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BRIEF COMMUNICATION

Testosterone and the Sex Difference in Blood Pressure in Mice

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SCHMALBACH, N. L. AND C. L. KUTSCHER. *Testosterone and the sex difference in blood pressure in mice.* PHARMAC. BIOCHEM. BEHAV. 4(3) 339–341, 1976. – Blood pressure of adult male SWR/J mice is approximately 20 mm Hg higher than that of females. Castrating males at weaning eliminated this sex difference. Testosterone restored blood pressure of castrated mice to the level of the intact males within 10 days indicating a pressor effect of testosterone. Ovariectomy had no effect on blood pressure.

Blood pressure Gonadectomy Sex Testosterone

ACTION of sex steroids on the modulation of blood pressure has been little studied in spite of the fact that in many animals there is a sex difference with male blood pressure exceeding that of females. Such a difference has been noted in certain inbred mouse strains [8,9], especially those with high blood pressure [17], in rat strains selectively bred for spontaneous hypertension [13, 14, 15, 19], in rat strains bred for sensitivity or resistance to hypertension induced by a high NaCl diet [4,5] and in chickens [21]. In man, a higher incidence of hypertension in males has been observed in subjects 25–64 years old [20]; however, blood pressure in males has been found in some studies to exceed that of females only during the first four decades of life [2,7].

It is not clear if these sex differences are mediated by action of male or female sex steroids, or both [1]. The higher blood pressure of males of the CBA mouse strain [8,9] has been attributed to a higher level of aggressive behavior mediated by testosterone among males when mice were maintained under group-housing conditions designed to maximize aggressive encounters. We present evidence here that in the SWR/J inbred mouse strain, the higher male blood pressure may be caused directly by testosterone independently of any testosterone-induced changes in aggressive behavior. Presence of normal levels of steroids in females has no effect on blood pressure.

METHOD

The SWR/J mice used in this experiment were bred in the laboratory from stock obtained from Jackson Laboratory, Bar Harbor, Maine. This strain was used because of the relatively high blood pressure level it exhibited [17] and because it may be particularly sensitive to actions of sex steroids [18] thus maximizing opportunity to observe steroid action. At 30 days of age, gonads were removed from 20 females and 20 males and a sham operation (gonads exposed during surgery, but not disturbed) was performed on 20 females and 20 males. Operations were performed under Nembutal anesthesia (65–70 mg/kg IP). Following the operation, half the mice in each sex/operation condition were housed in plastic cages 17.3 × 29.0 × 12.5 cm, 2–5 mice per cage (Group-reared condition). Half the mice were housed individually in steel cages 13.3 × 17.8 × 13.8 cm (Isolation-reared condition). Both types of cages had 0.6 cm mesh hardware cloth tops and solid floors covered with clay bedding. All mice were maintained on Purina Laboratory Chow and demineralized water. Temperature was maintained at 21.1 ± 1°C and lights were on for 12 hr/day.

At 4 months of age, blood pressure was measured indirectly in restrained, unanesthetized mice by means of a tail cuff, using a Narco Bio-Systems apparatus consisting of a Model DMP-4B physiograph and a Model PE-300 programmed electro-sphygmomanometer. The tail cuff was 17 mm wide. Restraint of the mice produced no overt signs of distress and, in our hands, no deaths have resulted from this technique contrary to the experience of others [16]. The blood pressure determination used for statistical analysis for each animal was the mean of at least 10 measurements made on two separate occasions 48 hours apart.

