ALTERATIONS IN BODY COMPOSITION WITH SODIUM LOADING AND POTASSIUM RESTRICTION IN THE RAT: THE TOTAL BODY SODIUM, NITROGEN, MAGNESIUM AND CALCIUM [1]

By DONALD B. CHEEK and CLARK D. WEST with the technical assistance of CATHERINE CARTER GOLDEN

(From The Children's Hospital Research Foundation and the Department of Pediatrics, University of Cincinnati, Cincinnati, O.)

(Submitted for publication October 27, 1955; accepted March 15, 1956)

Alterations of muscle composition and acid base balance during potassium deficiency have been the subject of previous investigations (1, 2). During metabolic alkalosis and potassium deficiency there is a loss of cellular potassium and a lesser or disproportionate increase in muscle cell sodium (3–5).

The changes in chloride metabolism have not been clearly defined. During potassium depletion and sodium loading in humans, the presence of edema has been recorded (6) and during experiments concerning potassium deficiency in rats a gain in weight was noted (7). The volume of distribution of bromide in malnourished infants with chronic diarrhea subjected to sodium chloride loading and potassium restriction predicted an increase in total body chloride on a total weight basis (8). On the other hand, balance studies on two patients with chronic diarrhea demonstrated significant deficits of body chloride (9) and data are at hand from rats indicating that muscle chloride content during potassium depletion may be low (2) or high (10). Analysis of the whole rat under these circumstances by one group disclosed no significant alteration of body chloride content (10). From a clinical viewpoint, the changes in body composition during potassium restriction assume importance in the management of therapy.

The present experiments were designed to examine the behavior of total electrolyte and serum electrolyte concentrations when groups of rats were subjected to sodium loading with and without potassium restriction, and in the presence or absence of desoxycorticosterone acetate (DCA). One group of animals was given sodium bicarbonate in the drinking water, two groups received sodium chloride for variable periods, and the fourth group received sodium chloride as well as injections of desoxycorticosterone (DCA). For controls the carcasses of rats on a normal diet and on a normal diet with saline for drinking, with and without DCA, have been analyzed. Because of the possibility that some of the changes observed were due to a diminished food intake, a study of the changes in carcass composition during starvation has also been included.

The results indicate that the severity of potassium deficiency, as measured by total carcass potassium content, cannot be accurately predicted by any specific change in body composition or by alteration in acid base balance. Total body chloride is usually decreased but, under certain conditions, total chloride may be increased. The status of total body chloride does not correlate with the serum chloride concentration. While total body sodium increased in all potassium deficient animals, the increment could not be correlated with the decrement in body potassium.

EXPERIMENTAL PROCEDURE AND METHODS

Male rats derived from the Wistar strain were used. Normal values for total body electrolyte, water and nitrogen, furnishing standards for comparison of the results on the experimental animals, were determined by carcass analysis of animals of normal health and vigor of varying ages and weights. The larger and older animals had been previously used for breeding purposes. All had been under observation for a period of at least two weeks prior to sacrifice and for one week had been maintained on a synthetic diet of composition previously described (11).

The 48 animals selected for the experimental groups were in most cases of the median weight of the normal animals, 210 to 250 grams. In some instances animals of

[1] This investigation was supported in part by a research grant (H-1638) from the National Heart Institute of the National Institutes of Health, United States Public Health Service.

greater weight were included. They were placed either on a normal or a potassium deficient diet. The normal diet is that described previously (11). The potassium deficient diet was similar to the normal in composition except that potassium was removed and a mixture of sodium chloride and sodium bicarbonate added. A kilogram of potassium deficient diet contained 300 mEq. of sodium, 150 mEq. each of chloride and bicarbonate and 0.5 mEq. of potassium. The animals were divided into 6 groups and subjected to the following conditions:

Group 0. Ten rats received a normal diet and isotonic sodium chloride for drinking during a 10-day period.

Group 1. Thirteen rats received the low potassium diet for three weeks and isotonic sodium bicarbonate for drinking during the last four days prior to sacrifice.

Group 2. Five rats received the low potassium diet for three weeks and isotonic sodium chloride for drinking during the last four days.

Group 3. Five rats received the same treatment as Group 2 but in addition injections of desoxycorticosterone acetate in oil, 2½ mg. daily, were given intramuscularly during the last four days.

Group 4. Four rats were subjected to the same treatment as those in Group 3 except that they were given a normal diet.

Group 5. Six rats received the low potassium diet for eight days and isotonic sodium chloride to drink for eight days prior to sacrifice.

Experiments were also done on a group of five rats to determine the effect of starvation on body composition. These animals were placed in individual cages and given 85 ml. of water daily in which were dissolved 1.7 mEq. of sodium chloride, 4.2 gm. of glucose and a mixture of vitamins. The daily allowance of fluid was completely consumed each day. After eight days of starvation, the animals were sacrificed.

The technique for reducing the rat carcass to a fine, homogeneous, fat free powder and the methods for estimation of carcass chloride, potassium and water and for serum chloride have been described previously (11). The serum CO_2 content was determined by the method of Van Slyke and Neill (12) on blood collected anaerobically from the aorta. Serum sodium and potassium levels were determined by flame photometry.

