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Successful Replacement Therapy in Lactating Thyro-parathyroidectomized. Rats.* (25746)

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Successful replacement therapy in thyro-parathyroidectomized (TPEC) rats requires optimal amounts of thyroxine (T4) as well as parathyroid hormone (PTH). Study of T4 secretion rate in lactating rats indicated 3 μg/100 g body weight (BW) was upper normal range(1). When this amount was injected into lactating rats from days 7-13 postpartum, milk yield increased 37% and when maximum removal was insured with oxytocin (OXT) milk yield increased 63.2%(2). With T4 and PTH at levels of 10, 2 x 20 and 3 x 20 USP parathyroid units/day partial replacement therapy of TPEC lactating rats was effected(3). Completely successful replacement therapy is here reported using both litter weight and milk yield on day 14 as indices. Technics are described by which animals with accessory parathyroid tissue (APTT) may be detected.

Material and methods. Primiparous lactating rats of Sprague-Dawley-Rolfsmeyer strain were housed in individual cages, fed Purina Lab Chow and water *ad lib.* Animal room was maintained at uniform temperature of 78 ± 1°F. Shortly after delivery, each litter was reduced to 6 young. TPEC was performed under ether anesthesia in less than 20 minutes on day 7 postpartum (PP). Replacement therapy was started shortly after operation: 3 μg T4/100 g/day and 2 x 40 USP units PTH‡/day were given subcutaneously. Parameters of lactational performance were mean litter weight and milk yield. Amount of milk removed by litter of 6 on day 14 under the influence of 1 USP unit oxytocin (OXT)§ was estimated by method described previously(4). To repeat this test at 24 hour intervals litters were weighed at end of 10 hours isolation, immediately replaced and allowed to nurse for 30 minutes. Litters were weighed again to nearest 0.5 g. Difference in litter BW before and after each nursing was used to represent amount of milk secreted during previous 10 hours. After this test, litters were allowed to stay over-night with mothers. To prevent feed intake other than mother's milk by young, mothers were allowed to eat only during 10 hour separation period from day 15 on. PTH was discontinued 5 hours prior to nursing period in 22 animals on day 14, in 12 animals morning of day 13.

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‡ Parathyroid hormone kindly supplied by Eli Lilly & Co., Indianapolis, Ind.

§ Oxytocin supplied by Armour Lab., Chicago.

Results. TPEC rats given replacement therapy of 3 μg T4/100 g/day and 2 x 40 USP units PTH/day from day 7-14 of lactation raised litters of 6 young successfully, reaching normal litter weights although weight gain from day 6-13, 14 was lower because of extremely high litter weights on day 6 (Table I). Similarly, lactational performance on day

TABLE I. Successful Parathyroid Hormone Replacement Therapy in Thyro-parathyroidectomized Rats.

*	Treatment (T4, PTH: Day 7-14, 15; OXT: Day 14, 15)	No. of rats	Killed on day	Day 6 total, g	Mothers' avg BW Day 14 Total, g	Mothers' avg BW Day 14 Gain or loss, %
A.	Control, no treatment†	30	14	297	300	+ 1.1 ± .4
B.	Control‡ T4: 3 μg/100 g/day, day 7-13 OXT: 1 USP unit, day 14	30	14	280	286	+ 2.1 ± .4
C.	TPEC: Day 4§ T4: 3.5 μg/100 g/day, day 4-14 PTH: None OXT: "	8	14	271‖	240	−11.0 ± 3.2¶
D.	TPEC: Day 7 T4: 3 μg/100 g/day, day 7-14 PTH: 2 × 40 USP units/day, day 7-14 OXT: 1 USP unit	10	14	296	275	− 7.3 ± 1.0
E.	TPEC: Day 7 T4: *Idem* PTH: " , day 7-12; 40 USP units day 13 OXT: None Died in tetany	10		283	(7) 242	−12.1 ± 1.6
F.	TPEC: Day 7 T4: *Idem*, day 7-15 PTH: *Idem* OXT: 1 USP unit, day 14, 15 APTT: Survived	2		276	273	− .9
G.	TPEC: Day 7 T4: *Idem* PTH: " , day 7-14 OXT: *Idem* 15 cases tetany, 13 fatal	15		276	271	− 2.0 ± .8
H.	TPEC: Day 7 T4: *Idem* PTH: " OXT: " APTT: Survived	7		282	272	− 3.5 ± 1.6

*	Litters' avg BW Day 6 Total, g	Day 13 Total, g	Day 13 Gain, %	Day 14 Prenursing total, g	Day 14 Gain, %
A.	79			164	112 ± 2
B.	74			166	124 ± 1
C.				93	
D.	86 ± 3[1]	173 ± 5	100 ± 5[5]	171 ± 4	99 ± 5[9]
E.	88 ± 2[2]	159 ± 4	(9) 80 ± 4[6]		
F.	84	168	99	173	106
G.	89 ± 2[3]	167 ± 4	88 ± 3[7]	166 ± 4	87 ± 3[10]
H.	81 ± 2[4]	171 ± 6	110 ± 6[8]	173 ± 6	113 ± 5[11]

(Continued on next page)

TABLE I (continued).

*	Avg milk yield — Day 14: Total, g	Day 14: Total as % of postnursing LW	Day 14: Increase over normal, %	Day 15: Total, g	Day 15: Total as % of postnursing LW
A.	6.2	3.8 ± .3			
B.	11.0	6.2 ± .1	63		
C.	1.6 ± .4	1.7 ± .3			
D.	10.6 ± .7	5.8 ± .2	53		
E.	No milk yield				
F.	8.0	4.4	18	13.0 (11)	6.9 (11)
G.	$10.2 \pm .4^{12}$	$5.8 \pm .2^{14}$	53	$12.2 \pm .8^{16}$	$6.8 \pm .4^{18}$
H.	$10.6 \pm .8^{13}$	$5.7 \pm .3^{15}$	50	$13.1 \pm .8^{17}$	$7.0 \pm .3^{19}$

* In the second and third parts of the Table, letters alone (A, B, C, etc.) are used for treatment, No. of rats and day killed.

