T H E S I S

for

THE DEGREE OF DOCTOR OF MEDICINE

THE COURSE OF PROTEIN METABOLISM IN DIABETES MELLITUS AND PHLORIDZIN DIABETES.

by

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P A R T I.

THE COURSE OF PROTEIN METABOLISM in DIABETES MELLITUS.

In spite of a large amount of work by many investigators our knowledge of diabetes is limited, so limited indeed that it is still defined in terms of its most important clinical symptom:- "a chronic disease in which 'grape-sugar' is excreted in the urine" (Von Noorden).

In many previous investigations the output of total nitrogen has been more or less thoroughly studied but without any very important results being obtained. In the majority of instances the patients investigated were not confined strictly to one diet during the period of experiment, with the result that great variations in the output of nitrogen were observed. Further, in many cases the diet during the experimental period was - owing to the nature of the disease - very rich in protein. This

This abundance of protein in the diet naturally leads to a very marked output of nitrogen in the urine, to such a degree indeed that any slight variations which might have occurred from alterations in metabolic processes would be entirely obscured.

Von Noorden, although he recognises that this high elimination of nitrogen is mainly due to the amount of protein ingested, still affirms that a diabetic excretes more nitrogen than does a healthy man on the same diet.

Two causes are put forward to account for this increased excretion of nitrogen:-

(a) Loss of the nitrogen from underfeeding; the diabetic being unable to utilise carbohydrate and thus save protein, an increased catabolism of the latter takes place.

(b) Loss of nitrogen from toxogenous protein catabolism; the clinical picture of diabetic coma suggests that this may occur.

It may be here stated that the results obtained in the present investigation do not bear out this statement of Von Noorden.

Still, as Lusk (1) and others have shown, the output of nitrogen, particularly in relation to this output, and that of the sugar, may yield results of great clinical (prognostic) importance. They have shown that after the removal of the pancreas

pancreas in dogs - whether these be fasting or on a meat diet - there is a constant elimination of nitrogen (N) and dextrose (D) in the urine. (Minkowski), and these two substances are always present in the same proportion every day; there are 2.8 gm.D for 1 gm.N - D: N :: 2.8:1. - Since each gram of nitrogen in the urine represents a destruction of 6.25 gm. of protein in the body, it is evident that 2.8 gm. of dextrose may arise from 6.25 gm. of protein, or protein yields 45 per cent of dextrose. Lusk (l.c.) determined the amount of sugar phlorhidzinised dogs would produce when gelatin was supplied, and found a higher D.N. ratio of 3.65 : 1. which represented a sugar production from protein of nearly 60%. This higher ratio has been found in human diabetes when the patient is on a diet of protein and fat (Mandel and Lusk), and also in dogs when both pancreas and thyroid have been removed (Falta).

The variability of the D.N.ratio in different animals and in the same animal under different circumstances cannot yet be definitely explained. Falta suggests that the extirpation of the pancreas and thyroid gland leaves the adrenals highly active furnishing a secretion which tends to promote the formation of sugar. Mandel and Lusk assume the existence of two different chemical combinations of protein and sugar - an alpha-colloid dextrose representing the sugar in the 2.8 ratio, and a beta-colloid

colloid dextrose representing the additional per centage of the protein when the 3.65 ratio is present, the ratio depending on the combustion or non-combustion of the latter. The higher ratio has been termed the "fatal ratio" as it shows a complete intolerance for carbohydrates, a lower ratio indicates hope for the patient. It is recommended as a matter of routine in practice that the patient should be placed on a strict carbohydrate free diet and on the second day the D.N.ratio determined.

Cathcart (2) in a recent paper has, as the results of many experiments using diets of varied nature, put forward the hypothesis "that carbohydrates are absolutely essential for endocellular synthetic processes in connection with protein metabolism". In his investigation he used the output of creatin - not a normal constituent of healthy urine - as the index of protein metabolism. This substance was chosen because it is constantly present in the urine during starvation (Cathcart (3) Benedict (4)). Cathcart (2) found that when the diet consisted only of carbohydrates or was one comparatively rich in carbohydrates, although it might be relatively low in calories, no creatin was ever found in the urine. He also found that even when the feeding followed a period of starvation,- which led of course to the appearance of creatin in the urine,- that on these diets the creatin immediately disappeared. On the other hand if

if carbohydrates were completely absent from the diet or present only in very small quantity, and although the intake of calories was abnormally high, creatin appeared in the urine. And, further, if the feeding followed a period of starvation the output of creatin instead of diminishing, as when carbohydrates were used, increased in amount. As a result of these experiments the hypothesis above mentioned was formulated, the suggestion being that the presence of carbohydrate in proper form is essential before the anabolic processes in connection with the utilisation of protein in the body can take place. In support of his contention he brought forward much collateral evidence.

An experiment performed on myself at Dr.Cathcart's request has considerable significance in connection with this point. A strict carbohydrate free and creatin-free diet was taken, consisting of 10 eggs, ½ lb. butter and one diabetic loaf (6 oz) The figures obtained were as follows:-

Exam.	Amt.urine c.c.	Sp.Gr.	React	T. N. gm.	Total Creatinin gm.	Creatinin gm.	Creatin gm.
1	sample	1018	Acid	1.3%	0.21%	0.21%	Nil
2	1400	1020	Acid	13.6	1.567	1.5	0.067
3	1200	1018	Acid	17.1	1.536	1.4	0.136
4	1150	1025	Acid	17.0	1.9	1.9	Nil

The first examination represents the analysis of a sample of urine when on ordinary mixed diet.

The second and third are the results obtained while the special diet was being taken. As will be seen creatin was found.

The

The fourth examination was made after the addition of carbohydrate to the diet. Creatin had disappeared.

Another exactly similar experiment carried out on Dr.Cathcart had the identical results.

It was thought that as diabetes is the disease in which carbohydrate metabolism is notoriously faulty (v.s.) that creatin would be present in the urine of diabetics in large amount if the above hypothesis of Cathcart were correct. This has proved to be the case. It was further thought that alterations in the output of creatin might prove of value from the prognostic standpoint; that a diminution in the output would indicate that the utilisation of carbohydrate was improving, or if the output increased, that the disease was increasing in severity. This expectation has not, however, been realised.

In the first place a brief mention will be made of the modern work on the source and the nature of the carbohydrate metabolism in diabetes.

In a paper entitled "Diabetes in the light of modern Physiology", Noël Paton (5) gives a resumé of Pflüger's paper "Glykogen". In this it is maintained that carbohydrates are the sole source of glycogen whether the carbohydrate is in the free condition or linked to proteins in the glycoprotein molecule. It

It is denied that proteins are a source of glycogen, and the possibility of fat being a source of the carbohydrates of the body is considered unproved. The conversion of glycogen to sugar is looked upon as being due to a zymin which is largely under the influence of nerve stimulation. Pancreatic diabetes is explained on the theory that the increased amount of sugar in the blood causes the secretion of a substance in the pancreas which decreases sugar production in the liver - either by acting on the sugar centre in the medulla or on the substance of the liver cells themselves. The conclusion that liver glycogen is changed to sugar as that substance is required by the muscles is in accord with Bernard's theory, and in opposition to that of Pavy. MacLeod (6) in the chapter on the Metabolism of the Carbohydrates, discusses at some length the derivation and fate of glycogen in the organism. Its distribution in the various tissues is widespread, but the liver and muscles are looked upon as the largest depôts, and it probably can be independently formed in the liver and other tissues in which it is found. The question of sugar formation from protein and fat is also discussed, and the evidence is clearly in favour of such a conversion. The primary cause of the accumulation of sugar in the blood is ascribed to the withdrawal from the organism of some influence

influence necessary for the utilisation of dextrose; the influence in question being an internal secretion furnished by the pancreas.

