JCI The Journal of Clinical Investigation

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J Clin Invest. 1961;40(8):1475-1485. https://doi.org/10.1172/JCI104378.

Research Article

EFFECT OF ORGANOMERCURIALS ON THE RENAL CONCENTRATING OPERATION IN HYDROPENIC MAN: COMMENTS ON SITE OF ACTION*

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(Submitted for publication February 3, 1961; accepted April 7, 1961)

The renal site of action of organomercurials has variously been placed within the proximal or distal tubule on the basis of numerous observations in dog and man (1–9). Experiments in maximally hydrated man recently reported from this laboratory showed that free water clearance remained relatively fixed throughout the major portion of the diuresis induced by the organomercurial, meralluride (10). In addition, free water clearance could generally be augmented by superimposing a nonspecific solute diuretic [1] during the mercurial diuresis. A hypothesis was suggested that placed a major inhibitory action of organomercurials at a late distal segment, beyond the water-clearing site where, it was assumed, a process of isosmotic salt and water reabsorption takes place.

Previous data concerning the effect of mercurial diuresis on the concentrating operation in hydropenic man have led to varied conclusions. Some have argued that a mercurial diuresis limits the renal concentrating capacity (11). Others have found that a mercurial diuresis superimposed upon or developing simultaneously with a nonspecific solute diuresis does not limit the capacity of the collecting duct to extract solute-free water (12, 13).

Medullary hypertonicity is established primarily by the deposition within the interstitium of salt absorbed from the ascending limb of the loop of Henle. Under conditions of maximal hydropenia, the variable hypotonicity produced in the ascending limb is dissipated by the outward diffusion of water through a permeable tubular membrane, so that isotonicity is reattained before the end of the distal tubule (14–16). The quantity of solute-free water (T^cH_2O) extracted from this isosmotic fluid as it courses through the collecting duct may afford some measure of the quantity of solute transported to the medullary interstitium.

It appeared reasonable that a comparison of the urinary concentrating characteristics during a mercurial diuresis and a nonspecific solute diuresis might elucidate further the site of action of organomercurials in man.

MATERIALS AND METHODS

Four separate groups of studies were performed in normal, maximally hydropenic subjects. In the first or control group, the characteristics of the urine were studied during the infusion of hypertonic mannitol or salt and after the administration of aminophylline. In the second series of experiments, after different rates of solute excretion had been established and maintained at a steady state, an organomercurial was administered intravenously. In the next group of studies, one of the nonspecific solute diuretics used in the control group was superimposed after a mercurial diuresis had developed. Finally, glomerular filtration rate (GFR) and effective renal plasma flow (ERPF) were reduced during the course of comparable diureses produced by the infusion of hypertonic salt or the administration of an organomercurial.

All subjects were normal young females, free of cardiovascular or renal diseases. Each was maintained on a regular diet but was deprived of food and water for 16 hours prior to the study, which began at 8:00 a.m. Twelve hours before the experiment, each subject received an intramuscular injection of 2.5 U of vasopressin (Pitressin tannate in oil). Throughout each experiment the infusion contained sufficient aqueous vasopressin to provide at least 200 mU per hour. When filtration rate or renal plasma flow or both were measured, quantities of inulin and para-aminohippuric acid sufficient to produce satisfactory plasma levels were included in the infusion following a priming dose of each agent. All subjects were recumbent during the experiments. Catheterization was performed with a no. 16 French catheter,

* Supported by Grant A-277 from the National Institute of Arthritis and Metabolic Diseases, Bethesda, Md.

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[1] The term "nonspecific solute diuretic" is used for those agents which act to increase the quantity of isosmotic fluid escaping proximal tubular reabsorption.

