

CHAPTER VI

CYSTINURIA

OUR knowledge of cystin and of cystinuria alike dates from the year 1810, in which year Wollaston¹ described a previously unknown variety of urinary calculus, of which two specimens had come into his hands. The first of these had been removed from the bladder of a young child. Seeing that both stones had been found in the urinary bladder Wollaston assigned to the new compound of which they were composed the name of cystic oxide.

It was Berzelius² who suggested the change of name from cystic oxide to cystin, and of this new name, which has since been universally adopted, Civiale wrote³ in 1838, that although it corrected an error of chemistry it perpetuated an error of physiology, for cystin is excreted by the kidneys and does not have its origin in the bladder.

The sediments of hexagonal crystals found in urine, upon which the diagnosis of cystinuria is so often based, were first observed by Stromeyer⁴ in 1824 and by Prout⁵ in 1825.

Prout made the earliest analysis of cystin, but, being unaware of the presence of sulphur in it, he reckoned the contained sulphur as oxygen. Baudrimont and Malaguti⁶ announced in 1837 that cystin contains sulphur, no less

¹ *Philosophical Transactions of the Royal Society*, 1810, c. 223.

² *Traité de Chimie*, 1833, vii. 424.

³ *Comptes rendus de l'Académie des Sciences*, Paris, 1838, vi. 897.

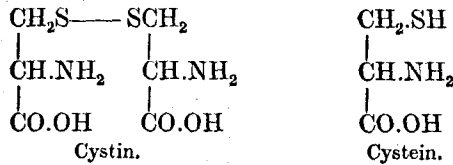
⁴ *Annals of Philosophy*, 1824, viii. 146.

⁵ *On Stomach and Urinary Diseases*, second edition, 1825, p. 166.

⁶ *Comptes rendus de l'Académie des Sciences*, Paris, 1837, v. 394.

than twenty-seven years after Wollaston's discovery ; and in the year following Thaulow ⁷ published a complete analysis which was in accord with that of Prout, save that half the oxygen of his reckoning was replaced by sulphur.⁸

To Goldmann and Baumann ⁹ we owe the recognition of the fact that cystin is an amino-derivative of thio-lactic acid, in which two molecules are linked together by their sulphur atoms, whereas cystein, which is easily obtained by reduction of cystin, contains an SH group in its single molecule. In Baumann's formula the sulphur atom and the amino-group were represented as being both attached to the α carbon atom, but Friedmann ¹⁰ has more recently shown that this is not the case, but that the amino-group occupies the α and the sulphur atom the β position. Cystin must therefore be regarded as di- α -amino- β -thio-lactic acid.



Since then the synthesis of this compound, which had repeatedly been attempted without success, has been accomplished by Erlenmeyer, jun.,¹¹ and its formula may be regarded as finally established.

⁷ *Annalen der Chemie* (Liebig's), 1838, xxvii. 197.

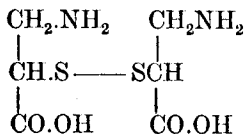
⁸ Baudrimont and Malaguti state (*Journal de Pharmacie*, 1838, xxiv. 633) that they presented a complete analysis to the Académie des Sciences in 1837, but I can find no published account of this. The *Comptes rendus* for that year contain only a brief note of their paper, in which the presence of sulphur in cystin is mentioned.

⁹ *Zeitschrift für physiologische Chemie*, 1888, xii. 254.

¹⁰ Hofmeister's *Beiträge zur chemischen Physiologie und Pathologie*, 1903, iii. 1.

¹¹ *Berichte der deutschen chemischen Gesellschaft*, 1903, xxxvi. 2720.

The revised formula renders possible the existence of an isomeric cystin, with the composition of di- β -amino- α -thio-lactic acid :



And this substance also has been prepared synthetically by Gabriel.¹²

Carl Neuberg and P. Meyer¹³ have put forward the view that some cystin calculi consist wholly or in part of this isomeric cystin, although they found that the cystin of sediments and that which is present in solution in the urines which they examined was of the ordinary kind. This would imply that some cystinurics excrete one and some the other cystin, for it cannot be supposed that urinary calculi are formed of a material which is not present in the urine of the patients who produce them. Some calculi which Neuberg examined, when dissolved in ammonia, yielded on evaporation of the ammonia acicular crystals which he believed to consist of iso-cystin. This view has not met with acceptance from other investigators. The great majority of the calculi are undoubtedly composed of ordinary cystin, which is deposited in hexagonal plates from ammoniacal solutions. That the materials obtained from proteins and from such calculi are identical in their chemical and physical properties, including their effect upon the polarized ray, has been proved by Rothera,¹⁴ Abderhalden,¹⁵ and J. Gaskell.¹⁶ Moreover, Emil Fischer

¹² *Berichte der deutschen chemischen Gesellschaft*, 1905, xxxviii. 637.

¹³ *Zeitschrift für physiologische Chemie*, 1905, xlv. 472.

¹⁴ *Journal of Physiology*, 1905, xxxii. 175.

¹⁵ *Zeitschrift für physiologische Chemie*, 1907, li. 391.

¹⁶ *Journal of Physiology*, 1907, xxxvi. 142.

and Zuzuki,¹⁷ who examined some of the same calculus material which Neuberg used, found that it gave a pronounced red colour with Millon's reagent, and they suggest that the acicular crystals described consisted of tyrosin. More recently, Abderhalden¹⁸ examined four cystin stones, all of which were shown to consist of ordinary protein cystin, and in three of these the presence of tyrosin was easily demonstrated by means of Millon's reagent. I have never obtained the acicular crystals from any cystin calculus which I have examined, although slender hexagonal prisms were not infrequently obtained. It is obvious that the occurrence of iso-cystin as a metabolic product is as yet very far from proven.

There are a few scattered records, of the last century, of the detection of cystin in animal tissues. Cloetta¹⁹ found it in the kidneys of an ox but failed to obtain it from other ox kidneys. Scherer²⁰ isolated cystin from the liver of a man who had died from typhoid fever, and Kulz²¹ obtained it on one occasion among the products of pancreatic digestion *in vitro*. Such observations lent support to the view, which had been held all along, that the sulphur of proteins was the source of the sulphur of cystin, which was regarded as an intermediate product of sulphur metabolism.

The year 1899 was marked by a great advance in our knowledge of the origin of cystin, for in that year K. A. H. Mörner²² showed that cystin could be obtained in abundance by the hydrolysis of hair. Soon afterwards G. Embden²³ obtained it, in like manner, by the hydrolysis of serum- and egg-albumins. Not only was there thus provided a ready

¹⁷ *Zeitschrift für physiologische Chemie*, 1905, xlv. 405.

¹⁸ *Ibid.*, 1919, civ. 129.

¹⁹ *Leibig's Annalen*, 1856, xcix. 299.

²⁰ *Jahresbericht für Chemie*, 1857, p. 561.

²¹ *Zeitschrift für Biologie*, 1884, xx. 1.