More recently Graham Lusk (l.c.) has contributed an interesting paper on Metabolism in Diabetes. Diabetes Mellitus is here defined as a condition in which the power to burn sugar within the organism is partly or completely destroyed, and must not be confounded with glycosuria.

Claude Bernard's celebrated experiment of puncture of the floor of the fourth ventricle led to the belief that Diabetes was essentially of nervous origin (cf. Pflüger's "Glykogen"). Dock disproved this by showing that puncture does not cause diabetes in fasting animals, i.e. in animals whose tissues are glycogen free. It is therefore evident that nerve impulses from the so-called diabetic centre simply reduce the capacity for holding glycogen on the part of the liver, and perhaps of other organs, with the result that the blood is flooded with sugar which is eliminated by the kidneys.

In support of this the experiments of MacLeod and Dooley (7) are of interest. They used injections of nicotine which inhibit the transmission of impulses through the sympathetic ganglia, and found that stimulation of the diabetic centre was not followed by glycosuria or loss of liver glycogen.

In

In phloridzin glycosuria - discovered by Von Mering - the blood passing through the kidneys has no power to retain its sugar. In this form of diabetes small quantities of ingested sugar are eliminated in the urine (8), but if large quantities are ingested the organism is found fully able to burn sugar.

Von Mering and Minkowski removed the pancreas in dogs and found a condition analogous to diabetes mellitus in man, i.e. there is hyperglycaemia, glycosuria, ingested dextrose is not burned but eliminated, acidosis and possibly death from coma. Diabetes does not develop if a part of the pancreas is left, nor if a part of the gland be ingrafted, so long as the graft remains functional.

In depancreatised dogs the liver is free from glycogen, and ingested dextrose cannot be converted into glycogen, but when levulose is given glycogen may be stored in the liver. The capacity for glycogen formation is therefore intact.

When dextrose cannot be burned in the organism the synthesis of glycogen from dextrose is in abeyance,,the conversion of glycogen into sugar is normal.

Cold, which brings about an increase of heat production by an increased combustion of fat - does not increase the output

output of sugar in pancreas or phloridzin diabetes, neither does mechanical work. Hence the quantity of fat metabolised is apparently without effect on the output of sugar.

In the acute forms of diabetes mellitus there is complete loss of power to burn dextrose, and probably the tissues do not retain glycogen. Such an organism must exist at the expense of protein and fat as sources of its potential energy. But in diabetes a major portion of the ingested protein is convertible into sugar which is carried away in the urine, and this may contain a large proportion of the potential energy of protein available for cell life. To compensate for this the protein metabolism increases (as much as three to five times); but fat metabolism remains the mainstay of the life of the diabetic as it does of the fasting individual. In diabetes the protein metabolism is abnormal, and conditions varying in severity also arise in which the end-products of fat metabolism, such as beta-oxybutyric acid, aceto-acetic acid and acetone, do not burn, but accumulate within the organism and are eliminated in the urine.

Voit believed that protein breaks up into a nitrogenous portion convertible into urea, and a non-nitrogenous portion which as sugar or fat can be used by the organism. Kossel, however, has pointed out that the amino-acids are convertible into

into dextrose, and that they probably are the source of urinary sugar in diabetes. Friedrich Müller is also of this opinion.

Stiles and Lusk (9) fed a phloridzinised dog with a pancreatic digest of meat which had been digested till it was biuret free, i.e. it consisted only of amino-acids. The result was a large production (40%) of sugar from the amino-acids ingested.

Embden and Salomon have given asparagin, glyco-coll, and alanin to partly depancreatised dogs, and have noted large increases in the amount of urinary sugar. Their experiments however, are wanting in completeness in that the pancreas diabetes was not a total diabetes, and the same criticism is justified concerning similar experiments on phloridzinised dogs.

The true situation was first appreciated by Neuberg who found glycogen in the liver and lactic acid in the urine of a normal rabbit following the ingestion of alanin. The amino-acid alanin is converted into lactic acid by hydrolysis with elimination of ammonia. The ammonia is converted into urea. Mandel and Lusk (10) have shown that d-lactic acid is completely converted into dextrose in the organism, and recently a diabetic dog fed with 20 gm. of i-alanin completely eliminated this in the form of urinary sugar.

Lusk (11) has further shown that glutamic acid is convertible

convertible into sugar in so far as it can form alanin in the organism.

The chemical process by which this synthesis of glucose from i-lactic acid is brought about is difficult to conjecture. If the lactic acid is converted into acetol, and finally into formaldehyd, the formation of dextrose could take place because the liver can build glycogen from formaldehyd.

Lusk (1) has dealt fully with the question of fat metabolism in diabetes, and the origin of beta-oxybutyric acid and its derivatives. These are specially found in the normal body during fasting, they cannot therefore be derived from dextrose as was formerly supposed. It has been shown that when the organism is thrown suddenly from a carbohydrate regimen to a combustion of fat the acetone bodies appear in the urine, and this condition is intensified in diabetes.

Von Noorden states that acetone is a by-product of protein via fatty acids, and is formed in the liver, its presence being due to an absence of proper carbohydrate metabolism.

In extreme diabetes the hopeless picture of perverted metabolism is described by Lusk (l.c.) as follows:- "Sugar cannot burn, fats burn only as far as beta-oxbutyric acid, and as for protein a part of its amino-acids are converted into sugar, and another part into beta-oxbutyric acid, neither of which can be burned".

The

The literature shows that a certain number of observations have been made in connection with the excretion of creatinin in diabetes. Creatin on the other hand has received less attention. Indeed the only reference found with regard to the latter in diabetes consists of the brief mention of two cases by Shaffer (12) in both of which it was found.

Prior to the introduction in 1905 of Folin's method of estimating urinary creatinin and creatin many observers had worked at the subject. The results are fully discussed by recent writers: Von Noorden, Mellanby (13) Shaffer (l.c.) and others. The latter gives an outline of the more important findings, and adds that these must be accepted with caution on account of the methods employed.

Von Noorden states that it was early recognised that the excretion of creatinin was increased in diabetes, up to 2 gm. per diem; an increase said to be due to the conditions of nitrogenous metabolism obtaining in diabetics; the patient eats a great deal of meat and possibly breaks down his own muscle substance. Im the same section this writer, referring to Folin's paper (14) in which this author demonstrated that the urinary creatinin has both an endogenous and an exogenous origin, remarks "that it would be extremely interesting to know whether or not the endogenous creatinin of diabetics ever shows considerable variations

variations from the normal 0.4 gm. - 0.6 gm. per diem."

Bunge (15) states that the patient does not excrete more than a healthy man on a purely meat diet, viz. 2.163 gm. per diem.

Folin (l.c.) first observed that the amount of creatinin excreted in the urine by a normal individual is quite independent of the amount of protein in the food, and of the total nitrogen in the urine; the amount excreted from day to day is practically constant for each individual under similar conditions of health and muscular activity. Shaffer (l.c.) has further found that the hourly excretion of creatinin is also a constant quantity, and is quite independent of the amount of urine passed, or the amount of protein ingested. Folin (l.c.) has also shown that the chief factor determining the amount of creatinin eliminated is the body weight of the individual. Wolf and Shaffer (16) have confirmed a further statement made by Folin and Klercker that the excretion of creatinin is wholly unaffected by the ingestion of creatin.

