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THE EFFECT ON URINE OSMOLARITY OF A TRANSIENT REDUCTION IN GLOMERULAR FILTRATION RATE AND SOLUTE OUTPUT DURING A 'WATER' DIURESIS

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It has been suggested that the change in urine concentration which occurs during an osmotic diuresis (McCance, 1945; Rapoport, Brodsky, West & Machler, 1949) is due to an increased quantity of isosmotic fluid (approximately 300 m-osmole/l.) flowing past the site in the tubule where the final concentration and dilution of urine takes place, and that the urine is therefore approaching the concentration of plasma (Rapoport *et al.* 1949; Smith, 1951; West, Kaplan, Fomon & Rapoport, 1952). It has recently been shown, however, that in certain circumstances a large increase in solute output may be associated with a change in the concentration of the urine from hyper- to hypotonic (de Wardener & del Greco, 1955). The following experiments were made to determine whether the reverse can also occur, i.e. whether a considerable decrease in solute output might be associated with a change in urine concentration from hypo- to hypertonic.

METHODS

The experiments were performed on six dogs. The rate of solute excretion was lowered by means of a transient reduction in glomerular filtration rate by either (1) partial aortic occlusion produced by inflating a rubber balloon in the thoracic aorta (Thompson, Barrett & Pitts, 1951), or (2) renal vaso-constriction induced by the inhalation of cyclopropane (Miles & de Wardener, 1952).

All animals were anaesthetized with intravenous pentobarbitone, 20–40 mg/kg as a priming dose. Further intravenous supplements were given as required to maintain a suitable depth of anaesthesia (spontaneous respiration and absence of reflex muscular movements). The trachea was intubated, and a rubber catheter was placed in the bladder. The urine was collected at intervals of 1–10 min, depending upon the urine flow and the rate at which changes were occurring. Blood samples were collected at 10–20 min intervals. When spontaneous respiration failed on administration of cyclopropane, ventilation was maintained by intermittent pressure (10–15 times/min) on a rubber bag joined to the endotracheal tube by a CO_2 absorber.

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The mean abdominal aortic pressure was measured by a mercury manometer via a polythene tube introduced through the right femoral artery. Exogenous creatinine clearances were measured after a priming infusion of 1 g of creatinine in 20 ml. of saline, followed by a continuous maintenance infusion sufficient to give a concentration of about 5 mg/100 ml. plasma. Creatinine was estimated by the method of Bonsnes & Taussky (1945). The osmolarity of the urine and plasma were calculated from the freezing-point depression determined by a thermistor incorporated in a bridge circuit. This machine is linear down to a temperature of $-0{\cdot}93°$ C, corresponding to 500 m-osmole/l., and the standard deviation of a single reading is $\pm 0{\cdot}0102°$ C corresponding to $\pm 5{\cdot}5$ m-osmole/l. Duplicate estimations were made, and as each can be performed in 2–4 min the first estimation was determined during the course of the experiment, as soon as possible after the urine had been collected.

Experiments in which the aorta was partially occluded. Dogs Nos. 1–3. A flow of hypotonic urine was promoted by (i) the administration of 500–700 ml. of water by stomach tube half an hour before the induction of anaesthesia, and subsequently by (ii) the intravenous infusion of 0·5% NaCl at 5–7 ml./min; the total volumes of fluid given varied between 80 and 130 ml./kg. The infusion of 0·5% NaCl was discontinued when the aorta was occluded. For the sake of brevity, this flow of hypotonic urine has been called a 'water' diuresis.

A double-lumen balloon catheter was introduced into the descending thoracic aorta via the right carotid artery. The balloon was inflated with saline and the abdominal aortic pressure adjusted to be as low as was compatible with a urine flow of about 0·5 ml./min. The difficulties of this manoeuvre were considerable, for unless the inflation of the balloon was continuously and rapidly adjusted, the abdominal aortic pressure showed pronounced fluctuations. In these experiments blood samples were venous.

Experiments in which cyclopropane was administered. Dogs Nos. 4–6. A flow of hypotonic urine was promoted by an intravenous administration of 0·5% NaCl at 9–17 ml./min beginning soon after induction of anaesthesia with pentobarbitone; the total volume given varying between 80 and 130 ml./kg. In these experiments no water was given by stomach tube.

When the urine had become hypotonic the saline infusion was stopped and 20–50% (v/v) cyclopropane in oxygen was administered in closed circuit. After the first few breaths of cyclopropane, respirations ceased, and it became necessary to carry out controlled respiration (with intermittent positive pressure) until spontaneous respiration returned 5–15 min after the cyclopropane administration had ended. The concentration of cyclopropane was adjusted so that the blood pressure should remain approximately unchanged while the urine flow gradually decreased to about 0·5 ml./min; if the anaesthetic depth was increased too rapidly there was a fall in blood pressure.

