9. West, H. D., Goldie, H., PROC. SOC. EXP. BIOL. AND MED., 1956, v92, 116.
10. Goldie, H., Cooper, E., Chess, R., Freeman, R., *Proc. Cancer Res. Assn.*, 1958, v2, 301.
11. Andrews, G. A., Root, S. W., Kniseley, R. M., *Cancer*, 1953, v6, 294.
12. Kniseley, R. M., Andrews, G. A., *ibid.*, 1952, v6, 303.

Received June 25, 1959. P.S.E.B.M., 1959, v102.

Prevention of Salt Hypertension by Propylthiouracil Treatment in Rats.* (25226)

MELVIN J. FREGLY (Introduced by A. B. Otis)
Dept. of Physiology, College of Medicine, University of Florida, Gainesville

The anti-thyroid drug, propylthiouracil, has been used successfully in rats to prevent development of renal hypertension(1,2); desoxycorticosterone-acetate induced hypertension(3) and adrenal-regeneration hypertension(4). Our studies were to determine whether this drug would also prevent a fourth type of experimental hypertension, namely that accompanying administration of hypertonic NaCl solution(5,6).

Methods. Holtzman rats were kept 2 or 3/cage in thermoregulated room at 25 ± 1°C, illuminated from 8 a.m. to 6 p.m. Blood pressures and body weights were measured during one-week control period and weekly thereafter for duration of experiment. Systolic blood pressure was measured by the microphonic manometer technic(7) but without anesthesia, as described previously(8). Immediately after the control period, 15 male rats (Exp. 1) were given 1.75% NaCl solution and 12 female rats (Exp. 2) 1.9% NaCl solution as sole drinking fluid. Eight rats in Exp. 1 and 6 rats in Exp. 2 received in addition ground Purina chow† containing 0.1% propylthiouracil (PTU)‡ The remainder of the rats received ground chow without the drug. After 11 weeks, all rats in Exp. 1 were killed by ether and heart, kidneys, testes, seminal vesicles, prostate, thyroid, thymus and adrenal glands were removed, trimmed, drained of excess fluid or blood, blotted on filter paper and weighed on torsion balance. Results are expressed as ratio of organ weight to body weight (mg/100 g B.W.). Since all rats in Exp. 2 lost nearly 50 g in 3 weeks, 1.75% NaCl solution was substituted for 1.9% NaCl solution. The substitution seemed to ameliorate weight loss. Only 4 rats in each group survived the total 14 weeks, then they were killed by ether and adrenals, thyroid, thymus and ovaries were removed and weighed as described above. Simultaneous means of control and experimental groups were compared using the "t" test for the 95% confidence limit (9).

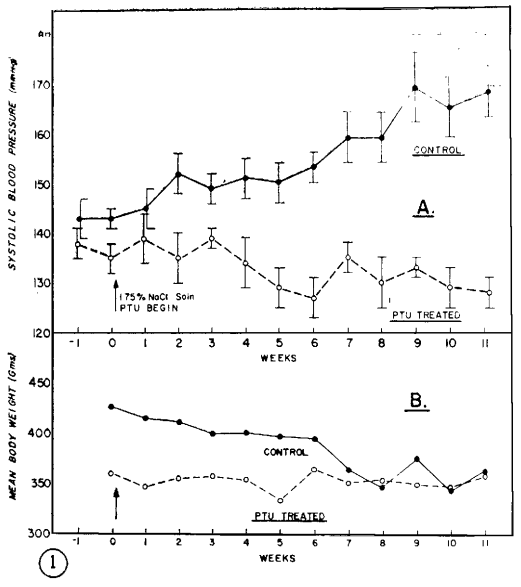
Results. Fig. 1A illustrates that 1.75% NaCl solution given male rats produced a marked elevation in systolic blood pressure from 143 mm Hg to 165 mm Hg at end of Exp. 1. In contrast, rats receiving PTU in addition showed a decrease from 138 mm Hg to 128 mm Hg. Differences were significant ($P = .02$) by the second week and highly significant ($P < .01$) by 5th week. Mean body weight of rats receiving chow and salt fell during the experiment while that of PTU-treated group remained constant (Fig. 1B).

Systolic blood pressures of female rats in Exp. 2 showed results similar to, but more variable than, those of comparably treated groups of male rats in Exp. 1 (Fig. 2A). Control, salt-treated rats increased mean systolic blood pressure from 137 mm Hg to approxi-

* Supported by Grants from Nat. Heart Inst., Bethesda, Md., and The American Heart Assn.

† Analysis of Purina laboratory chow by wet ashing with concentrated nitric acid and subsequently determining sodium and potassium content by flame photometry indicated a sodium content of 127 mEq./Kg and potassium content of 218 mEq./Kg.

‡ 6-n-propyl-2-thiouracil, Nutritional Biochemicals Corp., Cleveland O.