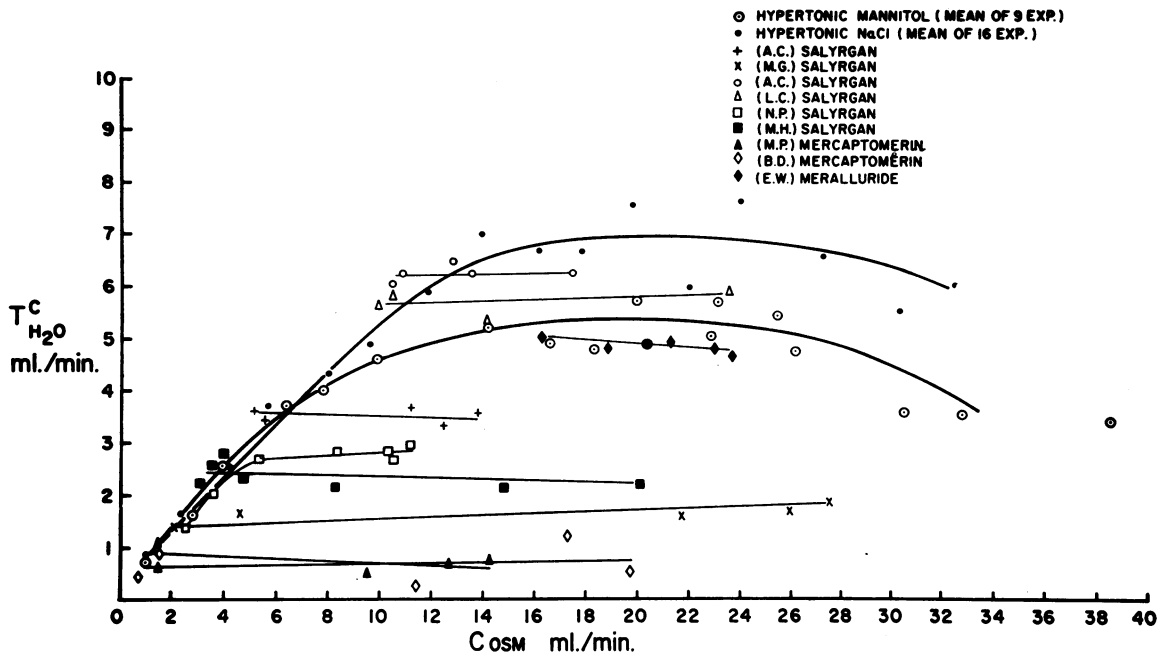

FIG. 1. EFFECT OF ORGANOMERCURIALS AND HYPERTONIC SALT AND MANNITOL INFUSIONS ON SOLUTE EXCRETION (C_{Osm}) AND FREE WATER REABSORPTION (T^cH_2O). In the mercurial experiments the first point represents the value obtained after the fall in T^cH_2O and C_{Osm} before diuresis which was noted in some experiments with Salyrgan and mercaptomerin, and after the first phase of the meralluride diuresis.

and air washouts were used to assure complete bladder emptying. A urine specimen was collected at the beginning of each experiment and at approximately 10 to 30 minute intervals thereafter, depending upon the rate of urine flow. A blood specimen was taken prior to the start of the infusion and every 45 to 60 minutes thereafter from an antecubital vein through a heparinized indwelling needle. The infusions were maintained at a constant rate by a Bowman infusion pump.

In the first group of studies, 9 subjects received an infusion of 10 per cent mannitol, administered at progressively increasing rates from 10 to 35 ml per minute. In a similar manner, 2.5 per cent salt was infused into 16 subjects. When possible, the experiments were continued until a urine flow of 25 to 40 ml per minute had been reached. In 4 subjects, 500 mg of aminophylline was administered intravenously over a 5-minute period to comparably hydropenic subjects.

In the second series of experiments, 26 similarly hydropenic subjects were infused with either normal saline at 1.5 ml per minute or 10 per cent mannitol at 1.5 or 4.5 ml per minute. After a steady state was achieved, three 20-minute urine samples were obtained. Thereafter, 3 ml of meralluride, mercaptomerin, or mersalyl (Salyrgan without theophylline) was injected intravenously. After administration of the mercurial, each experiment was continued for approximately 200 minutes.

In the third group of experiments, approximately 100 to 120 minutes after an organomercurial was administered and a considerable diuresis established, 12 subjects received an infusion of 10 per cent mannitol, 2.5 per cent salt or 500 mg of aminophylline, as in the control experiments. Thereafter the experiments were continued for another 60 to 90 minutes.

In the fourth group of experiments, the blood pressure was suddenly reduced during a Salyrgan- or meralluride-induced diuresis in 4 subjects, and during a diuresis produced by the infusion of 3 per cent salt at 6.5 ml per minute in 3 subjects. This fall in blood pressure was accomplished by the intravenous administration of 50 to 100 mg of SC1950 (1-ethyl-2,6-dimethylluptidine ethobromide),[2] a ganglionic blocking agent, after the application of venous tourniquets to the thighs. The hypotension was maintained for 30 to 60 minutes. Thereafter the blood pressure returned to control levels spontaneously or after the removal of the tourniquets.

All urine and plasma samples were analyzed for osmolality, sodium, potassium and chloride concentrations. When appropriate, inulin and para-aminohippuric acid determinations were also carried out. All determinations were done by methods previously described from this laboratory (17). Clearances were calculated by standard methods.

Solute clearance (C_{Osm}) was calculated from the formula $C_{Osm} = U_{Osm}\ V/P_{Osm}$, where U_{Osm} represents urine osmolality, V is urine flow and P_{Osm} is plasma osmolality.

[2] This drug was generously supplied by G. D. Searle Co., Chicago, Ill., through Dr. Irwin C. Winter, Clinical Director.

TABLE I

Summary of effect of organomercurials on solute excretion and free water reabsorption in hydropenic man

Subject	Mercurial		Solute excretion* (UV/P)	Free water reabsorption (T^cH_2O)
			ml/min	*ml/min*
N.P.	Salyrgan	I	2.2	1.3
		Ia	5.2	2.7
		II	11.1	2.9
M.G.	Salyrgan	I	4.6	1.6
		II	27.8	1.8
E.P.	Salyrgan	I	3.0	1.6
		II	9.2	2.0
H.B.	Salyrgan	I	2.5	1.6
		Ia	4.8	3.1
		II	10.3	2.5
M.H.	Salyrgan	I	3.7	2.6
		II	20.1	2.1
L.J.	Salyrgan	I	4.0	2.6
		II	23.7	2.9
A.C.	Salyrgan	I	4.3	3.0
		II	13.8	3.5
L.G.	Salyrgan	I	5.5	3.6
		II	17.4	3.3
A.S.	Salyrgan	I	8.3	4.7
		II	20.0	4.1
L.C.	Salyrgan	I	10.4	5.8
		II	23.6	5.9
A.C.	Salyrgan	I	10.8	6.2
		II	17.4	6.2
B.D.	Mercaptomerin	I	1.3	0.9
		II	19.7	0.5
M.P.	Mercaptomerin	I	1.4	0.6
		II	12.6	0.7
J.R.	Mercaptomerin	I	1.6	1.1
		II	11.0	0.8
M.R.	Mercaptomerin	I	3.4	2.4
		II	11.4	2.2
E.P.	Mercaptomerin	I	5.1	3.3
		II	16.0	4.0
T.C.	Mercaptomerin	I	9.7	4.2
		II	24.0	4.6
Q.R.	Mercaptomerin	I	12.5	7.0
		II	18.3	8.0
A.S.	Meralluride	I	2.9	1.8
		Ia	6.7	3.0
		II	10.5	3.4
H.L.	Meralluride	I	3.3	1.9
		II	9.0	1.8
R.M.	Meralluride	I	4.1	2.5
		Ia	7.5	3.6
		II	13.6	3.3
J.P.	Meralluride	I	6.2	3.4
		II	13.6	3.6
M.P.	Meralluride	I	5.5	3.5
		II	15.3	4.0
C.D.	Meralluride	I	5.8	4.0
		II	12.5	3.2
E.W.	Meralluride	I	16.2	5.0
		II	23.9	4.6
P.M.	Meralluride	I	13.3	6.9
		II	20.6	6.5

