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GENETIC INFLUENCE ON HYPERTENSION INDUCED BY CADMIUM IN DAHL HYPERTENSION-RESISTANT AND HYPERTENSION-SENSITIVE RATS*

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DISCLAIMER

GENETIC INFLUENCE ON HYPERTENSION INDUCED BY CADMIUM IN DAHL HYPERTENSION-RESISTANT AND HYPERTENSION-SENSITIVE RATS

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ABSTRACT

Dahl hypertension-resistant (R) and hypertension-sensitive (S) rats, with opposite genetically controlled propensities for hypertension, were used to determine if cadmium-induced hypertension was also dependent upon genetic predisposition. When intraperitoneal injections of cadmium (1 and 2 mg/kg) were given to male and female R and S rats maintained under ordinary laboratory conditions, female S rats manifested a significant increase ($p < 0.01$) in blood pressure which persisted for 16 weeks, whereas the R rats and the male S rats showed no significant changes in blood pressure. Cadmium had no effect on body weight. The concentrations of cadmium in hepatic and renal tissues of cadmium-injected male and female S rats were significantly higher ($p < 0.001$) than that of R rats. The kidneys of female S rats given cadmium showed renal vascular changes in accord with renal hypertension of moderate degree. We concluded that differences in genetic substrate influence the pathogenesis of experimental cadmium-induced hypertension.

INTRODUCTION

A positive correlation between cadmium (Cd) concentrations in the air of American cities and death rates from hypertension (HT) and arteriosclerotic heart disease has been reported (1, 12). Experimental HT has been produced by feeding traces of Cd in drinking water for long periods of time (17) or by injecting small doses of Cd (18) in the rat. However, there are also negative findings in regard to Cd and HT (13, 15). The present experiment was designed to investigate the influence of genetic predisposition on the pathogenesis of Cd-induced HT in HT-resistant (R) and HT-sensitive (S) rats maintained under ordinary laboratory conditions.

MATERIALS AND METHODS

All the rats were selected from one of two unique strains of rats originally derived from the same Sprague-Dawley ancestors by selective inbreeding. They were designated

* The authors note with deep sorrow the death of Dr. Lewis K. Dahl on November 26, 1975.

Dahl HT-resistant, or R, and HT-sensitive, or S strains respectively, because of their resistance or susceptibility to develop HT in response to chronic excess salt (NaCl) ingestion (5) as well as to other experimental procedures (9,11). Details on animal care, housing and the rationale for defining "HT" may be found in earlier papers (2-4). Specially prepared low-salt chow (Agway, Inc., Country Foods Division, Syracuse, N.Y.) containing 0.3% NaCl and tap water were available ad libitum.

The present experiment involved sixty weanling rats divided into 2 groups as follows: a) cadmium (5 male R, 10 female R, 5 male S, 10 female S) and b) controls (5 male R, 10 female R, 5 male S, 10 female S). The rats in the experimental group were injected with 2 mg of Cd per kilogram of body weight, as the acetate intraperitoneally (IP) in a concentration of 1 mg Cd/ml. The controls received the same volume of 0.9% aqueous NaCl IP. Systolic BP of experimental and control rats was measured under light ether anesthesia by tail plethysmography (2) at 3 weeks post-weaning (6 weeks old), and then at one to two week intervals depending upon the development and severity of HT. Body weights were recorded concomitantly with BP. BP data obtained from rats that appeared ill or that exhibited a weight loss in excess of 10 g from any previous maximum were not included in the data analysis. At necropsy, both kidneys and liver were removed. Kidneys were further bisected across the short axis. Two halves, one from each kidney, were fixed in formalin for light microscopy and the other two kidney halves and the liver were assayed for Cd by atomic absorption spectrophotometry (Perkin-Elmer 503). Statistical analysis of the data was performed with specially prepared computer programs. Data with p values of < 0.01 were considered significant. All p values < 0.001 were assigned that nominal value.