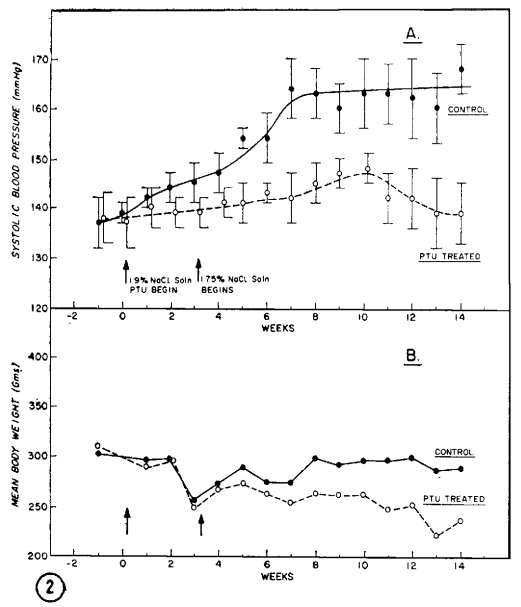


FIG. 1. A. Mean systolic blood pressures of control (●) and propylthiouracil treated (○) male rats throughout experiment. Both groups of rats were given 1.75% NaCl solution as the sole drinking fluid. ± 1 stand. error is set off at each mean. B. Mean body wts of control (●) and propylthiouracil-treated (○) male rats throughout experiment.

FIG. 2. A. Mean systolic blood pressures of control (●) and propylthiouracil treated (○) female rats. Both groups of rats were given 1.9% NaCl solution for 3 wk and then switched to 1.75% NaCl solution for remainder of experiment. ± 1 stand. error is set off at each mean. B. Mean body wts of control (●) and propylthiouracil-treated (○) male rats throughout experiment.

mately 164 mm Hg in 14 weeks. PTU-treated rats showed a maximal rise to 148 mm Hg (10th week). Thereafter, blood pressure fell and by end of experiment was the same as during control period. Differences between the 2 groups were of limited significance ($P < .05$) beginning with week 5 (with exception of weeks 9 and 10).

After change-over from 1.9 to 1.75% NaCl solution, control rats in Exp. 2 regained their lost weight and maintained a relatively constant body weight (Fig. 2B). PTU-treated rats continued to lose weight during the experiment in contrast to their male counterparts in the first experiment.

In Exp. 1, ratios of heart and kidney weight to body weight of the PTU-treated group were significantly ($P < .01$) smaller than those of comparable controls while thyroid and testis weight ratios were significantly larger (Table I). Seminal vesicle, prostate, thymus and adrenal weight ratios of PTU-treated rats were similar to those of controls. Results in female rats of Exp. 2 were similar except that gonad to body weight ratio of the latter was reduced significantly ($P < .01$) by PTU-treatment (30 ± 6.1, control, and 16.1 ± 2.7, PTU).

Discussion. Hypertension developed in male and female rats fed commercial laboratory ration and 1.75% NaCl solution as sole drinking fluid. Blood pressure was significantly elevated within 2 to 5 weeks and became maximal within 8 to 10 weeks (*cf.* also 5, 10, 11). The mechanism resulting in eventual hypertension is not understood but may involve sensitization by salt to circulating pressor agents(12,13) or accumulation of salt within arteriole walls(14) with subsequent reduction of lumen size. Renal lesions are ultimately observed under these conditions(11, 15). Hibbard(16) and Axelrad *et al.*(17) found that ingestion of large amounts of sodium chloride increased thyroid activity as assessed by hyperplasia, histological changes characteristic of hypersecretion and increased radioactive iodine uptake. Increase in thyroid activity may play a role in development of hypertension under these conditions, and the ameliorating effect of PTU would have a logical explanation. This drug also prevented

TABLE I. Organ/Body Weight Ratios of Rats in Exp. 1.

Treatment	No. of rats	Mean body wt (g)	Organ/body wt ratio (mg/100 g body wt)							
			Heart	Kidneys	Testes	S. vesicles	Prostate	Thyroid	Thymus	Adrenals
Chow + 1.75% NaCl sol.	7	342	441.9 ± 35.5*	1013.2 ± 43.8	834.4 ± 42.6	122.0 ± 16.2	90.4 ± 10.8	6.3 ± .6	51.4 ± 18.6	13.8 ± 1.1
			<.01†	<.01	<.01	.26	.23	<.01	.81	.34
PTU + 1.75% NaCl sol.	8	352	292.4 ± 7.9	810.4 ± 33.5	1002.6 ± 32.8	99.1 ± 10.5	112.0 ± 13.1	30.2 ± 2.1	46.5 ± 5.6	12.6 ± .5

* ± 1 stand. error of mean.
† Probability value.

cardiac and renal hypertrophy observed in non-PTU-treated controls. These results support earlier observations in which PTU treatment generally, although not always, prevented cardiac hypertrophy accompanying bilateral renal encapsulation with molded latex envelopes(2).

