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## A NEW METHOD FOR THE DETERMINATION OF THE REDUCING SUGARS.

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In the study of velocity of amyolytic action it became desirable to determine with the greatest possible accuracy the reducing sugars resulting from the digestion of starch. This led to the following study of some modifications of Fehling's reagent with a view to establishing the optimum medium and conditions for a gravimetric method of determining reducing power.

Benedict<sup>1</sup> and others have pointed out the fact that glucose is more readily destroyed with sodium hydroxide than with sodium carbonate, and that larger amounts of copper are reduced by the same weight of sugar if sodium carbonate is used in place of sodium hydroxide. A comparison of the results obtained with three of the more common methods and a copper solution where the sodium hydroxide is replaced with sodium carbonate follows.

50 mg. of glucose reduces according to:

Defren's method.	Allihn's method.	Munson and Walker's method.	Na <sub>2</sub> CO <sub>3</sub> solution replacing NaOH.
89.8 mg. Cu.	98.2 mg. Cu.	102.0 mg. Cu.	147 mg.

We thus see that the use of sodium hydroxide gives only about two-thirds of the amount of copper reduced which may be obtained with the carbonate.

After a series of experiments with the various alkalies, it was found that potassium carbonate was the one best suited to furnish the alkalinity. As the reducing power of glucose decreases with increase in the volume of the solution, it is necessary to have the volume of the solutions containing the copper and alkali which are added to the sugar solution as small as possible. Potassium carbonate is better than sodium carbonate, being much more soluble and having a slight advantage in giving more copper reduced for the same weight of glucose. Some experiments showing the relation between these two carbonates are as follows: 50 mg. of glucose gave with

- 5 g. sodium carbonate, 129.8 mg. copper.
- 10 g. sodium carbonate, 133.3 mg. copper.
- 15 g. sodium carbonate, 133.3 mg. copper.

All conditions being the same with

- 12 g. potassium carbonate, 140.4 mg. copper.
- 14 g. potassium carbonate, 142.8 mg. copper.
- 16 g. potassium carbonate, 140.4 mg. copper.

<sup>1</sup> *J. Biol. Chem.*, 3, 101 (1907); 5, 485 (1908).

Having found that potassium carbonate was best suited to furnish the alkalinity to the copper solution, it still remained to determine whether a better medium than Rochelle salts could be found to hold the copper in solution.

Some results according to Munson and Walker's method showed that unless the spontaneous reducing power of the alkaline tartrate solution is determined and allowed for, the results obtained will be considerably too high. Some results we obtained, using the method of Munson and Walker, but not allowing for the reducing power of the tartrate solution, are:

Sugar taken.	Copper found.	Sugar equivalent given in table.
60	126.5	62.4
60	124.6	61.4
100	203.0	102.5
100	202.7	102.3

In their original article<sup>1</sup> Munson and Walker give a series of figures showing the reducing power of their alkaline tartrate solution from day to day during the course of the investigation. These results varied from 0 to 2.0 mg. of cuprous oxide. Apparently the reducing power of the alkaline tartrate solution varies with different samples of Rochelle salts and it is imperative to make some correction for all samples which we have examined.

The amount of copper reduced by the alkaline tartrate seems to be much greater when the reduction takes place in a bath of boiling water than it does when the solution is heated for a short time over the flame as in Munson and Walker's method. By heating for 20 minutes in a bath of boiling water 50 cc. of the mixed Defren solution in a total volume of 150 cc. may reduce as much as 7 mg. of copper. Provided the reduction was caused by an impurity in the tartrate it would be possible to free the solutions from such impurities by treating the alkaline tartrate with a copper solution, reduce by heating in a bath of boiling water, filter, and use the resulting solution which would have no reducing power of its own. This was tried and it was then found that when such a solution was heated again in the boiling water a second reduction, as large as the first, took place. If the cuprous oxide was then filtered off and the solution again heated, a third reduction took place. Since this showed that the reduction is due to the tartrate itself, and that a previous reduction is not capable of removing the source of error, it seemed imperative to find some medium other than Rochelle salts for keeping the copper in solution.

Many compounds have been proposed for this purpose, among which

<sup>1</sup> THIS JOURNAL, 28, 663; 29, 541.

may be mentioned the bicarbonate solution of Soldaini<sup>1</sup> and the citrate solution proposed by Benedict.<sup>2</sup> In Benedict's volumetric method for the determination of sugar the disappearance of the blue color is taken as the end point of the titration and this method allows of an accurate determination of the sugar. Experiments were therefore made to see if Benedict's citrate solution could be adapted to a gravimetric method. It was then found that marked changes in the reducing powers of the sugars followed changes in concentration of the sodium carbonate and citrate.

Thus, using 50 mg. of glucose and 2 g. of copper sulfate in a volume of 150 cc., the following weights of copper, expressed in milligrams, were obtained under the conditions as given below, when heated for 20 minutes in a bath of boiling water:

Sodium citrate. Grams.	Sodium carbonate. 5 grams. Mg.	Sodium carbonate. 10 grams. Mg.	Sodium carbonate. 15 grams. Mg.
5	116.5	134.1	144.2
10	100.0	117.3	127.1
15	89.3	107.9	122.0
20	65.3	101.8	117.8

The change in the reducing power of sugar is explained only in part by assuming that the citrate solution dissolved the reduced cuprous oxide. This was shown by placing 150 mg. of Kahlbaum's cuprous oxide in each of four flasks and heating it under identical conditions of volume, time, and concentration of solution as in the above experiments. Oxidation of the cuprous oxide during the heating was prevented by displacing the air in the flask with illuminating gas and closing the flask with a two-hole stopper.

Determination of the copper content of 150 mg. of the cuprous oxide used showed on duplicate determination 125.6 and 126.1 mg. of copper, average 125.9 mg. All conditions being the same as above, the following weights of cuprous oxide were recovered after heating for 20 minutes:

Sodium citrate. Grams.	Sodium carbonate. 5 grams. Mg.	Sodium carbonate. 15 grams. Mg.
5	125.2	...
10	123.3	122.4
15	120.2	122.4
20	118.3	122.1

As 20 g. of sodium citrate in the presence of 5 g. of sodium carbonate could dissolve but 7.6 mg. of cuprous oxide the low results obtained with the sugar must be due to a depression of the reducing power of the sugar by the citrate. The results of other experiments in which the weights

<sup>1</sup> *Gaz. chim. ital.*, 6, 322.

<sup>2</sup> *J. Biol. Chem.*, 5, 485 (1908).

of sugar varied showed that a citrate solution does not furnish a satisfactory solution in a gravimetric method.

As the spontaneous reduction of Rochelle salts and the depression and variations caused in the reducing power of sugar by sodium citrate are serious objections to these two salts, further work was done to find some other agent for holding the copper in solution.

Theoretically, any organic compound having a carboxyl and alcohol group is capable of holding the copper in solution in an alkaline mixture. Glycerol and mannite have also been suggested as possible agents, but they do not furnish a convenient solution with which to work. Lactic acid will hold the copper in solution, but the reducing power of sugar is but slight in such a solution.

Among a number of organic compounds which were tried, salicylic acid was found to be one which will furnish a medium for the reduction of sugar, but which has no reducing power of its own and will not dissolve the cuprous oxide.

An alkaline salicylate solution replacing the alkaline tartrate showed no reduction of copper when heated in a bath of boiling water for 7 hours, and the following results show that there is no appreciable change in reducing power with small changes in concentration of the salicylic acid.

50 mg. glucose. 5 grams sodium carbonate in 150 cc. volume.

3 grams salicylic acid, 125.7 mg. copper.

4 grams salicylic acid, 126.0 mg. copper.

5 grams salicylic acid, 125.7 mg. copper.

6 grams salicylic acid, 124.8 mg. copper.

Further experiments showed that with the other reducing sugars, maltose, lactose, and invert sugar, the alkaline salicylate solution furnishes a satisfactory medium for the reduction of the copper.

It now remained to determine what weights of copper, potassium carbonate and salicylic acid give the optimum conditions for the reduction of the copper.

In Munson and Walker's conditions 1.858 grams of copper sulfate (crystalline) are used per determination, the largest weight of copper reduced being 435.3 mg. While larger amounts of copper give greater reducing powers to the sugars, it was decided to use two grams of copper sulfate (crystalline) per determination and limit the reduction to 450 mg. of copper.

The weights of potassium carbonate and salicylic acid which give the optimum conditions for maltose were determined and these weights were used for the determination of the reducing power of the other sugars.