²² *Zeitschrift für physiologische Chemie*, 1899, xxviii. 595.

²³ *Ibid.*, 1901, xxxii. 94.

source of supply of this compound, which had previously only been obtainable from the rare cystin calculi and from the no less rare urinary sediments, but also it was shown that cystin has a place among the numerous amino-acids of which the complex molecules of proteins are built up. Like other primary protein fractions it is contained in widely different proportions in different members of the protein group.

Whether or not cystin is the sole protein fraction which contains sulphur, and is responsible for the entire sulphur contents of proteins, save the small quantity of sulphate which they hold in some sort of combination, cannot be regarded as finally decided. Mörner's work shows that all the sulphur of some proteins is in this form, but it is not yet proved that other sulphur compounds obtained from certain substances of the group are derivatives of cystin. Thus the α -thio-lactic acid, which has frequently been obtained, offers obvious difficulties in this respect, for in it the sulphur atom occupies the α position, whereas in cystin it is attached to the β carbon atom.

Some account of what is known concerning sulphur metabolism forms a necessary preliminary to the discussion of the disturbance which it undergoes in cystinuria. However, our knowledge concerning it is far less advanced than that of the metabolism of nitrogenous compounds; the processes at work are evidently of considerable complexity and it appears certain that the cystin of the proteins broken down in the body does not all follow the same catabolic path. Almost the whole of the sulphur of the proteins of food and of tissues ultimately appears in the urine, for little of it is excreted by way of the alimentary canal. A small portion is present in the saliva and gastric contents as sulphocyanide, but as the sulphocyanide grouping is not represented in protein molecules it must be supposed that it is formed by synthesis within the organism.

This fraction is eventually reabsorbed from the intestine and is excreted, at least in part, unchanged in the urine. A more considerable sulphur fraction goes to form the taurin of the bile, but the bulk of the sulphur takes a more direct route and is excreted as sulphates. Of the urinary sulphur, by far the greater part is in this fully oxidized form, and only some 14-20 per cent. is contained in a number of unoxidized or incompletely oxidized constituents which go to make up what is known as the neutral sulphur. Of the sulphates, the output of which may be swelled by sulphates absorbed as such from the alimentary canal, some portion is in combination as aromatic or ethereal sulphates; the greater part, some 90 per cent. of the total, is in the form of simple salts. The formation of the aromatic sulphates is usually ascribed to the working of a protective mechanism, by means of which aromatic substances of harmful nature, such as are apt to be absorbed from the alimentary canal, are rendered harmless and inert. The ratio of aromatic to simple sulphates is therefore regarded as affording a valuable index of the amount of protein decomposition brought about by the bacteria which inhabit the alimentary canal. Folin has thrown doubt upon this prevalent view, and seeing that the output of aromatic sulphate is little affected by a change from a diet rich in proteins to one poor in such constituents, he classes these compounds among the products of tissue metabolism as distinguished from that of the proteins of the food. However, the large amount of evidence available upon the other side—the increased output of aromatic sulphates in cases in which intestinal decomposition is abnormally active, the conversion of the whole of the sulphate into aromatic sulphate in some cases of carboluria, and the effects of intestinal disinfection in limiting the output—appear to me to call for the production of stronger evidence than has yet been brought forward before the

accepted view need be given up. As F. G. Hopkins²⁴ has suggested, the comparative uniformity of the excretion of these compounds upon widely different diets may well be due to the want of conspicuous variations in the bacterial activity in the intestine, a factor which is not known to be dependent upon the amount of protein contained in the food.

That variations in the amount of protein in the diet, although they influence very conspicuously the output of sulphates in the urine, have little effect upon that of neutral sulphur is a well-established fact, from which Folin concludes that the latter is made up of products of tissue metabolism as distinguished from that of protein foods. He compares the neutral sulphur with the endogenous uric acid and kreatinin among nitrogenous waste products, whereas the sulphates may be compared to urea. Abstinence from food causes relative decrease of sulphates, and so does the increased breakdown of tissue proteins which is brought about by certain toxic substances, such as chloral and chloroform.

Of the materials which go to make up the so-called neutral sulphur our knowledge is as yet very incomplete. Some is probably derived from taurin; the cystin-like compound found by Baumann and Goldmann may contribute a fraction; minute quantities of sulphocyanide have already been mentioned, and among the sulphur-containing substances must be reckoned certain little-known acids of high molecular weight, but which do not yield the reactions of proteins, to which the names of uroproteic and uroferrie acids have been assigned. In the urine of animals there have been met with, also, products of the decomposition of cystin in the alimentary canal, such as thio-sulphates, methyl mercaptan, and ethyl sulphide.

Some interesting experiments carried out by Baumann

²⁴ *Guy's Hospital Gazette*, 1907, xxi. 424.

and Preusse²⁵ and Jaffe,²⁶ at a time when it was not yet known that cystin is one of the primary fractions of proteins, were of much importance as showing that cystein is present in the animal organism as an intermediate product of protein metabolism. These investigators found that when monochlorbenzene or monobrombenzene was administered by the mouth to a dog there appeared in the urine of the animal a compound known as a mercapturic acid, which has been said to be excreted in combination with glycuronic acid. The process which leads to the formation of the mercapturic acid may be classed among the chemical protective processes, cystein being taken into combination with the halogen-benzene, just as glycozell combines with benzoic acid to form hippuric acid. Mester²⁷ afterwards found that when a halogen benzene was taken by a man hardly any excretion of mercapturic acid resulted. When it was shown that the rational formula of cystin required revision the force of this evidence was apparently destroyed, assuming that the formula of the mercapturic acid as given by Baumann were correct ; but Friedmann²⁸ followed up his work on cystin itself by a reinvestigation of the mercapturic acids, and showed that their formulæ also required correction in a similar sense.

Zeller, Thomas, and Straczewski,²⁹ found that when brombenzene is administered by the mouth to a dog on protein-free diet, no mercapturic acid is excreted, but when brombenzene is injected *sub cutem* into a dog taking a diet rich in protein, mercapturic acid is excreted, but in

²⁵ *Zeitschrift für physiologische Chemie*, 1879, iii. 159. See also Marriott and Wolf, *Biochemische Zeitschr.*, 1907, vii. 213.

²⁶ *Berichte der deutschen chemischen Gesellschaft*, 1879, xii. 1093.

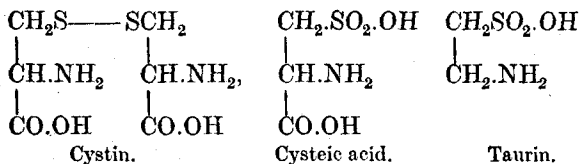
²⁷ *Zeitschrift für physiologische Chemie*, 1890, xiv. 109.

²⁸ Hofmeister's *Beiträge zur chemischen Physiologie und Pathologie*, 1903, iv. 486.