On the other hand the experiments of Noël Paton (17) and Cathcart (2) have shown that there is in all probability a very close connection between the output of creatinin and creatin. In one experiment of Cathcart's it was shown that with a rise in the output of creatin there was a marked fall in the output

output of creatinin. There is also a certain amount of evidence to show that under certain circumstances the ingestion of food containing creatin is followed by a rise in the output of creatinin. The following short experiment performed on myself bears on this point. A diet containing 480 gms. meat was taken on the day succeeding a creatin-free diet. The figures were as follows:-

Exam.	Amt. Urine c.c.	T. N. gm.	Total Creatinin. gm.	Creatinin gm.	Creatin	Remarks
1	1600	10.7	1.68	1.68	nil	Creatin-free diet.
2	1500	15.7	2.347	2.17	0.177	Flesh Diet

As will be seen creatin was excreted on this abundant flesh diet, and this occurrence was associated with a rise in the total nitrogen and creatinin outputs.

It has already been mentioned that Folin (l.c.) believes that the amount of creatinin excreted in the urine is a constant quantity for the individual and independent of the nitrogen intake or of the total nitrogen passed in the urine.

Noël Paton (18) on the other hand, working with dogs, found a certain relationship between the production of creatinin and the nitrogen intake, and his more recent paper (17) has further confirmed this view. He concludes therefore that the excretion of creatin varies directly but not proportionately with

with the excretion of total nitrogen. Cathcart's figures (2) are mentioned as lending additional support to this opinion.

The figures in the tables accompanying this paper also indicate a connection between the creatinin excretion and the nitrogen intake, and they further show that the output of creatinin in diabetics is by no means always greatly exaggerated even when a highly nitrogenous diet is being taken.

Mellanby (l.c.) found that creatin and creatinin feeding had no effect upon the creatin content of muscle after it had reached a certain saturation point.

Von Noorden gives conflicting figures when considering the effect of diet on the daily output of creatinin: with a mixed diet 0.8 gm. - 1.2 gm. per diem, with vegetable diet 0.61 gm. - 0.86 gm. He quotes MacLeod and Long. The former obtained higher values; with "mixed meat diet" 2.098 gm. and with "creatin-free" food 1.064 gm. The latter found 0.9 gm. in vegetarians. The dietaries of the patients examined for this paper were rarely excessive in flesh content, and in a large majority of the cases a creatin-free food was given; the persistence of creatin in the urine during these periods must have been due to causes other than the food ingested.

The earlier estimates of creatinin excretion during muscular

muscular work vary and are probably of little value on account of the defective methods employed. Von Noorden states that excessive muscular work may cause creatin to appear in the urine. Cathcart, Kennaway and Leather (19) found that even severe work does not cause a rise in the urinary creatinin when the diet is ample. Shaffer's work (20) led him to express the opinion that the amount of muscular activity is in itself entirely without effect upon the amount of creatinin excreted.

Quite recently Graham Brown and Cathcart (21) in a paper in which the literature on this point is carefully reviewed found that stimulation of muscle might actually decrease the amount of total creatinin (creatinin + creatin) extracted. Mellanby (l.c.) on the other hand states that work has no influence on the creatin content of muscle.

So far as this investigation is concerned the influence of muscular work on the urinary creatinin and creatin excretion may be ignored. The patients examined - with one exception - being all hospital in-patients either entirely at rest in bed or only doing the light work of the wards.

Creatin is a product of pure endogenous origin, and is never found in the urine as an excretory product under normal conditions (Folin). It can, however, be readily caused to appear, its chief source of origin being the muscle tissues.

Voit

Voit believed that it was present in urine alkaline when passed (Von Noorden). This has been shown to be incorrect (12).

The latest contribution to the literature on the subject of creatin excretion is an article by Noël Paton (17) who finds that the exact significance of creatin in the economy is still unexplained in spite of a considerable amount of investigation. He shows that in the urine of birds creatin takes the place of creatinin, and is an end product of metabolism, and that increased creatin excretion indicates an increase of muscle catabolism.

The urinary creatin excretion has been examined in certain diseases. In nephritis it is said to be increased and was at one time thought to accumulate in the blood and be a factor in uraemic poisoning (Von Noorden). This, however, has been denied as it probably never occurs in sufficient quantities and it is a less toxic substance than creatinin. Turner (22) refers to recent work by Donath who records the results of his experiments into the convulsion-producing properties of the substances found in the blood and urine of epileptics. When injected into guinea pigs and dogs creatin proved innocuous; creatinin on the other hand produced convulsions.

A sample of urine from a female patient who died of diabetic

diabetic coma was examined, and the creatin value was distinctly high. Total Creatinin 0.0448 gm. %. Preformed Creatinin 0.0224 gm.%, and Creatin 0.0224 gm.%. It has also been noticed that in those cases in which the acetone and diacetic acid tests were most marked the creatin values were highest. (e.g.Table VIII.)

Mellanby (l.c.) tabulates the excretion of creatinin and creatin in various pathological conditions affecting the liver; he found that the output of creatinin in hepatic disease is markedly subnormal on account he believes of depressed hepatic function, while in cancer of the liver creatin is excreted in large quantities; in cirrhosis and engorged livers on the other hand creatin excretion is not affected. This writer is of opinion that the liver is intimately connected with the production of creatin and the excretion of creatinin. The creatin excretion in pathological states other than those already mentioned has been examined by other workers. Shaffer (12) mentions the following conditions in all of which there is probably a breaking down of muscle tissue; acute fevers, the acute stages of exophthalmic goitre, tumour cachexia. The largest output found was in women during the first week post-partum. Finally in the graver forms of blood diseases the creatinin values are low, and creatin has been found in lymphatic leucaemia (Shaffer) and myelogenic leucaemia (Von Noorden).

The

The following then appear to be the conditions under which creatin may occur in the urine as an excretory product. Starvation, (?) excessive muscular work on an insufficient diet, heart disease with failure of compensation, certain forms of liver and blood diseases, nephritis, Grāves disease, fevers, tumour cachexia, epilepsy, and during the early days of the puerperium.

In the series of cases examined for this paper all these conditions may be excluded.

The methods employed for the various examinations connected with this paper were:- Total nitrogen (Kjeldahl), creatinin and creatin (Folin), Sugar (Pavy). In order to acquire familiarity with these methods three patients suffering from diabetes were taken. These were all undergoing hospital treatment and were improving. The figures are not tabulated, but it may be stated that creatin was always found in the urines, and that the total nitrogen and creatinin excretions were not high. An attempt was made - as will be noted in the various protocols - to test the various patients with a diet which was absolutely free from either creatinin or creatin. Such a diet, free from carbohydrate and yet palatable, was impossible. Accordingly from time to time a diet was used which - although free

free from creatinin and creatin - was rich in carbohydrate. As such a diet is quite unsuited for a diabetic patient the results obtained were on the whole very unsatisfactory. This diet consisted of:- eggs, milk, cream, oatmeal, cheese, butter, diabetic bread, water. The tables may now be appended.

TABLE I. Female aet.53. Weight 7 st. 10½ lbs.

Exam.	Amt urine cc	Sp.Gr.	React	Sugar gm.	Total Nitrogen gm.	Total Creatinin gm.	Creatinin gm.	Creatin gm.	Remarks
1.	7,360	1045	Acid	268	39.7	1.008	0.883	0.125	On ordinary mixed diet.
2.	4,460	1033	Acid	85.6	24.9	1.517	0.624	0.893	Third day after diabetic diet begun
3.	4,206	1034	Acid	91.0	23.9	1.345	0.715	0.630	do. do.
4.	4,265	1040	Acid	125.4	24.3	1.531	0.597	0.934	Creatin-free food previous day
5.	6,423	1039	Acid	122.8	27.3	1.284	0.661	0.623	do. do.
6.	3,922	1039	Acid	121.3	19.6	0.901	0.431	0.470	do. do.
7.	2,842	1032	Acid	67.6	21.5	1.705	0.824	0.881	After diabetic diet for 4 days.
8.	2,750	1032	Acid	65.2	20.7	1.682	0.815	0.867	do. do.
									Weight 7 stones 11 lbs.

