CLXXV. URINARY OUTPUT OF CREATINE AND CREATININE ASSOCIATED WITH PHYSICAL EXERCISE, AND ITS RELATIONSHIP TO CARBOHYDRATE METABOLISM

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THE first accurate quantitative estimations of creatine and creatinine were carried out by Folin [1904]. Since that time it has become generally accepted that creatinuria occurs regularly in both sexes during childhood and also during menstruation and pregnancy. It is also known to occur in such conditions as complete starvation, diabetes, Graves' disease and certain myopathies. Apart from such conditions as these, creatinuria has not been generally accepted as occurring in adult men. Thus Rose [1911-12] and Harding & Gaebler [1922] regard 16 years of age as the limit for creatinuria, whilst Light & Warren [1934] found creatinuria in males only up to the age of 19. Recently, however, it has been reported that creatinuria may occur under certain conditions at later ages. Djen & Platt [1934] found creatinuria in adult males of Chinese extraction, whilst Taylor & Chew [1936] described creatinuria, increasing after exercise and diminishing on resting, in students and laboratory workers between 20 and 34 years of age. Haldi & Bachman [1936] found that the ingestion of sugars, glucose and particularly fructose, produced a creatinuria which was further increased by exercise.

Folin [1905] studied the creatinine output in a number of "normal" individuals and noted that the daily creatinine excretion for a given individual was constant and independent of the amount of nitrogen excreted in the urine (the amount varying from 1050 to 1660 mg. per day). At this time he proposed the term "creatinine coefficient" to indicate mg. creatinine excreted daily per kg. body weight. It appeared that the more obese the individual the lower the creatinine coefficient. Shaffer [1908, 1] noted that the daily creatinine output in a large number of individuals seemed to be directly proportional to their muscular development. He also defined the creatinine coefficient in terms of mg. creatinine, or creatinine nitrogen, excreted daily per kg. body weight.

To obtain further information on these points, particularly the influence of age, muscular activity and carbohydrate intake, the daily excretion of creatine and creatinine in a group of students attending the Carnegie Physical Training College, Leeds, was investigated. These men, ranging in age from 19 to 30, were carrying out a fixed routine of physical activity including gymnastics, swimming, athletics and team games, extending over 9 months. The diet was such that the average daily supply of food amounted to 4200 calories per head. It contained large amounts of readily assimilable carbohydrate and appeared to be adequate from the point of view of its content of animal protein and vitamins. Because of the large amount of carbohydrate being metabolized by the subjects, and in view of the important role played by vitamin B₁ in intermediate carbohydrate metabolism, special attention was paid to the adequacy of the supply of this

vitamin. The diet was apparently adequate for the supply of energy, since the majority of students showed an increase in weight during the year.

Observations were made on 97 students who were dealt with in two groups. Group A consisted of 42 students on whom observations were made during October, November and December 1936. Group B consisted of 55 students on whom observations were undertaken during October, November and December 1937, and repeated in May and June 1938. In addition, a sub-group of 12 individuals was taken from Group B and fed for a period on a low carbohydrate diet, and another sub-group of 12 individuals was taken in December 1939 to determine the effect of exercise on creatine and creatinine output.

EXPERIMENTAL

Great care was taken in the collection of urine to ensure that an accurate 24 hr. sample was obtained. Urine was voided at 8.0 a.m., this sample being thrown away; thereafter all samples voided were collected, including a final one at 8.0 a.m. the following morning. Urine was collected at all times immediately before going to stool. A few drops of toluene were added to each collecting bottle.

Estimation of creatine and creatinine. Preliminary tests for acetone bodies and reducing sugar were carried out by Rothera's and Benedict's tests, since both acetone and glucose interfere with the accurate estimation of creatine. This is a precaution which appears to have been overlooked by a number of workers.

Quantitative estimation of creatinine was carried out by the micro-method of Folin and creatine was estimated by the modification of Folin's micro-method, described by Hunter [1928].

