TABLE II. Mean Outflow Facility (C) of 12 Pairs of Rhesus Monkey Eyes After Infusion of Progessively Increasing Amounts of Liquid. One eye of each pair was perfused with saline containing plasmin (Eye 1), while the contralateral eye was perfused with saline alone (Eye 2).

ml infused	.05	.10	.25	.50	.90-1.0
Eye 1	.58 ± .16*	.61 ± .16	.83 ± .20	1.01 ± .23	1.14 ± .27
(Plasmin)	.34 – .81†	.38 – .83	.48 – 1.07	.62 – 1.25	.63 – 1.36
Eye 2	.45 ± .15	.38 ± .12	.50 ± .13	.63 ± .17	.67 ± .15
(Saline)	.21 – .58	.18 – .63	.31 – .76	.41 – .88	.45 – .93
Mean ratio: Eye 1 / Eye 2	1.29	1.60	1.66	1.60	1.70

* Mean ± S.D. † Range.

of monkey (*Macaca mulatta*). This effect is immediate and striking and levels off at a lower value than reached with saline alone. The effect resembles that produced by hyaluronidase in some animal species.

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Effect of Sodium State on Reactivity of Renal Juxtaglomerular Cells And Adrenal Zona Glomerulosa. (30720)

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Sodium deficiency increases the degree of granulation of renal juxtaglomerular cells (JGI) in several species, including man, whereas the converse has been observed in situations characterized by sodium retention. These alterations of JGI have been considered to result from changes in blood volume and pressure attendant with these Na states; and the juxtaglomerular apparatus has been visualized as a stretch receptor(1). Recent studies in our laboratory with hypertensive and nephrotic rats subjected to sodium depletion or salt overload delineated pressor and electrolytic effects on the juxtaglomerular apparatus. The results suggested that this morphologic unit might also represent an osmoreceptor(2-4). These, as well as contemplated studies of the juxtaglomerular apparatus, disclosed the need for information concerning the rate and degree of change in JGI following sodium depletion or salt overload and the recovery from these states. Miller and Hartroft(5) briefly noted the occurrence of prompt degranulation followed by hypergranulation of juxtaglomerular cells after administration of a sodium deficient diet. A similar reaction has been observed following partial renal artery occlusion(6).

Lack of a systematic study of the responsiveness of juxtaglomerular cells to sodium depletion or salt overload and the recovery phases from these states has prompted this report. The changes in JGI, which reflect renal renin content, have also been correlated with the width of the zona glomerulosa of adrenal cortices (ZGI), a morphologic index of adrenal cortical aldosterone production, be-

TABLE I. Effect of Peritoneal Dialysis and Sodium Deficient Diet on Body Weight, Fluid Intake, Serum Na and Cl, Hematocrit, Blood Pressure and Adrenal Weight.

Time after onset of dialysis	No.	Change in body wt (g)	Avg daily fluid intake (ml)	Serum (meq/l) Na	Serum (meq/l) Cl	Hematocrit	BP (mm Hg)	Adrenal wt (mg)
1 hr	8	—	—	124 ± 6	90 ± 3	55 ± 4	85 ± 15	42 ± 6
4 "	9	—	—	128 ± 7	94 ± 5	52 ± 6	90 ± 20	40 ± 7
6 "	10	− 1	—	130 ± 8	99 ± 3	55 ± 4	88 ± 12	39 ± 9
1 day	9	− 3	—	135 ± 4	106 ± 5	43 ± 4	93 ± 10	45 ± 10
3 "	7	−11	—	135 ± 7	107 ± 5	49 ± 4	90 ± 15	50 ± 8
5 "	10	−10	—	145 ± 5	103 ± 1	46 ± 5	95 ± 12	50 ± 7
7 "	8	−10	—	145 ± 5	108 ± 1	43 ± 4	85 ± 16	48 ± 12
14 "	9	—	—	142 ± 6	105 ± 3	45 ± 2	85 ± 12	52 ± 10
28 "	12	—	24	145 ± 3	106 ± 1	45 ± 3	90 ± 15	50 ± 12
Control	19	—	52	142 ± 5	102 ± 4	44 ± 4	88 ± 16	50 ± 14

± Standard deviation.

cause of their relationship in the renin-angiotensin-aldosterone system.

