Failure of Benzydroflumethiazide and Hydrochlorothiazide to Influence Plasma Potassium in Nephrectomized Dogs.* (26441)

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The pharmacological basis of the hypotensive action of chlorothiazide and structurally related compounds continues to be a controversial problem. Evidence from several laboratories(1,2,3,4,5) indicates that the initial antihypertensive response of chlorothiazide is a result of reduction in plasma volume and consequent oligemia. These results are further supported by observation that restoration of plasma volume by intravenous dextran solutions, either salt free or salt containing, results in partial reversal of the antihypertensive action of chlorothiazide(6,7). In addition, chlorothiazide and mercurial diuretics have been reported to have a selective antihypertensive action which may be potentiated by a reduction in body sodium(8). The foregoing results strongly suggest that the principal action of the benzothiadiazine diuretics is dependent upon a reduction in plasma volume, resulting from the diuretic action of the drug. In contrast, it has been reported that the hypotensive response of chlorothiazide *per se* may depend upon antagonism to an unknown pressor substance (9). Chlorothiazide has also been shown to decrease "reactivity" to various types of pressor agents in normotensive and hypertensive subjects(4,10) as well as in normotensive dogs(11). This response is not specific for chlorothiazide since a similar effect concomitant with diuretic action has been observed with acetazolamide and mercaptomerin, indicating sodium excretion played a dominant role(12). Recently it has been reported that chlorothiazide and hydrochlorothiazide† significantly lowered plasma potassium levels with no effect on sodium levels in nephrectomized dogs(13) whereas, acetazolamide and mercaptomerin had no effect. These results suggest that the chlorothiazide derivatives have an extrarenal action and focus attention on both an extrarenal effect and a saluretic response contributing to the early hypotensive action observed with these diuretic agents, and that they exhibit a specific effect not shared by other diuretics.

During an investigation of benzydroflumethiazide‡ it seemed of interest to determine the effect of this diuretic on plasma sodium and potassium levels in acute nephrectomized dogs. It would be anticipated *a priori* that this structurally related agent would decrease plasma potassium if the extrarenal effect is specific for the benzothiadiazine derivatives. Our results indicate that benzydroflumethiazide does not alter plasma potassium in acute nephrectomized dogs.

Methods. Experiments were carried out with dogs of either sex weighing 9 to 20 kg that had been maintained on a diet of Friskies dog cubes for a minimum of 1 week prior to experiment. The animals were anesthetized with sodium pentobarbital (35 mg/kg) and arterial blood pressure was recorded with a 3 channel physiograph. Acute bilateral nephrectomies were performed *via* the ventral approach. Blood samples were obtained before nephrectomy(C), 1 hour post nephrectomy (PN-1 hr) immediately before administration of the diuretic, then 30 minutes (PD-30 min), 1 hour (PD-1 hr), 1.5 hours (PD-1.5 hr), 2 hours (PD-2 hr), 3 hours (PD-3 hr) and 4 hours (PD-4 hr) after beginning of drug administration. Blood hematocrits were determined and plasma sodium and potassium were measured with a Beckman flame photometer. A control group of 8 dogs received 10 ml 5% glucose in water 1 hour after nephrectomy followed immediately with a 30 minute infusion of 50 ml 5% glucose. In the control animals the 30 minute sample was not taken. All experimental animals received the same amount of glucose plus the diuretic that was being in-

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TABLE I. Plasma Electrolytes Values in Acute Nephrectomized Dogs.

