

ferments, and to which Euler has applied the term "Gärungsenzyme" (see footnote on page 1220).

10. The transformation products of glucose resulting from the action of this ferment are largely acids, none of which has so far been definitely identified. However, among the cleavage products of the sugar the presence of pentose and of formaldehyde could be ascertained.

I wish to express my thanks to Dr. T. Brailsford Robertson for his valuable counsel and continuous interest in this investigation, and also to Mr. C. B. Bennett for kind suggestions in carrying out some of the chemical work.

SOME ANALYSES OF URIN COMPOSITS.¹

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Several years ago, in the course of some metabolism experiments carried out in this laboratory, a large number of urin examinations were made daily with special reference to the nitrogen factors and the sulfur. At the same time composites were saved for a more detailed examination regarding certain constituents, and especially for the determination of the inorganic bases.

Much of the investigation was concerned with a study of the effects of certain diets on the composition of the urin, but the composites in question were collected during the preliminary or fore periods of the studies, and represent, therefore, urins corresponding to the usual or ordinary diets. The length of the fore periods from which urin could be saved from the same individuals amounted to a month or more, as will be shown in the detailed statements below.

The men whose urins were the objects of the investigation lived under the same general conditions and consumed a diet qualitatively the same in all cases. There were appreciable quantitative variations, however, in some of the factors of the food, which variations will be shown in the tables below, along with certain other data regarding the men, which may be considered as preliminary to the presentation of the special details of the urin analyses. The fecal excretion for two important factors is given also.

While the inorganic constituents of the food have not been directly determined, it will be recognized that the relative distribution of these bodies might be approximately found by calculation from the relative nitrogen distribution.

¹ Paper presented at the annual meeting of the American Chemical Society, Washington, December 27, 1911.

Subject No.	1	2	3	4	5	6
Time in days.	34	37	31	33	36	30
Subject's age.	24	21	20	26	22	28
Average wt. kg.	82.7	69.4	58.2	60.6	69.9	69.5
Nitrogen in food, grams.	12.64	11.27	11.31	12.65	14.03	12.84
N from bread, crackers, etc.	4.08	2.99	3.00	4.41	4.63	3.92
N from meats.	3.93	3.83	4.16	3.50	4.35	4.27
N from milk.	1.38	1.08	1.00	1.32	1.32	1.35
N from fruits and vegetables.	3.25	3.37	3.15	3.42	3.73	3.30
Nitrogen in feces.	1.53	1.49	1.72	1.40	1.81	1.77
Fat in food.	142.48	142.56	99.27	158.46	155.61	130.30
Fat in feces.	4.23	4.00	3.34	4.45	4.16	4.47

Analysis of the Urin.

The urin passed each day was preserved by means of toluene and always diluted to the constant volume of 2000 cc. A composit was made by taking 150 cc. in each case and saving the mixture to the end of the period of observation. All the determinations of the bases, and for total sulfates, phosphates and chlorides were made on this composit, while the nitrogen factors, the neutral sulphur and the ethereal sulfates were determined in the fresh urin from day to day and the means taken. Chloride, phosphate and sulfate determinations were also made on the fresh urin. The means of these determinations agreed very closely with those from the composites.

In the determination of calcium and magnesium, 400 cc. in each case was heated with hydrochloric acid and filtered, the filtrate made alkalin with ammonia and then slightly acid with acetic acid. This acid solution was precipitated with ammonium oxalate. The precipitate was filtered, washed, converted into oxide and weighed. In parallel determinations the well washed precipitate was dissolved in sulfuric acid and titrated with 0.1 N potassium permanganate, the results of the two sets of determinations agreeing very closely.

The filtrates from the oxalate precipitations were evaporated to dryness, heated to expel ammonium salts and the residues taken up with dilute hydrochloric acid. These solutions, after filtration, were precipitated in the usual manner with sodium phosphate, after addition of ammonia water and ammonium chloride. The precipitates were collected on Gooch funnels, washed, dried and converted into pyrophosphate in the usual manner.