²⁹ *Archiv f. Anatomie u. Physiologie* (Physiol. Abtheilung), 1914, 585 ; *ibid.*, 1919, 249.

diminished quantity. Kapfhammer³⁰ has more recently shown that even a dog upon protein-free diet excretes abundance of mercapturic acid, if cystin is injected *sub cutem* and brombenzene is given by the mouth. From this it would appear that the synthesis of mercapturic acid does not take place in the intestine, but probably in the liver.

Of the cystin formed in the breaking down of proteins within the body, a portion, which has been estimated at some 30 per cent. of the total amount, is set apart for the formation of taurin, which is found in the bile in combination with cholalic acid, as taurocholic acid. Taurin stands in a simple chemical relationship to cystin as the following formulæ show :



and it has actually been obtained from it, *in vitro*, by Friedmann.³¹ It is obvious that the cystin destined to follow this metabolic path must be set apart at a very early stage of catabolism, before either the sulphur atom or the amino-group has been removed, and there is evidence that the removal of both of these is early effected.

It was shown by von Bergmann³² that when cystin is given by the mouth to a dog with a biliary fistula the taurocholic acid of its bile is not appreciably increased, whereas if cholalic acid be administered at the same time a conspicuous increase is obtained. On the other hand,

³⁰ *Zeitschrift f. physiol. Chemie*, 1921, cxvi. 302.

³¹ Hofmeister's *Beiträge zur chemischen Physiologie und Pathologie*, 1903, iii. 1.

³² Hofmeister's *Beiträge zur chemischen Physiologie und Pathologie*, 1903, iv. 192.

sodium cholate when given alone caused an obvious but transitory increase of taurocholic acid. This suggests that no excess of cholalic acid is available in the canine organism, but that an excess of cystin is available, which is, however, quickly exhausted. Only when an excess of both cystin and cholalic acid were swallowed was taurocholic acid continuously formed in abnormal quantities. In rabbits, on the other hand, as Wohlgemuth³³ found, the administration of cystin alone suffices to cause an increased formation of taurocholic acid. Rothera found that in man the administration of cholalic acid alone, or of this acid and of cystin together, does not bring about any decrease of the sulphates of the urine such as might be expected to result if an excess of taurin were formed, always supposing that the taurin fraction finally appears in the urine as neutral sulphur. When cystin was swallowed with cholalic acid it was burnt completely to sulphate, just as was the case when no cholalic acid was given with it. The more recent investigations of Foster, Hooper, and Whipple³⁴ throw further important light upon this question. These observers found that when taurin and cholalic acid were given by the mouth to dogs with biliary fistulæ there was an increased formation of bile and of bile acids, such as occurs when taurocholic acid itself was swallowed. When, on the other hand, taurin alone was injected into a vein or given by the mouth it had no effect upon the output of bile acids.

Cholalic acid given alone produced a conspicuous increase of bile acids in the bile, and they also came to the conclusion that under normal conditions of diet and health there is always an excess of taurin available, and that the amount of available cholalic acid is the factor which determines the output of taurocholic acid.

It was further shown that when cystin is given intra-

³³ *Zeitschrift für physiologische Chemie*, 1903, xl, 81.

³⁴ *Journal of Biological Chemistry*, 1919, xxxviii, 355 et seq.

venously to a fistula dog, and at the same time cholalic acid is given by the mouth, there is a conspicuous increase of taurocholic acid in the bile, and this accords with the observations of von Bergmann, who gave cystin with cholalic acid by the mouth.

Lastly, Foster, Hooper, and Whipple conclude that there are both exogenous and endogenous factors concerned in the metabolism of bile acids; that taurin can be produced from cystin under physiological conditions, and that the taurin so formed is available for combination with cholalic acid to form taurocholic acid.

Goldmann's³⁵ investigations leave no doubt that the excretion of mercapturic acid, which follows the administration of halogen-benzenes to dogs, is at the expense of the sulphates of the urine and not at that of the neutral sulphur. In other words, the cystin which is in combination in the mercapturic acid is some of that which in ordinary circumstances would have been fully burnt to sulphuric acid. However, Blum³⁶ states that when an experimental cystinuria is induced in dogs with biliary fistulæ, presumably by the halogen-benzene method, although this is not stated, the taurin disappears almost completely from their bile, and this would suggest that the mercapturic cystin is that which ordinarily goes to form taurin. If this be so the inference is not to be avoided that under normal conditions the sulphur of taurin is mainly excreted as sulphate. But there is some evidence which points in a contrary direction and which tends to show that the taurin sulphur is excreted as neutral sulphur. Thus Salkowski³⁷ found that in dogs, and in man also, taurin introduced into the stomach did not increase the output of sulphates, and came to the conclusion that it was largely excreted as

³⁵ *Zeitschrift für physiologische Chemie*, 1885, ix. 260.

³⁶ *La Semaine médicale*, 1906, xxvi. 554.

³⁷ *Virchow's Archiv*, 1877, lviii. 460.

taurin-carbamic acid. In rabbits, on the other hand, and probably in the vegetivora in general, the administration of taurin by the mouth leads to an increase of the urinary sulphates. C. Schmidt, von Adelung, and Watson,³⁸ who have reinvestigated this question, found, as Salkowski had done, that when taurin is taken by the mouth, or is injected in Ringer's solution, the increased output of sulphur is almost wholly as neutral sulphur; but Schmidt and Allen found no evidence that it was excreted as taurin-carbamic acid, and suggest that the taurin-carbamic acid found by Salkowski was formed in the urine after it was passed. Kunkel's³⁹ observation, that the establishment of a biliary fistula in a dog, and the withdrawal of bile from the alimentary canal, conspicuously diminished the neutral sulphur in the animal's urine, points in the same direction, but Kunkel himself declines to base any generalization upon an experiment upon a single dog. The question of the ultimate fate of the sulphur which goes to form taurin is clearly one which calls for further investigation.

It is found that when cystin itself is introduced into the stomach of animals, even in very large doses, no unchanged cystin is excreted in their urine. In addition to an increase of the sulphates, an increase of neutral sulphur has been observed in animals so treated, and this has been largely in the form of thio-sulphates, which are probably formed by the decomposition of cystin in the intestine, and thence absorbed. When doses of a few grammes of cystin are swallowed by a normal man the cystin is wholly burnt to sulphate and no increase of the neutral sulphur has been observed.⁴⁰ Polypeptides containing cystin, such as di-alanyl-cystin and di-leucyl-cystin, were found by Abder-

³⁸ *Journal of Biological Chemistry*, 1918, xxxiii. 501. *Ibid.*, 1920, xlii. 55.

³⁹ *Archiv für die gesammte Physiologie* (Pflüger's), 1877, xiv. 344.

⁴⁰ Rothera, *loc. cit.*, sub 14.

halden and Samuely⁴¹ to be dealt with by normal man in the same way as free cystin is. By injecting cystin into the systemic veins of animals L. Blum⁴² caused cystin to appear in their urine, but when the injection was made into the branches of the portal vein it was apparently destroyed in the liver and no cystinuria resulted. However, neither Blum, nor Rothera⁴³ who also tried the experiment, was able to bring about the destruction of cystin by adding it to crushed hepatic tissue. Obviously the whole of the cystin ingested in protein foods, as distinguished from that derived from the tissues, is not burnt straightway to sulphates and excreted as such. Some, as we have seen, is probably utilized in the formation of taurin, and some must escape direct destruction and be built up into the tissue proteins, the cystin content of which must necessarily be maintained.

