THE RELATIONSHIP OF TOTAL EXCHANGEABLE POTASSIUM AND CHLORIDE TO LEAN BODY MASS, RED CELL MASS AND CREATININE EXCRETION IN MAN

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Potassium has long been known to be almost completely confined to the intracellular compartment, and the neutral fat present in muscle has been shown to have a low chloride content (1). It seemed probable, therefore, that the error associated with predictions of total exchangeable electrolyte from total body weight would diminish if the variable of depot fat was eliminated by the use of lean body mass as a reference standard. A close correlation of lean body mass with red cell mass (2) and urinary creatinine (3) has previously been shown. Accordingly, measurements of total exchangeable chloride and potassium have been correlated with lean body mass, red cell mass and creatinine excretion in a series of normal subjects.

MATERIALS AND METHODS

Sixteen normal males and fourteen normal females were studied, their ages ranging from 18 to 81 years and their body weights from 44.5 to 100.2 Kg. They were selected to include both lean and obese subjects, the range of body fat being 5 to 50.8 per cent of total body weight.

Total exchangeable potassium (K_e) measurements were carried out according to the method of Corsa, Olney, Steenburg, Ball, and Moore (4), four spot urine specimens being collected between twenty-two and twenty-six hours after injection. The urinary potassium concentration was estimated by flame photometry. Since the short half-life of Cl^{38} precluded its use, Br^{82} was used to measure the chloride space, one plasma sample being obtained after a period of equilibration of twenty-two to twenty-four hours (5). Plasma chloride was measured by the method of Van Slyke (6), and no correction was made for the Donnan equilibrium. The total exchangeable chloride (Cl_e) was derived from the equation:

$$\text{Total exchangeable chloride } (Cl_e) \text{ (mEq.)} = \text{chloride space (liters)} \times \text{serum chloride (mEq. per liter)}$$

The volume of distribution of Cl^{38} in man, two and one-half hours after injection, closely approximates that of Br^{82} (7) which has been used in this study. Moreover, it has been shown in rats that the bromide dilution method provides an estimate of total body chloride which agrees closely with that obtained by direct carcass analysis (8). It is concluded, therefore, that the use of Br^{82} provides a valid estimate of Cl_e which is in turn a true measure of total body chloride. One hundred fifty μc. of K^{42} (potassium chloride), irradiated as potassium carbonate, and 25 μc. of Br^{82} (sodium bromide), irradiated as ammonium bromide, were used irrespective of body weight. The radiation dose for a person weighing 70 Kg. is 0.11 rad[2] due to K^{42}, and 0.06 rad due to Br^{82}.

Values for lean body mass were derived from the Pace-Rathbun formula (9): Lean body mass = Total body water × 100/73. Total body water was derived from the antipyrine space, which was obtained from the antipyrine concentration in plasma using the method of Brodie, Axelrod, Soberman, and Levy (10). The plasma concentration of antipyrine was corrected for plasma proteins which were assumed to average 7 per cent of plasma volume. Values of lean body mass in Subjects 12, 14, 15, 16, 27 and 30 were derived from measurements of red cell mass, employing the correlation previously established (2).

To derive red cell mass, plasma volume measurements were performed by means of the Evans blue (cellulose extraction) method (11), as modified by Muldowney (2). Hematocrit figures were obtained with Wintrobe tubes spun at 3,000 r.p.m. for 55 minutes. A correction for trapped plasma was made according to the figures of Chaplin and Mollison (12). The true venous hematocrit was then corrected by the factor 0.91 to calculate total body hematocrit (13). Red cell mass was calculated from the plasma volume as follows:

$$\text{Red cell mass} = \text{plasma volume} \times (\text{total body hematocrit})/(100 - \text{total body hematocrit}).$$

Measurements of 24-hour urinary creatinine excretion were performed according to the method of Folin (14) as described by Hawk, Oser and Summerson (15).

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[2] One rad is equal to an absorbed dose of 100 ergs per gram of tissue.