On stopping the cyclopropane controlled respiration was continued with 100% oxygen. The return of the glomerular filtrations rate and solute output to control values was liable to take a few minutes longer than in the aortic occlusion experiments. In order to accelerate the recovery of the solute output, 25% mannitol was given to dogs 4 and 5 at the rate of 1·5 ml./min for 7–18 min, via a polythene tube which had been introduced into the descending thoracic aorta through the left femoral artery. In these experiments blood samples were taken from the abdominal aorta via another polythene tube introduced through the left femoral artery.

RESULTS

In both sets of experiments the following three urine collection periods are compared: (i) control, (ii) experimental, (iii) recovery. The control period refers to one or more urine samples collected immediately before the aorta was occluded, or cyclopropane given. The experimental period refers to the urine of highest osmolarity collected during occlusion of the aorta or inhalation of cyclopropane. The recovery period refers to the first hypotonic urine collected

after deflation of the aortic balloon or cessation of cyclopropane administration. The estimations of creatinine clearance and rate of solute excretion for the experimental and recovery periods can only be rough approximations, for these periods were of short duration (1–7 min), the rate of urine flow was changing, and during the experimental period was 0·2–0·7 ml./min. The times referred to below, and in Tables 1 and 2, are from the beginning or end of aortic occlusion, or cyclopropane administration, to the mid-point of the urine collection period mentioned.

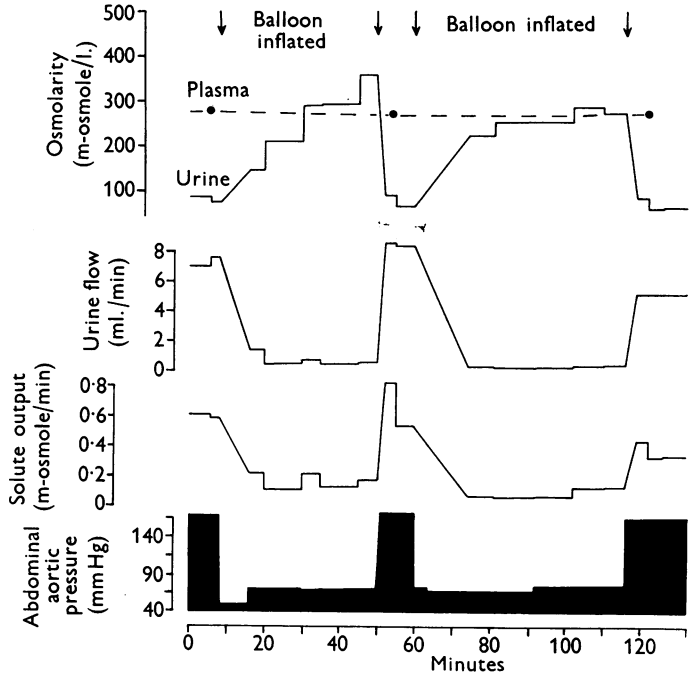

Fig. 1. Dog no. 2. The effect of transient occlusion of the thoracic aorta upon urine flow, urine osmolarity, solute output and mean abdominal aortic pressure during a water diuresis. The aorta was occluded on two occasions by inflation of a rubber balloon.

The effect upon hypotonic urine of occluding the aorta

The values obtained in the control, experimental and recovery periods in four experiments performed on three dogs are given in Table 1. The two experiments performed on dog 2 are illustrated in Fig. 1. On inflating the balloon in the aorta the urine became more concentrated than the plasma, and there was a conspicuous decrease in urine flow, solute output, creatinine clearance and abdominal aortic pressure. The mean urine osmolarity rose in 29·3 (12·5–45) min from 110 (63–197) to 346 (426–288) m-osmole/l., while the mean plasma osmolarity remained unchanged at 272 (256–286) m-osmole/l. On deflating the balloon there was a rapid return toward control values, the mean urine

TABLE 1. The effect upon urine flow, urine osmolarity, solute output, creatinine clearance and plasma osmolarity of partial aortic occlusion. The headings 'control', 'experimental' and 'recovery' are explained in the text

