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Survival of Litters from Adrenalectomized Rats Treated with Cortico-Adrenal Substitutes.

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Rats adrenalectomized several days prior to parturition will deliver normal litters but will not lactate sufficiently to raise them, if untreated, unless accessory adrenal cortical tissue is present.[1] Nelson and Gaunt[2] and Reece[3] have shown the adrenal cortical hormone to be a necessary factor in the initiation of lactation.

Preparations of the corpus luteum hormone[4, 5, 6, 7, 8] will prolong the survival period of young adrenalectomized rats. It has also been reported that adrenalectomized rats with ovaries heavily luteinized by gonadotropic hormones[7, 9] survive longer than untreated adrenalectomized controls. NaCl and other salts will help support life and lactation in adrenalectomized animals. The salt therapy is enhanced by injections of adrenal cortical hormone, while the cortical hormone alone in adequate dosage can maintain life and lactation in adrenalectomized rats.[10] The effect of testosterone propionate, directly or indirectly, on the female reproductive system[11, 12] suggested its use as a cortico-adrenal substitute.

Pregnant albino rats were bilaterally adrenalectomized on the 17th to 21st day of gestation. After delivery control and experi-

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2 Nelson, W. O., and Gaunt, R., PROC. SOC. EXP. BIOL. AND MED., 1937, **36**, 136.

3 Reece, R. P., PROC. SOC. EXP. BIOL. AND MED., 1939, **40**, 25.

4 Gaunt, R., and Hays, H. W., *Am. J. Physiol.*, 1938, **124**, 767.

5 Gaunt, R., Nelson, W. O., and Loomis, E., PROC. SOC. EXP. BIOL. AND MED., 1938, **39**, 319.

6 Greene, R. R., Wells, J. A., and Ivy, A. C., PROC. SOC. EXP. BIOL. AND MED., 1939, **40**, 83.

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8 Schwabe, E. L., and Emery, F. E., PROC. SOC. EXP. BIOL. AND MED., 1939, **40**, 383.

9 D'Amour, M. C., and D'Amour, F. E., PROC. SOC. EXP. BIOL. AND MED., 1939, **40**, 417.

10 Gaunt, R., and Tobin, C. E., *Am. J. Physiol.*, 1936, **115**, 588.

11 Starkey, W. F., and Leathem, J. H., PROC. SOC. EXP. BIOL. AND MED., 1938, **39**, 218.

12 Nathanson, I. T., Franseen, C. C., and Sweeney, A. R., Jr., PROC. SOC. EXP. BIOL. AND MED., 1938, **39**, 385.

mental litters were reduced to 5 young each to make their comparative growth records uniform. Since all the adrenalectomized females showed maternal care and attempted to suckle their litters, the day of death of the last rat in each litter was considered the time of complete lactation failure.

I. Normal lactation. No cases of lactation failure have been noted in this colony. As a standard for normal lactation, daily weight records were made on the litters of 5 unoperated, untreated mothers for 25 days. The average weight per young was 40.2 g on the 25th day.

II. Ovariectomized—untreated. To determine if the luteinization of the ovaries after delivery is necessary for normal lactation, 5 females were bilaterally ovariectomized on the day of delivery. One suckling rat was lost from each of 4 of these litters during the 25 day period. The average weight of the young surviving to the 25th day was 40.8 g. The loss of young from some of these litters indicates a slight disturbance in maternal care or lactation. The litters which survived to the 25th day were within the normal weight range of the controls.

III. Adrenalectomized—untreated. Since the survival[13] and lactation of untreated adrenalectomized rats varies in different colonies, ten females were adrenalectomized as untreated controls. These animals survived an average of 13.6 (8-17 days). Three of the 10 absorbed their young. The average survival of the remaining 7 litters was 5.7 (3-8) days.

IV. Adrenalectomized—1.05% or 2.0% NaCl drinking solutions. Seven females which were given 1.05% or 2.0% NaCl solutions to drink, survived an average of 37.3 (8-77) days and their litters 6.5 (3-10) days. Three of these females died on the 37th, 67th and 77th day after adrenalectomy, 3, 4 and 12 days respectively after the mothers were given tap water to drink in place of the salt solutions. The short survival of these litters indicates that although the NaCl solutions did prolong the survival time of the mothers, this therapy is not a complete substitute for the adrenal cortex of lactating rats.

V. *Adrenalectomized—crystalline progesterone.** Five females survived an average of 17.8 (10-29) days and their litters 6.6 (2-11) days when given single daily injections of crystalline progesterone totalling 2 mg over 2 days; 7 mg over 3 days; 22 mg over

[13] Gaunt, R., Gaunt, J. H., and Tobin, C. E., PROC. SOC. EXP. BIOL. AND MED., 1935, **32**, 888.