TABLE I

The data (males)

Subject	Age	Height	Weight	Anti-pyrine space	Lean body mass	Fat % of body weight	Red cell mass	Exchange-able K	Exchange-able Cl	Urinary creatinine
		cm.	*Kg.*	*liters*	*Kg.*		*ml.*	*mEq.*	*mEq.*	*mg./day*
1	22	168.0	61.3	35.6	48.8	20.4	1,818	2,960	1,710	1,485
2	27	183.0	72.7	45.5	58.5	14.3	2,090	3,680	2,004	1,852
3	22	177.0	68.2	45.1	56.15	9.4	2,060	3,060	2,020	2,110
4	36	175.5	97.4	45.0	61.6	36.7	2,110	3,560	2,270	2,060
5	22	177.0	65.4	35.45	48.6	25.6	1,740	3,030	1,850	1,970
6	28	184.0	71.8	44.6	61.1	15.0	2,223	3,530	2,162	
7	22	192.0	82.0	50.4	71.7	15.7	2,590	4,240	3,015	2,330
8	22	169.0	65.0	37.95	52.0	20.0	1,748	2,910	1,930	1,480
9	23	178.0	65.5	42.5	58.2	11.0			2,185	1,562
10	25	183.0	87.2	53.0	72.6	16.8	2,480		2,450	1,660
11	27	175.0	57.3	36.4	49.8	13.0	1,795	3,160	1,950	
12	33	178.0	77.2		59.5*	23.0	2,100	3,390		1,870
13	43	164.5	61.0	41.9	57.45	6.0	2,035		1,975	
14	29	168.0	55.8		53.0*	5.0	1,890	2,700	1,805	1,213
15	33	165.0	63.6		51.1*	19.7	1,830	2,650	1,776	1,580
16	59	183.0	82.7		55.1*	33.5	1,960	2,720	1,928	1,380

* Derived from red cell mass.

RESULTS

The complete data are given in Tables I and II and the statistical analysis is summarized in Table III.

The correlation of K_e with total body weight (Figure 1) was poor ($r = 0.45$) and the 95 per cent confidence limits were very wide ($\pm$ 53 per cent of mean). When correlating K_e with lean body mass (Figure 2), red cell mass (Figure 3) and 24-hour urinary creatinine (Figure 4), it was found that the correlation coefficients were equally good ($r = 0.90$ to 0.91), and that in each case the 95 per cent confidence limits agreed fairly closely ($\pm$ 25 per cent to 28 per cent of mean).

The correlation of Cl_e with total body weight (Figure 5) was better than that of K_e, the correlation coefficient ($r = 0.58$) being higher, and the 95 per cent confidence limits being narrower ($\pm$ 29 per cent of mean). In the correlation of Cl_e with lean body mass (Figure 6) and red cell mass (Figure 7), the correlation coefficients ($r = 0.93$) were again equally good, and the 95 per cent confidence limits ($\pm$ 14 per cent to 15 per cent of mean) agreed closely. The correlation of 24-hour urinary

TABLE II

The data (females)

Subject	Age	Height	Weight	Anti-pyrine space	Lean body mass	Fat % of body weight	Red cell mass	Exchange-able K	Exchange-able Cl	Urinary creatinine
		cm.	*Kg.*	*liters*	*Kg.*		*ml.*	*mEq.*	*mEq.*	*mg./day*
17	45	140.0	83.6	30.0	41.1	50.8	1,509	1,840	1,580	646
18	55	156.5	82.7	35.4	48.5	41.4	1,840		1,827	
19	43	153.0	86.4	33.0	45.2	47.7	1,688	2,670	1,730	
20	31	168.0	60.0	31.7	43.5	27.4	1,620	2,340	1,525	
21	57	158.0	93.6	38.0	52.0	44.5	1,868	2,400	1,980	890
22	38	155.0	70.5	33.5	45.9	34.8	1,695	2,100		1,168
23	81	152.0	44.5	24.3	33.3	25.2	1,200	1,190	1,248	527
24	55	156.0	66.0	33.1	45.3	31.4	1,605		1,680	1,230
25	18	173.0	100.2	45.0	61.5	39.7	2,080	2,910	2,440	1,310
26	50	157.5	57.2	33.0	45.2	26.7	1,695		1,728	
27	51	151.0	50.0		36.65*	16.0	1,355	1,800		1,262
28	52	152.0	63.6	34.3	47.0	26.0	1,735	2,040	1,720	1,024
29	37	170.0	55.0	33.42	45.8	16.7	1,760		1,830	
30	66	162.5	54.0		42.1*	22.0	1,535	1,610	1,369	810

* Derived from red cell mass.