Dog no.	Wt. (kg)	Fluid load (ml./kg)	Interval after balloon inflated (min)	Interval after balloon deflated (min)	Duration of period (min)	Urine flow (ml./min)	Urine osmolarity (m-osmole/l.)	Plasma osmolarity (m-osmole/l.)	Solute output (m-osmole/min)	Creatinine clearance (ml./min)	Abdominal aortic pressure (mm Hg)
						Control					
1	12	129	—	—	10	7·4	197	258	1·46	58	145
2	13	103	—	—	2	7·6	77	274	0·58	94	170
2	—	—	—	—	5	8·4	63	272	0·53	51	175
3	14	82·5	—	—	6	6·0	102	288	0·61	82	165
	Average	105	—	—	5·75	7·3	110	272	0·80	71	164
						Experimental					
1	—	—	12·5	—	5	0·7	344	256	0·24	6	50
2	—	—	42·0	—	5	0·5	327	273	0·17	49	70
2	—	—	45·0	—	7	0·4	288	273	0·12	31	75
3	—	—	17·5	—	7	0·6	426	286	0·25	44	52
	Average	—	29·3	—	6	0·5	346	272	0·19	32·5	62
						Recovery					
1	—	—	—	4·0	4	9·0	171	253	1·76	70	155
2	—	—	—	3·5	3	8·7	92	272	0·80	93	175
2	—	—	—	4·5	3	5·3	84	273	0·44	81	165
3	—	—	—	1·5	3	3·9	254	285	0·99	197	162
	Average	—	—	3·4	3·25	6·7	150	270	1·0	110	164

osmolarity falling in 3·4 (4·5–1·5) min to 150 (254–84) m-osmole/l. in the recovery period. In all experiments the urine osmolarity continued to decrease for 5–10 min after the end of the recovery period.

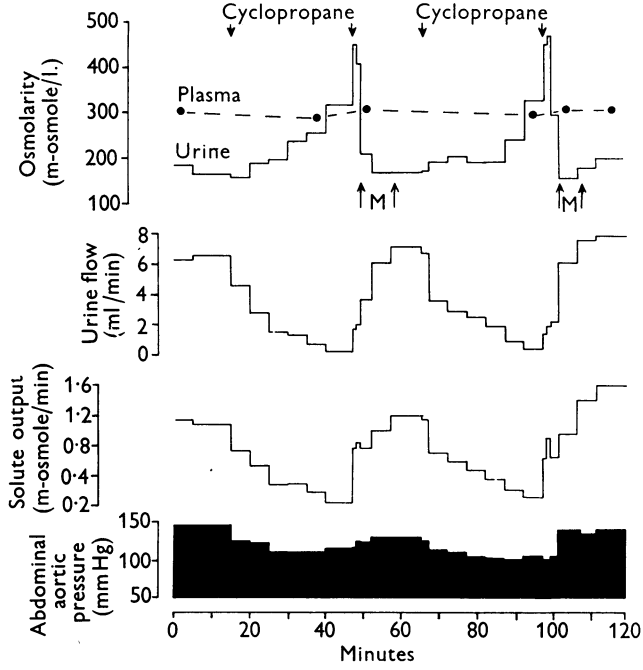

Fig. 2. Dog no. 5. The effect of transient renal vasoconstriction produced by inhalation of 20–50% cyclopropane upon urine osmolarity, urine flow, solute output and mean abdominal aortic pressure during a water diuresis. The experiment was performed twice. (M) represents the administration of 25% mannitol at 1·5 ml./min via a polythene catheter in the thoracic aorta.

The effect upon hypotonic urine of the inhalation of cyclopropane

The values obtained in the control, experimental and recovery periods in four experiments performed on three dogs are given in Table 2. The two experiments performed on dog 5 are illustrated in Fig. 2. After the administration of cyclopropane the urine became more concentrated than the plasma, there was a pronounced decrease in urine flow, solute output and creatinine clearance, and a minor fall in abdominal aortic pressure. The mean urine osmolarity rose in 29·8 (31·5–28·5) min from 193 (255–165) m-osmole/l. in the control period to 373 (500–316) m-osmole/l. in the experimental period, when the mean plasma osmolarity was 295 (284–309) m-osmole/l. The results obtained in the experimental period refer only to urine *collected* during the administration of cyclopropane (see methods); owing to the effects of renal dead space and delay time, however, in three of the four experiments the urine osmolarity continued to

TABLE 2. The effect upon urine flow, urine osmolarity, solute output, creatinine clearance and plasma osmolarity, of renal vasoconstriction produced by the inhalation of 20–50% cyclopropane. The headings 'control', 'experimental' and 'recovery' are explained in the text.

