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Effect of Spirolactone SC 8109 on Renal Function in Normal Human Subjects.* (24721)

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A natriuretic effect of synthetic steroids (spirolactones) has recently been shown in the rat, dog(1), and man(2). Kagawa, *et al.* (1) demonstrated that these spirolactones are virtually ineffective in the absence of mineralocorticoid and that effects upon urinary electrolyte excretion are constant so long as a constant ratio of spirolactone/mineralocorticoid is maintained. Similar observations have been reported by Liddle(2) in the human subject. Sodium diuresis has been induced in normal human beings on salt restriction and in patients with aldosterone-secreting adrenal adenoma despite increasing levels of urinary aldosterone(2,3,4). Although Salassa, Mattox, and Power(4) and Liddle(2) have reported that the 24-hour clearance of endogenous creatinine is unchanged in the sodium diuresis induced by spirolactones in man, the literature contains no precise measurements of renal hemodynamics during administration of these substances. The purpose of this study has been to examine more carefully renal function in the normal human subject under the influence of endogenous aldosterone with and without administration of spirolactone.

Materials and methods. The substance used was 3-(3-oxo-17*beta*-hydroxy-19-nor-4-androsten-17*alpha*-yl)propionic acid *gamma*-lactone known by the code name, SC 8109.‡ Drug administration was at a dosage level of 50 mg each 3 hours beginning at 6:00 to 8:00 a.m. on the day preceding study and continuing until the beginning of the clearance periods. Normal, young, male volunteers were fed a diet containing 20 to 30 meq of sodium, under the supervision of a dietitian. Water and potassium were allowed *ad libitum*. Following 5 to 7 days of sodium restriction, the subjects were studied to determine inulin, para-aminohippurate (PAH) and free water clearances and sodium, potassium and chloride excretion rates. Sodium restriction was continued until the 24-hour urinary excretion rate was in the range observed prior to the first study, then a second set of clearance studies was done. In 3 subjects the control studies were obtained prior to the test, while the remaining 2 were studied in the reverse order. Subjects were studied between 8 a.m. and noon following a 16-hour fast. The men were placed in bed with head elevated to 45° and were not allowed to smoke. A water diuresis was induced by oral administration of 20 ml of water/kilogram of body weight and diuresis was sustained by replacement of urine volume plus estimated unmeasured water loss every 15 minutes. Four control and 4 treatment clearance periods of 15 to 20 min. duration were obtained according to accepted technics of timing and collection of samples(5). Sustaining solutions were administered in a 0.9% solution of sodium chloride at the rate of 3.8 to 4.0 ml/min as controlled by a constant rate infusion pump. Following each experiment, the data were analyzed by Barnard's

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TABLE I. Renal Function before and during Therapy with Spirolactone (Mean of 4 Observations).

Subjects		ml/min. Glomerular filtration rate	ml/min. Effective renal plasma flow	Excretion, μeq/min. Na	K	Cl	Urinary Na/K	μl/min. free water clearance
R.H.	Control	134	642	86	93	30	.64	12.6
	Therapeutic	125	824	209	78	87	2.55	14.1
N.P.	Control	128	706	106	118	29*	.94	11.8
	Therapeutic	117	734	176	64	70	2.67	11.4
S.W.	Control	146	744	69	108	20	.74	10.9
	Therapeutic	143	718	156	60	34	2.75	11.7
C.S.	Control	97	500	120	101	38	1.19	13.4
	Therapeutic	109	511	174	36	52	4.8	14.8
G.S.	Control	149	861	134	154	61	.87	18.6
	Therapeutic	141	831	268	93	112	2.88	19.4

* Mean of 3 observations.

sequential "t" test(6) and experiments discontinued when changes in sodium and potassium excretion were shown to be significant at the 5% level of confidence. Chemical determinations of inulin were performed as described by Roe, Epstein, and Goldstein(7); PAH by Goldring and Chasis(8); chloride by Schales and Schales(9); and sodium and potassium by flame photometry, utilizing a lithium internal standard. Free water clearance was calculated as described by Wesson and Anslow(10). Osmolality determinations were performed cryoscopically on a Fiske osmometer.

