

XXII.—*The Solution-density and Cupric-reducing Power of Dextrose, Levulose, and Invert-sugar.*

By HORACE T. BROWN, F.R.S., G. HARRIS MORRIS, Ph.D., and J. H. MILLAR.

The Solution-densities.

IN a previous communication (Trans., 1897, 71, p. 72), we have entered fully into the necessity for the accurate determination of the solution-densities of the carbohydrates, in order that the correct factors for the estimation of the solid matter in carbohydrate solutions may be deduced. In the paper above referred to, we gave the results of the accurate determination of the solution-densities and divisors for various concentrations of maltose, soluble-starch, and the products of the conversion of starch by diastase. We have now carried out a series of similar determinations for dextrose, levulose, and invert-sugar.

The apparatus used for drying the respective sugars was that described in our former paper (this vol., p. 76). In this, crystallised dextrose can be rendered perfectly dry in 4 hours at 105–106° without the slightest decomposition or coloration taking place. Crystallised levulose can be dried still more readily, but the operation requires greater care, on account of the low melting point of this sugar. The temperature of the bath must not be allowed to rise

276 BROWN, MORRIS, AND MILLAR: THE SOLUTION-DENSITY AND

above 75° for the first three or four hours, but after that it may rise to 85—90°. The latter temperature must not be exceeded, or the substance fuses and begins to colour. A total period of 6 hours at the given temperatures is amply sufficient to drive off all moisture without the slightest discoloration taking place.

The sugars used in the experiments were prepared with the greatest possible care by the most approved methods, and were repeatedly recrystallised from alcohol until their properties were absolutely constant. The dextrose was prepared from three sources, namely, from pure maltose by acid hydrolysis; from cane-sugar by inversion with acid, and from a crystallised sample of dextrose obtained from Kahlbaum. The levulose was prepared in two ways, the one from inulin in the manner described by Wohl (*Ber.*, 1890, 23, 2107), and the other from a sample of Schering's crystallised product. Invert-sugar was obtained by the inversion of pure, crystallised cane-sugar with yeast, and in some experiments, by mixing equal quantities of perfectly pure dextrose and levulose.

In the results given in the following tables,

Column *a* gives the weight of dry substance taken.

Column *b* gives the total weight of solution.

Column *c* gives the specific gravity of the solution at 15·5°, referred to water at 15·5°.

Column *d* gives grams of sugar per 100 c.c. (reputed)*.

Column *e* gives the divisor for the determination of grams per 100 c.c. (reputed) from the specific gravity.

Table I.—Solution-density of Anhydrous Dextrose.

	<i>a.</i>	<i>b.</i>	<i>c.</i>	<i>d.</i>	<i>e.</i>
1	1·2988	49·8518	1010·12	2·6317	3·845
2	1·6324	30·5690	1020·95	5·4516	3·842
3	3·2659	51·2638	1025·07	6·5306	3·839
4	3·2751	31·1712	1041·94	10·9470	3·831
5	8·3380	53·7774	1062·93	16·4800	3·818
6	6·4414	36·5604	1072·03	18·8870	3·813
7	9·0925	39·9196	1094·66	24·9330	3·796

The above results are expressed graphically in the dextrose curve of Plate I. (p. 278),* and the true divisor for any particular concentration can be obtained by inspection of this curve, or from the following equation, in which *D* is the required divisor, and *G* the specific gravity of the solution when water = 1000.

$$D = 3·848 - 0·00028 (G - 1000) - 0·0000028 (G - 1000)^2.$$

* For the meaning of this expression see this vol., p. 77, footnote.

The divisors for dextrose deduced from the foregoing experiments agree very closely with those recently given by C. O'Sullivan and Stern (Trans., 1896, 69, 1695).