RESULTS

Blood Pressure. The pertinent BP data are summarized in Figures 1 and 2. An analysis of variance indicated the anticipated strain difference with S controls exhibiting significantly higher BP than R controls ($p < 0.001$). Among animals exposed to Cd, BP, as compared with appropriate controls, was not significantly affected

3 weeks after the first injection (week 6 or 6 weeks of age). Following a 2nd dose of Cd (1 mg/kg) on week 6, female S rats developed significantly higher BP than their respective controls by week 8. The Cd-induced HT continued through week 23, after which the BP returned to control levels. In response to a 3rd injection of 1 mg Cd/kg given to S rats on week 28, the difference in BP between the female S cadmium group and their counterpart controls once again achieved statistical significance ($p < 0.01$) by week 29 and continued therafter. Male S rats, however, showed no consistent Cd-induced BP elevations. Despite a 3rd and 4th injection of Cd (1 mg/kg), on weeks 23 and 28, respectively, R rats remained normotensive throughout the experiment. None of the controls developed HT. Morbidity and mortality commenced among Cd-injected males and females of both strains on weeks 29 and 32, respectively, and there were no healthy survivors to make a significant comparison thereafter. Of those rats examined post mortem, 90% of our animals in the Cd group died of internal hemorrhage which was markedly more massive in S than in R rats. An analysis of body weight showed the expected significant difference between males and females of both strains ($p < 0.001$). However, weights of rats in Cd groups and their respective controls were similar.

Cd concentrations in tissues. Figure 3 shows that the concentrations of Cd in kidney and liver tissues in S rats (4 males, 6 females) given Cd were significantly higher ($p < 0.001$) than that of R rats (4 males, 6 females). Since no significant differences appeared between tissues obtained from male and female rats, these values were pooled.

Histology. Histological kidney sections were evaluated without knowledge of strain and treatment. Microscopic changes of glomeruli (thickening of capillary basement membrane; hyaline deposits along the basement membrane; increase in number of epithelial cells; hyalinization and atrophy) arterioles and arteries (hyperplasia of intima; hypertrophy of media; sclerosis; narrowing of the lumen; necrotizing lesions) were graded as follows: normal = 0, slight = $\pm$, moderate = $+$ and marked = $+++$. Since no significant renal vascular changes were noted in the male R and S cadmium groups and their respective controls, this data was not presented. A summary of the renal vascular changes in female R and S rats is shown in Table I. Although changes occurred to some extent in all the groups except

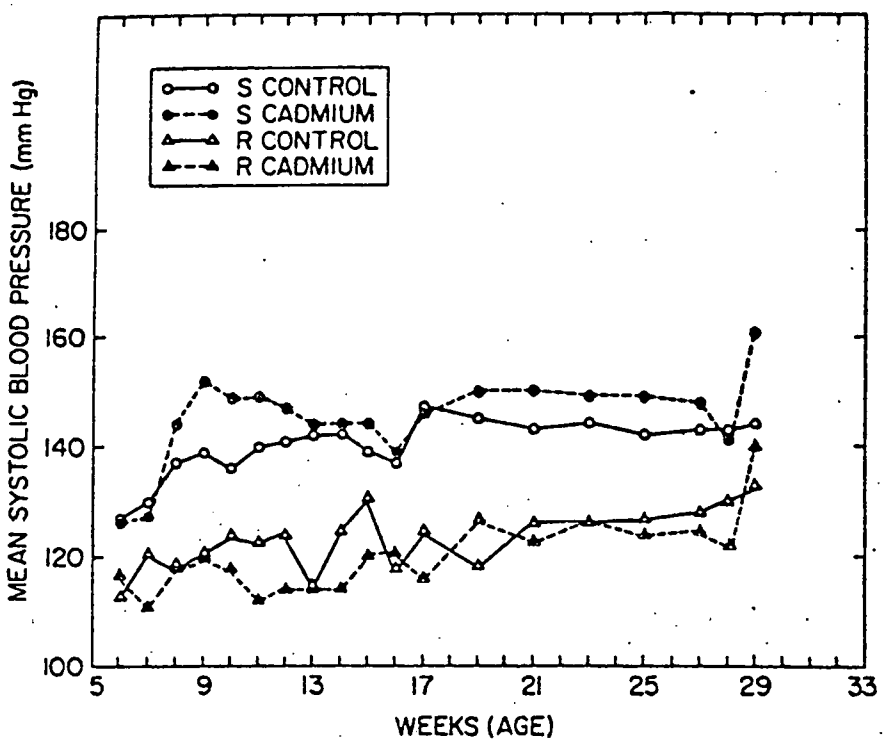

Figure 1 - Effect of cadmium injection on blood pressure of male R and S rats. For more details see text.

