THE UNIVERSITY *of* EDINBURGH

Title: Experimental and clinical studies in the physiology and pathology of carbohydrate and protein metabolism

Author: Robert Arno Krause

Digitisation notes:

p. 73 & 74 paginated twice

University of Edinburgh.

THESES PRIZE COMPETITION.

Thesis by Robert A. Krause

It is requested that this Thesis should be read and circulated in the following order, and thereafter returned to the Office of the Medical Faculty :—

		When Forwarded.
Professor	Schäfer	14th May
,,	Wyllie	21st May
,,	Smith	4th June
,,	Thomson	27th May
,,	Robinson	5th June

N.B.—When forwarding this Thesis to next Member of Committee, it should, if possible, be passed through the Office. When more convenient to hand direct to next reader, a Post Card should be sent to the Office giving date when Thesis forwarded.

EXPERIMENTAL AND CLINICAL STUDIES

in the

PHYSIOLOGY AND PATHOLOGY

of

CARBOHYDRATE AND PROTEIN METABOLISM.

by

Robert Arno Krause,
M.B., Ch.B., D.P.H.

for the degree of

Doctor of Medicine (Univ. Edin.)

and also for

the Milner-Fothergill Prize.

Presented March 31st., 1913.

CONTENTS.

INTRODUCTION.

EXPERIMENTAL AND CLINICAL STUDIES IN THE PHYSIOLOGY AND PATHOLOGY OF CARBOHYDRATE AND PROTEIN METABOLISM.

INTRODUCTION.

Views held regarding general metabolism have undergone great changes during the last ten years. Especially is this the case with regard to protein metabolism. The large number of theories regarding protein metabolism bears witness to the defective knowledge which has existed in connection therewith. Several factors have helped to bring about a radical change in our conceptions of the subject. Among the most important is the greatly improved technique of our present day methods, and especially does this apply to the examination of the urine. This factor has helped us greatly in getting information regarding the excretion of the various nitrogenous bodies in the urine, and has indicated to us that if we wish to study the protein metabolism and its expression in the nitrogen excretion, we have to make a more thorough study of the various nitrogenous substances in the urine, and to watch their percentage relationship to one another as well as any changes in their absolute amounts. This method has been applied to normal individuals, and, combined with more complete and detailed analyses of the diet, has given us many valuable data and has shed new light on

various obscure problems. But, in relationship to pathological conditions, this factor has not received general recognition and even less application. This probably is partly due to the fact that the methods in some cases are too elaborate and that they put too great a demand upon valuable time; but some recently described methods seeking to obviate these difficulties will be described in the sequel (v. Analytical methods).

Some other factors that have taken a prominent part in advancing our knowledge of the protein metabolism are the following.

a) Research upon enzymes has thrown some light upon a large number of processes; amongst others, upon the splitting up of the proteins in the intestine, into amino acids, and later the deaminisation of the last named into ammonia and the non-nitrogenous residue.

b) There have been researches upon the relation of digestion to metabolism; and the constitution of the protein has been worked out by Fischer, Kossel, Hofmeister, Abderhalden and others.

c) The importance of the energy requirements of the body has been specially emphasised by Rubner et al.

The three main theories regarding protein metabolism are those of Voit, Pflüger and Folin.

Voit held that there are two forms of protein, the one form being present in the cells as the fixed or tissue (organeiweiss) protein, in contradistinction to the other form dissolved in the nutrient fluids circulating around the cells.

The circulating protein, to which is added that coming from the intestine, is more easily split up than the tissue protein. He found that the protein decompos ition was not proportional to the total protein weight of the body, but to that of the intaken food. Further he showed that a starving dog, previously well fed with large quantities of protein, has a higher nitrogen output during the first days of starvation, as compared to the nitrogen output when the animal had been previously fed with a protein-poor diet and then starved.

Pflüger, on the other hand, held that the increased protein metabolism after a large meal of proteins is due to the cells being saturated (Sättigung mit Eiweiss) with protein; for the circulating fluid is left unchanged in protein concentration owing to the cells instantaneously absorbing the proteins from it. He therefore held that the protein of the food has first to be taken up by the cells and form an integral part of the protoplasm itself before it could be catabolised. Pflüger based his views upon the experiments of Schöndorff. Blood from a starving dog was circulated through the limb of a well fed dog; the urea content of the perfused blood was found to be increased; also, blood of a well fed dog and of a starved dog was perfused through the limb of a starved dog, but no increase in urea was noted in either case. When, however, it is considered that the total quantity of urea obtained was very small (in the one experiment it was only 25mgm. for $4\frac{1}{2}$hrs. whilst the dog was metabolising 35gms. of nitrogen per day) these experiments, as pointed out by Folin, are not sufficient to found a theory upon.

Folin put forward the theory of an exogenous and endogenous form of metabolism, as a result of the analysis of thirty urines of normal subjects, who received two markedly different diets; one a protein-rich, and the other a protein-poor diet. These diets were creatinin- and purin-free.* He found that the constitution of the urine was not such a constant one as physiologists had been led to believe from Voit's experiments. He states '' That quantitative changes in the daily protein catabolism are accompanied by pronounced changes in the distribution of the urinary nitrogen and sulphur, and that the variations occur according to laws that can be formulated with a fair degree of precision''.

In Table-I, we see the nitrogen and sulphur excretion in the urine of a man who has been on the two different diets referred to. The second diet contained only about 1gm. of nitrogen as compared to the 19gms. of the first. Each of those diets is sufficient in caloric value for the requirements of a man of about 70 kilos.

It is evident on looking at Table-I that the effect of these two diets on the partition- nitrogen** and sulphur excretion is not the same in each case. The quantities of certain of the nitrogenous and sulphur constituents in the urine are al-

* The first diet (protein-rich) consisted of milk(500gms.) cream(300gms.), eggs (450gms.), Horlick's malted milk(200gms.), sugar(20gms.), sodium chloride(6gms.) and water to make up to 2litres; also 900cc. of water to drink. The second consisted of pure starch in the form of pure arrowroot(400gms.) and cream (300cc.)

** i.e. nitrogen as variously distributed in the urine .

TABLE I - (normal man)

	July 13	July 20
Volume of urine	1170cc.	385cc.
Total nitrogen	16·8gm.	3.60gm.
Urea-nitrogen	14·70gm. = 87·5%	2·20gm. = 61·7%
Ammonia-nitrogen	0·49gm. = 3·0%	0·42gm. = 11·3%
Uric acid-nitrogen	0·18gm. = 1·1%	0·09gm. = 2·5%
Creatinin-nitrogen	0·58gm. = 3·6%	0·60gm. = 17·2%
Undetermined nitrogen	0·85gm. = 4.9%	0·27gm. = 7·3%
Total SO_3	3·64gm.	0·76gm.
Inorganic SO_3	3·27gm. = 90·0%	0·46gm. = 60·5%
Ethereal SO_3	0·19gm. = 5·2%	0·10gm. = 13·2%
Neutral SO_3	0·18gm. = 4·8%	0·20gm. = 26·3%

Diet on July 13 a protein rich diet.(119gms protein, about 148gms fat and 225 gm Carbohydrate

Diet on July 20 a protein poor diet.

tered and those of others are unaltered in consequence of changed diet.

The absolute amount of creatinin is unaltered (it is always unaffected by the quantity of nitrogen given in the diet, provided that it is creatin-free). This quantity of creatinin is constant for each person though it varies in different individuals. The uric acid is greatly reduced, but when compared to the diminution of the total nitrogen it is found not to be reduced in the same proportion: the percentage of uric acid-nitrogen is increased on the nitrogen-poor diet.

As regards ammonia nitrogen there is usually a diminution with a reduction of the total nitrogen, but not always. Not only does the urea-nitrogen fall with the total nitrogen, but the percentage it contributes to the total nitrogen also falls, as is seen in Table-I.

When we look at the sulphates we find that here the various forms of sulphates which together form the total sulphate excretion, do not behave alike under those two diets. The inorganic sulphates (which form the bulk of the total sulphates) are most definitely affected by variations in the protein metabolism, so that there is an absolute diminution as well as a percentage reduction on the nitrogen-poor diet. Baumann showed that part of the total sulphates -known as ethereal sulphates- are derived from aromatic products formed in the intestines by the action of bacteria on the protein of the food. These ethereal sulphates, as is seen is Table-I are increased in their percentage relationship to the total sulphates- when a diet poor in protein is given. Another

moiety of the sulphur excretion ''neutral sulphur'' behaves, on the other hand, more like the creatinin and bears no relation to the total amount of sulphur excreted.

From the preceding it will be seen that the excretion of urea, inorganic sulphates and to a less extent ammonia, are definitely altered in their absolute and relative amount by variations in the amount of protein intake. On the other hand, creatinin and neutral sulphates are unaffected by variations in the nitrogen intake (provided the diet is creatin-free). As regards uric acid, the amount excreted is not constant when the nitrogen intake is altered, yet it does not show the definite changes which characterise urea or even ammonia.

Reasoning from these observations, Folin propounded the theory that catabolism must be of at least two kinds, the two being essentially quite different. ''One kind is extremely variable in quantity, the other tends to remain constant. The one yields chiefly urea and inorganic sulphates, no creatinin and probably no neutral sulphur. The other, the constant catabolism, is largely represented by creatinin and neutral sulphur, and to a less extent, by uric acid and ethereal sulphates''. Further he says, ''I would therefore call the protein metabolism which tends to be constant, tissue metabolism or endogenous metabolism, and the other, the variable protein metabolism, I would call the exogenous or intermediate metabolism''.

Folin does not exclude urea or inorganic sulphates from the endogenous metabolism; they are, however, at once

affected by any alteration in the nitrogen intake. Creatinin is of great interest in throwing light upon any changes in endogenous metabolism, of which it is the clearest representative. A study of its excretion in pathological conditions is of great value in showing which part of the metabolism is affected and to what extent. It may also help to throw some more light upon the intermediate products of the metabolism.

In the muscles of the body creatin has been found to be present (Liebig et al.); as it can easily be converted into creatinin by hydrolysis with acids, many observers have looked upon creatin as a precursor of creatinin. It has been generally held and taught by various investigators that creatinuria never occurs under physiological conditions. It is said to be a pathological constituent of the urine, only appearing in the urine in cases of muscular atrohpies; and also in cancer of the liver.

The appearance therefore of creatin in a number of physiological conditions to be presently described is of considerable interest, as it may help to elucidate some of the problems concerning endogenous metabolism.

I therefore wish to present the results of observations extending over the last four years and carried out in this University, upon the creatin excretion under various physiological and pathological conditions, as well as the complete partition nitrogen analyses of the urines under those conditions.

I propose to consider the subject thus introduced under the following headings:-

Creatinuria

1) In various physiological conditions.

2) In various pathological conditions.

3) Under various experimental conditions.

4) Some problems in connection with the genesis of acidosis.

5) The effects of thyroid feeding on carbohydrate metabolism: and remarks on the relation of that metabolism to protein metabolism as evidenced in particular by creatinuria.

6) A description of the methods used in the researches with especial reference to their clinical application.

I here wish to thank Professor Schäfer for permission to carry out the work in his laboratory. I also thank him and Dr. Cramer for the help and advice which they gave me during the course of these researches. My thanks are also due to those Physicians and Surgeons of the Royal Infirmary of Edinburgh by whose courtesy I was enabled to utilise ~~much valuable~~ much valuable clinical material.

The expenses of the various researches were defrayed by grants from the Crichton Research Scholarship, the Moray Fund and the Carnegie Trust.

SECTION-I

EXPERIMENTAL RESEARCHES ON THE EXCRETION OF CREATININ AND CREATIN IN CERTAIN PHYSIOLOGICAL CONDITIONS.

SECTION-I.

EXPERIMINTAL RESEARCHES ON THE EXCRETION OF CREATININ AND CREATIN IN CERTAIN PHYSIOLOGICAL CONDITIONS.

In the introduction I have already pointed out how important it is to know the end products of the protein metabolism, so that we may get a clearer conception of the changes the proteins undergo in their passage through the body. The end products which promise to give the most interesting and definite results are those belonging to the endogenous metabolism, such as creatinin or the neutral sulphates.

There, is also, however, a substance found in the muscles and very closely allied to creatinin, into which it is easily converted; this is creatin. It is generally supposed, that this substance is not excreted in the urine of normal individuals, but only in certain pathological conditionsm such as muscular atrophy (Shaffer), or in disease of the liver (Mellanby) and some other conditions.

I have however found creatin in the urine of normal individuals, at certain age periods and under certain physiological conditions.

In the following researches I shall present my observations on the urines of children of both sexes and of various ages as well as those of adults.

a. Creatinuria in Children.

In investigating the problem of creatinuria, one has also to study the excretion of creatinin, owing to its close relationship to creatin. The following researches will deal with both those substances in detail.

The first to examine the urine of infants for the presence of creatinin were Hoffmann and also Pouchet, who failed to find any creatinin present. Grocco(1886) found a few crystals microscopically of creatinin zinc chloride in some cases. On the other hand Rietschel also using the creatinin zinc chloride method found no creatinin in the urine of normal infants, but a small amount was found to be excreted where a febrile condition was present. Those observers used methods which were unreliable and their results strictly speaking, are of no value.

Closson in 1906 using Folin's method, found creatinin in the urine of suckling kittens. His figures also show that creatin was excreted, but he makes no reference of this in his paper.

Amberg and Morrill, from their results, concluded that the creatinin in the urine of the newly born was present in too small amounts to allow of a conclusion being made regarding its definite presence. They therefore recommended concentrating the urine. Acting on this suggestion Funaro concentrated the urine and was able definitely to establish the presence of creatinin in the urine of infants. Amberg

and Morrill found creatin present in the urine of one of the babies (aged 13days).

Hoogenhuyze and Verploegh also found creatinin in the urine of infants, this was more distinct with Jaffe's reaction than with Weyl's test. The quantities found were:-

1·11mg.	(8days old)	Specimens of 10cc.
0·91mg.	(32days old)	
0·41mg.	(2months old)	
1·7 mg.	(2months old)	

From the foregoing literature it is evident that, using using Folin's method fo the estimation of creatinin, it is found to be present in the urine of infants and also creatin may be present.

As regards the presence of creatinin in the urine of children, Closson found it present in a boy of 6yrs. and also in one of 14.

Two years ago I examined the urines of a number of hospital cases (Table II) and also a few normal cases, and from the results I got, I concluded at the time, that normally, children do not excrete creatin, but do so very readily when ill. In Table II we see that out of the 7 children, two did not excrete creatin. The amounts of creatin excreted are variable, the largest amount being got with a girl (12yrs. old) suffering from orthostatic albuminuria. Here one-third of the total creatinin (i.e. creatinin+creatin) is excreted as creatin. Another point of interest is the high percentage the urea-nitrogen forms of the total nitrogen. All those children were on a creatin-free diet and therefore would not be getting protein-rich diets, yet the

TABLE-II.

Total Urine	Total Nitrogen	Urea-N	Ammonia-N	Uric ac.-N	Creatinin-N	Creatin-N	Note
cc.	gms.	gms. %	gm. %	gm. %	gm. %	gm. %	
1186	9·46	-	-	-	0·135 (1·4)	0·065 (0·7)	R.C. orthostatic albuminuria. Girl. Age-12yrs. Cin.Cofft.6·3.
835	8·76	7·4 (85)	0·33 (3·7)	·070 (0·8)	0·135 (1·4)	0·065 (0·8)	
450	5·11	4·27 (83)	0·27 (5·3)	·033 (0·6)	0·08 (1·5)	0·002 (·04)	M.G. orthostatic album. Girl. Age-10yrs.
925	4·69	4·09 (87)	0·19 (4·0)	·036 (0·7)	0·064 (1·3)	0·019 (0·3)	
500	3·74	3·23 (86)	0·17 (4·4)	·030 (0·6)	0·062 (1·6)	0·015 (0·4)	
573	5·72	4·92 (86)	0·19 (3·3)	·056 (0·9)	0·114 (1·9)	0·030 (0·5)	
600	8·33	7·19 (86)	0·52 (5·4)	·059 (0·7)	0·150 (1·8)	0·011 (0·1)	D.G.T. mentally weak. Boy Age-11yrs. Cin.Cofft.5·0
1610	8·02	-	0·13 (1·6)	-	0·129 (1·6)	0	H. albuminuria, girl. Age-11yrs. Cin.Cofft.2·4
770	4·45	3·85 (86)	0·213 (4·7)	·033 (0·7)	0·048 (1·0)	0·013 (0·2)	McL. convalescent, boy. Age-7yrs.
1050	6·55	5·38 (82)	0·37 (5·6)	·054 (0·8)	0·090 (1·3)	0·045 (0·8)	J.S. Convalescemt chorea. Age-7yrs. Boy. C.Ct.3·0
610	5·47	-	0·15 (2·6)	·053 (0·8)	0·127 (2·3)	0	L. Conv.chorea, boy. Age-10yrs.
600	8·55	7·51 (88)	0·29 (3·4)	·107 (1·2)	0·168 (1·9)	0	

percentage of urea-nitrogen is fairly high; whilst the creatinin-nitrogen when compared to the total nitrogen excretion is rather low.

Amberg and Morrill, in the analysis of the urine of an infant (13days old), got the following figures:-

Total nitrogen	=	0·411gm.
Urea-nitrogen		91%
Ammonia-nitrogen		4·1%
Creatinin-nitrogen		2·7%

Here also the urea-nitrogen forms a high percentage of the total nitrogen, whilst the creatinin-nitrogen is somewhat low. They conclude that this is because of the rich protein diet of the infant, which would cause a high urea-nitrogen percentage, and a low one for creatinin. This is however not so, for as Rubner out from the analyses of Hoffmann and of Camerer and Soldner, the protein content of the milk is low, in fact unusually poor in proteins (1·03% protein in the milk as compared to the old figure of 3%). This would mean that the caloric value of the protein of the milk is about 7·8 to 8·9 calories, or 50% less calories than formerly estimated. Further Camerer and Soldner and Munck show that in spite of the small nitrogen content of the mother's milk, a large amount of this nitrogen however does not belong to protein.

From this it will be evident that Amberg and Morrill's explanation is not a correct one.

The results obtained in Table II are somewhat indefinite in regard to the creatin excretion, and as I got

no creatinuria in two normal boys of 8 and 14 years of age, I decided to extend my observations on normal children, trying to get where possible, a number of children out of one family, for there the conditions would be more comparable, and any variation due to any external cause would be common to each.

Whilst engaged on this piece of research a number of papers appeared on the subject, but instead of throwing light upon it, they have only confused it still more, by the small number of cases examined and by their not keeping the children on a creatin-free diet.

Schwarz examined one normal boy, aged 5yrs., and found no creatin in the urine; whilst in a number of rickety children, creatinuria was always present. He concludes from his observations that normally, children do not excrete, creatin, but that creatinuria is always present in rickets.

Rose who examined the urine of 19 boys and 20 girls, ranging between 1½ and 21 years of age, found creatin in the urine of all the children except two. His results are however of no value as he does not know what food they were getting.

MacCrudden examined the urine of a number of cases of infantilism and found creatin present in each case. From his results he concludes that the metabolism of creatin and creatinin are independent of each other.

Wolf criticises the results of Rose owing to the quality of the food not being known, and supports Schwarz but brings forward no new evidence regarding the subject.

In a recent communication, Folin and Denis confirm the occurrence of creatin in the urine of children. Some ~~of~~ cases were not on a creatin-free diet however. They consider the presence of a creatinuria in children as remarkable, because as they say, "it only occurs in children and does not correspond to what is found in older people. In the latter it only occurs when creatin is taken in the food or when there is an unusual disintegration of tissue metabolism". This view cannot be held any longer as I will show later, for adults do excrete creatin under definite physiological conditions.

The results which I got in the following research were got from children, all of whom were apparently healthy at the time of examination, in only one case (G.M.) was the child not well and a few days later developed influenza. His urine was however examined at a later date. All those children were put on a creatin-free diet one or two days before the urine was collected(24hrs. specimens). The urine was at once examined and not allowed to stand for any length of time before being analysed. In Tables- III-IV-V, I have given the complete distribution of the urinary nitrogen, and the percentage contributed by each constituent to the total nitrogen. The members of a family are all grouped together. In Tables VI, VII I have taken all the girls and all the boys, and have arranged them according to their ages, starting with the youngest and going up. In these tables I give the figures for the total creatinin-nitrogen and creatin-nitrogen as well as the percentage they form of the total nitrogen.