Results. Increase in rate of urinary excretion of sodium was observed following administration of the steroid-17-lactone in each of the 5 subjects studied (Table I). Mean control excretion rates were of the order of 69 to 134 μeq/min of sodium. Under the influence of SC 8109 the subjects excreted sodium at a rate of 156 to 268 μeq/min. Experimental values were 1.45 to 2.4 times the control, regardless of the sequence of control and experimental studies. Chloride excretion rates paralleled those of sodium, despite a lower absolute value. Experimental rates were 1.4 to 2.9 times those of the control.

No increase occurred in the glomerular filtration rate or effective renal plasma flow. Indeed, in 4 subjects the clearance of inulin was lower during the drug-induced natriuresis.

Rate of potassium excretion fell significantly during administration of the spirolactone. Following administration of the drug lactone, there was a striking rise in the ratio of urinary Na/K from a mean control value of 0.88 to a mean of 3.1.

An increase in free water clearance was observed in 4 of the 5 subjects during administration of SC 8109. The changes were small (0.8 to 1.5 ml/min) and not statistically significant in this small group.

Discussion. The observation of an increase in urinary sodium and a simultaneous decrease in urinary potassium confirms previous work in the salt-restricted human subject treated with spirolactones. A disparity between the absolute rate of excretion of sodium and chloride is observed; however, the changes in sodium and chloride excretion parallel one another. Landau and Lugibihl(11) have shown a similar disparity between these 2 ions during the presumed mineralocorticoid inhibition by progesterone. The difference has not been explained completely.

The changes in rate of excretion of sodium and potassium reported here are consistent with a withdrawal of aldosterone effect. Several investigators have demonstrated increases in urinary aldosterone in subjects receiving spirolactones. This persistence of urinary aldosterone is not conclusive evidence against a decrease in elaboration of mineralocorticoid but the evidence does suggest that aldosterone production is unchanged by spirolactones. The constant drug/mineralocorticoid ratio necessary to give a constant effect and the lack of effect of spirolactone in absence of mineralocorticoid have suggested that the mechanism

of action of the spirolactones might represent competition with mineralocorticoid at a site not yet elucidated. Final acceptance of the thesis of a competitive inhibition must await further study.

This study shows that synthetic steroids do not exert their effect by increasing the filtered load of sodium. The data indicate that sodium diuresis is effected independently of changes in the glomerular filtration rate or effective renal plasma flow.

Summary. Five normal, male, human subjects have been studied by the classical clearance technics during salt deprivation before and after therapy with the steroid-17-spirolactone, SC 8109. No change in the glomerular filtration rate or effective renal plasma flow could be demonstrated but a definite sodium diuresis and potassium retention occurred in response to the spirolactone. A small increase in free water clearance was seen in 4 of 5 subjects studied. These observations localize the effect of spirolactone to the renal tubule.

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Reduction of Post-Occlusive Hyperemia in Cold Subjects.* (24722)

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Hyperemia that occurs in the forearm following arterial occlusion has been ascribed to anoxia of tissues and accumulation of metabolites(1). This view is supported by experiments of Goldschmidt & McGlone(2) and Jepson(3). However, Folkow(4) found the effect of vasodilator metabolites is slight in cats and suggested that reduction of stretch in the vascular walls during occlusion, resulting in reduction of vascular tone, is the major factor in vasodilation occuring in the first stage of reactive hyperemia. More recently, Patterson(5), in studies of reactive hyperemia in human forearms, has come to similar conclusions. The present experiments were performed to determine the effect of increased vasomotor tone on reactive hyperemia. For this purpose it is assumed that exposure to cold and performance of the cold-pressor test increase peripheral vascular tone.

Methods. Experiments were performed on 2 male subjects in room at about 20°C. The subjects were warm (covered with blanket) or cold (nude). Reactive hyperemia was produced by inflating to 220 mm Hg, a sphygmomanometer cuff wrapped around arm and maintaining pressure for 5 or 10 min. A point on upper forearm, free from veins, was marked and skin temperature recorded every 30 sec. with Stoll-Hardy radiometer accurate to 0.1°C. The results were divided into 4 groups: (W10) subjects warm, occlusion time 10 min.; (W5) subjects warm, occlusion time 5 min.; (C10) subjects cold, occlusion time

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