OBSERVATIONS ON THE METABOLIC EFFECTS OF THE CARBONIC ANHYDRASE INHIBITOR DIAMOX®: MODE AND RATE OF RECOVERY FROM THE DRUG'S ACTION

By T. HANLEY and M. M. PLATTS

(*From the Department of Medicine, University of Sheffield, Sheffield, England*)

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Ingestion of certain unsubstituted sulphonamides causes an increased excretion of sodium, potassium, and bicarbonate ions. The effects are probably due to reduction of the quantity of hydrogen ions available for secretion by the renal tubules, with consequent impairment of the $Na^+ - H^+$ exchange postulated by Pitts and Alexander as being responsible for the tubular reabsorption of bicarbonate (1).

Schwartz (2) suggested that the natriuretic effect of carbonic anhydrase inhibitors might prove to be of therapeutic value in patients with sodium retention and congestive heart failure, and for this purpose Miller, Dessert, and Roblin tested the inhibitory potency of a number of heterocyclic sulphonamides *in vitro* (3). Among the most powerful of their compounds was Diamox®, 2-acetylamino-1, 3, 4-thiadiazole-5-sulphonamide, the substance used in the present investigation.

The effects of Diamox® on the urinary excretion of water and electrolytes have been described by Nadell (4), and by Friedberg, Taymor, Minor, and Halpern (5) who noted that the drug's action is transient. The present study was undertaken primarily to determine the mechanism and rate of recovery from the disturbance of acid-base balance produced by the drug.

MATERIAL AND METHODS

Two normal adults, referred to as Subjects 'A' (male, 34 years, wt. 72 kg.) and 'B' (female, 31 years, wt. 61 kg.) maintained a fixed intake of food and fluid for 13 and 16 days, respectively. After control periods of six and seven days, Diamox® was ingested for four and three days in a dosage of 0.25 g. 6-hourly. Measurements were continued for recovery periods of three and six days, respectively. All urine was collected under oil in two 12-hour periods for each day, and a complete analysis was made of its acid-base composition. In addition more limited urinary studies were carried out on four normal adults who ingested Diamox® at daily intervals for 3 to 5 days. Blood samples were taken at approximately 10.00, 16.00, and 22.00 hours each day and all urine collected under oil over corresponding intervals. Observations were also made on eight patients with heart failure (six cor pulmonale, two mitral stenosis), seven of whom ingested 0.25 g. of Diamox® 6-hourly, the eighth 0.2 g. 4-hourly. Urine was collected over 4 to 6-hour periods until bicarbonate excretion ceased.

Blood samples were taken from an arm vein in which the blood had been rendered arterial in character by immersion of the limb in water at 118°F for ten minutes. Plasma for carbon dioxide analysis was obtained by anerobic centrifugation of the blood in the syringes employed for collection. Analytic methods used were as follows:

Sodium, Potassium	Flame Photometer
Ammonia	Aeration and titration (6)
Titratable acid	Electrometric titration to pH. 7.4
Magnesium	Titan yellow colorimetric (7)
Chloride (urine and plasma)	Electrometric titration (8)
Calcium	Oxalate precipitation, permanganate titration (9)
Phosphate	Molybdate colorimetric (10)
Sulphate	Benzidine titrimetric (11)
Organic acids	Titration (12)
Total carbon dioxide (urine and plasma)	Manometric (13)
pH (urine and whole blood)	Glass electrode, electrometric pH meter Marconi type T511D

The blood pH was corrected to 38°C, employing a correction factor of 0.014 units per degree C. No temperature correction was applied to urine pH.

The bicarbonate concentrations of plasma and urine were derived by insertion of the measured values of pH and total CO_2 in the Henderson-Hasselbalch equation.

Calculation of electrolyte "balance." It was assumed that a steady state of body electrolytes had been obtained when the daily excretion of electrolyte became constant (within the limits of inherent cyclical variation), while the intake remained unvaried. Changes in the total body content of electrolyte during the ingestion of Diamox®, or during recovery from its effects, were calculated as the difference between the observed rate of urinary electrolyte excretion for that day and the control rate.