Group and No. animals	C	PN-1 hr	PD-30 min.	PD-1 hr	PD-1.5 hr	PD-2 hr	PD-3 hr	PD-4 hr
				Na, meq/l				
Control	140 ±1.9	139 ±1.9		138 ±1.3	140 ±1.6	139 ±2.0	139 ±2.0	140 ±1.0
Benzydroflumethiazide 12	141 ±1.7	141 ±1.7	140 ±1.4	140 ±2.2	141 ±1.0	142 ±1.7	141 ±1.3	140 ± .9
Hydrochlorothiazide 12	141 ±1.8	140 ±1.4	140 ±1.9	141 ±1.5	141 ±1.6	142 ±1.6	143 ±1.7	142 ±1.5
				K, meq/l				
Control 8	4.1 ± .10	4.0 ± .14		4.0 ± .11	3.9 ± .10	3.9 ± .13	3.9 ± .14	4.0 ± .08

vestigated. The second group of 12 animals received a prime dose of 0.5 mg kg benzydroflumethiazide and the same dose of the drug during the 30 minute infusion. The first 6 animals that received benzydroflumethiazide were followed for only a period of 2 hours after nephrectomy, to mimic the procedure employed for hydrochlorothiazide by Beavers (13). Thereafter all experiments were extended from 2 to 4 hours and more blood samples obtained. The third group of animals received 2 mg/kg hydrochlorothiazide as a prime dose and the same amount in the infusion. Statistical analysis of the data was performed by the method of Mather(14).

Results. Table I summarizes plasma sodium levels in control animals and those administered benzydroflumethiazide and hydrochlorothiazide as well as plasma potassium levels in control animals. Tables II and III illustrate the effect of benzydroflumethiazide and hydrochlorothiazide, respectively, on plasma potassium levels. Data for potassium are presented in detail because they do not confirm previous data(13). There was no significant change in plasma sodium concentration after either diuretic ($P>0.05$). Similarly, no significant change in plasma potassium was observed in animals that received either diuretic ($P>0.05$).

Mean arterial blood pressure did not decrease more than 20 mm Hg after nephrectomy. Hematocrits for each group did not vary more than 10% from control values throughout the experiments.

Discussion. Results obtained in this study indicate that benzydroflumethiazide does not have any effect on plasma sodium and potassium levels in the acute nephrectomized dog. These results do not support the concept of an immediate extrarenal action which has been suggested for this type of diuretic(13). However, these acute experiments do not rule out the possibility that chronic administration of the drug may exhibit some extrarenal effect. Failure to observe a decrease in

TABLE II. Plasma Potassium Values in Acute Nephrectomized Dogs During and Following Administration of Benzydroflumethiazide.

Dog	C	PN-1 hr	PD-30 min.	PD-1 hr	PD-1.5 hr	PD-2 hr	PD-3 hr	PD-4 hr
1	3.7	3.7		3.7		3.7		
2	5.0	4.7		4.7		4.7		
3	4.7	4.6		4.2		4.4		
4	4.8	4.8		4.4		4.4		
5	3.7	4.0		3.7		3.7		
6	4.0	4.6		4.7		4.6		
7	3.9	4.0	3.7	4.0	4.3	4.1	4.0	4.4
8	3.6	3.6	3.8	3.5	3.7	3.9	3.9	3.6
9	4.3	4.1	3.9	4.2	4.0	4.2	4.2	4.4
10	4.5	4.7	4.8	4.4	4.3	4.6	4.5	4.4
11	3.8	3.7	4.1	4.0	3.6	3.7	4.0	4.1
12	4.2	3.8	3.9	4.1	3.7	3.9	3.9	3.9
Mean & S.E.	4.1 ±.14	4.2 ±.13	4.0 ±.16	4.1 ±.11	3.9 ±.12	4.2 ±.11	4.1 ±.10	4.1 ±.13

TABLE III. Plasma Potassium Values in Acute Nephrectomized Dogs During and Following Administration of Hydrochlorothiazide.

