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Influence of the Parathyroids on Composition of Rat Milk.* (28021)

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The demonstration of an effect of parathyroid hormone on lactation has been, in the past, based on differences produced in the rate of weight gain of the litters(1-7). The first report concerning the effect of this hormone on the composition of milk was that of Manunta and Mureddu(8) who reported an increase in calcium concentration of milk following parathyroid extract administration. Mosimann(9) ascribed the increased calcium in the milk of ewes in which lactation was induced by estrogens to an increase in parathyroid activity. However, recently, Toverud and Munson(10,11) noted a paradoxical increase in concentration of milk calcium of lactating parathyroidectomized rats in spite of a depressed serum calcium level. They ascribed most, but not all, of this increase to an increase in concentration of total milk solids. Administration of parathyroid extract immediately after parathyroidectomy prevented the decreased water content of the milk as well as the reduction in level of serum calcium, but the increase in calcium concentration of the milk was not so clearly prevented. Munson suggested that parathyroid hormone influenced the calcium concentrating mechanism of the mammary gland directly.

Studies were undertaken in this laboratory to determine whether the changes in composition of milk which are known to occur following parathyroidectomy in the rat could be attributed directly to a loss of the influence of parathyroid hormone on the mammary epithelium or are the result of changes in the serum calcium which occur following parathyroidectomy.

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Methods. Holtzman albino rats 2 to 3 months old were parathyroidectomized or sham-operated under light ether anesthesia on the 4th, 11th, 12th, or 13th day of lactation. In the days immediately preceding their milking, groups of rats were given diets which varied in calcium and protein content. Some groups were isolated from their litters immediately following parathyroidectomy, some were suckled for 24 hours, and some were returned to their litters for 9 to 10 days before isolation and milking. After 16 hours isolation from the litter, the unanesthetized rat was milked with a specially designed glass milking tube connected to a motor-driven suction apparatus. In order to insure milk letdown, the rat was injected with 1 USP unit of oxytocin or syntocinon prior to milking. Blood was taken by heart puncture at the conclusion of milking.

Calcium determinations were carried out on milk and serum by a method modified from that of Collip and Clark(12). This procedure involves precipitating the calcium with ammonium oxalate, redissolving in perchloric acid and reading on a Beckman Flame Spectrophotometer. Milk samples from groups 3 and 5 were ashed, dissolved in .5N HCl, and filtered before precipitation with ammonium oxalate. Milk samples from groups 1, 2, 4, 6, and 7 were analyzed for calcium directly by precipitating samples of whole milk with ammonium oxalate.

The percentage of total milk solids was determined by drying the samples to a constant weight in a 37°C oven. All calcium determinations on the milk were corrected for total solids.

The specific treatments of different groups

TABLE I. Effects of Parathyroidectomy on Composition of Rat Milk.

Group	Exp treatment	No. of animals	Plasma Ca, mg/100 ml	% total solids	Mg Ca/g solids
1	Sham	10	10.7 ± .5	26.5 ± .8	15.3 ± 1.1
	Ptx	8	10.7 ± .3	25.3 ± .6	13.9 ± .5
2	Sham	14	Not taken	21.9 ± .6	14.2 ± .4
	Ptx	12	9.8 ± .7	29.3 ± 1.9	14.2 ± .5
3	Sham	14	12.4 ± .3	23.0 ± 1.9	15.3 ± .3
	Ptx	11	9.3 ± .3	27.8 ± 1.3	16.0 ± .3
4	Sham	16	11.4 ± .3	23.4 ± .6	18.7 ± .6
	Ptx	24	9.1 ± .9	27.8 ± 1.2	18.1 ± .5
5	Sham	9	10.2 ± .4	24.5 ± .6	11.8 ± .8
	Ptx	5	8.5 ± .2	36.1 ± 2.9	14.9 ± 1.5
6	Sham	8	11.2 ± .6	21.3 ± 1.7	18.3 ± .5
	Ptx	4	8.1 ± .6	31.6 ± 1.9	14.3 ± .9
7	Sham	7	11.6 ± .4	26.4 ± 1.8	15.5 ± .9
	Ptx	11	8.2 ± .3	34.6 ± 1.3	12.7 ± .8

NOTES:
1. Ptx = parathyroidectomized.
2. Values given with S.E. $= \pm \sqrt{\frac{\Sigma d^2}{n(n-1)}}$.
3. Groups 2 and 3 are essentially duplicates run 6 months apart in which calcium analyses were determined in Group 2 on whole milk samples and in Group 3 from ashed milk samples.

of rats are outlined below:

Group 1—Parathyroidectomized on 13th day of lactation, isolated for 16 hours on normal stock diet, and milked on 14th day.

