CCIV. THE SUGARS OF URINE.
II. FACTORS AFFECTING THE EXCRETION OF
FERMENTABLE AND NON-FERMENTABLE
SUGARS IN URINE.

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THE first paper of this series by West and Peterson [1932] outlines an improved procedure for the determination of fermentable and non-fermentable sugars in urine. Although a number of workers have studied the factors which influence the excretion of these substances, the whole question is in a rather confused and unsatisfactory state. This is largely due to the fact that in much of the older work the methods of analysis were inadequate. This applies especially to the fermentation procedures used, which involved the use of unwashed yeast acting for many hours. Under such conditions it is well recognised that the yeast itself may add an amount of reducing material which may completely offset that removed by the yeast, leading to data which indicate the complete absence of fermentable sugar, or bacterial contamination may cause sugar destruction leading to values for fermentable sugar which are too high. Other sources of error lie in the incomplete removal of non-sugar reducing substances from the urine before analysis and the use of sugar methods incapable of detecting small differences.

Benedict and Osterberg [1918] using $\text{Hg}(\text{NO}_3)_2\text{-NaHCO}_3$ filtrates of urine and the picrate sugar method, studied the effect of diet and starvation upon the excretion of total sugar in the dog. They found this to be highest on a carbohydrate diet, appreciably decreased on a high protein diet and markedly less during starvation. They noted a maximum excretion 4-5 hours, and a minimum excretion about 15 hours, after feeding. They found the output to be independent of urine volume. These workers with Neuwirth [1918] studied both the fermentable and non-fermentable sugars in the urine of two normal men as affected by diet. They found the total excretion to be greater, in general, for a high carbohydrate than for a high protein diet, both fermentable and non-fermentable fractions being increased. Their fermentable values for all experiments on one individual averaged nearly the same as their non-fermentable, something over 500 mg. (calculated as glucose) per 24 hours. The other subject

showed less, but approximately the same proportion of fermentable and non-fermentable sugar. The large proportion of fermentable sugar was perhaps due to long periods of fermentation with unwashed yeast during which there may have been considerable destruction of sugar by bacteria. They concluded that fermentable sugar is always present in normal urine and advanced the idea of a continued "glycuresis" or passage of blood-sugar into the urine.

Folin and Berglund [1922] studied the total sugar of urine and blood after the ingestion of large quantities of various carbohydrates. In determining urine-sugar they used Lloyd-filtrates and analysed them by the method of Folin and Wu [1920]. Fermentable sugar was not determined. They noted an increase in total sugar (Lloyd-filtrates contain much non-sugar reducing material) when passing from a high protein diet to an ordinary mixed diet containing carbohydrate, but when pure sugars (glucose or its derivatives) were ingested in large amounts there was no increased output. Fructose was found to cause an increase, especially if previously heated.

Folin and Berglund concluded that there is no "glycuresis" normally of blood-sugar into the urine as stated by Benedict and that the sugars of normal urine are essentially composed of unassimilable carbohydrate substances of the food, occurring there either naturally or formed by heating.

Greenwald, Gross and Samet [1924-25] studied the effect of diet upon the excretion of urinary sugar in both the dog and man. They used $\text{Hg}(\text{NO}_3)_2$ - NaHCO_3 filtrates and analysed them by four different methods, the two picrate methods of Benedict and Osterberg [1918; 1921], the copper method of Folin and Berglund and that of Shaffer and Hartmann [1920-21]. Fermentable sugar was not determined. From their observations they conclude that the sugars of normal urine are composed of carbohydrates assimilable with difficulty or not at all and of reducing substances (probably pentose) derived from protein of the food and from endogenous sources. They considered at least one-half of the sugars to originate from food protein and endogenous sources. Their experiments tended to show that practically no reducing material arises from the metabolism of nucleic acids or endogenous nuclear metabolism, as shown by absence of increase in urinary sugar after the ingestion of nucleic acids or as the result of turpentine abscesses in the dog. The marked effect of heated carbohydrate material (grape-nuts) in increasing the reducing material of urine was also observed. Later Greenwald, Gross and McGuire [1927] carried out other experiments and used Eagle's [1926-27] fermentation procedure which consisted of incubation with unwashed yeast for 40 minutes. They concluded that while a part of the urinary sugar may arise from heat-altered carbohydrate of food, this fraction is rather small. The opinion expressed in the former paper that pentose derived from protein constitutes a part of the sugar of urine was abandoned. Also they considered the small fraction of fermentable sugar indicated by their data probably to represent a sugar other than glucose. They suggested the idea that some of the urinary sugar may arise from bacterial action in the intestines. No experiments were reported in support of this view.