The effect of varying amounts of potassium carbonate and salicylic acid is shown in the following table. The volume was 140 cc. and 2 g. of copper sulfate were present:

Sugar maltose. Mg.	Salicylic acid. Grams.	Potassium carbonate.		
		12 g.	14 g.	15 g.
100	4	151.7	153.8	154.9
100	5	151.0	157.1	155.0
100	6	149.1	154.6	154.8
100	7	135.7	150.4	154.9

These and other determinations showed that 15 grams of potassium carbonate, 5 grams of salicylic acid, and two grams of copper sulfate in a total volume of 140 cc. give satisfactory conditions for the determination of maltose. The least volume of water which will conveniently dissolve the copper sulfate is 15 cc. and the least volume for the 15 grams of potassium carbonate is 25 cc. While it would be possible to make one solution of the three compounds, it was found that both copper and potassium salicylate are so slightly soluble that the volume of such a solution would be too great to give satisfactory results. The potassium carbonate and copper sulfate are therefore dissolved in water and added separately to the sugar solution while the salicylic acid is added in the dry condition.

In regard to the method of heating, the following experiments were carried out to determine whether or not any cuprous oxide was lost during the heating by surface oxidation:

150 mg. of Kahlbaum's cuprous oxide were placed in each of four flasks, 15 grams of potassium carbonate, 2 grams of copper sulfate and 5 grams of salicylic acid were added in a volume of 140 cc.

The weight of copper in 150 mg. of the cuprous oxide used was found to be 125.9 mg. The weights of copper recovered from the four flasks after the treatment indicated below were as follows:

1. Solution boiled over free flame under conditions of Munson and Walker, 124.5 mg. copper recovered.

2. Solution heated 20 minutes in bath of boiling water, 126.3 mg. copper recovered.

3. Solution heated 20 minutes in bath of boiling water with surface covered with toluene, 125.7 mg. copper recovered.

4. Solution heated 20 minutes in bath of boiling water, air above solution being displaced with illuminating gas, 126.3 mg. copper recovered.

These results showed that there is no appreciable loss of cuprous oxide due to surface oxidation when the heating is continued for 20 minutes in the boiling water.

The two methods of heating which have been used for the reduction of copper with sugar are by heating over a free flame or in a bath of boiling water. In choosing between these two methods, ease of operation, time required, and accuracy of the results obtained were the factors considered.

The following results bear on this point:

The conditions of the solutions in each of the following sets were those found to be the optimum conditions for determining the reducing power of sugars. Volume 140 cc., potassium carbonate 15 grams, salicylic acid 5 grams, copper sulfate 2 grams.

#### HEATING IN BATH OF BOILING WATER.

Time. Min.	Glucose, 50 mg. Mg. Cu.	Invert sugar, 50 mg. Mg. Cu.	Lactose, 100 mg. Mg. Cu.	Maltose, 100 mg. Mg. Cu.
10	132.7	142.4	123.6	124.8
15	145.1	152.5	144.5	144.8
20	149.8	154.7	154.4	153.1
25	150.1	157.4	161.1	158.5
30	155.0	162.9	165.4	163.3
40	157.1	164.9	171.6	167.9
50	159.9	166.3	176.2	172.2

#### HEATING OVER FLAME.

Total time of heating. Min.	Time of boiling. Min.	Glucose, 50 mg. Mg. Cu.	Lactose, 100 mg. Mg. Cu.
6	2	112.2	103.0
8	4	134.1	132.8
9	5	137.1	139.0
12	8	143.8	154.1
14	10	143.0	161.1
16	12	148.3	163.6
18	14	150.4	167.3
20	16	153.4	169.4

The figures in the first line of the last table above give the weights of copper reduced under the conditions of Munson and Walker. It is apparent that the reduction under these conditions is far from complete and that the speed of reaction at this point is too great to allow of an accurate determination of reducing power. The reason for the incomplete reduction after two minutes' boiling is undoubtedly due to the slower reaction of the carbonate-salicylate solution than of the hydroxide-tartrate solution.

When the determinations of reducing power are done in sets of four or more time is saved per determination by making the time of heating as short as possible. However, it is evident that at least twelve minutes of boiling over a flame are required. When the time of heating is limited to twelve minutes it is impossible to filter one set while the following set is being heated, but if the time of heating be extended it is possible to give one's entire attention to filtering the reduced copper and hence there is no actual loss of time per determination.

To boil a solution over a flame for twelve to sixteen minutes requires more or less attention to maintain uniform conditions, but it is an easy

matter to duplicate conditions of heating in a bath of boiling water and no attention is required during the heating. Furthermore, it was found that the results obtained by heating in boiling water are more accurate than those obtained by boiling the solution. After twenty minutes' heating in boiling water the reaction is nearly complete for glucose and invert sugar and there is only a slow rate of reduction for lactose and maltose. As heating beyond 20 minutes would mean a needless expenditure of time, it was decided to limit the reduction for all of the sugars to that obtained during 20 minutes' heating in boiling water.

Although the salicylic acid is employed in the alkaline solution and must, therefore, exist as potassium salicylate, it was found impossible to replace the acid with sodium salicylate and obtain the same reducing power for maltose.

The following results show the difference between the free acid and the sodium salt:

Grams.	Sodium salicylate.	Salicylic acid.
3	117.4	141.3
4	126.0	143.1
5	131.9	148.4
6	136.2	148.7

The volume was 125 cc., 2 g. copper sulfate, 11.5 g. potassium carbonate, and 100 mg. of maltose being present. The figures are milligrams of copper reduced.

When the salicylic acid and sodium salicylate were kept constant and the potassium carbonate varied, the following results were obtained:

Potassium carbonate. Grams.	Sodium salicylate, 3 grams. Mg. Cu.	Salicylic acid, 3 grams. Mg. Cu.
15	123.1	145.6
18	125.7	146.8
21	130.4	148.1
24	135.8	151.9

The volume was 125 cc., 2 g. of copper sulfate and 100 mg. maltose were used.

Another series where more salicylic acid and sodium salicylate were used gave the following results:

Potassium carbonate. Grams.	Sodium salicylate. Grams.	Copper.
15	6	138.3
18	6	142.1
21	6	144.3
24	6	147.8
Potassium carbonate. Grams.	Salicylic acid. Grams.	Copper.
12	6	148.1
15	6	152.6
18	6	157.0
21	6	158.3

Although it would seem to make no difference when the sugar was added to the solution of potassium-copper salicylate, experiment showed that it is necessary to add the copper to the solution and not *vice versa*. A series where 15 g. of potassium carbonate and 5 g. of salicylic acid were used and 100 mg. of maltose were added to this solution gave 152.6, 146.2, 149.1, and 156.2 mg. of copper reduced. Under identical conditions, but where the copper solution was added to the sugar, the following weights of copper were obtained: 154.9, 155.0, 154.8, 154.9.

The effect of mixing the sugar and alkaline copper solutions and allowing to stand in the cold is shown by the following results: To four flasks, each containing 100 cc. of water, 100 mg. of lactose and 25 cc. of copper sulfate (2 grams) were added. At intervals of five minutes, 12 grams of potassium carbonate and 3 grams of salicylic acid were added to the four flasks in succession. The flask to which the potassium carbonate and salicylic acid was first added would have stood 15 minutes before the mixture had been added to the fourth flask. As soon as the fourth flask was ready they were all placed in the boiling water and allowed to remain 20 minutes; the following weights of copper were obtained:

Solution stood in the cold 15 min., 149.8.

Solution stood in the cold 10 min., 147.4.

Solution stood in the cold 5 min., 146.3.

Solution stood in the cold 0 min., 146.5.

These results show that the sugar can stand in the alkaline copper solution for 5-6 minutes in the cold without any appreciable change, but that a slight reduction will occur if they are allowed to stand 10-15 minutes. In practice 2-3 minutes is all that is needed to dissolve the salicylic acid and prepare the solutions for the boiling water.

The temperature at which the solutions are added to the boiling water is without appreciable influence between 18° and 50°. Four solutions, each containing 12 g. potassium carbonate, 3 g. salicylic acid, 2 g. copper sulfate, and 100 mg. lactose, when placed in the boiling water at the indicated temperature gave the following weights of copper reduced:

18°, 150.4 mg. copper; 30°, 150.7 mg. copper; 40°, 150.7 mg. copper; 50°, 151.5 mg. copper.

It is essential to have the boiling water heated with a flame large enough to cause the water to begin boiling within 1.5-2 minutes after the addition of the flasks containing the sugar-copper solutions.

Two flasks, containing 12 g. of potassium carbonate, 3 g. of salicylic acid, 2 g. of copper sulfate, and 120 mg. of lactose in 125 cc., were placed in the boiling water with a flame under the bath, which caused the water to boil within 1.5-2 minutes after the flasks were placed in the bath. The copper reduced at the end of 20 minutes was 175.5 and 176.3 mg.