TABLE- III

Urine	Total -N	Urea -N	Ammonia -N	Uric ac. -N	Creatinin -N	Creatin -N	Cin-N Cofft	Subject	Age	Sex	Weight
cc.	gms.	gms. %	gm. %	gm. %	gm. %	gm.%				yrs	kilos
680*	4·19	3·43 82	-	·022 ·5	·086 2·0	0	-	R.P.	12	Boy	-
655*	3·56	3·12 87	-	·040 1·1	·075 2·1	0	3-5	R.P.	11	Girl	35
540	4·12	3·36 81	·254 6·0	·076 1·8	·123 3·0	0	3·5	..	..	..	..
500*	2·33	2·03 87	-	-	·032 1·4	·003 .14	-	B.P.	6	Girl	18
201	1·45	-	-	-	·062 4·3	·012 .8	4·1	..	..	..	..
470	4·51	3·77 83	·271 6·0	·047 1·0	·079 1·7	·004 .08	..	..	..	..	..
500	4·20	3·39 81	·180 4·2	-	·069 1·6	·005 .11	-	M.G.	5	Girl	21·7
500	3·70	3·04 81	·170 4·7	-	·084 2·2	·009 .24	3·8	..	..	..	..
1005	6·05	5·03 83	·270 4·4	-	·134 2·2	0?	3·6	E.G.	10	Boy	36·7
500	3·9	3·16 81	·290 7·5	-	·135 3·4	0	..	..	..	..	..

* Not complete 24hrs. specimens.

Cin-N Cofft. = Creatinin-Nitrogen Coefficient.

TABLE IV

Urine	Total -N	Urea -N	Ammonia -N	Creatinin -N	Creatin -N	Cin-N Cofft.	Acidity	NH_3+ amido ac -N	Subject	Age	Sex	Weight
cc.	gms.	gms. %	gm. %	gm. %	gm. %		cc.	gm.		yrs.		kilos.
825	8·5	7·38 87	·365 4·3	·147 1·7	·020 ·23	4·7	–	–	E.Mk.	10	Girl	31
550	5·62	4·68 82	·369 6·5	·162 3·0	·015 ·26	5·2	220	·400	..	..	..	..
830	9·75	8·47 87	·427 4·3	·157 1·6	·066 ·67	6·2	–	–	D.Mk.	7+	Girl	25
450	4·51	3·79 84	·403 8·5	·126 2·8	·030 ·66	5·0	156	·411	..	..	..	..
510	8·68	–	·386 4·4	·201 2·3	·011 ·12	5·6	–	–	N.Mk.	12	Girl	36
470	4·58	–	–	·190 4·1	tr.	5·3	–	–	..	..	..	..
1000	6·70	–	·516 7·7	·224 3·3	·042 ·6	6·2	268	·716	..	..	..	..
600	5·54	4·08 74	·436 7·9	·208 3·8	·010 ·18	5·6	214	·470	..	..	..	..
500	8·34	–	·434 4·3	·246 2·9	· 0	5·2	–	–	H.Mk.	13+	Boy	47
925	6·14	4·68 76	·491 8·0	·268 4·3	0	5·7	230	·544	..	..	..	..
300	3·00	2·52 83	·163 5·4	·025 ·86	·013 ·43	–	–	–	H.A.	2	Boy	–
310	2·64	–	·120 4·5	·029 1·0	·009 ·34	–	–	–	H.P.	3	Boy	–
580	5·48	–	–	·057 1·0	·022 ·4	–	–	–	–	–	–	–
365	2·32	–	–	·024 1·0	·0098 ·4	–	–	–	–	.	–	.

TABLE V.

Urine	Total -N	Urea-N	Ammonia -N	Uric ac.-N	Creatinin -N	Creatin -N	Acidity	NH_3+ AA-N	Subject	Age	Sex
cc.	gms.	gms. %	gm. %	gm. %	gm. %		cc.	gm.			
1150	11·97	10·24 85	·46 3·8	·118 0·9	·0378 3·1	0	271	0·64	A.Ms.	15	Boy
650	7·94	6·91 87	·31 3·8	·103 1·3	·178 2·2	0	247	·38	M.Ms.	8	Boy
710	7·20	6·35 88	·15 2·1	·074 1·0	·163 2·2	0	–	–			
460*	5·31	–	–	–	·157 2·9	0	–	–	A.Ms.	12	Boy
500	5·74	4·55 79	·344 6·0	· –	·225 3·9	0	–	–			
620	6·10	5·18 85	·215 3·5	·066 1·0	·112 1·8	·025 0·4	161	·31	G.Ms.	5	Boy
740	6·22	5·25 84	·256 4·1	–	·124 2·0	·028 ·45	–	–			
500	4·50	3·58 80	·196 4·3	· –	·101 2·2	·021 ·46	–	–			
625	8·10	6·97 86	·455 5·6	·175 2·1	·268 3·3	0	364	·56	J.McD.	16	Boy
1120	11·7	9·67 82	·533 4·5	·174 1·4	·374 3·2	0	520	·61			
875	5·75	5·03 87	·240 4·1	·136 2·3	·136 2·3	0	189	·30	N.McD.	7	Boy
950	9·10	8·19 90	·308 3·2	·118 1·3	·189 2·1	0	342	·45			

* Some of the urine was lost. NH_3+AA-N= Ammonia + Amido acid-Nitrogen.

When we look at Tables- III-V we find, as in the hospital cases, the percentage of nitrogen excreted as urea tends to be high; more especially is this the case with the girls. The ammonia nitrogen in some of the cases is also somewhat high. The low ammonia excretions were got in cases with a tendency to be stout. In some, the percentage of uric acid nitrogen tends to be high as compared to adults. This may be due to a rapid nucleo-proteid metabolism. On looking back at Table-II, it will be seen that the uric acid excretion of the hospital cases is a somewhat low one.

As regards the excretion of creatinin and creatin I shall first deal with creatin. When TablesIII-V are examined it is seen that some children excrete creatin and others do not. In the case of the children P., neither the boy(aged 12) nor a girl (11yrs) had any creatinuria, whilst the youngest girl(aged 6)had. With the four brothers (M)only the youngest aged 5yrs. excreted creatin. In the family (Mk) consisting of three girls and one boy, all the girls excreted creatin. Only in the case of the eldest girl (N.aged 12yrs.) was there only a trace on one occasion. The girl N. had her urine tested on four different occasions, with an interval of about one week between the two first and the two last specimens, and about three weeks separating the second from the third. The amount of creatin excreted varied somewhat. The excretion of creatin was very like that in the case of menstruation (as I shall show later). The girl had not started to menstruate, but it is quite likely that she was nearing puberty, being a Semitic (in whom puberty very often develops earlier than in the Anglo-Saxon races). There may be another explan-

TABLE VI (GIRLS)

	Case	Age yrs.	Creatinin-N gm.	Creatin-N gm.	Cin-N Cofft.
1.	A.D.*	2	0·006	0·004	-
2.	M.G.	5	0·069	0·005	3·8
			0·085	0·009	
3.	B.Pl.	6	0·032	0·003	4·1
			0·062	0·012	
			0·079	0·004	
4.	D.Mk.	7+	0·158	0·066	6·2
			0·126	0·030	5·0
5.	E.Mk.	10	0·147	0·020	4·7
			0·162	0·015	5·2
6.	R.Pl.	11	0·075	0	3·5
			0·123	0	
7.	N.Mk.	12	0·201	0·011	5·6
			0·190	tr.	5·3
			0·224	0·042	6·2
			0·208	0·010	5·6

* Amount estimated in the specimen (not for 24hrs.)
the total nitrogen in sp. was ·45gm.

TABLE VII (BOYS)

	Case	Age yrs.	Creatinin-N gm.	Creatin-N gm.	
1.	H.A.	2	0·025	0·013	
			0·025	0·012	
2.	M.P.	3	0·029	0·009	
			0·024	0·009	
			0·057	0·024	
3	T.D. (only a specn.)	6	0·014	0·0009	
4.	G.Ms.	5	0·112	0·025	
			0·124	0·028	
			0·101	0·021	
5.	N.McD.	7	0·136	0·0	
			0·189	0	
6.	M.Ms.	8	0·178	0	
			0·163	0	Cin-N Cofft
7.	E.G.	10	0·135	0?	3·6
			0·135	0	
8.	R.Pl.	12	0·086	0	
9.	A.Ms.	12	0·157	0	
			0·225	0	
10.	M.Mk.	13+	0·246	0	5·2
			0·268	0	5·7
11.	A.Ms.	15.	0·378	0	
12.	J.McD.	16	0·268	0	
			0·374	0	

Cin-N Cofft.= Creatinin Nitrogen Coefficient.

ation for this. As I shall show later creatinuria is always present where there is a disturbance of the carbohydrate metabolism. Now it has been shown by v.Noorden and others that the Jews as a race are more afflicted with diabetes, than any other race. If that be so, then might not the creatinuria in this case at this age be due to a latent tendency towards a disturbance of the carbohydrate metabolism?

In the case of the brother and sister (G.) the girl(5) had a creatinuria, whilst the boy (aged 12yrs.) has not. The query in the creatin column of the boy means that there was a trace, but the amount was so much within the limits of error that it could not be looked upon as positive. In the case of the two brothers (MacD.) aged 7 and 16 no creatinuria was present in either.

When we examine those Tables(III-V) the first impression received is that children may or may not excrete creatin. But on closer examination it is found that boys after a certain age stop excreting creatin whereas girls continue to do so for a longer time. To make this perfectly evident, I have taken the creatinin and creatin figures of the boys and girls separately and arranged them chronoligically in Tables- VI and VII.

When the figures for the girls are compared with those of the boys it is at once seen that the creatinuria of the latter ceases apparently at about the age of 5 or 6, whereas with the girls the creatinuria continues till about or even after 10 years. The case N.Mk.is probably exceptional and the creatinuria here present is possibly due to puberty, even though there were no external signs, such as, menstruation.

At whatever age the creatinuria ceases in girls it certainly is at a later date than in the case of boys. That it does cease in girls is shown by the case R.P.(aged10 yrs.) here the urine was examined on two different occasions but each time no creatin was found.

Does the creatinuria in children show any gradual diminution as age advances? When we look at the absolute figures for the creatin in the tables, there is no evidence to show that there is any gradual transition from the state of creatinuria to one without and the disappearance of the creatinuria seems to be quite an abrupt one. The exact time when it takes place probably depends upon the condition of the individual; with healthy children it probably disappears more readily than in those who are ailing, yet the three Jewish girls were all strong and healthy. A factor which probably influences this creatinuria is the action of certain internal secretions (e.g. thyroid), but this aspect still requires to be worked out.

If we examine the figures for the creatinin+ creatin nitrogen (known as the total creatinin)nitrogen) and see what percentage of the total is constituted by the creatin, we find that in the younger children the percentage of ''total creatinin'' excreted as creatin is higher than in the older children with creatinuria

When Table VIII is examined we find that at the age of two, the percentage of total creatinin excreted as creatin is 34%, whilst at 5 years of age (the last boy with creatinuria) the percentage is only 6.

TABLE VIII(Boys)

age	percentage of total creatinin as Creatinin	Creatin.
2	66%	34%
3	77%	23%
	72%	28%
	73%	27%
5	82%	18%
	82%	18%
	83%	17%
	94%	6%
7	100%	0

When we look at the figures presented by the girls, we find very much the same thing.

TABLE IX (Girls)

age yrs.	percentage of total creatinin as Creatinin	Creatin.
2	65%	35%
5	93%	7%
6	90%	10%
	91%	9%
	84%	16%
7	72%	28%
	81%	19%
10	88%	12%
	90%	10%
11	100%	0
12	96%	4%
12	99%	1%
	85%	15%
	95%	5%

If we put out of count the three sisters (Jews, 7, 10 and 12), we find that there is the same diminution in the percentage of the total creatinin excreted as creatin as in the

case of the boys. The figures at the age of 2 being 35% and at 6 between 15 and 16. There is a certain variation, but broadly speaking, the figures are similar in both cases. As already indicated this may quite well be a racial characteristic. In the case of the eldest of the three girls, the variations in the percentage show a kind of periodicity, similar to what one gets after puberty due to menstruation.

If we now look at the creatinin excretion in the children, we find that there is a gradual increase in the absolute amount of the creatinin as the children get older. This is to be expected as they gain in weight and their muscles become more developed. The increase in the creatinin excretion, is out of proportion to the gain in body weight, so that the amount of creatinin nitrogen in milligrams per kilo of body weight (or the creatinin-nitrogen coefficient) is greater in children of 12yrs. of age than in those only 5years old.

The creatinin-nitrogen coefficient for adults according to Shaffer is 8·1 and upwards, the same figure as I got for normal adults in Table XI . According to Folin the chief factor determining the amount of creatinin eliminated, appears to be the weight of the person. The amount of fat must however be noted. The fatter the person the less creatinin will he or she excrete per kilo of body weight. He concludes that the amount of creatinin excreted primarily depends upon the amount of active protoplasm. Shaffer also emphasises the importance of the varying varying degrees of muscular development and more especially the muscular tonus

as affecting the creatinin elimination. This is also indirectly in accordance with the view expressed by Pekelharing and Hoogenhuyze regarding the effect of muscle tonus on the creatin excretion. Another view is held by Benedict and Myers who hold that the creatinin excretion is proportional to the body weight only and not to the active mass of protoplasmic tissue.

When we compare the creatinin coefficient of the adult, namely 8·1 to that of the child, it is found that in the the latter it is a good deal below this figure. In fact at the age of 5 or 6 years, the creatinin coefficient of a child is just half that of an adult. At the age of 12years the ratio of the creatinin nitrogen to one kilo of body weight has risen to about 6. The increase in the creatinin coefficient is more marked in young than in older children.

In some young rabbits (2months old) examined I noticed the increase of the creatinin coefficient even within three weeks. In those animals the increase of weight is also more rapid than in the case with human beings.

	Weight	Creatinin-N	Creatin-N	Cin-N Cofft.
I	607gms.	·0047gm.	·0014gm.	7·7
II	710	·0047	·0003	6·6
III	679	·0043	·0003	6·3

I	845gms.	·0079gm.	·0014gm.	9·3
II	890	·0072	·0006	8·0
III	855	·0067	·0018	7·8

Amberg and Morrill in their paper also got very low creatinin coefficients for infants (7-14days). Their fig-

ures are of interest as they show a rapid increase in the creatinin per kilo of body weight at that early period. Their figures are.-

	Age	Cin-Cofft.*
1.	7days	5·4
2.	10 ..	6·7
3.	10 ..	7·78
4.	13 ..	9·7
5.	14 ..	9·9

Their figures for the creatinin coefficient compared to Folin's (20-24) or Shaffer's (18-30) for adults, are found to be from one-third to one-fifth of that of the adult.

From the preceding it will be evident that the amount of creatinin per kilo of body weight is very low in the infant and it quickly increases in early childhood, and more gradually in later childhood.

I conclude that my figures for the creatinin coefficient support Shaffer's view of the importance of the muscular development and the muscular tonus as affecting creatinin excretion.

Folin in his paper (1905) showed that the excretion of creatinin per diem for each individual was constant, provided his diet was creatin-free. The food could contain large or small quantities of protein and yet no alteration in the amount of creatinin excreted would take place. This also applies to children, for if the three cases N.Mk. and G.M. and E.G. are referred to, one sees a constancy of the creatinin even although the specimens were not got closely following each other, but were collected at different time

* Cin-Cofft.=Creatinin coefficient, i.e. milligrams of creatinin per kilo of body weight.

intervals.

In the case of N.Mk. the figures are.-

Date	Cin-N
2·6	0·20gm.
10·6	0·19gm.
1·7	0·22gm.
8·7	0·21gm.

The figures show that the creatinin excretion is constant from day to day. Of course I do not wish to overlook the point that there is a gradual, but a comparatively slow increase of the creatinin excretion as the child grows older, but the increase at this age is not perceptible from day to day, hardly month by month, indeed.

In the case of the boy G.M.(aged 5) and the boy E.G. (aged 10) the figures are.-

	Date	Cin-N
G.M.	15·2	0·11gm.
	12·7	0·12gm.
	13·7	0·10gm.
E.G.	7·10	0·13gm.
	14·10	0·13gm.

b. CREATIN INGESTED BY CHILDREN

From the foregoing observations it is evident that normal children do excrete creatin. It is therefore also of interest to examine their behaviour towards creatin given with the food - in particular whether they are able to metabolise it or excrete it unchanged.

A number of observers have carried out experiments on the subject of the ingestion and excretion of creatin or creatinin by the adult. In 1868 Meissner published results, in which he showed that creatinin and also creatin when ingested or subcutaneously injected, were both totally (or nearly so) recovered in the urine, and in the form of creatinin. C. Voit also in 1869 feeding a dog with 8·6gm. of creatin with the food, recovered 4·2gm. of creatinin and 3·2gm. of creatin in the urine. Mallet in 1900 concluded from his experiments that the human body possessed a practically unlimited capacity for manufacturing and eliminating creatinin from creatin absorbed from the digestive tract.

More recent results have differed from the foregoing. Mendel stated that the amount of creatinin excretion bears a possible relationship to the quantity of protein metabolised. He makes his comparisons from different cases, and so the variations which he got in his tables may be due to individual differences in weight, sex, age etc.

Achaelis carried out experiments on men and dogs, giving large quantities of creatinin with the food and from his results he concluded that a large quantity of the ingested creatinin was destroyed. V. Klercher failed to find any indication that creatin given with the food is converted into creatinin before being eliminated.

He took large quantities of meat and found no apparent increase in the creatinin excretion. He holds that the urinary creatinin is of endogenous origin, as no creatinin was present in the food but a change from creatin to creatinin did not take place. A connection between the urinary creatinin and the creatin of the muscle is not probable; more likely it is formed in the general protein metabolism.

Folin in 1905 stated that creatin may be one of the nitrogenous substances which serve to maintain the nitrogen-equilibrium in the living body and which do not easily take part in the urea forming processes: he also suggested that it probably belongs to the endogenous metabolism, and is analogous to uric acid, which he found was likewise unaffected by a protein rich diet, a conclusion in which he agrees with Burian and Schur, as well as with Siven.

Folin (1906) as a result of various lines of inquiry, concludes that creatin in contradistinction to creatinin is a food and not a waste product. He adds that the results of feeding experiments with creatin, depend largely upon the character of the food. With a

TABLE X (Creatin Ingestion)

Urine	Total -N	Urea -N	Ammonia -N	Creatinin -N	Creatin -N	Uric ac.-N	Acidity	Ammonia + amino ac. -N	Subject	Age	Weight
cc.	gms.	gms. %	gm. %	gm. %	gm. %	gm. %	cc.	gm.		yrs.	kilos
650	7·94	6·91 87	·309 3·8	·178 2·2	·004 ·5	·103 1·3	347	·381	M.Ms.	8	-
710	7·20	6·35 88	·152	·163 2·2	·00	·074 1·0	139	·215			
740	5·30	-	-	·147 2·7	·042 ·79	·-	139	-			
620	6·10	5·18 85	·215 3·5	·112 1·8	·025 ·4	·066 1·0	161	·306	G.Ms.	5	-
330	3·35	-	·-	·063 1·8	·047 1·4	-	108	-			
740	6·22	5·25 84	·256 4·1	·124 2·0	·028 ·4	·-	-	-			
500	4·50	3·58 80	·196 4·3	·101 2·2	·021 ·4	-	-	-			
645	3·56	-	·-	·075 2·1	·0	·040 1·0	90	-	R.P.	11	35
540	4·12	3·37 82	·253 6·1	·123 3·0	0	·076 1·8	192	·330			
690	3·42	2·51 73	·247 7·2	·126 3·7	·032 ·9	·065 1·9	138	·305			
470	4·51	3·77 83	·271 6·0	·079 1·7	·004 ·09	·047 1·0	158	·306	B.P.	6	18
510	2·39	1·81 76	·206 8·6	·075 3·1	·060 2·5	·045 1·8	57	·248			

* Got 50cc.creatin soln.,comtaining ·098gmCreatin-Nitrogen and ·007gm Creatinin-N

** Got 50cc.creatin soln.,containing ·103gm.Creatin-N and ·007gm.Creatinin-Nitrogen.

TABLE XI (Creatin ingestion)

Urine	Total -N	Urea -N	Ammonia -N	Creatinin -N	Creatin -N	Acidity	Ammonia + amino ac.-N	Subject	Age	Weight	Cin-N
cc.	gms.	gms. %	gm. %	gm. %	gm.	cc.	gm.		yrs.	kilos	Cofft.
1300	10·20	–	·538 5·2	·541 5·2	0	317	··606	R.	26	67	8·3
1200	9·47	–	·524 5·5	·570 6·0	0	360	·687				
1400*	10·35	–	·659 6·3	·592 5·7	0	–	–				
1400	10·15	–	·624 6·6	·552 5·4	00	372	·902				
1460	3·27	1·83 56	·245 7·5	·485 14	0	228	·344	R.	25	66·5	7·2
1400**	3·14	1·39 44	·250 8·0	·498 15	0	168	·400				
1470	3·37	1·87 55	·263 7·8	·524 15	0	211	·428				
1760	3·45	2·03 59	·226 6·5	·515 14	0	218	·374				
1280	13·30	11·44 86	·574 4·3	·620 4·6	0	568	·674	B.	36	76	8·1
970***	12·07	10·14 84	·456 3·8	·610 5·0	0	–	–				

* 50cc.Creatin soln. given =0·126gm.Creatin-Nitrogen and ·002gm.Creatinin-Nitrogen.

