–15.5 and 10.5–15.0 ml fractions respectively. The eluate containing progesterone was evaporated to dryness and the ^{3}H and ^{14}C content determined by maximal discriminatory simultaneous counting, and corrected for background, cross-over counting, and procedural losses. The mean recovery of progesterone in all chromatographed samples was 73.5% ± 7.4% (SE, $n = 122$). The MCR was calculated as described by Tait (6) where

$$\text{MCR (liters/day)} = \frac{^{3}\text{H-progesterone infused, in cpm/min}}{^{3}\text{H-progesterone concentration in plasma, in cpm/ml}} \times 1.44.$$

Endogenous circulating progesterone was measured by radioimmunoassay (7) in serum samples taken just prior to the beginning of the infusion of ^{3}H-progesterone. The lower limit of detectability of progesterone was 0.1 ng/ml using 100 μl samples. The secretion rate of progesterone (μg/day) was calculated by multiplying the MCR (liters/day) by the circulating level of progesterone (μg/liter).

TABLE I. Metabolic Clearance Rates (MCR) and Production Rates of Progesterone in Normally Cycling and Lactating Rhesus Monkeys.

	Animal	Day of cycle	Serum progesterone, ng/ml	MCR, liters/day	Production rate, μg/day
Follicular phase	675	3	<0.1	321	<32
	600	4	<0.1	312	<31
	542	5	<0.1	259	<26
	655	5	<0.1	285 ± 248/18 (4) [a]	<25
Luteal phase	675	19	7.0	340	2380
	600	19	3.4	261	887
	656	20	6.8	312	2122
	648	21	13.3	303 ± 299/16 (4) [a]	3977
				294 ± 12 (8) [a]	
		Day postpartum			
Lactation	704	11	1.0	321	321
	613	13	<0.1	433	<43
	1557	14	0.4	320	128
	715	18	0.2	433	87
	1555	22	0.6	381 ± 399/26 (5) [a]	239

[a] Mean ± SE (number of observations).

Differences between groups were analyzed by Student's t test.

Results. The MCR of progesterone does not differ in the follicular and luteal phase of the normal menstrual cycle (Table I). This point is reemphasized by the comparison of the follicular and luteal phase MCR's performed on the same animals (600 and 675). Estimates for the production rate of progesterone can be calculated only during the luteal phase of the cycle when circulating progesterone levels are well above the limits of detection of the assay.

The MCR of progesterone in lactating animals is significantly greater than that seen in the follicular phase ($P < 0.02$), or luteal phase ($P < 0.05$), or when the follicular and luteal phase values are combined ($P < 0.02$). The mean production rate of progesterone during lactation is approximately 15 times less than that seen during the luteal phase.

Discussion. The 30% increase in the MCR of progesterone during lactation cannot account for the 20-fold difference between the circulating levels of this hormone during lactation and during the luteal phase (0.2 ng/ml vs 4.1 ng/ml, respectively) (1). Since the production rate of progesterone is at least one order of magnitude greater during the luteal phase than during lactation, while there is no significant difference in the progesterone concentration in the venous drainage of the active ovary (1), a marked diminution in ovarian blood flow must bear the major responsibility for the low circulating progesterone levels observed during lactation.

The reason underlying the increased MCR of progesterone during lactation is unknown, although it is possible that the rhesus mammary gland, as in the goat (8), may show an increased uptake of progesterone during lactation.

Summary. No difference in the MCR of progesterone was observed in the follicular and luteal phases of the menstrual cycle in the rhesus monkey. In lactating animals, however, the MCR was increased by 30%. This increase is insufficient to account for the 20-fold difference in circulating progesterone levels in luteal phase and lactating animals.

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