Table II.—Solution-density of Anhydrous Levulose.

	a.	b.	c.	d.	e.
1	1.2326	50.5868	1009.72	2.4676	3.939
2	3.1436	51.3039	1024.67	6.2784	3.929
3	2.5962	40.7573	1025.65	6.5330	3.926
4	2.0206	27.5294	1029.67	7.5576	3.925
5	6.5677	54.0685	1049.85	12.7530	3.909
6	4.8289	38.6476	1051.35	13.1360	3.909
7	5.0967	32.5304	1065.06	16.6870	3.898
8	7.0191	34.0505	1087.10	22.4000	3.886

The foregoing results are embodied in the levulose curve on Plate I., the equation for which is

$$D = 3.946 - 0.00068 (G - 1000) - 0.0000007 (G - 1000)^2.$$

We have already fully discussed (*loc. cit.*) the relation of the divisor we have hitherto employed (3.86) to the true divisors for the carbohydrates, and it is, therefore, unnecessary to again refer to the matter. We may, however, point out that here, as in the cases we have previously given, the divisor decreases as the concentration of the solution increases. So far as levulose is concerned, this is in direct opposition to the results obtained by Hönig and Jesser (*Monatshefte*, 1888, 9, 562), from which it appears that the divisor increases with the concentration.

Since it has been conclusively shown that invert-sugar is a mixture of equal amounts of dextrose and levulose, we calculated from the foregoing results the divisor to be applied to solutions of invert-sugar. This was done by taking the mean of the divisors for dextrose and levulose for solutions of one-half the gravity of that of the invert-sugar solutions; thus, the divisors for dextrose and levulose for a solution of sp. gr. 1010 are 3.845 and 3.939 respectively; the mean of these is 3.892, which was taken as the divisor for a solution of invert-sugar of sp. gr. 1020. We are aware that this is not quite correct, as solutions of the same specific gravity do not contain exactly equal amounts of dextrose and levulose, and, moreover, no allowance is made for the lower solution-density of the solution of double strength; but the error thus introduced is so small, being in the fourth place of decimals, that it falls well within the limits of experimental error. In this way, we obtained the following table.

Table III.—Divisors for Anhydrous Invert-sugar.

Gravity.	Divisor.	Gravity.	Divisor.
1010	3·894	1070	3·877
1020	3·892	1080	3·874
1030	3·889	1090	3·871
1040	3·886	1100	3·868
1050	3·883	1110	3·865
1060	3·880	1120	3·862

The foregoing values were checked at several points by determinations made with pure invert-sugar, and were found to be correct. They are expressed graphically in the invert-sugar curve of Plate I, the equation for which is

$$D = 3.897 - 0.00025 (G - 1000) - 0.0000004 (G - 1000)^2.$$

The Cupric-reducing Powers.

In the paper already mentioned (*loc. cit.*, p. 95, *et seq.*), we discussed, at considerable length, the conditions requisite for the accurate and concordant determination of the cupric-reduction of the carbohydrates, but it will be advisable to briefly recapitulate them here. They are (1) the use of a Fehling's solution of constant composition; (2) the maintenance of the same degree of dilution in all experiments; (3) the precipitation of an amount of copper oxide which shall fall between certain limits; and (4) an invariable method of determination, both as regards mode and time of heating.

The composition of the Fehling's solution we use is

Recrystallised copper sulphate ...	34.6	grams	per litre.
Rochelle salt	173.0	„	„
Anhydrous sodium hydroxide ...	65.0	„	„

Fifty c.c. of this solution, which should be freshly mixed, are placed in a beaker of about 250 c.c. capacity and having a diameter of 7.5 centimetres. This is placed in a boiling water bath, and when the solution has attained the temperature of the water, the accurately weighed or measured volume of the sugar solution is added, and the whole made up to 100 c.c. with boiling distilled water. The beaker, which is covered with a clock glass, is then returned to the water bath, and the heating continued for exactly 12 minutes. The precipitated cuprous oxide is now rapidly filtered off through a Soxhlet tube, washed first with hot water, then with alcohol and ether, and finally dried. When dry, the cuprous oxide is reduced to metallic copper by gentle heating in a stream of hydrogen, and weighed. The weight of copper obtained must be corrected for that due to the slight spontaneous reduction which always