For the alkali determination 100 cc. in each case was heated with sulfuric acid and barium permanganate to destroy organic matter, according to the method of Pribram and Gregor. The excess of permanganate was removed by oxalic acid and the sulfate by barium chloride. After treatment with ammonia and ammonium carbonate the solutions were filtered and evaporated, leaving a residue of chlorides. These chloride

Subject No.	1	2	3	4	5	6
Volume, for day	1171 cc.	1086 cc.	900 cc.	1092 cc.	1067 cc.	1107 cc.
Calcium	0.179 gm.	0.200 gm.	0.256 gm.	0.153 gm.	0.255 gm.	0.215 gm.
Magnesium	0.123	0.169	0.119	0.124	0.129	0.178
Sodium	5.606	4.312	3.776	5.110	4.947	4.168
Potassium	2.448	2.734	2.515	3.392	3.199	3.083
Ammonium, N	0.462	0.439	0.336	0.404	0.403	0.448
Inorganic sulfate, SO ₄	1.681	1.435	1.564	1.960	1.942	1.750
Ethereal sulfate, SO ₄	0.180	0.195	0.126	0.192	0.183	0.141
Neutral sulfur, S	0.149	0.151	0.127	0.122	0.148	0.131
Chlorine	9.017	7.952	7.242	9.869	9.230	8.378
Phosphoric acid, P ₂ O ₅	1.848	1.775	1.669	1.668	2.039	1.751
Total nitrogen	9.994	9.259	9.308	10.796	11.182	9.851
Urea nitrogen	7.982	7.408	7.753	9.105	9.247	8.026
Creatinine nitrogen	0.664	0.636	0.603	0.563	0.684	0.654
Uric acid nitrogen	0.199	0.147	0.166	0.169	0.184	0.181
Purine nitrogen	0.087	0.089	0.056	0.036	0.076	0.055
Rest nitrogen	0.600	0.540	0.394	0.519	0.588	0.487
Acidity	1.41	1.10	0.95	1.37	1.58	1.09
Indican	17	7	5	40	20	23

residues, as finally purified, were separated in the usual manner by platinum chloride.

The chlorides in the urin were found by titration according to the Volhard method. The phosphates were found by the uranium titration directly, controlled, in some cases, by the Pemberton titration. For the total sulfate determination 50 cc. was treated with hydrochloric acid, boiled, filtered, treated with barium chloride, boiled again and filtered. The precipitates were collected and weighed on Gooch funnels.

In the determination of the nitrogen factors the usual standard methods were employed. The urea was measured by the method of Benedict and Gephart, and the total sulfur by the method of Benedict and Gage.

In the tables of analytical results given above the numerical values express the means for the 24-hour excretion, usually in grams. The acidity, however, is given in terms of the oxalic acid equivalent, and the indican in terms of the arbitrary Fehling solution color standard. The volumes are given in cubic centimeters.

These figures, representing as they do mean results for rather long periods of time, and on known diets, have certain special values for the calculation of a number of interesting data, some of which will be given below. Among these the nitrogen distribution will be shown first, and this will be followed by other relations involving the nitrogen excretion of the men in different compounds.

PERCENTAGE DISTRIBUTION OF NITROGEN.

N of Urea.....	79.87	80.01	83.29	84.34	82.70	81.47
Creatinine.....	6.64	6.87	6.48	5.21	6.11	6.64
Ammonia.....	4.62	4.74	3.61	3.74	3.60	4.55
Uric acid.....	1.99	1.59	1.78	1.57	1.65	1.84
Purines.....	0.87	0.96	0.61	0.33	0.68	0.56
Rest.....	6.01	5.83	4.23	4.81	5.26	4.94

These distribution figures show the interesting relation, that in amount the various substances stand always in the same order. The ammonia nitrogen is always smaller than the creatinine nitrogen, and the rest—or undetermined—nitrogen is always between the creatinine and ammonia nitrogen values. Such relations do not hold universally, but in many similar investigations in this laboratory the same order has been generally followed. However, with some individuals there has been a departure as far as the relation of creatinine and ammonia is concerned. In this series of experiments the regularity is undoubtedly connected with the uniformity in diet.