** 40cc.Creatin Soln.given =0·079gm.Creatin-Nitrogen and ·001gm.Creatinin-Nitrogen.

*** 50cc.Creatin Soln.given =0·098gm.Creatin-Nitrogen and ·002gm.Creatinin-Nitrogen.

rich carbohydrate and fat diet, poor in proteins, a large amount of the ingested creatin was retained in the body than when a protein-rich diet was taken, and so the retention of the creatin when fed together with a nitrogen-poor diet, clearly indicates that creatin is not a waste product.

Klercker in 1907, as a result of experiments carried out with meat extract and pure creatin confirms his earlier as well as Folin·s views, that ingested creatin and creatinin are partially excreted by the kidneys as such, without any change. The only difference between the creatin of meat extract and pure creatin, is that when ingested none of the latter is recovered in the urine. This he was able to prove is due to protein pure diet. Wolf and Shaffer have confirmed these results by injecting creatin, and finding the excretion of creatinin wholly unaffected.

Weber tried feeding experiments with meat extract. He found that the increase of creatinin in the urine exceeded the amount given in the food, and that some of the creatin had been converted into creatinin. Hoogenhuyze and Verploegh (1908) recovered some of the ingested creatin as creatinin in the urine. Lefmann also made experiments on dogs with meat extract. When he gave a dog small amounts, the creatin and creatinin excretion was increased.

He agrees with Weber that the quantities of creatin and creatinin in meat extract are very variable.

He was not able to determine any conversion of creatin to creatinin, as the amount excreted corresponded with that ingested. He also shows that the creatinin excretion is controlled by factors other than those of uric acid and urea.

Twort and Mellanby in a recent communication describe a bacillus in the human faeces which has the power of destroying creatin; a number of other micro-organisms are described which have a similar power. Such being the case, they hold that the results of certain creatin feeding experiments cannot be properly valued without taking this factor into account. Mellanby in a previous paper (1907) had suggested the possibility of Folin's results being due to the action of intestinal bacteria. It seemed probable that, when the diet contained but little nitrogen, the intestinal flora lived at the expense of the creatin-nitrogen: and that on the other hand when plenty nitrogen was present in the diet, the creatin was untouched by the bacteria because more assimilable substances were present.

None of the previous investigators have, to my knowledge, tried creatin feeding experiments on children. I therefore carried out some observations on the subject, using comparatively small quantities of creatin. I considered that, if the children excreted creatin, then the additiom of small quantities would lead to an excretion of perhaps all the ingested creatin. I therefore did not use large quantities, but large enough to exclude any experimental error. For the experiments I chose two

sisters (P) and two brothers (M): the older of each sex was not excreting creatin. The children were put on a creatin-free diet during, as well as a few previous to, the experiment. I dissolved about 0·3 to 0·35gm. of creatin in 50cc. distilled water, and this quantity was given to each child, the first thing in the morning in the case of the boys (urine was collected from 10am. to 10am.) whilst the girls got theirs the last thing at night (the urine in this case was collected from the first passed in the morning to the last in the evening). The diet the children got was not a particularly proteid-rich diet, and consisted of porridge, milk, one or two eggs, bread and butter, potatoes and vegetables and some milk puddings.

The results as given in Table X show that creatin when given to children whether already excreting creatin or not, leads to an increased creatinuria (if creatin was already being excreted), or to a creatinuria where none existed previously). The amount so excreted when there was no previous creatinuria or the increased amount of creatin where there was already a creatinuria, leads to no other conclusion than that the experimental creatinuria originated in both cases from the ingested creatin. The two girls both got 103gm. creatin (as nitrogen): in the solution there was also ·007gm. creatinin (as nitrogen). Of the two girls the younger (6 yrs.) excreted 56mgms. of creatin (as nitrogen) more than she did on the previous day. This when calculated would mean an excretion as creatin of 54% of the ingested

creatin. In the case of the older girl (11) the same amount of creatin led to an excretion of 32mgms. of creatin nitrogen, which would mean that 31% of the ingested creatin was excreted as such.

The boys got a solution containing 0·098gm. creatin nitrogen, in which there was also a little creatinin present (representing about ·007gm. nitrogen)

The older boy did not previously excrete creatin, but when creatin was given him, about 43% of it was excreted in the urine as creatin. In the case of his younger brother (aged 5), (the evening portion was lost and so I did not get the whole 24 hours specimen, but from what there is,) there is also an increased excretion of creatin nitrogen. If we go by the creatinin only half of the available amount is present, if this also applied to the creatin then the amount of ingested creatin-nitrogen would probably work out at about 70% (estimating it in absolute figures at 69mgms. creatin-nitrogen).

This is certainly purely an assumption, but nevertheless, the case shows that a large amount of the ingested creatin was here also excreted.

Was any of the ingested creatin excreted as creatinin and was there a rise in the creatinin excretion? If we look at Table X and examine the creatinin figures, there is none in of the cases any apparent increase in the amount of creatinin excreted, so that in the case of the children certainly, although ingested creatin is in part

excreted again as creatin there is no evidence of any of this ingested creatin being converted into creatinin and excreted as such.

From an examination of Table XII we find, that the younger the child fed with creatin, the more does it excrete that creatin unchanged.

TABLE XII

Sex	Age	Percentage of ingested creatin excreted as creatin.
Boy	5 yrs.	(70%)
Girl	6 yrs,	56%
Boy	8 yrs.	43%
Girl	11 yrs.	31%

I have already shown that in the more recent literature it has been pointed out that creatin when given to adults is not excreted as such in the urine, nor is there any evidence of it having been converted into any particular constituent of the urine, for Folin failed to find any increase in any of the nitrogenous bodies after creatin had been given. As the evidence of some of the observers is rather conflicting I decided to carry out control experiments on adults. I gave similar quantities of creatin dissolved in 50cc. of water to two male adults, both being on a creatin-free diet and neither of them excreting creatin previous

to the experiment. The results I have collected together in Table XI.

The figures show that no creatin was excreted even after creatin was given with the food. The diet in neither case was a carbo-hydrate-poor diet, indeed the case B. had a diet which inclined somewhat to a protein-rich diet, and yet not a trace of creatin was excreted. Was any of the ingested creatin excreted as creatinin? If we look at case B. the answer is decidedly negative. In case K. there is a slight suggestion that a portion of the creatin may have been converted into creatinin and caused the slight rise noted on that day, but the quantity is so slight no definite conclusion is permissible.

On Table XI I give the figures of an experiment carried out on K. some time ago; on this occasion the diet was very poor in protein but contained plenty carbohydrates and fats. I gave 79mgm. of creatin nitrogen. Here again there was no creatin excreted, but as the rise in creatinin was so slight on the day on which creatin was given, and as it continued to rise for three days after (a total of·111gm. as against the possible 79mgm. of creatinin from creatin) one must come to the conclusion that the rise of creatinin here shown is probably not due to any ingested creatin but is rather a result of the diet fed to the subject.

That the conversion of creatin into creatinin is possible, however, is perhaps illustrated by another

case, the results of which are also given on the same Table (XI) as the last case.

The diet in this case (M) was a protein-rich one (including 4 or 5 eggs a day). The amount of creatin nitrogen given was ·103gm. + ·007gm. creatinin nitrogen. (Total nitrogen = ·110gm.) There is no creatin excreted, but there is a large increase in the creatinin, an increase which (·121gm. creatinin nitrogen) more than covers the amount which was available for that purpose by the ingested creatin. This is rather striking and I intend to repeat experiments using larger doses and in altered conditions. The explanation I would advance in this case, is, that it may be due to an intestinal condition. This patient had been troubled with spasm of the intestines last summer: and this was only removed after prolonged treatment with colon lavage.

This case is however interesting in the light of Twort and Mellanby's experiments, and may be an example, where owing to the large protein diet and to the presence probably of organisms, the creatin is converted into creatinin and excreted as such. Why no retention of creatin took place is due to the latter not being a food substance and so excreted in toto from the body. This latter conclusion, if correct, would point to the creatin having been converted into creatinin in the intestine before entering the body proper, for if the creatin had entered the tissues as creatin, even though there was plenty of protein present,

part of it would have been retained in the body and only part of it would have been excreted and then probably not as creatinin.

This experiment may therefore be of some interest in so far as by means of this method we may be able to throw some light upon the manner in which certain intestinal conditions may affect the creatin absorption; conversely, the examination of the urine for creatin and creatinin may become of diagnostic significance in these cases.

My results would therefore support the view that creatin when given in the food does not usually lead to an increased creatinin excretion, not even in those cases where creatin is already excreted normally (as occurs in children); but under certain abnormal conditions probably of an intestinal nature, creatin may be converted into creatinin, absorbed as such, and not being of any dietetic use to the organism, simply excreted.

PART II

THE PRESENCE OF CREATIN IN THE URINE OF ADULTS UNDER PHYSIOLOGICAL CONDITIONS

a. THE URINE OF NORMAL WOMEN DURING THE SEXUAL CYCLE

A few years ago whilst examining a number of urines of female patients in the Royal Infirmary, Edinburgh, I found that they always contained creatin, even although they were getting a creatin-free diet. I tried to find an explanation for it, but could not. I therefore decided to examine the urine of normal females, also on a creatin-free diet. The first specimens I examined contained no creatin, but in the next specimen I found creatin present. From this it seemed that women may or may not have a creatinuria. But what brought it about? As the last specimen was got about a week after menstruation, I decided to investigate the subject from the point of view whether menstruation had any relationship to the creatinuria or not.

The following analyses were carried out on the urines of two normal women, which were examined about once a week or a fortnight for a continued period of /

TABLE XIII.

Date	Urine	Total -N	Urea -N	Ammonia -N	Uric acid -N	Creatinin -N	Creatin -N	Acidity in cc. $\frac{N}{10}$ NaOH	Percentage of undetermined -N	Notes.
	cc.	gms.	gms.	gm.	gm.	gm.	gm.			
12·1	1275	8·568	7·45 (87)	·292 (3·4)	·123 (1·4)	·330 (3·8)	0	159	4·4	Case—F.M., unmarried.
13·1	1600	7·213	6·01 (83)	·214 (2·9)	·102 (1·4)	·297 (4·1)	0	144	8·6	Age—40yrs. Wt.—59·7kilos.
26·1	1500	8·98	7·56 (84)	·621 (6·9)	·135 (1·5)	·363 (4·0)	·015 (·17)	363	3·5	Menses on 21st.—22nd. January.
27·1	1200	7·79	6·38 (73)	·711 (9·1)	·104 (1·3)	·356 (4·5)	·019 (·24)	348	12·0	
2·2	1500	8·27	6·54 (79)	·595 (7·0)	·079 (0·9)	·328 (3·9)	·016 (·19)	540	17·0	
9·2	1500	8·94	—	·570 (6·3)	·104 (1·1)	·291 (3·3)	·016 (·18)	210	—	
10·2	1600	7·30	5·59 (76)	·366 (5·0)	·092 (1·2)	·280 (3·8)	·014 (·19)	186	13·3	
16·2	1500	7·56	5·26 (70)	·453 (5·9)	· —	·341 (4·5)	·022 (·29)	180	20·0	Constipated lately; headaches.
24·2	1700	6·85	5·40 (79)	·447 (6·5)	·071 (1·0)	·296 (4·3)	·015 (·2)	170	9·2	Slight menses on 22nd. February
9·3	1300	8·70	6·62 (76)	·440 (5·1)	· —	·349 (4·0)	·016 (·19)	237	14·6	Menses on 5th.
8·4	1500	8·44	—	·507 (6·0)	—	·349 (4·1)	0	247	—	
31·5	1500	6·80	5·07 (74)	·469 (6·9)	—	·378 (5·4)	trace	258	13·2	Menses on 1st.—3rd. June.
8·6	1400	4·86	—	·320 (6·5)	· —	·332 (6·8)	·017 (·35)	190	—	
9·6	1400	8·35	—	·455 (5·4)	—	·403 (4·8)	·012 (·1)	280	—	

of time (several months). The subjects of the experiment were always on a creatin-free diet for two days previous, the diet consisting of porridge and milk, bread and butter, vegetables, eggs, milk puddings tea, coffee or cocoa, but was not restricted in quantity. This diet was also given on the day of collection. All the analyses were carried out at once on receipt of the twenty-four hours specimen.

In each of the Tables XIII-XV , the results are given both as absolute amounts and relatively as to the proportion of the total nitrogen execreted in the form of the several substances. The absolute amounts are expressed in terms of the absolute amounts of nitrogen excreted in the form of those substances.

In Tables XIII-XV , it is seen that the urine of women differs from that of men qualitatively. This qualitative difference consists in the frequent presence of creatin in the urine of women.

It is always present after menstruation. It may be absent or present only in traces, two or three weeks after that period, but sometimes persists right through the menstrual period, especially when the menses are irregular. This is seen in Table XIII (case F.M.) during the period Jan. 23rd to Feb.24th.

The ammonia excretion shows similar, although not necessarily corresponding, variations. This is

most /

TABLE XIV.

Date	Total urine	Total -N	Urea -N	Ammonia -N	Uric acid -N	Creatinin -N	Creatin -N	Acidity in cc. $\frac{N}{10}$ NaOH	Percentage of undetermined-N	Notes.
	cc.	gms.	gms.	gm.	gm.	gm.	gm.			
16·1	1600	14·82	11·71 (79)	·697 (4·7)	·175 (1·1)	·427 (2·8)	·042 (·28)	488	12·2	Case–E. unmarried. Age–22 Wt.–70kilos.
22·1	1075	8·91	7·74 (87)	·324 (3·6)	·099 (1·1)	·243 (2·7)	·006 (·07)	252	6·0	Menses on 12–14th. Jan.
23·1	1320	9·50	7·36 (77)	·398 (4·1)	·106 (1·1)	·278 (2·9)	·018 (·19)	290	14·2	Menses irregular
30·1	1350	11·72	9·42 (80)	·672 (5·7)	·127 (1·0)	·335 (2·8)	·009 (·08)	370	10·5	
5·2	1500	8·90	5·82 (65)	·478 (5·3)	·106 (1·1)	·270 (3·0)	·012 (·13)	210	25·5	
6·2	3000	12·78	8·92 (70)	·570 (4·4)	· –	·336 (2·6)	trace	300	23·0	
26·2	(1000)	(6·44)	–	(·386) (5·9)	–	(·189) (2·9)	(·007) (·1)	(100)	–	Menses 18th–20th. Feb. (not complete spec.)
12·3	1700	13·04	10·25 (78)	·693 (5·3)	–	·401 (3·0)	·016 (·1)	314	13·0	
17·4	1300	8·62	6·82 (79)	·530 (6·1)	–	·363 (4·2)	·011 (·13)	292	10·6	
29·5	1000	6·55	5·03 (77)	·319 (4·7)	–	·302 (4·6)	0	232	14·0	Menses 2 weeks previou
5·6	1500	6·89	–	·352 (5·1)	–	·372 (5·4)	0	258	–	
12·6	1500	9·78	8·17 (83)	·436 (4·4)	–	·382 (3·9)	·008 (·08)	384	8·0	
25·6	1700	8·43	6·46 (77)	·484 (5·7)	–	·401 (4·7)	·017 (·2)	265	12·7	Menses 16–20th. June.
19·6	(500)	(3·41)	–	(·165) (4·8)	–	(·154) (4·5)	(·006) (·19)	(118)	–	Specn. passed during menstruation, sanguineous.

most apparent in the Case F.M., where the total nitrogen excretion remained throughout on a fairly constant level, and where therefore, both the absolute and the relative values for the ammonia excretion, which are to a great extent dependent upon the amount of total nitrogen metabolised, are directly comparable with each other. In this case the ammonia-nitrogen rises from 0·2 and 0·3gm.(=2·9 per cent. and 3·4 per cent.) excreted in the first intermenstrual period (Jan.12th-13th) to 0·6 and 0·7gm. (=6·9 per cent. and 9·1 per cent respectively) immediately after menstruation (Jan. 26th-27th), and falls again to 0·36gm. (5 per cent.) on Feb.10th. The subsequent rise is less pronounced and menses are irregular.

In Case E. (Table XIV.) the menses were irregular and the nitrogen excretion varied a good deal, so that it is not possible to draw definite conclusions from this case. Even in this case however was the ammonia execretion variable. According to Folin in a normal male the proportion of total nitrogen excreted as ammonia falls with an increase in the amount of total nitrogen excreted, other conditions remaining the same, in the above case there is a rise. On Jan. 30th, for instance, 5·7 per cent. of 11·7gms. of total nitrogen are excreted as ammonia, while a week previously, on Jan. 22nd, with a lower total nitrogen excretion of 8·9gms. only 3·6 per cent. were excreted as /

as ammonia nitrogen.

In Table XV, I have given the analyses also of another normal case (H.M., a multipara). The ammonia nitrogen per centage was as high as 7·9 per cent. immediately after menstruation on May 1st., with a total nitrogen excretion of 12·3gms. Twelve days later the percentage of ammonia nitrogen had fallen to 5.2 per cent., the total nitrogen excretion being 12.2gms.

The rise in the ammonia excretion is accompanied by a rise in the excretion of undetermined nitrogen and a fall in the urea excretion.

As already shown on pp. 21 Folin and others have found that the daily excretion of creatinin for an individual is constant; this also applies to female subjects. The case E. on Table XIV shows fluctuations, however, which I cannot explain. Also in Case F.M. (Table XIII) there is a distinct subnormal creatinin excretion on two successive days (Feb. 9th-10th). In interpreting these results it must be borne in mind that in the case of these women my observations were made at intervals during a prolonged period extending over half a year, and not during a short period of successive days, as is usually the case. That this explanation is correct, is borne out by the result of analyses carried out on case E. between the 11th and 19th of July, when a series of observations were made for / (Table XV.)

TABLE XV.

Date	Total Urine	Total -N	Urea -N	Ammonia -N	Uric acid -N	Creatinin -N	Creatin -N	Acidity in $\frac{N}{10}$ cc. NaOH	Notes.
	cc.	gms.	gms.	gm.	gm.	gm.	gm.		
11·7	1450	9·98	7·93 (79)	·406 (4·3)	–	·432 (4·3)	·0	435	Case-E. (contin.) Wt.=67kilos.
12·7	1275	9·07	6·77 (75)	·364 (4·0)	–	·458 (5·2)	0	316	
14·7	1300	9·43	7·77 (83)	·517 (5·4)	– –	·454 (4·8)	trace	374	
15·7	1250	8·68	6·72 (77)	·532 (6·1)	–	·409 (4·7)	0	375	
16·7	1600	9·18	7·45 (81)	·474 (5·2)	–	·498 (5·4)	0	352	
17·7	1480	8·46	6·19 (73)	·438 (5·2)	·110 (1·0)	·438 (5·1)	·009 (·16)	325	
18·7	1360	8·83	6·96 (78)	·389 (4·4)	·105 (1·1)	·414 (4·6)	·0	287	Menses began on the 18th. July.
19·7	1050	7·64	6·04 (79)	·411 (5·2)	· –	·379 (4·9)	·019 (·24) (24)	226	
25·7	925	9·14	8·03 (88)	·477 (5·2)	–	·368 (4·0)	·017 (·18)	303	
26·7	1650	10·07	6·92 (68)	·611 (6·1)	–	·454 (4·5)	·012 (·12)	442	
1·5	1700	12·29	9·16 (74)	·979 (7·9)	·145 (1·1)	·644 (5·2)	·014 (·11)	552	Case-H.M. Married woman, Aged 40. Miscarriage 2months ago, Apparently normal. Menses 28th. April. Wt.= 87kilos.
12·5	1800	12·19	9·19 (75)	·635 (5·2)	·148 (1·2)	·651 (5·3)	·015 (·12)	535	

for several successive days, here the creatinin, apart from slight variations, is fairly constant. That variations also occur in males when observations are carried out over a long period of a year or two, I shall be able to show later on.

Benedict and Myers have shown that the creatinin nitrogen coefficient (i.e. the number of milligrams of creatinin nitrogen per kilo of body weight) in women is lower than in men. In the latter it ranges between 7 and 11, whereas in my cases it ranges between 3·1 and 7·4. My results, therefore, confirm the observations of Benedict and Myers. I have tabulated my results in Table - XVI.

TABLE XVI.

Case	Creatinin-N	Creatinine-Coefficient
1	0·330	5·5
	0·291	4·6
2	0·427	6·0
	0·243	3·5
3	0·644	7·4
4	0·195	4·0
5	0·173	3·3
6	0·164	3·1

The following figures are from a bitch during the oestrous and anoestrous periods, The dog was fed on bread and milk for several days before the urine was examined for creatinin and creatin. Since in these

preliminary observations I wished to determine qualitatively the presence or absence of creatin, the total volume of urine was not collected, and the figures given below represent the amount of creatinin or creatin expressed as nitrogen excreted in a sample of 20 c.c. of urine. To each of the specimens collected, chloroform and thymol were added as a preservative. The analysis was, however, performed immediately.