PLATE I

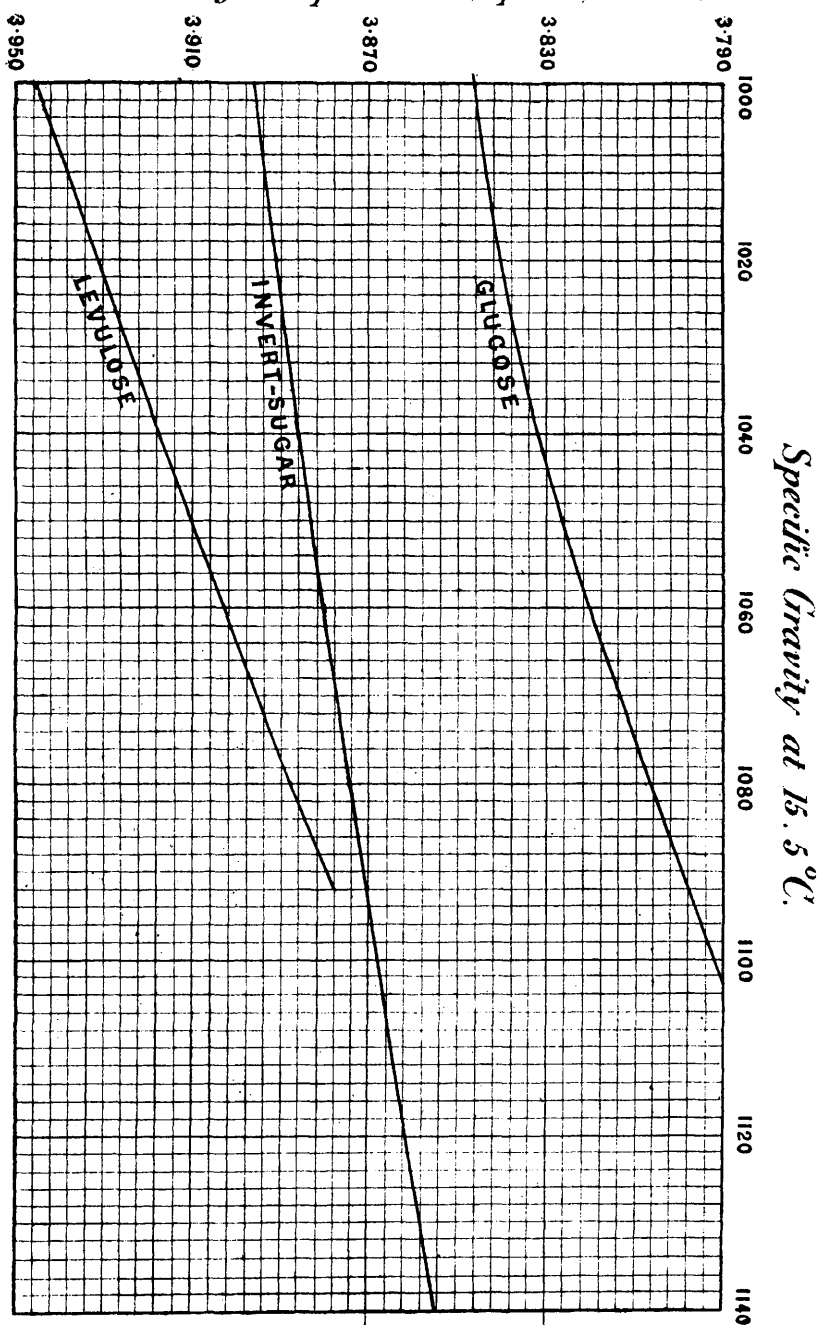
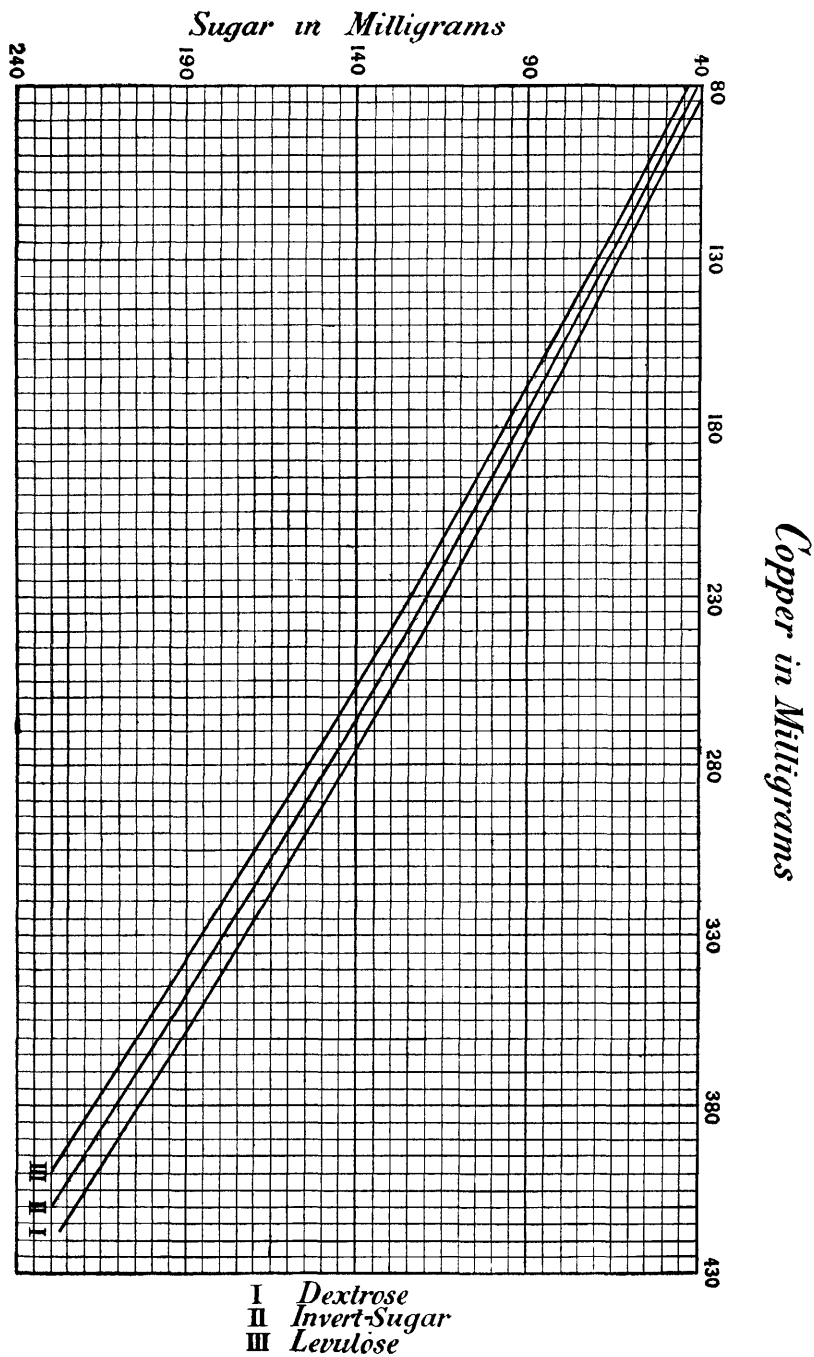
Divisor for grams per 100 c.c. (reputed) at 15.5°C.