Another relation involving the creatinine may be shown by considering this factor in connection with the body weights of the several individuals. Subject I shows the highest weight, but a large part of this is in the form of fat. On the other hand, the lightest man, number 3, is deficient in fat,

and number 4 is the least active physically, and probably in general strength somewhat below the other men. The creatinine values for the six men are as follows:

Number.....	1	2	3	4	5	6
Grams daily.....	1.783	1.708	1.619	1.512	1.836	1.756

These weights are fairly uniform. Number 5, with an excretion of over 1.8 grams daily, is still within the ordinary limits of excretion. If, by the aid of the weights of the men given above we calculate the excretion per kilogram of body weight we obtain the following relations:

Number.....	1	2	3	4	5	6
Mg. per kilo.....	21.56	24.61	27.83	24.95	26.28	25.27

The men were all students during the time of these experiments, and in no case were they performing any severe physical labor. Numbers 1 and 5, who show the highest gross excretion, had paper routes and carried bundles of papers some hours each morning. The marked difference in the excretion, as expressed in milligrams per kilo, is undoubtedly due to the larger fat content in the weight of 1. This man, in bone and muscle structure, is much like number 6, and with the excess of fat off might be expected to show the same proportional excretion. If the fat in all these men could be reduced to the condition which obtained with number 3 it is likely that all would show for the creatinine-weight relation a value in excess of 26 mg. per kilo.

The dependence of the creatinine excretion on muscular exertion has never been clearly established, although certain investigations have seemed to suggest it. On the other hand, a simple relation to weight would appear from some of the investigations of Folin,¹ as well as from figures pointed out in one of the long papers of Van Hoogenhuyze and Verploegh.² From the data of Folin somewhat lower proportional figures would be obtained than those which we have quoted, but in the case of the urins from five students examined by Van Hoogenhuyze and Verploegh the creatinine values run from 27-31.5 mg. per kilo of body weight, daily. Nothing is said about the fat of these men, and the determinations are apparently from single observations. It is proper to state in this connection that other extended experiments in this laboratory seem to indicate an excretion of about 30 mg. per kilo as what should be expected from men whose fatty tissue is relatively small.

If any further data were needed to show the non-dependence of the creatinine excretion on the total nitrogen the following figures would suffice. They are from studies as yet unpublished. Through a period of 10 days a group of students on a rather low protein diet excreted the following weights of nitrogen and creatinine daily:

¹ Folin, *Am. J. Physiol.*, 13.

² Van Hoogenhuyze and Verploegh, *Z. physiol. Chem.*, 46, 415.

Subjects.....	A	B	C
Total nitrogen.....	7.72	7.73	7.97
Creatinine nitrogen.....	0.64	0.56	0.64
Creatinine nitrogen, per cent.....	8.30	7.24	8.03

The same men through 20 days on a much higher protein diet, but qualitatively the same and with conditions of exercise or work the same, excreted, respectively:

Total nitrogen.....	12.36	12.86	13.24
Creatinine nitrogen.....	0.65	0.59	0.68
Creatinine nitrogen, per cent.....	5.26	4.59	5.11

With the variations in the total nitrogen the creatinine nitrogen is but little changed, but in percentage values the differences become, in consequence, very great. It is interesting to note, however, that for all the men the relation of the high to the low percentage is essentially the same, *viz.* 1.58, 1.58 and 1.57, respectively.

The Rest Nitrogen.—There has been much written on the question of the interpretation of the rest or undetermined nitrogen. Data on this point have value only when they are based on rather long series of analyses, made by uniform methods of the greatest possible accuracy. In the present case it is necessary to bear in mind that the urea values we report were obtained by the original Benedict and Gephart method and are therefore slightly in excess of those which would be secured by the Benedict modifications. But the excess is so trifling that it has no bearing on the question of relative values.