We may now turn to the consideration of the derangement of sulphur metabolism in cystinuria, in virtue of which the subjects of that anomaly excrete some part of their cystin as such in their urine, often to their serious disadvantage.

The liability to the formation of calculi composed of cystin, and to other urinary disorders such as cystitis, gives to cystinuria a practical as well as a theoretical importance, as its study may possibly lead to the discovery of some means of averting its evil consequences. It may even happen that cystin is deposited in crystalline form in the organs and tissues of a cystinuric, as sodium biurate is in those of a victim of gout. Only one case of the kind is as yet on record. An infant, described by Abderhalden,⁴⁴ died at the age of twenty-one months with symptoms of

⁴¹ *Zeitschrift für physiologische Chemie*, 1905, xlv. 187.

⁴² Hofmeister's *Beiträge zur chemischen Physiologie und Pathologie*, 1904, v. 1.

⁴³ *loc. cit.*, sub 14.

⁴⁴ *Zeitschrift für physiologische Chemie*, 1903, xxxviii. 557.

inanity. The child was one of a family which included a number of cystinurics. At the necropsy the internal organs showed innumerable white specks, visible to the naked eye, which were found to be deposits of cystin. From the spleen cystin was readily extracted by ammonia, and was deposited from the solution in hexagonal plates, the nature of which was fully confirmed by chemical methods. In the few other available records of post-mortem examinations of cystinurics no mention is made of such deposits in the tissues.

The urine of cystinurics has been said to possess an odour not unlike that of sweetbriar when fresh, and of putrid cabbage when decomposition sets in. It is also said to acquire a greenish tint on standing, and Golding Bird ⁴⁵ described one specimen which assumed a bright apple-green tint. I cannot say that I have ever noticed a smell recalling that of sweetbriar from such urines or any distinctly green tint, but the odour of sulphuretted hydrogen which is given off in decomposition is sufficiently obvious. The deposits of crystalline cystin which are thrown down from such urines are not very abundant, but may often be seen with the naked eye to consist of glancing crystals. Under the microscope these appear as hexagonal plates, the edges of which usually show paired inequalities of length. Hexagons of different sizes are often superposed upon each other, and with the plates longer or shorter hexagonal prisms are occasionally intermixed. In some specimens the crystals show a radiating striation and jagged edges, but they still retain roughly hexagonal forms. Fresh crystals are deposited after the urine has been passed, especially on addition of acetic acid, and Delépine ⁴⁶ came to the conclusion that, in a case which he investigated, their deposition was favoured by the presence of an organism, probably

⁴⁵ *Urinary Deposits*, London, 1851, p. 181.

⁴⁶ *Proceedings of the Royal Society*, 1890, xlvii. 198.

one of the blastomycetes, which could be separated by filtration. No confirmation of this has yet come from other observers.

The solubility of cystin in ammonia affords a ready means of identification of the crystals, which are insoluble in acetic acid. In case of doubt the following test may be applied, which was described by Wollaston in his original paper on Cystic Oxide. Some crystals are dried upon a glass slide and covered with a cover-slip. A drop of strong hydrochloric acid is then allowed to flow over the deposit, and as each crystal is bathed in the acid there springs from it a stellate cluster of delicate prisms, which grow rapidly under the eye and which are composed of cystin hydrochlorate. If now a drop of water be allowed to dilute the acid the prisms melt away as rapidly as they were formed. The urinary crystals which are most likely to be mistaken for the hexagons of cystin are the almost colourless plates of uric acid which are sometimes deposited from feebly pigmented urines, and which may assume roughly hexagonal forms.

The cessation of deposits must not be taken as evidence that a patient has ceased to excrete cystin, for when no crystals are found in the untreated urine, and especially in such as is alkaline in reaction, the addition of acetic acid may cause their deposition. Even in acid urine cystin is by no means insoluble. However, there can be little doubt that the excretion of cystin may be actually suspended, at least for a time. Thus Loewy and Neuberg⁴⁷ failed to detect any, either as sediment or in solution, in the urine of a medical man who had undoubtedly been cystinuric at a former period, and Lewis and Simon⁴⁸ have described a case in which cystin could no longer be found although cadaverin was isolated from the urine. It is noteworthy

⁴⁷ *Biochemische Zeitschrift*, 1907, ii. 438.

⁴⁸ *American Journal of Medical Sciences*, 1902, cxxiii. 838.

that in both cases the urine yielded hydrogen sulphide very readily, and this was attributed by Loewy and Neuberg to an excessive output of neutral sulphur. This suggests that possibly the error of sulphur metabolism persists, although the form of its manifestation is changed, and that in such cases some other intermediate product than cystin itself may come to excretion.

Thus Thorndike⁴⁹ described a case in which the urine readily became alkaline and smelt strongly of hydrogen sulphide. A specimen acidified strongly with acetic acid had deposited no crystals in three days, but after it had stood for a month crystals of cystin were found. Similarly in a case observed by Mackenzie Wallis and myself, the urine of a man who had passed cystin calculi, and which was infected with *Bacillus coli*, threw down no crystals when acidified with acetic acid, even when acetone was added. A large specimen was kept with thymol for months and was repeatedly examined, but only at the end of six months was a fairly abundant sediment of cystin crystals found. In this case the output of neutral sulphur was such as is observed in cases of cystinuria, and it seemed probable that the cystin was in combination, and that the compound became broken up after long standing.

Hitherto cystin has not been detected in the fæces of cystinurics, whereas diamines are sometimes present. In the sweat it is stated to be present in some cases by Dewar and Gamgee,⁵⁰ who observed that silver coins carried in the pockets of such patients are apt to be blackened; but these authors do not describe how cystin, as such, was detected, and in a single case, in which I examined some collected sweat, I failed to find any evidence of the presence of cystin therein.

⁴⁹ *Boston Medical and Surgical Journal*, 1898, cxxxviii. 367.

⁵⁰ *Journal of Anatomy and Physiology*, 1871, v. 142.

Abderhalden⁵¹ found that the cystin content of the hair and nails of a cystinuric was normal.

Desmoulière⁵² demonstrated the presence of cystin in the blood of a cystinuric, and obtained the amino-acid in crystalline form from 25 c.c. of the blood. The method employed, which is fully described in his thesis, is too long to be quoted here, but the evidence appears to be quite conclusive—far more so than the observation described by Achilles Müller,⁵³ in the same year, of the presence of a single crystal of cystin in the blood of a patient who was the subject of this anomaly.