RESULTS

Creatine excretion

The examination of a 24 hr. sample of urine from 97 subjects demonstrated creatinuria in 96 of them. The daily output of the 96 varied from 92 to 1200 mg., the mean being 637 mg. The results are expressed graphically in Fig. 1.

Fig. 1. The daily output (mg./24 hr.) of creatine (a) and creatinine (b) as observed in 97 subjects.

The present investigation showed a higher daily excretion of creatine in males than has hitherto been recorded, and it was surprising to find creatinuria in all cases examined except one, since most previous observations on normal adults indicated a complete absence of creatine from the urine. It appeared likely from the findings of other workers that one or both of two factors might be responsible for this high output: (a) high carbohydrate intake, which would accord with the findings of Haldi & Bachman [1936]; (b) hard intermittent muscular work, sufficiently severe to lead to oxygen debt several times a day.

Accordingly, it was decided first to investigate the effect of a low carbohydrate diet on the daily excretion of creatine.

A group of 12 individuals from Group B was placed for 1 week on a diet as low as possible in readily assimilable carbohydrate. All jam, sugar, honey, marmalade, cakes etc., were entirely eliminated and bread and potatoes cut down to a minimum. The diet consisted largely of meat and vegetables and the intake of these was increased to try and maintain the total calorie intake at the usual level. Unfortunately it was found impossible to estimate the calorie intake but none of the subjects showed a positive test for acetone bodies while they were on the diet. While living on this diet it became more difficult for the subject to perform his routine physical activities, fatigue came on early during the performance of work and there was a general disinclination to perform the various physical activities demanded of him. One individual collapsed and had to rest in bed. As can be expected, there was some difficulty in persuading the subjects to adhere strictly to the diet, particularly as they appeared to have an actual craving for sweet things. It is remarkable that normally a large amount of their diet was supplied by readily assimilable forms of carbohydrate, taken in the form of jam, sugar and honey. The daily excretion of creatinine and creatine was determined immediately before commencing the new diet, during the first day and during the fourth day. Observations at the end of the week were nullified by the inability of several individuals to adhere to the diet.

Table I. *Creatine and creatinine outputs of 11 individuals on a low carbohydrate diet*

High carbohydrate diet		Low carbohydrate diet 1st day		Low carbohydrate diet 4th day	
Creatinine mg.	Creatine mg.	Creatinine mg.	Creatine mg.	Creatinine mg.	Creatine mg.
2330	640	2446	290	1800	0
2000	750	2220	348	2053	370
1860	720	1974	628	1740	300
1780	615	2120	395	2116	380
2370	890	2040	464	2687	395
2040	270	1980	155	2052	230
2520	160	2160	464	2208	460
2096	880	2180	476	2134	0
2450	740	2225	314	2040	0
2220	1440	2000	880	2026	120
1870	590	2200	430	2320	300
Mean 2139	700	2140	440	2107	230

The results of the experiment are given in Table I and show a definite lowering in the daily excretion of creatine in all cases but one. In three cases the creatinuria had disappeared by the fourth day. This was in marked contrast to the findings in the 97 individuals who were assimilating a high carbohydrate diet, among which only one showed an absence of creatine. From calculations of the mean values and the standard deviations it can be shown that the results are statistically significant.

The explanation of these results was complicated by the fact that in addition to the decreased intake of carbohydrate there was an unavoidable decrease in the amount of physical exercise performed. In order to assess the effect which this decrease in activity might have had on the results, a control experiment was carried out to find the effect of decreased exercise on the excretion of creatine.

The excretion of creatine was determined in a group of 12 subjects on Thursday, Saturday and Sunday of one 7-day period. Although there was a considerable decrease in activity on Saturday and Sunday compared with Thursday, the small decrease noticed in the mean excretion of creatine on Saturday and Sunday was without significance and some subjects actually showed an increased creatinuria (see Table II).