TABLE III

K_e or Cl_e	Reference standard	r	95 per cent confidence limits	95 per cent confidence limits	Equation of regression line
			*mEq.**	*% of mean*	
K_e	Total body weight (TBW)	0.45	±1,440	±53	K_e = 21.71(TBW) + 1,181.5
K_e	Lean body mass (LBM)	0.90	± 680	±25	K_e = 73.98(LBM) − 1,063.3
K_e	Red cell mass (RCM)	0.91	± 670	±25	K_e = 2.21(RCM) − 1,341.1
K_e	Urinary creatinine (C)	0.90	± 740	±28	K_e = 1.36(C) + 745.6
Cl_e	Total body weight (TBW)	0.58	± 550	±29	Cl_e = 14.35(TBW) + 902.5
Cl_e	Lean body mass (LBM)	0.93	± 270	±14	Cl_e = 37.19(LBM) − 23.2
Cl_e	Red cell mass (RCM)	0.93	± 290	±15	Cl_e = 1.13(RCM) − 204.5
Cl_e	Urinary creatinine (C)	0.72	± 630	±33	Cl_e = 0.58(C) + 118.6

* Taken about the mean value of the reference standard.

creatinine (Figure 8) showed a lower degree of correlation (r = 0.72) and wide 95 per cent confidence limits (± 33 per cent of mean) were obtained.

The standard error of each individual measurement of K_e and Cl_e was ascertained and was found to be ± 5 per cent for K_e and ± 3 per cent for Cl_e. These represent errors in the physical and chemical methods involved, the largest contribution be-