Groups 2 and 3—Parathyroidectomized on 12th day, returned to litters for 24 hours on normal stock diet, isolated for 16 hours and milked on 14th day.

Group 4—Parathyroidectomized on 13th day, isolated for 16 hours on a calcium-free, protein-free diet, and milked on 14th day.

Group 5—Parathyroidectomized on 12th day, returned to litters on a calcium-free, 30% protein diet, isolated on 13th day and milked on 14th day.

Group 6—Parathyroidectomized on 11th day, isolated on the 12th day on calcium-free, protein-free diet, and milked on 13th day.

Group 7—Parathyroidectomized 9 days before milk samples were obtained. The females were suckled during this interval and were maintained on a normal diet.

A sham-operated control group given identical treatment was run with each of the above groups.

Results. The effects of the progressive changes in plasma calcium on the composition of rat milk are summarized in Table I. A complete description of the various groups is given in the preceding section. The groups are tabulated by treatments which progressively increased the calcium deficiency of the lactating females. The following observations can be made from these results:

(1) Interruption of milk loss to the litter and provision of sufficient dietary calcium prevented any reduction in plasma calcium following parathyroidectomy, and no significant differences in the milk could be demonstrated.

(2) An increase in calcium deficiency produced a decrease in plasma calcium values of the parathyroidectomized rats and an increase in the percent total milk solids.

(3) The milk of the parathyroidectomized rats under extreme calcium deficiency was markedly concentrated and often was released from the gland with difficulty if at all. Fifty percent of the parathyroidectomized rats in Groups 5 and 6 gave no milk.

(4) The effect of parathyroidectomy on calcium concentration of the milk solids was much less clear-cut. There appeared to be a decrease in the calcium concentration of the solids when the rats were maintained on a protein-free diet, and some tendency toward a higher calcium concentration if the diet contained 30% protein. However, when the decrease in plasma calcium was less than 2 mg %, there was no significant effect on the calcium content of the milk solids.

(5) In rats parathyroidectomized 9 days

before milk samples were taken there was a 22% decrease in calcium concentration of the milk solids.

Discussion. Definition of the role of the parathyroid glands in milk secretion was approached primarily by a study of the effects of parathyroidectomy on milk calcium and solids with and without concomitant plasma calcium changes. Experiments were carried out in which it was possible to maintain groups of animals, in the days immediately following parathyroidectomy, with plasma calcium levels ranging from normal to sufficiently low to produce tetanic symptoms. Maintenance was accomplished by varying the calcium and protein content of the diet and by varying the extent to which the lactating female was suckled to alter the calcium deficiency of the animal.

The most consistent effect of parathyroidectomy on rat milk was an increase in the total milk solids which was noted whenever the plasma calcium decreased relative to the controls. However, whether or not this increase in milk solids was accompanied by a net change in the calcium content of the solids depended on the availability of dietary calcium and protein as well as the amount of endogenous calcium removed by the litter.

Generally, the results of these experiments in lactating rats indicate that the effects of parathyroidectomy on milk secretion depend on fluctuations in the serum calcium occasioned by removal of parathormone from the circulation. This suggests that the parathyroid glands affect the quantity and quality of milk secretion indirectly and changes in milk yield, milk solids, and milk calcium which occur following parathyroid removal can be attributed to the consequences of a reduced level of serum calcium on the mammary gland.

Summary. 1. The effect of parathyroidectomy on the composition of rat milk was studied in animals maintained on diets varying in calcium and protein content. 2. The main effect of parathyroidectomy on milk composition was an increase in the total milk solids. However, maintenance of the plasma calcium at the control level by removal of litters and provision of dietary calcium abolished this effect. 3. The effect of parathyroidectomy on the calcium content of the milk solids depended on the magnitude of the decrease in plasma calcium and on the availability of dietary calcium and protein.

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