Höst [1923], using the method of Benedict and Osterberg, studied the excretion of urinary sugar by many human subjects after various diets. He determined fermentable sugar with unwashed yeast acting for 48 hours. He concluded that bread especially contributes to the sugar of urine and that glucose is not present.

The workers cited above have carried out the principal investigations relating to the effect of diet upon urinary sugar. In none of these cases have the methods of analysis employed been capable of giving an accurate estimate of the fermentable and non-fermentable fractions, and in some no attempt at determining fermentable sugar was made. In fact it was a debatable question until recent years as to whether urine contains any fermentable sugar at all. As noted above, Benedict, Osterberg and Neuwirth found considerable quantities; Shaffer and Hartmann [1920-21], using unwashed yeast, failed to observe fermentable sugar in urine, as also did Eagle [1926-27]. After Somogyi [1927] introduced the use of washed yeast for fermentation and the more sensitive sugar reagents had become available, several workers reported the presence of fermentable sugar in urine. Among these were Van Slyke and Hawkins [1929], and Peterson and West [1929]. At the present time, then, some investigators agree that normal urine contains both fermentable and non-fermentable sugar, though there still seems to be no evidence in the literature that the fermentable fraction is glucose. This represents a separate problem which will be considered in the third paper of this series by West and Steiner. Suffice it to say that evidence has been obtained from experiments on the rate of fermentation of isolated sugars of urine in the Warburg apparatus indicating that the fermentable sugar of urine is glucose.

In view of the conflicting opinions arising from this older work the writers have applied their methods of analysis, reported in the first paper, in an attempt to determine more certainly some of the factors influencing the excretion of fermentable and non-fermentable sugars in the urine. Evidently much of the work is repetition of that formerly done by the older methods of analysis. This seemed not only desirable, but necessary, in order that we might continue our studies as to the chemical nature of these sugars.

The factors specifically studied have included the effects of diet, starvation, renal damage, glandular stimulation and constipation.

FACTORS AFFECTING THE EXCRETION OF FERMENTABLE AND NON-FERMENTABLE SUGARS.

Diet.

Non-fermentable sugars. A diet entirely free from substances that will contribute to the non-fermentable fraction cannot be realised. Accordingly, a preliminary study was made to discover the foods which, taken in reasonable quantities, have no marked effect upon the excretion of non-fermentable substances. It was found in brief that white bread, milk, potatoes, sucrose

starch, fats, peas, string beans, tomatoes, citrus fruits, canned pineapple and pears, strawberries, bananas, lettuce, cabbage, celery, eggs, cheese, cream of wheat, meats cooked without cereal, and some other articles of food can be taken rather interchangeably in mixed diets with a practically constant elimination of non-fermentable reducing substances.

A number of experiments on different individuals in which the subjects ate mixed diets of these foods on successive days were run. The results on two individuals are illustrative. Over a period of 7 days, A.C.L. excreted daily a maximum of 360 mg. of non-fermentable sugar (as glucose), a minimum of 312 and an average of 335 mg.; a maximum of 136 mg. of fermentable sugar, a minimum of 96 and an average of 117 mg. In the case of E.S.W. over a period of 5 days a maximum of 351 mg. of non-fermentable sugar, a minimum of 312 and an average of 325 mg. were eliminated. The fermentable sugar output showed a maximum of 158 mg. and a minimum of 111 mg. The values can be made to vary appreciably from these by varying the relative proportions of the different foods taken, though it seems that significantly high values cannot be produced by quantities which the average person cares to eat. Urine volume was found to be not detectably related to the excretion of reducing substances, even with extreme diuresis induced by forced fluids. This is in accord with the observations of other workers.