The term "conservation" applied to electrolytes is used to mean retention of electrolyte with reduction of an existing negative balance. Urinary "hydrogen ion" refers to the sum of urinary ammonium ion and titratable acid, in accordance with current theories of hydrogen ion excretion (14–18).

RESULTS

The changes in urine flow and acid-base composition of the urine for the whole period of observation are shown in Tables I to IV.

I. *Balance Experiments in Normal Subjects*

Changes in electrolyte excretion and urine flow during ingestion of Diamox®

On the first day of ingestion of the drug both subjects showed an increase in flow of urine of approximately 1.2 liters; smaller increments of urine flow also occurred on the next two days. There was a corresponding weight loss, which finally amounted to 2.3 kg. and 1.7 kg., respectively.

Total daily electrolyte excretion rose from 400 to 550 m.Eq. in 'A' and from 310 to 500 m.Eq. in 'B' on the first day; on each day thereafter total electrolyte excretion was not materially different from the control period. The sequence of changes in the excretion of HCO_3^-, H^+, and of K^+, was almost identical in the two subjects. Excretion of bicarbonate was greatly augmented on the first day, but dwindled rapidly and attained normal values by the third day; the cumulative totals of bicarbonate excretion were 163 and 132 m.Eq. Excretion of H^+ was reduced to very low levels on the first day and was almost normal by the third. The cumulative buffer decrement (excess

TABLE I

Acid-base composition of the urine and daily electrolyte excretion during ingestion of Diamox® and during recovery from the drugs effects (Subject 'A')

	Day	pH	Na^+ m.Eq.	K^+ m.Eq.	Mg^{++} m.Eq.	Ca^{++} m.Eq.	NH_4^+ m.Eq.	Titr. Acid. m.Eq.	Total "Base" m.Eq.	Cl^- m.Eq.	HPO_4^- m.Eq.	SO_4^{--} m.Eq.	Org. Acid. m.Eq.	HCO_3^- m.Eq.	Total "Acid" m.Eq.	Body Wt. Kg.
CONTROL(mean)	1-6	-	180	108	8	17	40	45	398	171	91	74	54	4	394	72.48
Diamox 0.25 g 6-hrly.	7	7.31, 7.02	258	245	12	18	7	12	552	172	105	79	43	133	532	71.27
	8	6.58, 6.50	165	140	10	17	36	40	408	142	109	69	59	19	398	70.77
	9	6.21, 6.52	144	113	7	16	35	33	348	133	98	64	47	9	351	70.14
	10	6.21, 5.97	119	119	9	20	38	34	339	126	97	67	48	2	340	70.17
Recovery	11	5.01, 4.99	131	72	8	21	61	64	357	155	83	71	50	2	361	70.69
	12	4.99, 5.32	142	44	12	20	114	63	395	185	95	61	48	2	391	71.89
	13	5.15, 5.32	206	50	9	22	96	90	473	222	120	80	61	2	485	72.12

Urine pH values are given for 2 12-hour periods of each day

TABLE II

Daily changes in urinary acid-base excretion during ingestion of Diamox® and during recovery from the drug's effects, Subject 'A'

(Computed from Table I)

DAY	Wt. (Kg)	Water Balance (l) Daily	Water Balance (l) Cumulative	Na^+ m. Eq. Daily	Na^+ m. Eq. Cumulative	K^+ m. Eq. Daily	K^+ m. Eq. Cumulative	NH^+_4 plus Titr. Acid m. Eq. Daily	NH^+_4 plus Titr. Acid m. Eq. Cumulative	Cl^- m. Eq. Daily	Cl^- m. Eq. Cumulative	HCO^-_3 m. Eq. Daily	HCO^-_3 m. Eq. Cumulative	"Other Anions" m. Eq. Cumulative
7	-1.21	+1.22	+1.22	+78	+78	+137	+137	-66	-66	+1	+1	+129	+129	
8	-1.71	+0.60	+1.82	-15	+63	+32	+169	-9	-75	-29	-28	+15	+144	
9	-2.34	+0.39	+2.21	-36	+27	+5	+174	-17	-92	-38	-66	+5	+149	
10	-2.31	+0.06	+2.27	-61	-34	+11	+185	-13	-105	-45	-111	-2	+147	+9
11	-1.88	-0.70	+1.57	-49	-83	-36	+149	+40	-65	-16	-127	-2	+145	
12	-1.39	-0.46	+1.11	-38	-121	-64	+85	+92	+27	+14	-113	-2	+143	
13	-0.36	-0.47	+0.64	+26	-85	-58	+27	+101	+128	+51	-62	-2	+141	+21

+ = Increase
- = Decrease
} of urinary excretion as compared with control value.