Materials and methods. Three hundred forty-two adult female Wistar rats weighing 150-200 g survived the required experimental periods. Groups of rats were sacrificed at 1, 4, 7 and 10 weeks after being placed on a sodium-free diet (General Biochemicals Co., Chagrin Falls, Ohio) and deionized water as drinking fluid. Others were sacrificed 1 hour after onset of peritoneal dialysis and 1, 4 and 6 hours and 1, 3, 5, 7, 14 and 28 days after this procedure and start of the sodium-free diet. Rats subjected to this procedure for 1 and 4 weeks were also sacrificed at 3, 7, 14 and 21 days after realimentation with a stock diet containing .3% sodium (Lab-Blox) and tap water.

Groups of rats receiving the stock diet, but 2% saline as drinking fluid, were sacrificed at 1, 4, 7 and 10 weeks. Five mg of deoxycorticosterone (DCA) were administered subcutaneously daily to groups of rats that received the stock diet and either 1% saline or tap water as drinking fluid for 3, 5, 7 and 14 days. Others received the stock diet and DCA and 1% saline for 2 weeks and were sacrificed at 3, 7, 10 and 14 days after cessation of treatment.

Peritoneal dialysis was performed by withdrawal of 12 ml of peritoneal fluid 3 hours after instillation of 25 ml of 5% glucose in water. With this technic the dialysates contained 15-20 meq/l of sodium.

Blood pressure was estimated at the beginning and termination of the experiment by a microphonic technic. Fluid intake of each rat was measured daily.

At autopsy kidneys were fixed in Helly's fluid and sections prepared and stained for estimation of juxtaglomerular index (JGI) according to the method of the Hartrofts(7). Adrenals were blotted, weighed and fixed in 10% formalin. Frozen sections were stained with oil red O and the average of 4 measurements of the width of the zona glomerulosa expressed as percentage of cortex (ZGI).

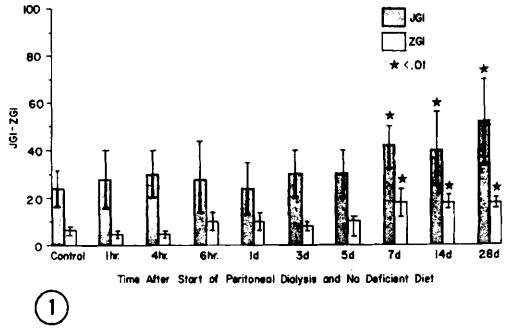
Serum Na, Cl, K and hematocrit were determined at time of sacrifice as described previously(2-4).

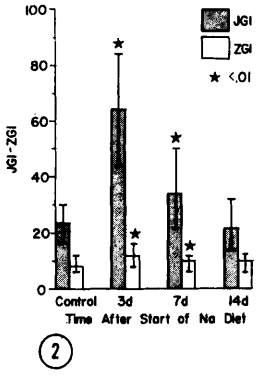
Results. Rats subjected to peritoneal dialysis and sodium deficient diet, as well as those receiving 1% or 2% saline as drinking fluid, exhibited only slight weight gain during the experimental periods, whereas those treated with DCA and saline experienced a slight loss (Tables I, II, III). Those maintained on the sodium deficient diet without dialysis and rats receiving DCA alone gained weight comparable to untreated rats in this laboratory.

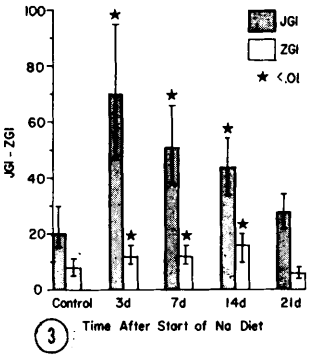
A statistically significant increase ($P<.01$) in JGI and ZGI was first observed at 1 week following peritoneal dialysis and administration of sodium-free diet and was most pronounced after 4 weeks of this regimen (Fig. 1). Repletion of dietary sodium in rats depleted for 1 and 4 weeks reduced JGI and ZGI to normal ($P>.1$) after 2 and 3 weeks, respectively (Fig. 2, 3). Administration of the sodium-free diet without peritoneal dialysis for as long as 10 weeks failed statistically

to alter JGI and ZGI (Table II).

