Effects of Thyroxine and Exercise on the Glandular and Plasma Levels of Corticosterone in the Male Rat

B. W. Craig and D. R. Griffith

Wichita State University, Department of Biology, Wichita (Kansas 67208, USA); and Iowa State University, Ames (Iowa, USA), 26 January 1976.

Summary. The results presented here demonstrates that the thyroid gland is essential for normal corticosterone production. They further show that exercise stimulates this production whether the thyroid gland is present or not. The release or metabolism of corticosterone seems dependent upon an intact thyroid gland since plasma levels of corticosterone are decreased during exercise if the thyroid is absent. The administration of thyroxine is not sufficient to renew these levels.

Short-term exercise (3 weeks of swimming) can increase plasma levels of corticosterone in the rat, whereas prolonged exercise (6 weeks of swimming) can decrease plasma corticosterone[1]. These glucocorticoid changes are best demonstrated when exhaustive exercise is employed[2,3]. Prolonged exercise (10 weeks of swimming) has also been shown to decrease cholesterol levels in the blood of rats[4], and it was suggested that this was the result of increased concentrations of thyroid stimulating hormone (TSH). It has been theorized[5] that an inverse secretory mechanism exists between the stimulating hormones or the releasing factors of the thyroid gland and the adrenal cortex. Therefore, if TSH levels are elevated by exercise or any other factor, then adenocorticotropic hormone (ACTH) concentrations will drop. This inverse relationship has been demonstrated in rabbits[6] and cattle[7] but not in the rat[8]. However, it has been established that hypothyroidism within the rat causes a decrease in adrenal cortical activity[9–11]. This study was undertaken to investigate the effects and interactions of exercise and thyroxine on corticosterone levels of the rat.

Materials and methods. Two groups of young (70-day-old) male rats were subdivided into 3 treatment types: control-received no treatment, thyroid intact; euthyroid-thyroidectomized and given 1.0 µg L-thyroxine (T_4)/100 g body weight/day s.c.; and athyroid-thyroidectomized and no replacement given.

Table I. Corticosterone concentrations of adrenal glands (µg/mg)

	NEx	Ex
Control	4.17 ± 0.62[a]	4.83 ± 0.66
Euthyroid	4.79 ± 0.62	5.52 ± 0.62
Athyroid	2.54 ± 0.66	3.67 ± 0.62
lsd	0.456[b]	

[a] Mean ± standard error of that mean ($n = 10$). [b] Least significant difference at $p < 0.05$ for comparison of any two means within the appropriate parameter.

Table II. Corticosterone concentrations of plasma (µg/ml)

Control	0.783 ± 0.078[a]	0.804 ± 0.086
Euthyroid	1.063 ± 0.070	0.928 ± 0.072
Athyroid	0.681 ± 0.070	0.553 ± 0.070
lsd	0.065[b]	

[a] Mean ± standard error of that mean ($n = 9$), [b] Least significant difference at $p < 0.05$ for comparison of any two means within the appropriate parameter.

Each treatment group was separated into nonexercise and exercise animals. The exercised animals were subjected to exhausive exercise by swimming for 5 days a week for a 10-week period. Swimming was carried out in 25 gallon plastic barrels with the water temperature being held constant at 37 ± 1 °C and a wetting agent added to each tank. To insure exhaustive swimming lead weights amounting to 4% of the body weight were added to the tail of each rat, and the animal was allowed to swim until unable to surface after an 8–10 sec period[12]. At the end of the experimental period the animals were anesthesized with nembutal and decaptitated. Both the glandular and plasma concentrations of cortcosterone were analyzed fluorometrically[13]. The data was first compared using an analysis of variance test of significance and then subjected to a least significance difference test.

Results and discussion. Removal of the thyroid gland with no T_4 replacement given (athyroid) causes a significant decrease in glandular (Table I) and plasma (Table II) levels of corticosterone. When T_4 is given (euthyroid) these corticoid concentration are renewed thus indicating the importance of the thyroid gland in adrenal cortical activity. As was pointed out earlier, an inverse secretory relationship between TSH and ACTH does not seem to function in the rat[8]; therefore, the role of thyroxine would be of a stimulatory nature either through ACTH or directly on the adrenal cortex.