TABLE XVII

Date	Creatinin-N	Creatin-N	Period
	mgms.	mgms.	
21:5:11	2·502	·319	Oestrous
22:5:11	2·502	·511	do.
23:5:11	2·708	·383	do.
24:5:11	1·919	1·959	do.
25:5:11	1·883	·482	do.
17:6:11	4·66	0	Anoestrous
18:6:11	4·225	0	do.
19:6:11	5·16	0	do.

I also append (Table XIII) some observations on the same dog in which the total volume of urine was collected. This table, therefore, gives the total quantities excreted per diem expressed in terms of nitrogen; it also gives the percentage of this nitrogen to the total nitrogen. The weight of the dog was 7·7 kilos.

TABLE XVIII

Date	Total nitrogen	Creatinin-N	Creatin-N	Period
	gms.	gm.	gm.	
20:6:11	4·865	·081 (1·6%)	0	Anoestrous
7:7:11	3·920	·080 (2·0%)	·018 (·4%)	Oestrous
8:7:11	3·848	·065 (1·7%)	·009 (·24%)	do.

According to Marshall, small dogs may have a recurrence of heat after four months. In the above case, however, the bitch was on heat in May and again in July. This may be due to the dog, which had been kept in the laboratory for over a year, having been prevented from breeding, in which case, according to Heape, the periods tend to become irregular,

b. THE URINE OF WOMEN IN CASES OF NORMAL PREGNANCY

In the following five cases of normal pregnancy only qualitative examinations were made. The patients were on a creatin and creatinin-free diet.

TABLE XIX

Case	Creatinin	Creatin (as Creatinin)
	mgm.	mgm.
1	5·74	·3
2	12·01	1·59
	12·4	1·56
3	5·54	·32
	6·13	·20
4	7·64	·62
5	6·43	·10

In the following seven cases an attempt was made to estimate the total amount of creatin excreted per diem. Some difficulty was experienced in getting the total volume of the urine carefully collected, so that the following figures only give fairly approximate data.

TABLE XX

Case	Urine	Creatinin-N	Creatin-N
	c.c.	gm.	gm.
6	1192	·395	·024
	1420	·364	·042
7	1079	·337	·073
	992	·302	·047
	1363	·322	·021
8	965	·254	·025
	1363	·348	·047
9	965	·197	·015
10	1533	·174	·022
11	2044	·355	·022
	2050	·293	·039
12	1760	·456	·043
	1910	·442	·023

All these cases were expected to be delivered within one month from the date of examination. They were all healthy young women between 18 and 30 years. Their weights were not obtained.

In the next case (Table XXI) a complete analysis of the different nitrogenous substances in the urine was carried out. The case was that of a young woman of about 20 years of age.

No complaint of any sort. Delivery normal, The observations were made three weeks ante-partum.

TABLE XXI

- Case 13. Age 18 Years, Weight about 38 to 45 Kilos

Date	Volume of urine	Total N.	NH3 -N.	Urea -N.	Uric acid -N.	Creatinin -N.	Creatin -N.	Total acidity in c.c. of $\frac{n}{10}$ NaOH.	Diet
			%	%	%	%	%		
	c.c.	gms.	gm.	gms.	gm.	gm.	gm.		
16:12:10	2526	7·709	·452	5·701	·117	·254	·054	334	Creatin-free
			5·8	73·9	1·5	3·3	·69		
17:12:10	2500	9·725	·545	7·080	·151	·271	·031	387	do.
			5·6	72·8	1·5	2·7	·3		
18:12:10	2600	8·226	·364	6·260	·058	·212	·035	318	do.
			4·4	76·1	·7	2·5	·4		
19:12:10	3182	8·731	·461	6·571		·376	·137	318	Fish
			5·2	75·2		4·3	1·5		
20:12:10	3068	7.903	·429			·219	·051	337	Creatin-free
			5·4			2·7	·65		
22:12:10	3324	7·725	·485	6·032	·077	·208	·019		do.
			6·2	78	1	2·6	·25		
23:12:10	3290	7·461	·493	5·379	·028	·233	·028	329	do.
			6·6	72	·3	3·1	·37		

If this Table (XXI) is compared with the table giving the composition of the urine of normal women (Tables XIII/XV), the following differences will be found. Creatin is always present in amounts which are higher than the maximal amounts observed in the urine of normal women. The urea excretion, if compared with that of normal women, is on the whole diminished, and is distinctly less than that of men. The undetermined nitrogen is correspondingly increased, and reaches figures above those observed by Ewing and Wolf in normal pregnancy. The greater part of the undeternined nitrogen may be amido-acid nitrogen, which according to v. Leersum may amount to 10 per cent. of the total nitrogen in pregnant women living on the ordinary hospital diet. The acidity is also high.

The ammonia-excretion is fairly constant and not higher than the average ammonia-excretion observed in normal women on a similar excretion of total nitrogen. My values for the ammonia excreted in the urine of two normal pregnancies (cases 13 and 14) range from 4·4 per cent. to 6·9 per cent. of the total nitrogen, the average being 5,9 per cent, This does not include the figures obtained for two days on a fish diet in Case 18, on which days the percentages of ammonia excreted were 13·9 per cent. and 9·8 per cent. The amounts found are slightly higher than the figures found by Folin in men. If one selects from his tables those figures which refer to an excretion of from 7 to 8 gms. of total nitrogen, one finds on an average about 4·8 per cent. of the total nitrogen

excreted in the form of ammonia, while 79 per cent. are excreted bound up in urea. The values for the ammonia excretion found by me in normal pregnancies agree with the observations of Ewing and Wolf on four cases of normal pregnancy, their ammonia values ranging from 2·8 per cent. of the total nitrogen to 8·6 per cent., their average figure for ammonia being about 5·1 per cent. In eight cases of eclampsia these observers found the ammonia excretion raised to very much higher figures (from 5·3 per cent. to 18 per cent. of the total nitrogen, or an average of 9·6 per cent.) On the other hand, some of the figures obtained by Hoogenhuyze and Doeschate in their recent investigations are considerably lower than those obtained by Ewing and Wolf or by myself. Some of the values for the ammonia excretion obtained by Hoogenhuyze and Doeschate are even lower than those observed in male subjects on a similar nitrogen excretion. Their average ammonia-excretion for four cases of normal pregnancy is 3·8 per cent. of the total nitrogen. Their figures range from 1·3 per cent. to 9·2 per cent., and it is to be noted that these extreme variations occur in the same case, and with a similar total nitrogen excretion, namely, 22·8 gms. and 24·9 gms. respectively. In some of the cases of eclampsia which they examined the ammonia excretion does not even exceed the lower figures found by them in cases of normal pregnancy, the average ammonia excretion for seven cases of eclampsia being 5·6 per cent, of the total nitrogen (ranging from 2·2 per cent. to 15·1 per cent.). It is possible that the diet or the water may be responsible

for the low figures found by the Dutch observers. In this connection it is interesting to note that recently one observer (Taylor), using the same method (Folin's method), but working in two different localities (California and Philadelphia), has obtained entirely different results, which he is inclined to ascribe to the factors mentioned above. These factors would not, however, account for the extreme variations observed in some of the cases of normal pregnancy (Table XIV) recorded by the Dutch observers, and to which I have referred above. It must also be borne in mind that Hoogenhuyze and Doeschate carried out their estimations on eclamptic urines after removal of the protein by boiling. It appears possible that some of the ammonia may have been lost on boiling the urine, unless special precautions were taken. In some ammonia estimations which I carried out on albuminous urine before and after removal of the protein, I have always obtained lower values after the urine had been freed from protein by boiling.

TABLE XXII.

(Case 14)

Date	Urine	Total -N.	NH_3 -N. %	Urea -N. %	Uric Acid -N. %	Crea-tinin -N. %	Creatinin -N. %	Total acidity	Diet
1910.	c.c.	gms.	gm.	gms.	gm.	gm.	gm.		
27·11	1363	12·362	·845 6·8	9·568 77	·163 1·3	·394 3·1	·065 5	–	Creatin-free.
28·11	1335	9·638	·667 6·9		·132 1·3	·308 3·2	·055 ·57	–	,,
1·12	1000	8·100	·800 9·8	6·03 74	·151 1·8	·374 4·6	·212 2·5	–	Fish
2·12	1000	6·916	·968 13·9	4·683 67·6	·168 2·4	·336 4·8	·243 3·5	–	,,
6·12	1335	10·412	·431 5·1	6·899 81·7	·167 1·6	·303 3·5	·059 ·7	–	Creatin-free.
7·12	1364	7·370	·446 6·0	5·686 77	·096 1·3	·218 2·9	·026 ·3	160	,,
1911.				POST - PARTUM		(Delivery on 28th.Dec. 1910.)			
5·1	206	(2·693)	(·108) 4	(2·446) 90		(·054) 2	(·016) ·6	(91·1)	Creatin-free.
7·1	435	(4·287)	(·115) 3·6	(3·620) 84·5	(·050) 1·1	(·083) 1·9	(·023) ·5	(152)	,,
9·1	440	(4·201)	(·189) 4·5	(3·568) 84·9	(·038) 0·9	(·097) 2·2	(·015) ·3	(169)	,,
7·2	1221	7·384	·498 6·7	5·485 74·2	·093 1·2	·211 2·8	·010 ·14	402	,,
20·2	1000	8·540	·649 7·5	6·127 71·7	·115 1·3	·372 4·3	·012 ·1	550	,,

In determining the influence which normal pregnancy has on the distribution of the urinary nitrogen, several factors must be taken into account. Individual variations and external experimental variations play a great part and may account for some of the contradictory results mentioned above. Further, the conditions obtaining in pregnancy must be compared, not with observations on normal men as has hitherto been done, but with observations on normal non-pregnant women. This is all the more necessary since the variations in metabolism due to the factor of sex are, as I have shown above, in the same direction as those observed in pregnancy.

All these fallacies can be excluded by taking observations on the same woman both before delivery and at various dates for a prolonged period afterwards. I have had an opportunity of doing so in one case (Case 14). The dietary conditions both in this case and in the previous case were the same as those observed for normal non-pregnant women. This case shows (Table XXII.), as the previous case did, a constant creatin excretion, which before delivery exceeded in amount that of normal non-pregnant women and also that of the same woman several weeks after delivery. The ammonia excretion ante-partum is distinctly increased, the urea excretion diminished. The values for the undetermined nitrogen do not go beyond the normal limit.

Post-partum the ammonia excretion falls and the urea excretion rises. The high ammonia excretion and acidity a month and three months after delivery is perhaps due to a pathological condition, since the patient was troubled with a persistent irregular discharge. It, unfortunately, not possible to obtain a complete twenty-four hours' specimen during the first week after delivery, because the urine was admixed with discharge, so that the figures given for that period have only a relative value. Nevertheless the percentage figures show sufficiently clearly that even a week after delivery, when the ammonia excretion had fallen to the normal level, there is a fairly high creatin excretion. This so-called post-partum creatinuria was first pointed out by Shaffer, who did not, however, recognise the fact that it was only an exaggeration of a process taking place throughout the whole of pregnancy. He attributed it to the involution of the uterus. Similarly Murlin, experimenting on a dog, found that creatin appeared in the urine two days before parturition (he examined the urine during the first and last weeks of pregnancy as well as one week post-partum, the dog being on a creatin-free diet), and that the creatinuria reached its maximum on the fifth day after parturition. He regarded this latter point as probably marking the maximum of the involution process.

In another case which I examined, a primipara aged 21, estimations of the total creatin nitrogen and creatinin nitrogen four and six days after delivery were carried out, the patient being on a creatin-free diet. The results obtained are;-

	Vol. of urine	Creatinin N.	Creatin N.
4 days after	1562	·795	·242
6 days after	0680	·296	·098

In a paper which has recently appeared Mellanby gives another explanation of post-partum creatinuria. He points out that the post-partum creatinuria in women, stands in relationship to lactation, and if any disturbance of lactation takes place, such as abscess formation, and consequently non-production of milk, then no creatinuria occurs. He gave some lactose to one of the cases, but got no diminution in the creatinuria, he therefore concludes that this creatinuria is not due to a disturbance of the carbohydrate metabolism; this I hold is not correct, for if glucose be given to a diabetic, the creatinuria would not be reduced owing to the sugar not being available for the organism. Similarly in the case of lactation, the giving of the comparatively small amounts of lactose (as practised by Mellanby) will not prevent the protein catabolism in the mammary gland which brings about a formation of the lactose of the milk. It is doubtful whether even very large amounts of lactose

TABLE XXIII.

Date	Urine	Total -N	Urea -N %	Ammonia -N %	Creatinin -N %	Creatin -N	Acidity	Notes
								Creatin-free diet.
26·4	1500	9·49	7·22 76	·672 7	·346 3·6	0	247	Woman, aged 54yrs.
27·4	2000	9·41	7·69 82	·592 6·2	·350 3·7	0	275	Weight=65kilos.
6·6	2200	10·72	–	·602 5·6	·360 3·3	0	290	
7·6	1500	9·07	–	·520 5·7	·304 3·3	0	348	
8·6	1500	9·78	–	·604 6·1	·396 4·0	0	426	
9·6	1500	8·90	–	·504 5·6	·345 3·8	0	354	
9·8	1000	10·02	–	·242 2·4	·448 4·4	0	336	Man, aged 52yrs.
22·11	1100	10·85	9·22 85	·419 3·8	·493 4·5	0	501	Weight=72kilos
23·11	1000	10·41	8·65 83	·403 3·8	·422 4·0	0	508	
16·11	1250	4·62	3·56 77	·287 6·2	·222 4·5	0	100	Man, aged 76yrs.
17·11	1932	7·03	5·22 74	·364 6·4	·365 5·1	0	162	Weight=69·4kilos.
18·11	1705	5·50	4·04 74	·363 6·5	·281 5·1	0	116	(Slight bronchitis)

would prevent it.

From the preceding researches it will be evident that creatin is normally excreted by the female, viz. after menstruation, in pregnancy and post-partum. Why this is so is difficult to explain as yet, but most probably it is related to the action (one or more internal secretions). Whether the thyroid gland is partly responsible is difficult to say;* it certainly has been very often noticed to become larger and more swollen during menstruation in cases of exophthalmic goitre. The liver function is also supposed by some to be altered during pregnancy (Bartels), and if so this would perhaps explain the creatinuria present in that condition.

I think however the problem is a little more complicated than this, and it will require further investigation before any more definite conclusion can be arrived at.

I shall conclude this chapter by giving the results of the analysis of the urine of a normal woman after climacteric, also the figures of the urines of two men above 50. It will be seen that there is no creatinuria present. Another point of note is that the creatinuria coefficient is low, especially in the old man of 70. (Table XXIII).

---

* Recently is has been shown by Engelhorn that the thyroid undergoes hypertrophy in menstruating and pregnant women. He holds that normally the ovary influences the function of the thyroid but the presence of a corpus luteum inhibits the action of the ovary and so hypertrophy of the thyroid takes place. „Schilddrüse u. weibliche Geschlechtsorgane." Inaug. Diss. Erlangen, 1912.

SECTION II.

THE APPEARANCE OF CREATIN IN THE URINE

in

PATHOLOGICAL CONDITIONS.

THE APPEARANCE OF CREATIN IN THE URINE IN PATHOLOGICAL CONDITIONS

Since Folin (1901-1905) noted the creatinin excretion in man remained constant without regard to the quantity or quality of the food (provided it was creatin free), and that creatin was absent from normal urine, a number of investigators have confirmed his observations, and have also endeavoured to find out how the creatin and creatinin excretion would be altered, if at all, under various pathological conditions.

I shall only mention those workers who carried out the research with the new method and shall not refer to the older results, as they are only of historical interest.

Hoogenhuyze and Verploegh ('08) examined urines of different kinds of patients, and got the following results. In Fevers, and in cases of pathological exaltation, the output of creatinin was increased. It was diminished where the vitality is lowered e.g. marasmus, whether due to disease or old age, and in such cases creatin may be found in the urine. They conclude that creatinin is formed from creatin in the body, especially in the liver.

Benedict and Myers ('07) published results

of /

of examination of twenty-five urines of insane women, and found that the form of insanity had no marked influence on the creatinin elimination. In another paper of the same issue they publish work on urines, in various pathological conditions, in which creatin was also found in the urine.

If the hypothesis of Folin is corrent, that the creatinin excretion is dependent on the metabolism, it may be expected that an alteration in the metabolism will cause a change in the excretion of the creatinin. An increase where the metabolism is increased and a diminution where there is lessened proteid catabolism. Various papers have been published concerning the creatinin elimination in fevers in which there is generally an increase in the metabolism. The most important are those of Leathes ('06) Hoogenhuyze and Verploegh ('08), Schottin ('06), Shaffer ('08) and Munk (1862). Generally speaking a rise of body temperature is associated with an increase in the creatinin output. After prolonged fever when the organism has reached a state of inanition, a fall in the creatinin content of the urine was noticed sometimes. Also an appearance of creatin in the urine. Leathes from the figures of the febrile cases he examined, found that although in each case there was increased tissue metabolism, the proportion of the nitrogen leaving the body in the form of creatinin was low. /

low. His average $\frac{CN}{TN}$ % being about 2.0. He injected into himself anti-typhoid vaccine. With a rise of temperature, he got an increase of about 20% in the output of creatinin, but an increase in the total nitrogen of about 50%, so that relatively there was a diminution rather than an increase in the creatinin. Hoogenhuyze, who had a rise of temperature for several days (Influenza), noted an increase in the creatinin of about 50%, without any corresponding rise in the total nitrogen.

In 1908 Shaffer (~~101~~) published his papers dealing with creatinin and creatin excretion in health and disease. He states that a creatinin coefficient below 7 is normal only in elderly, inactive, poorly developed or excessively fat subjects. He also emphasises that a low creatinin excretion is found in a large number of diseased conditions - in Chronic Nephritis, flat foot (very inactive), Diabetes, and Lymphatic Leukaemia. When the excretion is abnormally low, it is not peculiar to any one disease. Creatin he shows to be an abnormal product of endogenous metabolism, and not normally found in urine unless it has been taken in the food. It may be excreted by subjects of acute fevers, in the acute stage of exophthalmis goitre, and also in other conditions, where there is a rapid loss of muscle protein; also in women during the post-partum resolution of the uterus. /

uterus. He concludes that the source of endogenous creatin is probably the creatin of the muscle tissue, and its appearance in the urine most likely indicates an absorption of the muscle proteid.

As regards creatinin, this he does not regard as an index of the total endogenous proteid metabolism, for in cases of exophthalmic goitre in which this metabolism is probably greatly increased, very low creatinin excretion may be obtained. The creatinin which is slightly increased in acute fevers, is not in these cases regulated by the muscular efficiency of the patient. The creatinin excretion appears to be the result of a normal metabolism, of which the greater part, if not all, takes place in the muscles. The muscular efficiency of the person seems to depend upon the intensity of this process. In another paper which he published dealing with a patient with a permanent biliary fistula, he also noticed a low creatinin excretion.

Spriggs (07-8) examined a number of conditions in which the muscular system was either directly or indirectly affected. He found that the creatinin excretion is lowered where the bulk of the muscle tissue is diminished, e.g. in the primary myopathies. The same is seen where the muscular activity and the muscular tone are depressed by an affection of the muscle or motor apparatus, e.g. Myasthenia gravis and amyotonia /

amyotonia congenita. But in cases where the muscular tone is lowered by an interference with the sensory path, e.g. locomotor ataxia, it is unaffected. In cases of abnormal muscular activity e.g. in tetanus and spasticity, he was unable to note more than a slight increase in its excretion. His conclusions are that "creatinin is probably connected with the "nutritional metabolism of the muscle fibre and is "not a substance formed in the act of contraction."

Mellanby performed some incubation experiments on chicks. He studied the growth, the development of the liver and the increase of creatin, in their relationship to one another and was able to show that these three developed synchronously up to near hatching, when an increase in the creatin formation was observed corresponding with the growth of the liver. The muscular growth on the other hand had almost ceased. This Mellanby held suggests that the liver plays a very important part in the formation of creatinin, and consequently he examined a number of patients with disease of the liver. He found that the excretion of creatinin in disease of the liver is low. Patients suffering from cancer of the liver excrete a large amount of creatin, whereas in cirrhosis and engorged livers there is no, or practically no, creatin in the urine. Mellanby states that the diminished creatinin excretion is more likely to be due /

due to depressed liver activity than to any circulatory disturbance. This small amount of creatinin in liver disease would give additional support to the suggestion that the liver is responsible for the formation of creatinin. As regards the presence of creatin in the urine, he concludes that in carcinoma of the liver with accompanying loss of body weight it is probable that the creatin set free by the breaking down of the muscle cells is excreted without being changed to creatinin.