For total carcass sodium estimation the method of Butler and Tuthill was used (13). One gram of fat free dry carcass was weighed out in a platinum crucible, covered with a nickel lid, and placed in a muffle furnace at 550°C for 20 hours. After cooling the ash was dissolved in 1.5 ml. of 2 N HCl. One drop of alcoholic phenophthalein solution was added and the mixture transferred to a 5-ml. volumetric flask and made up to volume. The solution was then poured into a 10-ml. centrifuge tube and 200 mgm. of Ca $(OH)_2$ were added. The procedure of Butler and Tuthill was then completed. The estimation of total nitrogen was carried out by the method of Conway (14) using 50 mgm. of fat free dry carcass for digestion in a 50-ml. Kjeldahl flask. Each digestion was done for at least eight hours in duplicate and microdiffusion technique was carried out in triplicate. Blanks and a standard solution were run simultaneously. For digestion 3.5 ml. of Conway's acid digestion mixture were employed. For magnesium and calcium determinations 500 mgm. of carcass were ashed and the ash dissolved in weak HCl. To a 2.0-ml. aliquot of the extract of the ash was added 2.0 ml. of saturated ammonium oxalate and the calcium precipitated. To 2.0 ml. of the supernatant fluid were added 2 ml. of 2 per cent KH_2PO_4 and 2 ml. of concentrated ammonia to precipitate magnesium as the triple salt—ammonium magnesium phosphate. After repeated washing and centrifuging the triple salt was dissolved in 0.2 N HCl and the method of Fiske and Subbarow (15) was then employed to determine the amount of phosphate present. The content of magnesium was calculated from a standard curve for the triple salt derived from estimations of standard magnesium solutions. Calcium was estimated by the permanganate titration method, after the calcium oxalate precipitate had been repeatedly washed and dissolved in 2.5 ml. of 30 per cent sulphuric acid.

CALCULATIONS

The chloride space has been used for appraisal of change of extracellular volume and no attempt has been made in the calculation to correct for intracellular chloride or for the slightly greater chloride concentration in connective tissue water as compared with that of a serum ultrafiltrate. It is assumed that neglect of these corrections will produce the same magnitude of error in the experimental as in the normal rats so that the values obtained provide an index of change in extracellular volume. A value of 0.95 is used as the Donnan factor and it is assumed that serum water represents 930 ml. per liter of serum.

The total sodium of the carcass has been partitioned into two fractions, that present in the chloride space and that deposited in the bone and cells. The calculation is based on the assumption that the concentration of sodium throughout the chloride space is the same as that in an ultrafiltrate of serum. That which is not in the chloride space serves as an index of the amount of intracellular and bone sodium. The actual amount in bone and cells is probably greater than that calculated, because the chloride space has not been corrected for factors mentioned above, and is consequently larger than it should be. Hence, it should be emphasized that the calculated values for sodium in the non-chloride space serve only as an index of bone and cell sodium.

RESULTS

Body composition of normal rats

In normal rats total body electrolyte (sodium, potassium, chloride, calcium and magnesium), nitrogen and water were found to be linearly related to the fat free dry solid content of the car-

cass with relatively small scatter. The regression equations expressing the relation of these parameters to fat free dry solid together with the standard errors of estimate are given in Table I. Also included in the table is the regression for bone and cell sodium on fat free dry solid and the average and range for body fat content.

The rats sacrificed to establish the normal values for body electrolyte, water and nitrogen ranged in body weight from 70 to 410 grams, and in fat free dry solid weight from 13 to 96 grams.

TABLE I

Body composition in the normal rat: Regressions of total body electrolyte, nitrogen and water on fat free dry solid *

Regression equations		Standard error of estimate	Number of rats
Na_t (mEq.)	$=0.193$ FFDS$+$ 0.78	$\pm$ 0.64	36
Na_{b+c} (mEq.)	$=0.038$ FFDS$-$ 0.38	$\pm$ 0.55	28
K_t (mEq.)	$=0.283$ FFDS$+$ 0.36	$\pm$ 1.08	47
Cl_t (mEq.)	$=0.131$ FFDS$+$ 1.04	$\pm$ 0.41	35
Mg_t (mEq.)	$=0.101$ FFDS$+$ 0.59	$\pm$ 0.32	32
Ca_t (mEq.)	$=2.20$ FFDS$-$ 1.74	$\pm$11.50	35
N_t (gms.)	$=0.131$ FFDS$+$ 0.07	$\pm$ 0.20	40
H_2O_t (ml.)	$=2.725$ FFDS$+$13.2	$\pm$ 3.70	36
Average fat content: 9.6% body wt. Range 15.3 to 3.3%			36

* Subscript "t" indicates amount in total carcass; Subscript "b+c" indicates amount in bones plus cells; FFDS = fat free dry solid.