It has been claimed that this rest nitrogen is largely a matter of diet, and recently Bouchez and Lambling¹ have published data to show that when the diet consists largely of milk the rest nitrogen may be reduced to a negligible quantity, or brought within the limits of the errors of experimentation. In one set of observations reported by them it appears that a change in the diet from much meat to little meat reduced the undetermined nitrogen from 1.17 grams per day to 0.3 gram, or in percentage amount from 6.26 to 2.82% of the total nitrogen excreted.

Our experiments were not so conducted as to show the effect of a diet free from meat, but they have this important bearing, however, that they present a comparison of men on nearly similar diets through long periods. It will be noted in the case of subject number 1 that he took a higher proportion of his nitrogen in the form of milk than was the case with the other men, while the nitrogen in the form of meat was not as large as with some of the others. Yet the rest nitrogen of this man is the highest in gross and percentage amount. On the other hand, number 3 took relatively much of his nitrogen in the form of meat and relatively less as milk, yet with him the rest nitrogen reaches the lowest figure in the group.

¹ Bouchez and Lambling, *Compt. rend. soc. biol.*, 71, 435, 486.

These results do not support the contention of Bouchez and Lambling, and it may be added that a few data drawn from the results of Folin¹ in his experiments with a starch and cream diet indicate that, although the gross rest nitrogen may be reduced on the non-meat diet, the percentage amount is increased just as the percentage of creatinine nitrogen is increased under like circumstances.

In the study of a large number of relations obtained in other experiments we are led to the view that the rest nitrogen is to a considerable degree an individual peculiarity, depending possibly on the weight of the subject, and partly on the gross protein of the diet, rather than on the kind of protein. We have found for some men, with a varied diet, always a relatively high rest nitrogen fraction, while with others with the same diet the fraction is always relatively low, when the periods are long enough to eliminate accidental variations. In some cases observed by us this has been so marked that the identity of the subject could be discovered by a consideration of the rest nitrogen alone. It is possible that a part of the rest nitrogen depends on some peculiar metabolism of endogenous protein, as in the case of creatinine, while another part may increase and decrease with the ingested protein. Hippuric acid nitrogen is always a fraction of this undetermined residue and it is evident that any diet which would increase this form of excretion would naturally increase the rest nitrogen. There are doubtless similar cases.

Indican.—This was always determined in one one-hundredth of the urin for 24 hours by addition of an equal volume of Obermeyer's reagent and shaking out with 5 cc. of chloroform. The values here reported as means of long series of determinations have this interest that they show about the same variations which may be expected from any group of normal individuals on ordinary diet. In spite of all that has been written on the subject many physicians persist in considering such amounts of indican as we are here dealing with as pathological. In our experience in the laboratory with large groups of students the color reaction may run from practically zero to 100 or over on the arbitrary Fehling solution standard. While it is true that a starch and cream diet, kept up long enough, will bring the indican down to practically zero, it is also true that with some individuals on a high protein diet the indican may be again zero, or nearly so. From the observations recorded above it is evident that there is no relation between indican and a meat diet, or high protein in general, or with the amount of ethereal sulfates in the urin. In most cases the indican forming group must make up but a small fraction of the sulfate excreted in this way. This point is mentioned here because the relation is still frequently taken for granted in some of our recent literature.²

¹ Folin, *Loc. cit.*

² For some of the literature on these points see, for example, the recent hand-book by C. Neuberg, *Der Harn*, etc., p. 903.