PLATE II



CUPRIC-REDUCING POWER OF DEXTROSE, LEVULOSE, ETC. 279

takes place, and which should be determined for each batch of Fehling's solution.

Working in this way, we have determined with great care the cupric-reducing power of dextrose, levulose, and invert-sugar, taking in each series of experiments gradually increasing amounts of the sugar, and determining the copper reduced by each amount. The results are embodied in the following tables, which are given in the same form as those in our previous paper, to which reference has been made. The sugars used were prepared as indicated in the first part of this paper, and were most carefully and thoroughly purified. In the dextrose table, the source from which the sugar was obtained is indicated in the first column; the determinations marked *a* were made with dextrose prepared from Kahlbaum's product; *b*, from maltose, by acid hydrolysis; and *c*, from cane-sugar, by inversion with acid.

Table IV.—Cupric-reduction of Dextrose.

Source.	Dextrose by 3·86 divisor.	Dextrose absolute.	Cu weighed.	CuO per gram of 3·86 dextrose.	CuO per gram of absolute dextrose.	K3·86.	Kabsolute.
<i>b</i>	0·0457	0·0459	0·0938	2·572	2·562	116·6	116·2
<i>a</i>	0·0474	0·0476	0·0983	2·599	2·589	117·9	117·4
<i>c</i>	0·0491	0·0493	0·1024	2·614	2·604	118·5	118·1
<i>c</i>	0·0736	0·0739	0·1513	2·577	2·567	116·8	116·4
<i>b</i>	0·0884	0·0888	0·1809	2·564	2·554	116·3	115·8
<i>a</i>	0·0950	0·0954	0·1953	2·577	2·567	116·8	116·4
<i>c</i>	0·0980	0·0984	0·1996	2·553	2·543	115·8	115·3
<i>c</i>	0·1180	0·1185	0·2369	2·516	2·507	114·1	113·7
<i>b</i>	0·1338	0·1344	0·2648	2·480	2·470	112·4	112·0
<i>a</i>	0·1425	0·1431	0·2836	2·494	2·485	113·1	112·7
<i>c</i>	0·1585	0·1592	0·3089	2·442	2·433	110·7	110·3
<i>b</i>	0·1786	0·1793	0·3397	2·385	2·375	108·1	107·7
<i>a</i>	0·1911	0·1919	0·3635	2·384	2·375	108·1	107·7
<i>c</i>	0·1968	0·1976	0·3732	2·377	2·368	107·8	107·4
<i>b</i>	0·2105	0·2114	0·3920	2·334	2·325	105·8	105·4

The above results are expressed graphically in curve I of Plate II (p. 280), which shows that the copper oxide reduced by 1 gram of dextrose appreciably decreases as the amount of copper reduced increases.

Table V.—*Cupric-reduction of Levulose.*

Levulose by 3·86 divisor.	Levulose absolute.	Cu weighed.	CuO per gram of 3·86 levulose.	CuO per gram of absolute levulose.	K _{3·86} .	K _{absolute} .
0·0403	0·0396	0·0750	2·329	2·375	105·6	107·7
0·0443	0·0435	0·0822	2·323	2·369	105·3	107·4
0·0446	0·0438	0·0831	2·332	2·379	105·7	107·9
0·0473	0·0466	0·0887	2·323	2·360	105·4	107·0
0·0507	0·0500	0·0939	2·318	2·355	105·2	106·8
0·0964	0·0949	0·1765	2·295	2·332	104·1	105·7
0·0964	0·0949	0·1765	2·295	2·332	104·1	105·7
0·0964	0·0949	0·1770	2·301	2·338	104·4	106·0
0·0964	0·0949	0·1787	2·324	2·361	105·3	107·0
0·1455	0·1432	0·2612	2·251	2·287	102·1	103·7
0·1443	0·1421	0·2601	2·260	2·295	102·4	104·0
0·1438	0·1416	0·2585	2·254	2·289	102·2	103·8
0·1890	0·1861	0·3290	2·182	2·217	99·0	100·5
0·2011	0·1980	0·3507	2·186	2·221	99·2	100·7
0·2022	0·1991	0·3526	2·186	2·220	99·2	100·7
0·2043	0·2012	0·3576	2·194	2·228	99·5	101·0

From the foregoing numbers, curve III of Plate II has been constructed. It will be seen that, throughout the series, any given amount of levulose reduces appreciably less copper than the same weight of dextrose.