Significance of weight changes with potassium deficiency

Animals used in the experiments to determine the effect of potassium deficiency weighed 210 to 250 grams initially. In some of the groups larger animals were included. The range of fat free dry solid weights at sacrifice extended from 33 to 70 grams. In all groups except Group 5 the experimental conditions were such that weight changes during the experiments were relatively slight. While on the potassium deficient diet the animals initially gained weight. Subsequently when electrolyte solution replaced tap water for drinking, some of the groups showed a slight loss of weight. Thus the animals of Groups 0, 1, 2 and 3 underwent a slight weight loss while those of Group 4 receiving a normal diet, DCA and saline to drink showed a small weight gain. In all cases the changes in weight amounted to less than 8 per cent of the initial weight. It seems likely that in these groups the weight changes were mainly the result of changes in body fat and water content with little loss of tissue (measured as fat free dry solid). Evidence of water loss was seen on carcass analysis in Groups 0, 1 and 2. Body water in these animals (as compared with the normal animal of the same fat free dry solid content) was reduced by 4.6, 5.8 and 2.6 per cent, respectively (Figure 5 and Table II). Presumably there were also small changes in body fat content. As compared with an average fat content of 9.6 per cent of body weight in the normal animals, the fat content of Group 0 animals averaged 8.6 per cent, the Group 1 animals, 9.2 per cent, Group 2, 7.7 per cent and Group 3, 9.5 per cent. In the Group 4 animals, which gained weight during the experimental procedure, there was a gain in water of 2.2 per cent (Table II) and fat content at sacrifice averaged 12.5 per cent of body weight. The data would indicate that the experimental conditions to which the animals of Groups 0 to 4 were subjected produced little change in their nutritional status. Weight changes appeared to result primarily from small changes in body water and fat. It would appear that body composition in these groups can be adequately assessed by comparing it with that of a normal rat of the same fat free dry solid content.

In contrast, the experimental conditions to which the animals of Group 5 were subjected were more rigorous. In the course of the experiment a weight loss averaging 16.7 per cent of the initial weight occurred. A fat content at sacrifice averaging 5 per cent of body weight suggested that their caloric intake had been low. The fat loss, however, is not of sufficient magnitude to account for their total weight loss and losses of fat free dry solid together with commensurate amounts of water would be predicted. Reduction of fat free dry solid complicates the assessment of the data of this group. Therefore, an attempt at assessment of the body composition changes occurring in these animals has been made not only by comparing body composition at sacrifice with that of a normal animal of the same fat free dry solid content but also by comparing composition with that calculated to exist at the outset of the experiment. Further assessment is provided by comparison with the data for animals subjected to simple starvation.

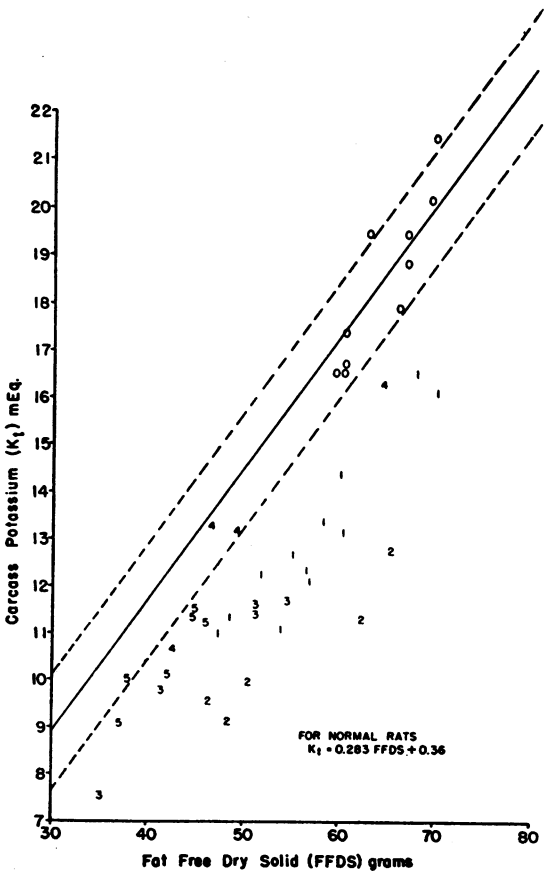

FIG. 1. CARCASS POTASSIUM OF THE RAT VERSUS CARCASS CONTENT OF FAT FREE DRY SOLID

The points for the experimental animals are designated by numbers corresponding to the experimental group. The straight line represents the regression of carcass potassium on fat free dry solid for 47 normal animals fed a normal diet and the broken lines, the standard error of the estimate.

Effect of potassium deficiency, sodium loading and DCA on body composition

In Figure 1 total carcass potassium content of the experimental animals has been plotted against their fat free dry solid content at sacrifice. The values have been indicated by numbers corresponding to the group to which the animal belongs. The potassium content of normal animals is indicated by the line bounded by the broken lines (standard error of estimate). In comparison with a normal animal of the same fat free dry solid content it is evident that all animals on a potassium restricted diet drinking either saline (Groups 2, 3 and 5) or sodium bicarbonate (Group 1) were potassium depleted. Four animals receiving a normal diet, saline to drink and injection of DCA (Group 4) fall in or close to the normal range in potassium content. Animals receiving a normal diet and saline to drink over a ten-day period (Group 0) demonstrated no deficit of carcass potassium.

Carcass chloride and sodium content are shown in similar plots in Figures 2 and 3. Again, the average and standard error of the estimate for normal animals is indicated by the line bounded by the broken lines. According to this method of assessment, body composition with respect to chloride showed considerable variation with the various regimens employed. Animals receiving a potassium restricted diet and additional sodium loads for drinking for a four-day period, with or without DCA (Groups 1, 2 and 3), had a low total chloride while the animals of Group 5 receiving eight days of saline with only eight days of potassium restriction had a high total chloride per