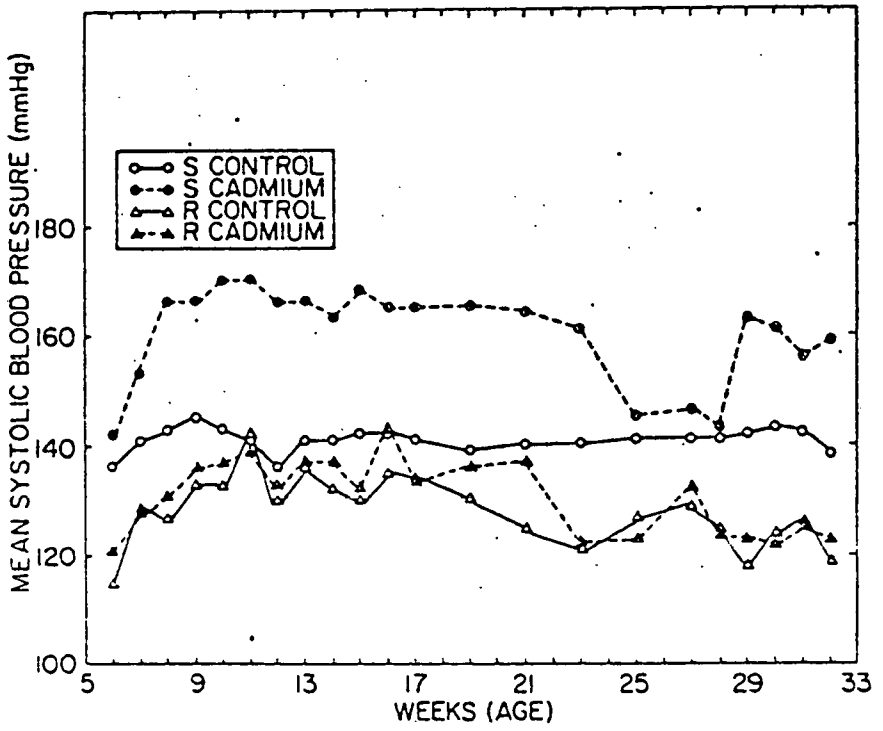

Figure 2 - Effect of cadmium injection on blood pressure of female R and S rats. For more details see text.

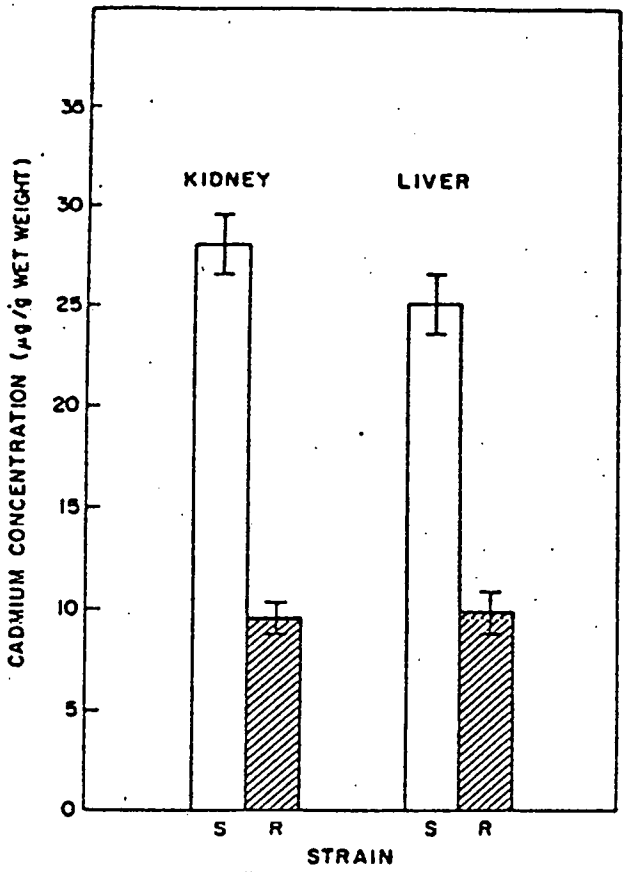

Figure 3 - Cadmium concentrations in the kidney and liver of R and S rats injected with cadmium.

TABLE I. RENAL VASCULAR CHANGES IN FEMALE R AND S RATS INJECTED WITH CADMIUM

GROUP	GLOMERULI		ARTERIOLES		ARTERIES	
	0	± - ++	0	± - ++	0	± - ++
CONTROL						
R (10)	10	0	10	0	10	0
S (10)	8	2	7	3	9	1
CADMIUM						
R (10)	8	2	7	3	8	2
S (10)	1	9	2	8	4	6

The number of rats examined is indicated in parenthesis. For more details see text.

in the R controls, S cadmium group exhibited significantly more changes than any other group ($p < 0.01$).

