most recently emphasized by Teshan(6) the types of experiment in which renal functions and urine flow are rapidly changing, put clearance methods at their greatest disadvantage. This is particularly true when hemorrhagic shock and mannitol infusions are both important parts of the protocol. Direct or electromagnetic flowmeter measurements of renal blood flow may offer a considerably more accurate assessment of change in the design of future experiments.

Summary. Unilateral renal artery infusions of hypertonic mannitol increase ipsilateral urine flow, but do not change PAH or creatinine clearances in the anesthetized dog, when infused at 300 mg/min for 10 minutes. In the dog with hemorrhagic hypotension, unilateral renal artery infusion of 480 mg/min of hypertonic mannitol produces a 50% increase in renal blood flow ipsilaterally and a 6% increase contralaterally. These effects are seen in the absence of any change in plasma volume or any increase in the hypotensive arterial pressures.

1. Barry, K. G., Cohen, A., LeBlanc, P., *Surgery,* 1961, v50, 335.
2. Barry, K. G., Cohen, A., Knochel, J. P., Whelan, T. J., Jr., Beisel, W. R., Vargas, C. A., LeBlanc, P. C., Jr., *New Engl. J. Med.,* 1961, v264, 967.
3. Harvey, R. B., *Am. J. Physiol.,* 1960, v199, 31.
4. Lilienfield, L., Braun, W., *Clin. Research,* 1962, v10, 67.
5. Braun, W., Lilienfield, L., *ibid.,* 1962, v10, 67.
6. Teschan, P. E., Gagnon, J. A., Murphy, G., *ibid.,* 1963, v11, 241.
7. Goldberg, A., Lilienfield, L., *ibid.,* 1963, v11, 242.
8. Bonsnes, R. W., Taussky, H. H., *J. Biol. Chem.,* 1945, v158, 581.
9. Smith, W. W., Finkelstein, N., Smith, H. W., *ibid.,* 1940, v135, 231.
10. Selkurt, E. E., *Am. J. Physiol.,* 1945, v145, 699.
11. Barry, K., Doberneck, R., McCormick, G., *Clin. Research,* 1962, v10, 245.
12. Bounous, G., Shumacker, H., King, H., *Ann. Surg.,* 1960, v151, 47.
13. Marshall, R., Shepherd, J. T., Jr., *Am. J. Physiol.,* 1959, v197, 951.

Received May 17, 1963. P.S.E.B.M., 1963, v113.

Effect of Crowding on Hypertension and Growth in Rats Bearing Regenerating Adrenals.* (28541)

LEE L. BERNARDIS AND FLOYD R. SKELTON
Department of Pathology, State University of New York, Buffalo, N. Y.

Increased population density has been shown to have both a deleterious effect on growth(1,2,3) and a stimulating action on the anterior pituitary-adrenocortical system (4,5,6,7). Deranged response of the latter system to stress has been implicated rather widely in the genesis of hypertension, further support for which was found in the observation(8) that rats bearing regenerating adrenals develop hypertension and vascular changes similar to human hypertensive disease when renal mass is reduced and excess sodium provided.

The high level of stress incident to modern civilization is blamed by some for the increasing frequency of hypertensive disease and, although evidence is not conclusive, high population density and psychic factors arising therefrom may enter into the genesis of hypertension(9). In fact, Weiss(10) has stated that emotional stress seems at times to precede the onset of hypertension and Ayman(11,12) postulated that the early symptoms of essential hypertension were of psychic origin.

The present study was done to determine whether the stress of population density affected the development of high blood pressure in rats bearing regenerating adrenals.

Methods. Seventy-two female Charles River rats were divided into 6 groups and treated in the following manner: Group 1 (6 rats in single cages) and Group 2 (18 rats, 3 per

* This investigation was supported by a research grant from Nat. Heart Inst., Nat. Inst. Health, P.H.S.