Hoogenhuyze and Verploegh (08) noted a low excretion of creatinin in some cases of liver disease while in others the excretion was normal. In certain cases it was even excessive. They also found creatin in the urine in cases of carcinoma of liver where the disease had destroyed the greater part of the organ. But in patients with liver disease, where the function of the organ was depressed, creatin was only present in the urine in small quantities, or not at all. The same is true with carcinoma of any other part of the body, when the liver is unaffected. These investigators add that the presence of creatin might be explained by the metabolic processes being reduced to a minimum in all the organs, and the liver consequently rendered unable to convert the creatin into creatinin. But they favour the idea according to which the muscle disintegration is increased and an increased /

increased amount of creatin set free, which owing to the liver being functionless as such (because of the cancer) passes on and is excreted with the urine. This would also explain the diminution of the creatinin, for in the above disintegration, the creatinin formation in the various tissues will be diminished if not stopped, and so the amount excreted from this source will be small. Underhill and Kleiner (1908) as well as Richards and Wallace (1908) got similar results with cases of liver disease which they examined.

Leffmann (08) induced organic disease of the liver in a dog by giving it amylalcohol as well as phosphorous, in order to study the proper relationship of creatin and creatinin to the liver function. His conclusions are:-

(1) The creatin and creatinin excretion in a well nourished animal is fairly constant. When creatin or creatinin are given with the food in such an animal, they are again completely excreted.

(2) Creatin given per os or injected is never changed to creatinin. In hunger this creatin is, however, practically all retained.

(3) When the liver is damaged, and there is increased proteid breakdown, there is a larger amount of excreted creatinin, followed by a diminution. With the diminished creatinin excretion an increased output of /

of creatin is got. From this he concludes that the liver is probably the seat of formation of creatinin. Lef mann forms the following hypothesis from this:- When the muscle requires a supply of creatin, a ferment comes into action, which converts the necessary amount of creatinin into creatin. This he holds is proven by the feeding experiments with meat extract, in which case because of the large amount of proteid present, the creatin is mostly excreted and not retained. Whereas when pure creatin is ingested, there being no abundance of proteid with it, such creatin is retained in the body.

He also poisoned the kidneys of dogs with potassium chromate. The output of creatinin was continually lowered with the progress of the lesions, but, in proportion with the fall in the creatinin, the output of creatin rose so that ultimately the ratio of $\frac{\text{creatinin}}{\text{creatin}} = 1{\cdot}2$. A still greater increase in creatin output was observed after intravascular injection of creatin or after a beef diet.

The author concluded that creatinin and creatin formation have to be regarded as two phases in the metabolism of one substance.

Cathcart (07) puts forward another theory according to which the liver is the organ most deeply concerned in one stage at least of the carbohydrate metabolism. /

metabolism. If the glycogen storing capacity of the liver were interfered with there would no longer be a proper supply of sugar available, with the result that faulty and incomplete synthesis would take place.

As regards blood diseases apart from Myelogenous and lymphatic leukaemia, Hofmann studied Chlorosis ('67), and Stejskal and Erben cases of Pernicious Anarmia ('00). They found a low creatinin excretion in these conditions.

Levene and Kristeller (1909) examined a number of pathological conditions, classifying them under three headings:-

(1) Those which are associated with a cellular activity of a very high intensity, e.g. convulsions, maniacal conditions, fever, etc.

(2) In which the cellular activity is depressed, e.g. paralysis, fasting, etc.

(3) Conditions in which a deficiency in the function of an individual organ is marked, e.g. liver and kidney diseases.

From their results they hold that there are various factors which regulate the output of creatinin, such as the formation of the substance, and its oxidation. Any disturbance of either of these two factors may lead to an abnormal creatinin output. The second factor may only be partially deficient, so that ingested creatin fails to be further oxidised.

Levene /

TABLE- XXIV

(Cases of Progressive Muscular Atrophy)

Case	Urine	Creatinin	Creatin as Creatinin	Diet	Age
1.	1705cc.	0·321gm.	0·145gm.	C.F.D.	45yrs.
	1705,,	0·451,,	0·121,,	,,	
	1378 ,,	0·025,,	0·162,,	,,	
2.	1136cc.	0·752gm.	0·008gm.	,,	60yrs.
	1420,,	0·568,,	0·027,,	,,	
3.	852cc.	1·232gm.	0·120gm.	,,	38yrs.
	625,,	0·713,,	0·145,,	,,	
	852,,	1·069,,	0·430,,	,,	
	426,,	0·547,,	0·013,,	,,	
	852,,	1·077,,	0·133,,	,,	

The average creatinin coefficient for case 3 was 4·6, which is rather low when compared with that of the normal adult at this age, namely 8·1or above. Especially is the low creatinin coefficient striking in this case, as the patient previous to the attack, was a man of great muscular development. The creatinin-nitrogen when compared to the total nitrogen, shows a constancy in the percentage, which the creatin does not show.

Spec.	Total Nitrogen	Creatinin-N	Creatin-N
2	8·88gms.	0·260gm. (2·9)	0·053gm. (·58)
3.	13·92 ,,	·399,, (2·8)	·015,, (1·1)
4.	6·86 ,,	·199,, (2·9)	·005 ,, (·07)

Levene and Kristeller also hold that creatin and creatinin are different phases of one substance, for they observed that a diminution in the creatinin excretion was accompanied by a rise in creatin. They also found that a high proteid diet (creatin free) caused in some patients an increased excretion of both creatin and creatinin. They explain the normal creatin excretion during conditions of high muscular activity by assuming that the tissues have greater power in oxidising the creatin, even though it is produced in a larger amount than under normal conditions.

I have also examined the urines of a number of patients suffering from progressive muscular atrophy. The results obtained have been collected in Table XXIV. There is nothing of special note in those cases, except that the creatin excretion is rather variable, and the creatinin coefficient is rather low, being on the average about 4·6. The creatin excretion in the first case for the first specimen was rather high, and is due to the patient not yet having been put on a creatin-free diet, and is of interest in so far as it indicates that the patient was retaining very little of the ingested creatin, if any.

From the foregoing survey of the literature on the subject of creatinuria in pathological conditions, and in what manner the creatinin excretion is affected if at all by those conditions, it is evident that there are three different views regarding the factors which may affect the creatinin metabolism and bring about a creatinuria.
The view chiefly held in America is that creatin is formed

from the breaking down of muscle, and that the creatinin excretion in the urine is an expression of the state of the muscles. This theory would account for the presence of creatin in the urine of patients suffering from progressive muscular atrophy. It also tends to look upon the creatin and creatinin having each its distinct origin.

Then there is the theory put forward by Mellanby, namely, that the liver is the seat of formation for the creatinin. It is then carried by the circulation to the muscles where it is converted into creatin. In the developing muscle the creatinin is changed to creatin and stored, while after the muscle has reached a saturation point, creatinin is continuously excreted. He supports his view by the fact that in liver disease the creatin excretion is lowered, whilst in cancer of the liver creatin is excreted, due to muscular break down, and so the creatin suffers no change to creatinin.

The third view put forward is that of Cathcart, who holds that the carbohydrates play an important part in the production of creatinuria. Cathcart and also Benedict and Diefendorf have shown that when a person fasts, creatin appears in the urine, the amount increasing as the fast proceeds, as well as a gradual diminution in the creatinin excretion. As has already been shown, the ingestion of creatin in adults may lead to a partial excretion of the ingested creatin, if the diet be proteid-rich, whereas on a carbohydrate-rich diet ~~and~~ poor in proteids, none of the ingested creatin is excreted, but retained in the body.

Lüthje had shown that the decomposition products of the proteins which are formed as a result of digestion, cannot be made use of, unless carbohydrates be present. Cathcart therefore concluded that in starvation there must be an absence of some substance which given in the ordinary diet, would prevent the breaking down of the tissue proteins, and so also prevent the occurrence of the creatinuria.

In later experiments, he found, that if a fasting person be given a sufficiency of fat (to meet the caloric requirements of the body), he was not able to reduce the creatinuria definitely, neither was he able to do so when only proteids were given instead of fats, but if only carbohydrates were given, a very definite reduction in the creatinuria resulted.

Cathcart therefore concludes that the carbohydrates are necessary for the assimilation of the proteins and if absent the protein decomposition products, produced by the breaking down of the tissues, are prevented from being resynthetised, and so creatin is set free.

If the conclusion arrived at by Cathcart is correct, then there ought to be a creatinuria in all those conditions in which there is a disturbance of the carbohydrate metabolism, such that the carbohydrates, although given in the diet, are not available for the tissues. A disease in which such a condition is present, is Diabetes Mellitus. I therefore decided to investigate this disease for the presence of creatin and any other changes present in the urine due to alterations in the endogenous metabolism.

TABLE- XXV

Case	Urine	Creatinin	Creatin (as Cin)	Cin-Cofft.	Diet	Weight	Sex	Age
1.	3182	·795	·238	5·2	Creatin-free	54·6	Woman	41
	3921	1·117	·337	7·6	,,			
	3694	·443	·109	2·9	,,			
	3654	·706	·142	3·4	,,			
2.	2273	1·702	1·115	7·3	Meat	58·3	Woman	38
	2273	1·266	·454	7·9	Creatin-free			
	2483	·811	·561	5·0	,,			
3.	4404	·920	·745	4·9	Creatin-free	68·1	Woman	
4.	5115	1·102	·644		Fish	53·9	Man	
	6194	2·156	·483	14·6	Creatin-free			
	4830	·454	·391	3·0	,,			
	2855	1·048	·604		Half-meat			
	2955	1·573	·642		,, ,,			
	2728	1·096	·842		Full-meat			
5.	2955	·564	1·060	4·0	Creatin-free	50·8	Woman	20
	2614	·732	·397	5·2	,,			
	3012	·844	·107	6·0	,,			
	3182	·792	·423		,,			
	7 weeks later							
	4546	·663	·181		,,			
	4092	·630	·433		,,			
6.	3410	1·079	·218	6·1	Creatin-free	64·1	Man	45
	2671	·940	·197	5·3	,,			
	3410	·723	·229	4·1	,,			
	2784	1·256	·207	7·1	Fast			
7.	3636	·483	·306	3·3	Creatin-free	52·8	Man	47
	4204	·744	·487	5·1	,,			
	3124	·474	·481	3·2	,,			
8.	4887	·723	·504	5·0	Creatin-free	51·8	Man	22
	4460	·551	·436	3·8	,,			
	4602	1·196	·739		Meat			

Cin.-Cofft. = Creatinin Coefficient or the number of milligrams of creatinin per kilo of body weight.

In all the cases that I examined, the patient was put on a creatin-free diet, that is to say, the patient got no meat, fish or broth, but got plenty porridge and milk, bread and butter, vegetables and potatoes. This is certainly not a strict "diabetic diet", but whether a strict non-carbohydrate diet is generally advisable, is very doubtful, if recent researches are of any value. I shall deal with the point later.

In Table XXV the amounts of creatinin and creatin excreted, per diem, by each patient are given. The creatin is stated as creatinin, for it is estimated as such. The creatinin coefficient, or the number of milligrams of creatinin nitrogen per kilp of body weight, is also stated.

These cases varied in severity, but none of them was of the simple type (alimentary glycosuria). Two cases terminated fatally. The disturbing influences which some of the acetone bodies have on the estimation of creatinin, are dealt with under the analytical methods, and I there present the results of my researches on this subject..

As will be seen from the Table XXV , the period during which a creatin-free diet was given was not lengthy, and it might be urged that creatin ~~had been retained~~ had been retained in the body from the previous meat diet, and was only slowly being excreted. This is not so, for when a diabetic had meat added to the diet, a marked increase in the creatin excretion as well as an

TABLE- XXVI

	Date	Urine	Creatinin	Creatin (as Cin.)	Cin.-Cofft.	Sugar	Notes.
		cc.	gm.	gm.		cc.	
Case 9	Feb. 5	2387	·977	·294	4·0		Creatin-free diet.
	,, 6	2671	1·374	·667	5·6		
	,, 7	2271	1·416	·585	5·3	137	
	,, 8	2216	1·136	·311	4·6	144	
	,, 9	2230	1·374	·375	5·6	-	
	,, 10	2729	1·674	·390	6·8	102	
	,, 11	2955	1·300	·360	5·2	68	
	Mar. 2	2102	·944	·273	3·8	-	
	,, 3	2216	1·090	-	4·2	15	Fast from 8pm of 4th.March t
	,, 5	1657	1·143	·267	4·6	18	to 8am. of 6th March.
	,, 6	1300	·703	·095	2·8	0	
	,, 7	909	·681	·118	2·7	trace	
	,, 8	1192	1·149	·083	4·6	,,	
	,, 9	994	·936	·058	3·8	,,	

N.B.-Only a trace of aceton present in the above specimens.

Cin-Cofft. = Creatinin Coefficient.

increase in the creatinin excretion was at once noted showing that there could not be a great retention, if there was any at all. This view is also negatived by the case (No. 9) in Table XXV, for the patient was kept on creatin-free diet for five weeks, and at no time was creatin absent from the urine.

When the Tables XXV-XXVI are examined, we find that creatin is excreted in every case of diabetes, not in small, negligible quantities, but in some cases in very large amounts, in fact, very nearly as high as the creatin figures which Mellanby got in the cases of carcinoma of the liver.

The creatinin excretion is not so constant as in normal cases: this is best seen in case 9 (Table XXVI) in which the observations were carried out over a prolonged period. Also the amount of creatinin excreted per diem is less than with normal persons, as is best seen when we look at the creatinin coefficient. The creatinin coefficient of the cases here examined gives an average of about 7; the higher figures are probabln due to some creatin still being excreted from the previous meat day, the higher results being got on the first creatin-free day. The creatinin coefficient for normal individuals, is 8·1 and over. Case 9 is a good deal below this, Some of the older observers (Maly, Senator and others) who used the old Newbaur method for the estimation of creatinin, state that in diabetes the excretion of creatinin is increased. These results are obviously due to the large amount of meat given in the diabetic diet, the creatin contents of which

was not taken into account. Senator observed a decrease of the urinary creatinin in a few cases, although a meat diet was given. This he ascribed to a fallacy in the method of estimating creatinin (Neubauer method). Several authors (Hofmann, Winogradoff, Stopczanski and Gathgens) state that creatinin is decreased in the urine of diabetics, but none of them give figures, as the presence of sugar in the urine seems to interfere with the proper deposition of creatinin as the zinc chloride salt. By none of these observers nor by any other has the presence of creatin been determined in diabetic urine. The creatin figures of case 9 in Table XXVI are of interest when compared with the figured given for the sugar. Here there is a relationship between the amount of creatin excreted and that of the sugar. As the sugar gradually diminishes from 144gms. to a trace, so the creatin also diminishes, falling from 0·585gm. to 0·058gm. The fall of the one is not directly parallel to the other, still it is apparent that with a definite fall of the one there is also a fall of the other. This relationship is also seen in the more extended analysis of the case in Table XXVII. Here there is a fall in the amount of sugar 18th. onwards from 154gms. to below 30gms. the creatin also falls from 0·647gm. to below 0·200gm. Later when this recurs the creatin does not respond; this may be due to the presence of the meat on the 1st of May.

This case of diabetes is of further interest

when it is considered that the subject was not put on a rigid diabetic diet during the whole time she was in the hospital. During the first part of her stay in the Infirmary she was on a creatin-free diet, consisting mainly of porridge and milk, toast and butter, potatoes and vegetables. When a sugar excretion is examined during this period it is seen that it falls only slowly to a trace. This fall in the sugar excretion is accompanied by a diminution in the amount of creatin excreted in the urine; a result ~~of~~ which is interesting as it shows that every case of diabetes need not be put on a carbohydrate-poor diet, and given a protein-rich diet.

The latter diet was first introduced by Dr. Rollo, in 1780, who tried it on a man of 36 years of age with good results. Since then the protein-rich diet has been very carefully worked out by Prout (1825), Bouchardat (1835), Seegen, Pavy and others. So that now it is, as one might say, an "official" method. Various other methods of treatment have been suggested during the past century, but none of them has been in vogue to any great extent.

In 1903 v. Noorden introduced his porridge treatment. It was primarily intended for those diabetics with dyspeptic symptoms who therefore could not digest large amounts of protein. But when it was applied it was found that the porridge diet led to a reduction in the output of sugar per diem. The value of this form of treatment was at once questioned, and several clinicians were able to show bad results from it, but in spite of this,

the now numerous papers on this subject go to show that with this method very good results can be got in a large number of diabetics; Lampe, Lüthje, Minkowski, Magnus-Levy and others report good results and give particulars as to how the diet should be given. Some keep strictly to the directions given by v. Noorden; others introduce modifications. Some hold that the porridge diet should only be used in cases of medium severity, others only in the severe forms, especially when coma is impending or has set in; in the latter case this diet sometimes acts like a charm. Most of them think it unnecessary for the mild forms, in fact some think it makes these cases worse. Others doubt that and recommend this diet with vegetable days interpolated on the more strict diet. In a number of the cases oedema set in, commencing at the eye-lids but was readily removed when theocin was given to the patient. V. Noorden holds that this oedema is due to a toxic substance in the oats. This view is probably not correct, for it has been shown by Blum that a pronounced water retention is got when wheaten flour is used instead of oats. It therefore seems as if in diabetes the organism retains water; just as, in the case of underfed infants, when starches are added to the milk they improve in weight owing to a water retention (the increase of weight is too rapid to be explained in any other way).

How does the porridge treatment effect an improvement?

Of the various theories put forward, the theory of

Klotz most commends itself. Naunyn and his pupil Lipetz hold that the action of porridge is not due to an elective tolerance of the carbohydrates present in the oats, but to the formation of fermentative products of the oats in the intestine. Klotz (1912) says that the carbohydrate in oats is relatively specific but not absolutely. It is relatively specific in that it is more easily broken down than that got in the various other cereals. This he has shown to be the case by experiments on phlorhizinized dogs. According to Rosenheim, Phlorhizinized dogs fed with dextrose, do not get fatty liver; on the other hand, gluconic acid, glycosamin and saccharic acid when given to such animals do not prevent fatty degeneration of the liver. Klotz used the starch of oats or of barley for such animals, and found that it did not prevent the fatty degeneration of the liver, whilst the flour of wheat or rice or the starch of potatoes caused glycogen to be deposited in the liver and did not lead to the same degree of fatty liver.

Naunyn, as already mentioned, in criticising the porridge treatment stated that its beneficial action was due to the products of the fermentation that took place in the intestine.

His pupil Lipetz found that in the cases of diabetes which were successfully treated by the porridge diet the bacterial content of the faeces was increased, whereas, in the unsuccessful cases there were very few bacteria present. Klotz, (as the result of a number of researches) holds that a successful result in a diabetic,

put on the porridge treatment, can only be got if an appropriate intestinal flora is present, or if not present in sufficient amount, if it can be augmented and improved by the repeated use of the porridge treatment just as in an adult dog, though there is no ferment in the intestine to convert lactose into dextrose, if milk be given to the dog for a prolonged period, a ferment is gradually developed which carried out this action (Lüthje). Klotz therefore ascribes the want of success in the unsuccessful cases to the absence of an appropriate intestinal flora and to the seeming inability some patients have of being able to form one when the above diet is used. This inability may be due to the improper application of the treatment or it may be an inherent deficiency in the patient.

As long as there are large numbers of proteolytic bacteria in the intestine, so long will the above treatment be unsuccessful; therefore those bacteria must be removed; this can be done by means of a fast preceding the porridge diet. The preparatory treatment with vegetables is probably even better, in so far as they have a double function, first in removing the proteolytic bacteria, and secondly, in bringing about an increase of the fermenting agents. It will also be apparent, that if the above view is correct the presence of meat in the porridge diet will favour the presence of the proteolytic bacteria and so hinder the fermenting agents from developing and carrying out their fenction.

Although Klemperer states, that one is not

TABLE. XXVII (contin.)