Table VI.—*Cupric-reduction of Invert-sugar.*

Invert-sugar by 3·86 divisor.	Invert-sugar absolute.	Cu weighed.	CuO per gram of 3·86 invert- sugar.	CuO per gram of absolute invert- sugar.	K _{3·86} .	K _{absolute} .
0·0506	0·0502	0·0979	2·426	2·445	110·0	110·9
0·0860	0·0854	0·1681	2·449	2·468	111·0	111·9
0·1095	0·1087	0·2085	2·387	2·405	108·2	109·0
0·1303	0·1293	0·2511	2·416	2·435	109·5	110·4
0·1582	0·1570	0·2965	2·350	2·368	106·5	107·4
0·1847	0·1833	0·3386	2·298	2·316	104·2	105·0
0·2167	0·2152	0·3890	2·250	2·267	102·0	102·8

The above results are expressed graphically in the invert-sugar curve II of Plate II, which falls midway between the curves for dextrose and levulose.

From the curves obtained when the foregoing experimental results are plotted on a system of rectangular co-ordinates, we have constructed the following table showing the reducing powers of the three

CUPRIC-REDUCING POWER OF DEXTROSE, LEVULOSE, ETC. 281

sugars in question. We have given, in the first column, the quantities of sugar from 50 to 205 milligrams, at intervals of 5 milligrams, and, opposite to these, the amounts of Cu and CuO precipitated by each quantity, together with the weight of CuO corresponding to 1 gram of each sugar, when the respective quantities are oxidised under the conditions of our method.

Table VII.

Sugar mngs.	Dextrose.			Levulose.			Invert-sugar.		
	Cu grams.	CuO grams.	CuO corresponding to 1 gram.	Cu grams.	CuO grams.	CuO corresponding to 1 gram.	Cu grams.	CuO grams.	CuO corresponding to 1 gram.
50	0.1030	0.1289	2.578	0.0923	0.1155	2.310	0.0975	0.1221	2.442
55	0.1134	0.1422	2.585	0.1027	0.1287	2.341	0.1076	0.1349	2.453
60	0.1238	0.1552	2.587	0.1122	0.1407	2.345	0.1176	0.1474	2.457
65	0.1342	0.1682	2.589	0.1216	0.1524	2.346	0.1275	0.1598	2.459
70	0.1443	0.1809	2.585	0.1312	0.1645	2.350	0.1373	0.1721	2.459
75	0.1543	0.1935	2.580	0.1405	0.1761	2.349	0.1468	0.1840	2.454
80	0.1644	0.2061	2.577	0.1500	0.1881	2.351	0.1566	0.1963	2.454
85	0.1740	0.2187	2.572	0.1590	0.1993	2.345	0.1662	0.2084	2.451
90	0.1834	0.2299	2.555	0.1686	0.2114	2.349	0.1755	0.2200	2.445
95	0.1930	0.2420	2.547	0.1774	0.2224	2.341	0.1848	0.2317	2.439
100	0.2027	0.2538	2.538	0.1862	0.2331	2.331	0.1941	0.2430	2.430
105	0.2123	0.2662	2.535	0.1952	0.2447	2.331	0.2034	0.2550	2.429
110	0.2218	0.2781	2.528	0.2040	0.2558	2.325	0.2128	0.2668	2.425
115	0.2313	0.2900	2.522	0.2129	0.2669	2.321	0.2220	0.2783	2.420
120	0.2404	0.3014	2.512	0.2215	0.2777	2.314	0.2311	0.2898	2.415
125	0.2496	0.3130	2.504	0.2303	0.2887	2.310	0.2400	0.3009	2.407
130	0.2585	0.3241	2.493	0.2390	0.2997	2.305	0.2489	0.3121	2.400
135	0.2675	0.3354	2.484	0.2477	0.3106	2.300	0.2578	0.3232	2.394
140	0.2762	0.3463	2.473	0.2559	0.3209	2.292	0.2663	0.3339	2.385
145	0.2850	0.3573	2.464	0.2641	0.3311	2.284	0.2750	0.3448	2.378
150	0.2934	0.3673	2.448	0.2723	0.3409	2.273	0.2832	0.3546	2.364
155	0.3020	0.3787	2.443	0.2805	0.3517	2.269	0.2915	0.3655	2.358
160	0.3103	0.3891	2.432	0.2889	0.3622	2.264	0.3002	0.3764	2.352
165	0.3187	0.3996	2.422	0.2972	0.3726	2.258	0.3086	0.3869	2.345
170	0.3268	0.4098	2.410	0.3053	0.3828	2.252	0.3167	0.3971	2.336
175	0.3350	0.4200	2.400	0.3134	0.3930	2.245	0.3251	0.4076	2.329
180	0.3431	0.4302	2.390	0.3216	0.4032	2.240	0.3331	0.4177	2.320
185	0.3508	0.4399	2.377	0.3297	0.4134	2.234	0.3410	0.4276	2.311
190	0.3590	0.4501	2.369	0.3377	0.4234	2.228	0.3490	0.4376	2.303
195	0.3668	0.4599	2.358	0.3457	0.4335	2.223	0.3570	0.4476	2.295
200	0.3745	0.4689	2.344	0.3539	0.4431	2.216	0.3650	0.4570	2.285
205	0.3822	0.4792	2.338	0.3616	0.4534	2.211	0.3726	0.4672	2.279