Distribution of Sulfur.—The absence of any close relation between indican and ethereal sulfur is shown in the following table of the percentage distribution of sulfur:

Number	1	2	3	4	5	6
Inorganic sulfur	72.9	69.0	75.5	77.9	75.6	76.6
Ethereal sulfur	7.8	9.3	6.1	7.6	7.1	6.2
Neutral sulfur	19.3	21.7	18.4	14.5	17.3	17.2

It will be noticed that the highest ethereal sulfur value, in percentage amount, is found in a composit in which one of the lowest indican values was recorded. The same fact is brought out just as clearly by the consideration of the gross weights instead of the percentage weights. That the sulfur values, in gross, are regular and normal for the nitrogen content is shown by the following nitrogen-sulfur ratios:

Number	1	2	3	4	5	6
N/S ratio	13.1	13.3	13.4	12.9	13.0	12.9

The distributions given are pretty uniform and the variations naturally follow from the variations in the ingested protein. It will be noticed from the first general table given that the gross weights of neutral sulfur excreted are nearly the same for subjects 1, 2 and 5, while the gross ethereal sulfur is nearly the same for 1, 2, 4 and 5. We have observed in numerous other composites, as well as here, that a low total sulfur excretion is not necessarily accompanied by a low excretion of neutral or ethereal sulfur. The diminution commonly falls on the inorganic sulfur, as it does on the urea on a low protein diet, as has been suggested by Folin in the paper referred to above. These relations can not be easily recognized except in a long series of determinations, or in composites covering many days, because the lag in excretion after protein ingestion is not perfectly uniform for the different sulfur fractions.

Neutral sulfur and creatinine vary in about the same manner, and we find a nearly constant ratio between them in a large number of determinations made. For the six composites in question the following ratios obtain:

Number	1	2	3	4	5	6
Creat. N						
Neut. S	4.5	4.2	4.7	4.6	4.6	5.0

In the case of the three composites from subjects A, B and C mentioned above, we find the relations for high and low protein periods, as follows, and first for the ten days on low protein:

	A.	B.	C.
Total S.	0.610	0.607	0.635
Neut. S.	0.144	0.114	0.121
Neut. S per cent.	23.6	18.8	19.0

For the high nitrogen period of twenty days we have:

Total S.....	0.943	0.982	1.034
Neut. S.....	0.157	0.132	0.138
Neut. S per cent.....	16.7	13.4	13.3

The ratio of the percentage increase is, like the creatinine case, nearly constant, and for the three cases is:

1.41 1.40 1.43

Now, if we consider the relation of the creatinine nitrogen to the neutral sulfur we find:

		A.	B.	C.
Creat. N	} high protein.....	4.1	4.5	4.9
		Neut. S	} low protein.....	4.4

The ratios are not exactly the same for the two periods because, while the creatinine is essentially the same for each man under the two conditions, the neutral sulfur increases slightly with increased protein ingestion. If the creatinine and neutral sulfur came from exactly the same kind of metabolism they should vary together, and the fact that we find this slightly augmented neutral sulfur with changed metabolism suggests that probably two kinds of metabolism are concerned in the liberation of part of the sulfur in this so-called neutral form. It is possible that the larger part of the neutral sulfur, like the creatinine, is of endogenous origin, while a smaller fraction may come from the breaking down of the food protein directly, and would, therefore, increase with the amount of the protein in the diet. That the diet is of great moment here is certainly not the case. It is said that in starvation this neutral sulfur is increased both relatively and in absolute amount.¹ If this is true it would point to the breaking down of muscle as the probable source of the main portion, and would relate it to the creatinine. This assumption offers a plausible explanation of the interesting relation between the two pointed out above.

Relation of Bases and Acids. Acidity.—Acidity determinations were made for purposes of comparison from day to day, rather than with the aim of finding anything like absolute values. In fact, such determinations seem impossible with our present means of study, and all attempts to find a measure of the acidity are at best but approximations, and are further complicated by the interpretation of the behavior of various indicators. In recent years accurate methods have been gradually developed for the determination of the ionic, or what may be called the true hydrogen acidity, based on measurements of the observed electric potential developed in concentration cells of which the urin is one element. But Hoeber² has shown, and the same thing follows from other recent discussions, that there is no close relation between this ionic acidity and the titration acidity, however important the determination is for other purposes.