Muscular exercise appears to be unrelated to the output of the non-fermentable fraction as found by experiments on a subject exercising normally and then resting in bed.

In order to determine the effect of the ingestion of large quantities of different proteins, carbohydrates, fruits and other substances, restricted diets of the above foods were employed. The subject was fed with the restricted diet for 2-3 days previous to the administration of the food or substance to be tested and also for 1-2 days thereafter if a positive effect was noted.

A. The following combinations either supplementing or replacing the restricted diet caused no significant increase and in some instances a decrease of the 24-hour excretion of non-fermentable reducing substances: thirty egg-whites; 280 g. of cheese and 12 eggs; 1000 g. of herring roe; 1125 g. of brains (pig); 900 g. of boiled kidney (beef); 900 g. of boiled liver (pig); 675 g. of pancreas (ox); 675 g. of boiled beefsteak; 560 g. of canned sweet corn (Del Monte); 200 g. of dextrin (Merck's white), 100 g. at 10.30 a.m. and 100 g. at 3.30 p.m.; 600 g. of glucose in 4 portions of 150 g. each during the day; 200 g. of starch and 250 g. of sucrose; 14 g. of yeast nucleic acid; 1200 g. of bananas (net); 1350 g. of white seedless grapes.

B. The foods listed below did cause a marked increase in the output of non-fermentable substances: 500 g. of grape-nuts, 936 mg., 125 % increase; 450 g. of blue label Karo syrup (dark), 643 mg., 68 % increase; 125 g. of caramelised sucrose, 534 mg., 68 % increase; 400 g. of honey in 3 portions, 555 mg., 83 % increase; 450 g. of raisins and 280 g. of dates, 884 mg., 115 % increase; 1350 g. of unpeeled apples, 554 mg., 40 % increase.

It will be seen that the non-fermentable sugars are increased by the ingestion of such things as grape-nuts, dark Karo syrup, honey, caramelised sugar, dates, raisins and apples, but unaffected by pure carbohydrates as starch, sucrose, dextrin and glucose. These findings are in accord with the ideas of Folin and Berglund and others that much of the reducing material of normal urine represents unassimilable foreign and altered carbohydrate substances from the diet. Non-fermentable sugars are not increased beyond the values of the restricted diets by high protein intake. It was noted, however, that meat rolled in flour and cooked so as to yield caramelised carbohydrates greatly increases these substances. In the experiments reported, this was avoided, the meat being either boiled or fried plain. The effect of liver may be large if it is fried instead of boiled, presumably owing to alteration of the carbohydrate contained in it. Nucleoprotein diets and nucleic acid (yeast) caused no definite

increase of non-fermentable sugars. These results are in agreement with the findings of Greenwald, Gross and Samet [1924-25].

Non-fermentable sugars have been determined in several of the foods used. In $\text{HgSO}_4\text{-BaCO}_3$ filtrates of thoroughly macerated beef kidney, liver and muscle 9, 60 and 25 mg. (as glucose) per 100 g. of tissue were found respectively. These were increased by hydrolysis of the tissues for 5 hrs. in 0.5 N H_2SO_4 to 282 mg. in kidney, 229 in liver and 220 in muscle per 100 g. Karo corn syrup (dark), honey and caramelised cane sugar were found to yield considerable amounts of non-fermentable reducing substances both before and after hydrolysis. Ordinary commercial sucrose also gave appreciable non-fermentable reducing materials upon hydrolysis. Human faeces from a restricted diet were found to give about 20 mg. per 100 g. (as glucose) of non-fermentable reducing substances in $\text{HgSO}_4\text{-BaCO}_3$ filtrates.

It seems likely that practically all foods contribute something to this non-fermentable fraction which is apparently made up of a very heterogeneous group of substances essentially non-nitrogenous, and probably chiefly carbohydrate in nature.