Table II. *Effect of reduced amount of physical exercise (Saturday and Sunday) on the creatine and creatinine output of 12 subjects*

Thursday		Saturday		Sunday	
Creatinine	Creatine	Creatinine	Creatine	Creatinine	Creatine
2194	760	1960	476	1910	554
2320	1749	2230	321	2304	444
1924	1661	2157	438	2130	374
2162	1238	2214	160	2133	248
2465	168	2542	101	2460	610
1763	240	1800	325	1617	408
2222	180	2266	104	1786	408
2190	0	2270	0	2060	476
2074	146	1966	184	2206	367
2191	182	2282	516	2000	414
1873	387	2065	385	1930	405
2180	291	1883	217	1810	488
Mean 2130	583	2136	269	2029	400

These results therefore suggest that a diet rich in readily assimilable carbohydrate is associated with creatinuria which is quickly reduced after a change to a low carbohydrate diet.

Creatinine excretion

The daily excretion of creatinine in the 97 subjects examined varied from 1430 to 3800 mg. (see Fig. 1). Folin's observations on 30 subjects showed a variation from 1050 to 1660 mg.

The high creatinine excretion of these subjects is reflected in the "creatinine-nitrogen coefficient" [Shaffer, 1908, 2] which varies from 7.8 to 19.6 mg. creatinine-nitrogen per kg. of body weight with a mean of 11.6 (Fig. 2). Shaffer gives a range of 5.4 to 11.7 with a mean of 8 to 9 for "supposedly normal men", whilst Folin (on asylum inmates) gives a range of 3.5 to 9.8 with a mean of 6.

It is of interest to note that the subject having the highest creatinine coefficient of 19.6 could jump 6 ft. in the high jump and 12 ft. in the pole vault; in addition, his amount of daily practice was well above the average. This same individual also showed the highest daily excretion of creatine, i.e. 1200 mg.

The daily excretion of creatinine was determined in the selected groups of individuals referred to in the previous section. Neither low carbohydrate diet nor diminished amounts of exercise were accompanied by any significant change in the creatinine excretion (Tables I and II).

Fig. 2. Distribution of creatinine-nitrogen coefficients (mg. creatinine-nitrogen/kg./24 hr.) amongst 97 subjects.

*Variations in the output of creatine and creatinine associated with season
and increased fitness for exercise*

In order to determine if the course of physical training affected the creatinuria and high creatinine coefficients, estimations of creatine and creatinine outputs were repeated on 45 subjects of Group B during May and June 1938, i.e. towards the end of the year's training.

Although in many cases there were variations in figures for both creatine and creatinine from those obtained during October, November and December 1937, nevertheless, the high creatinine coefficients and creatinuria were maintained.

These individual variations in excretion were not all in the same direction either for creatinine or creatine, neither was any significant shift shown in the frequency distribution (Figs. 3 and 4).

Fig. 3.

Fig. 4.

Fig. 3. (Upper): The daily output (mg./24 hr.) of creatine (a) and creatinine (b) observed in 45 subjects [Group B] in October 1937. (Lower): The same group examined in May 1938.

Fig. 4. Distribution of creatinine-nitrogen coefficients amongst the 45 members of Group B, (a) October 1937, (b) May 1938.

DISCUSSION

Creatinuria

There is evidence that muscular exercise and the ingestion of large amounts of glucose can both contribute towards the production of creatinuria, and from results described in this paper it appears that the latter factor may be the more important.

The production of creatinuria following the ingestion of glucose might be explained by experiments reported by Brentano [1932] and by Cori & Cori [1931], who found that the subcutaneous injection of insulin produced creatinuria, for the intake of glucose, especially in large amounts, presumably evokes a secretion of insulin by the pancreas.

The results presented here are in agreement with those of Haldi & Bachman [1936] who produced creatinuria by feeding glucose and fructose, but their results were obtained on only two adult subjects and the diet throughout the 24 hr. prior to and during the experiments was not indicated.