¹ See C. Neuberg, *Der Harn*, etc., p. 139 for literature.

² *Beitr. chem. Physiol. Pathol.*, 3, 525.

We found the acidity of the urins in question, from day to day, by means of the simple titration with 0.1 normal alkali and phenolphthalein. While a higher value is always obtained by the addition of sodium oxalate, as is frequently recommended, it has appeared to us and to several analysts who have helped in the titrations that the end reaction is not as certain and sharp as in the simpler scheme. In this connection it is interesting to note that in the rather lengthy review of the recent literature in the Neuberg handbook this Naegeli method is referred to as still the most certain for securing comparable results.

Our values, which are expressed in terms of grams of anhydrous oxalic acid daily, are as follows for the composites, the results being averages from the daily determinations on the fresh urins:

Number.....	1	2	3	4	5	6
Acidity.....	1.41	1.10	0.95	1.37	1.58	1.09

Certain relations may be noted here. Numbers 1 and 4 with nearly the same acidity correspond to diets containing essentially the same weights of protein, as shown in a previous table. Number 5 with the highest acidity corresponds to the highest protein. Number 3, with the lowest acidity, has not only a low total nitrogen, but low bread and milk consumption. The urin of this man was often cloudy when voided, and from the table is shown to contain the highest calcium content in the group, although the general food consumption was among the lowest. The previous history of this man pointed to a urin approaching alkalinity and frequent cloudiness for years, and it is evident that the low titration acidity is connected with the very common precipitation of calcium phosphate, since the sulfur and phosphorus ingestion, while lower than in the cases of other men in the group, are not low enough to explain it.

The general relation of the bases and acids in the composites is best shown by a tabulation such as follows, where each basic and acid radical is given in terms of its hydrogen equivalent, in grams, daily. In this way we can express the values for the five common basic elements, calcium, magnesium, sodium, potassium and ammonium and the five most important acid combinations, chlorides, inorganic and ethereal sulfates, phosphates and urates. Carbonates were not determined. The amount of carbonic acid held in the urin is often an important factor in the balance.

No. 1.			
Calcium	0.00890	Chlorine	0.25440
Magnesium	0.01006	Inorg. sulfur SO_4''	0.03498
Sodium	0.24322	Ether. sulfur RSO_4'	0.00187
Potassium	0.06253	Phosphoric acid PO_4'''	0.07809
Ammonium	0.03290	Uric acid $C_5H_2N_4O_3''$	0.00709
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	0.35761		0.37643

		No. 2.	
Calcium	0.00995	Chlorine	0.22430
Magnesium	0.01390	Inorg. sulfur SO_4''	0.02988
Sodium	0.18710	Ether. sulfur RSO_4'	0.00202
Potassium	0.06983	Phosphoric acid PO_4'''	0.07500
Ammonium	0.03130	Uric acid $\text{C}_5\text{H}_2\text{N}_4\text{O}_3''$	0.00523
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	0.31208		0.33643
		No. 3.	
Calcium	0.01275	Chlorine	0.20430
Magnesium	0.00973	Inorg. sulfur SO_4''	0.03256
Sodium	0.16382	Ether. sulfur RSO_4'	0.00131
Potassium	0.06424	Phosphoric acid PO_4'''	0.07052
Ammonium	0.02393	Uric acid $\text{C}_5\text{H}_2\text{N}_4\text{O}_3''$	0.00591
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	0.27447		0.31460
		No. 4.	
Calcium	0.00761	Chlorine	0.27840
Magnesium	0.01016	Inorg. sulfur SO_4''	0.04080
Sodium	0.22169	Ether. sulfur RSO_4'	0.00200
Potassium	0.08664	Phosphoric acid PO_4'''	0.07048
Ammonium	0.02878	Uric acid $\text{C}_5\text{H}_2\text{N}_4\text{O}_3''$	0.00602
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	0.35488		0.39770
		No. 5.	
Calcium	0.01272	Chlorine	0.26037
Magnesium	0.01060	Inorg. sulfur SO_4''	0.04043
Sodium	0.21459	Ether. sulfur RSO_4'	0.00190
Potassium	0.08169	Phosphoric acid PO_4'''	0.08616
Ammonium	0.02871	Uric acid $\text{C}_5\text{H}_2\text{N}_4\text{O}_3''$	0.00655
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	0.34831		0.39541
		No. 6.	
Calcium	0.01072	Chlorine	0.23635
Magnesium	0.01457	Inorg. sulfur SO_4''	0.03643
Sodium	0.18082	Ether. sulfur RSO_4'	0.00147
Potassium	0.07875	Phosphoric acid PO_4'''	0.07400
Ammonium	0.03190	Uric acid $\text{C}_5\text{H}_2\text{N}_4\text{O}_3''$	0.00645
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	0.31676		0.35470