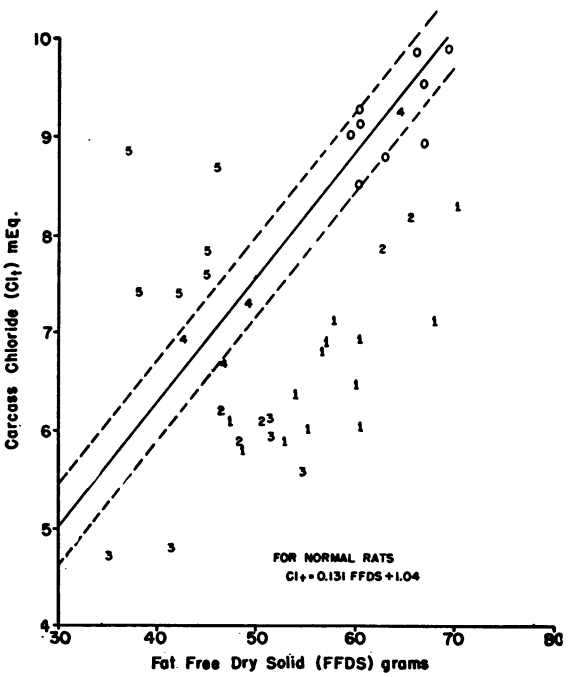

FIG. 2. CARCASS CHLORIDE OF THE RAT VERSUS CARCASS CONTENT OF FAT FREE DRY SOLID

The regression of carcass chloride on fat free dry solid for 35 normal animals fed an adequate diet is indicated by the straight line and the standard error of the estimate, by the broken lines.

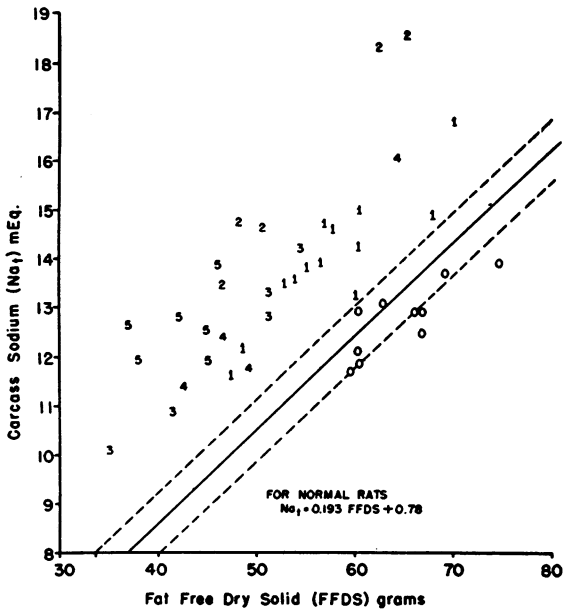

FIG. 3. CARCASS SODIUM VERSUS CARCASS CONTENT OF FAT FREE DRY SOLID

The straight line represents the regression of carcass sodium on fat free dry solid for 36 normal animals on a normal diet and the broken lines, the standard error of the estimate.

unit of fat free dry solid. As will be indicated later the high carcass chloride in this group is probably secondary to the poor nutritional state of the animals rather than the effect of potassium depletion. Both groups on a normal diet (Groups 0 and 4) had a normal total chloride.

The changes in body sodium content, as may be seen in Figure 3, were, on the other hand, more constant. An increased body sodium content was present in all animals receiving a potassium restricted diet (Groups 1, 2, 3, and 5), as well as in those receiving a normal diet, saline and DCA (Group 4). A normal or slightly low body sodium content was seen only in animals on a normal diet with saline for drinking (Group 0).

Values serving as an index of sodium in bone and cells are shown in Figure 4. By the method of calculation employed, the average bone and cell sodium for normal rats, indicated by the solid line, is about 12 per cent of the total in smaller animals and about 17 per cent in larger animals. The rats of Groups 1 and 2 receiving a potassium deficient diet and sodium bicarbonate or saline for drinking had a marked increase in the amount of sodium deposited outside of the chloride space. In the remaining groups the amount of sodium in cells and/or in bone was normal or only slightly altered.

None of the experimental animals showed a deviation from the normal in the relation of total body nitrogen, magnesium, or calcium to fat free dry solid. The graphs expressing these relationships are not shown.

Carcass water content is shown in Figure 5. Total water was definitely altered in the animals receiving a normal diet and saline to drink (Group 0) and in those subjected to a potassium deficient diet and sodium bicarbonate for drinking (Group 1). In these groups body water tended

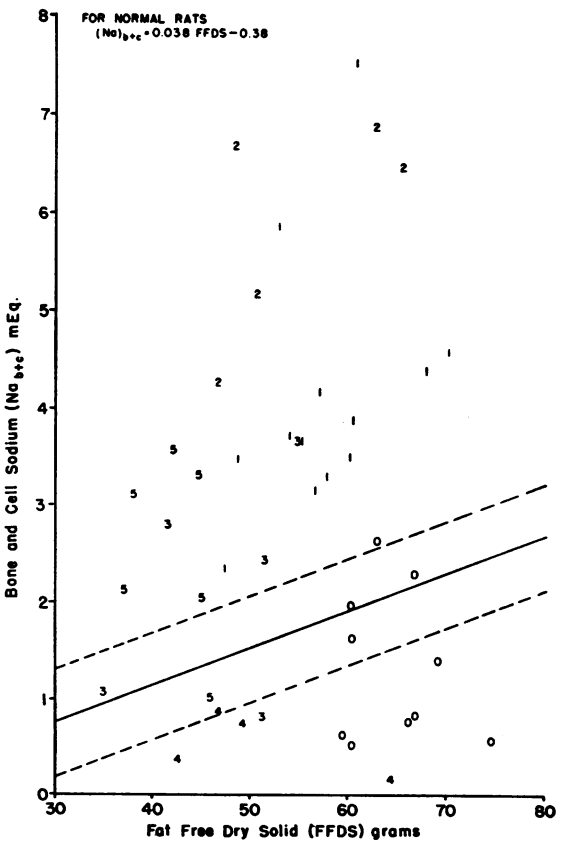

FIG. 4. THE BONE AND CELL SODIUM OF THE RAT (CALCULATED AS THE AMOUNT DEPOSITED OUTSIDE THE CHLORIDE SPACE) VERSUS THE CARCASS CONTENT OF FAT FREE DRY SOLID