† Data from Grosvenor & Turner(5). ‡ Data from Grosvenor & Turner(2). § Data from von Berswordt-Wallrabe & Turner(3). ‖ Day 4. ¶ Day 4-14.

Figures in parentheses = No. of rats surviving, healthy.

APTT, accessory parathyroid tissue; TPEC, thyro-parathyroidectomy; BW, body wt; LW, litter wt; OXT, oxytocin; PTH, parathyroid hormone; T4, l-thyroxine.

Student's "t" test

	p value		*p value*
6–8, 7–8, 10–11	<.001	5–7, 12–16, 14–18, 15–19	.01 < P <.025
5–6	.001 < p <.005	2–4, 3–4, 13–17	.025 < p <.05

14 aided by OXT was comparable to milk yield of normal rats administered T4 plus OXT.

To determine completeness of operation, PTH was withdrawn on day 13 in 1 group. Ten of 12 animals had fatal attacks of tetany within 6 to 18 hours which caused depressed body and litter weights. These animals were not separated from young. On day 14 mothers of a second group were separated from litters for 10 hours for milk yield test. Last PTH injection was given 5 hours before this test. Of 22 animals, 13 had fatal attacks of tetany and 2 others severe attacks between days 15 to 18. Animals free of tetany until day 18 were assumed to possess APTT. Of the group of completely parathyroidectomized rats, milk yield test was repeated on day 15. In 4 animals showing heavy signs of tetany, only 6.2 g milk was produced whereas in 11 others, milk yield was significantly greater than on day 14. The same was true of group of rats with presumed APTT.

All operated mother rats lost body weight during lactation in contrast to normal lactating rats which gain slightly.

Discussion. These data demonstrate that PTH plays an important role in the lactation process. Limitations in amount of hormone administered reduced capacity of rats to secrete milk normally(3) whereas with 2 x 40 USP units/day lactation levels higher than those of normal rats were attained. This was made possible by concurrent administration of T4 (3 μg/100 g/day). Lactation sustained by replacement therapy was actually in excess of that which might have been produced by animals on lower level of T4.

Amount of PTH which successfully maintained lactation may be considered tentatively as estimated equivalent PTH secretion rate of this strain of lactating rats (29-30 USP units/100 g/day).

Although discontinuation of PTH in 1 group on day 13 proved that successful replacement therapy was achieved, it was not possible to run a test for milk yield on day 14, because all animals which did not have APPT

succumbed to tetany in less than 24 hours, whereas at lower levels withdrawal of PTH did not generally result in fatal attacks of tetany by this time(3). Another group of animals was injected until day 14 with PTH. Young were separated from mothers for 10 hours and milk yield was determined. Survival time of mothers was considerably prolonged and milk yield on day 15 obtained by same technic was significantly higher in animals not yet affected by tetany. Increased survival time may be explained by gradual inhibition of lactation and corresponding reduction in blood Ca removal during 10 hour separation period, whereas in previous group, young nursed continuously. More difficult to explain is the mean increase in milk yield on day 15 as compared to day 14, 29 hours after last injection of PTH. The increase was observed even in animals which then died in tetany a few hours later.

Summary. Thyroparathyroidectomy was performed in 44 lactating rats on day 7 postpartum. Successful replacement therapy was started immediately after surgery with optimal level of T4 (3 μg/100 g/day), 2 x 40 USP units PTH/day (corresponding to 29-30 USP units/100 g/day) and 1 USP unit OXT for complete milk removal on days 14-18. Litter weight and milk yield on day 14 reached levels of normal animals given optimal amounts of T4 (3 μg/100 g/day) and 1 USP unit OXT for milk yield test. Discontinuation of PTH on day 13 resulted, within 6-18 hours, in 10 fatal attacks of tetany in a group of 12. Withdrawal of PTH on day 14 coincided with 10 hour isolation period for milk yield test. A group of 15 such animals survived considerably longer than previous group, due, it is believed, to gradual inhibition of lactation and reduction in blood Ca removed during separation period. In 11 rats which succumbed to tetany later, milk yield was significantly higher on day 15 as compared to day 14, 29 hours after last PTH injection. Nine rats which lactated normally without exhibiting signs of tetany during 4 day period after withdrawal of PTH, were considered to have (accessory) parathyroid tissue. It is tentatively proposed to consider 29-30 USP units of PTH/100 g/day as estimate of parathyroid secretion rate in this strain of intensively lactating rats under beneficial influence of optimal T4 level.

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Non-Obstructive *Escherichia coli* Pyelonephritis in the Rat.* (25747)

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Pyelonephritis has not resulted from intravenous injection of coliform organisms in the experimental animal, in most reported studies, unless preceded by renal injury(1,2,3,4). A "virulent" strain of *E. coli* previously described(5) will, however, produce renal infection in some normal rats when injected intravenously. Pyelonephritis produced by *E. coli* tends to run a self-limited course of only a few weeks even though damage and inflammation are said to persist after infection has terminated(6). The course of pyelonephritis in the animal has been assessed by postmortem cultural and histological methods and, in some instances, by infrequent urine samples obtained by laparotomy and needle aspiration of bladder(3,6,7). "Clean-voided" urine collections with quantitative urine cultures have come into widespread use for evaluation of

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