Excretion of creatinine

Since Folin [1905] suggested that the daily creatinine output might be taken as an index of endogenous metabolism, other views regarding its significance have been put forward.

Shaffer [1908, 2], Burger [1919] and McClugage *et al.* [1931] found a direct relationship between muscular development and creatinine excretion in adult men, and both Burger and Shaffer regarded the creatinine coefficient as a measure of the "participation of the muscles in the total body weight". In agreement with this view Hodgson & Lewis [1928] found that women have lower creatinine coefficients than men, with the exception that women whose muscles had been developed by muscular exercise had creatinine coefficients comparable with those of men. Talbot [1936] found in children that low coefficients were associated with obesity and high coefficients with lack of fat and well-developed muscles.

Myers & Fine [1915], working with cats, dogs and rabbits, found a relationship between the concentration of creatine in the muscles and the creatinine coefficient, and described the creatinine coefficient as being "an index of the concentration of creatine in the muscles or body". These findings have been confirmed by Palladin [1924] but have been denied by Benedict & Osterberg [1914], Chanutin [1932] and Garot [1930].

It would appear that the creatinine coefficient bears a relationship to the proportion of muscular tissue in the total body weight.

The experimental findings recorded in this paper are in agreement with these results. The subjects used in the experiment consisted of individuals picked from a large number of applicants for their ability at gymnastics and athletics. Their muscular development in proportion to total body weight was above the average and, accordingly, a high creatinine coefficient was anticipated. But the daily output of creatinine and the creatinine coefficients (creatinine-nitrogen) are higher than any hitherto recorded. The experimental findings offer no support for the idea that increased activity increases the creatinine coefficient, since there was no fall in the creatinine output in the two groups of subjects whose muscular activities were curtailed.

In view of the fact that there was no significant variation in the excretion of creatinine when subjects were placed on a low carbohydrate and high protein diet, it is not considered that the protein intake of the individuals was responsible for the high creatinine output.

Folin's original experiments were carried out on 30 inmates of an asylum, and in his paper no data are given concerning the types of patient or the nature of their muscular activity. It is more than likely that a group of such subjects taken at random for experimental purposes would contain some suffering from melancholia and probably characterized by immobility for long periods with consequent diminution in muscular development. Moreover, there is no indication of the diets or ages of these patients. Following the demonstration of the dependence of creatinine excretion upon muscular development and from a consideration of the opinions of other workers, it seems very difficult to take these figures of Folin as representative of average normal individuals, and although they are still quoted in physiological textbooks as figures for normal individuals, their revision is obviously required.

In conclusion it may be stated that an estimation of the creatinine coefficient can afford valuable evidence regarding the muscular development and obesity of the individual. Such a test should prove useful to those associated with physical

training and the assessment of the state of nutrition of individuals, including children. But the results of investigations of the excretion of creatine and creatinine should always be correlated with the diet and the amount of physical activity undertaken.

SUMMARY

1. The excretion of creatine and creatinine was investigated in 97 male students resident in a physical training college.

2. The subjects showed a higher creatinine-nitrogen coefficient (range 7.8–19.6 mg./kg./24 hr.; mean 11.6) than the accepted “normal” figure (range 5.4–11.7; mean 8–9).

3. Although creatinuria is not usually found in adult males it occurred in 96 of the 97 subjects, the range for the 24 hr. excretion being 92 to 1200 mg. with a mean of 637 mg.

4. No correlation was detected between increasing physical fitness and the magnitude of the creatine or creatinine excretion.

5. Subjects placed on a low carbohydrate diet showed a disinclination to undertake muscular work, and fatigue ensued more rapidly than usual. In these circumstances creatinuria diminished, and in some cases disappeared.

6. The probable causes of the creatinuria and the significance to be attached to the creatinine-nitrogen coefficient are discussed.

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