Two other flasks containing identical solutions were placed in the boiling water with a flame under the bath which caused the water to boil in 5-6 minutes after the addition of the flasks. After 20 minutes from the time the flasks were placed in the water the copper reduced was 167.9 and 171.8 mg., showing lower and irregular results.

In order to show the variations caused by slight differences in the weights of potassium carbonate and salicylic acid added, the following series of determinations were made under identical conditions of time and volume. The volume was 140 cc. and time 20 minutes:

Potassium carbonate. Grams.	Salicylic acid. Grams.	Glucose. 50 mg.	Invert sugar. 50 mg.	Lactose. 100 mg.	Maltose. 100 mg.
15	4	148.9	155.2	160.0	154.9
15	5	149.2	155.7	157.2	155.0
15	6	...	156.7	154.9	154.9
15	7	150.0	158.0	151.4	154.9
13	5	151.4	...	152.9	153.0
14	5	151.4	158.3	154.5	154.6
15	5	149.4	155.7	157.2	155.0
16	5	149.4	156.1	156.9	156.3
17	5	147.3	155.5	160.1	158.4

These results show that glucose, maltose, and invert sugar vary but slightly for differences in amounts of potassium carbonate and salicylic acid present and that lactose is more sensitive in this respect.

In practical determinations of sugar, the variations in the weights of potassium carbonate and salicylic acid can be controlled within  $\pm 0.2$  g. without taking any special precautions and it is apparent that such a variation causes no appreciable change in the reducing power of any of the sugars.

#### Determination of the Copper Reduced.

In a recent number of THIS JOURNAL the writer described a method for the determination of copper by means of the iodide method. The method, as there described, was devised primarily for the determination of copper obtained by the reduction with sugar. It differs from the original iodide method in that the solutions are prepared for titration in the cold, thus overcoming the delay caused by boiling the solution or evaporating to dryness.

During the course of this investigation several hundred determinations of copper have been made by this modification of the iodide method, and these results show that, if the conditions prescribed are followed, the determination of copper can be made by this method with great accuracy. Irrespective of the way the reduced copper is determined it has to be removed from its filter, and the most convenient way to do this is to dissolve it in nitric acid. The iodide method allows of the accurate

determination of the copper *thus dissolved*, hence doing away with drying and weighing, which is time-consuming and laborious.

The cuprous oxide reduced by the sugar is filtered on a glass funnel such as is usually employed to hold a Gooch crucible. The filter is made by placing a perforated porcelain disk in the bottom of the funnel and making an asbestos felt 6 to 8 mm. in thickness. A porcelain disk should be used to hold the asbestos, as glass wool retains traces of the alkaline copper solution. After the solution has been filtered with suction and washed with hot water, the funnel and rubber stopper are removed from the suction flask, washed free from any copper solution which may adhere to the outside, and placed on a 350 cc. suction flask. If the stopper does not fit, the top of the flask is ground smooth on a carborundum hone so that when suction is applied it will hold the stopper down tightly over the mouth. Before the suction is applied to the flask the cuprous oxide is dissolved in not less than 10 cc. of *hot* nitric acid (1 part of acid to 3 of water). It is imperative to have the nitric acid hot and it should be contained in a wash bottle which delivers a small stream. The flask in which the reduction takes place and the sides of the funnel are washed with the hot acid and then the asbestos is stirred up by the jet from the wash bottle. It is best not to have a porcelain disk on the surface of the asbestos. The hot nitric acid will dissolve only a trace of nitrous oxide, but if cold nitric acid is used large amounts of nitrous acid will be dissolved, which will prevent the accurate determination of the copper. When all of the cuprous oxide has been dissolved the suction is applied and the funnel is washed with several additions of small amounts of water—not more than 10–12 cc. at a time. The wash water is sucked through each time before more is added. All of the copper, when washed in this way, can be removed with 40–50 cc. of wash water. The copper in solution may now be determined as described in the method.<sup>1</sup>

#### Preparation and Analysis of the Sugars Used.

The four sugars used to determine the relation between sugar and copper given in the table below were prepared and analyzed as follows:

The glucose of highest purity, furnished by Merck & Co., when analyzed for moisture and rotating power showed 0.16% of moisture and a specific rotating power of 52.68°. As the specific rotating power was in accordance with that given by Tollens for pure glucose no further purification was considered necessary.

The sucrose was prepared from Kahlbaum's C. P. saccharose by the method outlined by the International Commission for the Unification of Sugar Analysis. The sample thus prepared contained 0.13 per cent. of moisture and showed a specific rotating power of 66.5°.<sup>2</sup>

<sup>1</sup> THIS JOURNAL, 33, 1947.

<sup>2</sup> The sucrose was inverted essentially by the method used by Munson and Walker

The lactose was prepared from Kahlbaum's crystallized lactose by dissolving in boiling water, filtering, and allowing the lactose to crystallize for seven days from this solution. These crystals were dried in a vacuum over sulfuric acid for three days, ground into a powder, and again dried in vacuum over sulfuric acid. The determination of moisture showed 5.62% of water. One molecule of water, corresponding to the formula  $C_{12}H_{22}O_{11} \cdot H_2O$ , requires 5.0% of water, therefore the sample thus prepared contained 0.62% excess water calculated as lactose hydrate. Its specific rotating power calculated as  $C_{12}H_{22}O_{11} \cdot H_2O$  was  $52.51^\circ$ .

The maltose was prepared by letting barley diastase act on soluble starch as described by Baker and Day<sup>1</sup> and Baker.<sup>2</sup> The maltose thus prepared was dried in an electric oven at  $70-80^\circ$  for 15-18 hours, and was then ground and passed through a 60-mesh sieve. The moisture determination showed 5.33% of water. As maltose hydrate,  $C_{12}H_{22}O_{11} \cdot H_2O$ , requires 5% of water there was present 0.33% excess water. The specific rotating power was  $137.3^\circ$ .

The rotating power of all preparations was determined with sodium light, using a 4 decimeter tube in a Schmidt and Haensch polariscope at  $20^\circ$ . The solutions were 10% of sugar, except for maltose, which was 5%. The solutions, except for sucrose, were allowed to stand 24 hours at room temperature to destroy the multirotation.

The water content of all preparations were determined as follows: Two grams of the sample in a small weighing bottle was placed in the bottom of a 4-inch desiccator which had a suction outlet in the lid. Around the weighing bottle was placed a wire gauze collar about 1.75 inches in diameter. This wire gauze supported a Petri dish of 3 inches diameter which contained phosphorus pentoxide. A second Petri dish of almost the same diameter as the desiccator was supported above the first Petri dish by three wire supports at a distance of 0.5 inch above the lower Petri dish. This top Petri dish acted as a cover and prevented the phosphorus pentoxide from dusting when the suction was released. After placing the cover on the desiccator it was placed inside an electric oven and connected with stout suction hose to a Gaede pump. The temperature of the oven was kept for lactose at  $130^\circ$ , maltose  $110^\circ$ , sucrose and glucose  $100^\circ$ , for 4-5 hours. The suction was maintained during the entire time of heating. At intervals of one hour the weighing bottle was taken out of the desiccator and weighed. When heating for one hour produced a change of less than 0.5 mg. the heating was discontinued. Duplicate determinations were made in all cases.

by heating for 30 minutes in boiling water with  $N/10$  HCl, using 20 cc. for every 100 cc. final volume. The solution was barely neutralized with  $N/10$  sodium hydroxide, cooled and filled at  $20^\circ$  to the mark of graduation.

<sup>1</sup> *Analyst*, 33, 393 (1908).

<sup>2</sup> *J. Chem. Soc.*, 1902, 1177.

### Determination of Relation between Sugar and Copper.

For maltose and lactose the sugar was weighed out in such quantities as to make 5 mg. per cc. of solution, allowance being made for the presence of the water; the weights of sugar were calculated as maltose and lactose hydrate,  $C_{12}H_{22}O_{11} \cdot H_2O$ .

For invert sugar and glucose, solutions were made which contained 2.5 mg. of sugar calculated as  $C_6H_{12}O_6$ .

The temperature of graduation of both flasks and burets was  $20^\circ$ .