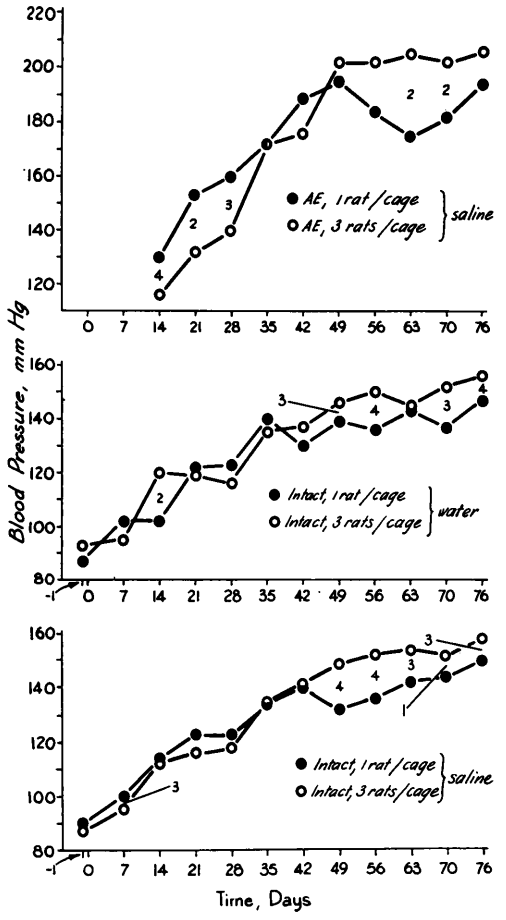

FIG. 1. Systolic blood pressure changes in adrenal-enucleated and intact rats under crowded and non-crowded conditions. (1) $p < .05$, (2) $p < .02$, (3) $p < .01$, (4) $p < .001$.

cage) were subjected to right-sided nephrectomy and adrenalectomy and left-sided adrenal enucleation. Group 3 (6 rats in single cages) and Group 4 (18 rats, 3 per cage) consisted of intact rats; these animals as well as those of Groups 1 and 2 were given 1% saline to drink *ad libitum*. Groups 5 and 6, arranged in a corresponding manner, also consisted of intact rats but were given tap water instead of saline.

The cages in which the animals were maintained measured 228 × 178 × 178 mm and were fitted with spill-proof food trays. All animals were fed a synthetic diet, containing 0.8% NaCl and calculated to yield 4.2 Cal/g. Food and drinking fluid consumption were measured for 3 consecutive days at weekly intervals.

Systolic blood pressure was determined weekly by the microphonic manometer method of Friedman and Freed(13) with the animals under light ether anesthesia. Body weight and length were measured while the rats were anesthetized. The animals were killed by decapitation after 11 weeks and their organs removed, trimmed and weighed.

Results. Fig. 1 shows that mean blood pressure of all grouped animals (Groups 2, 4 and 6) was higher than that of corresponding controls housed singly (Groups 1, 3 and 5). Deaths in Group 2 reduced to 3 the number of animals on which data could be used for statistical evaluation.

Of the rats bearing regenerating adrenals (Groups 1 and 2) the grouped animals weighed significantly less than those caged singly. The difference in body weight between grouped (Groups 4 and 6) and non-grouped intact rats (Groups 3 and 5) was smaller and not significant. There was little, if any, difference in the mean body length between grouped and non-grouped animals, regardless of treatment.

Without exception, food intake of all grouped animals (Groups 2, 4 and 6) was less than that of controls housed in single cages (Groups 1, 3 and 5). The grouped intact rats given saline (Group 4) drank consistently less than intact controls maintained in single cages (Group 3).

The higher blood pressure of the grouped, adrenal-enucleated animals (Group 2) was reflected in the significantly greater cardiac hypertrophy ($p<0.02$). Kidneys, brains and adrenals were also heavier, but these changes were not significant (Table I). It is noteworthy that the pituitary and thyroid glands of these animals also were significantly larger (both $p<0.02$) than those of control rats kept in single cages. No significant differences were seen in the weight of the thymus in any group.

Discussion. It is apparent that crowding caused a modest, significant increase in the level of blood pressure over that which occurred in non-crowded animals bearing regenerating adrenals. The presence of pituitary enlargement and slightly greater size of

TABLE I. Organ Weights (% Body Weight) of Adrenal Enucleated and Intact Rats Under Crowded and Non-crowded Conditions.