Exercise resulted in a significant increase in glandular levels of corticosterone in all 3 of the treatment groups; the largest increase being in athyroid rats and the smallest in control animals. The effects of exercise on plasma levels of corticosterone varies with the treatment. In control animals there is an insignificant increase while the athyroid and euthyroid animals show significant decreases due

[1] R. Freinkl and L. Csalay, J. Sports Med. phys. Fitness *1962*, 2207.
[2] E. F. Korenskaya, Probl. Endokr. *13*, 65 (1967).
[3] T. Suzuki, K. Otsuka, H. Matsui, S. Ohukuzi, K. Sakai and Y. Harada, Endocrinology *80*, 1148 (1967).
[4] J. A. Story and D. R. Griffith, Horm. Metab. Res. *4*, 380 (1972).
[5] G. W. Harris, Ciba Found. Coloq. Endocr. *8*, 531 (1955).
[6] J. Leppaluoto, Acta endocr., Copenh. *70* (Suppl. 165), 7 (1974).
[7] W. J. Vanjonack and H. D. Johnson, Proc. Soc. exp. Biol. Med. *149*, 638 (1975).
[8] P. Ducommun, E. Sakiz and R. Guillemin, Am. J. Physiol. *210*, 1257 (1966).
[9] M. X. Zarrow, L. M. Harger and J. L. McCarthy, Proc. Soc. exp. Biol. Med. *94*, 348 (1957).
[10] J. L. McCarthy, R. C. Corley and M. X. Zarrow, Am. J. Physiol. *197*, 693 (1959).
[11] B. G. Steintz and U. L. Beach, Endocrinology *72*, 45 (1962).
[12] C. A. Dawson and S. M. Horvath, Med. Sci. Sports *2*, 51 (1970).
[13] J. Vander Vies, Acta endocr., Copenhagen *33*, 59 (1960).

to exercise. The results suggest that exercise increases glucocorticoid biosynthesis but does not affect plasma levels in animals with an intact thyroid gland. Exercise only affects the plasma levels of thyroidectomized animals regardless of whether T_4 is present or not. This indicates that the thyroid is important in maintaining corticosterone levels during exercise but that T_4 is not involved in this regulation.

The data presented here demonstrates that the thyroid gland is essential for normal glucocorticoid production. It further shows that exercise stimulates this production in both intact control and thyroidectomized animals. The release or metabolism of corticosterone seems dependent upon an intact thyroid gland since plasma levels of the corticoid are decreased during exercise if the thyroid is absent. The administration of T_4 is not sufficient to renew these levels. At the present time there are no clearcut answers to some questions raised by these findings, but with research we are presently conducting, we hope to provide the needed solutions.

Testosterone in Human Saliva

A. D. LANDMAN, L. M. SANFORD, B. E. HOWLAND, C. DAWES and E. T. PRITCHARD[1]

Department of Oral Biology, The University of Manitoba, Winnipeg (Manitoba R3E OW3, Canada), 14 January 1976.

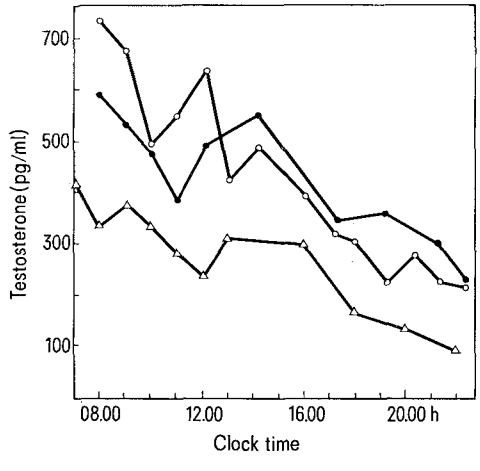
Summary. Testosterone has been detected in whole human saliva. Levels averaged (± SE) 295 ± 36 and 195 ± 25 pg/ml in adult males and females, respectively, and usually were undetectable in children. In adult males, the excretion of testosterone in saliva appeared to follow a circadian rhythm.