The mercury filtrates of urine from different diets have been hydrolysed for 5 hrs. with 0.5 N H_2SO_4 and fermentable and non-fermentable sugars determined. Table I records data obtained and shows that the non-fermentable

Table I. *Effect of hydrolysis for 5 hrs. with 0.5 N H_2SO_4 upon the non-fermentable and fermentable sugars of urine.*

Mercury filtrates of urine				Diet
Non-fermentable sugar total (mg.)		Fermentable sugar total (mg.)		
Before hydrolysis	After hydrolysis	Before hydrolysis	After hydrolysis	
644	620	146	1684	A.C.L. Restricted + 450 g. dark Karo syrup
316	399	142	198	E.S.W. Restricted
515	545	136	1160	E.S.W. Restricted + 125 g. of caramelised sucrose
885	862	243	256	A.C.L. Restricted + raisins and dates
274	279	68	129	E.S.W. Second day of starvation.
305	354	102	230	E.S.W. 1000 g. herring roe
936	826	72	107	E.S.W. 500 g. grape-nuts

fraction is not, on the average, appreciably increased by hydrolysis and is, for the most part, not of a polysaccharide character. Crystalline osazones (apparently a mixture) have been prepared from fermented mercury filtrates of normal urine, controls being run on the yeast. This material melted at 165-170° (uncorr.) with decomposition at 175°, and seemed to resemble the product

obtained by Patterson [1926]. It is interesting to observe that the fermentable substances in HgSO_4 - BaCO_3 filtrates are appreciably increased by hydrolysis, even during starvation. We have found this to be true in a number of experiments not listed in the table. It should be noted that Folin and Berglund [1922] observed an increase in total sugar of urine after hydrolysis as did Greenwald, Gross and McGuire [1927]. These workers, however, did not determine that the increase is chiefly concerned with the fermentable fraction of the sugars. Since this apparently cannot arise from the non-fermentable fraction, the conclusion seems warranted that diffusible non-reducing polysaccharides are normally excreted. It will be pointed out in the third paper that there is probably very little if any fermentable reducing polysaccharide in urine and, bearing the above facts in mind, it seems that the carbohydrates of both normal and starvation urines may be chiefly grouped into the following three classes:

A	B	C
Non-fermentable simple sugars or derivatives	Non-reducing polysaccharide material	Fermentable sugar
Unchanged by acid hydrolysis	Converted into fermentable sugar by hydrolysis	Glucose

The kidneys must effect a relatively very high concentration of the polysaccharide substances from the blood because Scharles and West [1931] have shown the absence of detectable hydrolysable sugar from tungstic acid blood-filtrates. The same thing is true for the non-fermentable reducing fractions of mercury filtrates.

It has regularly been observed that the ingestion of large quantities of sucrose leads to increased values for fermentable sugar. This can be accounted for in part or entirely by the excretion of unchanged sucrose in the urine, some of which is inverted during the acid mercuric sulphate precipitation used in the preparation of filtrates. Sucrose solutions treated with the mercuric sulphate reagent as in the preparation of urine-filtrates have been shown to be considerably hydrolysed. Values for fermentable sugar after the ingestion of quantities of sucrose sufficient to cause sucrosuria, are, therefore, not significant when the urine is precipitated by a strongly acid reagent such as HgSO_4 or $\text{Hg}(\text{NO}_3)_2$.

Fermentable sugar. The state of activity of the pancreas and general ability of the body to metabolise glucose are recognised as important controlling factors in the elimination of fermentable sugar in the urine. For a discussion of this effect the papers of Greenwald, Gross and Samet [1924-25], and those of Heinbecker [1928] and Dann and Chambers [1930] may be consulted. It is generally recognised that carbohydrate starvation as induced by fasting and to a less degree by protein and fat diets decreases the ability of an animal to metabolise carbohydrates. This condition followed by a carbohydrate meal may lead to a glycosuria which is easily detectable by the usual qualitative sugar tests. This effect on the small quantity of fermentable sugar normally excreted, which would not be recognised as glycosuria by the ordinary tests, seems to be clearly shown in a series of experiments on E.S.W.

