

intact, it had previously been found^{9,10} that total doses of 16-48 mg of progesterone with or without estrogen over periods of 8-20 days were ineffective in stimulating mammary development above that of estrogen alone. Inasmuch as the growth obtained with similar doses in the present work with hypophysectomized rats was relatively minimal compared to normal growth, possibly any effect that might have been present in the normal rat would have been masked by the greater overall development of the normal gland. That this is likely is suggested by the fact that doses of 150 mg of progesterone over 10 days in spayed rats, or 225 mg over 15 days in rats spayed 9 days before injections, produced distinct mammary development with lobule formation.¹¹

Other workers have reported negative effects of progesterone on hypophysectomized rat mammary glands.^{4,5} In the latter work a total amount of 100 mg of progesterone over a 10-day period or 200 mg over a 20-day period was used without securing growth. As the weights of the rats employed were some-

what heavier than those used in the experiments reported here, it would seem that the conditions necessary for new growth with progesterone are similar to those required for estrogen stimulation.

Progesterone exhibited androgenic activity in that it prevented the normal rate of atrophy of the testes following hypophysectomy, an action similar to that of testosterone. Although testosterone³ and progesterone were found capable of increasing the thickness of the mammary ducts, testosterone was found to be more potent. A dosage of 6.25 mg of testosterone gave better results than 30 mg of progesterone in this respect.

These studies, while indicating that the hypophysectomized rat mammary gland may be stimulated by certain steroids, emphasize the necessity of adequate pituitary hormones to produce a fully developed mammary gland.

Summary. Hypophysectomized rats were treated immediately or several days after the operation with progesterone alone or in combination with estrogen and with desoxycorticosterone acetate. The mammary glands of these animals were stimulated with progesterone and estrogen alone or in combination only when treatment was begun immediately. The effects of the combined hormones revealed a summation of the individual responses. Desoxycorticosterone was ineffective.

⁹ Selye, H., Browne, J. S. L., and Collip, J. B., *Proc. Soc. Exp. Biol. and Med.*, 1936, **34**, 472.

¹⁰ Astwood, E. B., Geschichter, C. F., and Rausch, E. O., *Am. J. Anat.*, 1937, **61**, 373.

¹¹ Selye, H., *Proc. Soc. Exp. Biol. and Med.*, 1940, **43**, 343.

14326 P

Local Maintenance of Spermatogenesis in Hypophysectomized Rats with Low Dosages of Testosterone from Intratesticular Pellets.*

SAMUEL DVOSKIN. (Introduced by P. E. Smith.)

From the Department of Anatomy, College of Physicians and Surgeons, Columbia University.

The maintenance of spermatogenesis in hypophysectomized rats by androgen administration, first reported by Walsh *et al.*,¹ has been repeatedly confirmed. Reinitiation of

spermatogenesis by this treatment has also been reported by Nelson.² In all of these experiments the androgen has been administered in regions of the body distant from the testes. It seemed not improbable that androgen of testicular origin (testosterone) might

* Aided by a grant, administered by Dr. P. E. Smith, from the Rockefeller Foundation.

¹ Walsh, E. L., Cuyler, W. K., and McCullagh, D. R., *Am. J. Phys.*, 1934, **107**, 508.

² Nelson, W. O., *Anat. Rec.*, 1941, **79**, Suppl., 2, 48.

TABLE I.

Type of pellet	Days treated	Decrease in pellet wt (mg)	No. of animals	Vent. prostate wt, avg and range (mg)	Wt of treated testis, avg and range (mg)	Wt of untreated testis, avg and range (mg)
Cholesterol	20	0.00	3	23 (17-29)	289 (270-312)	296 (279-319)
Testosterone	20	4.46	4	276 (only 1 weighed)	885 (769-990)	868 (758-978)
"	33%	0.73	10	161 (81-269)	851 (642-1093)	570 (391-827)
"	33%	0.73	11	164 (65-250)	524 (338-715)*	507 (351-699)
"	15%	0.25	6	94 (54-110)	746 (674-948)	493 (420-617)
"	5%	0.05	6	57 (37-78)	556 (467-649)	408 (342-475)
"	20%	0.42	2	52 (51-52)	412 (392-431)	203 (202-203)
Untreated normal controls			45	186 (50-310)	1318 (795-1655)†	1302 (758-1638)
Untreated hypophysect. controls			5	16 (8-26)	228 (144-346)†	235 (136-355)

* The pellets were implanted in the subcutaneous tissue of the flanks, not in the testes.

† No pellets implanted.

be quantitatively more effective if administered intratesticularly because of a local or regional action. The greater efficacy of local application of hormones is well established in many conditions. Also, intratesticular administration would simulate, though crudely, the normal liberation of the hormone. It also might furnish evidence as to whether or not androgen in maintaining spermatogenesis acts directly on the testis. The extratesticular administration heretofore employed may be responsible for the relatively high dosages of androgen (usually 1 to 3 mg per day) required to maintain spermatogenesis in hypophysectomized rats.

