

On the cause of the non-precipitation of oxide of copper in testing certain cases of diabetic urine / by T. Lauder Brunton.

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THE CAUSE OF THE NON-PRECI- OF OXIDE OF COPPER

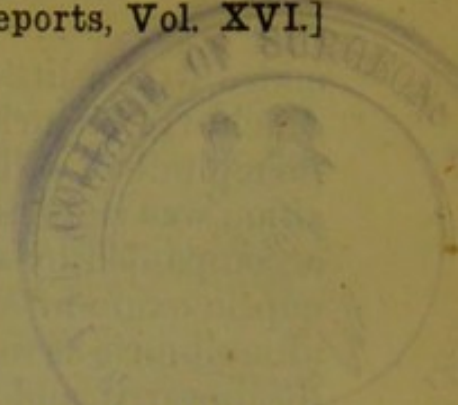
IN TESTING CERTAIN CASES OF DIABETES

BY

T. LAUDER BRUNTON, M.D., F.R.C.S.

In testing for sugar in urine, I have frequently on boiling, after the addition of sulphate of copper, or of Fehling's solution, the blue colour of the precipitate, and was replaced by a brownish-yellow colour than that of the urine itself, but without whatever being thrown down. When this has I have generally tested the urine by Moore's test, or by Fehling's solution, in order to ascertain whether sugar was present. As a rule, I have attributed little importance to the non-precipitation of oxide of copper, and have usually caused by the presence of excess of some reducing or organic compound of unknown nature. However, on examining the urine of a patient known to be diabetic, I found to my astonishment that no precipitate of oxide of copper, although the mixture of urine and cupric solution was placed taken by a tolerably bright yellow opalescent colour, as I did that sugar was generally present in urine, it occurred to me that there might be some cause present in the urine which kept the reduced oxide of copper in solution. It seemed not improbable that if I were to allow the solvent power of the oxide to be exhausted, it might be, as to allow of the oxide being thrown down, I should be able to look about the third of a test-tube full

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ON
THE CAUSE OF THE NON-PRECIPITATION
OF OXIDE OF COPPER

IN TESTING CERTAIN CASES OF DIABETIC URINE.

BY

T. LAUDER BRUNTON, M.D., F.R.S.

In testing for sugar in urine, I have frequently observed that on boiling, after the addition of sulphate of copper and caustic potash, or of Pavy's solution, the blue colour of the mixture disappeared, and was replaced by a brownish-yellow one, somewhat darker than that of the urine itself, but without any precipitate whatever being thrown down. When this has been the case, I have generally tested the urine by Moore's test with liquor potassæ alone, in order to ascertain whether sugar were present or not. As a rule, I have attributed little importance to this decolouration without precipitation of oxide of copper, and have considered it to be usually caused by the presence of excess of uric acid, or to some reducing or organic compound of unknown nature. A few months ago, however, on examining the urine of a patient whom I knew to be diabetic, I found to my astonishment that I got no precipitate of oxide of copper, although I got the blue colour of the mixture of urine and cupric solution destroyed, and its place taken by a tolerably bright yellow opalescence. Knowing as I did that sugar was generally present in specimens of this urine, it occurred to me that there might be some other substance present in the urine which kept the reduced oxide of copper in solution. It seemed not improbable that if I were to dilute the urine, I should so lessen the solvent power of this substance, whatever it might be, as to allow of the oxide being precipitated. I therefore took about the third of a test-tube full of water, and

added to it a little of Pavy's solution, poured into it about half a drachm of the urine, and boiled it; and this time got an abundant precipitate of yellow cupric oxide, clearly showing that my supposition was right, and that the urine contained abundance of sugar, accompanied by some substance which had the power to retain cupric oxide in solution. My patient had been suffering from albuminuria as well as glycosuria, but the albumen had completely disappeared, and the sugar had become greatly diminished under a skim-milk diet. I tested the urine with cold nitric acid, in order to see whether the albumen was still absent, and found that it was so; but I got a purple colouration with the acid, such as is usually attributed to indican. It seemed possible, then, that the substance in the urine which kept the cupric oxide in solution was either indican or some substance accompanying it. I have therefore tested a great number of specimens of urine in the out-patient department of St. Bartholomew's Hospital, and I find, when I get a well-marked purple reaction on the addition of cold nitric acid, that the urine will, to a great extent, prevent the precipitation of oxide of copper. The method I have adopted for demonstrating this fact is the following:—I add to about two drachms of the urine half a drachm of syrup of grape-sugar, containing 10 per cent., and then test for sugar in the ordinary way, either with Pavy's solution or with sulphate of copper and caustic potash. I then find that on boiling I get the colour destroyed and replaced by a yellow, but that no precipitate occurs. Not unfrequently I obtained a dark yellow opalescence, which, by its contrast with the blue solution, might at first be mistaken for an actual precipitate; but, on looking through it, it is found that no precipitate exists, and that the fluid is quite transparent though opalescent, almost like a solution of glycogen. Its transparency is perhaps more readily demonstrated by allowing the light to shine through it upon a piece of white paper held behind it. It is then found that the yellow part of the solution allows the light to pass as readily as the blue, and concentrates it as a bright yellow band upon the paper behind. If a comparative experiment be made with a similar mixture of ordinary urine not giving the purple reaction with nitric acid and grape-sugar, or of water and grape-sugar, the difference will be at once perceived, because the part of the tube in which the oxide has been thrown down is quite opaque, so that one cannot see through it; and when the light is allowed to fall upon it, it throws a dark shadow upon the paper behind, while the part of the solution in which no precipitation has taken place allows the light to pass freely. The curious opalescence resembling that of glycogen is suggestive, but I have made no observations at present in this direction. I cannot at present say that indican is the substance which keeps the oxide of