HCO_3^- excretion plus reduction of H^+ excretion) was 252 and 234 m.Eq. in 'A' and 'B', respectively, when the drug was withdrawn. In both persons a considerable negative balance of K^+ occurred, amounting finally to 185 and 153 m.Eq.

There were considerable differences in the effect of the drug on sodium excretion in the two individuals. Subject 'A' (Tables I and II) showed a negative balance of sodium only on the first day; thereafter there was a small positive balance for each individual day, and the cumulative balance ultimately became positive. The sodium balance in this subject appeared to be largely dependent on changes in chloride excretion (Figure 1), whereas potassium balance seemed independent of chloride. In subject 'B' there was a large negative balance of sodium on the first day, and a small negative balance on each subsequent individual day, with a final deficit of 196 m.Eq. No material change in chloride excretion occurred in this subject, but a large increment of excretion of "other anions" (phosphate, sulphate and organic acids considered jointly) occurred.

Electrolyte excretion during recovery from the effects of Diamox®

The relevant data are contained in Tables I–IV.

During the recovery periods the urine became strongly but not maximally acid. The excretion of ammonium ion and titratable acid was greatly augmented in 'A', in whom the average daily increment of H^+ excretion was 78 m.Eq. Subject 'B' showed an average H^+ increment of only 45 m.Eq. however, and the urine pH tended to be higher than in 'A'. It may be seen from Tables II and IV that the final deficit of fixed cation (Na^+ plus K^+) was 349 m.Eq. in 'B' and only 151 m.Eq. in 'A'. This fact, coupled with the lower increment of H^+ during recovery in 'B', determined the relative duration of the recovery periods; after six days subject 'B' had repaired the cation deficit by only 55 per cent, whereas three days sufficed for almost complete recovery of K^+ balance in 'A', the net (Na^+ plus K^+) balance at this time being slightly positive in this subject owing to sodium retention.

The different patterns of disturbance of anion excretion observed during the drug period was also evident during the initial stages of recovery.

TABLE III

Acid-base composition of the urine and daily electrolyte excretion during ingestion of Diamox® and during recovery from the drug's effects (Subject 'B')

	Day	pH	Na^+ m.Eq.	K^+ m.Eq.	Mg^{++} m.Eq.	Ca^{++} m.Eq.	NH_4^+ m.Eq.	Titr. Acid. m.Eq.	Total "Base" m.Eq.	Cl^- m.Eq.	HPO_4^- m.Eq.	SO_4^{--} m.Eq.	Org. Acid. m.Eq.	HCO_3^- m.Eq.	Total "Acid" m.Eq.	Body Wt. Kg.
CONTROL(mean)	1-7	-	143	53	7	18	43	45	309	150	73	53	32	2	310	61.24
Diamox 0.25 g 6-hrly.	8	7.41, 7.29	303	152	5	18	14	5	497	168	95	54	66	117	500	59.95
	9	6.71, 5.55	155	84	8	18	25	36	326	139	63	62	42	14	320	59.55
	10	5.78, -	167	76	7	18	35	41	344	142	94	80	27	1	344	59.55
Recovery	11	6.21, 5.15	113	65	8	18	63	40	307	145	84	59	32	2	322	59.70
	12	5.00, 5.25	74	40	7	18	103	52	294	131	98	51	30	2	312	60.30
	13	5.22, 5.41	103	27	5	18	98	50	301	137	82	55	27	2	303	60.55
	14	6.33, 5.72	151	25	5	18	90	50	339	174	75	53	34	2	338	60.65
	15	5.43, 5.65	153	26	7	18	85	45	334	163	73	34	45	2	317	60.55
	16	5.51, 5.41	175	33	5	18	73	47	351	177	70	50	39	2	338	-