In the experiments here reported, testosterone was administered intratesticularly by means of pellets.† The pellets were placed just beneath the tunica albuginea of the testis. A wide range of dosages, most of which were much lower than those heretofore reported, were used. In order to secure these low dosages of testosterone, its absorption was slowed by mixing it with cholesterol in varying proportions to produce mixtures containing 5, 10, 15, 20, 25, and 33% testosterone. Pellets containing only testosterone and only cholesterol were also used. The pellets were cylindrical and measured 1.7 mm in diameter by approximately 2 mm in length. They were weighed to the nearest 0.1 mg, and ranged from 4-5 mg. All implantations were made at the time of hypophysectomy into rats of

the Long-Evans strain. They were autopsied after 20 or 40 days. The rats were 70 to 120 days old at the beginning of the experiment, most of them being between 90 and 120 days old.

With pellets containing only testosterone, the weight and structure of the testis not receiving the implant was as well maintained as was the one into which the pellet had been inserted. The accessory sexual glands were much heavier than normal. With a pellet of 33% testosterone, the testis into which it had been implanted was heavier than the opposite testis; its seminiferous tubules were better maintained, but sperm were also present in the other testis. The ventral prostate was maintained at about normal weight. The most pronounced differential effects were obtained with pellets containing the lowest percentages of testosterone used, namely, 5%. With these pellets, the seminiferous tubules of the testis into which an implant had been made contained sperm, whereas those of the other testis showed no spermatozoa whatsoever as in the untreated and cholesterol implanted controls. In some cases the spermatozoa were present throughout the treated testis; in other cases they were more numerous or present only in the neighborhood of the pellet. There was thus a local or regional effect. The size of the ventral prostate was much less than normal. Since the largest of these pellets contained only 250 μ g of testosterone, there could be during a 20-day period an average absorption per day of only 12.5 μ g, even if all the hor-

† The testosterone was kindly supplied by Dr. Erwin Schwenk of the Schering Corporation.

mone were absorbed. The complete absorption of all the testosterone in the pellets seems improbable for the melting point of the center was different from that of the periphery of the removed pellets. Furthermore, the weight loss of these pellets averaged but 0.05 mg (an average of 2.5 μ g per day), though the errors due to weighing and the absorption of substances from the body fluids makes an exact determination of the weight loss impossible.

The local or regional maintenance effected by the lower dosages indicates that testos-

terone acts directly on the testis and not by scrotal or general systemic stimulation.

Summary. Very small dosages of testosterone, administered intratesticularly to hypophysectomized rats, cause a regional maintenance of spermatogenesis. These dosages are much less than those necessary for complete maintenance of the sexual accessory glands. Testosterone has a direct action on the testis in the maintenance of spermatogenesis.

14327

Factors Influencing Ability to Counteract Gravitational Interference with Circulation, in the Rabbit.

ARTHUR LOCKE, R. B. LOCKE, AND A. P. McILROY.

From the Western Pennsylvania Hospital Institute of Pathology, Pittsburgh.

This is a report of a comparative study, in the rabbit, of a series of substances and procedures affecting ability to counteract the gravitational interferences with circulation imposed by the standing position. The substances studied were: paredrine, ephedrine, d-amphetamine, coramine, nicotinic acid amide, ascorbic acid, desiccated thyroid, sulfanilamide, sulfathiazole, and sodium nitrite. The procedures studied were: chilling, salt restriction, and bleeding from the heart and ear.

Method. New Zealand White male rabbits were used, between 3.0 and 4.5 kg in weight. The rabbits were chilled, in cool water,¹ until the rectal temperature had fallen to a point between 94 and 95°F. They were then dried and allowed to warm up again, spontaneously. When the rectal temperature had reached 96°F, they were placed in boxes forcing the rabbits to stand upright for one-half hour.

During this half hour of forced standing, temperature recovery proceeded without retardation in the rabbits able to counteract the

gravitational interferences with circulation imposed by the standing position. In rabbits with an impaired capacity for counteracting gravitational interference, the rate of temperature recovery slowed down during the period of forced standing and, in extreme instances, reached negative values (impending gravitational shock).¹

The extent of the temperature rise during the half hour of forced standing was measured: (1) in the absence of preliminary preparations other than were required to provide a true control for the test determination, and (2) one week later, during the period of action of the substance or procedure under study. Whenever practicable, an additional control determination was done one or 2 weeks after the test determination and averaged with the earlier control determination as a way of minimizing the magnitude of error due to spontaneous change.

An average test rise significantly greater than that observed during the control determinations was considered to be an indication of possible improvement as a result of the action of the substance or procedure tested. A significant decrease in rise was taken as an indication of converse impairment. A change

¹ Locke, A., Locke, R. B., McIlroy, A. P., and Chung, L., *PROC. SOC. EXP. BIOL. AND MED.*, 1942, **51**, 184.