

Metabolism of Ca^{45} in Blood, Bones, and Young of Lactating Rats Treated with Estradiol.* (23088)

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In dairy cattle, with the rise in milk secretion following parturition, a condition known as "milk fever" or "parturient-paresis" may occur. In these animals both serum calcium and phosphorus are low with the result that the animal passes into a state of coma. Injection of calcium salt brings about rapid and dramatic recovery. The cause of this condition has been attributed to rapid withdrawal of blood calcium and phosphorus in synthesis of milk by the mammary gland before the parathyroid hormone secretion rate can increase to meet the greatly increased need of calcium by the mammary glands(1). In the previous study(2), the data indicated that estrogen may play a more important role in mobilizing serum calcium than had been appreciated previously. It is well known that increasing amounts of estrogen are secreted with the advance of pregnancy in dairy cattle and other mammals(3). However, following parturition and removal of the placenta, the secretion of estrogen (as shown by the excretion of estrogen in the urine and feces) is greatly reduced.

It seemed of interest, therefore, to study the calcium metabolism of normal lactating rats and compare them with similar lactating rats injected daily with an amount of estrogen estimated to equal that secreted in late pregnancy(4).

Procedure. Twelve pregnant albino rats were used. During the pre- and post-partum period, the animals were fed as previously described(2). As soon as possible after parturition (1-10 hours) the number of sucklings was reduced to 6. Six mothers were kept as controls and 6 received daily sub-

cutaneous injections for 4 days of 1.5 μg of estradiol benzoate in 0.35 ml of olive oil. All mothers were injected intraperitoneally with total dose of 2.65 μc of Ca^{45} /100 g body weight, in the form of CaCl_2 (Specific activity >0.2 mc/g of Ca) over a 3 day period, starting on day of parturition. The daily gain in litter weights was recorded. Five days after parturition the mothers were killed by exsanguination from the left carotid artery. Blood and bones were treated as previously described(2). The offspring were eviscerated to eliminate variation in radioactivity that might be due to difference in amounts of milk present in the digestive system. Each newborn was digested in 5 ml hydrochloric acid. The filtrate was diluted to 25 ml with distilled water as described for bone, and 2 aliquots of each solution (1 newborn) measured for radioactivity. An effort was made to match the weights of the 2 groups of mothers. Corrections were not made for sample self-absorption. The *in vitro* addition of 17 μc of Ca^{45} to 1 ml of normal digested femur of 0.750 g (same procedure as above) gave 10.400 scintillations/minute.

Results. Average gain in weight of litters in the 2 groups was similar, indicating that estrogen did not influence secretion of milk adversely (Table I). Data for control and estrogen-treated groups show that radioactivity (Ca^{45}) in both serum and bone of the treated group was significantly higher than the controls. It would appear that in spite of intense lactation and rapid withdrawal of Ca^{45} in the milk, estrogen favorably influenced retention of Ca in the bones and in the blood serum as shown in our previous work(2). However, in the male rat the retention of Ca^{45} in bones was about twice as great as in the lactating female even though the amount of Ca^{45} injected was relatively higher. The radioactivity in blood serum in lactating animals was much lower also as compared to the male. These differences between male and lactating

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TABLE I. Effect of Estradiol upon Calcium Metabolism of Lactating Rats Injected with Ca^{45} . (Animals sacrificed on 5th day of lactation.)

Body wt, g	Serum count, min./ml	Bone count, min./g	Gain wt, g	Radioactivity count/min./g
Control mothers			Litters of 6	
255	55	7579	20.0	7737
236	60	8608	18.0	7002
196	48	7789	18.0	8747
176	41	9950	22.5	6803
166	50	7679	20.5	6639
146	53	10192	17.0	5847
Avg 195.8	51	8632	19.3	7129
Estrogen-treated mothers			Litters of 6	
238	81	11839	28.0	7563
213	84	8525	22.8	7815
191	68	13694	18.7	5969
166	74	19227	17.2	5639
164	73	12660	16.6	8167
144	72	16666	15.5	6219
Avg 186.0	75	12676	19.8	6895

female are due to the large amounts of calcium secreted in the milk.

Due to greater retention of Ca^{45} in the lactating rat treated with estrogen, it might have been expected that litters from treated groups would have shown lower radioactivity. However, the differences were slight. The lack of difference between the two groups may be due to adequacy of the calcium in milk in either group or it might be due, in part, to the presence of estrogen in milk which might favorably influence the retention of calcium in the nurslings.

Radioactivity in one femur represented 8% of total radioactivity in the body. The amount of radioactivity in the total litter was higher than that of the mother.

Discussion. The present experiment in conjunction with previous observations(2) suggests that estrogen plays a role in calcium metabolism hitherto unsuspected. Estrogen aids in the maintenance of higher levels of

Ca^{45} in the blood and in lactating rats permitted greater deposition of calcium in bones in spite of great withdrawal of calcium in the milk. The mechanism of its action is not known but the observations suggest the possibilities of a higher renal or fecal threshold for calcium. Since Ca^{45} was injected intraperitoneally, effect of estrogen upon calcium absorption in the digestive tract was eliminated.

If estrogen has a similar effect upon calcium metabolism of the dairy cow, administration of estrogen post-partum to maintain the estrogen level at that time might aid in prevention of parturient paresis caused by the rapid decline in blood calcium associated with increasing milk secretion.

Summary. Estrogen, in an amount estimated to equal that secreted in late pregnancy, was injected daily into lactating rats for 4 days along with Ca^{45} . On the 5th day, radioactivity of blood serum, bones and young was compared to a similar group of lactating rats not treated with estrogen. Average gain in weight of the litters of the 2 groups was similar. However, radioactivity in serum and bones of treated group was significantly higher than in the controls. The radioactivity in litters of the 2 groups was similar. These data indicate that estrogen plays a role in the retention of calcium by the animal body.

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