Urine pH values are given for 2 12-hour periods of each day

TABLE IV

Daily changes in urinary acid-base excretion during ingestion of Diamox® and during recovery from the drug's effects, Subject 'B'

(Computed from Table III)

DAY	Na^+ m. Eq.		K^+ m. Eq.		NH^+_4 plus Titr. Acid m. Eq.		Cl^- m. Eq.		HCO^-_3 m. Eq.		"Other Anions" m. Eq.
	Daily	Cumulative	Daily	Cumulative	Daily	Cumulative	Daily	Cumulative	Daily	Cumulative	Cumulative
8	+160	+160	+99	+99	-69	-69	+18	+18	+115	+118	
9	+12	+172	+31	+130	-27	-96	-11	+7	+12	+127	
10	+24	+196	+23	+153	-12	-108	-8	-1	-1	+126	+109
11	-30	+166	+12	+165	+15	-93	-5	-6	-		
12	-69	+97	-13	+152	+67	-26	-19	-25	-		
13	-40	+57	-26	+126	+60	+34	-13	-38	-		
14	+8	+65	-28	+98	+52	+86	+24	-14	-		
15	+10	+75	-27	+71	+42	+128	+13	-1	-		
16	+32	+107	-20	+51	+32	+160	+27	+26	-	+126	+152

+ = Increase
- = Decrease
} in urinary excretion as compared with control value

There was a further extension of "other anion" losses in 'B' but none in 'A', while the chloride retention observed in 'A' during the drug's action was temporarily increased during recovery (Tables II and IV).

The electrolyte balances at the end of the drug period are compared with those prevailing at the end of the recovery study, in Figure 2.

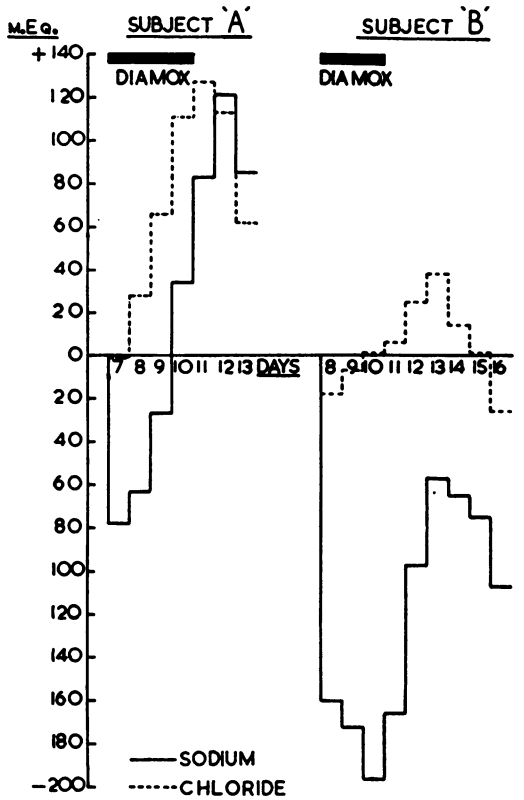

FIG. 1. RELATION OF SODIUM AND CHLORIDE BALANCES DURING ACTION OF DIAMOX® AND DURING RECOVERY OF SUBJECTS 'A' AND 'B'

II. *The Effects of Single Daily Doses of Diamox®*

The changes in urinary excretion of HCO_3^- and H^+ which resulted from ingestion by a normal person of four doses of Diamox® at intervals of one day, are shown in Figure 3. Details of observations in this person and three other normal subjects are shown in Table V.