Notes on Case I.

This patient had been the subject of diabetes for six years. Before its onset she was a big well-nourished woman weighing 15 stones (her own statement).

Polyuria was often excessive, and the output of sugar large. Acetone frequently occurred. There was well marked double cataract. The urine was first examined soon after her admission to hospital while she was still on an ordinary mixed diet. The sugar and total nitrogen eliminated were both high. The total creatinin on the other hand rather low. The influence of withholding carbohydrate is seen at the 2nd. and 3rd. examinations, the total creatinin values are distinctly higher, the creatin readings showing a marked increase.

On creatin free food a gradual fall in the excretion of creatinin and creatin may be observed, but the glycosuria increased and the diet was not persisted in.

The last two examinations were made after a return to diabetic diet; the total creatinin values were again increased. The weight was fully maintained during the period of observation.

TABLE II. Female aet 16. Weight 7 st. 6½ lbs.

Exam.	Amt. Urine c.c.	Sp.Gr.	React.	Sugar gm.	Total Nitrogen gm.	Total Creatinin. gm.	Creatinin. gm.	Creatin gm.	Remarks
1.	960	1034	Acid	28.5	7.4	0.562	0.531	0.031	Diabetic Diet
2.	1,620	1033	Acid	44.9	14.7	0.465	0.453	0.012	Creatin- -free diet
3.	1,136	1033	Acid	39.8	12.2	0.370	0.329	0.041	do. do.
4.	1,648	1035	Acid	41.2	15.9	0.388	0.316	0.072	do. do.
5.	995	1029	Acid	14.2	8.4	0.576	0.548	0.028	Diabetic diet
6.	950	1028	Acid	12.1	7.9	0.583	0.554	0.029	do. do.
									Weight 7 st. 6¼ lb.

Notes on Case II.

The first examination was made after the patient had been in hospital for three weeks. The glycosuria still persisted but the other symptoms were in abeyance. The history dated back some ten months, but at no time had the illness been acute and she was still fairly well nourished. On ordinary diabetic diet the first analysis showed a low output of total nitrogen and creatinin. On creatin-free food the amount of total nitrogen eliminated suddenly shot up, and this was associated with a rise in the output of sugar and creatin. Creatinin itself rather diminished. The sudden rise in the total nitrogen and creatin excretions during the period creatin free food was being given may be explained by the fact that the diet was badly borne, and weight rapidly lost (five and one half lbs. in five days). The two final examinations were made after the loss in weight had been made good, and the patient again on strict dietetic treatment. The total nitrogen output was again low, as was also that of creatinin.

TABLE III. Male aet. 39. Weight 13 stones, 9 lbs.

Exam.	Amt. urine c.c.	Sp.Gr	React.	Sugar gm.	Total Nitrogen gm.	Total Creatinin gm.	Creatinin gm.	Creatin gm.	Remarks
1.	3,000	1028	Acid	47.8	32.4	2.88	1.68	1.2	Diabetic Diet
2.	2,500	1031	Acid	35.4	37.5	3.74	2.22	1.52	do. do.
3.	2,600	1027	Acid	36.3	42.6	3.85	2.22	1.63	do. do.
4.	13,000	1030	Acid	335.6	56.6	3.21	1.98	1.23	Ordinary Mixed Diet
5.	15,690	1033	Acid	506.7	47.07	2.28	1.47	0.81	do. do.
6.	10,000	1038	Acid	240.	39.2	3.78	1.60	2.18	do, do,
7.	11,080	1035	Acid	250.	65.3	4.66	2.59	2.07	do. do.
8.	10.520	1037	Acid	311.5	37.8	2.89	1.77	1.12	do. do.
									Weight 13 st.8 lbs.

Notes on Case III.

This was a case of Acromegaly Diabetes. Acromegaly and Diabetes are so frequently associated that some close connection between the two diseases must exist. Von Noorden mentions five cases of acromegaly, four of which were complicated with diabetes. Various explanations to account for this relationship have been put forward. Some maintain that two entirely separate pathological conditions co-exist; others believe the glycosuria occurring in acromegaly to be produced by the pressure of the pituitary tumour on the fourth ventricle. A third view is that it is due to loss or increase of an internal ferment. Mayo Robson and Cammidge (23) state that chronic pancreatitis has been repeatedly observed in association with acromegaly, and are of opinion that the glycosuria in this disease is due to morbid changes in the pancreas.

When the patient first came under observation there was no glycosuria, but it soon appeared with all the classical symptoms of diabetes. The first three examinations were made when diabetic treatment was being tried, and the values of total nitrogen, creatinin and creatin were all high. The patient then developed diphtheria and it was four months later that the subsequent

subsequent examinations were made. The enormous polyuria was the most striking symptom on his return, together with an appetite and thirst that can only be described as insatiable. On account of the latter no diet could be rigidly adhered to. He passed great quantities of sugar, the maximum being reached at the fifth examination. The total nitrogen eliminated reached great heights, the figure at the eighth examination being particularly noticeable, at the same time the total creatinin (creatin + creatinin) touched the highest level.

Creatin was always found in considerable amount.

It is worth observing that in spite of the man's symptoms the weight did not alter materially, and apart from the excessive craving for food he was quite comfortable.

TABLE IV. Male aet. 30. Weight 9 st. 12 lbs.

Exam.	Amt. Urine c.c.	Sp. Gr.	React.	Sugar gm.	Total Nitro-gen gm.	Total Creat-inin gm.	Creat-inin gm.	Creatin gm.	R e m a r k s.
1.	1,050	1020	Acid	nil	18.1	1.843	1.57	0.273	Rich meat diet carbohydrate free
2.	920	1022	Acid	nil	16.7	1.267	1.16	0.107	do. do.
3.	1,365	1018	Acid	nil	21.8	0.84	0.84	0.00	Eggs, milk, less meat.
4.	1,194	1020	Acid	nil	21.4	0.79	0.79	0.00	do do.
5.	910	1025	Acid	5.0	15.7	0.618	0.58	0.038	Creatin free diet ?
6.	1,120	1027	Acid	8.3	16.3	0.613	0.57	0.043	do. do.
7.	870	1018	Acid	nil	16.9	0.948	0.948	0.00	Mixed diet with limited carbohydrate. Weight 9 st. 12½ lbs.

Notes on Case IV.

This was apparently a case of so-called alimentary glycosuria, one in which a fair tolerance for carbohydrates existed. Lusk (v.s.) has pointed out that this condition must not be confounded with true diabetes. It occurs when the sugar holding capacity of certain organs is reduced or over-strained as when an excessive quantity of sugar is ingested.

When the patient first came under observation sugar had been absent from the urine for eight days; this disappearance took place after twelve days treatment. The diet consisted of protein and fat, no carbohydrate being given. The presence of creatin at this time may have been entirely due to the excess of meat ingested (v.s.), but it is probable that the want of carbohydrate in the diet was also a determining factor. When eggs were substituted for some of the meat and milk added the creatin disappeared, and at the same time the total creatinin diminished.

A creatin free diet was now given and sugar again appeared in the urine. Creatin also reappeared, but the total creatinin excretion was somewhat reduced.

It is difficult to explain the reappearance of creatin on this carbohydrate rich diet, especially when the experimental work on dogs, referred to later, is taken into account

account. But in this case there is probably some definite pathological state underlying the condition, a state which only permits of the combustion of a certain (limited) amount of carbo-hydrate. If this limit be over-stepped hyperglycaemia and glycosuria occur, and it may be that at the same time some catabolic process (possible toxic) is originated which leads to a breaking down of muscle protein and the appearance of creatin in the urine. The last examination made in this case was during a period of ordinary diet with limited carbohydrate. Both sugar and creatin were absent, and the total creatinin value was lower than that found when no carbohydrate was allowed.