Similarly, substitution of 1% or 2% saline as drinking fluid to rats maintained on the sodium-containing diet for a comparable period did not provoke a significant change in these parameters ($P>.1$) (Table II). On the other hand, DCA and 1% saline as drinking fluid significantly decreased JGI and ZGI ($P<.01$) after the 10th day of treatment. JGI and ZGI returned to normal 2 weeks







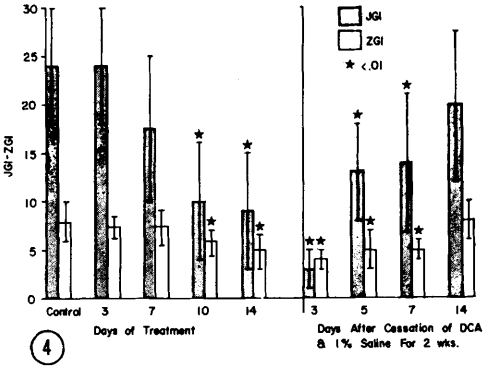


FIG. 1. Effect of peritoneal dialysis and Na-deficient diet on JGI and ZGI.
FIG. 2. Effect of Na repletion on JGI and ZGI after 1 week of depletion.
FIG. 3. Effect of Na repletion on JGI and ZGI after 4 weeks of depletion.
FIG. 4. Effect of DCA and 1% saline on JGI and ZGI (left) and changes after cessation of treatment (right).

TABLE II. Effect of Dietary Sodium on Change in Body Weight, Fluid Intake, Serum Na, Cl, Hematocrit, Blood Pressure, Adrenal Weight, JGI and ZGI.

Duration	No.	Change in body wt (g)	Avg daily fluid intake (ml)	Serum (meq/l) Na	Serum (meq/l) Cl	Hematocrit	BP (mm Hg)	Adrenal wt (mg)	JGI	ZGI
1% saline* 10 wk	10	+40	79	144 ± 3	108 ± 6	42 ± 6	88 ± 16	48 ± 12	30 ± 10	7 ± 3
2% saline† 10 wk	10	+28	88	146 ± 7	104 ± 8	47 ± 6	90 ± 15	47 ± 6	23 ± 8	8 ± 2
Na defic‡ 10 wk	10	+35	50	140 ± 7	104 ± 6	43 ± 5	85 ± 16	50 ± 12	27 ± 7	8 ± 1

± Standard deviation.
* Groups of 12, 12, 10 also sacrificed at 1, 4, 7 wk
† " " 10, 11, 7 " " " 1, 4, 7 "
‡ " " 8, 8, 10 " " " 1, 4, 7 "
No difference in results from those at 10 wks.

TABLE III. Effect of DCA and 1% Saline on Body Weight, Fluid Intake, Serum Na, Cl, Hematocrit, Blood Pressure and Adrenal Weight.

	No.	Change in body wt (g)	Avg daily fluid intake (ml)	Serum (meq/l) Na	Cl	Hematocrit	B P (mm Hg)	Adrenal wt (mg)
DCA + saline								
3 days	8	− 2	—	140 ± 6	106 ± 4	46 ± 3	94 ± 12	52 ± 8
7 "	8	− 2	—	160 ± 8	110 ± 8	42 ± 5	97 ± 8	46 ± 12
10 "	8	− 4	—	156 ± 7	112 ± 9	47 ± 5	140 ± 14	58 ± 16
14 "	8	−10	162	155 ± 10	111 ± 6	46 ± 4	145 ± 10	50 ± 8
DCA only*								
14 days	8	+15	53	148 ± 12	109 ± 8	48 ± 6	108 ± 15	52 ± 6
After DCA + saline for 14 days								
3 days	8	+ 8	—	163 ± 7	108 ± 6	46 ± 4	120 ± 18	54 ± 12
7 "	8	+20	—	150 ± 4	107 ± 6	47 ± 6	90 ± 16	50 ± 8
14 "	8	+32	56	142 ± 6	105 ± 4	44 ± 2	101 ± 12	56 ± 16

± Standard deviation.
* Group of 8 also sacrificed at 3 and 7 days. No difference in results from those at 14 days.

following cessation of DCA and saline administration for 2 weeks (Fig. 4). DCA without saline failed significantly to alter these parameters ($P>.01$).

A significant increase in blood pressure was encountered 10 days after onset of DCA and saline treatment ($P<.01$) (Table III). No alteration of pressure was observed in other groups ($P>.1$).

Serum Na and Cl were significantly decreased ($P<.01$) in rats subjected to peritoneal dialysis only during the initial 72 hours following such treatment and an increase in hematocrit and decrease in adrenal weights ($P<.01$) (Tables I, II, III, IV) during the first 6 hours. A moderate but significant elevation of Na and Cl was noted in rats receiving DCA and saline ($P<.01$) (Table III).