Group	Heart	Kidney	Adrenals	Thymus	Brain	Pituitary	Thyroid
1 Adrenal-enucleated (saline) 1 rat/cage	328 ± 5*	566 ± 5	12.2 ± .5	160 ± 16	565 ± 37	4.1 ± .6	5.3 ± .3
2 Adrenal-enucleated (saline) 3 rats/cage	410 ± 3	776 ± 88	14.3 ± 1.9	156 ± 15	766 ±110	5.8 ± .3	7.2 ± .6
3 Intact (saline) 1 rat/cage	328 ± 9	686 ± 38	19.8 ± .7	142 ± 9	671 ± 32	5.6 ± .3	6.1 ± .6
4 Intact (saline) 3 rats/cage	325 ± 9	712 ± 19	19.3 ± .8	148 ± 10	706 ± 13	5.8 ± .1	5.4 ± .3
5 Intact (tap water) 1 rat/cage	326 ±21	656 ± 27	21.5 ± 1.1	163 ± 12	670 ± 49	5.2 ± .2	4.9 ± .3
6 Intact (tap water) 3 rats/cage	307 ± 6	676 ± 20	20.2 ± .7	152 ± 10	706 ± 17	5.2 ± .6	5.3 ± .4

* ± S.E.M. Underlined values, $p < .02$.

the regenerated adrenal in the grouped animals suggests increased ACTH secretion, but the absence of thymic involution in these animals, normally a manifestation of increased glucocorticoid secretion, strongly suggests that the regenerating adrenals were not responding to such ACTH stimulation with the secretion of corticosterone, the normal glucocorticoid in this species. The growth retardation of these animals also may reflect this functional change in accordance with the suggestion of Selye and Collip(14), that under stress the pituitary shifts its hormone production to more ACTH and less STH.

These observations and the report of Skelton and Hyde(15) that the plasma level of corticosterone required to produce hypertension is greater than that attained in rats bearing regenerating adrenals even in response to severe stress further suggest that adrenal regeneration hypertension results from secretion of a steroid other than corticosterone.

Summary. 1. Uninephrectomized, salt-treated rats bearing regenerating adrenals and grouped 3 to a cage developed significantly higher blood pressure than corresponding rats caged singly. Absence of thymic involution in the grouped, adrenal-enucleated rats lends further support to the thesis that the development of hypertensive disease under these conditions is through the secretion of a steroid other than corticosterone. 2. The enhancement of hypertension by crowding occurred in the face of lower dietary and saline consumption and smaller weight gain than was observed in controls housed singly. Similar, but much less consistent changes occurred in intact grouped rats drinking saline or tap water. 3. These observations support the hypothesis that increased population density augments blood pressure levels attained in rats bearing regenerating adrenal glands.

The authors are indebted to Miss Mary Hyde, Mr. Luther Joseph, Miss Anne Simpson, Mr. Peter Poliszcuk and Mr. David Zandor for technical assistance in this investigation. The illustrations were prepared by Mr. Melford Diedrick, Dept. of Medical Illustration, and by Mr. Emmanuel Kaufman, Dept. of Pathology.

1. Goetsch, W., *Biol. Zbl.,* 1924, v44, 529.
2. Vollmer, H., *Zschr. Kinderheilk.,* 1926, v41, 205.
3. Adolph, E. F., *Biol. Bull.,* 1931, v61, 350.
4. Christian, J. J., *Am. J. Physiol.,* 1955 (a) v182, 292.
5. ———, *ibid.,* 1955 (b) v181, 477.
6. Davis, D. E., Christian, J. J., PROC. SOC. EXP. BIOL. MED., 1958, v94, 728.
7. Flickinger, G. L., Ratcliffe, H. L., *Fed. Proc.,* 1961, v20, 176.
8. Skelton, F. R., PROC. SOC. EXP. BIOL. AND MED., 1955, v90, 342.
9. Alexander, F., *Psychosomat. Med.,* 1939, v1, 139.
10. Weiss, S., *Emanuel Libmann Anniversary Vol.,* 1932, v3, 1181.
11. Ayman, D., *JAMA,* 1930, v95, 246.
12. ———, *ibid.,* 1931, v96, 2091.
13. Friedman, M., Freed, S. C., PROC. SOC. EXP. BIOL. AND MED., 1949, v70, 670.
14. Selye, H., Collip, J. B., *Endocrinology,* 1936, v20, 667.
15. Skelton, F. R., Hyde, P. M., PROC. SOC. EXP. BIOL. MED., 1961, v106, 142.

Received May 23, 1963. P.S.E.B.M., 1963, v113.