TABLE V. Female aet. 3. Weight 27½ lbs.

Exam.	Sp.gr.	React.	Sugar gm. per 1000	gm. % Total Nitrogen	gm. % Total Creatiin	gm. % Creatinin	gm. % Creatin	R e m a r k s
1.	1028	Acid	20.0	0.42	0.023	0.015	0.008	Ordinary Mixed diet
2.	1028	Acid	20.8	0.33	0.022	0.014	0.008	do. do.
3.	1029	Acid	21.7	0.40	0.056	0.017	0.039	Creatin free diet
4.	1032	Acid	25.0	0.53	0.022	0.010	0.012	do. do.
5.	1033	Acid	29.4	0.58	0.0267	0.0087	0.018	do. do.
6.	1035	Acid	33.2	0.73	0.0134	0.0056	0.0078	do. do.
7.	1033	Acid	30.0	0.79	0.0159	0.007	0.0089	do. do.
8.	1030	Acid	28.7	0.99	0.0236	0.015	0.0086	After two days on ordinary diet Weight 28 lbs.

Notes on Case V.

The first point of interest here is the age of the patient. On account of this the output of urine for the twenty-four hours couod not be ascertained, and the table represents percentage values except for the sugar which is expressed in grams per 1000 c.c. Diabetes at this age is rare. Amongst 1,360 private patients who consulted Pavy the disease began under ten years of age in only eight cases. The youngest patient Pavy had under his care was an infant aged 12 months (24)

The history in this case was of 8 weeks duration only, the chief symptom noted by the mother being excessive thirst.

It was easy to keep this patient on a creatin-free diet, but the glycosuria tended to increase. The total creatinin output, however, diminished.

The course of the illness was rapid, death ensuing from coma, at the end of six weeks after she first came under observation.

TABLE VI. Male aet. 32. Weight 7 st.13 lbs.

Exam.	Amt. Urine c.c.	Sp.Gr.	React.	Sugar gm.	Total Nitro-gen gm.	Total Creat-inin. gm.	Creat-inin gm.	Creatin gm.	R e m a r k s
1.	2,103	1025	Acid	55.1	10.6	0.455	0.428	0.027	Diabetic diet
2.	2,160	1028	Acid	50.5	15.4	0.619	0.591	0.028	do. do.
3.	2,100	1028	Acid	44.6	13.8	0.612	0.584	0.028	do. do.
4.	3,520	1030	Acid	102.0	18.3	0.564	0.529	0.035	Creatin-free Diet
5.	3,200	1032	Acid	112.0	10.9	0.331	0.280	0.051	do. do.
6.	3,240	1032	Acid	111.7	10.4	0.298	0.247	0.051	do. do.
7.	2,100	1028	Acid	41.4	9.7	0.425	0.402	0.023	Diabetic Diet, modified Weight 8 st.0 lbs.

Notes on Case VI.

This patient had been in hospital for five weeks before he came under observation. He was doing well on a strict diabetic diet, but the glycosuria was still marked. Although the intake of flesh was considerable, the readings are not high; the total creatinin excretion being low, and the nitrogen elimination well within normal limits. On a creatin-free diet the amount of urine and sugar passed both increased; the total creatinin output on the other hand was lower. On the day following the application of creatin-free food there was a rise in the output of total nitrogen; this rise was, however, transitory.

The final examination shows the effect of a diabetic diet with the addition of small quantities of carbohydrate. It is again evident from this table that a diabetic even on a rich nitrogenous diet need not excrete excessive amounts of nitrogen and creatinin.

TABLE VII. Male aet.33. Weight 8 st. 6½ lbs.

Exam.	Amt Urine c.c.	Sp.Gr.	React.	Sugar gm.	Total Nitro-gen gm.	Total Creat-inin gm.	Creat-inin gm.	Creatin gm.	R e m a r k s
1.	4,830	1039	Acid	182.4	14.8	0.710	0.648	0.062	Ordinary Diet. Ace-tonuria.
2.	3,500	1042	Acid	115.5	15.3	1.070	0.850	0.220	do. do. do.
3.	2,700	1040	Acid	91.8	10.05	0.820	0.653	0.167	Ordinary Diet, no acetone
4.	800	1055	Acid	45.0	7.9	0.759	0.338	0.421	Diabetic Diet do.
5.	1,140	1040	Acid	23.1	14.6	1.084	0.534	0.550	do. do. Aceto-nuria
6.	800	1040	Acid	17.6	6.9	0.608	0.242	0.366	do. do. marked acidosis
7.	760	1039	Acid	16.1	6.2	0.787	0.242	0.545	Carbohydrate and alkali do.
8.	1,650	1042	Acid	82.5	11.7	1.025	0.557	0.468	do. do. acet-nuria
9.	3,500	1045	Acid	157.5	22.0	1.869	0.942	0.927	Ordinary diet, no acetone. Weight 8 st. 0½ lbs.

Notes on Case VII.

For some days this patient was seriously ill, and unfortunately left hospital (of his own accord) before his condition was considered safe, and before a creatin free diet could be tried.

The amount of urine passed showed considerable variations as did also the output of sugar and total nitrogen, the latter being generally low.

The acetonuria present at the outset disappeared, and a carbohydrate free diet was given. This reduced the glycosuria but led to an increase in the output of creatin, and a marked acidosis, the breath having the typical smell. Carbohydrates and alkalis were pushed and the acidosis gradually disappeared. The various readings in this case were somewhat irregular, probably on account of the extreme perversion of metabolism present.

TABLE VIII. Male aet. 37. Weight 7 st. 9 lbs.

Exam.	Amt. Urine c.c.	Sp.Gr.	React.	Sugar gm.	Total Nitro-gen gm.	Total Creat-inin gm.	Creat-inin gm.	Creatin gm.	R e m a r k s
1.	3,400	1038	Acid	170	11.9	0.669	0.350	0.319	Ordinary diet; trace acetone
2.	3,400	1035	Acid	163.2	17.0	0.800	0.498	0.302	do. no acetone
3.	3,444	1036	Acid	61.0	17.1	0.986	0.478	0.508	Diabetic Diet do.
4.	3,410	1037	Acid	92.0	15.6	1.031	0.470	0.561	do. do. do.
5.	2,850	1036	Acid	81.2	18.8	1.184	0.652	0.532	do. do. do.
6.	3,000	1040	Acid	78.1	17.9	1.447	0.698	0.749	do. do. Aceto-nuria
7.	3,350	1039	Acid	75.3	17.7	1.517	0.609	0.908	do. do. do.
8.	3,750	1038	Acid	92.5	27.0	1.728	0.738	0.990	Carbohydrate & Alkali - acidosis.
9.	3,700	1040	Acid	85.1	25.2	1.913	0.773	1.14	do. do.
10.	3,100	1037	Acid	71.3	19.9	1.573	0.550	1.023	do. do. (less)
11.	3,500	1038	Acid	157.5	14.7	0.741	0.413	0.328	Ordinary diet. No Acetone Weight 7 st. 6 lbs.

Notes on Case VIII.

Like the preceding case this patient was acutely ill for some days, and in the same way left hospital before a creatin free diet could be given.

He had suffered from diabetes for four years with well-marked symptoms, and at the time of examination was considerably reduced in weight.

On ordinary diet he was progressing favourably, though a large amount of sugar was still being passed. When the urine became free of acetone a diabetic diet was tried. The immediate result of this was to reduce the glycosuria, but a marked and progressive rise in the excretion of creatin took place, together with all the signs of acid intoxication. This high elimination of creatin was most marked when the acidosis was at its height. The effect of a nitrogenous diet on the total creatinin excretion is well shown.