The error of metabolism of which cystinuria is a manifestation is clearly far more complex than that which underlies alcaptonuria, and far less uniform in character. Although the excretion of cystin in the urine is apparently a constant feature, and is that by which it has hitherto always been recognized, cystin is by no means the only protein fraction which is implicated and in some cases other amino-acids or their derivatives are to be found in the urine. The commonest of these are the diamines cadaverin and putrescin, which stand in intimate chemical relationship to the diamino-acids lysin and ornithin respectively, of which the former is a primary constituent of the protein molecule, whilst the latter enters into the composition of another such fraction, arginin. More rarely leucin and tyrosin are excreted unchanged, and it is probable that other amino-acids, less easily detected, will be found to be excreted in some cases. Thus it comes about that cases of cystinuria differ widely among themselves, not only in the number of protein fractions which are implicated but also in their behaviour as regards the individual fractions, so that what is true of one cystinuric may be quite untrue of another.

⁵¹ *Zeitschrift f. physiol. Chemie*, 1919, civ. 129.

⁵² *Thèse de Paris, La Cystinurie*, 1911, No. 328.

⁵³ *Wiener med. Wochenschrift*, 1911, lxi. 2364, 2488.

The cases hitherto investigated admit of classification upon the following lines : 1. In some no diamines and no primary protein fractions, other than cystin have been found in the urine. 2. In some the urine has contained cadaverin or putrescin or both, in addition to cystin, but the excretion of diamines is apt to occur in an intermittent manner. In one case lysin has been found unchanged. 3. In a very few cases leucin or tyrosin or both have been excreted with or without diamines. 4. In a single case, that of Loewy and Neuberg,⁵⁴ cystin was present in the urine, but no diamine, leucin, or tyrosin; yet when diamino-acids were given by the mouth the patient excreted the corresponding diamines, and when tyrosin and aspartic acid were given these were excreted unchanged. In other cases, in which similar feeding experiments have been tried, no such results have followed the swallowing of diamino- or monamino-acids, and the patient of Loewy and Neuberg was also exceptional in his method of dealing with cystin introduced by the mouth.

Seeing that leucin and tyrosin are usually found in the urine of sufferers from grave hepatic diseases, such as acute yellow atrophy, we might expect that in some cases other protein fractions, and amongst them cystin, might escape destruction and be excreted. Umber⁵⁵ actually observed such an excretion of cystin, in association with leucin and tyrosin, in a case of acute yellow atrophy of syphilitic origin in which recovery was brought about by salvarsan. The urine threw down the characteristic hexagonal crystals, and their nature was established by chemical tests. This case, although so far it stands alone, leaves little room for doubt that cystinuria occurs as a morbid event, and not only as a manifestation of an inborn anomaly.

⁵⁴ *Zeitschrift für physiologische Chemie*, 1904, xliii. 338.

⁵⁵ *Münchener med. Wochenschrift*, 1911, lviii. 2,499.

The investigation of the pathogeny of cystinuria has been greatly impeded by the lack of a simple and reliable method for the estimation of cystin in urine, and the methods employed by different observers have been of very different values. The naphthalene-sulphonic-chloride method employed by Abderhalden⁵⁶ is not easy of application. Concentration of the urine under reduced pressure, with the addition of acetic acid, gives results which are presumably reliable, and J. Gaskell's⁵⁷ method, in which the deposition of cystin is aided by the addition of acetone to the urine acidified with acetic acid, is easily carried out, and is satisfactory save for the detection of very small quantitative variations, in connexion with which the degree of dilution of the urine has a disturbing effect upon the results. The mere addition of acetic acid, without concentration, does not suffice, for, as with Heintz's old method of estimating uric acid, the results obtained are capricious and quite unreliable. The method of Mester,⁵⁸ which has been extensively employed, is based upon the assumption that an increase of the ratio of neutral sulphur to total sulphur, beyond the average ratio for normal urines, is due to cystin excreted as such. The objections to this indirect method are obvious, in view of the conspicuous effect of diet upon the excretion of sulphates, whereas the output of neutral sulphur is little affected by the amount of protein in the food. Alsberg and Folin⁵⁹ adopt a different plan of reckoning, by which any increase of neutral sulphur above the average normal output, which they estimate at a lower figure than most other observers, is reckoned as cystin sulphur. This plan avoids the error due to fluctuations of the sulphate excre-

⁵⁶ *loc. cit.*, sub 44.

⁵⁷ *Journal of Physiology*, 1907, xxxvi. 142.

⁵⁸ *loc. cit.*, sub 26.

⁵⁹ *American Journal of Physiology*, 1905, xiv. 54.

tion, but involves the assumptions that the cystin is excreted wholly at the expense of the sulphates, that the normal neutral sulphur remains intact, and that the increase of neutral sulphur is wholly in the form of cystin.

As regards the disturbance of sulphur metabolism in cystinuria it is certain that only a portion of the total cystin of the proteins broken down is excreted as such, and that the error is in no sense complete. Cystinurics always excrete sulphates, and neutral sulphur other than that contained in the cystin. In no single recorded instance has any approach been made to a maximal excretion, which would attain to some five grammes per diem. Nevertheless, a comparison of the average daily outputs determined by various observers suggests that the amount of cystin excreted tends to be approximately uniform. These results have been obtained by a variety of different methods of very different degrees of accuracy, and there has been no uniformity in the diets of the patients, but the outputs tend to be more uniform the more accurate the methods of estimation. On a diet containing moderate quantities of protein cystinurics usually excrete from 0.3 to 0.5 gramme in the twenty-four hours.

Figures from twenty-five cases are embodied in the accompanying table. In most instances averages have been calculated from the observations on several days, and in Caracciolo's case the amount of cystin has been worked out according to Mester's method from his published figures. As an example of the uncertainty of the acetic acid method, Leo⁶⁰ obtained by its means an average daily output of 0.14 gramme, but as calculated from his figures for sulphate and neutral sulphur excretion the average output according to Mester was 0.607, and according to Alsberg and Folin 0.72 gramme.

If further investigations by reliable methods should

⁶⁰ *Zeitschrift f. klin. Medizin.*, 1889, xvi. 325.

show that the uniformity thus foreshadowed is an actual fact, and that the quantity of cystin excreted in different cases of cystinuria does not overstep a definite limit, it will be necessary to suppose that some definite fraction of the proteins broken down escapes its usual fate.

There is a consensus of opinion among those who have worked at the subject that the unchanged cystin is excreted at the expense of the sulphates rather than at that of the neutral sulphur. The high ratios of neutral sulphur to total sulphur in the urine of cystinurics, which far exceed the normal ratios, bear witness to this, and it is upon the assumption that the neutral sulphur is not implicated that the indirect methods of estimating cystin suggested by Mester and by Alsberg and Folin are based. That the output of sulphates is diminished is a fact which is established beyond question, but the immunity of the normal neutral sulphur does not appear to me to be so well established. Loewy and Neuberg obtained in their case normal values for the neutral sulphur after removal of cystin by concentration of the urine acidified with acetic acid, but Thiele⁶¹ who employed the same method for the removal of cystin, obtained unduly high values of neutral sulphur after its removal. Desmoulière⁶² estimated the cystin in his patient's urine at 0.4916 grammes per litre, by a modification of Gaskell's method and at 0.4815 per litre by a method based upon the ratio of neutral to total sulphur. On the other hand, Williams and Wolf⁶³ found that after subtraction of the cystin sulphur the output of neutral sulphur in the urine of their patient was abnormally high, and conclude that in cystinuria the neutral sulphur fraction is increased either by the excretion of an excess of the substances which are normally present in that form, or of

⁶¹ *Journal of Physiology*, 1907, xxxvi. 68.