Before commenting on our results, it will be necessary to refer briefly to the method at present in use of expressing the cupric-reducing power of the sugars. In 1876, C. O'Sullivan (Trans., 1876, ii, 125),

defined this "to be the amount of cupric oxide, calculated as dextrose, which 100 parts reduce." The cupric reduction of dextrose would then be 100, and 1 gram of this sugar was considered to reduce 2.205 grams of copper oxide. O'Sullivan expressed this value by K , and the symbol κ had previously been used by one of us and Heron (Trans., 1879, 35, 607) with the same significance.

It has long been known that the 2.205 value for dextrose was not quite correct, being, in fact, too low, and consequently that the true cupric-reduction of dextrose was distinctly above 100 when referred to this standard. As we have, however, already pointed out at length, in the paper previously referred to (this vol., p. 97), this is immaterial if it is clearly and definitely understood to what basis the cupric-reducing power is referred.

From the foregoing experiments with dextrose, it will be seen that we have obtained considerably higher values than those mentioned above. We find that the amount of copper oxide reduced by 1 gram of the sugar ranges from 2.562 to 2.325 grams, according to the extent to which reduction of the Fehling's solution is carried; and on the 2.205 basis, these numbers correspond to a κ of 116.2 and 105.4 respectively. Recently, and after our experiments were finished, Heron (*Journal Federated Institutes Brewing*, 1896, 2, 443) and C. O'Sullivan and Stern (Trans., 1896, 69, 1691) pointed out the higher reducing power of dextrose. The former states that 1 gram reduces 2.26 grams of cupric oxide, but he does not mention the precise conditions under which this value was obtained. The latter authors give 2.306 grams of CuO as the equivalent of 1 gram of sugar, and the mean reducing value as $\kappa = 104.6$; they use 30 c.c. of Fehling's solution diluted to at least 90 c.c., and take from 0.12 to 0.13 gram of sugar. Under these conditions, we should expect that their results would be somewhat lower than ours.

The method employed by Kjeldahl in his investigations on the cupric-reduction of the sugars (*Résumé du Compte-rendu des travaux du laboratoire du Carlsberg*, 4^{me} vol., 1^{re} livr., 1895), very closely resembles ours, with the exception that the Fehling's solution is heated for 20 minutes, and the reduction carried on in an atmosphere of hydrogen. As would be expected from this variation in the conditions, his values for the cupric-reduction of dextrose are uniformly higher than ours; but the difference is fairly constant throughout the series. This, we have already shown, was also the case with maltose (*loc. cit.*).

It has always been generally held that the reducing powers of levulose and invert-sugar were less than that of dextrose. This fact is well shown in the foregoing results, and also in Table VII., which gives the amount of copper oxide reduced by 1 gram of each sugar at different stages of reduction. It has been stated by J. O'Sullivan

(Trans., 1892, 61, 408) and by Heron (*loc. cit.*) that the reducing power of invert-sugar is the same as that of dextrose, but this is negatived by our results, which are, on this point, in accord with those of Kjeldahl (*loc. cit.*), Ost (*Ber.*, 1890, 23, 3003), Hönig and Jesser (*Monatshefte*, 1888, 9, 562), Soxhlet (*J. pr. Chem.*, 1880, 21, 227), and other workers.