TABLE IX. Male aet. 50. Weight 9 st.4 lbs.

Exam.	Sp.Gr.	React	Sugar gm. per 1000 c.c.	Total Nitrogen gm %	Total Creatinin gm %	Creatinin gm. %	Creatin gm %	Remarks
1.	1038	Acid	28.0	0.30	0.0144	0.012	0.0024	Modified Diabetic Diet
2.	1045	Acid	23.4	0.40	0.0289	0.025	0.0039	do. do.
3.	1042	Acid	37.02	0.36	0.0167	0.013	0.0037	Creatin free diet
4.	1047	Acid	32.06	0.58	0.0328	0.029	0.0038	do.
5.	1043	Acid	35.7	0.40	0.0249	0.021	0.0039	do.
6.	1040	Acid	36.3	0.36	0.0212	0.016	0.0052	do.
7.	1042	Acid	31.2	0.33	0.0197	0.014	0.0057	do.
8.	1040	Acid	33.2	0.40	0.0265	0.019	0.0075	do.
								Weight 9 st.4 lbs.

Notes on Case IX.

This patient was the only case in the present series who was not an hospital in-patient. He was a tailor to trade, and attended at the out-patient department. When first seen he was on a modified diabetic diet, carbohydrates being restricted as far as possible, and under this treatment he was doing well. An effort was made to measure the quantity of urine passed in the 24 hours, but the results were unsatisfactory, so that only percentage amounts are represented in the table. It was apparent, however, that there was considerable polyuria. His illness dated back five years, but had never been severe, and he was always able to follow his employment. The figures obtained are fairly uniform. Creatin was always found, and tended to increase slightly with creatin-free diet, this increase being associated with a rise in the output of sugar.

Male

TABLE X. Aet. 22, Weight 8 st.6 lbs.

Exam.	Amt Urine c.c.	Sp.Gr.	React.	Sugar gm.	Total Nitro-gen gm.	Total Creat-inin gm.	Creat-inin. gm.	Creatin gm.	Remarks
1.	2,900	1039	Acid	80.0	17.1	1.155	0.715	0.440	Diabetic Diet
2.	2,950	1038	Acid	81.2	17.1	1.173	0.716	0.457	do. do.
3.	2,950	1034	Acid	83.6	23.6	0.982	0.581	0.401	do. do.
4.	2.950	1035	Acid	97.3	20.0	0.805	0.486	0.319	Creatin free diet
5.	2.850	1036	Acid	88.3	19.9	0.943	0.561	0.382	do. do.
6.	2,850	1035	Acid	91.2	19.9	0.741	0.451	0.290	do. do.
7.	2,800	1040	Acid	100.8	18.4	0.696	0.414	0.282	do. do.
8.	3,200	1036	Acid	102.4	19.9	0.708	0.464	0.244	do. do.
9.	3,000	1035	Acid	84.0	21.4	1.020	0.504	0.516	Diabetic Diet.
10.	2,800	1038	Acid	81.2	20.6	1,004	0.506	0.498	do. do.
									Weight 8 st.7 lbs.

Notes on Case X.

A history extending over two years was given by this patient. When first seen he was on diabetic diet, but the polyuria and glycosuria were still abundant. The total creatinin output on this diet cannot be considered large, nor was the elimination of nitrogen excessive. On creatin free food the excretion of total creatinin fell, both creatinin and creatin being diminished in amount. With a return to diabetic diet these values both became higher.

TABLE XI. Male aet. 23. Weight 7 st. 8½ lbs.

Exam.	Amt. Urine c.c.	Sp.Gr.	React.	Sugar gm.	Total Nitro-gen gm.	Total Creat-inin gm.	Creat-inin gm.	Creatin gm.	R e m a r k s
1.	7,000	1038	Acid	140	27.3	1.484	0.714	0.770	Ordinary diet, Carbohydrate limited
2.	6,000	1035	Acid	150	24.0	1.512	0.672	0.840	do. do.
3.	6,000	1036	Acid	204	18.0	1.260	0.588	0.672	Creatin free diet
4.	5,500	1037	Acid	191.4	19.2	1.072	0.566	0.506	do. do.
5.	6,500	1036	Acid	208.	19.8	1,084	0.576	0.508	do. do. Weight 7 st. 7½ lb.

Notes on Case XI.

Although this man had only been in hospital for a week he refused further treatment, and a more extended examination was impossible.

Like the previous case he was a poorly nourished man, with a history of about two years duration. He passed a large amount of urine, and the glycosuria was also great.

On ordinary diet the total nitrogen excretion was large, but the total creatinin output was not high. The large proportion of creatin is noticeable. On creatin-free diet the output of total nitrogen diminished, and the creatinin and creatin values were also reduced.

The patient was losing weight, and the prognosis was considered unfavourable.

The above tables represent the majority of the cases examined for this investigation. It will at once be seen that creatin was always present - sometimes to a considerable extent - when sugar occurred in the urine, and this may be taken as evidence of the endogenous protein catabolism which occurs in diabetes. The fact that creatin was found even after a patient had been on a creatin-free diet for some days is sufficient indication that it must be derived from the tissues and not from the food ingested.

The effect of a creatin-free diet was generally to decrease the output of both creatinin and creatin, but the latter was sometimes increased. This is probably due to the fact that a suitable creatin-free diet is (as has already been noted) of necessity rather rich in carbohydrate, and tends to increase the glycosuria, and in some instances upset the patient.

With the exception of the case of acromegaly diabetes the total creatinin eliminated by these patients cannot be considered high; this is not in agreement with the statement of Von Noorden (v.s.) that creatinin excretion is exaggerated in diabetes on account of the amount of meat consumed; even on strict diabetic diet these cases did not eliminate an excessive amount of creatinin.

Folin's

Folin's statement that the amount of creatinin excreted is a constant quantity for the individual and independent of the nitrogen intake or total nitrogen excreted in the urine, is not supported by the figures in the above tables. It must be remembered, however, that Folin refers to the normal individual.

In the same way the greatly heightened total nitrogen output said to occur in diabetes (v.s.) is not observed in this investigation; in table I, the excretion at the first examination was high (39.7 gm.); but this was quite the exception; the majority of the readings were within normal limits, and in a few instances were even lower than might be expected.

The case of acromegaly diabetes must be taken separately; the figures in this case reached great heights; but it is reasonable to suppose that the diseased pituitary gland contributed to this great increase in metabolism, either of itself, or in association with a morbid condition of the pancreas.

As a result of this investigation the following conclusions may be drawn:-

(1) Creatin is constantly found in the urine of diabetics.

(2) The presence of creatin in diabetic urine is an expression of increased endogenous protein catabolism due to inability on the part of the tissues to burn sugar.

(3)

(3) Total creatinin excretion is not abnormally increased in diabetes even when a highly nitrogenous diet is being ingested.

(4) The amount of creatinin excreted depends directly on the nitrogen intake.

(5) The total nitrogen eliminated in diabetes is not always greatly increased even with a comparatively high protein intake, and may even be low.

P A R T II.

PROTEIN METABOLISM IN PHLORIDZIN DIABETES.

As it was found impossible to utilise patients for the investigation of this problem, especially with regard to the influence of various modifications of diet on the output of creatin, it was decided to experiment with dogs on which glycosuria had been induced by injections of phloridzin. The injections were performed by Dr.Cathcart.

The experiments were designed to test the effects of varying quantities of carbohydrate alone, and carbohydrate and fat combined on the protein metabolism during the period of glycosuria.

As is well known phloridzin given by the mouth or injected subcutaneously gives rise to glycosuria in a few hours. The amount of sugar excreted varies considerably in well fed animals, and the D.N.ratio is very inconstant. MacLeod (l.c.) discusses the difference between phloridzin and other forms of diabetes.