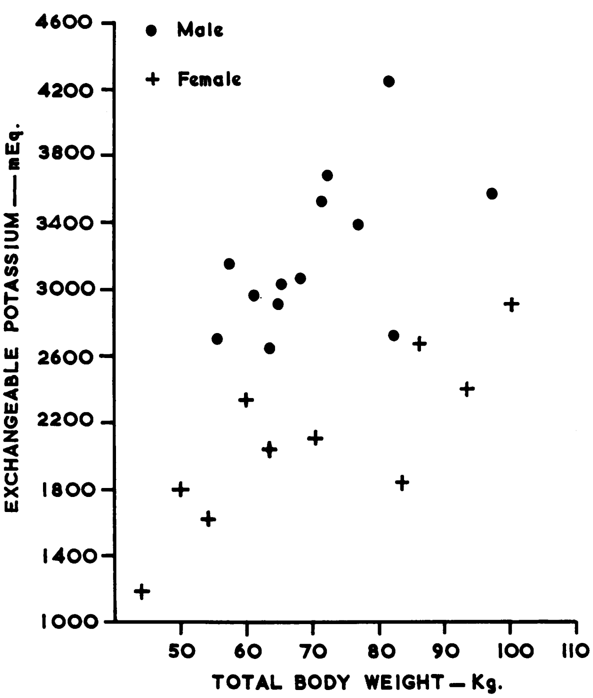

FIG. 1. CORRELATION OF TOTAL EXCHANGEABLE POTASSIUM WITH TOTAL BODY WEIGHT

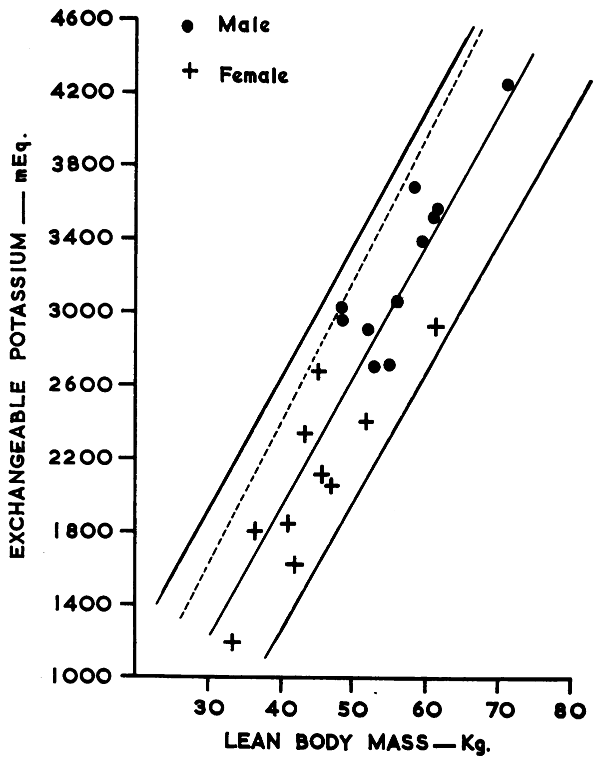

FIG. 2. CORRELATION OF TOTAL EXCHANGEABLE POTASSIUM WITH LEAN BODY MASS

The continuous lines are the regression line of K_e on lean body mass and its 95 per cent confidence limits. The interrupted line is the regression line derived from the data of Ikkos and associates (17).

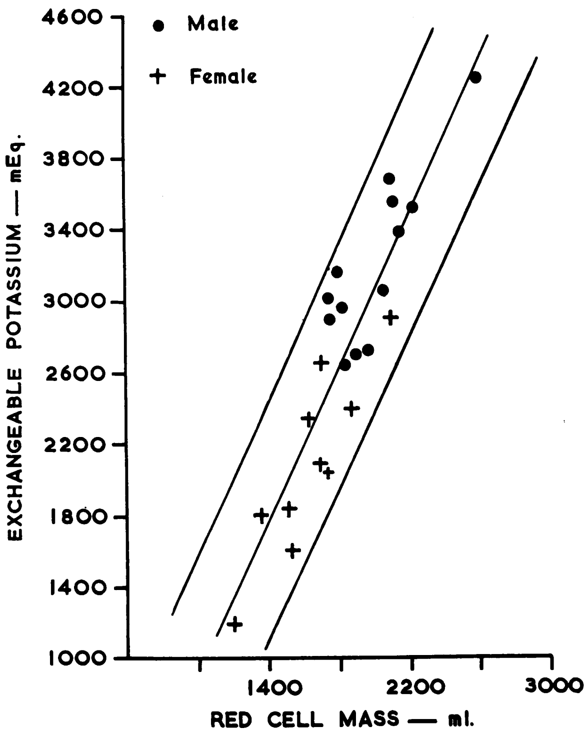

FIG. 3. CORRELATION OF TOTAL EXCHANGEABLE POTASSIUM WITH RED CELL MASS

The regression line of K_e on red cell mass and its 95 per cent confidence limits are shown.

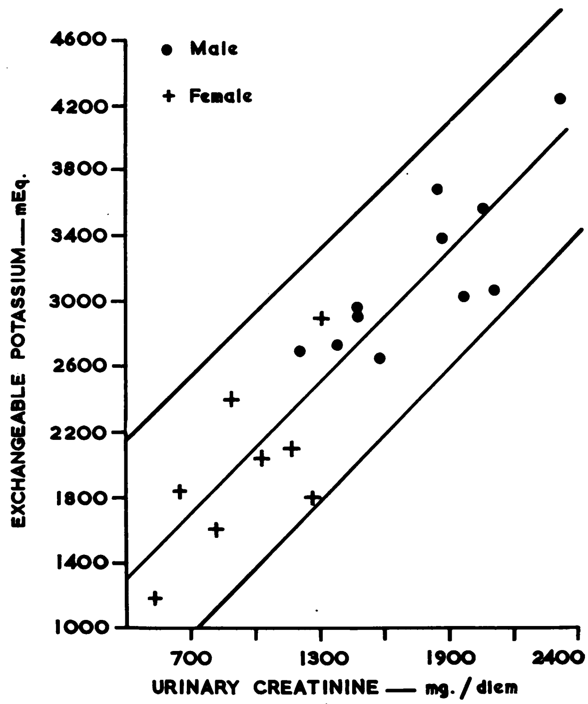

FIG. 4. CORRELATION OF TOTAL EXCHANGEABLE POTASSIUM WITH URINARY CREATININE

The regression line of K_e on urinary creatinine and its 95 per cent confidence limits are shown.

ing the statistical error in counting. Repeated estimations of lean body mass and red cell mass in the same subject agreed to within 4 per cent.