The initial dose of Diamox® caused an excretion of bicarbonate which ranged from 78 to 97 m.Eq. in the first six hours; during this period the urine pH ranged from 7.50 to 7.70, ammonia excretion was negligible and titratable alkali replaced titratable acid. In the succeeding six hours the urine pH was much reduced, and bicarbonate excretion was less than half that of the first six-

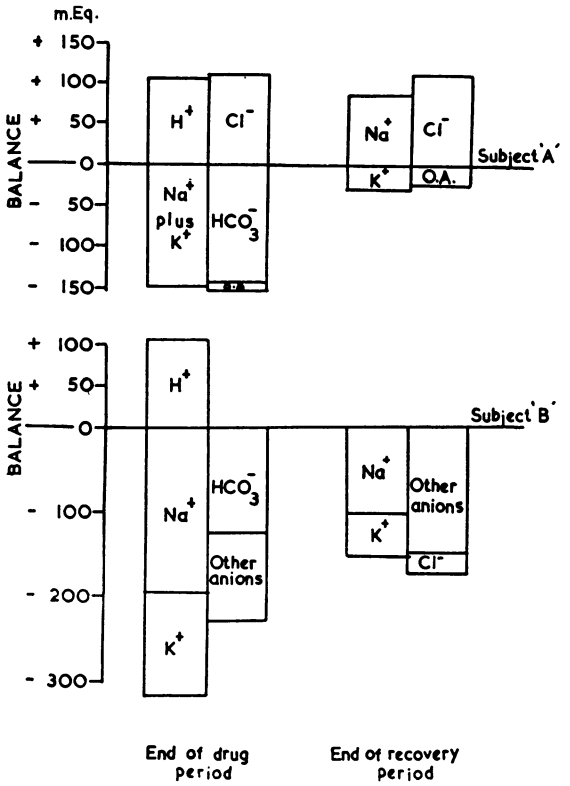

FIG. 2. CHANGES IN BALANCE OF ELECTROLYTES OCCURRING AS A RESULT OF INGESTION OF DIAMOX®, AND DURING RECOVERY FROM THE DRUG'S EFFECTS

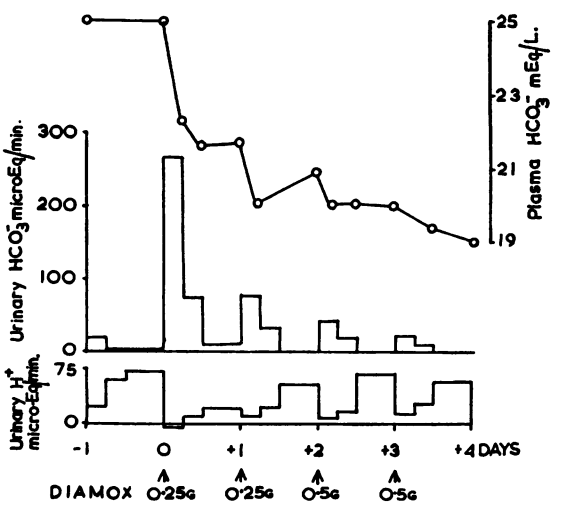

FIG. 3. CHANGES IN PLASMA BICARBONATE AND URINARY EXCRETION OF H^+ DURING ACTION OF SINGLE DAILY DOSES OF DIAMOX®

TABLE V

Effects of single daily doses of Diamox® on urine and plasma of four normal persons