The straight line represents the regression of bone and cell sodium on fat free dry solid for 28 normal rats on a normal diet and the broken lines, the standard error of the estimate.

TABLE II

Determined and derived values for body composition and serum electrolytes in potassium depleted and control rats *

	Group 0	Group 1	Group 2	Group 3	Group 4	Group 5	Normal rats
Number of rats	10	13	5	5	4	6	
Diet	Normal	K def. 3 wks.	K def. 3 wks.	K def. 3 wks.	Normal	K def. 8 das.	
Drinking fluid	NaCl 10 das.	$NaHCO_3$ 4 das.	NaCl 4 das.	NaCl 4 das.	NaCl 4 das.	NaCl 8 das.	
DCA				2.5 mg. 4 das.	2.5 mg. 4 das.		
FFDS, gms./rat	64.7	57.6	54.7	46.8	50.8	42.2	50
Serum Na, mEq./L.	145	153	151	172	165	143	145
Serum Cl, mEq./L.	107	90	93	84	94	102	110
Serum K, mEq./L.	4.6	2.0	1.8	1.8	2.3	2.5	4.7
Serum CO_2 content, mEq./L.	24.0	38.5	36.2	45.6	34.3	33.6	24.0
Na_t, % change	−3.8	+17.5	+40.7	+25.0	+23.4	+41.9	10.4 mEq.
Cl_t, % change	−1.0	−23.1	−15.0	−24.0	−2.0	+20.0	6.7 mEq.
Na_t/Cl_t ratio	1.35	2.12	2.32	2.26	1.71	1.62	1.37
K_t, % change	+1.1	−27.5	−33.2	−23.1	−9.1	−14.0	14.5 mEq.
H_2O_t, % change	−4.6	−5.8	−2.6	+0.3	+2.2	+3.7	149.4 ml.
Cl space, % change	+1.8	−5.8	+0.3	+0.2	+15.0	+31.5	60.8 ml.
Na in Cl space, mEq.							
found	11.45	10.10	9.86	10.00	11.82	10.10	
expected†	11.17	10.13	9.62	8.44	9.05	7.70	8.90
Bone Na + cell Na, mEq.							
found	1.31	3.90	6.08	2.30	1.10	2.54	
expected†	2.10	1.80	1.71	1.38	1.53	1.22	1.50

* FFDS = Fat free dry solid; Subscript t means "total"; DCA = Desoxycorticosterone acetate.

† By expected sodium is meant the amount of sodium either inside or outside the chloride space that is present in the normal rat possessing the same fat free dry solid as that of the experimental group under consideration. Mean values for FFDS from the experimental groups are taken for comparison. The expected sodium is derived from the equations for total chloride and sodium for normal rats from 70 to 410 grams in weight.

to be low while in the others there was no significant alteration.

A summary of the changes in body composition in the experimental animals is given in tabular form in Table II. Values for electrolyte and water composition for each group are expressed as percentage of the expected normal. The expected normal is calculated from the regression equations using a fat free dry solid content equal to that of the mean for the group under consideration. Also shown in the table are the serum contents of sodium, chloride, potassium and carbon dioxide as well as derived values as to the amount of sodium in the chloride space and in bone and cells.

The rats receiving a normal diet and saline for drinking over a ten-day period (Group 0) demonstrated no gross change in body composition except for a slight reduction in total body water. There was no change in the $\frac{\text{total Na}}{\text{total Cl}}$ ratio or in the volume of the chloride space.

In rats of Group 1 subjected to potassium restriction for three weeks with four days of sodium bicarbonate for drinking, body potassium was reduced by 27 per cent. Also there was a moderately severe hypochloremic alkalosis with a slight elevation of serum sodium concentration. The total carcass sodium was elevated and total chloride reduced, resulting in a ratio of total chloride to total sodium of 2.12 as compared with a normal value of 1.37. There was a significant reduction of total water as well as in the volume of the chloride space; a small water loss from both extra- and intracellular compartments would be pre-

dicted. The sodium content of the chloride space was calculated as unchanged so that the increment in total sodium apparently resided entirely in cells and/or bone.

The rats of Group 2 were subjected to a regimen similar to that of Group 1 except that sodium chloride rather than sodium bicarbonate was given for drinking. Body composition resembled the Group 1 animals with respect to the loss of potassium and chloride and the degree of alkalosis. The increment in total body sodium was, on the other hand, much greater, and again, nearly all the excess was located in cells and/or in bone.

When DCA was added to the regimen of potassium restriction and saline loading (Group 3) the severity of the potassium depletion was not enhanced. Indeed the potassium loss was less appreciable than that of animals receiving the same regimen without DCA (Group 2). Alkalosis was marked with a very high serum sodium and a low serum chloride concentration. While the gain in body sodium and loss of body chloride was similar to that of the previous groups (Groups 1 and 2), the increment of sodium remained largely in the chloride space with only a small gain in cell or bone sodium.