DISCUSSION

The results show that the use of total body weight as a standard of reference for total exchangeable potassium and chloride is unsatisfactory, the 95 per cent confidence limits being ± 53 per cent and ± 29 per cent of the mean, respectively. Our figures agree with those of other workers when both males (4) and females (16) are included. When, on the other hand, total exchangeable potassium and chloride are correlated with lean body mass, the relationship is closer, the 95 per cent confidence limits being ± 25 per cent and ± 14 per cent of the mean, respectively. Furthermore, the relationship is the same for both males and females, the apparent sex difference, based on total body weight, being due to the greater fat content of the female subjects.

These results are supported by experiments carried out by other workers both in human subjects and in animals. Thus, Ikkos, Ljunggren, and Luft (17) in Sweden, as part of an investigation into the relation between extracellular and intracellular water in acromegaly, measured total body water and K_e in a control group of 33 normal subjects. From their data it is possible to derive antipyrine space measurements and, after correcting for the water content of plasma, to estimate lean body mass by means of the Pace-Rathbun formula (9). When this is done, a regression line of K_e on lean body mass is obtained which has been included in Figure 2, the coefficient of the correlation being 0.92. This regression line lies within the 95 per cent confidence limits of the present series, although a slight systematic difference is apparent. Data for K_e, Cl_e and total body water given by Corsa and colleagues (4) and Ikkos, Ljunggren, Luft, and Sjögren in a later paper (18) are consistent with the present results when lean body mass is derived from their values for total body water.

Carcass analysis in rats provides direct evidence of a close correlation of total body potassium and total body chloride with lean body mass (8). Weir (19) had previously demonstrated that total exchangeable chloride was directly related to the lean carcass in dogs, while Hastings and Eichelberger (1) commented on the necessity for analyzing fat-free muscle to allow comparison of the electrolyte content of muscle samples from different animals, or even from different muscles of the same animal. This evidence suggested, therefore, that chloride is confined to lean tissue, and is not present in depot fat. Forbes and Lewis (20), however, have analyzed two human cadavers and found an appreciable chloride content in adipose tissue. This appears to conflict with the results of the present series, and with the data from animals quoted above. The discrepancy is one of definition, however. It must be clearly stated that depot fat, as referred to here and in the preceding communication (2), refers only to the lipid content of adipose tissue. Keys and Brozek (21) estimate that lipids form 62 per cent, water 31 per cent and cell solids 7 per cent of adipose tissue. It is important to note that these relative proportions of water and cell solids are similar to those obtained in muscle, and that the non-lipid portion of adipose tissue is therefore included in the estimate of lean body mass derived from total body water. One would expect that adipose tissue as a whole should contain a percentage of chloride and potassium in proportion to its non-lipid content.

In view of the high degree of correlation which has been shown to exist between lean body mass and red cell mass (2) it is not surprising to find a similar close relationship between red cell mass and both K_e and Cl_e. Provided hematological abnormality can be excluded, it is possible that this relationship may have an application in states of acute electrolyte disturbance which are so often associated with changes in total body water which invalidate the determination of lean body mass by the antipyrine dilution method. Further work,