### The Reducing Power of the Sugars

was determined as follows: The varying weights of sugar as indicated below were measured into 200 cc. Erlenmeyer flasks and the volume in each case made up to 100 cc. with distilled water. 5 grams of salicylic acid were now added to each of four flasks containing the sugar to be determined. Fifteen cc.<sup>1</sup> of copper sulfate solution and then 25 cc. potassium carbonate<sup>2</sup> solution were added to each of the flasks without any agitation of the solution. It was found necessary to observe this order for the addition of the reagents. The flasks were then shaken with a rotary motion. The precipitate of copper carbonate dissolved, forming a dark green solution. As soon as the salicylic acid dissolved the four flasks were put in a holder and placed in a bath of boiling water.<sup>3</sup> The

<sup>1</sup> The copper sulfate solution is prepared by dissolving 133.33 grams of  $CuSO_4 \cdot 5H_2O$  per liter of water; 15 cc. of such a solution contains 2 grams of copper sulfate.

<sup>2</sup> The potassium carbonate solution contains 600 g. of anhydrous potassium carbonate per liter. 25 cc. of this solution contains 15 g. of potassium carbonate. As potassium carbonate is hygroscopic it is necessary to drive off all water before making up the solution. This is done by heating the carbonate for 3-4 hours at a temperature from  $190^\circ$  to  $200^\circ$ . A sample of the salt thus treated when heated in a small test tube should not give any evidence of liberation of water. The carbonate solution should be made up in a liter or other size graduated flask and should be diluted almost to the proper volume well mixed, and then adjusted to the mark of graduation.

<sup>3</sup> The water bath used during this investigation was one 10 inches in diameter and  $6\frac{1}{2}$  inches deep. It contained 4 liters of water. The level of the water could be maintained constant by using the ordinary constant water level bath, or by means of a syphon acting between the water bath and a reservoir of water of constant level. The syphon for constant use must have a T or Y tube inserted at its highest point. A 20 cc. pipet closed at one end with rubber hose and pinchcock is connected with the T-tube. To start the syphon both ends are placed under water and (the pinchcock being open) the air is withdrawn from the pipet. When the pipet is full of water the pinchcock is closed. Any bubbles of air liberated from the water in the tube of the syphon will rise to the highest point of the tube and will there be caught in the pipet, displacing the water. Should the pipet become filled with air the syphon may be re-established by again withdrawing the air through the rubber tube and pinchcock. The end of the syphon which is in the water bath must have a small opening (about  $\frac{1}{8}$  in.). If a large opening is used the water when boiling may syphon out of the bath. The diameter of the rest of the syphon may be of any sized tubing.

The flame under the water bath was supplied by one large-sized Fletcher and two Bunsen burners. The water should be boiling vigorously when the flasks are placed

flame under the bath must be of such size that boiling begins within 1.5-2 minutes after addition of the flasks. At the end of twenty minutes from the time the flasks were placed in the bath the solutions were filtered and the copper determined as described above.

TABLE I.

Weight of sugar. Mg.	Glucose.		Invert sugar.		Weight of sugar. Mg.	Lactose.		Maltose.	
10	30.4	30.2	30.9	30.7	20	31.9	32.1	31.0	30.3
20	60.9	61.8	63.2	62.4	40	64.0	62.5	61.0	61.3
30	90.2	90.6	94.4	94.7	60	95.5	95.2	93.4	92.3
40	120.0	120.1	126.5	126.8	80	124.5	125.0	123.6	123.6
50	148.1	149.9	157.5	156.7	100	157.7	157.7	154.6	154.6
60	176.8	179.2	188.2	186.4	120	287.4	286.3	187.9	187.2
70	206.1	207.4	{217.5	219.7}	140	218.5	217.0	219.2	218.5
			{217.5	215.6}					
80	233.8	233.8	{245.3	245.3}					
			{245.6	*248.0}	160	246.3	246.7	250.6	250.4
90	261.3	263.3	276.1	276.1	180	279.0	278.4	278.6	280.1
100	288.0	289.2	302.6	302.6	200	306.8	305.9	{309.6	*313.6
								{310.9	*312.4
110	316.2	314.7	330.8	330.7	220	338.7	337.1	342.2	342.2
120	343.1	341.5	359.4	358.6	240	{364.6	*363.4	373.3	372.9
						{364.9}			
130	367.7	367.7	385.5	386.4	260	397.3	397.1	403.2	401.6
140	391.2	393.0	410.9	410.9	280	424.9	427.0	431.4	{431.4
									*429.4
150	418.5	418.5	438.1	438.1	300	455.6	454.3	{459.4	458.1
								*456.2	*454.3
160	443.3	442.1	462.1	460.9	...	...	...	...	...

in the water and all of the flames should be burning. As soon as the water has begun to boil after the addition of the flasks (which should be in less than two minutes) the two bunsen burners are turned out as the Fletcher burner is sufficient to maintain the boiling.

In the practical application of this method for the determination of the reducing sugars, it is most convenient to make a set of 4 determinations at once. A rack is made to hold the 4 flasks as follows: a brass rod  $\frac{1}{4}$  inch in diameter and 14 inches long is erected at the center point and perpendicular to a brass disk 8 inches in diameter and  $\frac{3}{32}$  inches thick. This disk forms the bottom upon which the flasks are placed. Another disk of the same size as the bottom one, but with a hole  $\frac{5}{16}$  inch in diameter, slides up and down the rod. Four holes  $1\frac{1}{2}$  inches in diameter are drilled through this second disk, the centers of the holes being arranged symmetrically  $2\frac{1}{4}$  inches from the center of the disk. This top disk being lifted up the four flasks are placed on the bottom of the holder. When the top disk is lowered the flasks will pass through the four holes, and as the opening is only sufficiently large to allow the neck of the flask to pass through, the flasks are held secure. A hook at the end of the rod is used to hang the entire holder from a support above the bath at such a height that the rack sinks in the water up to the level of the top disk. In the bottom disk a number of  $\frac{1}{2}$ -inch holes are drilled so that the rack may be placed in and removed from the water with ease. When the flasks are removed from the rack the solutions are filtered immediately on four suction flasks.

TABLE II.

Glucose.			Invert sugar.			Lactose.			Maltose.		
Found. Mg.	Calculated. Mg.	Error. Mg.	Found. Mg.	Calculated. Mg.	Error. Mg.	Found. Mg.	Calculated. Mg.	Error. Mg.	Found. Mg.	Calculated. Mg.	Error. Mg.
30.3	30.6	0.3	30.8	31.43	0.63	32.0	32.07	0.07	30.6	30.2	-0.40
61.4	60.9	-0.5	62.8	63.55	0.75	63.3	63.38	0.08	61.2	61.96	0.76
90.4	90.8	0.4	94.6	95.24	0.64	95.4	94.52	-0.88	92.9	93.57	0.67
120.1	120.3	0.2	126.7	126.44	-0.26	124.8	125.48	0.68	123.6	125.02	1.42
149.0	149.4	0.4	157.1	157.15	0.05	157.7	156.28	-1.42	154.6	156.32	1.72
178.0	178.1	0.1	187.3	187.37	0.07	186.9	186.90	0	187.6	187.47	-0.13
206.8	206.4	-0.4	217.7	217.10	-0.60	217.8	217.34	-0.46	218.9	218.47	-0.43
233.8	234.3	0.5	245.5	246.33	0.83	246.5	247.60	1.10	250.5	249.32	-1.18
262.3	261.7	-0.6	276.1	275.07	-1.03	278.7	277.70	-1.00	279.4	280.04	0.64
288.6	288.8	0.2	302.6	303.33	0.73	306.4	307.63	1.23	310.5	310.59	0.29
315.5	315.5	0	330.8	331.09	0.29	337.9	337.38	-0.52	342.2	341.00	-1.20
342.3	341.7	-0.6	359.0	358.35	-0.65	364.8	366.95	2.15	373.1	371.25	-1.85
367.7	367.6	-0.1	386.0	385.14	-0.86	397.2	396.35	-0.85	402.4	401.35	-1.05
392.1	393.0	0.9	410.9	411.43	0.53	426.0	425.58	-0.42	431.4	431.31	-0.09
418.5	418.1	-0.4	438.1	437.23	-0.87	455.0	454.64	-0.36	458.7	461.12	2.42
442.7	442.7	0	461.5	462.55	1.05	...	....	..	...	....	..

TABLE III.