copper in solution ; indeed, I am strongly inclined to believe it is some substance which may be, and often is, accompanied by indican, but not indican itself. All I can yet do is to mention the non-precipitation of oxide of copper in urines which give a purple reaction with nitric acid, and to draw the attention of the profession to this fallacy in testing for sugar in urine, and to say that one mode of avoiding it is to use more tests than the potash alone ; because I found that the presence of this body, whatever it may be, does not prevent the brown colour occurring on boiling urines containing sugar with caustic potash. The other is, in cases of suspected urine, to use only a small quantity, well diluted, along with Pavy's solution, instead of using a mixture of the solution with pure urine.

Urines which give a purple reaction with nitric acid do not all possess an equal power of retaining oxide of copper in solution, although, so far as my experience goes, they all have this to a greater extent than ordinary urine. Not unfrequently, after a mixture of Pavy's solution with urine giving this reaction has been boiled and allowed to stand for a few minutes, it becomes of a dark-brown colour, and a precipitate may then occur.

According to Kühne,¹ the amount of sugar present in normal urine varies considerably, but one difference can always be observed between the normal which is the richest, and the diabetic urine which is the poorest in sugar. This difference does not consist in the amount of the reduced cuprous oxide, but in the precipitation of this substance. In normal urine the cuprous oxide may be reduced, but it is not precipitated, and this, says Kühne, is what must always be carefully noted in the examination of urine for diabetes. Normal urine must, therefore, in addition to sugar, contain substances which, when mixed with free alkali, are able to retain the cuprous oxide in solution. Cuprous oxide, when once precipitated, is not readily soluble, but certain substances will retain it in solution in considerable quantity if they are present while it is being formed. Ammonia, however, can hardly be the agent by which cuprous oxide is retained in solution in ordinary urine, for although it is produced by heating caustic alkalies with urea, it escapes almost immediately ; and as it is found in diabetic urines as well as in normal, it cannot be the substance which retains the cuprous oxide in solution in normal urine. Moreover, normal urine, when treated with soda and cupric sulphate, does not deposit any cuprous oxide or metallic copper when treated with dilute sulphuric acid, as an ammoniacal solution of cuprous oxide would do. Consequently substances must be present in normal urine which are absent in almost cases of diabetes, and these substances must be excreted by

¹ Kühne's *Physiologische Chemie*, p. 519.

healthy persons in large quantities, for normal urine will often dissolve ten times as much cuprous oxide as it can itself form by reduction. It is only necessary to add to normal urine measured quantities of diabetic urine, or weighed quantities of sugar, in order to convince one's self that even when the amount of sugar is increased by one-half per cent., and sometimes even by 1 per cent., Trommer's test gives a negative result, if one judges by the precipitation of the cuprous oxide. This has hitherto always been the case, and the numerous researches which have been made on the excretion of sugar by the kidneys, after feeding with sugar or its injection into the blood, are therefore to some extent valueless, especially as the experiments have generally been made with dogs, the urine of which animals contains enormous quantities of the substances which dissolve cuprous oxide. Of one of these substances we can speak more definitely. It is, according to Winogradoff, creatinine, an alkaline solution of which retains in solution the cuprous oxide which has been reduced by means of sugar in such a way that it is not precipitated by dilute sulphuric acid. Creatinine, however, is in all probability not the only substance concerned in preventing the precipitation of cuprous oxide in normal urine. In diabetes these substances which precipitate are generally absent, and Winogradoff found in fact, that the excretion of creatinine in many cases of diabetes was diminished or completely arrested. But this is not always the case, for there are some kinds of diabetic urine which allow only a part of the cuprous oxide to be precipitated, and retain another part in solution. Indeed, there are some where the precipitation of cuprous oxide is absent even when the urine contains 1 to $1\frac{1}{2}$ per cent. of sugar. We can readily see that in such cases, when the amount of sugar sinks under 1 per cent., Trommer's test would apparently indicate the entire absence of sugar. "After many years' observation," says Kühne, "I am inclined to suspect that those substances which retain cuprous oxide in solution are found in those cases of diabetes which run a slow course, and in which, with the exception of the high percentage amount of sugar in the urine, the symptoms of diabetes are absent or only slightly marked, and the urine retains its normal depth of colour."

In the most typical cases, on the other hand, with excessive secretion of a very pale urine, with a dry skin, and frequently with the formation of cataract, these substances are either entirely absent from the urine or present in very small quantities. Such cases must, therefore, in addition to the increased formation of sugar, be complicated with another pathological process, which consists in the non-formation or non-excretion of these substances. It would be desirable to investigate how far this last process is

secondary, and produced through the long imbibition of the tissues with sugar.

The case which I have just narrated serves, I think, to show that the non-appearance of these substances in urine is not due to the long-continued action of sugar on the tissues, as Kühne thinks it might possibly be, but is rather owing to a close connection between their formation and the production of sugar itself in the normal manner in the body; for in this case, while the sugar was present in large quantities, the urine presented the ordinary appearance of diabetic urine, but as the amount of sugar excreted in the urine diminished, these substances again made their appearance.

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