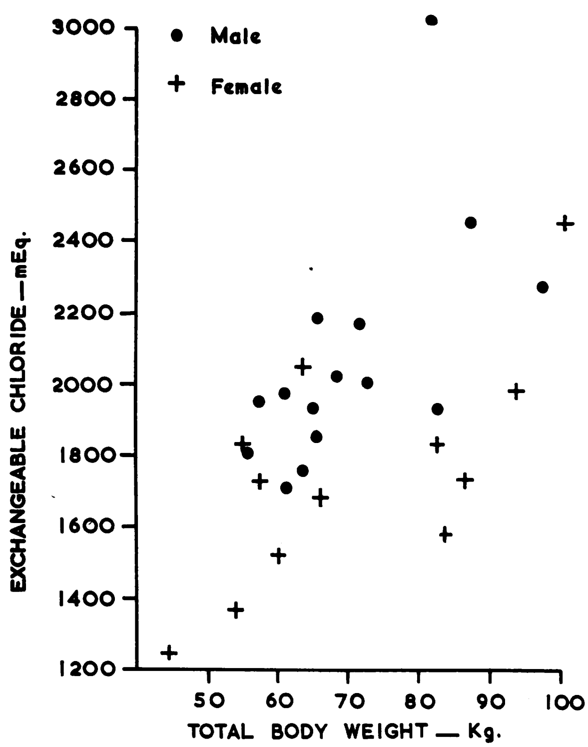

FIG. 5. CORRELATION OF TOTAL EXCHANGEABLE CHLORIDE WITH TOTAL BODY WEIGHT

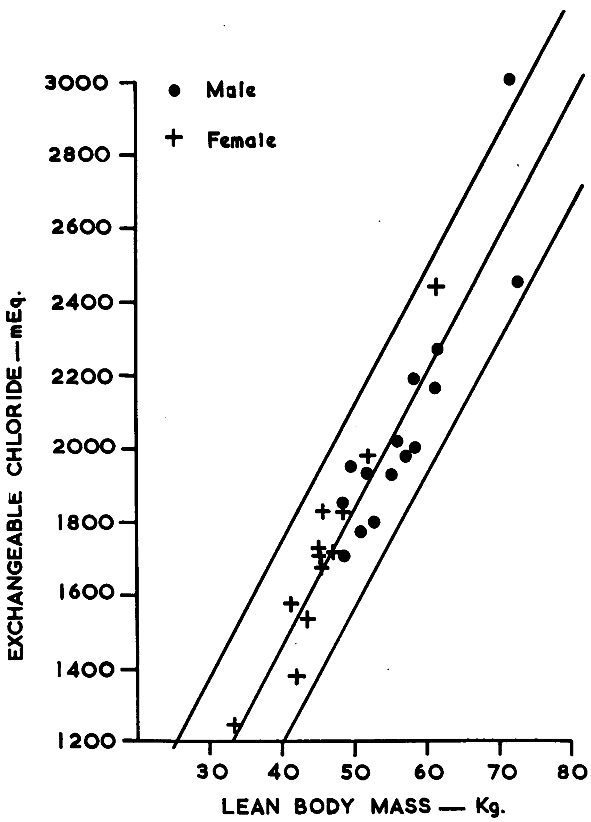

FIG. 6. CORRELATION OF TOTAL EXCHANGEABLE CHLORIDE WITH LEAN BODY MASS

The regression line of K_e on lean body mass and its 95 per cent confidence limits are shown.