Method.	Dextrose.		Invert sugar.		Lactose.		Maltose.	
	50 mg.	100 mg.	50 mg.	100 mg.	100 mg.	200 mg.	100 mg.	200 mg.
Defren.....	89.8	178.1	...	...	125.7	248.4	109.5	216.5
Munson and Walker.....	102.2	198.4	98.5	192.3	132.7	261.6	108.7	215.4
Allihn.....	98.2	195.0	...	...	...	...	...	...
Lehmann.....	...	...	101.4	190.0	...	...	...	...
Meissl.....	...	...	96.0	189.2	...	...	...	...
Soxhlet.....	...	...	...	...	138.3	269.6	...	...
Wein.....	...	...	...	...	...	...	115.1	226.8
Ost.....	165.6	294.3	170.0	298.0	...	...	167.5	331.8
Kjeldahl.....	107.2	197.7	101.7	190.2	...	...	...	...
Brown, Morris and Millar.....	103.0	202.7	97.5	194.1	...	...	...	...
This method.....	149.3	288.7	157.0	303.3	156.3	307.6	156.3	310.6

Using this method of procedure the results in Table I were obtained for the indicated weights of sugar. All of the results which were obtained are given, but the ones which were not used in the calculation of the curve are starred:

From these results the following equations showing the relation between sugar and copper were deduced by means of the method of least squares.<sup>1</sup>

In the equations X refers to weights of copper reduced, Y to weights of sugar reacting:

$$\begin{array}{l} \text{Glucose} \dots\dots\dots X = -0.17 + 3.0923 Y - 0.002026 Y^2 \\ \text{Invert sugar} \dots\dots\dots X = -1.30 + 3.2918 Y - 0.002455 Y^2 \\ \text{Lactose} \dots\dots\dots X = 0.59 + 1.5786 Y - 0.000217 Y^2 \\ \text{Maltose} \dots\dots\dots X = -1.69 + 1.5988 Y - 0.000187 Y^2 \end{array}$$

The weights of sugar given in Table IV were substituted in the proper equation and the corresponding values of X were found. The differences between the values thus calculated and actually found is given in Table II.

By means of the equation the copper equivalent to weights of maltose and lactose from 20 to 300 mg. were calculated for every 4 mg. of sugar. With glucose and invert sugar the copper equivalent for every two mg. of sugar from 10 to 160 was found. This gave a series of points which differed by about six mg. of copper. These figures were changed to integral weights of copper and hence decimal weights of sugar. The figures lying between each six mg. of copper were interpolated, the interpolations being carried to the second decimal place. The complete table giving the relation between the four reducing sugars and copper for every mg. of copper from 30 to 450 mg. is given in Table IV.

A comparison of the reducing power of the sugars obtained by this method with that obtained with other methods in use is given in Table III.

TABLE IV.

Copper. Mg.	Cuprous oxide. Mg.	Cupric oxide Mg.	Glucose. Mg. C <sub>6</sub> H <sub>12</sub> O <sub>6</sub> .	Invert sugar. Mg. C <sub>6</sub> H <sub>12</sub> O <sub>6</sub> .	Lactose. Mg. C <sub>12</sub> H <sub>22</sub> O <sub>11</sub> .H <sub>2</sub> O.	Maltose. Mg. C <sub>12</sub> H <sub>22</sub> O <sub>11</sub> .H <sub>2</sub> O.
30	33.8	37.6	9.8	9.6	19.3	19.9
31	34.9	38.8	10.2	9.9	20.0	20.5
32	36.0	40.1	10.5	10.2	20.6	21.1
33	37.2	41.3	10.8	10.5	21.2	21.7
34	38.3	42.6	11.1	10.8	21.8	22.4
35	39.4	43.8	11.5	11.1	22.5	23.0
36	40.5	45.1	11.8	11.4	23.1	23.6
37	41.7	46.3	12.1	11.8	23.7	24.3
38	42.8	47.6	12.4	12.1	24.4	24.9
39	43.9	48.8	12.8	12.4	25.0	25.5
40	45.0	50.1	13.1	12.7	25.6	26.1

<sup>1</sup> A good example of the use of this method is given in Allihn's original article, *J. prakt. Chem.*, 22, 46 (1880).



TABLE IV (Continued).

Copper. Mg.	Cuprous oxide. Mg.	Cupric oxide. Mg.	Glucose. Mg. $C_6H_{12}O_6$ .	Invert sugar. Mg. $C_6H_{12}O_6$ .	Lactose. Mg. $C_{12}H_{22}O_{11} \cdot H_2O$ .	Maltose. Mg. $C_{12}H_{22}O_{11} \cdot H_2O$ .
41	46.2	51.3	13.4	13.0	26.3	26.8
42	47.3	52.6	13.8	13.3	26.9	27.4
43	48.4	53.8	14.1	13.6	27.5	28.0
44	49.5	55.1	14.4	13.9	28.1	28.7
45	50.7	56.3	14.8	14.2	28.8	29.3
46	51.8	57.6	15.1	14.5	29.4	29.9
47	52.9	58.8	15.4	14.8	30.0	30.6
48	54.0	60.1	15.7	15.2	30.6	31.2
49	55.2	61.3	16.1	15.5	31.3	31.8
50	56.3	62.6	16.4	15.8	31.9	32.4
51	57.4	63.8	16.7	16.1	32.5	33.1
52	58.5	65.1	17.1	16.4	33.2	33.7
53	59.7	66.3	17.4	16.7	33.8	34.3
54	60.8	67.6	17.7	17.0	34.4	35.0
55	61.9	68.8	18.1	17.3	35.0	35.6
56	63.0	70.1	18.4	17.6	35.7	36.2
57	64.2	71.3	18.7	17.9	36.3	36.9
58	65.3	72.6	19.1	18.3	36.9	37.5
59	66.4	73.9	19.4	18.6	37.6	38.1
60	67.6	75.1	19.7	18.9	38.2	38.8
61	68.7	76.4	20.0	19.2	38.8	39.4
62	69.8	77.6	20.4	19.5	39.4	40.0
63	70.9	78.9	20.7	19.8	40.1	40.7
64	72.1	80.1	21.0	20.1	40.7	41.3
65	73.2	81.4	21.4	20.5	41.3	41.9
66	74.3	82.6	21.7	20.8	41.9	42.5
67	75.4	83.9	22.0	21.1	42.6	43.2
68	76.6	85.1	22.4	21.4	43.2	43.8
69	77.7	86.4	22.7	21.7	43.8	44.4
70	78.8	87.6	23.0	22.0	44.4	45.1
71	79.9	88.9	23.4	22.3	45.1	45.7
72	81.1	90.1	23.7	22.7	45.7	46.3
73	82.2	91.4	24.0	23.0	46.3	47.0
74	83.3	92.6	24.4	23.3	46.9	47.6
75	84.4	93.9	24.7	23.6	47.5	48.2
76	85.6	95.1	25.0	23.9	48.1	48.9
77	86.7	96.4	25.4	24.2	48.8	49.5
78	87.8	97.6	25.7	24.5	49.4	50.1
79	88.9	98.9	26.0	24.9	50.0	50.8
80	90.1	100.1	26.4	25.2	50.7	51.4
81	91.2	101.4	26.7	25.5	51.3	52.0
82	92.3	102.6	27.1	25.8	51.9	52.7
83	93.4	103.9	27.4	26.1	52.6	53.3
84	94.6	105.1	27.7	26.4	53.2	53.9
85	95.7	106.4	28.1	26.8	53.9	54.6
86	96.8	107.6	28.4	27.1	54.5	55.2
87	97.9	108.9	28.7	27.4	55.1	55.8
88	99.1	110.1	29.1	27.7	55.8	56.5

TABLE IV (Continued).