Dog	C	PN-1 hr	PD-30 min.	PD-1 hr	PD-1.5 hr	PD-2 hr	PD-3 hr	PD-4 hr
1	4.7	4.6	4.6	4.5	4.6	4.7	4.9	4.4
2	3.8	3.8	3.4	3.6	3.9	4.0	4.2	4.0
3	3.7	3.5	3.6	3.8	3.7	3.7	4.2	4.2
4	3.8	3.9	3.7	3.5	3.7	4.3	4.0	3.8
5	4.1	4.2	4.1	4.5	3.9	4.2	4.1	4.4
6	3.8	3.7	3.8	4.0	4.2	4.2	4.2	4.2
7	3.8	3.5	4.1	3.8	3.7	4.0	3.5	3.5
8	4.1	3.8	3.8	3.6	4.0	4.0	4.3	4.4
9	4.2	3.8	3.8	4.3	4.2	3.9	4.3	4.1
10	3.8	3.7	3.9	3.6	3.8	3.8	3.9	3.9
11	3.6	3.6	3.7	3.8	3.8	3.7	3.7	3.8
12	3.9	4.0	3.9	3.9	4.1	4.1	4.2	4.0
Mean & S.E.	3.9 ±.11	3.8 ±.09	3.9 ±.09	3.9 ±.10	4.0 ±.08	4.1 ±.08	4.1 ±.10	4.1 ±.08

plasma potassium cannot be explained on insufficient dosage since benzydroflumethiazide is at least 5 to 10 times as potent as hydrochlorothiazide in producing a saluretic effect. The only conclusion that can be obtained from this study is that benzydroflumethiazide along with acetazolamide and mercaptomerin exhibits no extrarenal action when evaluated in acutely nephrectomized dogs.

The cause of the failure to obtain a decrease in plasma potassium with hydrochlorothiazide remains obscure. The data do not show any trend toward a decrease in potassium after nephrectomy. Extending the period of observation from 2 to 4 hours should have made it more apparent if there was a migration of potassium from the plasma; this did not occur. Therefore, results obtained in this study indicate that hydrochlorothiazide does not cause any extrarenal effect in acute experiments under the conditions described.

Summary. Benzydroflumethiazide and hydrochlorothiazide did not have any effect on plasma sodium or potassium levels when administered intravenously to acute nephrectomized dogs. These results indicate that under the conditions described benzydroflumethiazide and hydrochlorothiazide do not have an extrarenal effect on sodium or potassium which could explain the mechanism of their antihypertensive action.

1. Freis, E. D., Wanko, A., Wilson, I. M., Parrish, A. E., *Ann. N. Y. Acad. Sci.,* 1958, v71, 450.

2. Tapia, F. A., Dustan, H. P., Schneckloth, R. S., Corcoran, A. C., Page, I. H., *Lancet,* 1957, v2, 831.

3. Dollery, C. T., Harington, M., Kaufmann, G., *ibid.,* 1959, v1, 1215.

4. Freis, E. D., Wanko, A., Schnaper, H. W., Frohlich, E. D., *J. Clin. Invest.,* 1960, v39, 1277.

5. Conway, J., Lauwers, P., *Circulation,* 1960, v21, 21.

6. Freis, E. D., *Hypertension,* F. R. Skeleton, Ed., New York Am. Heart Assn., 1959, v7, 9.

7. Wilson, I. M., Freis, E. D., *Circulation,* 1951, v20, 1028.

8. Hollander, W., Chobanian, A. V., *J. Clin. Invest.,* 1958, v37, 902.

9. Wilkins, R. W., Hollander, W., Chobanian, A. V., *Ann. N. Y. Acad. Sci.,* 1958, v71, 465.

10. Merrill, J. P., Guinand-Baldo, A., Geordano, C., *Clin. Res.,* 1958, v6, 230.

11. Beavers, W. R., Blackmore, W. P., PROC. SOC. EXP. BIOL. AND MED., 1958, v98, 133.

12. Blackmore, W. P., Beavers, W. R., *ibid.,* 1959, v101, 128.

13. Beavers, W. R., *ibid.,* 1960, v103, 711.

14. Mather, K., *Statistical Analysis in Biology,* Interscience Publishers, Inc., New York, 1945.

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