On 8. xi. 29, with a restricted diet, the fermentable sugar excretion was 159 mg. The following day the subject fasted and the fermentable sugar dropped to 62 mg. The next day 900 g. of boiled pig-liver constituted the sole diet and the fermentable fraction was 76 mg. The following day an ordinary restricted diet was taken which normally would have caused the excretion of not over 150 mg. of fermentable sugar. Actually 445 mg. were found, about three times the expected amount. The reverse effect seems to be shown in another series on the same subject. On 19. xi. 29, E.S.W. excreted 131 mg. of fermentable sugar on a restricted diet. The next day the restricted diet *plus* 100 g. of sucrose at breakfast, 50 g. at lunch and 100 g. at dinner was taken, a total of 250 g. of sucrose. The day following, a restricted diet *plus* 525 g. of sucrose was taken. The next day, a restricted diet *plus* 150 g. of glucose at breakfast, 150 g. at 10 a.m., 150 g. at lunch and 150 g. at 4.30 p.m. was taken, a total of 600 g. of glucose. The fermentable sugar excretion was 76 mg. and the following day on a restricted diet it was 65 mg. Here by high carbohydrate feeding and stimulation of the pancreas and possibly other carbohydrate-metabolising apparatus on successive days the excretion of fermentable sugar was reduced below the normal on a restricted diet.

Such experiments suggest that the very small quantity of fermentable sugar normally excreted is controlled by the factors operating in general carbohydrate metabolism and that, consequently, the sugar is glucose. Data presented in the third paper substantiate this conclusion. It seems, then, necessary to return to Benedict's view of a continuous "glycuresis" or passage of blood-sugar into the urine. We wish to emphasise, however, that the quantity of sugar passing is so small that it is not detectable by the ordinary qualitative tests and that it is of no clinical importance.

Protein diet preceded by starvation and purgation.

The object of this experiment was to correlate the rate and amount of excretion of fermentable and non-fermentable sugar with that of nitrogen elimination after a protein diet. Greenwald, Gross and McGuire [1927] studied the effect of the ingestion of a protein-fat diet upon the hourly excretion of sugar and nitrogen. They fed mainly eggs as the source of protein and observed no increase in the total sugar output. It should be noted that their total protein intake was relatively small, and also that the present writers have found the non-fermentable sugars from high egg diets quite small. In our experiments preliminary starvation and purgation were employed to remove the residues of previous meals from the intestinal tract. Subject A.C.L. fasted on 8. ii. 29, and took a dose of Epsom salts at 2 p.m. The next day urine samples were collected in 2-hour periods from 5 a.m. to 9 p.m. and 450 g. of lean beef were eaten at 9 a.m. The excretion of non-fermentable and fermentable sugars and of nitrogen for the 2-hour periods was determined. Fig. 1 shows the relations. Non-fermentable sugar was very definitely increased during the 2-hour period following the meat ingestion, while nitrogen remained at about the same level. Both non-fermentable sugar and nitrogen output reached a maximum during the second 2 hours, and fell off sharply 6 hours after the meat and gradually returned towards the basal values. The parallelism of the curves suggests that the non-fermentable sugars arise directly from substances contained in the meat or else are a result of the glandular or other activity concerned with its digestion and absorption. The promptness with which

elimination of non-fermentable sugar begins after meat feeding suggests that some of it may arise endogenously from the organs of digestion. This point has been tested in experiments on the dog from which we conclude that glandular activity does not contribute. These experiments will be given later in the discussion.

A sharp increase in fermentable sugar occurred during the third period after the meat was taken. It is probable also that the glucose of the blood reached a maximum about this time.

Fig. 1.