Dog no.	Wt. (kg)	Fluid load (ml./kg)	Cyclopropane inhalation interval: From onset (min)	Cyclopropane inhalation interval: After termination (min)	Duration of period (min)	Urine flow (ml./min)	Urine osmolarity (m-osmole/l.)	Plasma osmolarity (m-osmole/l.)	Solute output (m-osmole/min)	Creatinine clearance (ml./min)	Abdominal aortic pressure (mm Hg)
					Control						
4	16	79	—	—	10	5·6	255	307	1·40	70	158
5	22	132	—	—	6	6·6	165	299	1·09	101	145
5	—	—	—	—	8	7·1	168	303	1·19	84	130
6	10	107	—	—	2	1·5	184	293	0·27	48	175
	Average	106	—	—	6·5	6·7	193	300	0·99	76	152
					Experimental						
4	—	—	31·5	—	5	0·4	500	309	0·23	34	150
5	—	—	31·0	—	7	0·2	316	288	0·04	19	115
5	—	—	28·5	—	5	0·4	328	298	0·12	22	105
6	—	—	28·5	—	5	0·2	344	284	0·09	15	150
	Average	—	29·8	—	5·5	0·3	372	295	0·12	25	130
					Recovery						
4	—	—	—	8·5	7	5·7	286	317	1·63	37	153
5	—	—	—	3·5	3	3·7	209	304	0·77	134	123
5	—	—	—	6·5	2	6·1	157	306	0·95	123	140
6	—	—	—	9·0	4	2·6	237	280	0·62	41	155
	Average	—	—	6·9	4	4·7	222	302	0·99	84	143

rise steeply for 1–3 min after the cyclopropane ceased (Fig. 2). It appears, therefore, that often the concentration of the urine *formed* during the experimental period was greater than that which was collected.

On ceasing the administration of cyclopropane there was a return towards control values, the mean urine osmolarity falling in 6·9 (9·0–3·5) min to 222 (286–157) m-osmole/l. in the recovery period. In three of the four experiments the urine osmolarity continued to decrease for 5–10 min after the end of the recovery period.

DISCUSSION

The results show that when these animals were passing hypotonic urine, partial occlusion of the aorta or inhalation of cyclopropane for 12·5–45 min produced a rise in concentration of the urine above that of the plasma. Subsequently, when the aortic occlusion or the cyclopropane inhalation was discontinued the urine again became hypotonic within 1·5–9·5 min.

Leaf, Kerr, Wrong & Chatillon (1954) have recently reported that they were unable to obtain any increase in urine osmolarity by compressing the renal artery of the unanaesthetized dog during a water diuresis. A comparison of their data with those reported in this paper, however, suggests that the difference between the two is due to the renal artery not having been sufficiently occluded.

It is considered that the changes in urine concentration which occurred in the experiments reported here were not due to changes in the concentration of circulating antidiuretic hormone for the following reasons: (1) the rapidity with which the urine osmolarity changed from hyper- to hypotonic after the balloon was deflated, or the cyclopropane anaesthesia discontinued; and (2) the demonstration that, following the administration of large quantities of intravenous fluids such as were given in these experiments, the kidney became relatively resistant to the effect of intravenous vasopressin ('Pitressin', Parke, Davis & Co.) or nicotine. This form of Pitressin resistance was first described by Wesson, Anslow, Raisz, Bolomey & Ladd (1950), who expanded the extracellular fluid volume with smaller quantities of fluids than those used in the experiments described in this paper. The mechanism of this teleologically reasonable phenomenon is not known, but its presence in the experiments reported here was confirmed on three occasions by the intravenous injection of 100 mU of Pitressin. Fig. 3 illustrates the results obtained in one of two further experiments designed solely to show this effect.

There is a further possibility that the changes in urine concentration may have been due to renal anoxia, but the hypertonic values reached (in some instances 200 m-osmole/l. above those of plasma), and the speed of recovery, make this unlikely. Nor does the fall in arterial pressure seem to be the determining factor, for changes in urine concentration were as great in the cyclo-

propane experiments (when the renal perfusion pressure changed relatively little) as when the aorta was occluded.

It is suggested that the change in urine concentration from hypo- to hypertonic was due to the decrease in glomerular filtration rate, and related to the consequent reduction in solute output. It has been shown on several occasions (Selkurt, 1951; de Wardener & Miles, 1952) that a fall in renal perfusion pressure below 90–100 mm Hg in the dog, or the inhalation of cyclopropane (Habif, Papper, Fitzpatrick, Lowrance, Smythe & Bradley, 1951; Miles & de Wardener, 1952), reduces renal blood flow, glomerular filtration