Subject	Urine: Interval (days)	Urine: HCO_3^- m. Eq.	Urine: NH_4^+ m. Eq.	Urine: Titr. acid m. Eq.	Plasma (arterialized): Time (days)	pH	HCO_3^- m. Eq./L.	pCO_2 mm. Hg	Doses of Diamox
	(control) −1–0	6	41	41	0	7.40	25.0	42	Control
1. (75 kg.)	0–1	129	11	5	1	7.35	21.8	41	Diamox 0.25 g. at 0, 24 hrs. 0.5 g. at 48, 72 hrs.
	1–2	41	26	25	2	7.36	21.0	38	
	2–3	22	22	36	3	7.38	20.0	35	
	3–4	11	27	28	4	7.37	19.0	34	
	4–5	—	84	62	5	7.40	20.8	35	
	5–6	—	87	49	6	7.41	22.0	36	Recovery
	6–7	—	91	43	7	7.43	23.6	37	
	−1–0	3	41	45	0	7.40	26.3	44	Control
2. (72 kg.)	0–1	120	9	8	1	7.35	21.5	40	Diamox 0.4 g. at 0, 24, 48 hrs. 0.8 g. at 72, 96 hrs.
	1–2	22	25	27	2	7.36	20.1	37	
	2–3	49	17	13	3	7.39	19.0	33	
	3–4	11	31	33	4	7.38	18.3	32	
	4–5	21	30	32	5	7.38	18.9	33	
	5–6	—	73	56	6	7.42	20.3	32	
	6–7	—	64	50	7	7.43	21.9	34	Recovery
	7–8	—	88	51	8	7.43	23.4	36	
	−1–0	7	24	28	0	7.41	21.8	36	Control
3. (61 kg.)	0–1	97	14	11	1	7.37	18.4	33	Diamox 0.25 g. at 0, 24, 48 hrs. 0.5 g. at 72, 96 hrs.
	1–2	24	24	25	2	7.32	16.9	34	
	2–3	25	29	22	3	7.37	16.4	29	
	3–4	25	20	19	4	7.34	16.6	32	
	4–5	13	18	28	5	7.39	17.6	30	
	5–6	—	70	45	6	7.38	20.3	36	Recovery
	6–7	—	70	41	7	7.39	20.8	36	
	−1–0	3	39	39	0	7.39	24.0	41	Control
4. (66 kg.)	0–1	138	20	6	1	7.38	19.0	33	Diamox 0.25 g. at 0, 24, 48 hrs.
	1–2	25	18	10	2	7.40	18.8	31	
	2–3	17	30	10	3	7.38	18.1	32	

hour period. In the last 12 hours of that day, the urine was acid, bicarbonate excretion was negligible, and H^+ excretion rose to an almost normal value for that period of the diurnal cycle.

Each succeeding dose of Diamox® caused a similar daily sequence of qualitative changes in the urine, but the magnitude of the effects rapidly diminished, even though we doubled the dose of Diamox® in two subjects on the third day. Excretion of bicarbonate on the final day (see Table V) ranged from 11 to 21 m.Eq. as compared with 97 to 138 m.Eq. on the first day. The excretion of bicarbonate on the 4th or 5th day was still significantly higher than that of the control day, however, in contrast with the subjects who ingested Diamox® at 6-hour intervals.

The diminishing effect of single daily doses of Diamox® on urinary bicarbonate excretion appeared to be related in a reasonably constant manner in the group of four subjects to concomitant falls of the plasma bicarbonate concentration (Table V). The initial dose of Diamox® caused a fall of plasma bicarbonate concentration which ranged from 3.2 to 5.0 m.Eq. per L. During the second 12-hour ("drug-free") period the plasma bicarbonate rose only very slightly or not at all (Figure 3), and in consequence the next dose was given while the plasma bicarbonate was still depressed. A similar but less pronounced sequence occurred with succeeding doses.

The total H^+ excretion for each drug day was always less than that of the control period, al-

though the deviations grew less each day. The cumulative decrement of H^+ excretion ranged from 50 to 205 m.Eq. while the total buffer decrements varied from 199 to 413 m.Eq., with a mean value of 313 m.Eq.

During recovery from the drug's action the urine became strongly acid, and NH_4^+ excretion was augmented. Subjects 1, 2, and 3 of Table V showed a mean daily increment of 57, 41, and 61 m.Eq. of H^+, respectively. In none of the three had the plasma bicarbonate concentration returned to normal within three days. Interpretation of changes in plasma bicarbonate as an index of recovery from the effects of Diamox® acidosis are rendered difficult by concomitant changes in pCO_2, however.

The data of Table V indicate the extent of disturbance of plasma acid-base balance caused by single daily doses of the drug. The arterial pH fell on the first day by 0.04 or 0.05 units in three of the four subjects; no compensatory fall of arterial pCO_2 occurred during this time, but by the fourth day a reduction of pCO_2 ranging from 6 to 11 mm. Hg had occurred, and had greatly minimized the potential rise of plasma hydrogen concentration.

III. *Effects of Diamox® in Patients with Heart Failure*

The curves of cumulative bicarbonate excretion against time which we observed in eight patients with heart failure, are shown in Figure 4. The curves were of roughly exponential form, and approached a limiting value within the range 150 to 250 m.Eq. The data indicate that the magnitude of the total urinary bicarbonate excretion which occurs before total "resistance" to the action of Diamox® is finally established, is comparable in patients with heart failure to that of normal persons. No measurements of NH_4^+ or titratable acid excretion were made in these subjects; it is probable, however, that the total deficit of buffer caused by the drug was similar to that of normal persons.