* I represents the period prior to the onset of the mercurial diuresis (after early transient diuresis with meralluride and after initial decrease in C_{Osm} and T^cH_2O in ten of the Salyrgan and mercaptomerin experiments); Ia represents the period showing the small increase in T^cH_2O with the initial rise in C_{Osm} during the mercurial diuresis; II represents the period of peak mercurial effect.

When the urine is concentrated above isotonicity, C_{Osm} is greater than V. Thus, the amount of solute-free water (T^cH_2O) absorbed from isosmotic fluid to concentrate the urine is calculated from the formula $T^cH_2O = C_{Osm} - V$.

RESULTS

Nonspecific solute diuretics (Figure 1). In these control studies, all C_{Osm} and corresponding T^cH_2O values obtained for each nonspecific diuretic were grouped about the closest successive 2-ml increment in C_{Osm}. The data represent the mean values for each group.

During hypertonic mannitol infusions,[3] as C_{Osm} increased from 1 to 14.0 ml per minute, there occurred a progressive increase in T^cH_2O from 0.7 to 5.4 ml per minute. Between a C_{Osm} of 14.0 and 23.1 ml per minute, T^cH_2O remained essentially unchanged and thereafter as C_{Osm} increased to 38.6 ml per minute, T^cH_2O declined to 3.6 ml per minute. With hypertonic salt qualitatively similar results were obtained, so that as C_{Osm} increased from 1.3 to 13.7 ml per minute, T^cH_2O increased

[3] Similar results were obtained with isotonic mannitol infusions.

from 0.9 to 6.9 ml per minute. Between a C_{Osm} of 13.7 and 27.3 ml per minute, T^cH_2O remained relatively fixed, and then as C_{Osm} was further increased to 32.6 ml per minute, T^cH_2O tended to diminish slightly to 6.1 ml per minute. After the administration of aminophylline, as C_{Osm} increased from 2.0 to 12.4 ml per minute, T^cH_2O rose from 1.4 to 6.6 ml per minute, producing a curve very similar to that observed after the administration of hypertonic salt.

Organomercurials (Tables I and II, Figure 1). The major diuresis induced by organomercurials began approximately 70 minutes after intravenous administration. The peak effect occurred at 140 minutes and a considerable diuresis was still present after 3 hours.

In 10 of 18 experiments the administration of Salyrgan or mercaptomerin caused a slight fall in C_{Osm} and T^cH_2O, averaging 0.9 ml per minute for each, prior to the onset of the diuresis (Table II; M.G., L.C.). In 4 experiments (Tables I and III, N.P., H.B., A.S., R.M.; Figure 1, N.P.), at the onset of the major mercurial diuresis, an increase in T^cH_2O averaging 1.3 ml per minute (range, 1.1 to 1.5) was observed in association with an initial mean increase in urine flow of 1.8 ml per minute (range, 0.8 to 2.6) and C_{Osm} of 3.1 ml per minute (range, 2.3 to 3.8).

In each meralluride experiment an immediate but transient increase in C_{Osm} averaging 4.3 ml per minute (range, 2.7 to 6.5) was associated with a mean increase in T^cH_2O of 2.2 ml per minute (range, 1.5 to 3.2) (Table II, J.P.; Figure 2).

TABLE II

The effect of organomercurials on solute excretion and free water reabsorption in hydropenic man (three typical experiments)

Subject	Time	Urine flow (V)	Urine osmolality (U)	Solute clearance (UV/P)	Free water reabsorption (T^cH_2O)	Inulin clearance (UV/P)
	min	*ml/min*	*mOsm/kg*	*ml/min*	*ml/min*	*ml/min*
M.G.	Control	1.3	801	3.8	2.5	110
		Salyrgan (3 ml)				
	0 – 29.5	0.7	813	2.1	1.4	72
	29.5 – 58	3.0	430	4.6	1.6	93
	58 – 74	20.1	301	21.7	1.6	125
	74 – 82.5	26.0	298	27.8	1.8	115
	82.5 – 89	24.3	300	26.0	1.7	98
J.P.	Control	1.1	730	2.9	1.8	112
		Meralluride (3 ml)				
	0 – 12.5	4.1	580	8.9	4.8	160
	12.5 – 25	3.3	662	8.0	4.7	
	25 – 42	3.4	687	8.6	4.8	
	42 – 55.5	2.7	677	6.9	4.2	
	55.5 – 65.5	3.2	653	7.9	4.7	
	65.5 – 75	3.3	628	7.8	4.5	131
	75 – 89	2.8	590	6.2	3.4	100
	89 – 100.5	3.4	532	6.9	3.5	
	100.5 – 111	4.9	463	8.5	3.6	
	111 – 122	6.7	420	10.6	3.9	
	122 – 135.5	8.3	383	11.9	3.6	
	135.5 – 145.5	9.0	365	12.3	3.3	104
	145.5 – 155	10.0	359	13.6	3.6	111
	155 – 164	9.9	362	13.5	3.6	
	164 – 173.5	9.9	362	13.5	3.6	
L.C.	Control	4.6	668	11.5	6.9	111
		Salyrgan (3 ml)				
	0 – 11.5	4.3	622	9.9	5.6	83
	11.5 – 31.5	4.6	610	10.4	5.8	80
	31.5 – 56	4.9	559	10.5	5.1	69
	56 – 81.5	8.8	439	14.1	5.3	81
	81.5 – 99	17.7	364	23.6	5.9	114