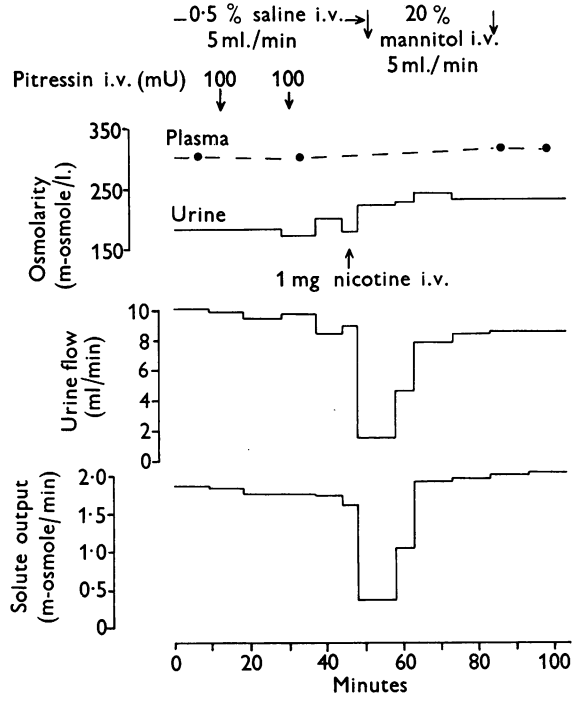

Fig. 3. Dog no 7. The effect upon urine osmolarity, urine flow and solute output of two intravenous injections of 100 mU Pitressin and 1 mg nicotine acid tartrate during a water diuresis induced by the administration of 0·5 % saline (90 ml./kg). The saline infusion started 110 min before the first injection of Pitressin. After the nicotine injection a mannitol infusion was given to increase the speed of recovery of the solute output to control levels.

and solute excretion rate; the decrease with cyclopropane being proportional to the depth of anaesthesia (Miles, de Wardener, Churchill-Davidson & Wylie, 1952). It is probable that, owing to delay time and dead space factors during the experimental period, the estimated changes in glomerular filtration rate and solute output in the observations reported here are inaccurate, but there can be little doubt that they were both considerably decreased.

It has been demonstrated in man that in certain circumstances a sudden *increase* in solute output may be associated with a change in urine concentration from hyper- to hypotonic (de Wardener & del Greco, 1955). These observations were made in subjects suffering from diabetes insipidus in whom the urine concentration was gradually increased by a continuous infusion of Pitressin. When the initially hypotonic urine had become slightly hypertonic, a rapid infusion of mannitol produced an increase in solute output and urine flow, and a decrease in urine osmolarity to a value below that of plasma, though the infusion of Pitressin was continued throughout. In these experiments, and in those described in this paper, changes in urine concentration from hyper- to hypotonic or vice versa were related to variations in the solute excretion rate. It seems probable, therefore, that they were due to modifications in tubular function caused by the changing rate at which solutes were being presented to the tubule. The nature of these modifications is not known, but it is reasonable to suppose that they are similar both in the human experiments and in those performed on the dog.

These results are not consistent with either of the two most widely discussed contemporary hypotheses concerning the concentration and dilution of urine (Smith, 1951; West *et al.* 1952), for neither concept allows that changes in the rate of solute output may be related to alterations in urine osmolarity from hyper- to hypotonic or vice versa. It has been suggested therefore (de Wardener & del Greco, 1955) that the change in urine osmolarity observed in the human experiments is due either to (1) an increase in the rate of flow of a *hypotonic* tubular fluid into the site in the tubule where concentration takes place, so that the urine approaches the concentration of this fluid (Shannon, 1942), or (2) that an increase in the rate of flow of an *isotonic* fluid into the concentration site reverses the proportions of water and solutes reabsorbed (Mudge, Foulks & Gilman, 1949). Either of these mechanisms, working in the opposite direction, could be responsible for the increase in urine osmolarity which occurred in the dog experiments.

SUMMARY

1. The effect of partial occlusion of the aorta, or inhalation of cyclopropane, on urine osmolarity, urine flow, solute excretion rate and creatinine clearance has been studied in the anaesthetized dog during a 'water' diuresis.

2. The urine became hypertonic within 12·5–45 min, and there was a decrease in urine flow, solute output and creatinine clearance.

3. Release of the aortic occlusion, or discontinuation of the cyclopropane was followed by a rapid return to control values, the urine becoming hypotonic within 1·5–9·5 min.

4. The evidence suggests that the changes in urine concentration were related to changes in solute output consequent upon a reduction of glomerular

filtration rate rather than to changes in the concentration of circulating antidiuretic hormone or renal anoxia.

5. It is concluded that in certain circumstances changes in the rate at which solutes are presented to the tubule may determine whether the urine is hyper- or hypotonic.

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