The results obtained by these workers, although not exactly the same as ours, owing to the differing conditions of experiment, yet show that levulose has an appreciably lower reducing power than dextrose, and that the cupric-reduction of invert-sugar stands intermediate between the two.

If we take the results expressed in Table VII, and calculate from them, at certain points, the cupric-reducing power, κ , expressed on the old basis of 2·205, we get the following values for the three sugars.

Milligrams Sugar.	Dextrose κ .	Levulose κ .	Invert-sugar κ .
50	116·9	104·7	110·7
100	115·1	105·7	110·2
150	111·0	103·1	107·2
200	106·3	100·1	103·6

If, however, we take the value of dextrose at each of the above points as 100, and express the values of levulose and invert-sugar as percentages on this number, we get the following results.

Milligrams Sugar.	Dextrose.	Levulose.	Invert-sugar.
50	100	89·60	94·72
100	100	91·84	95·74
150	100	92·85	96·56
200	100	94·11	97·05

We see, then, from these numbers, that if the reducing power of dextrose be taken as 100, when what may be regarded as the usual amount of copper is reduced (150 to 200 milligrams), the values to be assigned to levulose and invert-sugar closely approximate to those which have been usually taken for these sugars.

In our former paper, to which reference has repeatedly been made, we showed the great influence which the amount and nature of the alkali in the Fehling's solution exercised on the quantity of copper reduced by a given weight of maltose, or of the starch-transformation products. With dextrose and levulose, the influence is far less. Kjeldahl has shown (*loc. cit.*), and we have confirmed the observation, that the amount of sodium hydroxide per litre may be varied within fairly-wide limits, without producing any considerable alteration in the amount of copper reduced by a given weight of dextrose; whilst

Glendinning has proved (Trans., 1895, 67, 999) that an equivalent amount of potassium hydroxide may be substituted for the sodium compound without causing any alteration in the reducing power.

The variant which has the greatest influence in the case of dextrose and levulose is the state of dilution of the Fehling's solution. If the 50 c.c. of Fehling's solution is diluted with 100 c.c., 150 c.c., or 200 c.c. of water, instead of with the 50 c.c. of our standard method, the reducing power is appreciably lower at all stages of reduction, and the greater the dilution, the greater the difference. This is well seen in the following table, in which the results are given in terms of κ absolute.

Table VIII.—Reducing Power of Dextrose and Levulose at different Degrees of Dilution.

Sugar grams.	Dextrose.			Levulose.		
	Dilution 1 : 2.	Dilution 1 : 3.	Dilution 1 : 4.	Dilution 1 : 2.	Dilution 1 : 3.	Dilution 1 : 4.
0.0725	115.7	115.2	113.0	—	—	—
0.0846	—	—	—	105.0	102.6	102.9
0.1120	114.6	113.9	111.8	—	—	—
0.1275	—	—	—	104.1	102.2	101.9
0.1697	—	—	—	101.9	100.9	99.8
0.1830	108.1	107.8	107.2	—	—	—
0.1901	—	—	—	101.0	99.7	98.9

The degree of dilution has, however, a much greater influence on the reducing power if the experiments are made in a different manner, namely, by keeping the total volume of solution constant, and decreasing the amount of Fehling's solution in such volume. Kjeldahl made a series of experiments in this way, using 15, 30, 50, and 75 c.c. respectively of Fehling's solution and making up the volume in each case to 100 c.c. When nearly the maximum amount of copper was precipitated from the more dilute solutions, much lower values than those given above were obtained for the cupric-reducing powers, and we have confirmed these observations by direct experiment.

In our former paper (this vol., p. 106), we fully discussed the limits of error of the methods employed for the determination of solution density and cupric-reducing power, and they, therefore, need not be recapitulated here.