Date	Total Urine	Total -N	Creatinin	Creatin	Creatinin Coefficient	Sugar	Diet
	cc.	gms.	gm. %	gm. %		gms.	
Apr. 24	1932	13·60	·909 2·4	·228 ·61	3·7	120	Creatin-free
.. 25	1932	–	·948	·192	3·8	93	,, ,,
.. 26	2046	13·58	·845	·190	3·4	59	Porridge and buttermilk diet.
.. 27	2841	12·24	·913 2·2	·237 ·51	3·9	21	,, ,, ,, ,,
.. 28	2273	10·91	1·036 2·6	·114 ·6	4·1	13	Fast (lasting 30hours).
.. 29	1363	10·15	·862 3·4	·292 ·38	3·5	4·5	Milk
.. 30	1705	13·50	·987 3·1	1·04 ·256	4·1	12	Milk
May 1	1590	20·80	1·341 2·6	·331 ·6	5·7	22·1	Meat
.. 2	1705	21·99	1·381 2·2	·345 ·57	5·7	5·1	Milk and porridge.
.. 3	2484	30·38	1·584 2·2	·556 ·57	6·4	43·4	Milk and porridge.
.. 4	1932	21·40	1·167 1·9	·429 ·66	4·6	43·0	Milk and porridge
				·72			

Patient's weight on 27th.April . . . 88·8kilos.
,, ,, ,, 29th. ,, . . . 87·4 ,,
,, ,, ,, 3rd.May . . . 89·8 ,,
,, ,, ,, 4th. ,, . . . 91·1 ,,

TABLE- XXVII

Date	Urine	Total -N	Creatinin	Creatin	Creatinin Coefficient	Sugar	Diet.
	cc.	gms	gm. %	gm. %		gms.	
Apr. 5	2273		1·736	·482	7·2	98·4	Creatin-free
10	2427		·895	·432	3·6	-	,, ,,
11	2273		-	-		145	,, ,,
12	2386	19·03	1·123 2·1	·517 ·86	4·6	138	,, ,,
.. 13	2386	22·12	1·098 1·8	·458 ·66	4·5	133	Boiled, extracted meat
.. 14	2841	23·86	1·307 2·0	·755 1·1	5·3	139	,, ,, ,,
.. 15	2841	24·09	1·343 2·0	·827 1·1	5·5	126	Broth
.. 16	2784	21·05	1·149 1·9	·777 1·3	4·7	121	Creatin-free
.. 17	2557	18·82	1·030 1·9	·573 1·1	4·1	132	,, ,,
.. 18	2727	21·76	1·162 1·9	·647 1·1	4·7	158	,, ,,
.. 19	2614	35·49	2·640 2·7	·775 ·79	10·8	28	Fast (36hours)
.. 20	509	9·364	·808 3·1	·351 1·3	3·3	2·1	Cream, 1·5pints
.. 21	624	12·96	·835 2·3	·201 ·56	3·4	11	Cream, butter, cheese, buttermilk.
.. 22	738	11·88	·790 2·4	·174 ·53	3·1	31·1	Cream, butter, cheese, buttermilk and toast.
.. 23	1533	14·55	·970 2·4	·183 ·45	3·9	105	Creatin-free

limited to the use of porridge and wheat, but may try any cereal, yet it is undoubted that the porridge treatment is superior to them all.

Other cures for diabetes have been recommended, such as the milk treatment recommended by Donkin in 1863 and again brought forward by Oettinger as well as by Winternitz and Strasser towards the end of the nineties. For this treatment milk only should be given. It is indicated in young persons and in patients with nephritis. Sour milk is recommended by some as the lactose of the milk is supposed to be split up in this case, but according to certain authorities, only 10-15% of the milk sugar is split up when the milk becomes sour.

Potato -treatment in diabetes was first recommended by Mosse in 1902.

As already stated the patient, case 9, although on carbohydrate diet, showed a gradual diminution of sugar. The effect of a fast (19th April) is also well seen in this case, as a result of which the sugar falls from 158gm. per diem to 28gms, and the following day to 2·1gms. whilst the patient was on a fat diet (cream). Porridge and butter milk also had a good effect in reducing the sugar output. The creatin did not however remain low in the last two experiments but went up again.

This patient since she has left hospital takes large quantities of cream and butter; she says she feels better with this diet. She has had no relapse to her more serious condition since.

SECTION III.

C R E A T I N U R I A

under

VARIOUS EXPERIMENTAL CONDITIONS.

SECTION III

CREATINURIA UNDER VARIOUS EXPERIMENTAL CONDITIONS

In sections I and II, I have dealt with the presence of creatinuria in physiological and pathological conditions. In this section I shall deal with creatinurias induced by experimental methods.

I shall first treat with experiments, in which the drug, phlorhizin, was used, as the condition thereby induced is similar in some respects to diabetes mellitus. In the second part of this section I shall present a large series of experiments in which I studied the effect of thyroid feeding on the metabolism.

§ I. Phlorhizin Experiments

In the previous section, it has been shown that creatin is excreted in the urine of diabetics. It was therefore important to see whether in experimental diabetes there is also a creatinuria. The form of experimental diabetes I chose was the diabetic Glycosuria induced by the injection of phlorhizin. This form of experimental diabetes was first discovered by v. Mering, who described the course of the diabetes in a series of papers, (1886-9). In this form of diabetes the glycogen in the liver disappears (after repeated injections), the sugar content of the blood is not above normal (Pavy. Coolen and Kolisch) and

T A B L E - XXVIII

(Phlorhizin Experiments on Dogs.)

TABLE XXVIII

Dog	Urine cc.	T.N. gms.	Ammonia-N. gm.	%	Creatin-N. gm.	%	Creatin-N. gm.	%	Notes.
1 .	280	3·78	0·19	5.0	0·069	1.8	0		Bread and milk
	[1]450	2·89	0·10	3.6	0·051	1.7	·007	.2	Sug.=30gms.
	[11]610	5·11	0·22	4.3	0·089	1.7	·014	.27	Sug.=47gms.
	700	8·47	0·54	6.2	0·147	1.7	·0		Two days specn.
	350	4·39	0·30	6.9	0·080	1.8	0		
	250	4·47	0·138	2.9	0·078	1.7	0		Wt.= 11kilos.
2 .	300	2·43	0·169	6.4	0·085	3.3	0		Bread and milk.
	[2]610	4·45	0·24	5.4	0·088	1.9	·008	.17	Sug.present.
	[2]550	4·16	0·29	6.9	0·083	1.9	·016	.39	Sug.present.
									Wt.= 9·5kilos.
3 .	2300	9·66	0·69	7.1	-		-		Porridge and milk.
	720	2·12	0·15	5.3	0·074	2.5	0		(part specimen)
	[3] 2130	13·12	0·463	3.5	0·253	1.9	·017	.12	Sug.= 107gms.
	[3]-*1725	9·99	0·665	6.6	0·187	1.8	·020	.2	Sug.= 104gms.
	1950	9·88	0·897	9.0	0·196	1.9	·006	.06	Sug.=trace.
	320	-	-		0·033		· 0		Part specimen.
									Wt.= 20·4kilos

* Some of the urine lost.

[1] 1gm.of phlorhizin injected. [11] 1gm.at 10am. and 1gm.at 1pm. of phlorhizin injected.

[2] Same as preceding ([1] and [11]).

[3] 2gms.of phlorhizin injected at 12noon, and 2gms.at 5pm. of the same day, the third injection([3]-)was made the next day at 10am.(also 2gms.)

ligature of the ureters or ablation of the kidneys, causes no rise in the sugar content of the blood. From these results it was held that the drug in some way acted upon the kidney cells. v. Mering supposed that the kidney by means of the phlorhizin was made more permeable to sugar. Whilst Minkowski held that the phlorhizin was broken up in the kidney into dextrose and phloretin and that the latter again got into the circulation and combined with the sugar of the blood. Sugar thus excreted according to this theory would simply be the blood sugar. From work done by Pavy, Brodie and others it is now held that the phlorhizin in the kidney acts upon some substance in the blood and forms sugar from it. The substance which is so acted upon is the serum proteid.

The following experiments were carried out on two dogs. The dogs were first put on a creatin-free diet, consisting of bread and milk. There was no creatin in the urine when the injections were made. Each solution injected contained 1-2gms. of phlorhizin (with a sufficiency of sod. bicarbonate to dissolve the phlorhizin).

When Table XXVIII is examined we find that creatin was excreted in each case, on the day on which phlorhizin was injected and sugar appeared in the urine. An increase or diminution in the sugar was accompanied by an increase or a decrease in the creatinuria. The creatinin excretion also seems to be increased slightly; it is rather difficult to decide from the results got from those three dogs. There

There is also as a result of the injection a distinct increase in the total nitrogen excreted, as well as a relative increase in the ammonia excretion. The increase of these two seems to extend over several days after the injections; especially is this the case with the ammonia, and as a result the percentage amount of ammonia nitrogen of the total nitrogen is increased a few days after the last injection. Acetone and diacetic acid were also excreted, not in large amounts. The excretion of these acetone bodies occurs generally the day after the injection, and from this point of view it corresponds with the increased ammonia excretion. These results show that the increase of ammonia is in part due to the presence of the organic acids (oxybutyric and diacetic) which it has to neutralise. This Table (XXVIII) is also interesting as it points out that the creatinuria seems to precede the excretion of the acetone bodies and the increase in the ammonia output. This would explain why creatin is generally excreted in persons who are prone to get acidosis, without the latter condition being present. For instance, children are very liable to get an acetonuria, so are women during pregnancy, e.g. in the cases of toxaemia as has been pointed out by Ewing and Wolf, Williams and others.

From the above we can conclude that creatinuria very often occurs as a result of a disturbance of the carbohydrate metabolism. It is generally accompanied by an increase in the ammonia excretion, and may be or may not be succeeded by an acetonuria, according to the severity of the condition.

TABLE. XXIX

Urine cc.	Total Nitrogen gms.	Creatinin-N gm.	Creatin-N gm.	Dog
300	2·80	·044	0	Bitch N.
		1·5		
325	2·46	·038	·002 *	
		1·5	·08	
280	2·46	·037	·017	
		1·5	·7	
270	3·25	·056	·012	
		1·7	·3	
620	2·91	·069	·014	Bitch F.
		2·3	·45	
850	3·00	·073	·018	
		2·4	·6	
710	2·76	·060	·014	
		2·1	·5 **	
820	3·76	·073	·035	
		1·9	·9	
590	3·67	·065	·036	
		1·7	·9	

Diet- Both dogs were on a creatin-free diet consisting of bread and milk.

* Besides above diet got 4gms. dessicated thyroid. (in two parts one at 11am. and the other at 5pm.)

** Besides bread and milk got 4 lobes of sheep's thyroids.

§ 2. EXPERIMENTS WITH THYROID GLAND

As a result of experiments carried out in the Physiological Laboratory of this University, it was found,* that, when dogs, whose normal assimilation limit for glucose has been determined, had fresh thyroid gland fed to them, the limit for the assimilation for glucose was markedly lowered. That exophthalmic goitre is sometimes associated with glycosuria has been shown in cases recorded by Dumont-pellier, Brunton, Wilks, O'Neill and others. But the relation of the thyroid gland to normal metabolism has, so far as I know, been hitherto little studied.

It was therefore of interest to study the effect of feeding dogs with thyroid, on the nitrogen distribution in the urine. To do this the dogs were put on a creatin-free diet over an extended period, and the 24 hrs. specimens of urine analysed. In TableXXIX I give the results obtained from two bitches.

The urine of one dog was examined just before the oestrous period, and in her case creatin was constantly found. The analyses carried out in those two cases gave the total nitrogen of the urine, as well as the creatinin- and creatin-nitrogen. In the case of the dog N., the adminis-

*1. Communication to the Physiological Society by Robertson and Cramer, June 1911.
2. Communication to the Physiological Society by Krause and Cramer, Proceeds, Journal of Physiology, Vol.XLIV 1912.

TABLE XXX

Urine	Total -N	Ammonia -N	Creatinin -N	Creatin -N	Acidity	Ammonia+ amino ac. -N	Notes.
cc.	gms.	gm.	gm.	gm.		gm.	
650	5·24	·563 (10)	·090 (1·7)	trace	161	·564	Male dog J. Porridge, bread and milk.
770	5·26	·474 (9)	·095 (1·8)	trace	157	·476	Got 5 lobes of thyroid.
530	4·80	·395 (8·2)	·068 (1·4)	·025 (·5)	108	·403	
1400	3·33	·281 (8·4)	·118 (3·5)	·0	119	·343	Male dog N(2). Porridge, bread and milk.
1000	3·36	·246 (7·2)	·115 (3·4)	0	115	·329	
1200	3·86	·300 (7·7)	·118 (3·0)	0	75	· –	Got 1 lobe of thyroid.
1200	6·11	·388 (6·3)	·116 (1·8)	·011 (·18)	183	·462	Got 4 lobes of thyroid.
340	2·58	·177 (6·8)	·038 (1·4)	·0	89	·22	Bitch. (D) Potatoes and milk.
350	2·69	·164 (6·0)	·038 (1·4)	0	77	·18	
460	3·14	·193 (6·1)	·040 (1·2)	0	82	·22	Got 1·5 gm. Dessicated thyroid.
330	2·97	·136 (6·2)	·040 (1·3)	0	60	·17	
240	2·72	·199 (7·3)	·044 (1·6)	0	45	·20	

administration of the fresh thyroid gland caused the appearance of creatin in the urine. This creatinuria was still present in the urine of the next two days. The amount of total nitrogen does not show any marked immediate change, but there is a rise in the third day after administration of the thyroid. In the case of the dog F. the amount of creatin excreted is increased as the result of the administration of thyroid gland. The total nitrogen is also definitely increased.

Table XXX gives analyses of urine of three other dogs which were given thyroid. These dogs were also on a creatin-free diet. The examination included estimations of total ammonia, creatinin and creatin nitrogen: also the acidity and the ammonia + amino-acid nitrogen.

One of the dogs (D) did not excrete creatin as a result of thyroid administration, this being probably due to the small amount given, for in the case of the dog N., one lobe of fresh thyroid also had no effect. In both those cases however there was a slight increase in the total nitrogen as well as in the ammonia nitrogen. When however the dog N. was fed with a larger amount of thyroid, he excreted creatin. The rise in total nitrogen and ammonia-nitrogen is also a marked one. In the dog J. creatin is also excreted in a large amount as a result of giving thyroid. The acidity as well as the ammonia + amino acid nitrogen is also increased.

After these preliminary observations, I extended the analyses of the different constituents of the urine.

TABLE XXXI

Date	Urine	Total N	Urea N	Ammonia N	Creatinin N	Creatin N	Acidity	Ammonia + amino ac. N	Uric ac. N	Total P_2O_5	Ca	Mg	Total SO_3	Cl
Ap.	cc.	gms.	gms. %	gm. %	gm. %	gm. %	cc.	gm.						
8 *	1020	6·82	4·426 83	·317 5·8	·139	0	163	·494	–	–	–	–	–	
9	605	5·29	4·427 83	·317 5·8	·139 2·6	0	90	·414	·002	·872	–	–	–	
10	745	3·68	2·96 80	·327 8·8	·120 3·2	0	89	·336	·002	·818	·021	·035	–	
11	1025	6·02	5·13 85	·367 6·0	·141 2·3	·022 ·37	158	·447	·003	1·358	·032	·061	–	
Dec.														
19 **	510	4·33	–	·311 7·1	·082 1·9	0	–	·–	·–	–	–	–	–	
20	600	4·75	3·88 81	·426 8·9	·089 1·8	0	125	·427	·028	–	·025	–	·512	2·4
21	600	4·87	3·89 80	·440 9·0	·074 1·5	tr.	125	·476	·028	–	·035	–	·501	3·8
22	600	5·70	4·73 83	·430 7·5	·093 1·6	·029 ·50	138	·433	·028	–	·020	–	·571	3·3
23	500	3·95	–	·358 8·9	·082 2·0	·0059 ·14	98	·358	–	–	–	–	–	2·5

* Male dog (N) was fed with potatoes, bread, and butter thrice daily. On 10th. Ap. got 8 lobes thyroids.

** Male dog (J) was fed with porridge, bread and milk, twice daily. On the 20th. got about 2gms. of dessicated thyroid (=5 lobes of fresh gland).

In Table XXXIare given the results of thyroid feeding on the two dogs J. and N. They comprise estimations of the various nitrogenous substances and also of various inorganic constituents, such as calcium, phosphoric acid, magnesium, chlorides and total sulphates.

When Table XXXIis examined it is seen that the increase in the total nitrogen excretion, as a result of thyroid feeding, is almost wholly accounted for by the increased urea excretion. The increase in the ammonia excretion is not so marked as that of the urea. The uric acid excretion in dogs is very low, as in those animals the uric acid is oxidised a stage further and is excreted as allantoin.

The increase of the creatinin excretion is not very pronounced. In both cases creatin appeared in the urine after the administration of the thyroids. The acidity is also increased.

The inorganic constituents were also increased in amount after the animal was fed with thyroid.

Against the foregoing it might be contended that the creatin that appears in the urine as a result of thyroid feeding, is simply the creatin present in the thyroid ingested. I therefore made an estimation of the amount of creatin present in a gland and found that even a kilo would only contain 10·8mgm. creatin nitrogen : the amount in a lobe is therefore negligible.

I also carried out similar experiments on thyroid feeding on the human subject. In Tables XXXII-XXXIV are the results of the urine analyses from various such

TABLE XXXII (Expts. 1-2)

Date	Urine	Total -N	Urea -N	Ammonia -N	Creatinin -N	Creatin -N	Acidity	Ammonia + amino ac. -N.	Total -N	Notes.
	cc.	gms. %	gms. %	gm. %	gm. %	gm. %	cc.	gm.	gm.	Expt. 1
26·10	1300	14·20	11·38 80	·604 4·2	·606 4·2	·035 ·24	369	·851	1·638	Diet- Porridge, toast, fat, butter, vegetables, bread, apples and 6oz. meat
27·10	1600	15·01	12·24 82	·582 3·8	·623 4·1	·034 ·2	288	·762	1·401	
28·10	1225	14·68	11·87 79	·612 4·1	·567 3·8	·047 ·32	417	·824	3·507	2 lobes of thyroid given.
										Expt. 2
2·11	1500	12·26	9·47 77	·605 4·9	·547 4·4	·0	366	·783	2·970	Diet- Same as above ex- the meat which was
3·11	1450	12·31	10·24 83	·592 4·8	·543 4·4	0	278	·804	3·000	left out.
4·11	1900	12·77	10·04 78	·766 6·0	·562 4·4	·021 ·16	403	·979	4·100	3 lobes of thyroid given.

TABLE XXXIII (Expt. 3)

Date	Urine	Total -N	Urea -N	Ammonia -N	Uric ac. -N	Creatin -N	Creatin -N	Acidity	Ammonia + amino ac. -N	Total P_2O_5	Total SO_3	Calcium	Mg.	Chlorides
Jan.	cc.	gms.	gms.	gm.	gm.	gm.	gm.	cc.	gm.	gms.	gms.	gm.	gm.	gms.
24	1100	10·08	7·82 (78)	·579 (5·7)	–	·485 (4·8)	0	255	·707	2·42	–	[illegible]	–	10·45
25	1000	11·27	8·78 (78)	·767 (6·8)	·132 (1·1)	·509 (4·5)	0	392	·918	2·90	2·11	·086	·148	10·80
26	1100	11·65	8·98 (77)	·720 (6·1)	·120 (1·0)	·486 (4·8)	0	374	·843	2·99	2·22	·069	·148	10·40
27	1500	13·72	10·86 (79)	·815 (5·9)	·213 (1·5)	·538 (3·9)	·010 (·07)	570	1·008	4·65	2·59	·107	·160	11·76
28	1500	15·75	12·21 (77)	·998 (6·3)	·217 (1·3)	·573 (3·6)	·131 (·8)	660	1·243	4·57	2·62	·129	·154	11·76
29	1250	15·20	11·74 (78)	1·078 (7·0)	·216 (1·4)	·534 (3·5)	·150 (·98)	670	1·330	4·87	2·53	·182	·182	11·75

Diet- Was the same each day and consisted of porridge, milk, toast, butter, eggs, potatoes, vegetables, apples and jam.

On the 26th. (spec. of 27th) besides the diet got Thyroids, 3lobes at 8.30am. 4 at 1.0pm. and 3 at 6pm.

The figures for the N : S ratio from the 25th. onwards are - {19.4 : 1, 19.0 : 1, 18.8 : 1, 16.6 : 1, 16.7 : 1}

experiments carried out on myself. The diet in the first experiment (TableXXXII) included 6oz. of meat along with potatoes, vegetables, bread and butter, eggs, porridge and milk. In the second part the meat was excluded so that the diet was creatin-free. This latter diet was also used for the experiment in Table XXXIII. In the last experiment, (TableXXXIII) a carbohydrate- and fat-rich diet was used, which was very poor in protein. After I had been on a particular diet (constant in daily quantity) for some time in each experiment, I took two or more lobes of the fresh sheeps thyroid gland. In the experiments on Table XXXIII, fairly constant quantities of water were taken per diem in order to see how the quantity of urine would be affected by the ingestion of thyroid gland.

On examining Tables XXXII-III it is seen that a creatinuria is induced in experiments 1, 2 and 3 but not in 4. This may be related to the change to the carbohydrate and fat diet. The amounts of creatin excreted in experiments 1 and 2 are not very great, because of the comparatively small amounts of thyroid ingested, for in experiment 3 in which the diet is practically the same as in experiment 2, but included a larger dose of thyroid (10 lobes), the creatinuria reaches a very high figure. The creatinin excretion in this experiment is also increased in amount.