We see from this method of presenting the urin composition a relation which is generally overlooked, that is the very important part which ammonia plays in combining with the urin acids. It is usually recognized that in acidoses from various causes an increased ammonia excretion follows, but we have here in ordinary normal urin an ammonia excretion which in combining power exceeds in every case the calcium and magnesium excretions combined.

In all cases the acid radicals exceed the basic, as should be expected. In numbers 1 and 2 the excess is not very great, and if an attempt were made to calculate the possible combinations the urin would appear about neutral. Carbonic acid, and organic acids not determined, must play an important part here. In the other cases the excess of acid radicals over basic is more marked.

The mean value of the titration acidity is equivalent to 1.25 grams of oxalic acid daily, or on the hydrogen scale to 0.0277, while the mean excess of acidity from the tables is 0.0335. This, however, is a misleading value, as it is based on a comparison with a group containing phosphoric acid assumed to act with three valences of hydrogen. For a comparison of this kind the acidities due to carbonic and organic acids should be added, and one-third of the phosphoric acid value substituted. A better comparison is obtained by assuming that the acidity which may be titrated is due to the conversion of diacid into monacid phosphate, which would correspond with one-third of the phosphoric acid found, and on the hydrogen scale, in the mean, would amount to 0.0252, which is not far from the mean of the titration acidities actually found.

For the individual composites the relations appear as follows:

	1.	2.	3.	4.	5.	6.
Titration.....	0.0313	0.0246	0.0211	0.0304	0.0351	0.0242
One-third PO_4	0.0260	0.0250	0.0235	0.0235	0.0287	0.0246

The agreement is in some cases very close, and is in general good, when we consider the character of the titration reaction. It may be taken as confirming the assumption that the greater part of the phosphate, at all events, is in the dihydrogen rather than in the monohydrogen form. In numbers 4 and 5 the titration acidity is so much in excess of that from the phosphoric acid as to suggest that other acids must play some important part in the result, but even without the other acids phosphoric acid seems to be nearly sufficient to combine in the acid salt form. An arbitrary combination of the analytical results for number 4 will illustrate this, the combination being made so as to balance the various radicals, without regard to the most probable form of union, if indeed such a calculation were possible in a solution.

An almost exact balance is obtained by assuming, at the end, after all other acids are combined, that four-fifths of the phosphoric acid is held as dihydrogen phosphate and one-fifth as monohydrogen phosphate. Phenylpotassium sulfate is taken as the type of ethereal salt present, and uric acid is assumed to combine with two valences.

The table below gives the results so calculated in terms of grams in 24 hours.

Sodium chloride.....	12.969
Potassium chloride.....	4.230
Calcium sulfate.....	0.518
Magnesium sulfate.....	0.612
Ammonium sulfate.....	1.521
Ammonium urate.....	0.584
Potassium urate.....	0.029
Potassium phenyl sulfate.....	0.424
Potassium dihydrogen phosphate.....	2.556
Potassium monohydrogen phosphate.....	0.860

This arbitrary combination, which is based on the factors actually determined, gives an explanation of the observed acidity. If hippuric and other organic acids, which although weak, form well defined salts, were taken into consideration the result would be still more marked. In normal urin hippuric acid may make up 0.75 gram, or more, daily, and on a vegetable diet may be twice that. Three-fourths of a gram is a hydrogen equivalent of 0.0042, which is as important as the ethereal sulfate or the uric acid in some cases. Any attempt to balance the bases should therefore include this acid and probably others, as well as carbonic. As some of these acids contain nitrogen they have additional interest from the standpoint of the rest nitrogen.