6 days; 42 mg over 10 days; and 55 mg over 7 days. The apparent failure of lactation indicated by the early death of the litters was probably due to the androgen-like action of progesterone[14] which would tend to inhibit lactation. The survival time of the females was increased and found to be proportional to the total progesterone dosage even though the injections were not continued throughout the whole survival period of the females but were stopped with the death of the litters.

VI. *Adrenalectomized—crude progestin.** Five females survived an average of 7.8 (5-10) days and their litters 4.0 (2-5) days with daily injections of progestin totalling 2.4 mg over 4 days; 3.6 mg over 6 days; 10.8 mg over 5 days; 15.2 mg over 6 days; and 22.5 mg over 5 days. Injections were stopped with the death of the litters. The failure of this substitute therapy to support lactation and prolong the survival period of the females was probably due to the estrogenic material and other impurities present in this crude progestin preparation. It has been shown that estrogens are toxic to adrenalectomized animals[4] and the inhibitory effect of estrogens on lactation is well established.

VII. *Adrenalectomized—Antuitrin S.** Nine females were treated with Antuitrin S: one injected with 25 r.u. and one with 50 r.u. on the day before parturition; 4 animals, one injection on the day of parturition: 10 r.u., 20 r.u., 25 r.u., and 50 r.u.; 3 animals, one injection on the day of parturition followed by another injection 2 or 4 days later: 10 and 15 r.u., 25 and 25 r.u., and 50 and 50 r.u. Two of the females which were killed on the 51st day had accessory adrenal cortical tissue, but neither raised its litter beyond the 9th day. One was autopsied on the 8th day after injection, 4 days after her litter died. The other 6 females survived an average of 18.3 (13-30) days and their litters 6.4 (3-13) days. One female of this group which was injected the day before parturition had a survival period of 30 days and her litter, 13 days. The ovaries of these females were heavily luteinized at the time of death or autopsy.

IX. *Adrenalectomized—testosterone propionate.** Five females survived an average of 18.4 (13-31) days and their litters 3.0 (1-5)

[14] Greene, R. R., Burrill, M. W., and Ivy, A. C., *Endocrinol.*, 1929, **24**, 351.

* The author is indebted for the gifts of the following material which made this work possible: Crystalline progesterone and testosterone propionate, Dr. Erwin Schwenk, The Schering Corporation; Antuitrin S, Parke, Davis & Company; crude progestin, Dr. W. M. Allen, The University of Rochester, School of Medicine and Dentistry.

TABLE I.
Survival of Adrenalectomized Females and Their Litters After Cortico-adrenal Substitute Therapy.

Treatment	No. of animals	Avg survival Range of litters, days	Avg survival Range of mother, days
Adrenalect. untreated	10	5.7 (3-8)	13.6 (8-17)
1.05% and 2.0% NaCl sol.	7	6.5 (3-10)	37.3 (8-77)
Crystalline progesterone	5	6.6 (2-11)	17.8 (10-29)
Crude progestin	5	4.0 (2-5)	7.8 (5-10)
Antuitrin S	9	6.4 (3-13)	18.3 (13-30)
Testosterone propionate	5	3.0 (1-5)	18.4 (13-31)

days after daily injections of testosterone propionate (1-1.5 mg daily) totaling 11 mg over 11 days; 12.5 mg over 13 days; 14 mg over 13 days; 14 mg over 15 days; and 15 mg over 16 days. Injections were started one or two days before and continued 10 to 14 days after parturition. The failure of lactation after these injections is similar to that reported by Folley and Kon[15] for normal rats after testosterone propionate injection. This experiment indicates that the dosage of testosterone propionate used can slightly prolong the survival of adrenalectomized mothers.

In other experiments[5, 6, 8, 9] young adrenalectomized rats were used to test the effect of cortico-adrenal substitutes (their survival time being shorter and less variable than that of the older animals used in this experiment). The average length and range of survival of these adult adrenalectomized females (Table I) indicates that the dosage of some of these substitutes for the hormone of the adrenal cortex will prolong their survival time as compared with that of untreated adrenalectomized females. In none of these experiments was the survival time of the litters (Table I) enhanced by this therapy.

Conclusions. These experiments indicate that NaCl drinking solutions, crystalline progesterone, crude progestin, testosterone propionate, and Antuitrin S, in the dosages used as cortico-adrenal substitutes, will not enhance lactation as determined by the survival of the litters from adrenalectomized females. As compared with the survival of untreated adrenalectomized controls, crude progestin reduced the survival time of the females, whereas the other substitutes prolonged the survival time of the females.

[15] Folley, S. J., and Kon, S. K., *Proc. Roy. Soc. London*, 1038, sB, **124**, 476.