In Tables XXXII-XXXIII, it is seen that the ingestion of thyroid causes an increased excretion of the total nitrogen. According to Scholz and also Richter this can be avoid-

de

TABLE XXXIV (Expt.4)

(Inorganic Constituents)

Date	Total P_2O_5	Sulphates		N : S	Calcium	Magnesium	Chlorides	Diet.
		Total	Inorganic	ratio				
	gm.	gm.	gm.		gm.	gm.	gms.	
12·2	1·920	1·230	1·01	24 : 1	·037	·139	14·40	Potatoes, cheese (Cream) apples and butter.
13·2	2·210	1·098	–	25 : 1	·047	·156	16·30	
14·2	2·200	1·250	1·140	34 : 1	·059	·150	18·00	Cheese discontinued.
15·2	1·580	1·320	1·175	38 : 1	·040	·297	14·85	
16·2	2·190	1·270	1·140	39 : 1	·043	·237	–	
17·2	2·100	1·170	1·060	37 : 1	·039	·350	14·00	Also a soln. containing 79mgm. Creatin-Nitrogen.
18·2	2·056	1·135	1·058	33 : 1	·033	·269	14·70	
19·2	2·100	1·127	1·087	32 : 1	·043	·396	14·80	
20·2	2·240	1·220	1·116	31 : 1	·035	·364	16·03	3 at 9 am
21·2	3·340	1·155	1·092	25 : 1	·063	·377	20·80	Diet & Thyroids. 3 at 1pm. 1 at 10 pm.
22·2	2·530	1·107	0·998	25 : 1	–	–	19·10	
23·2	2·160	1·102	0·925	25 : 1	·064	·400	–	
24·2	–	1·350	1·208	28 : 1	·061	–	–	

By thyroid is meant a lobe of a sheep's thyroid

TABLE XXXIV (Expt.4)

Date	Urine	Total -N	Urea -N	Ammonia -N	Uric acid -N	Creatinin -N	Creatin -N	Acidity	Ammonia + amino acid -N	Weight in kilos	Notes
cc.	cc.	gms.	gms. %	gm. %	gm. %	gm. %	gm. %	cc.	gm.		
12·2	1200	4·60	2·82 61	·296 6·4	·138 3·0	·455 10·0	0	163	·423	–	
13·2	1700	4·28	2·74 64	·352 8·2	·132 3·0	·492 11·5	0	211	·485	–	
14·2	2000	3·70	2·11 57	·291 7·9	·137 3·7	·473 12·0	0	170	·452	–	
15·2	1400	3·45	1·94 55	·219 6·3	·144 4·1	·470 13·6	0	224	·353	–	
16·2	1460	3·27	1·84 56	·245 7·5	·127 3·9	·485 14·0	0	228	·344	66·5	
17·2	1400	3·14	1·39 44	·250 8·0	·151 4·8	·498 15·0	0	168	·400	66·3	(Creatin)
18·2	1470	3·37	1·87 55	·263 7·8	·160 4·7	·524 15·0	0	211	·428	–	
19·2	1760	3·45	2·03 59	·226 6·5	·159 4·6	·515 14·0	0	218	·375	66·0	
20·2	1370	3·37	2·02 59	·268 8·0	·175 5·2	·520 15·0	0	230	·421	–	
21·2	1760	4·54	2·46 55	·394 8·6	·175 3·8	·536 11·0	0	373	·611	–	(Thyroids)
22·2	1950	4·37	2·38 54	·404 9·2	·170 3·9	·507 11·0	0	320	·579	65·0	
23·2	1200	4·37	2·39 55	·369 8·4	·166 3·8	·500 11·0	0	302	·571	–	
24·2	1300	4·73	2·67 56	·386 8·1	·182 3·8	·480 10·0	0	–	–	–	

ed by supplying an increased number of calories in the food. I incline to the view that if sufficient thyroid be given, there will always be a loss of total nitrogen; the amount of nitrogen lost may vary, but this I hold is not due to the quantity of the food, but to its quality as regards carbohydrates and protein.

Schöndorff states that, where there is a negative nitrogen balance after the administration of thyroid, the loss in nitrogen is primarily due to an excretion of nitrogenous extractives. If Tables XXXII-IV are examined, it is at once evident that the increased excretion in the total nitrogen is in great part due to the increased excretion of urea, as a result of which the percentage contributed by the urea to the total nitrogen remains the same. If the phosphorus and sulphur output be also examined, it will be seen that the excretion of those substances is also increased; this leaves little doubt that the increased loss of nitrogen is due to a breaking down of protein, which may be further demonstrated as follows: the ratio of the nitrogen to the sulphur in muscle is 14 to 1; now if the nitrogen : sulphur ratio in Table XXXIII be examined it will be seen that whereas the ratio is 19 : 1 and above before the administration of the thyroid; now after the ingestion of thyroid the ratio falls to 16·6 : 1; this can only have occurred as a result of an increased decomposition of protein.

Experiment 4 is of great interest, as demonstrating the contrary effect of a diet rich in carbohydrate (and of low

nitrogen content-about 3·0 to 3·5gms.), on the composition of urine. In this experiment the nitrogen : sulphur ratio is very high, as the protein metabolism is reduced to a minimum. This N : S ratio fluctuates between 31 : 1 and 38 : 1, but on thyroid being administered, the resulting protein breakdown, causes a fall in this ratio to about 25 : 1 and this persists for three days, when it again begins to rise, as the thyroid loses its effect.

The other nitrogenous substances which participate in the increase of the total nitrogen, besides urea, are ammonia, amino acids, creatinin and uric acid. The excretion of calcium, magnesium and chlorides after thyroid feeding is likewise increased.

Experiment 4 apart from the feeding with thyroid is also interesting as showing to what degree the nitrogen excretion can be reduced in health. Clinically it is held that an excretion of 2·5 gms. of urea is only found in pathological conditions; here however is a normal person, on a diet with a caloric value (more than 3000 calories) above what was really necessary, and yet for a period of about a week the amount of urea of urea excreted was below 4gms. per diem-even as low as 2·8gms. on the 17th. Feb.(i.e. 1·37gms. of urea nitrogen). The percentage of total nitrogen formed by the urea is also of interest. On an ordinary diet containing 10 to 15gms. of nitrogen, the percentage of the total nitrogen represented by the urea will be between 80 and 85. In experiment 4 , in which the total excretion is reduced to about 3-4 gms., the per-

centage of the total nitrogen excreted as urea-nitrogen varies between 45-55.

Another fact to which attention may be drawn is that on the carbohydrate diet given in experiment 4 , there is an increase in the excretion of uric acid. Whether this increase is due to a specific action of the potatoes present in the diet, or simply due to the diet being protein-poor and carbohydrate-rich, further experiments will have to decide. In this connection, the observations of Hindhede are of interest, for he found that when a person gets a latge amount of potatoes in the diet, the urine excreted has a very high dissolving power for uric acid, and this was not the case when meat or bread was given instead of potatoes. One must not forget of course that potatoes contain much water and the large amount of water excreted may be partly the cause of Hindhede's results.

In experiment 4 , electrocardiograms, pulse-rate, and blood pressure readings were taken by Drs. Ritchie and Croom. The electrocardiograms showed no abnormality of the heart's action beyond the tachycardia, as a result of thyroid administration. The pulse-rate (when lying) rose from 64 beats per minute to 80. The blood-pressure rose from 115 and 120mm. of mercury to 130mm. on the first day after thyroid feeding, to 140mm. on the second day after. [This result differs from that obtained in experiments upon animals.] The subjective symptoms were, a poor appetite, a feeling of closeness and discomfort and also headache. I felt rather thirsty,

TABLE XXXV

Expt.	Urine	Total -N gms.	Urea -N gms.	Ammonia -N gm.	Creatinin -N gm.	Creatin -N gm.	Acidity	Notes (Male dog J. on Bread and milk diet)
1.	365	4·90	4·32 88	·29 6·0	·081 1·6	trace	138	
	280	3·60	2·99 82	·21 5·9	·094 2·6	·022 ·6	134	Fast–Water+Thyroids
	425	5·28	4·38 83	·27 5·1	·085 1·6	·051 ·9	97	
2.	312	4·16	3·54 85	·28 6·8	·066 1·7	·0	91	
	411	4·65	3·90 84	·34 7·2	·072 1·5	0	111	
	620	2·81	2·29 81	·23 8·2	·085 3·0	0	84	Fast–Only Water.
	350	3·88	3·37 88	·13 3·4	·062 1·6	0	43	
3.	383	4·02	3·23 80	·27 6·8	·056 1·4	?	81	
	590	2·77	2·32 79	·22 8·1	·068 2·4	0	112	Fast–Only Water
	670	4·54	3·59 79	·35 7·6	·068 1·5	·02 ·4	179	Water + Thyroids.
	500	5·16	4·41 85	·28 5·4	·090 1·7	·02 ·3	106	
4.	400	3·47	2·92 84	·17 5·1	·061 1·7	0	–	
	716	3·25	– 78	·47 13	·075 4·6	0	–	Fast–Water only
	550	2·90	2·27 78	·28 9·5	·106 3·6	·0	–	Fast–Water only
	350	4·10	3·63 88	·23 5·7	·076 1·8	0	–	

and exposure to the sun or to a fire caused discomfort.

In experiments 3 and 4, in which a constant quantity of water was taken per diem, it will be seen that there is an increase in the volume of urine excreted per diem as a result of thyroid feeding.

In order to demonstrate the effect of thyroid feeding on the endogenous metabolism, I carried out a number of experiments on a dog, which was starved for one or two days; whilst fasting thyroid was given. The results are tabulated in Table XXXV.

It will be seen that the effect of thyroid on the total nitrogen is to increase it, the increase being in the main due to the urea. When the dog was allowed to fast, only getting water, the total nitrogen fell, and generally there was no excretion of creatin (the fasting was not long enough to bring about a creatinuria); if however thyroid was given as well as the water, then the fall in the total nitrogen excretion was not so marked, and creatin was excreted on each occasion. The effect on the creatinin excretion is not definite enough to allow of any conclusions being formulated.

To summarise, it has been shown in this section that the injection of phlorhizin or the administration of thyroid, bring about a creatinuria. Those conditions are also associated with a disturbance of carbohydrate metabolism. It will have been noticed that the administration of thyroid brought about an increased calcium excretion. An increased excretion of calcium is also noted in Diabetes*; the excretion of cal-

* Increase of about 2or 3 times.

TABLE XXXVI

Date	Urine cc.	Total -N gm.	Calcium gm.	Sugar gm.	Notes.
18·12	490	8·87	-	-	Bitch D. Bread and milk (constant quantity)
20·12	440	6·30	·029	0	Acetone and diacetic acid not present.
21·12	605 *	9·73	·021	36	Acetone and diacetic acid present.
18·12	885	13·90	·036	0	Male dog J. Meat and Bread (constant quantity)
19·12	815	11·90	lost	0	Acetone and diacetic acid not present.
20·12	1020 **	12·94	·031	42	Acetone and diacetic acid (doubtful)
21·12	1050 ***	10·77	·029	80	Acetone and diacetic acid present.

* = 3 gms. of phlorhizin were injected.

** - 1·5gm. of ,, ,, ,,

*** - 2 gms. of ,, ,, ,, (twice).

TABLE XXXVII

Expt.	Urine cc.	Total -N gms.	Ammonia -N gm.	Creatinin -N gm.	Creatin -N gm.	Acidity	Ammonia + amino ac.-N gm.	Notes.
1.	650	3·21	·163 (5·1)	·047 (1·4)	·003 (·09)	67	·178	Dog N. (female) Bd. and milk
	360	2·89	·145 (5·0)	·037 (1·2)	·002 (·06)	35	·153	Got anterior pity.
	400	3·29	·177 (5·3)	·044 (1·3)	· 0	43	·171	
2.	300	2·74	·26 (9·5)	·046 (1·7)	·001 (·03)	64	·158	Dog D. (female) Bd. and milk
	290	2·62	·190 (7·2)	·047 (1·8)	·002 (·07)	61	·191	Got post. pituitary
	425	3·38	·200 (5·9)	·054 (1·6)	0	76	·216	
3.	500	3·766	·176 (4·6)	·053 (1·4)	·005 (·12)	56	·196	ibidem Bd. and milk.
	620	4·147	·196 (4·6)	·054 (1·3)	· 0	57	·237	Got comlpete pit[illegible].
	600	3·90	·201 (5·1)	·054 (1·3)	0	67	·225	
4.	425	4·76	·367 (7·7)	·078 (1·6)	0	124	·357	Dog J. (male). Bd. and milk
	350	4·22	·225 (5·3)	·068 (1·6)	tr.	88	·210	Got post. pituitary
	475	4·59	·268 (5·8)	·076 (1·6)	tr.	104	·266	

cium here may be twice or even thrice that found normally. As it seems to be increased in conditions associated with a disturbance if carbohydrate metabolism, it became of interest to see whether it was also increased by the injection of phlorhizin. The experiment was carried on two dogs, but from the results obtained (Table XXXVI) there seems to be no increase in the calcium excretion.

A number of feeding experiments with pituitary body (analogous to those with thyroid described in this section) have been carried out. In these, dogs had added to their diet the dessicated gland (total, or anterior or posterior lobe) and the urine examined to determine any change in nitrogenous metabolism. In Table XXXVII are seen the results of these investigations ; in no case did creatinuria definitely occur ; nor indeed did any variations of significance take place at all.

SECTION IV.

SOME PROBLEMS

IN CONNECTION WITH THE GENESIS OF

A C I D O S I S.

SECTION IV.

SOME PROBLEMS IN CONNECTION WITH THE GENESIS OF ACIDOSIS.

In the experiments detailed in the previous sections, it was shown that creatinuria generally occurred in conditions under which the subject is prone to an acidosis, eg. diabetes, phlorhizin diabetes, infancy and early childhood, pregnancy. In the experiments carried out with thyroid feeding, I was not able to induce an acidosis, probably owing to the period of administration being only one day.

It was therefore of interest to see what changes occur in the urinary nitrogen distribution as a result of a physiological acidosis. Such a physiological acidosis can quite easily be induced by eliminating the carbohydrates from the diet.

The experiment which I carried out on myself was as follows; weighed quantities of cream cheese, butter, Cox's gelatine and eight eggs formed the basis of the diet (which is protein- and fat-rich, but contains very little carbohydrate); on the first day besides the above diet, I also took potatoes (to supply the carbodrates); on the second and third days of the experiment the potatoes were left out of the dietary. The diet which was taken on those three days was creatin- and purin-free.

TABLE XXXVIII

Date	Urine	Total -N	Urea -N	Ammonia -N	Uric acid -N	Creatinin -N	Creatin -N	Acidity	Ammonia + amino ac. -N	Total SO_3	Calcium	Weight in
	cc.	gms.	gms. %	gm. %	gm. %	gm. %	gm. %	cc.	gm.	gm.	gm.	kilos
23·12	1550	13·33	10·89 82	·729 6·0	·174 1·3	·587 4·4	·017 ·10	372	·937	2·415	·164	–
			Note– no acetone or diacetic acid present. N:S ratio = 18·1 : 1									
24·12	1250	14·49	12·55 86	·950 6·5	·150 1·0	·486 3·3	·038 ·19	577	1·123	2·583	·165	66·5
			Note– acetone present, diacetic acid doubtful. N:S ratio = 17·8 : 1									
25·12	1450	17·66	15·28 86	1·348 7·6	·112 0·6	·516 2·9	·093 ·52	684	1·494	2·761	·137	65·8
			Note– acetone present, also diacetic acid. N:S ratio = 15·6 : 1									

Diet on the 23rd. Dec. – 8 eggs, butter($\frac{1}{2}$lb.), cream cheese($\frac{1}{2}$lb.), Cox's gelatine($\frac{1}{2}$lb.), water and potatoes.

Diet on the 24th. and 25th. Dec. same as the foregoing except for the potatoes.

The results of the analyses of the various nitrogenous and other constituents of the urine are given in Table XXXVIII

The first point of special interest noticed on examining this table, is that although the nitrogen intake is the same in amount for each of the three days, yet, as soon as the carbohydrates, in the form of potatoes, are eliminated from the diet, the total nitrogen excretion rises, not so very much on the first day of the carbohydrate-free diet, but very markedly on the second day of that diet. Which are the nitrogenous substances in the urine which participate in this increase? If Table XXXVIII be examined it will be seen that both the absolute amount and the percentage amount (of the total nitrogen) of the urea nitrogen have increased. Whereas on the first day of the experiment the percentage of total nitrogen contributed by the urea is 82, on the second and third days of the experiment, it rises to 86: showing therefore an absolute as well as a relative increase in the urea nitrogen. The same applies to the ammonia. Creatin, which was present in slight amount on the first day of the experiment, increases on the second and still more on the third day, owing to the increased endogenous metabolism.

The other nitrogenous substances of the urine, namely, uric acid, creatinin and amino-acids, all show a diminution as a result of the privation of carbohydrates. It will be remembered that in Table XXXVIII the uric acid showed an increase in amount as a result of a diet mainly consisting of carbohydrates; here on the other hand, the giving of a protein

and fat diet results in a decrease.

A piece of evidence which shows that in this experiment there was an increased protein catabolism, is the nitrogen : sulphur ratio. This ratio on the first day of the experiment was 18·1, whilst on the second day it fell ti 17·8 and on the third day to 15·6, a gradual approximation to the ratio which Cathcart got in a fasting subject, namely 15. In this fasting man, he also got a diminution of the uric acid, creatinin and calcium, just as is seen in Table XXXVIII In fact the results of my experiment resemble very closely those of Cathcart's in his observations on the fasting individual: viz., on the second and third days of the fast, there was a temporary rise in the total nitrogen, mainly represented by an increase in the urea and ammonia, and a diminution in the other nitrogenous substances of the urine, except creatin which gradually increases. The preliminary fall, on the first day of starvation in the total nitrogen , according to Prausnitz is due to the organism using its store of carbohydrate (glycogen). As the amount of carbohydrate which can be stored is strictly limited, it is soon used up. The amount of rise in the total nitrogen on the second and third days depends upon the amount of protein taken immediately previous to the fast; if a large protein intake has preceded fast, then the rise on the second and third days will be a high one, and vice versa. The above experiment is therefore of interest in that although the subject was getting a sufficiency of proteins and fats, yet after the carbohydrates

TABLE XXXIX

Urine	Total -N	Urea -N	Ammonia -N	Uric acid -N	Creatinin -N	Creatin -N	Acidity	Ammonia+ amino ac. -N	Notes
cc.	gms.	gms. %	gm. %	gm. %	gm. %	gm. %	cc.	gm.	C.F.D.
1150	11·39	–	·506 4·4	–	·459 4·0	·006 ·06	230	·672	
1275	11·96	9·84 82	·548 4·6	·182 1·5	·450 3·7	·008 ·06	223	·718	
2000	12·60	9·61 76	·694 5·5	·149 1·1	·450 3·6	·013 ·10	345	·868	About 7cc. $\frac{N}{1}$HCl taken.
3200	13·08	10·41 79	·697 5·3	·139 1·1	·440 3·3	·023 ·17	288	·851	About 12cc. $\frac{N}{1}$HCl ,,
3000	13·44	10·79 80	·972 7·2	·148 1·1	·448 3·3	·019 ·10	525	1·134	About 30cc ,, ,,
2000	11·032	9·06 82	·290 2·6	·154 1·3	·411 3·3	0	160	·448	Two portions of $NaHCO_3$ take 3gms. at 11am. and 5gms. at 4 4pm.
1500	13·48	10·04 74	·795 5·9	–	–	–	315	·913	New experiment.
2000	14·22	10·66 75	·980 6·8	·141 0·9	·407 2·8	0	480	1·211	
2000	15·12	11·33 71	1·152 7·6	·158 1·0	·422 2·7	0	490	1·386	25cc. $\frac{N}{1}$ HCl taken.
2200	14·78	10·67 72	1·403 9·5	·139 0·9	·428 2·8	0	495	1·578	50cc. ,, ,, ,,
2000	14·33	10·44 73	1·157 8·4	·146 1·0	·422 2·9	0	440	1·414	30cc. ,, ,, ,,
2000	13·05	–	1·288 9·8	–	·406 3·1	0	500	1·589	40cc. ,,,, ,,

25cc. $\frac{N}{1}$ HCl = 0·35gm. NH_3-N.

30cc. $\frac{N}{1}$ HCl = ·42gm. NH_3-N

No acetone and no diacetic acid found in the above specimens of urine.
Creatin-free diet throughout

were removed from the diet, the organism was behaving just as a fasting subject on the second and third days of a fast; showing that there is an increased catabolism of the proteins.

In the above experiment, traces of acetone appeared in the urine on the first protein day, whilst diacetic acid and B-oxybutyric acids were also excreted on the second protein day.

Benedict and Higgins have recently shown that the respiratory quotient got with a carbohydrate-free diet is as low as the ordinary quotients got in diabetes, if not lower, and that the respiratory quotient could be elevated persistently by increasing ~~by increasing~~ the quantity of carbohydrates in the diet.