In phloridzin diabetes the organism has not lost the power of utilising dextrose to the same extent as in pancreatic diabetes, or in diabetes mellitus (cf.Lusk). In this form of diabetes

diabetes there is no hyperglycaemia, the sugar being formed in the kidney itself out of some precursors contained in the blood. Evidence has been furnished to show that blood protein has a sugar-carrying function, and the serum protein may be regarded as the mother substance of the sugar.

In well fed animals this sugar can be split off from the serum protein without any disruption of the protein molecule occurring, and hence there is no rise in the excretion of nitrogen, provided always that the supply of carbohydrate is sufficient. The protein thus deprived of its sugar in phloridzin poisoning apparently becomes recombined with more of it during its circulation through the rest of the body.

When, however, the animal is starved and no fresh supply of carbohydrate is available for the reconstruction of the gluco-protein molecule, then the poisoned kidney cells attack the protein molecule itself, and thereby liberate not only the sugar bound up in it, but also the nitrogen. That an excessive breaking down of proteins does occur in advanced phloridzin poisoning during starvation is shown by the fact that beta oxybutyric acid and its derivatives make their appearance in the urine.

The tables showing the results of the various experiments are now given:-

TABLE XII. Collie Bitch; Weight 15.050 kilos.

Diet:- Meal 75 gm. Milk 250 cc. Water 500 cc.

Exam.	Amt. Urine c.c.	Sp.Gr.	React.	Sugar gm.	Total Nitrogen gm.	Total Creatinin gm.	Creatinin gm.	Creatin gm.	Remarks
1.	1,000	1012	Acid	nil	5.7	0.424	0.409	0.015	After fast
2.	750	1010	Acid	nil	3.2	0.319	0.319	0.00	
3.	700	1010	Acid	nil	3.0	0.305	0.305	0.00	
4.	925	1028	Acid ft.	15.8	3.35	0.383	0.339	0.044	After injection .75 gm. Phloridzin
5.	1,050	1015	Acid	1.08	4.2	0.360	0.336	0.024	
6.	870	1008	Acid faint	nil	4.8	0.325	0.325	0.00	
7.	1,010	1008	Acid	nil	2.95	0.337	0.337	0.00	
8.	850	1006	Acid faint	nil	2.25	0.260	0.260	0.00	
									Weight 13.750 kilos

Notes on Table XII.

The first analysis in this table shows the result of fasting; there was an increased output of total nitrogen, and creatin was found. On the diet allowed the dog lost in weight but no creatin was found till the glycosuria was established. This occurred six hours after injection, and during this period there was a slight rise in the total nitrogen eliminated. With the disappearance of sugar from the urine creatin also disappeared, the nitrogen output, however, did not fall till the succeeding day. It was evident that sufficient carbohydrate was not being supplied to reconstruct the gluco-protein molecule, and that the protein molecule itself was being attacked. In the next experiment an addition of 25 gm. oat meal was added and more milk allowed.

TABLE XIII. Collie Bitch: Weight 15.100 kilos.

Diet allowed:- Meal 100 gm.
Milk 500 cc.
Water 700 cc.

Exam.	Amt. Urine c.c.	Sp.Gr.	React.	Sugar gm.	Total Nitrogen gm.	Total Creatinin gm.	Creatinin gm.	Creatin gm.	Remarks.
1.	470	1020 concent.	Acid	nil	5.4	0.485	0.482	0.003	After fast
2.	950	1008	Acid	nil	3.9	0.391	0.391	nil	
3.	770	1008	Acid	nil	3.8	0.398	0.398	nil	Fed 10 gm.Glycosamin hydrochlor.
4.	800	1010	Acid	nil	3.7	0.380	0.380	nil	
5.	800	1007	Acid	nil	3.3	0.339	0.339	nil	
6.	1,150	1023	Acid	16.6	4.9	0.488	0.458	0.030	Injected .75 gm. phloridzin previous day
7.	860	1009	Acid	nil	3.9	0.327	0.327	nil	
8.	550	1012	Amph.	nil	3.24	0.304	0.304	nil	Fed 10 gm.glucose
9.	1,100	1010	Acid	nil	5.08	0.486	0.486	nil	
10.	800	1010	Acid	nil	4.5	0.436	0.436	nil	
									Weight 13.50 kilos

Notes on Table XIII.

This table represents the result of the increased allowance of carbohydrate, and it shows that there was still a deficient supply as evidenced by the rise in the total nitrogen excretion and the appearance of creatin after the phloridzin injection. The effects of feeding with 10 gm. glucosamin hydrochloride, and 10 gm. glucose is also shown; the former had no effect on the urinary constituents examined; the latter was followed by a rise in the output of total nitrogen and creatinin, the cause of which has not yet been established.

As in the previous table the result of fasting is shown at the first examination.

TABLE XIV. Collie Bitch: Weight 13.430 kilos.

Diet allowed:- Meal 100 gm.
Tapioca 100 gm.
Milk 500 cc.
Water 700 cc.

Exam.	Amt. Urine c.c.	Sp.Gr.	React.	Sugar gm.	Total Nitro-gen gm.	Total Creat-inin. gm.	Creat-inin. gm.	Creatin gm.	Remarks
1.	355	1030	Acid	nil	4.9	0.374	0.374	nil	Urine scanty and highly concentrated
2.	1,700	1008	Acid	nil	5.9	0.467	0.467	nil	
3.	1,400	1007	Acid	Nil	3.8	0.371	0.371	nil	
4.	1,350	1007	Acid	nil	3.1	0.371	0.371	nil	
5.	1,700	1030	Acid	27.6	3.7	0.482	0.482	nil	Injected .75 gm. phloridzin previous day.
6.	1,400	1017	Amph.	16.4	3.5	0.356	0.356	nil	
7.	1,550	1007	Acid	nil	5.0	0.438	0.438	nil	
8.	1,200	1006	Acid	nil	3.3	0.302	0.302	nil	
9.	1,400	1006	Acid	nil	3.7	0.352	0.352	nil	
									Weight 14 kilos.

Notes on Table XIV.

In this table the result of a considerable increase in the amount of the carbohydrate intake is shown. In contrast to the two preceding tables more sugar was excreted, but the total nitrogen and creatinin eliminations were unaffected, and no creatin was found.

The addition of 100 gm. tapioca to the diet must have supplied the blood with sufficient carbohydrate to prevent the break-down of the protein molecule. The next experiment (table XV.) was a repetition of this one performed on a different dog. The results were practically the same. No creatin was found and there was no rise in the output of nitrogen.

TABLE XV. Retriever Bitch; Weight 23,565 kilos.

Diet allowed:- Meal 100 gm.
Tapioca 100 gm.
Milk 500 cc.
Water 700 cc.

Exam.	Amt. Urine c.c.	Sp.Gr	React.	Sugar gm.	Total Nitrogen gm.	Total Creatinin. gm.	Creatinin. gm.	Creatin gm.	Remarks
1.	1,400	1007	Acid	nil	4.3	0.359	0.359	nil	
2.	1,150	1010	Amph.	nil	4.1	0.450	0.450	nil	
3.	1,600	1008	Amph.	nil	5.1	0.498	0.498	nil	
4.	1,550	1008	Acid	nil	5.1	0.489	0.489	nil	
5.	1,450	1007	Acid	nil	4.7	0.482	0.482	nil	
6.	1,400	1020	Acid	19.19	4.6	0.463	0.463	nil	Injected 1.0 gm. phloridzin previous day
7.	1,300	1008	Acid	nil	3.6	0.418	0.418	nil	
8.	1,300	1008	Acid	nil	4.2	0.514	0.514	nil	
9.	1,400	1007	Acid	nil	4.2	0.465	0.465	nil	
10.	1,300	1006	Acid	nil	3.8	0.526	0.526	nil	
									Weight 22.150 kilos.