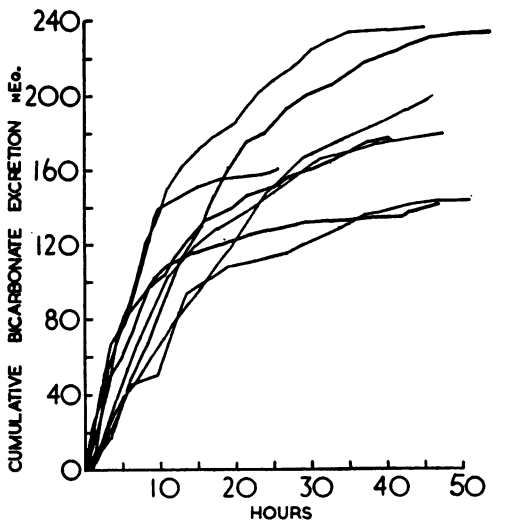

FIG. 4. CUMULATIVE URINARY EXCRETION OF BICARBONATE IN EIGHT PATIENTS WITH CONGESTIVE HEART FAILURE INGESTING DIAMOX® (SEE TEXT)

DISCUSSION

The diminishing effect of Diamox® on urinary bicarbonate excretion has been previously noted by several workers (4, 5, 19), but the nature of the mechanism which limits the drug's action is not entirely clear. The studies of Counihan, Evans, and Milne (20) suggest that lowering of the plasma bicarbonate is the limiting factor, operating through reduction of the load of bicarbonate filtered at the glomeruli. Maren, Wadsworth, Yale, and Alonso (21, 22), however, consider that plasma bicarbonate is "at best a semiquantitative indication of the degree of whole body acidosis following treatment with Diamox®." Irrespective of the ultimate controlling mechanism, it seems probable that Diamox® ceases to cause urinary bicarbonate excretion when a certain total quantity of "bicarbonate-bound base" has been lost from the body. Nevertheless, a number of reports of continued diuretic action of Diamox® have appeared (5, 23, 24). It seems probable that if such sustained diuresis is genuinely due to the drug, then either there must be some mode of recovery which avoids the limiting effect of a loss of bicarbonate ion and permits continued urinary excretion of "bicarbonate-bound base," or else the natriuresis and diuresis cannot be wholly dependent on bicarbonate excretion. The present observations were made to study the possibility that under suitable circumstances of dosage the mechanism and rate of recovery from the metabolic effects of Diamox® would avoid the self-limiting action of the drug.

The augmentation of urinary ammonia and titratable acid excretion which occurred during recovery from Diamox® acidosis also characterizes recovery from other forms of metabolic acidosis (14–18). Both changes represent secretion by the tubule cells into the glomerular filtrate of H^+ derived from H_2CO_3, with parallel regeneration of HCO_3^- for reconstitution of the blood and extracellular bicarbonate. Two well-recognized variations of such compensation for metabolic acidosis exist. In the first mechanism ('a') an exchange of Na^+ or K^+ for the secreted H^+ occurs. This cation exchange would tend to restore any depletion of fixed cations which had occurred during the development of acidosis, and together with the parallel increment of tubular water reabsorption necessary to maintain plasma osmolarity, would result in restoration of both the volume and composition of the extracellular fluid. The balance studies of our two normal subjects suggest that recovery in a normal person is effected predominantly by this mechanism 'a', since approximately 75 per cent of the increment of H^+ excretion during recovery was associated with conservation of fixed cation.