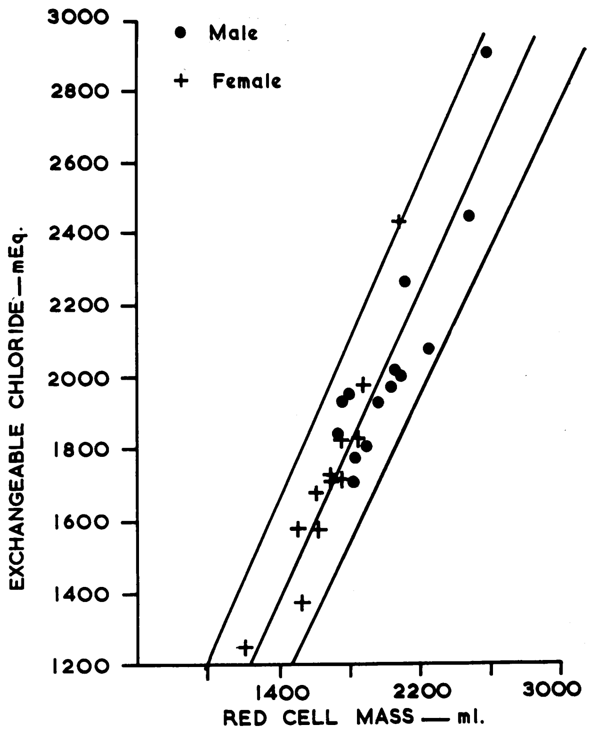

FIG. 7. CORRELATION OF TOTAL EXCHANGEABLE CHLORIDE WITH RED CELL MASS

The regression line of K_e on red cell mass and its 95 per cent confidence limits are shown.

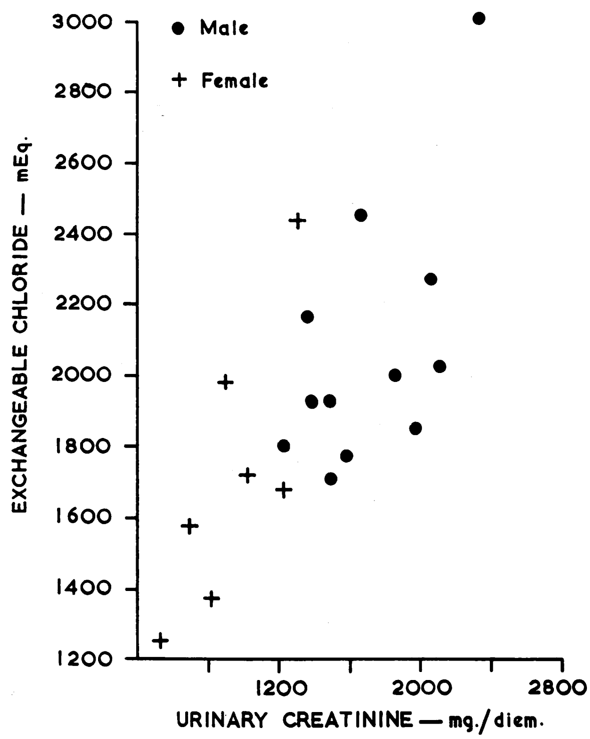

FIG. 8. CORRELATION OF TOTAL EXCHANGEABLE CHLORIDE WITH URINARY CREATININE

however, is required to evaluate the place of this relationship in such states.

The high correlation between K_e and creatinine excretion found by Corsa and associates (4) in five male subjects has been confirmed in this larger series containing both males and females. The closeness of this correlation was unexpected since only single 24-hour specimens of urine, primarily collected for electrolyte measurements, were used for creatinine estimations and dietary protein intake was not controlled. This technique may result in the occasional large errors mentioned by Miller and Blyth (3) and could account for the lean body mass–creatinine correlation ($r = 0.73$, 95 per cent confidence limits $\pm$ 28 per cent of mean) of this series as compared with that of Miller and Blyth ($r = 0.826$, 90 per cent confidence limits $\pm$ 13.1 per cent of mean). This, however, could not wholly explain why Cl_e did not correlate any better with creatinine than with total body weight. The value of creatinine excretion as a standard of reference for K_e lies in its relative ease of estimation, particularly where facilities for measuring lean body mass or red cell mass are not available. It suffers from the disadvantage that our knowledge of disturbances of creatinine metabolism in pathological states is incomplete.

The assessment of electrolyte disturbance by means of serum electrolyte concentration has been shown to be fallacious insofar as serum concentration does not necessarily reflect changes in total body content (22). It is highly desirable that measurements of total exchangeable electrolyte should be employed in such studies. The application of these measurements has, however, been limited by the lack of a suitable reference standard. It is hoped that the provision of these narrower ranges of normal values for total exchangeable potassium and chloride may be of value in the study of electrolyte disturbances.

SUMMARY

1. The correlation of total exchangeable potassium (K_e) and chloride (Cl_e) with total body weight in normal subjects is poor.

2. A close correlation of K_e and Cl_e with lean body mass and red cell mass in normal subjects is established.

3. There is a close relationship between 24-hour urinary creatinine and K_e, but not between 24-hour urinary creatinine and Cl_e.