Starvation. So far as we are aware there are very few data in the literature on the effect of starvation upon the excretion of non-fermentable and fermentable sugar by human subjects. Harding and Selby [1931] have recently reported the absence of fermentable sugar from normal fasting urines within the limits of the analytical methods employed. We have pointed out in our first paper that the method used by these workers is incapable of estimating with sufficient accuracy the fermentable sugar of starvation urines. We have never failed to find fermentable sugar in the starvation urine of both human beings and the dog, although often in concentrations which the method of Harding and Selby would have failed to detect. We have made observations upon several individuals employing periods of starvation of 1 and 2 days. The results for three subjects are given in Table II. The decrease in non-fermentable sugar was rather less than we had expected and on the second day of starvation this fraction generally showed a slight rise. Fermentable sugar was continually

Table II. *Showing the effect of short periods of starvation upon the excretion of fermentable and non-fermentable sugars of urine.*

Date	Subject	Total N.F. (as glucose) mg.	Total F. (as glucose) mg.	Diet
10. vi. 29	E.S.W.	349	326	Normal
11. vi. 29	"	265	308	Starvation
12. vi. 29	"	274	68	Starvation. True blood-sugar 74 at 5 p.m.
16. vi. 29	H.L.W.	368	142	Normal
17. vi. 29	"	225	38	Starvation
18. vi. 29	"	279	160	Starvation
5. xii. 29	R.H.W.	245	—	High protein
6. xii. 29	"	165	33	Starvation

present. The non-fermentable sugar of starvation must have come from either endogenous sources or food residues remaining in the intestinal tract or both. If food residues were the major factor, a sharp falling off in the output might be expected on the second day of starvation, especially as experiments on the rate of excretion after the ingestion of food show that non-fermentable sugar is eliminated promptly.

Experiments on the dog. In order to establish the effect of prolonged periods of starvation upon the excretion of non-fermentable and fermentable sugar a series of experiments was carried out upon the dog. A female weighing 11.3 kg. and thoroughly house-broken was selected. The animal was kept in a metabolism cage and the urine collected by catheter in 12 or 24-hour periods and preserved in the usual way. No trouble with bladder infection was experienced either on an all-meat diet or during starvation.

The dog was taken from the animal quarters and placed in the cage for several days preliminary to beginning the experiment. Determinations of the daily excretion of non-fermentable and fermentable sugars were then made for a period of 10 days on a diet of 1 lb. of lean horse meat daily in order to establish the normal output. This was followed by 11 days' starvation and then 9 days with a diet of meat as before starvation. Fig. 2 shows the daily excretion of non-fermentable and fermentable sugar throughout the 31 days of the experiment. Nitrogen excretion is also shown for the starvation period and a few days before and after. Several points of interest are brought out by this experiment. In the first place the great variation in non-fermentable sugar on a constant diet in the control period was unexpected, the range being from about 130 mg. to 230 mg. daily, much more than we have ever observed in human subjects on a carefully controlled diet. A chance observation seemed to throw some light upon this anomaly. It was noted that the animal showed much lower excretion of non-fermentable sugar when first taken from the animal quarters and placed in the cage than she did after some days in the cage on practically the same diet. It was also found that there was a rather gradual increase in the excretion upon confinement in the cage for several days with a kind of alternating high and low output but a general trend upward until an

average excretion of about 160 mg. daily was attained. These findings seem to be correlated with the general condition of constipation of the dog. When first taken from the animal quarters where life had been active and bowel movements more frequent and regular, the non-fermentable sugar excretion was low (about 110 mg.). In the cage the dog gradually became constipated so that after a time defaecation occurred only every four or five days, whereupon the non-fermentable sugar excretion increased. More evidence in this direction is to be found in Fig. 2 where it is seen that after the period of starvation was

Fig. 2.

terminated, a week was required for the daily excretion of non-fermentable sugar to reach the general level of the pre-starvation period. Presumably this was about the time required to restore the normal movements of the intestinal tract.

It seems justifiable to conclude then that not only do foods contribute directly to the non-fermentable fraction, but that the processes of putrefaction and bacterial action taking place in the intestinal tract determine, in no small degree, the amount and possibly the kind of these substances excreted. These observations seem to represent the first concrete evidence presented in support of the view of Greenwald, Gross and McGuire [1927] that bacterial action in the intestine may contribute to the sugars of urine.