TABLE III

The effect of nonspecific solute diuretics superimposed on organomercurials on solute excretion and free water reabsorption (two typical experiments)

Subject	Time	Urine flow (V)	Urine osmolality (U)	Solute clearance (UV/P)	Free water reabsorption (T^cH_2O)	Inulin clearance (UV/P)
	min	*ml/min*	*mOsm/kg*	*ml/min*	*ml/min*	*ml/min*
E.P.	Control	2.0	861	5.8	3.8	95
			Mercaptomerin (3 ml)			
	0 – 34.5	2.0	842	5.9	3.9	102
	34.5 – 51	1.8	801	5.1	3.3	95
	51 – 68.5	5.0	553	8.5	3.5	
	68.5 – 79.5	6.6	446	10.3	3.7	90
	79.5 – 91.5	9.5	395	13.2	3.7	96
	91.5 – 102	10.5	377	14.0	3.5	92
	102 – 114.5	11.5	372	15.2	3.7	94
	114.5 – 126.5	12.0	375	16.0	4.0	96
			Aminophylline (500 mg i.v.)			
	126.5 – 137.5	17.5	361	22.6	5.1	117
	137.5 – 149	16.2	387	22.4	6.2	117
N.P.	Control	1.0	657	2.4	1.4	122
			Salyrgan (3 ml)			
	0 – 15	0.9	657	2.2	1.3	105
	15 – 37	0.8	682	2.1	1.3	62
	37 – 61.5	1.5	634	3.6	2.1	62
	61.5 – 83.5	2.5	528	5.2	2.7	
	83.5 – 110	5.5	401	8.3	2.8	
	110 – 128	7.4	366	10.2	2.8	62
	128 – 139	7.8	355	10.4	2.6	63
	139 – 147.5	8.2	355	11.1	2.9	69
			Mannitol (10%)			
	147.5 – 164	12.5	311	15.7	3.2	68
	164 – 175.5	21.8	315	25.5	3.7	76
	175.5 – 189	25.9	315	30.0	4.1	77
	189 – 200.5	32.2	326	38.1	5.9	104

Thereafter C_{Osm} and T^cH_2O returned toward control levels within 30 to 60 minutes.

After the minor variations in T^cH_2O occurring before the major solute diuresis, the characteristics of all 26 mercurial diureses were similar. Specifically, C_{Osm} rose an average of 10.1 ml per minute (range, 3.8 to 23.2) with no mean change in T^cH_2O. T^cH_2O increased a mean of 0.4 ml per minute (range, 0.1 to 1.0) in 13, decreased a mean of 0.4 ml per minute (range, 0.1 to 0.8) in 12, and did not change in 1 subject. This constancy of T^cH_2O obtained despite the marked variation in the levels of C_{Osm} (range, 1.3 to 16.2 ml per minute) and T^cH_2O (range, 0.9 to 7.0 ml per minute) that existed prior to the onset of the major mercurial diuresis. There were no measurable differences among the three mercurials in their effect on T^cH_2O despite a mean increase in C_{Osm} after Salyrgan of 11.8 ml per minute, after mercaptomerin of 11.3 ml per minute and after meralluride of 6.8 ml per minute.

The glomerular filtration rate was determined in 15 of 18 experiments with Salyrgan and mercaptomerin. In 14, a mean decrease in GFR of 26 per cent (range, 13 to 45 per cent) occurred within 40 minutes of administration, prior to the onset of the solute diuresis. This was associated with a fall in C_{Osm} and T^cH_2O in 10 cases, as noted above (Table II, M.G., L.C.). In 8, the GFR slowly returned toward control levels and in 6 the decrease in GFR persisted throughout the experiment. In 6 meralluride experiments in which glomerular filtration rate was measured, an immediate increase occurred, averaging 39 per cent (range, 11 to 64 per cent) (Table II, J.P.). This rise generally disappeared within 50 minutes.

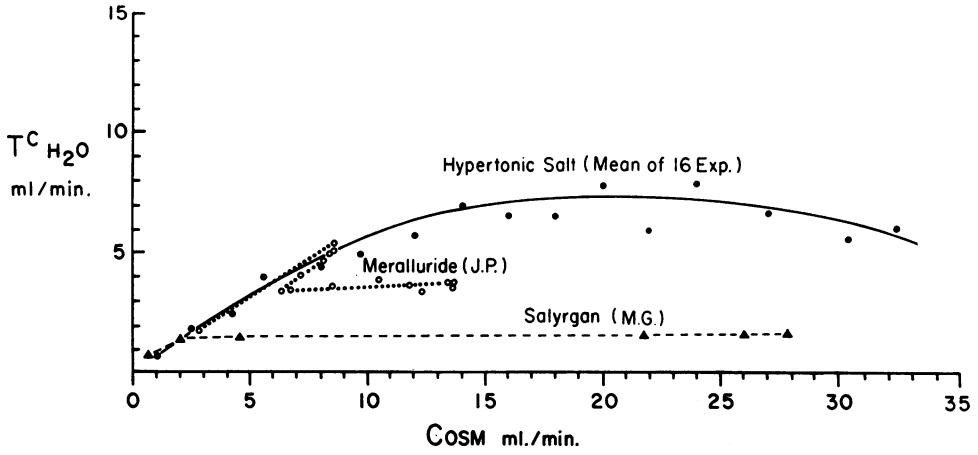

FIG. 2. COMPARISON OF THE 2 PHASES OF A MERALLURIDE DIURESIS WITH THE DIURESIS PRODUCED BY A NONTHEOPHYLLINE-CONTAINING MERCURIAL (SALYRGAN) AND BY A NONSPECIFIC SOLUTE DIURETIC (HYPERTONIC SALT).