RESULTS AND DISCUSSION

For Group-housed animals (Table 1), blood pressure determinations were analyzed by a 2 × 2 completely random-

TABLE 1

BLOOD PRESSURE (BP) AND BODY WEIGHT AT FOUR MONTHS OF AGE FOR MICE WHICH RECEIVED GONADECTOMY OR SHAM OPERATION AT 1 MONTH OF AGE

Animals	Group Reared n	BP (mm Hg)	Weight (g)	Isolation Reared n	BP	Weight
Intact females	10	84.4±3.8*	20.9±0.41	10	84.9±1.5	22.2±0.51
Ovex females	10	85.6±3.5	21.8±0.55	10	88.1±2.0	25.3±0.80
Intact males	10	104.6±3.3	24.0±0.29	10	104.0±2.3	25.8±0.42
Castrated males	10	81.4±4.1	23.5±0.70	10	83.6±3.2	25.7±0.40

*Mean ± the standard error.

ized factorial design (sex × operation). Blood pressures differed as a function of sex ($p<0.05$) and operation ($p<0.01$), but the interaction was also significant ($p<0.01$). Pairwise comparisons between all cell means were made with Scheffe's test [12]. Blood pressures of intact males exceeded ($p<0.01$) those of intact females by approximately 20 mm Hg. Castration at 1 month of age completely eliminated this sex difference as the castrated males became statistically indistinguishable from the intact females. Ovariectomy had no significant influence on blood pressure. No significant correlations were found between body weight and blood pressure.

The blood pressure measurements for isolation-reared mice were given the same kind of statistical analysis as the data for group-reared mice. The group means for the isolation-reared mice were very similar to those for the group-reared mice and the same significant differences were found in the statistical analysis. Mean blood pressure of intact males was higher than that of the other three groups which did not differ from each other. No significant differences were found between group-reared and isolation-reared mice. Thus the presence of male gonads mediates the attainment of a higher level of blood pressure than is possible in the absence of the gonads. The presence or absence of the ovaries appears to be irrelevant.

In order to determine whether the action of the testes on blood pressure is due to the action of testosterone or to some other factor concomitant with a functioning testes, male SWR/J mice castrated at 30 days of life were implanted when 7–7.5 months old with one 75 mg pellet of testosterone (Oreton; Schering Corp.) under the skin of the back dorsal to the forelimbs. As a control for surgical procedures, a group of intact males and a group of castrated males of the same age were given sham implantation operations in which mice received anesthesia, incisions and suturing, but no pellets. In the implanted group, pellets remained in place for 20 days at which time they were removed under anesthesia.

Measurements made within 5–10 days of implantation showed that testosterone raised blood pressure in the castrated males to a level not significantly different from that of intact males and produced a significant weight gain (Table 2). Measurements made 15 days after removal of the pellets indicated that blood pressure had returned to the preimplantation level.

In order to estimate the daily absorption rate of testosterone from the pellet, preimplantation and post-implantation weights of the pellets were made. Amount of hormone absorbed per unit time declines with the decreasing size of the pellet with the rate markedly slowed after 50% of the pellet has been absorbed [6]. In this study, average decrement in pellet size was 30%. If the absorption of the pellet is taken to be linear during the 20 days of implantation, the mean daily absorption was estimated to be 1120 μg/day, an amount which probably exceeds the normal daily testosterone secretion of the mouse [3].

Clearly, testosterone has a mild pressor effect in the castrated SWR/J mouse, but the mode of action is unknown. Possibly it acts by causing expansion of the extracellular fluid space, especially plasma volume, because of sodium retention. Edema is a complication of androgen therapy in man [10,11]. These results suggest that the action of testosterone on blood pressure deserves further elaboration.

TABLE 2

THE EFFECT OF TESTOSTERONE PELLETS ON BLOOD PRESSURE (BP) OF CASTRATED MICE

Animals	n	Implantation	Measure	Before Implant	5-10 Days After Implant	15 Days After Removal
Intact males	5	Sham-op	BP	101.9 ± 0.5 mm Hg*	104.6 ± 2.7	—
			Weight	28.6 ± 0.4 g	27.2 ± 0.3	—
Castrated Males	5	Sham-op	BP	81.1 ± 2.9	84.5 ± 1.3	—
			Weight	25.7 ± 1.0	25.3 ± 0.8	—
Castrated males	5	Testosterone	BP	77.7 ± 1.7	98.3 ± 3.8	79.8 ± 2.6
			Weight	25.9 ± 0.5	28.5 ± 0.7	27.7 ± 0.1

*Mean ± the standard error.

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