Copper. Mg.	Cuprous oxide. Mg.	Cupric oxide. Mg.	Glucose. Mg. $C_6H_{12}O_6$ .	Invert sugar. Mg. $C_6H_{12}O_6$ .	Lactose. Mg. $C_{12}H_{22}O_{11} \cdot H_2O$ .	Maltose. Mg. $C_{12}H_{22}O_{11} \cdot H_2O$ .
89	100.2	111.4	29.4	28.0	56.4	57.1
90	101.3	112.7	29.7	28.3	57.1	57.7
91	102.4	113.9	30.1	28.7	57.7	58.3
92	103.6	115.2	30.4	29.0	58.4	58.9
93	104.7	116.4	30.8	29.3	59.0	59.6
94	105.8	117.7	31.1	29.6	59.7	60.2
95	106.9	118.9	31.4	29.9	60.3	60.9
96	108.1	120.2	31.8	30.2	60.9	61.5
97	109.2	121.4	32.1	30.6	61.6	62.2
98	110.3	122.7	32.4	30.9	62.2	62.8
99	111.5	123.9	32.8	31.2	62.8	63.4
100	112.6	125.2	33.1	31.5	63.5	64.1
101	113.7	126.4	33.5	31.8	64.2	64.7
102	114.8	127.7	33.8	32.2	64.8	65.3
103	116.0	128.9	34.1	32.5	65.4	66.0
104	117.1	130.2	34.5	32.8	66.1	66.6
105	118.2	131.5	34.8	33.1	66.8	67.2
106	119.3	132.7	35.2	33.4	67.4	67.8
107	120.5	134.0	35.5	33.8	68.0	68.5
108	121.6	135.2	35.9	34.1	68.7	69.1
109	122.7	136.5	36.2	34.4	69.3	69.8
110	123.8	137.7	36.6	34.7	70.0	70.4
111	125.0	139.0	36.9	35.0	70.6	71.1
112	126.1	140.2	37.2	35.4	71.3	71.7
113	127.2	141.5	37.5	35.7	71.9	72.3
114	128.3	142.7	37.9	36.0	72.6	73.0
115	129.4	144.0	38.2	36.3	73.2	73.6
116	130.6	145.2	38.5	36.7	73.8	74.2
117	131.7	146.5	38.9	37.0	74.5	74.9
118	132.8	147.7	39.2	37.3	75.1	75.5
119	134.0	149.0	39.6	37.6	75.8	76.2
120	135.1	150.2	39.9	37.9	76.4	76.8
121	136.2	151.5	40.3	38.3	77.1	77.4
122	137.4	152.7	40.6	38.6	77.7	78.1
123	138.5	154.0	40.9	38.9	78.4	78.7
124	139.6	155.2	41.3	39.2	79.0	79.4
125	140.7	156.5	41.6	39.5	79.7	80.0
126	141.9	157.7	42.0	39.9	80.3	80.6
127	143.0	159.0	42.3	40.2	81.0	81.3
128	144.1	160.2	42.6	40.5	81.6	81.9
129	145.2	161.5	43.0	40.8	82.3	82.5
130	146.4	162.7	43.3	41.2	82.9	83.2
131	147.5	164.0	43.7	41.5	83.6	83.8
132	148.6	165.2	44.0	41.8	84.2	84.5
133	149.7	166.5	44.4	42.1	84.9	85.2
134	150.9	167.7	44.7	42.5	85.5	85.8
135	152.0	169.0	45.1	42.8	86.2	86.5
136	153.1	170.2	45.4	43.1	86.8	87.1

TABLE IV (Continued).

Copper. Mg.	Cuprous oxide. Mg.	Cupric oxide. Mg.	Glucose. Mg. $C_6H_{12}O_6$ .	Invert sugar. Mg. $C_6H_{12}O_6$ .	Lactose. Mg. $C_{12}H_{22}O_{11} \cdot H_2O$ .	Maltose. Mg. $C_{12}H_{22}O_{11} \cdot H_2O$ .
137	154.2	171.5	45.7	43.4	87.4	87.7
138	155.4	172.7	46.1	43.8	88.1	88.3
139	156.5	174.0	46.4	44.1	88.7	88.9
140	157.6	175.2	46.8	44.4	89.4	89.6
141	158.7	176.5	47.1	44.7	90.0	90.2
142	159.9	177.7	47.5	45.1	90.7	90.8
143	161.0	179.0	47.8	45.4	91.3	91.5
144	162.1	180.2	48.1	45.7	92.0	92.1
145	163.2	181.5	48.5	46.0	92.6	92.8
146	164.4	182.7	48.8	46.4	93.3	93.4
147	165.5	184.0	49.2	46.7	93.9	94.0
148	166.6	185.2	49.5	47.0	94.6	94.7
149	167.7	186.5	49.9	47.4	95.3	95.3
150	168.9	187.8	50.2	47.7	95.9	96.0
151	170.0	189.0	50.6	48.0	96.6	96.6
152	171.1	190.3	50.9	48.3	97.2	97.2
153	172.3	191.5	51.3	48.7	97.9	97.9
154	173.4	192.8	51.6	49.0	98.5	98.5
155	174.5	194.0	52.0	49.3	99.2	99.2
156	175.7	195.3	52.3	49.6	99.8	99.8
157	176.8	196.5	52.7	50.0	100.5	100.4
158	177.9	197.8	53.0	50.3	101.1	101.1
159	179.0	199.0	53.4	50.6	101.8	101.7
160	180.1	200.3	53.7	50.9	102.4	102.4
161	181.3	201.5	54.0	51.3	103.1	103.0
162	182.4	202.8	54.4	51.6	103.7	103.6
163	183.5	204.0	54.7	51.9	104.4	104.3
164	184.6	205.3	55.1	52.3	105.0	104.9
165	185.8	206.5	55.4	52.6	105.7	105.6
166	186.9	207.8	55.8	52.9	106.3	106.2
167	188.0	209.0	56.1	53.3	107.0	106.8
168	189.1	210.3	56.5	53.6	107.6	107.5
169	190.3	211.5	56.8	53.9	108.3	108.1
170	191.4	212.8	57.2	54.2	108.9	108.8
171	192.5	214.0	57.5	54.6	109.6	109.4
172	193.6	215.3	57.9	54.9	110.2	110.1
173	194.8	216.5	58.2	55.2	110.9	110.7
174	195.9	217.8	58.6	55.6	111.6	111.3
175	197.0	219.0	58.9	55.9	112.2	112.0
176	198.1	220.3	59.3	56.2	112.9	112.6
177	199.3	221.5	59.6	56.6	113.5	113.3
178	200.4	222.8	60.0	56.9	114.2	113.9
179	201.5	224.0	60.3	57.2	114.9	114.5
180	202.6	225.3	60.7	57.6	115.5	115.2
181	203.8	226.5	61.0	57.9	116.1	115.8
182	204.9	227.8	61.4	58.2	116.8	116.5
183	206.0	229.0	61.7	58.6	117.4	117.1
184	207.1	230.3	62.1	58.9	118.1	117.8

TABLE IV (Continued).

Copper. Mg.	Cuprous oxide. Mg.	Cupric oxide. Mg.	Glucose. Mg. $C_6H_{12}O_6$ .	Invert sugar. Mg. $C_6H_{12}O_6$ .	Lactose. Mg. $C_{12}H_{22}O_{11} \cdot H_2O$ .	Maltose. Mg. $C_{12}H_{22}O_{11} \cdot H_2O$ .
185	208.3	231.5	62.4	59.2	118.8	118.4
186	209.4	232.7	62.8	59.6	119.4	119.
187	210.5	234.0	63.1	59.9	120.1	119.7
188	211.7	235.3	63.5	60.2	120.7	120.3
189	212.8	236.5	63.9	60.6	121.4	121.0
190	213.9	237.8	64.2	60.9	122.0	121.6
191	215.0	239.0	64.6	61.2	122.7	122.3
192	216.2	240.3	64.9	61.6	123.3	122.9
193	217.3	241.5	65.3	61.9	124.0	123.6
194	218.4	242.8	65.6	62.2	124.7	124.2
195	219.5	244.0	66.0	62.6	125.3	124.8
196	220.7	245.3	66.3	62.9	126.0	125.5
197	221.8	246.5	66.7	63.2	126.6	126.1
198	222.9	247.8	67.0	63.6	127.3	126.8
199	224.0	249.0	67.4	63.9	127.9	127.4
200	225.2	250.3	67.8	64.2	128.6	128.1
201	226.3	251.5	68.1	64.6	129.2	128.7
202	227.4	252.8	68.5	64.9	129.9	129.4
203	228.5	254.0	68.8	65.2	130.6	130.0
204	229.7	255.3	69.2	65.6	131.2	130.6
205	230.8	256.5	69.5	65.9	131.9	131.3
206	231.9	257.8	69.9	66.2	132.5	131.9
207	233.0	259.0	70.2	66.6	133.2	132.6
208	234.2	260.3	70.6	66.9	133.8	133.2
209	235.3	261.5	71.0	67.3	134.5	133.9
210	236.4	262.8	71.3	67.6	135.2	134.5
211	237.6	264.0	71.7	67.9	135.8	135.2
212	238.7	265.3	72.0	68.3	136.5	135.8
213	239.8	266.5	72.4	68.6	137.1	136.5
214	240.9	267.8	72.7	69.0	137.8	137.1
215	242.1	269.0	73.1	69.3	138.5	137.8
216	243.2	270.3	73.4	69.6	139.1	138.4
217	244.3	271.5	73.8	70.0	139.8	139.1
218	245.4	272.8	74.2	70.3	140.4	139.7
219	246.6	274.1	74.5	70.7	141.1	140.3
220	247.7	275.4	74.9	71.0	141.8	141.0
221	248.7	276.6	75.2	71.4	142.4	141.6
222	249.9	277.9	75.6	71.7	143.1	142.3
223	251.0	279.1	76.0	72.0	143.7	142.9
224	252.1	280.4	76.3	72.4	144.4	143.6
225	253.3	281.6	76.7	72.7	145.1	144.2
226	254.4	282.9	77.0	73.1	145.7	144.9
227	255.6	284.1	77.4	73.4	146.4	145.5
228	256.7	285.4	77.8	73.7	147.0	146.2
229	257.8	286.6	78.1	74.1	147.7	146.8
230	258.9	287.9	78.5	74.4	148.4	147.5
231	260.1	289.1	78.8	74.8	149.0	148.1
232	261.2	290.4	79.2	75.1	149.7	148.8

TABLE IV (Continued).