The effective renal plasma flow, when measured, varied as the GFR, i.e., there was a prompt fall averaging 31 per cent when the GFR fell after administration of Salyrgan or mercaptomerin and there was a coincident rise averaging 29 per cent when GFR transiently increased with meralluride.

During the mercurial diuresis the increase in sodium and chloride excretion was commensurate with the increase in C_{Osm}. In the majority of experiments potassium excretion fell.

Nonspecific solute diuretics superimposed on organomercurials (Tables III and IV, Figure 3). In the 12 experiments in which a nonspecific agent (hypertonic mannitol, hypertonic salt or aminophylline) was superimposed upon a mercurial diuresis, an increase in T^cH_2O was noted in each instance. In 10 of these 12 experiments T^cH_2O increased at least 1.4 ml per minute, with a mean of 2.0 (range, 1.4 to 3.0). This increase in T^cH_2O was associated with a mean increase in C_{Osm} of 15.5 ml per minute (range, 6.4 to 30.5). The in-

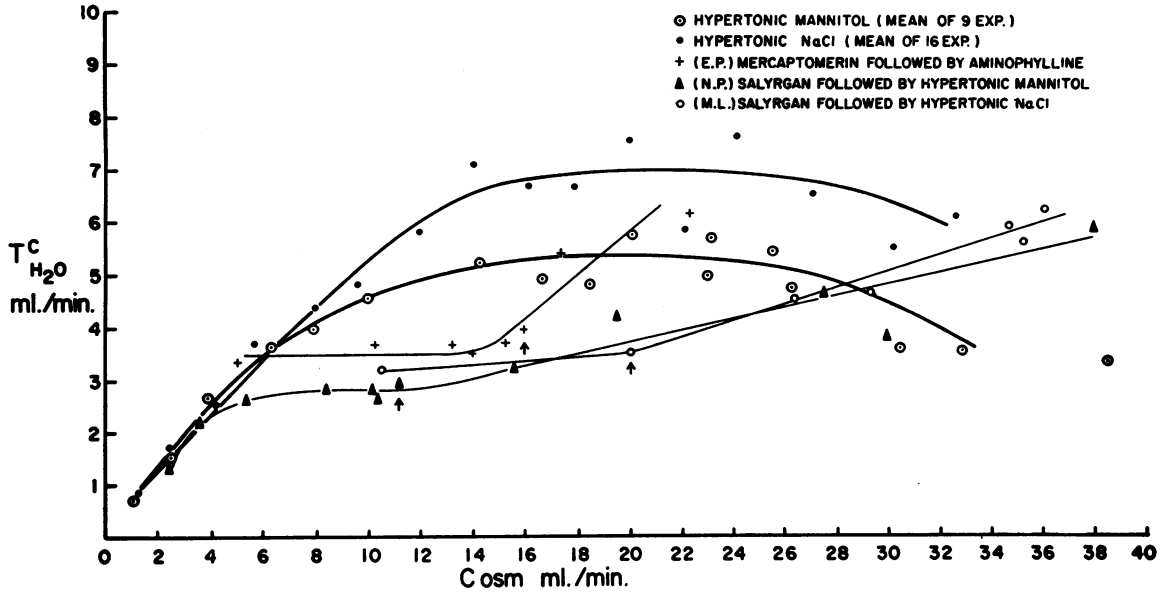

FIG. 3. EFFECT OF A NONSPECIFIC SOLUTE DIURETIC SUPERIMPOSED ON ORGANOMERCURIAL DIURESIS. The arrow indicates the point of administration of the nonspecific agent.

TABLE IV

Effect of nonspecific solute diuretics superimposed on organomercurials on solute excretion and free water reabsorption in hydropenic man

Subject	Mercurial		Solute excretion* (UV/P)	Free water reabsorption (T^cH_2O)
			ml/min	*ml/min*
M.G.	Salyrgan	I	26.0	1.7
		IIa	41.7	3.4
N.P.	Salyrgan	I	11.1	2.9
		IIb	38.1	5.9
A.C.	Salyrgan	I	12.4	3.3
		IIc	21.5	5.1
L.G.	Salyrgan	I	17.4	3.3
		IIc	26.1	4.2
M.L.	Salyrgan	I	20.0	3.5
		IIa	36.3	6.3
B.D.	Mercaptomerin	I	19.7	0.5
		IIb	36.6	2.4
M.P.	Mercaptomerin	I	12.7	0.7
		IIa	26.5	1.2
M.R.	Mercaptomerin	I	10.7	2.1
		IIc	17.8	3.5
E.P.	Mercaptomerin	I	16.0	4.0
		IIc	22.4	6.2
A.S.	Meralluride	I	9.9	3.4
		IIa	23.6	4.8
H.L.	Meralluride	I	8.1	1.7
		IIb	38.6	3.3
R.M.	Meralluride	I	10.0	2.6
		IIb	32.1	4.3

* I represents the period during the maximal mercurial diuresis just prior to the administration of the superimposed agent; II represents the period of peak solute excretion and free water reabsorption with the superimposed agent; a = 2.5% sodium chloride; b = 10% mannitol; c = aminophylline.

crease in T^cH_2O produced by the superimposed solute diuretics was comparable for the three organomercurials.