The output of non-fermentable sugar during the period of starvation dropped on the first day to somewhat less than half the previous level and changed very little, on the average, for 11 days. There seems to be a division into two periods, the first of 6 days and the second of 5 during each of which the average excretion was constant but somewhat higher in the first period than in the second. The constancy of the amount throughout starvation seems to suggest an endogenous origin for about a third or half of the non-fermentable sugar normally excreted. It may be that bacterial activity in the intestines produced these substances (or substance) though the constancy of the excretion would seem to preclude this action from playing more than a minor rôle.

During starvation it will be seen that fermentable sugar was continuously present in the urine though in greatly decreased amounts. Immediately after starvation the level rose to a slightly higher average, with less daily variation than before, and after about 6 days the curve resumed the shape of the pre-starvation period. The slightly higher average excretion after starvation may be a reflection of the impairment of carbohydrate metabolism by starvation.

There seems to be some alternation in the daily quantity of both non-fermentable and fermentable sugar excreted, that is, a high value followed by a low. This is not entirely regular and is disrupted for about a week after starvation when it seems to be resumed.

It is of interest to note that hydrolysis of $\text{HgSO}_4\text{-BaCO}_3$ filtrates of dog urine from the seventh day of starvation caused no appreciable change in the non-fermentable sugar and increased the fermentable from 12 to 22 mg. This is in general agreement with results obtained on human subjects and is possibly further evidence for the normal excretion in the urine of non-reducing polysaccharide material.

Glandular stimulation.

Attention has been directed to the promptness with which non-fermentable reducing sugar appears in the urine after taking food. This suggested that some of it may arise from glandular activity of the digestive organs and specifically from the catabolism of nucleic acids. In order to test this hypothesis experiments were made on the fasting dog in which the glands of digestion were stimulated by the sight and smell of food and by the ingestion of pilocarpine. Table III shows clearly that the excretion of both non-fermentable and fer-

Table III. *Effect of stimulation of the digestive glands of the dog upon the excretion of fermentable and non-fermentable sugars.*

Date	N.F. (as glucose) mg.	F. (as glucose) mg.	Remarks
1. iii. 30	57	11	Starvation
2. iii. 30	54	10	Starvation
3. iii. 30	56	8	Starvation. Stimulated from 9-15-11-15 a.m. by placing hot meat near animal
4. iii. 30	54	10	Starvation. Injected 6 mg. of pilocarpine nitrate intramuscularly at 9 a.m. Severe salivation

mentable sugar remained unchanged. It appears from these data then that the non-fermentable fraction of urine is unrelated to nuclear catabolism, a conclusion reached by Greenwald, Gross and Samet [1924-25] from a study of dogs with turpentine abscesses.

Kidney impairment.

A large number of experiments have been carried out on hospital patients afflicted with various pathological conditions. The excretion of non-fermentable sugar for a number of these cases is given in Table IV. It will be noted that

Table IV. *Non-fermentable reducing sugars in the urines of hospital patients.*

No.	Diuretic	24 hour urine cc.	Non- fermentable reducing substances. Total (mg.)	Remarks
1	Salyrgan 1 cc.	1040	161	Patient in hospital with chronic nephritis for 20 weeks
		540	81	
	Salyrgan 0.75 cc.	640	51	Successive days 4 days later
		640	107	
		540	38	
	Salyrgan 2 cc.	700	151	Successive days
		500	105	
		—	99	
2		900	81	Cardiac arteriosclerosis
		800	80	
3	Digitalis, 2 days Digitalis	810	154	Cardiac decompensation
		3900	292	
		1400	266	
4		1310	203	Adult rickets
5		1250	400	Anaemia. P.S.P.* normal
6	Digitalis	2800	420	Aortitis. P.S.P. normal
7		1250	356	
8		830	0	Haemorrhagic nephritis. Non-protein N normal
		600	42	
9	Digitalis	750	382	Mitral stenosis. No arteriosclerosis. Re- stricted fluids
		225	166	
10		850	0	Cardiac nephritis. Non-protein N 56
11	Digitalis	760	167	Cardiac arteriosclerosis
12		1550	163	
13		400	188	Post-hypercalcaemia
14				Diabetes, nephritis. Generalised oedema. P.S.P. 50 %
		750	382	
14				Determination, 15. iii. 29. Coronary infarct. Non-protein N 48, 13. iii. 29. Non-protein N 100, 18. iii. 29
		600	324	
Average (14, 9, 5, 6, 7 excluded)			138	
Normal—average of 58 cases			395	