Animals receiving a normal diet with saline and DCA (Group 4) also showed considerable change in body composition. Carcass potassium content tended to be low in two of the four animals, and alkalosis was present. The origin of the alkalosis appeared to differ from that of potassium restricted animals in that it was mainly the result of sodium retention in the extracellular phase with little change in body chloride content. The average value for bone and cell sodium was, if anything, somewhat reduced. The serum sodium concentration was elevated and serum chloride low; the latter apparently resulted from the expansion of the chloride space in the face of a normal body chloride content.

It is of interest to compare the changes in composition of the rats in Group 3 receiving the combination of potassium restriction and DCA with those in Group 2 receiving only potassium restriction and those in Group 4 receiving only DCA (all 3 groups received saline for drinking). DCA alone (Group 4) appeared to cause a slight loss in body potassium but when given with potassium restriction (Group 3) there was no augmentation of potassium loss. DCA alone caused accumulation of sodium in the chloride space while potassium deficiency resulted in a deposition of sodium outside the chloride space. When the two are given together as in the Group 3 rats, both extracellular and intracellular and/or bone sodium are increased. DCA alone produced no change in body chloride content (Group 4), and potassium deficiency alone caused a moderate reduction. When the two were given together (Group 3), there was a marked reduction in body

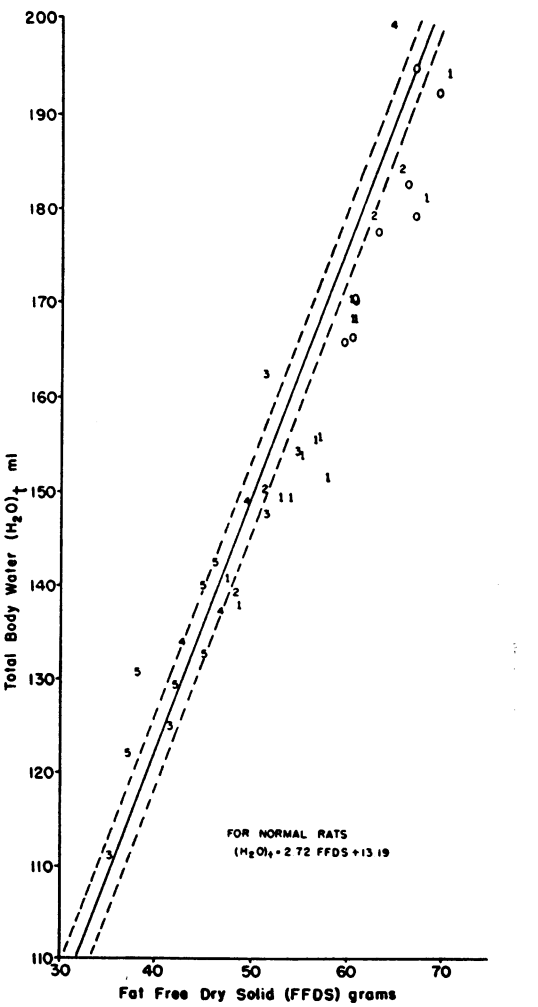

FIG. 5. TOTAL BODY WATER OF THE RAT PLOTTED AGAINST THE CARCASS CONTENT OF FAT FREE DRY SOLID

The straight line represents the regression of body water on fat free dry solid for 36 normal animals on a normal diet.

TABLE III

Calculated initial and final body composition of animals in Group 5 and of animals subjected to starvation—average values *

Body component	Calculated initial composition† A	Composition at sacrifice: Found B	Composition at sacrifice: Change from initial $\frac{B-A}{A} \times 100$	Composition of normal animal with FFDS content = Exper. group: Predicted C	Composition of normal animal with FFDS content = Exper. group: Deviation of exper. group $\frac{B-C}{C} \times 100$
Group 5. Initial wt., 215 gms. Final wt., 179 gms. Wt. loss, 16.7%. N = 6					
Cl_t (mEq.)	7.74	7.82	+ 1.0%	6.58	+20.0%
Na_t (mEq.)	10.66	12.64	+18.6%	8.93	+41.9%
K_t (mEq.)	14.86	10.58	−28.8%	12.31	−14.0%
Na_t/Cl_t	1.38	1.62		1.36	
H_2O_t (ml.)	152.7	133.0	−12.9%	128.2	+ 3.7%
FFDS (gms.)	51.2	42.2	−17.6%	42.2	
Fat (% B.Wt.)	10.0	5.0			
Starvation Group. Initial wt., 160 gms. Final wt., 121.5 gms. Wt. loss, 24%. N = 5					
Cl_t (mEq.)	5.70	5.40	− 5.3%	4.77	+13.2%
Na_t (mEq.)	7.65	7.11	− 7.1%	6.27	+13.4%
K_t (mEq.)	10.42	7.54	−27.7%	8.42	−10.5%
Na_t/Cl_t	1.34	1.32		1.31	
H_2O_t (ml.)	110.2	90.5	−17.7%	90.8	− 0.3%
FFDS (gms.)	35.6	28.5	−20.0%	28.5	
Fat (% B.Wt.)	10.0	2.1			

* Subscript "t" = total electrolyte or water; FFDS = fat free dry solid; N = number of rats.
† FFDS calculated from equation given previously (11),

$$LBM = 3.69\ FFDS + 12.61$$

assuming the lean body mass (LBM) to equal 90 per cent of body weight (body fat = 10 per cent of body weight)

chloride. The reason for the potentiation of chloride loss by DCA in potassium deficient animals is not apparent.