Copper. Mg.	Cuprous oxide. Mg.	Cupric oxide. Mg.	Glucose. Mg. $C_6H_{12}O_6$ .	Invert sugar. Mg. $C_6H_{12}O_6$ .	Lactose. Mg. $C_{12}H_{22}O_{11} \cdot H_2O$ .	Maltose. Mg. $C_{12}H_{22}O_{11} \cdot H_2O$ .
233	262.3	291.6	79.6	75.4	150.3	149.4
234	263.1	292.9	79.9	75.5	151.0	150.1
235	264.6	294.1	80.3	76.1	151.7	150.7
236	265.7	295.4	80.6	76.5	152.3	151.4
237	266.8	296.6	81.0	76.8	153.0	152.0
238	268.0	297.9	81.4	77.2	153.6	152.6
239	269.1	299.1	81.7	77.5	154.3	153.3
240	270.2	300.4	82.1	77.8	155.0	153.9
241	271.3	301.6	82.5	78.2	155.6	154.6
242	272.5	302.9	82.8	78.5	156.3	155.2
243	273.6	304.1	83.2	78.9	157.0	155.9
244	274.7	305.4	83.5	79.2	157.6	156.5
245	275.8	306.6	83.9	79.6	158.3	157.2
246	277.0	307.9	84.3	79.9	159.0	157.8
247	278.1	309.1	84.6	80.2	159.6	158.5
248	279.2	310.4	85.0	80.6	160.3	159.1
249	280.3	311.6	85.4	80.9	160.9	159.8
250	281.5	312.9	85.7	81.3	161.6	160.4
251	282.6	314.1	86.1	81.6	162.2	161.1
252	283.7	315.4	86.5	82.0	162.9	161.7
253	284.8	316.6	86.8	82.3	163.6	162.4
254	286.0	317.9	87.2	82.7	164.2	163.0
255	287.1	319.1	87.6	83.0	164.9	163.7
256	288.2	320.4	87.9	83.4	165.6	164.3
257	289.3	321.6	88.3	83.7	166.2	165.0
258	290.5	322.9	88.7	84.1	166.9	165.6
259	291.6	324.1	89.0	84.4	167.6	166.3
260	292.7	325.4	89.4	84.8	168.2	166.9
261	293.8	326.6	89.8	85.1	168.9	167.6
262	295.0	327.9	90.1	85.5	169.5	168.3
263	296.1	329.1	90.5	85.8	170.2	168.9
264	297.2	330.4	90.8	86.1	170.9	169.6
265	298.3	331.6	91.2	86.5	171.6	170.2
266	299.5	332.9	91.5	86.8	172.2	170.9
267	300.6	334.1	91.9	87.2	172.9	171.5
268	301.7	335.4	92.3	87.5	173.5	172.2
269	302.8	336.7	92.7	87.9	174.2	172.8
270	304.0	338.0	93.1	88.2	174.9	173.5
271	305.1	339.2	93.4	88.6	175.5	174.1
272	306.2	340.5	93.8	88.9	176.2	174.8
273	307.3	341.7	94.2	89.3	176.9	175.4
274	308.5	343.0	94.5	89.6	177.5	176.1
275	309.6	344.2	94.9	90.0	178.2	176.7
276	310.7	345.5	95.3	90.3	178.9	177.4
277	311.9	346.7	95.6	90.7	179.5	178.0
278	313.0	348.0	96.0	91.1	180.2	178.7
279	314.1	349.2	96.4	91.4	180.9	179.3
280	315.2	350.5	96.7	91.8	181.5	180.0

TABLE IV (Continued).

Copper. Mg.	Cuprous oxide. Mg.	Cupric oxide. Mg.	Glucose. Mg. $C_6H_{12}O_6$ .	Invert sugar. Mg. $C_6H_{12}O_6$ .	Lactose. Mg. $C_{12}H_{22}O_{11} \cdot H_2O$ .	Maltose. Mg. $C_{12}H_{22}O_{11} \cdot H_2O$ .
281	316.4	351.7	97.1	92.1	182.2	180.6
282	317.5	353.0	97.5	92.5	182.9	181.3
283	318.6	354.2	97.9	92.8	183.5	181.9
284	319.7	355.5	98.2	93.1	184.2	182.6
285	320.9	356.7	98.6	93.5	184.9	183.2
286	322.0	358.0	99.0	93.9	185.5	183.9
287	323.1	359.2	99.4	94.2	186.2	184.6
288	324.2	360.5	99.7	94.6	186.9	185.2
289	325.3	361.7	100.1	94.9	187.6	185.9
290	326.4	363.0	100.5	95.3	188.2	186.5
291	327.5	364.2	100.8	95.6	188.8	187.2
292	328.7	365.5	101.2	96.0	189.	187.8
293	329.9	366.7	101.6	96.3	190.1	188.5
294	331.0	368.0	101.9	96.7	190.8	189.1
295	332.1	369.2	102.3	97.1	191.4	189.8
296	333.3	370.5	102.7	97.4	192.1	190.4
297	334.4	371.7	103.1	97.8	192.8	191.1
298	335.5	373.0	103.4	98.1	193.5	191.8
299	336.6	374.2	103.8	98.5	194.2	192.4
300	337.8	375.5	104.2	98.9	194.9	193.1
301	338.9	376.7	104.6	99.2	195.6	193.7
302	340.0	378.0	105.0	99.6	196.2	194.4
303	341.1	379.2	105.3	99.9	196.9	195.0
304	342.3	380.5	105.7	100.3	197.6	195.7
305	343.4	381.7	106.1	100.6	198.3	196.3
306	344.5	383.0	106.4	101.0	198.9	197.0
307	345.6	384.2	106.8	101.3	199.6	197.6
308	346.8	385.5	107.2	101.7	200.3	198.3
309	347.9	386.7	107.6	102.1	200.9	199.0
310	349.0	388.0	107.9	102.4	201.6	199.6
311	350.1	389.2	108.3	102.8	202.3	200.3
312	351.3	390.5	108.7	103.1	202.9	200.9
313	352.4	391.7	109.1	103.5	203.6	201.6
314	353.5	393.0	109.5	103.8	204.3	202.2
315	354.6	394.2	109.8	104.2	204.9	202.9
316	355.8	395.5	110.2	104.6	205.5	203.6
317	356.9	396.7	110.6	104.9	206.2	204.2
318	358.0	398.0	111.0	105.3	206.8	204.9
319	359.1	399.2	111.4	105.6	207.5	205.5
320	360.3	400.5	111.7	106.0	208.2	206.2
321	361.4	401.7	112.1	106.4	208.9	206.8
322	362.5	403.0	112.5	106.7	209.6	207.5
323	363.7	404.2	112.9	107.1	210.3	208.2
324	364.8	405.5	113.2	107.5	211.0	208.8
325	365.9	406.7	113.6	107.8	211.7	209.5
326	367.0	408.0	114.0	108.2	212.3	210.1
327	368.2	409.2	114.4	108.5	213.0	210.8
328	369.3	410.5	114.8	108.9	213.6	211.5

TABLE IV (Continued).