⁶² *Thèse de Paris*, 1911, No. 328.

⁶³ *Journal of Biological Chemistry*, 1909, vi. 337.

DAILY EXCRETION OF CYSTIN IN 25 CASES OF CYSTINURIA.

No.	Sex and Age.	Observers.	Dates.	Methods of Estimation of Cystin.	Average daily Output of Cystin.	Average Neutral Sulphur as SO ₃	Remarks.
1	M. 18	Niemann	1876	Precipitation by acetic acid	0.52 grm.	—	extremes 0.42-0.59.
2	M. adult	Loebisch	1876	" "	0.393 grm.	—	mean of 10 observations.
3	M. 25	Ebstein	1879	" "	0.241 "	—	
4	F. 23	"	—	" "	0.430 "	—	
5	M. 13	Stadthagen	1885	" "	0.186 "	0.258 grm.	mean of 4 obs.
6	F. 41	Leo	1889	" "	0.140 "	0.483 "	
7	F. 29	Picini e Conti	1891	" "	0.19-0.3 grm.	—	
8	F. 65	Marrott and Wolf	1906	" "	0.464 grm.	0.82 grm.	mean of 2 obs.
9	M. 27	A. Conti	1922	" "	0.39 "	—	
10	F. adult	"	1922	" "	0.21 "	—	
11	M. adult	Mester	1889	Mester's method	circ. 1 "	0.940 grm.	mean of 13 obs.
12	F. 50	Percival	1902	" "	0.415 "	—	mean of 8 obs.
13	F. adult	Moreigne	1899	" "	0.454 "	—	
14	M. 23	Caracciolo	1907	" (calculated)	0.469 "	0.615 "	
15	M. adult	Thiele	1904	concentration and acetic acid	0.561 "	—	
16	M. adult	Loewy and Neuberg	1904	" "	0.471 "	—	
17	M. adult	Abderhalden and Schittenhelm	1905	" "	0.4 "	—	
18	M. adult	Alsberg and Folin	1905	Alsberg and Folin's method	—	{ 0.87 " } { 0.47 " }	on protein-rich diet 1 grm., on protein-free diet 0.5, on protein-rich diet 1.31 grm., on protein-poor diet 0.7. mean of 16 obs.
19	M. 44	Wolf and Shaffer	1908	" "	0.776 "	—	" " 5 "
20	F. 22	Hele	1909	Gaskell's method	0.314 "	—	" " 8 "
21	M. 63	"	1909	" "	0.412 "	—	" " 5 "
22	M. 32	"	1909	" "	0.556 "	—	on protein-rich diet 0.703.
23	M. 39	Williams and Wolf	1909	" (modified)	0.435 "	—	on protein-free diet 0.078.
24	F. 27	Klemperer and Jacoby	1914	" "	0.476 "	—	
25	F. 42	Ploos van Amstel	1910	Gaskell's method	0.43 "	—	

substances which, like cystin itself, are not present in normal urine in any appreciable quantities. That the urine of a cystinuric may at times contain no demonstrable cystin, but may yet yield a large excess of neutral sulphur, has already been pointed out. In a few cases in which cystin excretion has ceased, or nearly, ceased, for a time, the excretion of neutral sulphur has remained much above the normal. This was so in Loewy and Neuberg's second case, and the ready formation of hydrogen sulphide in the case recorded by Lewis and Simon suggests that the same was true for it. Bödtker⁶⁴ also obtained ratios of neutral to total sulphur as high as in any investigated case of cystinuria, even when the urine of his patient no longer deposited cystin, even with acetic acid, and when the presence thereof could only be detected by benzylation after concentration of the urine. The case observed by Mackenzie Wallis and myself, in which no cystin could be detected by Gaskell's method, but in which cystin was deposited after six months, affords a still more convincing example. Unless we are prepared to admit that precipitation methods may wholly fail even when cystin is abundantly present as such in urine, the inference cannot be avoided that under certain conditions cystinurics excrete a sulphur compound which is not free cystin but a compound or unoxidized derivative thereof. If this be so, estimations of cystin cannot be safely based upon variations, either relative or absolute, of the neutral sulphur.

The next question which calls for discussion—namely, whether or not the excretion of cystin is influenced by the amount of protein in the diet—is one of much interest. That the output is in part endogenous and derived from the tissue proteins cannot be doubted, for Alsberg and Folin found cystin in the urine of their patient after he had been for thirteen days upon a diet which was practically

⁶⁴ *Zeitschrift für physiologische Chemie*, 1905, xlv. 393.

protein-free. They found, however, that, although the ratio of neutral to total sulphur was naturally far higher on a protein-free diet, the actual quantity of neutral sulphur excreted was considerably greater on a diet rich in protein. As the excretion of neutral sulphur remains almost constant in normal individuals, even on such extremes of dietary, they infer that the increase was in the form of cystin and that the output of that substance, which averaged 0.5 gramme on a nitrogen-free diet, rose to 1 gramme on one rich in protein. In support of this view they adduce the fact that whereas upon the latter diet there was an abundant deposit of crystals, these almost ceased to be deposited when protein was withheld. Wolf and Shaffer⁶⁵ have obtained results which are in complete accord with those of Alsberg and Folin. With a high protein diet, and an excretion of 14.63 grammes of nitrogen in the urine, the cystin as estimated by the increase of neutral sulphur amounted to 1.31 grammes on an average. With an output of 3.53 grammes of nitrogen the cystin fell to 0.47 gramme. They also observed a greatly increased deposition of crystalline cystin when a diet rich in protein was taken. Thiele,⁶⁶ whose observations were unavoidably restricted to isolated days, and are therefore open to the objection that an unequal rate of excretion of nitrogen and sulphur, such as indicated by the results of other observers, may come into play, concluded that the output of cystin by his patient was not affected by the nature of the food taken. The cystin extracted by concentration and addition of acetic acid hardly varied in amount on a day of complete abstinence from food with a nitrogen excretion of 5 grammes, on a day of diet poor in protein on which 9 grammes of nitrogen were excreted, and on one of protein-rich diet on which the

⁶⁵ *Journal of Biological Chemistry*, 1908, iv. 444.