4. The correlations hold throughout a wide range of age and body weight and are the same for both males and females.

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REFERENCES

1. Hastings, A. B., and Eichelberger, L., The exchange of salt and water between muscle and blood. I. The effect of an increase in total body water produced by the intravenous injection of isotonic salt solutions. J. Biol. Chem., 1937, **117**, 73.
2. Muldowney, F. P., The relationship of total red cell mass to lean body mass in man. Clin. Sc., 1957, **16**, 163.
3. Miller, A. T., Jr., and Blyth, C. S., Estimation of lean body mass and body fat from basal oxygen consumption and creatinine excretion. J. Applied Physiol., 1952, **5**, 73.
4. Corsa, L., Jr., Olney, J. M., Jr., Steenburg, R. W., Ball, M. R., and Moore, F. D., The measurement of exchangeable potassium in man by isotope dilution. J. Clin. Invest., 1950, **29**, 1280.
5. Bradley, J. E. S., Davidsson, D., MacIntyre, I., and Rapoport, A., Estimation of extracellular volume using ^{82}Br (abstract). Biochem. J., 1956, **62**, 33.
6. Van Slyke, D. D., The determination of chlorides in blood and tissues. J. Biol. Chem., 1923, **58**, 523.
7. Gamble, J. L., Jr., Robertson, J. S., Hannigan, C. A., Foster, C. G., and Farr, L. E., Chloride, bromide, sodium and sucrose spaces in man. J. Clin. Invest., 1953, **32**, 483.
8. Cheek, D. B., and West, C. D., An appraisal of methods of tissue chloride analysis: the total carcass chloride, exchangeable chloride, potassium and water of the rat. J. Clin. Invest., 1955, **34**, 1744.
9. Pace, N., and Rathbun, E. N., Studies on body composition. III. The body water and chemically combined nitrogen content in relation to fat content. J. Biol. Chem., 1945, **158**, 685.
10. Brodie, B. B., Axelrod, J., Soberman, R., and Levy, B. B., The estimation of antipyrine in biological materials. J. Biol. Chem., 1949, **179**, 25.
11. Bedwell, G. A., Patterson, J., and Swale, J., The estimation of Evans blue in plasma. J. Clin. Path., 1955, **8**, 61.
12. Chaplin, H., Jr., and Mollison, P. L., Correction for plasma trapped in the red cell column of the hematocrit. Blood, 1952, **7**, 1227.
13. Chaplin, H., Jr., Mollison, P. L., and Vetter, H., The body/venous hematocrit ratio: its constancy over a wide hematocrit range. J. Clin. Invest., 1953, **32**, 1309.
14. Folin, O., On the determination of creatinine and creatine in urine. J. Biol. Chem., 1914, **17**, 469.
15. Hawk, P. B., Oser, B. L., and Summerson, W. H., Practical Physiological Chemistry, 12th ed. London, J. & A. Churchill, Ltd., 1951, p. 840.
16. Aikawa, J. K., Harrell, G. T., and Eisenberg, B., The exchangeable potassium content of normal women. J. Clin. Invest., 1952, **31**, 367.
17. Ikkos, D., Ljunggren, H., and Luft, R., The relation between extracellular and intracellular water in acromegaly. Acta. endocrinol., 1956, **21**, 211.
18. Ikkos, D., Ljunggren, H., Luft, R., and Sjögren, B., Content and distribution of potassium and chloride in adults. Metabolism, 1955, **4**, 231.
19. Weir, E. G., Further observations on total chloride content; the relationship between body fat and body chloride. Am. J. Physiol., 1940, **130**, 608.
20. Forbes, G. B., and Lewis, A. M., Total sodium, potassium and chloride in adult man. J. Clin. Invest., 1956, **35**, 596.
21. Keys, A., and Brozek, J., Body fat in adult man. Physiol. Rev., 1953, **33**, 245.
22. Moore, F. D., Bodily changes in surgical convalescence. I. The normal sequence—observations and interpretations. Ann. Surg., 1953, **137**, 289.