The animals in Group 5 subjected to eight days of potassium restriction and given isotonic saline for drinking were the most difficult to prepare. Originally it was intended that these animals be maintained on the potassium restriction-saline loading regimen for three weeks. However, of 13 animals, only one survived this ordeal and during the period the animals showed gross weakness and failure to thrive. With an eight-day period of potassium restriction and saline loading six of the seven animals survived and the data from these animals are recorded in Tables II and III. It can be noted from Table II that hypochloremic alkalosis was not severe when compared with the results of the other groups. In Table III the initial and final weights of the animals in Group 5 have been recorded and, on the assumption that 10 per cent of their initial weight is fat, the total electrolyte, water and fat free dry solid present at the outset of the experiment has been calculated (column A). The same parameters have been recorded for the actual analysis of the carcass at the time of sacrifice (column B). In column C are given the values for total electrolyte and water which one would expect to find in a normal rat of the same fat free dry solid content as the experimental animals at sacrifice. In the lower half of the table the same procedure has been followed to record data obtained from rats subjected to starvation for an eight-day period. Percentage differences between the predicted and estimated values are shown for convenience.

In both the Group 5 animals and in those subjected to starvation, fat free dry solid and total water were lost with progress of time. The loss of water was commensurate with the loss of fat free dry solid so that the ratio of total water to lean body mass remained essentially the same as that described by Pace and Rathbun for the normal animal (16). In both groups the fat free dry solid at the time of sacrifice is of different composition from that of a normal animal of the same fat free dry solid content. In the starved group, potassium per unit of fat free dry solid is low and sodium and chloride increased. An increase in extracellular water per unit of total water would be predicted. With respect to the elec-

trolyte content of the animals at the outset of the experiment, starvation produced a loss of potassium and only a slight fall in sodium and chloride. The Na_t/Cl_t ratio is maintained at the normal level. In Group 5 the changes in carcass composition resemble those produced by starvation, but the increase of carcass sodium is disproportionately greater than the increase of carcass chloride. Since the main difference between these two groups is the difference in sodium intake, it would seem reasonable to ascribe the disproportionately high carcass sodium of Group 5 and the alkalosis to sodium loading with an inability to excrete sodium effectively.

DISCUSSION

The imposition of various deleterious situations on the homeostatic mechanisms that regulate electrolyte balance appears to produce a wide spectrum of complex changes in body composition that cannot be fully explained. The present investigations pertaining to sodium loading and potassium deficiency emphasize this complexity and reveal the variety of changes in body composition that are possible with metabolic alkalosis. Determination of the electrolyte content of individual tissues may not portray the alterations in total body electrolyte. Analyses of the muscle of potassium deficient animals have suggested that total body chloride is commonly normal (7), whereas in the present study it was usually found to be diminished. It would appear that carcass composition can be considerably influenced by the method used to induce potassium deficiency. The administration of DCA, inadequate caloric intake and the kind of sodium salt administered with the potassium restriction regimen all appear to superimpose alterations in the basic changes in body composition and acid base balance produced by potassium deficiency. It would appear that a consistent program must be followed if results are to be critically compared.

The study would indicate that adequate potassium intake is essential to the preservation of homeostasis. Administration of large loads of saline as drinking water to animals in which potassium intake was adequate (Group 0) caused no change in body composition, while administration of saline to potassium restricted animals produced marked changes in body composition. While the experiments represent extreme conditions, the results offer a warning from a clinical standpoint against the unrestricted use of saline during periods of diminished potassium intake.

The consistent changes in body composition which occurred in all groups of potassium restricted, sodium loaded animals were a diminution in carcass potassium and an increase in carcass sodium. The changes in carcass chloride were not so consistent. Chloride was diminished in all groups in which body weight remained relatively constant during the experiments, while animals which showed a marked weight loss in the course of the experiments (Group 5) had an increase in carcass chloride. The data suggest that starvation may significantly affect body composition and changes in nutritional status of the animal must always be considered when interpreting the results of carcass analysis. In the starved animal with progress of time, there appears to be little change in the body content of sodium and chloride but at sacrifice, per unit of fat free dry solid, sodium and chloride are increased. The animals of Group 5 differed little in carcass chloride content from those subjected to starvation without sodium loading. It seems logical to attribute the high chloride content per unit of fat free dry solid in the animals of Group 5 to their poor nutritional status. With respect to carcass sodium, the Na_t/Cl_t ratio and the status of acid base balance, the Group 5 animals differed markedly from the starvation group.

The observations with respect to chloride in the present study suggest that the correction of potassium deficits by the administration of potassium alone without chloride, as described by Cooke, Segar, Reed, Etzwiler, Vita, Brusilow, and Darrow (7) would not under all circumstances be successful. The frequent occurrence of a low total body chloride would indicate that for complete repair of body deficits, chloride must be given with potassium.