* Phenolsulphonephthalein kidney test.

generally conditions leading to impaired kidney function cause a decreased output of non-fermentable sugar as would be expected. An interesting effect of the drug salyrgan¹ upon the output of non-fermentable sugar by nephritics is shown in that it causes a marked increase. It seems impossible to attribute this action to simple diuresis, because we have found, as did Harding and Van Nostrand [1930], that the output is independent of urine volume.

¹ Salyrgan is the trade name for the diuretic, sodium {o-[hydroxymercuric-methoxy-propyl-carbamyl] phenoxy} acetate, manufactured by H. A. Metz Laboratories, Inc., New York.

By way of summary it seems that the physiological factors affecting the excretion of fermentable sugar in normal urine are quite unrelated to those controlling the excretion of the non-fermentable fraction. The fermentable sugar appears to be related to the efficiency of the general carbohydrate metabolism, and probably represents a trivial loss of blood-sugar which escapes from the normal kidney. The non-fermentable fraction, on the other hand, varies with the diet, intestinal stasis and some unknown endogenous process.

Studies relating to the chemical nature of the non-fermentable sugars of urine are planned for the future.

SUMMARY.

A study of factors influencing the excretion of fermentable and non-fermentable sugars in urine by human beings and the dog has been made. The methods of analysis seem to offer more reliable data than have been obtained previously.

Studies on the relation of diet to the elimination of sugars have been made. Non-fermentable sugars apparently are derived in some measure from most common foodstuffs but in particular from dried fruits, apples, honey, *etc.* and carbohydrate-containing foods which have been subjected to high temperatures. This is in agreement with Folin and Berglund's [1922] conclusion that much of the reducing material of normal urine arises from altered carbohydrates of the food. Meat proteins, eggs, cheese, milk, purified carbohydrates, white bread, many fruits and common vegetables contribute somewhat, but not very much, to this fraction.

The excretion of non-fermentable sugar is not increased by high nucleoprotein diets or by the ingestion of yeast nucleic acid. Stimulation of the glands of digestion does not increase the excretion of non-fermentable sugar. Nucleoprotein metabolism seems to be entirely unrelated to this material.

In starvation experiments on both man and the dog, the excretion of non-fermentable sugar continued but at a somewhat lowered level. In the dog one-third to one-half may be endogenous and in man possibly more.

From experiments on the dog it is shown that intestinal stasis causes an increased output of non-fermentable sugar and it is concluded that bacterial action and putrefaction in the intestine may be considerable factors.

The non-fermentable sugar of urine is very little changed by hydrolysis.

Hospital patients with kidney impairment show a decreased output of non-fermentable sugar in the urine.

Fermentable sugar was found in all normal urines and in decreased amounts in the urines of persons during starvation. It was present in dog urine throughout a prolonged period of starvation.

The excretion of fermentable sugar in normal urine seems to be related to the condition of activity of the pancreas and of the general carbohydrate

metabolism. Conditions which lower the carbohydrate tolerance increase the fermentable sugar excreted and *vice versa*. This suggests that the small quantity of fermentable sugar of normal urine is glucose and that Benedict's idea of a continual passage of blood-sugar into the urine is correct. The amount present is undetectable by ordinary qualitative tests and of no clinical importance.

Evidence is presented that normally there is excreted in urine non-reducing polysaccharide material which upon hydrolysis yields fermentable sugar.

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