The above results are of interest when compared to the results got with diabetic patients. Clinical experience with diabetic patients suggests that an acidosis of moderate severity when produced by the sudden withdrawal of carbohydrates, is more dangerous than a more severe acidosis produced by a gradual elimination of carbohydrates from the diet. The result therefore of a sudden withdrawal of carbohydrates from the diet of a diabetic patient, when taken into hospital, may very readily bring about an acute attack of coma. On the other hand, it has been found that severe forms of diabetes when put upon a carbohydrate- poor diet for a prolonged period, may have a very large degree of acidosis and yet show no tendency towards the onset of coma.

Therefore from the foregoing it is evident that the restricting of the diet as regards carbohydrates, in diabetics,

must be undertaken with caution and with a thorough understanding of the physiology and pathology of metabolism, combined with a careful study of each individual case. No schematic treatment should be employed but rather each case treated upon its own merits.

The next series of experiments carried out, consisted in the taking of acid, in addition to the diet, to determine the effect on the metabolism.

In two experiments (Tables XXXIX) the diet was a mixed one, but creatin-free, in the third it was a protein-poor , but a carbohydrate- and fat-rich diet also creatin-free. To these diets were then added definite quantities of standard hydrochloric acid, as described in the tables.

When Tables XXXIX-XL are examined it is seen that there is a rise in the total nitrogen on the acid days; this increase of the total nitrogen was due to an increase in the urea-and ammonia-nitrogen. The other nitrogenous constituents were unaffected by the ingestion of acid. In TableXXXIXthere is a slight excretion of creatin at the beginning of the first experiment; the excretion of that substance is increased as a result of the administration of the hydrochloric acid ; it disappears,however,on the addition to the diet of sodium bicarbonate, in place of the acid. The rise of the urea and ammonia induced by the acid, is changed to a fall as a result of giving sodium bicarbonate on the 29·1.

The results of Tables XXXIX are interesting when compared to Table XL in that the increase of the urea

TABLE XL

Urine	Total -N	Urea-N	Ammonia -N	Uric acid -N	Creatinin -N	Creatin -N	Acidity	Ammonia+ amino ac. -N	Notes.
cc.	gms.	gms. %	gm. %	gm. %	gm. %	%	cc.	gm.	
2000	12·09	–	·794 6·5	–	·365 3·0	0	360	1·050	Diet-Porridge, toast and eggs.
2000	9·13	–	·520 5·6	–	·451 4·9	0	220	·686	Diet- Tapioca, potatoes
1500	6·09	–	·285 4·6	·095 1·5	·432 6·9	0	142	·404	butter and apples.
1500	5·67	–	·294 5·0	–	·395 6·9	0	157	·409	,,
1700	4·59	2·70 58	·246 6·9	·105 2·3	·374 8·1	0	102	·412	,,
1500	4·11	2·61 63	·285 7·2	·095 2·3	·392 9·5	0	142	·430	,, Wt.-65·4kilos.
2000	5·09	3·21 63	·368 8·4	·102 2·0	·374 7·3	00	200	·476	* ,,
2300	4·89	2·87 59	·411 9·1	·092 1·8	·390 8·0	0	207	·603	** ,,
2200	4·86	3·04 62	·466 6·5	·082 1·6	·371 7·6	0	247	·697	*** ,,
2000	3·92	2·37 60	·256	·102 2·6	·369 9·1	0	125	·469	,,

* besides the diet got 15cc. HCl at noon and 15cc. HCl (normal) at 5.30p

** ,, ,, ,, ,, 25cc. ,, ,, ,, ,, 25cc. HCl (,,) at 5pm.

*** ,, ,, ,, ,, 25cc. ,, ,, ,, ,, 25cc. HCl (,,) at 5'30p

25cc. $\frac{N}{1}$HCl (or normal) is equal to 0.35gm. NH_3-N.

and ammonia is not so marked in the experiment with a carbohydrate-rich diet as with a protein and carbohydrate diet after the administration of acid, even although the amount of acid given in the former exceeded per diem that given on the protein and carbohydrate diet.

SECTION V.

THE EFFECT OF THYROID FEEDING

on

CARBOHYDRATE METABOLISM.

SECTION V.

THE EFFECT OF THYROID FEEDING ON CARBOHYDRATE METABOLISM.

As has been pointed out in Section III., thyroid administration to a normal dog, causes a lowering of the limit of assimilation for carbohydrates. Apart from this experimental evidence and clinical observations on diseased conditions of the thyroid, in certain of which there has been an associated glycosuria, there is no definite knowledge of the influence of the thyroid on carbohydrate metabolism.

In order to elucidate this subject I have carried out a number of experiments on rats and cats; in these I examined the glycogen content of the liver, and the sugar-content of the blood after the administration of thyroid gland by mouth. The experiments were carried out in the following manner; the animals were put on the ordinary diet, and then one or more thyroid lobes were given. This was again repeated the next day. The animals were then fed the following day, the food taken away after they had eaten as much as they cared for, and the animal then killed three or more hours after this. The liver was at once removed, weighed and treated as described in the analytical methods.

The sugar content of the blood was only carried out in the case of cats, from whom the blood was first removed,

TABLE XLI

Rat	Weight of rat	Weight of liver.	Total Glycogen	Hours killed after meal.	Notes.
	gm.	gm.	gm.		
1.	182	7·5	·128	2·5	Control rat.
2.	204	8·5	·125	5·0	,,
3.	147	5·5	·029	7·0	,,
4.	143	7·0	·300	3·0	,,
5.	130	5·1	·192	5·5	,,
6.	130	4·5	·084	7·5	,,
7.	180	6·0	·175	3·0	,,
8.	164	6·1	0	3·0	Thyroid rat.
9.	160	5·1	0	5·0	T.R.
10.	193	6·6	0	7·0	T.R.
11.	200	7·5	·046	9·0	Control rat.
12	135	6·3	·368	2·5	Control rat.
13.	122	6·3	·292	2·5	Pituitary(ant.)rat.
14.	140	5·1	·048	7·0	Control rat.
15.	100	3·5	·011	7·0	Pituitary(ant.)rat.
16.	171	5·9	·191	3·0	Control rat.
17.	147	5·6	·140	3·0	Corpus luteum rat.
18.	130	4·9	·018	7·0	Control rat.
19.	122	4·0	·023	7·0	Corpeus luteum.

before removal of the liver.*

In Table XLI the glycogen content of the liver is given for various control rats, killed at different periods after the last meal. It will be seen that there is a gradual diminution of the glycogen in the liver, the longer the animal is allowed to live after the meal before being killed. When the rats had been fed with thyroid, there was a total absence of glycogen from the liver even if the animal was killed three hours after the last meal, whilst with the control rats there always was a large amount of glycogen present. This experiment has been repeated since with the same result.

On Table XLI will be seen the results of experiments on rats, previously treated with corpus luteum, or with anterior lobe of the pituitary body; in neither case is there any disappearance of glycogen from the liver, as a result of the ingestion of those substances.

In Table XLII will be found estimations of the glycogen content of the liver of cats, 1) normal and 2) after treatment with thyroid. It will be seen that here also the amount of glycogen present in the liver of the cat after the administration of thyroid was so small as to be inestimable. In one case(11) there is still some glycogen present in the liver: the cat in this case was killed 12 hours after the last meal; also in the case of the cat 66 (killed 18 hours after the last meal), there was a trace of glycogen present.

On this table are given the figures for the sugar con-

* These blood examinations were in the main carried out by Mr Horne, whose assistance I wish to acknowledge.

TABLE XLII

Cats.	Percentage of blood sugar.	Percentage of plasma sugar.	Notes.	Weight of liver.	Percentage of liver glycogen.
1.	·154	·235?	C.C. killed 3hrs. after last meal	–	–
2.	·235	·357	C.C. ,, ,, ,, ,, ,,	82	0·79 0·66
3.	·2085 ·237	·370	C.C. ,, ,, ,, ,, ,,	78	2·62 3·03
4.	·258	·403	T.C. ,, ,, ,, ,, ,,	78	0
5.	–	·418	T.C. ,, ,, ,, ,, ,,	67	0
6.	–	·597	T.C. ,, 18hrs. ,, ,, ,,	89	Trace.
7.	–	·490	C.C. ,, 18,, ,, ,, ,,	72	3·74 3·78
8.	–	·596	T.C. ,, 3hrs. ,, ,, ,,	–	–
9.	–	·423	T.C. ,, ,, ,, ,, ,,	98	0
10.	–	·367	C.C. ,, ,, ,, ,, ,,	81	3·88
11.	–	·358	T.C. ,, 12hrs. ,, ,, ,,	104	0·37
12.	–	·332	C.C. ,, 12 ,, ,, ,, ,,	72	2·96

T.C. = Thyroid cat.
C.C. = Control cat.

tent of the blood plasma (a few estimations of the blood sugar were also carried out). Those figures show that the sugar content of the blood plasma is higher in cats after thyroid feeding than in the controls untreated with thyroid.

The urine was also tested in some of those cases, but no sugar was found to be present. The results of those experiments are therefore rather striking in that they show a hyperglycaemia of the blood, and yet no sugar excreted in the urine. A similar condition has been noted in diabetics (Falta) in whom the urinary sugar has disappeared, yet on examination, the sugar content of the blood is found to be still very high and remains so for a long period. The explanation given is that "apparently after long continued hyperglycaemia, the kidneys lose their high degree of sensitiveness to a slight increase in the sugar content of the blood" (Liefman), and so do not react when even large quantities are present.

The above researches are being further extended and give promise of a number of interesting results. But even the few results so far obtained show that the thyroid has a very profound effect on carbohydrate metabolism.

It has been shown that the feeding of thyroid leads to certain alterations in the urine indicative of changes in protein metabolism; summarised these are: an increase of the urea, ammonia and creatinin: a decrease of uric acid: and the appearance of creatin, a constituent formerly absent.

Now in starvation or in carbohydrate hunger as was mentioned in the section upon acidosis, there occurs an in-

crease of urea and of ammonia excretion: a fall of creatinin and uric acid (eventually an increase of uric acid) and, again, the appearance of creatin. Creatinuria occurs in experimental acidosis.

I have shown that creatinuria occurs in diabetes= a disorder of carbohydrate metabolism in particular, also in experimentally induced glycosuria (with Phlorhizin).

Further, in feeding thyroid there also occur carbohydrate changes: glycogen is mobilised from the liver: and an increase of the sugar content of the blood - comparable to the anomalous fact that one may have considerable hyperglycaemia even when the glycosuria has disappeared by treatment.

A consideration of these facts may give some information on a question little discussed in the literature namely, what relations if any do these two types of metabolism (carbohydrate and protein) bear one to another ? It is obvious that some that some relation does exist and a probable hypothesis to explain it is the following one -a modification of Cathcart's: under normal conditions the tissues must find a certain amount of carbohydrate to carry on the life processes: now if the carbohydrate fed as such be insufficient, what happens is that the required amount is split off the protein molecule : the nitrogenous residue is,as it were ''unfixed,, and is excreted -partly as creatin. Such a state of affairs exists in diabetes when for some reason the carbohydrate is not available; the protein must therefore be used and this divergence from ordinary metabolism is expressed partly by

creatinuria.

Again, thyroid is known to stimulate metabolism - witness the increased nitrogen excretion etc.: the mode of its action may be that it lessens the stability of the nitrogen-carbohydrate (i.e.protein) molecule - certainly it causes a mobilisation of carbohydrates from liver to blood: again a nitrogenous residue is released, and concurrently creatin appears in the urine. This view was tested in the experiment above described in which abundant carbohydrate was supplied while thyroid was being ingested: the disruption of the carbohydrate moiety from the protein molecule was then unnecessary seeing so much carbohydrate was already available: and accordingly creatinuria did not occur.

SUMMARY.

1. Creatin occurs in the urine of infants and children: it persists later in the female than in the male.

2. The creatinin coefficient is much lower in children than in adults.

3. Creatin ingested by children is partly excreted as such: in the case of the adult none of the ingested creatin appears as such.

Taking these results as indicative of protein metabolism I conclude that the child exhibits differences in that metabolism from the adult.

4. Creatin is excreted by normal women after menstruation, in pregnancy and post partum: but not after the climacteric.

5. Creatinuria occurs in diabetes mellitus, phlorhizin glycosuria, after thyroid feeding, and in acidosis experimentally induced.

6. The feeding of thyroid is associated with changes in the amount of glycogen in the liver and sugar of the blood.

The cause of creatinuria in these conditions is shown to be a disturbance of carbohydrate metabolism.

7. The interference with the estimation of crea-

tinin in urine (and so of creatin) by certain acetone bodies (viz., acetone, diacetic acid, diacetic ether) is described (Analytical methods).

8. The use of chloroform-thymol as a preservative of urine may be followed by misleading results in the estimation of uric acid.

9. The dietetics of diabetes is considered in the light of experimental and clinical work.

The practical significance of these observations is apparent.

Creatin is thus found to be an indicator of protein metabolism: it is further observed to undergo variations in accordance with changes in carbohydrate metabolism: one must therefore conclude that these two species of metabolism meet here at least on common ground and that they should be less regarded as each sui generis and more studied from the point of view of their dependence, as I have attempted in this contribution to the subject.

BIBLIOGRAPHY.

Hofmann, K.B.	Arch.f.path.Anat.	48,	1869
Pouchet,	Jour.de Therap.	7,	
Grocco, P.	Annali di Clinica e di Farmacol,	4,	1886
Rietschel,	Jahrb.f.Kinderheilkunde,	61,	1905
Closson,	Amer.Jour.of Physiol.,	16,	1906
Amberg and Morrill,	Jour.of Biol.Chem.,	3,	1907.
v.Hoogenhuyze u.Verploegh,	Zeit.f.physiol.Chem.,	46,	1905
Schwarz,	Jahrb.f.Kinderheilk.,	72,	1910
Rose,	Jour.of Biol.Chem.,	10,	1911.
Wolf,	ibidem,	10,	1911
MacCrudden,	Jour.of Experiment.Med.,	15,	1912.
Folin and Denis	Jour.of Biol.Chem.	11,	1912
v.Noorden,	Die Zuckerkrankheit,	4te.Aufl.	1907
Shaffer,	Amer.Jour.of Physiol.,	22,	1908
Folin, O	ibidem,	13,	1905
Pekelharing u. Hoogenhuyze,	Zeit.f.physiol.Chem.,	64,	1910
Benedict and Myers	Amer.Jour.of Physiol.	18,	1907
Meissner,	Zeit.f.rat.Med.	31,	1865
Voit, C.	Zeit.f.Biologie,	4,	1868
Mallet,	Bulletin U.S.office of Experiment.Stan.	66,	1899
Mendel,	Amer.Jour.of Physiol.	13,	1904
Achaelis,	Zeit.f.physiol.Chem.	50,	1906
v.Klercker,	Biochem.Zeit.,	3,	1907
Siven,	Skand.Arch.f.Physiol.	11,	1901

(SECTION-I, §1)
continued.

Burian u.Schur,	Archiv.f.d.gesamt.Physiol.	87 u.94	
Folin,O.	Festschrift f.Hammerston,	3,	1906
Wolf and Shaffer,	Jour.of Biol.Chem.	4,	1908
Weber,	Arch.f.Path.u.Pharm.	57,	1907-8
Hoogenhuyze u. Verploegh,	Zeit.f.physiol.Chem.	57,	1908
Lefmann,	ibidem,	57,	1908
Twort and Mellanby,	Jour.of Physiol.	54,	1912
Mellanby,E.	ibidem,	36,	1907-8

(§2)

Bartels,F.	Zentralbl.f.Biochem.u. Biophysik.	2,	1911
Benedict and Myers,	Amer.Jour.of Physiol.	18,	1907
Marshall,	The Physiology of Reproduction,		1910
Heape,	cited by Marshall, l.c.		
Ewing,J.and Wolf,C.G.L.	Amer.Jour.of Obstetrics,	55,	1907
v.Hoogenhuyze,u. ten Doeschate,	Annales de Gynecologie et d'Obstetrique, Jan.-Fevriei,		1911
v.Leersum,	Biochem. Zeit.(Festsft.Hanburger)		1908
Taylor,A.E.,	Jour.of Biol.Chem.	11,	1911
Shaffer,	Amer.Jour.of Physiol.	23,	1908
Murlin,T.R.,	ibidem,	23,	1908
Mellanby,E.,	Proceeds.of the Royal Soc.,Series B,585,	86,	1913.

(SECTION-II).

Folin, O.	Zeit.f.physiol.Chem.	41,	1901
..	Amer.Jour.of Physiol.	14,	1905
v.Hoogenhuyze u. Verploegh,	Zeit.f.physiol.Chem.	57,	1908
Benedict and Myers	Amer.Jour.of Physiol.,	18,	1907
Leathes,	Jour.of Physiol.	35,	1906-7
Schottin,	cit.by Kraus in V.Noorden's Handb.d.Path.d.Stoffwechsels, (pp.137, vol.2)		1906
Shaffer	Amer.Jour.of Physiol.,	23,	1908
Munk,	Deutsch.Klinik,	30,	1862
Spriggs,	Quat.Jour.of Med.	1,	1907-8
Mellanby,	Jour.of physiol.,	36,	1907-8
Lefmann,	Zeit.f.physiol.Chem.,	57,	1908
Cathcart, E.P.,	Biochem.Zeit.,	6,	1907
..	Jour.of Physiol.,	39,	1909
Hofmann,	Archiv.f.Path.Anat.,	48,	1869
Stejskal u.Erben,	Zeit.f.d.klin.Med.,	40,	1900
Levene and Kristeller,	Amer.Jour.of Physiol.,	24,	1909
Underhill and Kleiner,	Jour.of Biol.Chem.,	4,	1908
Richards and Wallace,	ibidem,	4,	1908
Benedict and Diefendorf,	Amer.Jour.of Physiol.,	18,	1907
Maly, R.L.,	Wiener med.Wochenschr.,	20,	1862
Senator,	Ziemssen's spez.Path.u.Ther. Vol.XIII,		1875

(SECTION-II), contin.d

Hofmann, K.B.,	Virchow's Archiv,	48,	1869
Winogradoff,	ibidem,	27,	1863
Stopczanski, u. Jähtgens,	cited by Senator.		
Rollo, J.	On Diabetes Mellitus, London,		1797
Prout,	On the nature and treatment of stomach and renal diseases,		1848
v. Noorden, C.	Berlin. klin. Wochenschr.,	36,	1903
Lampe,	Zeit. f. phys. u. diät. Therap.		1909-10
Minkowski,	Med. Klinik,		1911
Falta,	Ergebnisse d. inneren Med. u. Kinderheilkunde,	2,	
Magnus-Levy,	Berlin. klin. Wochenschr.		1911
Naunyn,	Diabetes Mellitus, 2te. Aufl.,		1906
Lipetz,	Zeit. f. klin. Med.,	56,	
Luthje,	Therap. d. Gegenwart,		1910
Blum,	Münchener med. Wochenschr.		1911
Klemperer,	Therap. d. Gegenwart,		1911
Falta,	Verhandl. d. Kongresses f. inn. Med., Wiesbaden,		1911
Donkin,	Brit. Med. Jour.,	June,	1874
Winternitz u. Strasser,	Zentralb. f. innere Med.		1899
Mosse,	Revue de Medicine,		1902
Klotz,	Naturforscherkongress in Konigsberg,		1910
,,	Kohlehydratkuren bei Diabetes, Die Würzburger Abhandlungen,		1912

v.Mering, J.	Verhandl.d.Kongr.f. innere Med.		1886-87
	Zeit.f.klin.Med.	14,	1888
	ibidem	16,	1889
v.Mering, J.u. Minkowski, O.	Zentralbl.f.klin.Med.	10,	1889
Pavy, F.W.	Journ.of Physiol.	20,	1896
	''The physiology of the carbohydrates.'' London.		1893
Pavy, Brodie and Sian,	Journ,of Physiol.	29,	1903
Ewing and Wolf,	Amer.Jour.of Obstetrics.	55,	1907
Williams,	Johns Hopkins Hosp. Bullet.	17,	1906
Dumontpellier,	Goitre Exophalmique et Glycosurie, Paris.		1869
Brunton,	St.Bartholomew's Hosp. Reports.	10,	1874
Wilks,	The Lancet.	1,	1875
O'Neill,	ibidem.	1,	1878
Scholz,	Kongr.f.innere Med.		1902
Richter,	Centralb.f.innere Med.		1896
Schöndorf,	Pflüger's Archiv.	67,	1895
Hindhede, M.	Skand.Arch.f.Physiol.	26,	1912
Prausnitz,	Zeit.f.Biol.	29,	1892
Benedict and Higgins,	Amer.Jour.of Physiol.	30,	1912
Cathcart, E.P.	Biochem.Zeit.	6,	1907
Liefman,	Cited by Falta, Harvey Lectures.		1908-9.