Summary.

This paper presents a series of complete analyses of the urin of six men on a controlled diet, qualitatively the same and quantitatively nearly the same for all members of the group. The inorganic constituents were determined in composites of over thirty days, the other determinations being made daily and the means taken for the same days.

In the distribution of nitrogen the urea N varied between 79.87 and 84.34% of the whole for the six men, but the order of distribution was always the same, *viz.*, urea N, creatinine N, ammonia N, uric acid N, purine N. It is pointed out that in many other long series of experiments in the same laboratory the order is very generally the same, with occasionally, however, the ammonia N in excess of the creatinine N.

The creatinine varied between 21.56 and 27.83 mg. daily per kilo of body weight, the highest value being found in the case of a rather lean man, and the lowest in the case of a fat man.

It is shown in the consideration of another group of three men kept for 10 days on a low protein diet, and then for 10 days on a higher protein diet that while the percentage amount of the creatinine N varied in the usual manner, the *relation* of the percentage amounts for the three men was remarkably constant, being 1.58, 1.58 and 1.57.

From the results of these experiments it appears evident that a decrease in the fraction of the nitrogen intake from meat, with a corresponding increase in that from milk, does not result in a decrease in the rest nitrogen,

either in gross or percentage amount. In the value of the rest nitrogen individual peculiarity is an important factor.

There appears to be no relation between the amount of meat in the diet and the indican excretion. To some extent this excretion is an individual peculiarity.

A rather close relation seems to exist between creatinine and neutral sulfur. Both vary in percentage amount in essentially the same manner with the change from low to high protein in the diet. It is possible that a considerable part of the neutral sulfur, like the creatinine, may be of endogenous origin.

Tables are given showing the relation of the acid to the basic ions, expressed on the hydrogen scale, and a comparison is made between the apparent acidity and the titration acidity.

NORTHWESTERN UNIVERSITY MEDICAL SCHOOL.
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THE CHEMISTRY OF STEAM HEATED SOILS.¹

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The effect which heat has upon soils is a subject rather prominently before agricultural investigators at the present time. This subject has received increasing attention during the past decade, prominently from the point of view of sterilization and partial sterilization. There has been much valuable information obtained along the biological lines concerned. The biological factors cannot and should not be ignored, but it must be remembered that they are only of significance or interest in the light of the biochemical changes which they produce in the soil. Moreover, any chemical changes produced in the soil through other means, be they cultural or by the addition of manures and fertilizers, or by the influence of steam heating, in turn affect the biological activities. The chemistry, therefore, of the soil both before and after heating in sterilization work becomes of the greatest significance to the biological worker.

Since heat activates the changes going on normally in soils, it is obvious that a study of heated soils also throws light upon the biochemical changes taking place in soils under field conditions. The present paper is a contribution to the knowledge concerning soil organic matter and the changes which it undergoes. Notwithstanding the fact that some of the results obtained by some of the investigators on this subject² are contradictory.

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² In this connection see: Franke, *Ber. botan. Ges.*, 6, 89 (1888); Liebscher, *Deut. Landw. Presse*, 20, 976 (1893); Schmoeger, *Ber.*, 26, 386 (1893); Pfeiffer and Franke, *Landw. Vers.-Sta.*, 46, 117 (1896); Dehérain and Demoussy, *Ann. agron.*, 22, 305 (1896); Richter, *Landw. Vers.-Sta.*, 47, 269 (1896); Schulze, *Jahresber. Ver. Vertreter angew.*