TABLE XVI. Collie Bitch. Weight 15.870 kilos

Diet allowed: Meal 100 gm.
Lard 100 gm.
Milk 500 cc.
Water 250 cc.

Exam.	Amt. Urine c.c.	Sp.Gr.	React.	Sugar gm.	Total Nitro-gen gm.	Total Creat-inin gm.	Creat-inin. gm.	Creatin gm.	Remarks
1.	700	1010	Acid	nil	2.9	0.365	0.365	nil	no acetone
2.	1,000	1011	Acid ft.	nil	4.6	0.393	0.393	nil	do.
3.	700	1015	Acid ft.	nil	3.8	0.376	0.376	nil	do.
4.	500	1012	Acid	nil	2.6	0.318	0.318	nil	do.
5.	900	1020	Acid	13.6	3.2	0.476	0.448	0.028	Injected .75 gm. phlorhizin previous day do.
6.	500	1011	Amph.	2.5	2.1	0.337	0.309	0.028	do.
7.	800	1007	Acid	nil	3.04	0.468	0.468	nil	do.
8.	400	1010	Amph.	nil	1.5	0.274	0.274	nil	do.
9.	300	1011	Acid	nil	1.2	0.204	0.204	nil	Weight 15.870 kilos do.
10.	300	1012	Acid	nil	5.4	0.395	0.282	0.113	Fed raw meat 350 gm
11.	600	1021	Acid	nil	9.6	0.445	0.424	0.021	Ordinary food ad lib
12.	900	1009	Acid	nil	4.4	0.442	0.442	nil	
13.	1,100	1008	Acid	nil	3.8	0.519	0.519	nil	

Notes on Table XVI.

In this experiment 100 gm. of lard were substituted for the tapioca given in the two preceding ones; 100 gm. of oatmeal being still allowed. This diet was arranged to test the value of fat as a protein sparer. It will be seen from the table that this diet did not prevent the animal breaking down her own tissue protein as indicated by the appearance of creatin during the time the glycosuria lasted. There was, however, no marked increase in the output of total nitrogen.

Towards the end of this experiment the dog was fed with 350 gm. raw meat. Abundant creatin appeared together with a marked rise in the elimination of total nitrogen, and a rise in the output of preformed creatinin. This experiment is evidence against the contention of Folin and others that the output of creatinin is unaffected by the ingestion of creatin in the food. (No preformed creatinin, only creatin, is present in raw meat).

ft. = faint.

TABLE XVII. Collie Bitch, weight 16,852 kilos.

Diet allowed:- Meal 50 gm.
Lard 150 gm.
Milk 500 cc.
Water 250 cc.

Exam.	Amt. Urine c.c.	Sp.Gr.	React.	Sugar gm.	Total Nitro-gen gm.	Total Creat-inin. gm.	Creat-inin. gm.	Creatin gm.	Remarks
1.	800	1005	Acid	nil	2.8	0.372	0.372	nil	no Acetone
2.	600	1006	Acid ft.	nil	2.9	0.415	0.415	nil	do.
3.	500	1009	Acid ft.	nil	2.5	0.387	0.366	0.021	do.
4.	Sample	1009	Acid ft	nil	0.54 %	0.088 %	0.081 %	0.007 %	Total urine passed lost do.
5.	600	1010	Acid ft.	nil	3.3	0.564	0.528	0.036	do.
6.	800	1022	Acid ft.	21.6	3.0	0.604	0.568	0.036	Injected .75 gm.phlorhizin previous day do.
7.	300	1021	Amph.	7.8	2.4	0.258	0.247	0.011	do.
8.	800	1008	Acid	nil	3.4	0.522	0.487	0.035	do.
9.	650	1007	Amph.	nil	2.2	0.456	0.436	0.020	do.
									Weight 16.250 kilos.

TABLE XVIII. Collie Bitch. Weight 16 kilos.

Diet allowed:- Meal 75 gm.
Lard 150 gm.
Milk 500 cc.
Water 250 cc.

Exam.	Amt. Urine c.c.	Sp.Gr.	React.	Sugar gm.	Total Nitrogen gm.	Total Creatinin gm.	Creatinin. gm.	Creatin gm.	Remarks
1.	500	1014	Acid	nil	4.5	0.315	0.284	0.031	no acetone
2.	500	1011	Acid	nil	3.06	0.332	0.307	0.025	do.
3.	400	1013	Acid	nil	2.8	0.463	0.396	0.067	do.
4.	300	1013	Acid	nil	1.7	0.321	0.297	0.024	do.
5.	900	1019	Acid	16.2	3.6	0.715	0.611	0.104	Injected .75 gm.phlorhizin previous day do.
6.	400	1022	Amph.	6.0	2.5	0.442	0.404	0.038	do.
7.	500	1010	Amph.	nil	2.8	0.467	0.436	0.031	do.
8.	600	1010	Acid	nil	3.8	0.438	0.406	0.032	do. Weight 17 kilos.

The two last tables (XVII. & XVIII.) show the result of an increased allowance of fat with varying quantities of carbohydrate. The increased supply of fat was given to see if the tissue break-down which occurred in experiment XVI. could be prevented. The condition, however, was aggravated as will be seen from the figures.

Creatin appeared both before and after the injection; during the glycosuria it was slightly increased in amount, especially in table XVIII., and in this experiment a rise in the total nitrogen output is also seen. In both tables a decided rise in the total creatinin excretion occurred on the day following the injection.

It appears then that fat, even in considerable quantity cannot prevent protein catabolism, or be a substitute for a deficiency of carbohydrate intake. The calorie intake here was far higher than in the experiments in which the food consisted of carbohydrate. This of course points to the fact that there is some specific action in the various food stuffs, and that the diet cannot be, as certain modern German writers contend, merely reduced to a question of the intake of calories, it being immaterial whether these be obtained in the form of protein, carbohydrate or fat.

Cathcart (2) has fully discussed this question of the specific action of the various food stuffs, and considerable evidence

evidence is adduced to show that one is no longer justified in valuing a diet simply on its calorific value and its content of fat, protein and carbohydrate. The experiments of Cathcart and Landergren show that the power of carbohydrate and fat to spare the break-down of tissue protein is by no means equal. If a diet rich in protein is given and carbohydrate is removed, the calorie value being made up with fat, there is an immediate increase in the endogenous protein catabolism. Landergren found that if he gave a diet made up of pure carbohydrate, practically nitrogen free, the output of total nitrogen steadily fell, but when the carbohydrate was replaced by a diet consisting solely of fat, again nitrogen free, the nitrogen output steadily rose in amount. The experiments of Cathcart fully confirm these observations.

The above tables (XII. - XVIII.) show the important part played by carbohydrates in the metabolism of protein. In animals poisoned by phloridzin an insufficiency of carbohydrate permits of the breaking down of the protein molecule with liberation of its nitrogen. If sufficient carbohydrate is supplied no such destruction takes place.

When an amount of carbohydrate already shown to be insufficient (vide XII. and XIII.) is combined with fat, disruption of the protein molecule still proceeds, and this disruption continues even when the quantity of fat supplied is sufficient

sufficient to increase the weight of the animal.

The conclusions arrived at from these experiments are strongly in support of the hypothesis enunciated by Cathcart and already quoted.

Conclusions.

(1) That a sufficiency of carbohydrate alone is necessary to prevent the destruction of the protein molecule and permit of the complete metabolism of protein.

(2) That fat, alone or combined with an insufficiency of carbohydrate, is unable to save the protein molecule from disruption.

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