The average daily fluid intake of rats subjected to the sodium deficient diet only was comparable to that of controls, whereas that of those receiving 1% or 2% saline with or without DCA was markedly increased (Tables I, II, III).

Comment. Change in granularity of juxtaglomerular cells (JGC), *pari passu* renal renin, in rats under the conditions utilized herein is dependent upon a greater stimulus than that afforded by regulation of dietary intake of sodium. This information confirms previous impressions obtained in this regard during the course of related experiments(2-4). Although an increase in plasma renin has been noted following dietary restriction of sodium and the converse with salt overloading in healthy humans(8) no mention of the serum sodium content was made. In this study changes in JGI were encountered only in those instances in which augmentation of sodium deficiency or salt overloading also

TABLE IV. Effect of Na Repletion on Serum Na, Cl, Hematocrit, Blood Pressure, and Adrenal Weight After 1 and 4 Weeks of Na Depletion.

	Time after repletion	No.	Serum (meq/l) Na	Cl	Hematocrit	B P (mm Hg)	Adrenal wt (mg)
1 wk depletion:							
	3 days	9	146 ± 6	107 ± 3	40 ± 6	85 ± 12	45 ± 12
	7 "	12	146 ± 8	106 ± 3	43 ± 3	90 ± 10	50 ± 8
	14 "	11	145 ± 6	108 ± 3	46 ± 4	88 ± 12	50 ± 4
4 " "							
	3 "	8	147 ± 6	104 ± 3	42 ± 8	90 ± 8	55 ± 13
	7 "	8	142 ± 7	108 ± 3	41 ± 3	93 ± 8	45 ± 8
	14 "	7	149 ± 8	108 ± 4	43 ± 3	88 ± 14	52 ± 10
	21 "	7	142 ± 8	106 ± 7	43 ± 4	86 ± 16	46 ± 10

± Standard deviation.

resulted in alteration of serum Na levels. That the level of serum Na may be highly significant in inducing changes in JGI is also emphasized by our recent observations in cirrhotic patients(9). JGI and ZGI were elevated only in those which exhibited ascites (sodium retention) accompanied by hyponatremia. An inverse relationship between serum sodium and JGI and ZGI has also been previously observed in unselected patients at necropsy(10). These findings are consonant with the view signifying the JGC as an osmoreceptor(2-4,11). The decrease in JGI and ZGI observed in rats treated with DCA and saline prior to the onset of hypertension in this study further substantiates this view. On the other hand, the normal levels of serum Na observed after 3 days in dialyzed rats receiving the sodium deficient diet indicate that the JGC response may not be invariably correlated with the level of this electrolyte in the serum. Changes in JGI do not appear related to alteration in body weight induced by the various regimens since these were comparable in sodium deficient rats exhibiting elevated JGI and ZGI and those receiving DCA and saline in which the converse of these parameters was encountered.

Although increased dietary sodium administered to rats over prolonged periods may result in hypertension(12), no alteration of blood pressure was encountered in this study in those animals subjected to 1% or 2% saline as drinking fluid. However, such a response is not invariable and, as noted here and emphasized by Hall and Hall(13), may be related to amount of saline ingested. Hypertension was observed only in those rats receiving saline which were also treated with DCA and exhibited a marked increase in fluid intake.

It is of interest that restoration of JGI and ZGI to normal levels in rats previously subjected to augmented depletion or overload is delayed and, in part, dependent upon the duration of such electrolyte change. This finding does not detract from the view signifying the secretory nature of JGC since delay in functional and structural recovery from pathologic states is often encountered in other endocrine structures, *viz.*, thyroid. We have not observed the prompt degranulation of JGC after inducing sodium depletion as noted by Miller and Hartroft(5). Perhaps this type of reaction is characteristic of the weanling rats which were utilized by these investigators.

Summary. Changes in degree of granularity of juxtaglomerular cells (JGI) and width of zona glomerulosa of adrenal cortices (ZGI) of adult Wistar rats subjected to dietary restriction of sodium or salt overload was encountered only when these regimens were supplemented by peritoneal dialysis or administration of DCA, respectively. Increase in these parameters following the former and decrease after the latter both exhibited delay in onset as well as their recovery to control values following sodium repletion or cessation of salt overloading. Decrease in JGI and ZGI occurred prior to the onset of hypertension in rats receiving DCA and saline. These findings are consonant with views concerning the "endocrine" nature of JGC as well as their role as osmoreceptors.

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