Reduction in glomerular filtration rate during organomercurial and hypertonic salt diureses (Figure 4). In the experiments in which the blood pressure was suddenly reduced during a mercurial diuresis, GFR decreased a mean of 68 per cent and ERPF a mean of 60 per cent. Coincidentally, a prompt decrease in C_{Osm} occurred, averaging 11.5 ml per minute, but urine osmolality increased only slightly (mean increase, 27 mOsm per kg). As solute excretion diminished, urine osmolality and T^cH_2O values were significantly below those noted at comparable levels of solute clearance as the diuresis was developing.

When the blood pressure was abruptly lowered during the diureses induced by hypertonic salt infusions, GFR and ERPF were reduced an average of 42 and 48 per cent, respectively. Coincidentally, C_{Osm} decreased a mean of 8.7 ml per minute, but with this reduction in solute excretion an appreciable rise in urine osmolality occurred, averaging 237 mOsm per kg. As solute excretion fell, urine osmolality and calculated T^cH_2O values were similar to those observed at comparable C_{Osm}'s during the development of the diuresis. The reductions in solute excretion persisted for approximately 30 minutes in both the mercurial and salt-infusion studies.

DISCUSSION

In those experiments with Salyrgan and mercaptomerin in which an early reduction in solute excretion and T^cH_2O values was observed, a simultaneous fall in filtration rate was noted. Although the recorded mean reduction in GFR of 26 per cent is exaggerated by the abrupt fall in urine flow (18), a modest reduction in filtration rate appears to be responsible for these changes in C_{Osm} and T^cH_2O. The early but transient increase in filtration rate noted with meralluride was probably also exaggerated by the abrupt increase in urine flow from control hydropenic levels. However, this rise in filtration rate may in part explain the transient first phase of a meralluride diuresis in which C_{Osm} and T^cH_2O increased (Figure 2).

Apart from these early minor variations, it is evident from the data that T^cH_2O remains relatively constant during a mercurial diuresis at that level which obtained immediately prior to the development of the major saluresis (Tables I and II, Figure 1). This finding is also apparent in the data presented by Au and Raisz (11) and explains the diverse conclusions drawn by these and other authors concerning the effect of mercurials on the concentrating operation. When the mercurial is administered under conditions of relatively low solute clearance, the persistence of low T^cH_2O levels is reflected as an apparent defect in the concentrating capacity of the kidney (11). However, when the mercurial is administered to a

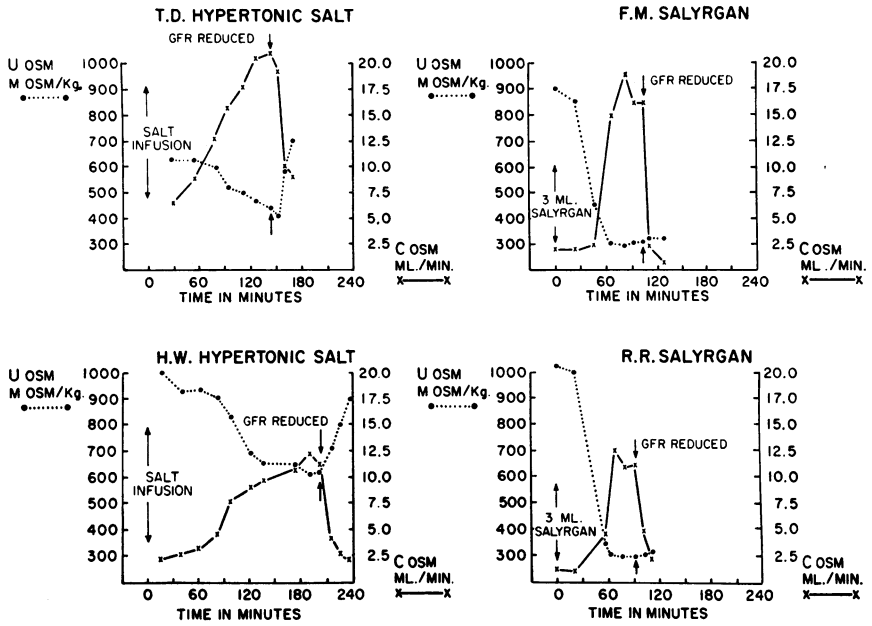

FIG. 4. CHANGES IN URINE OSMOLALITY (U_{Osm}) AND SOLUTE EXCRETION (C_{Osm}) DURING THE DIURESIS PRODUCED BY ORGANOMERCURIALS OR HYPERTONIC SALT INFUSIONS AND AFTER THE EXPERIMENTAL REDUCTION IN GFR. The arrows indicate the time at which GFR and C_{Osm} were reduced.

subject excreting large quantities of solute and extracting appreciable quantities of solute-free water, impaired concentrating capacity is not demonstrable (12, 13).

At very low rates of solute flow, a limiting factor in obtaining a maximal value for T^cH_2O appears to be the meager quantity of water (and solute) reaching the collecting duct. The early increase in T^cH_2O noted with very small increments in solute clearance in both the control studies and several of the mercurial experiments (Tables I and III, N.P., H.B., A.S., R.M.; Figure 1, N.P.) may therefore be attributed to the increased quantity of water presented to this site. However, on the basis of mercurial data, it is apparent that the presentation of increasing quantities of water (and solute) to the collecting duct does not in itself assure the development of a maximal T^cH_2O. No substantial evidence is available to suggest that mercurials inhibit the action of antidiuretic hormone or limit tubular permeability to water (1, 5, 11, 19–21). Therefore, the rising T^cH_2O in the control studies, contrasted with the constancy of this parameter during a comparable mercurial diuresis, suggests that during the former more osmotically active particles were present within the medullary interstitium. An agent which inhibits proximal tubular salt and water reabsorption would present more salt to the loop of Henle, a fraction of which could be transferred into the interstitial fluid (14). By this means, additional osmotic activity would be provided for the extraction of water from the isosmotic fluid in the adjacent collecting duct. At comparable levels of solute excretion, salt as the loading solute would be expected to present more absorbable particles than mannitol, and thereby produce a higher maximal value for T^cH_2O. Such was the case in the control studies reported here and apparent in the data reported by others in the hydropenic hamster and rat (14). Accordingly, peak T^cH_2O values will be achieved in hydropenic man when the dynamics of flow within the loop provide a maximal quantity of solute for transport into the interstitium and, simultaneously, adequate quantities of isosmotic fluid reach the collecting duct.