Both the structure and function of the submandibular gland in the rodent[2–5] and pig[6–8] are influenced by androgens, indicating that this gland is a target organ for these hormones. Moreover, the submandibular gland in the rat[9,10], mouse[11] and dog[12] exhibits some steroid synthetic ability, involving either the oxidative or reductive metabolism of testosterone. In spite of the numerous investigations of androgen-dependent characteristics of salivary glands and the studies on steroid metabolism by gland tissue, there appear to be no reports of the presence of testosterone in human saliva. There is reason to suspect that the major androgens found in serum might also be secreted in saliva since the glucocorticoids cortisol and cortisone have been reported present in human parotid saliva[13].

The present investigation was designed to determine the influence of sex and age on the concentration of testosterone in whole human saliva, and the possibility of a circadian rhythm in salivary levels of testosterone in the adult male.

Materials and methods. Between 10.00 and 15.00 h, a sample of unstimulated whole saliva (1–2 ml) was collected from each of 12 males (25–55 years of age), 12 females (25–35 years of age) and 12 children (5–10 years of age) of both sexes. To determine if levels of testosterone in saliva varied with time of day, samples were collected from 07.00 to 22.00 h at hourly intervals from 2 adult males. One of the males was sampled on 2 consecutive days. Saliva was stored at −20 °C until assayed for testosterone. Testosterone concentrations were determined in duplicate by radioimmunoassay as previously described[14], except that an antiserum raised in sheep immunized with testosterone-3-carboxy-methyloxime conjugated to bovine serum albumin was employed in the assay system. Differences between mean salivary levels of testosterone in males, females and children were tested for significance by analysis of variance followed by a Duncan's new multiple range test.

Results and discussion. Testosterone in whole saliva averaged (± SE) 295 ± 36 pg/ml for adult males and



The concentration of testosterone in whole saliva from 2 adult males at various times of the day. Saliva samples were collected hourly for 16 h from subject No. 1 on 2 consecutive days (●—●) and (○—○) and from subject No. 2 on 1 day (△—△).

[1] Acknowledgments. This study was supported by grant No. MA-4454 from the Medical Research Council of Canada. The authors wish to thank Mr. D. B. BEATON for excellent technical assistance.

[2] C. R. LEESON, in *Handbook of Physiology*, Section 6: Alimentary Canal (Ed. C. F. CODE; American Physiol. Soc., Washington, D.C. 1967), vol. 2, p. 463.

[3] J. R. BLAIR-WEST, J. P. COGHLAN, D.A. DENTON and R. D. WRIGHT, in *Handbook of Physiology*, Section 6: Alimentary Canal (Ed. C. F. CODE; Amer. Physiol. Soc., Washington, D.C. 1967), vol. 2, p. 633.

[4] E. J. ZEBROWSKI, Archs. oral Biol. *18*, 567 (1973).

[5] E. H. CHARREAU, Acta physiol. latinoam. *19*, 188 (1969).

[6] W. D. BOOTH, M. F. HAY and H. M. DOTT, J. Reprod. Fert. *33*, 163 (1973).

[7] W. D. BOOTH, J. Endocr. *55*, 119 (1972).

[8] W. D. BOOTH, J. Reprod. Fert. *23*, 533 (1970).

[9] J. M. ROSNER, J. C. MACOME and D. P. CARDINALI, Endocrinology *85*, 1000 (1969).

[10] A. BALDI and E. A. CHARREAU, Endocrinology *90*, 1643 (1972).

[11] J. C. COFFEY, Steroids *22*, 247 (1973).

[12] A. L. WEINER, P. OFNER and E. A. SWEENEY, Endocrinology *87*, 406 (1970).

[13] F. H. KATZ and I. L. SHANNON, J. clin. Invest. *48*, 848 (1969).

[14] L. M. SANFORD, J. S. D. WINTER, W. M. PALMER and B. E. HOWLAND, Endocrinology *96*, 627 (1974).