PTU-administration prevented growth in male and caused weight loss in female rats. In addition, testicular weight ratio increased significantly. Both effects have been reported earlier(1,2) while the latter has also been reported for thiouracil(18). Testicular effect of PTU may be due to increased gonadotrophin secretion by anterior pituitary gland or increased sensitivity of testicular tissue to normal gonadotrophin levels. Others observed increased testicular weight responses to exogenous gonadotrophin administered to thiouracil-treated rats(18) and increased compensatory hypertrophy upon removal of one testis of propylthiouracil-treated rats(19). These observations support the idea that sensitivity of testicular tissue to gonadotrophin increases. If one assumes increased pituitary secretion of gonadotrophin under these conditions, then testicular response may be due to increased secretion of FSH and not a combination of FSH and ICSH (LH), since evidence for increased secretion of testicular hormone was not observed. For example, prostate and seminal vesicle size were normal and kidney size reduced (Table I). Renotrophic effect of testicular hormone was absent(20). If ovarian weight ratio is a measure of FSH secretion, PTU-treated female rats in Exp. 2 showed evidence of decreased FSH secretion rather than an increase. No complete explanation can be given for the difference in gonadal response to PTU between the 2 sexes. Similar unpublished results have been observed previously for surgically thyroidectomized female rats whose kidneys were encapsulated with latex envelopes.

Weight ratios of adrenal and thymus glands in both male and female rats following PTU treatment are well within the range of normal, untreated rats kept under our laboratory conditions(2). These gross measurements suggest that adrenal function was unaltered either by salt treatment or by treatment with both

salt and PTU. Furthermore, it has been shown that salt hypertension develops in absence of adrenal glands(21), and is maintained when adrenal glands are removed after hypertension has developed(22). These results suggest that adrenal glands do not play an active role either in development or maintenance of this type of hypertension.

While the anti-thyroid drug, propylthiouracil, prevents development of salt hypertension, the mechanism by which hypertension is prevented is unknown. Whether the results are due to direct effect of this drug on thyroid gland; to indirect effects of lack of thyroid hormone or to extra-thyroidal effects cannot be stated. However, daily intake of NaCl solution by PTU-treated rats was at least the same as that of control group. Other unpublished studies have shown that spontaneous salt intake of PTU-treated rats is considerably greater than that of normal rats. Studies are being carried out to determine effect of PTU on sodium balance since a natriuretic effect of this drug could account, at least in part, for its anti-hypertensive effect. Other studies are designed to determine whether cardiac output is reduced by PTU administration.

Summary. Dietary administration to rats of the anti-thyroid drug, propylthiouracil, prevented development of hypertension accompanying administration of hypertonic (1.75%) NaCl solution as sole drinking fluid. Propylthiouracil administration prevented not only the expected rise in systolic blood pressure but also cardiac and renal hypertrophy. Although the results suggest the possibility that thyroid gland may play a role in salt hypertension, they could be accounted for by extra-thyroidal effects of the drug.

It is a pleasure to acknowledge technical assistance of W. Pickels.

1. Fregly, M. J., *Am. J. Physiol.,* 1958, v194, 149.
2. Fregly, M. J., Hood, C. I., *Circ. Research,* 1959, v7, 486.
3. Salgado, E., *Endocrin.,* 1954, v55, 377.
4. Chappel, C. I., Rona, G., Revesz, C., Cahill, J., PROC. SOC. EXP. BIOL. AND MED., 1958, v98, 23.
5. Sapirstein, L. A., Brandt, W. L., Drury, D. R., *ibid.,* 1950, v73, 82.
6. Masson, G. M. C., Corcoran, A. C., *Methods in Medical Research,* 1952, v5, 263.
7. Friedman, M., Freed, S. C., PROC. SOC. EXP. BIOL. AND MED., 1949, v70, 670.
8. Fregly, M. J., *Am. J. Physiol.,* 1954, v176, 275.
9. Fisher, R. A., *Statistical Methods for Research Workers* (10th ed.), Hafner, N. Y., 1948, p114.
10. Heroux, O., Dugal, L. P., *Canad. J. Med. Sci.,* 1951, v29, 164.
11. Koletsky, S., *Lab. Invest.,* 1958, v7, 377.
12. Gordon, D. B., Drury, D. R., Shapiro, S., *Am. J Physiol.,* 1953, v175, 123.
13. Vick, J., Ederstrom, H. E., Verger, T., PROC. SOC. EXP. BIOL. AND MED., 1956, v93, 536.
14. Tobian, L., Jr., Binion, J., *J. Clin. Invest.,* 1954, v33, 1407.
15. Grollman, A., *Perspectives in Biol. and Med.,* 1959, v2, 208.
16. Hibbard, J. S., *Arch. Surg.,* 1933, v26, 648.
17. Axelrad, A. A., Leblond, C. P., Isler, H., *Endocrin.,* 1955, v56, 387.
18. Meites, J., Chandrashaker, B., *ibid.,* 1949, v44, 368.
19. Horn, E. H., Lo Monaco, M. B., PROC. SOC. EXP. BIOL. AND MED., 1958, v98, 817.
20. Kochakian, C. D., *Recent Prog. Horm. Res.,* 1947, v1, 177.
21. Fregly, M. J., *The Physiologist,* 1958, v1, 24.
22. Brandt, W. L., Dubin, W. M., Sapirstein, L. A., *Am. J. Physiol.,* 1951, v164, 73.

Received June 29, 1959. P.S.E.B.M., 1959, v102.