The failure of additional solute to gain access to the medullary interstitium during the course of a mercurial diuresis tends to exclude the likelihood that mercurial agents exert their sole inhibitory effect within the proximal tubule. The inhibition of salt absorption at this site would be expected to

present at least as much solute to the ascending limb of the loop of Henle as mannitol and salt infusions at comparable levels of solute excretion. Progressive inhibition of salt reabsorption in the ascending limb of the loop of Henle during a mercurial diuresis might explain the failure of T^cH_2O to rise. However, the constancy of T^cH_2O in the experiments in which this parameter was elevated prior to the mercurial diuresis, noted here and by others (11) argues against this hypothesis. Furthermore, this explanation is not consistent with the increase in T^cH_2O produced by the administration of a nonspecific solute load during a mercurial diuresis (Tables III and IV, Figure 3). This finding suggests that when additional quantities of salt are presented to the loop of Henle during a mercurial diuresis, additional solute can be transported into the interstitial fluid. Moreover, progressive inhibition of salt reabsorption at this site conflicts with the observations in hydrated man that free water clearance remains relatively fixed as a mercurial diuresis develops (10). Although these data do not exclude some minor inhibition of salt reabsorption at the proximal segment or at the ascending limb of Henle's loop, they do weaken the possibility that the major site of action of organomercurials resides at either of these loci.

Another explanation for the failure of salt to be deposited in the interstitial fluid during a mercurial diuresis would presume that organomercurials enhance medullary blood flow and so limit the trapping characteristics of the medullary circulation. This hypothesis is not consistent with the observation that nontheophylline-containing mercurials tend to decrease total renal plasma flow. It seems unlikely that opposite qualitative changes in renal plasma flow and medullary flow would occur simultaneously. Pyrogenic reactions, which enhance renal plasma flow and presumably medullary blood flow, consistently reduce T^cH_2O (22, 23), an action different from that produced by mercurials.

A hypothesis compatible with the observations reported here would place the major inhibitory action of mercurials at a site distal to the loop of Henle. Such an action would explain the relative constancy of T^cH_2O during a mercurial diuresis, since the major increase in the flow of isosmotic fluid through the collecting duct would derive from a site beyond the loop of Henle. The effective osmotic gradient between interstitial and collecting duct fluid during any steady state is dependent upon the balance established between solute supply to the interstitium from the ascending limb and water supply from the collecting duct. For each level of solute supply there apparently exists a critical rate of collecting duct flow which will provide a quantity of water for back diffusion adequate to dissipate interstitial fluid tonicity. Without changes in loop solute supply, an increase in collecting duct flow up to this critical rate may produce a rise in T^cH_2O (Tables I and III, N.P., H.B., A.S., R.M.; Figure 1, N.P.) Beyond this critical rate, after medullary tonicity is largely dissipated, it appears that any increase in the flow of isosmotic collecting duct fluid will not in itself measurably augment the T^cH_2O value. This proposal is consistent with the increase in T^cH_2O produced by a superimposed nonspecific solute diuretic during a mercurial diuresis (Tables III and IV, Figure 3) or by the simultaneous administration of a nonspecific agent (13). Either technique would provide additional absorbable solute for loop transport into the interstitium during the mercurial diuresis so that more osmotically active particles would be available to enhance the back diffusion of water from the collecting tubule.

A distal site of mercurial action would also explain why a modest increment in C_{Osm} at the onset of the diuresis may be associated with a marked fall in urine osmolality (Table II, M.G.; Figure 4, R.R. and F.M.) In this view, a mercurial diuresis provides additional isotonic fluid to the collecting duct without appreciably increasing the quantity of solute presented to the loop of Henle. The extraction of a small additional quantity of solute-free water at the collecting duct at this time will rapidly attenuate medullary and, therefore, urinary concentration. This postmercurial diminution in medullary tonicity would be most conspicuous when control rates of loop solute flow were low or further reduced by the spontaneous reduction in filtration rate noted after the administration of nontheophylline-containing organomercurials.

This hypothesis can also explain the results obtained when filtration rate was reduced so that solute clearance fell comparably during a mercurial diuresis or a diuresis produced by a salt in-

fusion. Insofar as the diuresis produced by a salt infusion probably derives from a proximal source, it would be expected that loop solute supply would increase as the diuresis develops. When filtration rate is decreased, loop solute supply will return to the same levels which obtained at similarly low flow rates during the development of the diuresis. Therefore, urine osmolality will rise to levels commensurate with reduced levels of solute excretion. On the other hand, during the development of a mercurial diuresis, it has been hypothesized that loop solute supply is not augmented. When the filtered load is reduced during this state, loop solute supply would be expected to fall below those levels which obtained at similarly low flow rates during the development of the diuresis. This fall in loop solute supply might explain the failure of urine osmolality to rise.

The two-phase response produced by meralluride can best be explained by an early and transient increase in the quantity of salt presented to the ascending limb by the theophylline contained within meralluride, followed by a typical organomercurial diuresis. This dual effect would agree with previous observations in hydrated man where the theophylline contained in meralluride also produced an immediate but transient diuresis qualitatively different from that produced by the organomercurial itself (10).