⁶⁶ *loc. cit.*, sub 61.

nitrogen amounted to 17.29 grammes. Moreover, the residual neutral sulphur, excluding that of the separated cystin, showed very slight variations, amounting to 0.359 gramme SO_2 on the fasting day and to 0.428 on that of protein-rich diet. Abderhalden and Schittenhelm⁶⁷ give two estimations of cystin by the same method in a case described by them. With an excretion of 16.6 grammes of nitrogen the extracted cystin amounted to 0.31 gramme, with 12 grammes of nitrogen to 0.42 gramme. Of earlier observers Leo and Mester expressed the belief that the excretion of cystin in cystinuria is not influenced by diet. In a case under my care, which was investigated by Hele, it was not possible to give any extremes of diet—and the capricious appetite of the patient even prevented the maintenance of a constant scale of feeding, but the results obtained in observations extending over series of days pointed to a scanty augmentation of cystin, as extracted by Gaskell's method, and also of neutral sulphur, when the protein of the food was increased. In another case which Hele investigated the neutral sulphur of the urine rose slightly when protein food was increased; but the rise was small and not proportional to the large rise of oxidized sulphur.

However, the more recent observations, and especially those of Williams and Wolf,⁶⁸ lend very strong support to the conclusions of Alsberg and Folin, and leave little room for doubt that the output of cystin by cystinurics is influenced profoundly by the amount of protein in the diet. The average daily excretion of cystin by the patient of Williams and Wolf was 0.435 gramme, but during four days of protein-rich diet the daily average rose to 0.703 gramme, which corresponds to an increase of 0.268 gramme of cystin per diem. In this case the cystin was estimated by a

⁶⁷ *Zeitschrift für physiologische Chemie*, 1905, xlv. 468.

⁶⁸ *loc. cit.*, sub 63.

modified Gaskell's method, and the inorganic, ethereal, and neutral sulphur were also determined. During the first period the average output of nitrogen was 9.62 grammes, and during the protein-rich diet was 11.28.

Klemperer and Jacoby,⁶⁹ who also employed Gaskell's method, observed a great reduction of the urinary cystin on a protein-free diet. We have seen how valuable is the information afforded by the study of the Homogentisic acid : Nitrogen ratio in cases of alcaptonuria, and a similar study of the variations in the Cystin : Nitrogen ratio in cystinuria would be of much interest. However, the materials for such a study are as yet scanty.

In the table which follows the C : N ratio has been calculated from the records of a number of cases, but the value of the figures is greatly impaired by the diversity of

TABLE SHOWING VARIATIONS OF THE C : N RATIO IN CYSTINURIA.

<i>Names of Observers.</i>	<i>Total N in 24 Hours.</i>	<i>C : N Ratio.</i>	<i>Method of Estimating Cystin.</i>
	gram.		
Moreigne . . .	{ 4.13	7.4 : 100	Mester's method.
	{ 16.8	4.8 : 100	
Percival . . .	{ 3.98	5.9 : 100	" "
	{ 15.1	3.1 : 100	
Alsberg and Folin . . .	{ Mean 5.19	9.6 : 100	Absolute increase of neutral sulphur above normal average.
	{ Mean 14.84	6.7 : 100	
Wolf and Shaffer	{ Mean 3.53	13.3 : 100	" "
	{ 14.63	8.9 : 100	
Abderhalden and Schittenhelm .	{ 12.0	3.3 : 100	Concentration and acetic acid.
	{ 16.6	1.08 : 100	
Thiele . . .	{ 5.15	9.4 : 100	Concentration and acetic acid.
	{ 9.1	6.2 : 100	
	{ 17.29	2.9 : 100	
Hele I . . .	{ Mean 4.85	5.5 : 100	Gaskell's method.
	{ 8.83	4.3 : 100	
Hele II . . .	{ Mean 10.03	4.3 : 100	" "
	{ 16.47	2.3 : 100	
Williams and Wolf . . .	{ Mean 9.62	4.5 : 100	Gaskell's method (modified).
	{ 11.28	6.2 : 100	

⁶⁹ *Die Therapie der Gegenwart*, 1914, lv. 101.

the methods employed for the estimation of the urinary cystin, and the lack of information as to the nature of the diet in the several cases included.

One point which is brought out by these figures is that in different cases the C : N ratio is more or less upon the same plane, and this is specially noticeable in the cases of Hele and Williams and Wolf, in which the cystin was directly estimated by Gaskell's method.

It will be noticed, moreover, that in every case save that of Williams and Wolf the ratio fell conspicuously when the protein intake was increased. This is what might be expected if the output of cystin were independent of the amount of protein in the diet, always supposing that there is no difference in the quality of the proteins taken.

It would be unwise to draw any definite conclusions from the comparison of these figures, in view of the unequal value of the methods employed and the uncertainty as to the nature of the diets given in the several cases, and in the same case at different periods.

Some observers⁷⁰ have noted differences in the quantities of cystin in the day and night urines respectively, but their results have been contradictory, and Hele was unable to find any constant difference in the cases which he investigated. In one of Hele's cases the rate of excretion of neutral sulphur was very constant during the day and night, whereas that of nitrogen and oxidized sulphur underwent great variations.

Much interest attaches to the effects of the administration of cystin, as such, to cystinurics. As we have seen, a normal man burns cystin so given to sulphate, and Alsberg and Folin found that their patient behaved in this respect

⁷⁰ Beale, *Kidney Diseases, Urinary Deposits, and Calculous Disorders*, third edition, 1869, p. 385. Ebstein, *Deutsches Archiv*, 1881, xxx. 594. Piccini e Conti, *Lo Sperimentale*, 1891, xlv. 353. Bartels, *Virchow's Archiv*, 1863, xxvi. 419.

as does a normal man. Two doses of cystin, obtained by hydrolysis of hair, amounting to 1.2 and 6 grammes respectively, were given by the mouth at different times. The neutral sulphur output was unchanged and the increased excretion of sulphur was wholly as sulphates. In one of his cases Hele obtained a similar result from the administration of 5 grammes of cystin from hair. Thiele gave 4.6 grammes of the patient's own cystin thoroughly purified. The cystin excreted was estimated by concentration of the urine and addition of acetic acid, and showed no increase worthy of mention. The quantities for the day before the cystin day, for the cystin day itself, and for the day following were 0.577, 0.604, and 0.561 gramme respectively. However, on the cystin day there was a very conspicuous increase of neutral sulphur as well as of sulphate, and the total increase of sulphur of both kinds corresponded to 3.526 grammes of cystin, of which increase 35 per cent. was in the form of neutral sulphur. It will be remembered that in this case the output of neutral sulphur, above the normal, was always higher than was accounted for by the cystin extracted from the urine. On Alsberg and Folin's reckoning this patient would have been taken as having excreted some of the cystin given as such. Williams and Wolf, who administered 5 grammes of cystin prepared from hair to their patient, found an increase of the inorganic sulphur of 77 per cent. and of the neutral sulphur amounting to 16.3 per cent. This they interpret as signifying a moderate degree of intolerance, but it was obvious that the greater part of the cystin absorbed was excreted as inorganic sulphate, as is the case with a normal individual.