In the second mechanism ('b'), excess H^+ secreted during recovery from acidosis is eliminated in the urine in association with chloride ion withdrawn from the extracellular fluid. Chloride ion withdrawn in this way is replaced by bicarbonate ion derived from H_2CO_3, and "freed" by elimination of the excess H^+. This chloruresis would tend to restore the anion pattern of the plasma; furthermore, no Na^+ or K^+ exchange for H^+ need occur in order to eliminate an excess of H^+ in this instance, and there would, therefore, be no tendency for increased water reabsorption, unless plasma osmolarity had been changed by the original diuresis. Exclusive operation of mechanism 'b' during recovery would allow regeneration of body bicarbonate and thus repetition of the natriuresis and diuretic effects of Diamox® while the extracellular fluid volume was still lower than the pre-Diamox® value. In their balance studies of six subjects, Leaf, Schwartz, and Relman (19) observed an augmented excretion of both ammonium and chloride ions during recovery from the effects of Diamox®, a finding which suggests that mechanism 'b' may at least be partly responsible for recovery from the effects of Diamox® in patients with heart failure.

The present observations were made on normal persons, and it is impossible to draw from them any conclusions as to which qualitative mode of recovery from Diamox® acidosis would be likely to occur in edematous subjects. Certain quantitative aspects of the problem may be discussed in the light of these data, however. The actual time needed for full recovery from the effects of Diamox® will depend on the relative magnitudes of the original buffer depletion and the rate at which excess H^+ can be secreted by the renal tubules during recovery.

The observed daily increments of H^+ excretion ranged from 40 to 80 m.Eq., with a mean of 56 m.Eq. Recovery of the average buffer decrement of 290 m.Eq. in our six normal subjects will thus require 5 to 6 days; the recovery time is highly unlikely to fall below three days, and may be as long as 7 to 9 days.

Similar considerations may be applied to recovery from a solitary dose. The initial dose of Diamox® (0.25 to 0.4 g.) caused a decrement of base which ranged from 117 to 189 m.Eq. (Table V). The mean rate of H^+ increment, 56 m.Eq. per day would thus necessitate, on average, 2 to 4 days for recovery. This finding is in general agreement with Maren, Wadsworth, Yale, and Alonso's (21) statement that "it takes 3 to 5 times longer in the dog to repair a HCO_3^- or base deficit than to create it."

Our observations on the self-limiting action of Diamox® in single daily doses in human subjects, are wholly at variance with the results of Maren, Wadsworth, Yale, and Alonso (21) in dogs. These workers found that Diamox®, in a dosage of 5 to 10 mg. per kg. body weight, did not cause sustained depression of plasma bicarbonate. This was due to the fact that extra acidification during the "drug-free" period after each dose was associated with an H^+ increment sufficient for regeneration of bicarbonate lost in the initial phase of the drug's action, and the plasma bicarbonate was normal when each dose of Diamox® was given. The results already described, were different in all our subjects, and the self-inhibiting effect of Diamox® was apparently determined by the fall of plasma bicarbonate, as described by Counihan, Evans, and Milne (20).

A notable feature of one balance study (subject 'A') was the considerable retention of sodium ion which occurred and the large concomitant positive balance of chloride. It is improbable that this was caused by a fortuitous normal cyclical variation in body salt content as no comparable variation was observed in the 6-day control period. The erratic nature of the changes in urinary chloride excretion caused by Diamox® has also been noted by Maren, Wadsworth, Yale, and Alonso (21). It is probable that a chloruresis may be caused by Diamox® in some circumstances: Schwartz, Relman, and Leaf (24) noted that the large diuresis (and presumed natriuresis) which they observed in some of their edematous subjects, could only be accounted for by chloruresis.

SUMMARY

1. Detailed studies have been made in two normal persons of the changes in urinary electrolyte excretion which occur during ingestion of Diamox®, and during recovery from the drug's effects. It seems probable that recovery of normal subjects is effected by mechanisms similar to those seen in other forms of metabolic acidosis, *i.e.*, exchange of fixed cation for H^+. Alternative mechanisms of recovery exist and the therapeutic implications of the various mechanisms are discussed.

2. The self-limiting effect of Diamox® in single daily doses is described.

3. The total buffer decrement caused by Diamox® (when the drug was ingested to the point where it ceased to cause urinary bicarbonate excretion) ranged from 250 to 400 m.Eq. The rates of urinary H^+ increment observed during recovery from the drug's effects suggest that 5 to 6 days will be required on average for full recovery from a series of doses of Diamox®, and 2 to 4 days for recovery from an initial single dose.

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