The conclusions drawn from these experiments concerning the site of action of organomercurials in hydropenic man are consistent with previous reports from this laboratory which suggested that the organomercurials exert a major inhibitory influence on an isosmotic salt-absorptive process beyond the ascending limb of the loop of Henle (10).

SUMMARY

1. In the course of a nonspecific solute diuresis in hydropenic man T^cH_2O rises progressively as C_{Osm} increases. With hypertonic salt and aminophylline, T^cH_2O reaches a somewhat higher peak than with mannitol.

2. During an organomercurial diuresis T^cH_2O remains relatively fixed at whatever level prevailed prior to the development of the major solute diuresis.

3. The administration of a nonspecific solute diuretic during the organomercurial diuresis increases T^cH_2O.

4. When filtration rate is experimentally reduced in the course of a mercurial diuresis or of a comparable diuresis produced by a salt infusion, urine osmolality rises appreciably only in the salt-infused subjects.

5. These findings suggest a disparity in the supply of solute to the loop of Henle, at every level of solute excretion, between a mercurial diuresis and a comparable diuresis produced by a nonspecific solute diuretic. This disparity supports the hypothesis that the major action of organomercurials resides at a site distal to the loop of Henle.

ACKNOWLEDGMENT

The valuable technical assistance of Demetra Polimeros, Edith Neubert and Antonio Santana is gratefully acknowledged.

REFERENCES

1. Wesson, L. G., Jr., and Anslow, W. P., Jr. Effect of osmotic and mercurial diuresis on simultaneous water diuresis. Amer. J. Physiol. 1952, **170**, 255.
2. Brodsky, W. A., and Graubarth, H. N. Mechanism of mercurial diuresis in hydropenic dogs. Amer. J. Physiol. 1953, **172**, 67.
3. Welt, L. G., Goodyer, A. V. N., Darragh, J. H., Abell, W. A., and Meroney, W. H. Site of saluretic action of an organic mercurial compound. J. appl. Physiol. 1953, **6**, 134.
4. Grossman, J., Weston, R. E., Borun, E. R., and Leiter, L. Factors influencing the course of mercurial diuresis during Pitressin infusion in normal subjects. J. clin. Invest. 1955, **34**, 1611.
5. Capps, J. N., Wiggins, W. S., Axelrod, D. R., and Pitts, R. F. The effect of mercurial diuretics on the excretion of water. Circulation 1952, **6**, 82.
6. Duggan, J. J., and Pitts, R. F. Studies on diuretics. I. The site of action of mercurial diuretics. J. clin. Invest. 1950, **29**, 365.
7. Dale, R. A., and Sanderson, P. H. The mode of action of mercurial diuretics in man. J. clin. Invest. 1954, **33**, 1008.
8. Vander, A. J., Malvin, R. L., Wilde, W. S., and Sullivan, L. P. Localization of the site of action of mercurial diuretics by stop-flow analysis. Amer. J. Physiol. 1958, **195**, 588.
9. Kessler, R. H., Hierholzer, K., Gurd, R. S., and Pitts, R. F. Localization of diuretic action of chlormerodrin in the nephron of the dog. Amer. J. Physiol. 1958, **194**, 540.
10. Goldstein, M. H., Levitt, M. F., Hauser, A. D., and Polimeros, D. Effect of meralluride on solute and water excretion in hydrated man: Comments on site of action. J. clin. Invest. 1961, **40**, 731.
11. Au, W. Y. W., and Raisz, L. G. Studies on the renal concentrating mechanism. V. Effect of diuretic agents. J. clin. Invest. 1960, **39**, 1302.

12. Zak, G. A., Brun, C., and Smith, H. W. The mechanism of formation of osmotically concentrated urine during the antidiuretic state. J. clin. Invest. 1954, **33**, 1064.
13. Ladd, M. Renal excretion of sodium and water in man as affected by prehydration, saline infusion, Pitressin and Thiomerin. J. appl. Physiol. 1952, **4**, 602.
14. Gottschalk, C. W., and Mylle, M. Micropuncture study of the mammalian urinary concentrating mechanism: Evidence for the countercurrent hypothesis. Amer. J. Physiol. 1959, **196**, 927.
15. Wirz, H. The localization of antidiuretic action in the mammalian kidney *in* The Neurohypophysis, H. Heller, Ed., Proc. Eighth Symp. of the Colston Research Society. New York, Academic Press, 1957, p. 157.
16. Wirz, H. Der osmotische Druck in den corticalen Tubuli der Rattenniere. Helv. physiol. pharmacol. Acta 1956, **14**, 353.
17. Levitt, M. F., Levy, M. S., and Polimeros, D. The effect of a fall in filtration rate on solute and water excretion in hydropenic man. J. clin. Invest. 1959, **38**, 463.
18. Smith, H. W. The Kidney: Structure and Function in Health and Disease. New York, Oxford Univ. Press, 1951, p. 61.
19. Pitts, R. F., and Sartorius, O. W. Mechanism of action and therapeutic use of diuretics. J. Pharmacol. exp. Ther. 1950, **98**, 161.
20. Garby, L., and Linderholm, H. The permeability of frog skin to heavy water and to ions, with special reference to the effect of some diuretics. Acta physiol. scand. 1953, **28**, 336.
21. Pitts, R. F. Some reflections on mechanisms of action of diuretics. Amer. J. Med. 1958, **24**, 745.
22. Brandt, J. L., Zumoff, B., Castleman, L., Ruskin, H. D., Jones, A., and Zuckerman, S. Studies on the effects of large doses of bacterial pyrogen in the dog. I. The renal handling of salt and water. J. clin. Invest. 1956, **35**, 1080.
23. Eisner, G. M., Porush, J. G., Goldstein, M. H., and Levitt, M. F. An appraisal of free water reabsorption (T^cH_2O) in man. In preparation.