Although the potassium restricted groups were comparable in the direction of change in sodium and, with the exception of Group 5, in chloride, they were not comparable in many other respects. There was no correlation, for example, between the extent of potassium loss and of sodium gain in

the various groups. It is now well recognized that in the potassium deficient animal, part of the increment of the total body sodium is located in muscle cells and the replacement of sodium for potassium in these cells has been set at a ratio of approximately two sodium ions for three potassium ions (3, 17). It is apparent, however, from the present study that in the whole rat there is no consistent correlation between potassium loss and sodium gain. In the rats of Group 1 potassium loss averaged 4.58 mEq. and the sodium gain was less than half this amount, 2.08 mEq. In Group 2 the sodium gain (4.62 mEq.) was nearly as great as the potassium loss (5.25 mEq.) and in Group 5 the sodium increment of 4.75 mEq. greatly exceeded the potassium loss of 1.72 mEq. These discrepancies do not, of course, contradict the observation that in the muscle cell, three potassium ions are lost for every two sodium gained. Changes in extracellular volume, and perhaps deposition of sodium in bone, appear to cause the variations in this ratio in the whole animal. Use of the calculated values for sodium outside the chloride space (bone and cell sodium) to determine the cellular sodium: potassium ratio is dubious in view of the approximate nature of the calculations and because of the fact that some of the increment in non-extracellular sodium may be in bone.

The inconsistent changes in sodium and potassium balance in the whole animal were paralleled by a similar lack of uniformity in total cation balance. In agreement with earlier muscle analyses (18), the body content of magnesium and calcium was unaltered in potassium deficiency so that changes in the fixed cation in the body were solely dependent on changes in sodium and potassium. In the various groups the fixed cation balance varied from − 3.2 to + 2.3 mEq. per 50 gm. of fat free dry solid.

A lack of correlation is also apparent between the changes in acid base balance and the severity of the potassium deficiency. The increment in serum bicarbonate concentration was approximately the same in groups with potassium losses varying from 9 to 33 per cent. In potassium restricted animals receiving DCA, potassium loss was not enhanced, yet there was a marked elevation in serum bicarbonate. The serum level of potassium was roughly correlated with the severity of the potassium deficit but the total range of variation in the deficient groups was only 0.7 mEq. per L. The carcass content of sodium and chloride was in no way reflected in the serum levels of these ions.

A number of factors appears to influence the pattern of body composition in potassium deficient animals. As mentioned above, starvation seems to be a factor of great importance. Likewise the kind of sodium salt administered appears to have some effect on composition. Animals receiving supplementary sodium bicarbonate for four days with potassium restriction (Group 1) had a much smaller gain in total body sodium than those on the same regimen but receiving sodium chloride (Group 2) although the severity of potassium depletion in the two groups was approximately the same. DCA could be considered a third factor altering the results of carcass analysis. Animals restricted in potassium intake and receiving this steroid had a marked increase in the sodium content of the extracellular phase (Group 3) as compared with animals on a similar regimen but not receiving DCA (Group 2). Because the extracellular volume of the animals receiving DCA was not expanded, a severe hypernatremia resulted which in turn potentiated the hypochloremic alkalosis of potassium deficiency. The results indicate that this steroid not only superimposes further alterations in body composition but also is of little value in facilitating the production of potassium deficiency in the experimental animal. In fact animals on a potassium deficient diet receiving DCA had a slightly less severe potassium depletion than the comparable group not receiving DCA.

The effects of DCA in animals on a normal diet (Group 4), as seen in the present study are in complete agreement with the observations of others. Hypernatremia (19–21) with an increase in exchangeable sodium (21, 22) is also seen following the administration of DCA to dogs. Exchangeable chloride remains normal (21). Muscle potassium (20) as well as total body potassium (22) is decreased and serum potassium levels are low. Similar to the present study, this steroid may also produce an alkalosis in dogs which appears to be independent of potassium loss.

Gamble (21) found that the alkalosis would disappear when DCA was withdrawn without potassium being present in the diet. The average potassium loss in the Group 4 animals in the present study approximated the amount that has been termed labile potassium—an amount that can be lost without alteration in acid base balance.

SUMMARY

1. To ascertain the changes in body composition produced by potassium restriction, groups of normal rats were given diets low in potassium and either isotonic sodium chloride or sodium bicarbonate for drinking for variable periods of time. DCA was administered to some of the groups. The control groups consisted of animals on a normal diet with isotonic saline for drinking with and without DCA and of animals subjected to starvation. The results were assessed by comparing the body composition of the experimental animals with that of normal animals.

2. The basic change in the composition of the potassium restricted-sodium loaded animals was an increase in total body sodium, a reduction in potassium, and, in those animals remaining in good nutrition, a fall in body chloride. Animals losing weight in the course of the experiments, and control animals subjected to starvation without sodium loading, had a definite increase in chloride per unit of fat free dry solid but little change in body chloride content as compared with the content prior to experimentation. Starvation appears to alter markedly body composition with respect to chloride, sodium and potassium.

3. Animals given saline to drink and a diet adequate in potassium had no changes in body composition. With sodium loading, the administration of potassium is necessary for preservation of homeostasis.

4. The potassium deficient animals showed no deviation from the normal in their content of calcium, magnesium and nitrogen per unit of fat free dry solid.

5. The pattern of change in body composition produced by potassium restriction is altered by administration of DCA. DCA causes a retention of sodium in the chloride space, thus greatly potentiating the alkalosis, but does not enhance potassium loss.

6. The extent of the potassium depletion could not be correlated with the gain in body sodium, with the changes in acid base balance or with the serum levels of potassium.

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