A REAGENT FOR THE DETECTION OF REDUCING SUGARS.

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It has already been shown that the hydroxides of the alkali metals have a greater destructive action upon dextrose and various other carbohydrates than have the carbonates,[1] and in accordance with this fact, a copper-containing solution in which the alkalinity is secured by sodium carbonate makes a more delicate and specific test for the detection of dextrose than does a copper solution which contains sodium hydroxide. A reagent of this nature, containing copper sulphate, Rochelle salt, and sodium carbonate, was suggested in a previous paper.[2] This reagent affords a delicate test solution for dextrose, but it has the disadvantage common to so many of the alkaline copper solutions, viz: that after mixing, it rapidly deteriorates and soon becomes useless for detecting small quantities of sugar. For this reason it seemed desirable to obtain a solution in which the alkalinity is secured by carbonate, and which shall at the same time be permanent after mixing.

Rochelle salt is the constituent of the alkaline copper solutions which undergoes change upon standing, and forms products which cause a spontaneous reduction of the solution. As a substitute for the tartrate we may (theoretically) use any aliphatic compound which carries two or more OH radicals, and which is in itself incapable of reducing the copper at a boiling temperature. Substances in great number and variety have been proposed in the literature as substitutes for the Rochelle salt in Fehling's solution. In the present case, where the alkalinity is to be secured by carbonate, it was found that none of the substances usually employed could be used with advantage. Thus glycerol and mannite almost always carry sufficient reducing substance as

[1] Benedict: This *Journal* iii, p. 101, 1907.

[2] Benedict: *loc. cit.*

impurity to affect the carbonate reagent, even where there is not enough present to reduce Fehling's fluid.

Citric acid (in the form of its salts) should, from the theoretical point of view, be capable of holding cupric hydroxide in solution in an alkaline medium. Upon practical test it has been found to be a most satisfactory substance for this purpose. The following formula yields a satisfactory reagent:

Copper Sulphate (pure crystallized)	17.3	grams.
Sodium Citrate.[1]	173.0	"
Sodium Carbonate (anhydrous)	100.0	"
Distilled Water	to 1000.0	cc.

With the aid of heat dissolve the sodium citrate and carbonate in about 600 cc. of water. Pour (through a folded filter if necessary) into a graduate and make up to 850 cc. Dissolve the copper sulphate in about 100 cc. of water and make up to 150 cc. Pour the carbonate-citrate solution into a large beaker or casserole and add the copper sulphate solution slowly, with constant stirring. The mixture is ready for use.

This reagent is more sensitive to dextrose either in pure solution or in urine than is Fehling's fluid, is not reduced by uric acid (or appreciably by chloroform, chloral, or formaldehyde), and appears to suffer no deterioration on standing. The solution is not caustic and may be kept in cork or glass stoppered bottles. Samples of this solution prepared somewhat over a year ago appear to be in as good condition, in every respect, as when freshly prepared. These were kept in partially filled, uncolored glass bottles, exposed to light, heat, etc. A recent examination of these samples showed that not only had they undergone no spontaneous reduction, but that no sign of reduction or other alteration occurred upon heating for twenty-four hours in a bath of boiling water. (The heating was not continued longer.) Fehling's solution, freshly prepared and mixed, subjected to this treatment showed a marked precipitation of cuprous oxide after three hours heating, and this precipitate increased continuously during the subsequent heating.

[1] The ordinary sodium citrate of the drug trade appears to be sufficiently pure for use in this reagent. An examination of several samples purchased in the open market failed to reveal the presence of objectionable substances.

The following points may be mentioned in connection with the use of this reagent. No strongly dehydrating substance (such as potassium hydroxide) is present; hence upon reduction this solution is more apt to yield the hydrated oxides than is Fehling's solution. Thus the reduction product is frequently yellow or green, rather than red, as in Fehling's test. The reagent is not dark colored, like the hydroxide-containing solutions, and even the slightest precipitates may readily be observed without waiting for them to settle. For general work the solution is used just as is Fehling's fluid, save that it is desirable to continue the boiling for from one to two minutes, and then let the tube cool spontaneously.

The following is the procedure for the detection of dextrose in the urine. To about 5 cc. of the reagent in a test tube are added 8 (not more) drops of the urine to be examined. The fluid is then heated to boiling, kept at this temperature for from one to two minutes, and allowed to cool *spontaneously*. In the presence of dextrose the *entire body of the solution will be filled with a precipitate*, which may be red, yellow, or green. If the amount of dextrose is small, the precipitate forms only upon cooling. If no dextrose is present the solution either remains absolutely clear, or a very faint turbidity, due to precipitated urates, may be apparent. Even small quantities of dextrose in urine (0.1 per cent) yield precipitates of surprising bulk with this reagent, and the positive reaction consists in the filling of the entire body of the solution with a precipitate so that the solution becomes opaque. Since bulk, rather than color, of the precipitate is made the basis of the reaction, this test may be applied, even for the detection of small quantities of dextrose, as readily in artificial, as in day light. Urines containing 0.08 per cent dextrose give a very positive reaction with this test. Fehling's solution requires the presence of about 0.12 per cent of dextrose in urine to yield an equally positive test.

It is hoped to present a later paper in which certain other applications of this reagent will be discussed, including its employment in quantitative processes.

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