DISCUSSION

Previously, Schroeder and his co-workers (18) reported that IP injection of Cd resulted in transient (2 to 4 weeks) HT in female Long-Evans rats which were raised in a Cd-free environment on Cd-free food.

Results of earlier studies led us to conclude that in S rats the development of experimental HT required both the appropriate genetic substrate as well as non-genetic determinants for full expression. In this experiment we investigated the chronic effect of injected Cd on systolic BP in R and S rats maintained on ordinary laboratory conditions. The following observations were made: 1) When male and female R and S rats were exposed to Cd by injection, significant and persistent (16 weeks) HT developed in female S rats but not in R rats of either sex. Although Cd failed to produce consistent HT in male S rats, it did increase the mortality rate; 2) The concentrations of Cd in hepatic and renal tissues of Cd-injected S rats of both sexes were significantly higher than those of R rats; and 3) The female S kidney exposed to Cd by injection showed renal vascular changes well in accord with experimental HT of moderate degree.

Schroeder and Vinton (17) observed the development of HT in young female rats given trace amounts of Cd in drinking water from the time of weaning. On the other hand, male rats were largely resistant to this disorder. They had no explanation for this difference. Similarly, female S rats injected with Cd manifested an increase in BP, whereas the R rats and the male S rats showed no changes in BP. In contrast to these results, our previous studies have repeatedly shown that, while the ultimate BP levels were similar in both sexes on a high sodium chloride diet, the males generally developed HT more rapidly than the females. Furthermore, castration of males had no effect on BP (10). This study indicates that genetic factors play a role in experimental HT induced by Cd injection similar to that observed with other hypertensinogenic stimuli. However, at this juncture, insufficient data are available to explain the observed sex difference in S rats. Shaikh and Lucis (19) showed that

metallothionein, a Cd-binding protein was synthesized in mammalian tissues in response to the feeding or injection of Cd. Liver and kidney were the most active synthesizers of metallothionein. In this experiment, the reason(s) for this striking and unexpected difference in Cd-binding between R and S rats could be due to a) a difference in the concentration of metallothioneins, and b) a difference in Cd-binding properties of metallothioneins. The pathological changes in the kidney were not necessarily correlated with the renal Cd content which was similar in both male and female S rats, but they appeared to be the result of Cd-induced HT.

Dahl et al. (6) demonstrated that among animals which were on low salt regimen and were normotensive, S rats had significantly higher BP response to vasoconstrictor substances such as angiotensin II and norepinephrine than did R rats. Using the techniques of parabiosis and renal transplant between rats from R and S strains, we have shown that the kidneys play an important, genetically-determined role in the development of HT, or lack thereof, following exposure to any one of several hypertensinogenic stimuli commonly used to invoke experimental HT (7, 8). Recently, we reported that the acute pressor response to intra-arterial Cd injection was significantly higher in S rats than in counterpart R rats and consequently we concluded that the vascular response to Cd ion was, as with other hypertensinogenic stimuli, determined to a great extent by genetic predisposition (14). In this experiment we speculate that Cd may act on vascular smooth muscle through the kidneys in order to induce HT in female S rats.

Since most human subjects dying from complications due to HT showed in their kidneys either increased concentrations or increased ratios of Cd to zinc, compared to subjects dying of other major diseases (16), the principal interest in experimental Cd-induced HT lies in the possibility that it may be a contributing factor in the genesis of human"essential" HT. It appears that the genetic substratum may be a critical determinant of whether experimental HT develops after exposure to cadmium. If our experimental models have bearing on the process of "essential" HT in man, this study suggests that even if current environmental Cd pollution is not of immediate concern to "all" individuals, it may be a genuine health hazard to "sensitive" individuals with a family history of HT.

CONCLUSION

This experiment demonstrated that in rats the pathogenesis of Cd-induced HT was influenced to a great extent by genetic predisposition. When Dahl HT-resistant (R) and Dahl HT-sensitive (S) rats were exposed to cadmium by injection, the following observations were made: 1) Hypertension developed in S rats but not in R rats; 2) The concentrations of Cd in hepatic and renal tissues of S rats were higher than those of R rats; and 3) The kidneys of S rats showed renal vascular changes in accord with renal hypertension. In addition to the experimental implications, these findings may have relevance in the problem of human "essential" hypertension.

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