In Loewy and Neuberg's case,⁷¹ which stands wholly apart from all others hitherto investigated, the administration of cystin by the mouth produced far more remarkable results. When 5 grammes of cystin from hair were so taken

⁷¹ *Zeitschr. f. physiol. Chemie*, 1904, xliii. 338.

the excreted cystin, estimated, as in Thiele's case, by concentration with acetic acid, rose from 0.388 gramme per diem to 7.04 grammes in the collected urine of the three days following the administration, an increase which corresponds to the whole of the cystin swallowed. In other words, this cystinuric was quite unable to burn protein cystin given by the mouth, and simply added it to his ordinary daily output. Whether increase of protein food influenced the excretion in this case we are not told. Still more extraordinary was the result when cystin obtained from a calculus was given. The taking of 3.52 grammes of this material caused no increase of cystin in the urine, but there resulted a conspicuous increase of the neutral sulphur, mainly in the form of thio-sulphate. Why cystin obtained from hair and from a calculus respectively should have been dealt with in such wholly different ways by this patient remains a mystery, no solution of which can be suggested unless these authors are right in regarding them as different substances. The excretion of thio-sulphate suggests the possibility that some of the cystin taken by the mouth may undergo decomposition in the intestine, and that this may account for the increased output of neutral sulphur, other than cystin, which has sometimes followed its administration.

If it be established that the cystin of protein foods increases the urinary output of cystinurics, whereas cystin taken as such is fully burnt, it must be inferred that cystin is not absorbed as such from protein foods, but in some higher combination. This is in accord with Loewy and Neuberg's converse observation that their patient disposed of amino-acids given in proteins, whereas he excreted unchanged amino-acids given as such.

Wolf and Shaffer⁷² found that cystin and cystein subcutaneously injected into their patient caused a marked

⁷² loc. cit., sub 65.

increase of neutral sulphur in the urine, and a smaller increase of sulphate sulphur. If the increase of neutral sulphur was, as they believe, in the form of cystin, this would indicate that the cystin or cystein injected was in part excreted as cystin and in part destroyed. It was at least shown that the patient had the power of oxidizing part of the injected material.

The same observers had the exceptional opportunity of investigating a case of cystinuria in which a biliary fistula persisted after an operation for gall-stone. Unfortunately special difficulties were encountered in connexion with this case, and the results obtained were incomplete. However, they were able to show that the ratio of sulphur to nitrogen in the bile which came from the fistula lay within the normal limits, as indicated by comparison with the results obtained by Shaffer in another case of biliary fistula. The actual amounts of sulphur and nitrogen in the bile were greater than in that of the control subject. The administration of cystin by the mouth to the cystinuric patient appeared to increase the sulphur of the bile, and disturbed the S : N ratio. No such observations on a non-cystinuric patient with a biliary fistula are available.

It was a remarkable feature of the case in question that shortly after the operation cystin disappeared from the urine, and the ratio of neutral to total sulphur, which had been very high, fell at the same time to within the normal limits. It is difficult to suppose that this event can have been connected with the drainage of the bile, but the coincidence was, at least, a remarkable one. Even when the cystin had disappeared from the urine the output of undetermined nitrogen therein remained very high.

Owing to the great liability of cystinurics to the formation of urinary calculi, and the dangers and inconveniences which such calculi involve, efforts have naturally been directed to control of the anomaly by treatment.

Von Bergmann's observations upon the effects of administration of cholalic acid to dogs with biliary fistulae suggested to Simon and Campbell,⁷³ and independently to Alsberg,⁷⁴ that the excretion of cystin in the urine might possibly be due to deficient formation of cholalic acid, and that the cystin set apart for the formation of taurocholic acid might on this account fail to be utilized and be excreted unchanged. If this were so the administration of cholalic acid should have the effect of restricting or of abolishing the output of urinary cystin. If it be the case that much of the neutral sulphur of urine is derived from taurin, the fact that cystinurics apparently excrete normal quantities of neutral sulphur in addition to cystin would suggest that the formation of taurocholic acid is not interfered with.

There are obviously great difficulties in arriving at any definite conclusions as to the influence of cholalic acid upon taurocholic formation in the absence of a biliary fistula, and for this reason the observations of Simon and Campbell are not conclusive. They could obtain no evidence that in their cystinuric the taking of cholalic acid had any influence upon the output of cystin, but this was not estimated by any direct method, and the fluctuations of the excretion of neutral sulphur during the period over which the observations extended were very wide, so that it is not easy to gauge the effect of the cholalic acid upon the ratio of neutral sulphur to sulphate.

All that we know of the pathology of cystinuria renders it highly improbable that the formation of cholalic acid is primarily at fault, but these observations are of special interest as embodying a rational attempt to influence by treatment a condition which can give rise to grave morbid events.

Alsberg and Folin and some other observers have

⁷³ *Johns Hopkins Hospital Bulletin*, 1904, xv. 164.

⁷⁴ *Journal of Medical Research*, 1904, xiii. 105.

recommended reduction of protein intake as the only means at present available for reduction of the output of cystin in the urine, and in their hands the effects of such treatment have been encouraging.

The administration of an alkali in doses sufficient to keep the urine alkaline is a form of treatment which was suggested long ago, and is based upon the solubility of cystin in alkalis which prevents the formation of crystals in alkaline urine. Klemperer and Jacoby⁷⁵ have recorded some remarkable results of a trial of such treatment. In their case a conspicuous reduction of the cystin output, estimated by Gaskell's method, was brought about by the administration of a protein-free diet. They then gave 6 to 10 grammes of sodium bicarbonate daily, with the result that the output of cystin decreased from day to day, until on the fifth day no cystin could be found either in the sediment or in solution. The diet was then relaxed and the drug was stopped, and ten days later the cystin output had returned to its ordinary level of about 0.4 gramme per diem. No estimations of nitrogen or of sulphur excretion are recorded by these observers.

Smillie,⁷⁶ who repeated the experiments of Klemperer and Jacoby, was unable to confirm their observations. As estimated by the output of total, inorganic and neutral sulphur, the excretion of cystin was not diminished by the administration of as much as 40 grammes of sodium bicarbonate per diem. On the other hand no crystals of cystin were deposited from the alkaline urine. Smillie recommends a low protein diet with addition of sufficient alkali to keep the urine alkaline.

Since it is highly probable that infection of the urinary tract plays a very important part in the formation of calculi, whether of cystin or other materials, and that a cystinuric

⁷⁵ *loc. cit.*, sub 69.

⁷⁶ *Archives of Internal Medicine*, 1915, xvi. 503.

whose urine is sterile may live for many years without ever suffering from calculus, therapeutic measures directed against urinary infection may find a place in the rational treatment of the anomaly under consideration.

As has been already pointed out, the excretion of cystin in the urine is, after all, only one of the manifestations of the metabolic anomaly which we know as cystinuria. Other protein fractions than cystin may also be implicated in the error, with the result that cadaverin and putrescin, leucin and tyrosin may be excreted side by side with cystin, but the further aspects of the subject will best be considered in a separate chapter.