Copper. Mg.	Cuprous oxide. Mg.	Cupric oxide. Mg.	Glucose. Mg. $C_6H_{12}O_6$ .	Invert sugar. Mg. $C_6H_{12}O_6$ .	Lactose. Mg. $C_{12}H_{22}O_{11} \cdot H_2O$ .	Maltose. Mg. $C_{12}H_{22}O_{11} \cdot H_2O$ .
329	370.4	411.8	115.1	109.3	214.3	212.1
330	371.5	413.1	115.5	109.6	214.9	212.8
331	372.7	414.3	115.9	110.0	215.6	213.4
332	373.8	415.6	116.3	110.3	216.3	214.1
333	374.9	416.8	116.7	110.7	217.0	214.7
334	376.0	418.1	117.0	111.1	217.7	215.4
335	377.2	419.3	117.4	111.5	218.4	216.1
336	378.3	420.6	117.8	111.8	219.1	216.7
337	379.4	421.9	118.2	112.2	219.8	217.4
338	380.5	423.1	118.6	112.5	220.4	218.0
339	381.7	424.4	119.0	112.9	221.1	218.7
340	382.8	425.6	119.4	113.3	221.7	219.3
341	383.9	426.9	119.8	113.6	222.4	220.0
342	385.0	428.1	120.1	114.0	223.0	220.7
343	386.2	429.4	120.5	114.4	223.7	221.3
344	387.3	430.6	120.9	114.7	224.4	222.0
345	388.4	431.9	121.3	115.1	225.1	222.6
346	389.6	433.1	121.7	115.5	225.8	223.3
347	390.7	434.4	122.1	115.8	226.5	224.0
348	391.8	435.6	122.4	116.2	227.2	224.6
349	392.9	436.9	122.8	116.6	227.9	225.3
350	394.0	438.1	123.2	117.0	228.5	225.9
351	395.2	439.4	123.6	117.3	229.2	226.6
352	396.3	440.6	124.0	117.7	229.9	227.3
353	397.4	441.9	124.3	118.1	230.6	227.9
354	398.6	443.1	124.7	118.4	231.2	228.6
355	399.7	444.4	125.1	118.8	231.9	229.2
356	400.8	445.7	125.5	119.2	232.6	229.9
357	401.9	446.1	125.9	119.5	233.2	230.6
358	403.1	448.1	126.3	119.9	233.9	231.2
359	404.2	449.4	126.7	120.3	234.6	231.9
360	405.3	450.6	127.1	120.7	235.3	232.6
361	406.4	451.9	127.5	121.0	236.0	233.2
362	407.6	453.1	127.9	121.4	236.6	233.9
363	408.7	454.4	128.2	121.8	237.3	234.5
364	409.8	455.6	128.6	122.1	238.0	235.2
365	410.9	456.9	129.0	122.5	238.7	235.9
366	412.1	458.1	129.4	122.9	239.4	236.5
367	413.2	459.4	129.8	123.2	240.1	237.2
368	414.3	460.6	130.2	123.6	240.7	237.8
369	415.4	461.9	130.6	124.0	241.4	238.5
370	416.6	463.1	131.0	124.3	242.1	239.2
371	417.7	464.4	131.4	124.7	242.8	239.8
372	418.8	465.6	131.8	125.1	243.5	240.5
373	420.0	466.9	132.1	125.5	244.1	241.2
374	421.1	468.1	132.5	125.8	244.8	241.8
375	422.2	469.4	132.9	126.2	245.5	242.5
376	423.3	470.6	133.3	126.6	246.2	243.1

TABLE IV (Continued).

Copper. Mg.	Cuprous oxide. Mg.	Cupric oxide. Mg.	Glucose. Mg. $C_6H_{12}O_6$ .	Invert sugar. Mg. $C_6H_{12}O_6$ .	Lactose. Mg. $C_{12}H_{22}O_{11} \cdot H_2O$ .	Maltose. Mg. $C_{12}H_{22}O_{11} \cdot H_2O$ .
377	424.5	471.9	133.7	127.0	246.8	243.8
378	425.6	473.0	134.1	127.4	247.5	244.5
379	426.7	474.3	134.5	127.7	248.2	245.1
380	427.8	475.6	134.9	128.1	248.9	245.8
381	429.0	476.8	135.3	128.5	249.6	246.5
382	430.1	478.1	135.7	128.8	250.2	247.1
383	431.2	479.3	136.1	129.2	250.9	247.8
384	432.3	480.6	136.5	129.6	251.6	248.5
385	433.5	481.8	136.9	130.0	252.3	249.1
386	434.6	483.1	137.3	130.3	253.0	249.8
387	435.7	484.3	137.7	130.7	253.6	250.4
388	436.8	485.6	138.0	131.1	254.3	251.1
389	438.0	486.9	138.4	131.5	255.0	251.8
390	439.1	488.2	138.8	131.9	255.7	252.4
391	440.2	489.4	139.2	132.2	256.4	253.1
392	441.3	490.7	139.6	132.6	257.0	253.8
393	442.4	491.9	140.0	133.0	257.7	254.4
394	443.6	493.2	140.4	133.4	258.4	255.1
395	444.7	494.4	140.8	133.8	259.1	255.8
396	445.9	495.7	141.2	134.2	259.8	256.4
397	447.0	496.9	141.6	134.5	260.5	257.1
398	448.1	498.2	142.0	134.9	261.1	257.8
399	449.2	499.5	142.4	135.3	261.8	258.4
400	450.3	500.7	142.8	135.7	262.5	259.1
401	451.5	502.0	143.2	136.1	263.2	259.8
402	452.6	503.2	143.6	136.4	263.9	260.4
403	453.7	504.5	144.0	136.8	264.5	261.1
404	454.8	505.7	144.4	137.2	265.2	261.8
405	456.0	507.0	144.8	137.6	265.9	262.4
406	457.1	508.2	145.2	137.9	266.6	263.1
407	458.2	509.5	145.6	138.3	267.3	263.8
408	459.4	510.7	146.0	138.7	267.9	264.4
409	460.5	512.0	146.4	139.1	268.6	265.1
410	461.6	513.2	146.8	139.5	269.3	265.8
411	462.7	514.5	147.2	139.9	270.0	266.4
412	463.8	515.7	147.6	140.2	270.7	267.1
413	465.0	517.0	148.0	140.6	271.4	267.8
414	466.1	518.2	148.4	141.0	272.1	268.4
415	467.2	419.5	148.8	141.4	272.8	269.1
416	468.4	520.7	149.2	141.8	273.5	269.7
417	469.5	522.0	149.6	142.2	274.1	270.4
418	470.6	523.2	150.0	142.6	274.8	271.1
419	471.8	524.5	150.4	143.0	275.5	271.8
420	472.9	525.7	150.8	143.3	276.2	272.4
421	474.0	527.0	151.2	143.7	276.9	273.1
422	475.1	528.2	151.6	144.1	277.6	273.8
423	476.2	529.5	152.0	144.5	278.3	274.4
424	477.4	530.7	152.4	144.9	278.9	275.1



TABLE IV (Continued).

Copper. Mg.	Cuprous oxide. Mg.	Cupric oxide. Mg.	Glucose. Mg. C <sub>6</sub> H <sub>12</sub> O <sub>6</sub> .	Invert sugar. Mg. C <sub>6</sub> H <sub>12</sub> O <sub>6</sub> .	Lactose. Mg. C <sub>12</sub> H <sub>22</sub> O <sub>11</sub> .H <sub>2</sub> O.	Maltose. Mg. C <sub>12</sub> H <sub>22</sub> O <sub>11</sub> .H <sub>2</sub> O.
425	478.5	532.0	152.1	145.3	279.6	275.8
426	479.5	533.2	153.2	145.7	280.3	276.5
427	480.7	534.5	153.6	146.1	280.9	277.1
428	481.9	535.7	154.0	146.4	281.6	277.8
429	483.0	537.0	154.4	146.8	282.3	278.5
430	484.1	538.2	154.8	147.2	282.9	279.1
431	485.3	539.5	155.3	147.6	283.6	279.8
432	486.4	540.7	155.7	148.0	284.3	280.5
433	487.5	542.0	156.1	148.4	285.0	281.1
434	488.6	533.2	156.5	148.8	285.7	281.8
435	489.7	544.5	156.9	149.1	286.4	282.5
436	490.9	545.7	157.3	149.5	287.1	283.1
437	492.0	547.0	157.7	149.9	287.8	283.8
438	493.1	548.2	158.1	150.3	288.5	284.5
439	494.3	549.5	158.5	150.7	289.2	285.2
440	495.4	550.7	158.9	151.1	289.9	285.8
441	496.5	552.0	159.3	151.5	290.6	286.5
442	497.6	553.2	159.8	151.9	291.3	287.2
443	498.8	554.5	160.2	152.3	292.0	287.8
444	499.9	555.7	160.6	152.7	292.7	288.5
445	501.0	557.0	161.0	153.1	293.4	289.2
446	502.1	558.2	161.4	153.5	294.1	289.8
447	503.2	559.5	161.8	153.9	294.8	290.5
448	504.4	560.7	162.2	154.3	295.5	291.2
449	505.5	562.0	162.6	154.7	296.2	291.9
450	506.6	563.3	163.0	155.1	296.9	292.5

## NEW BOOKS.

**Famous Chemists.** By E. ROBERTS, B.Sc. New York: The MacMillan Company. pp. 243. Price, \$0.80 net.

The book contains very brief accounts of the lives and researches of Stahl, Boyle, Black, Cavendish, Priestley, Scheele, Lavoisier, Berthollet, Dalton, Davy, Gay-Lussac, Berzelius, Faraday, Dumas, Wöhler, Liebig, Graham, Bunsen, Hofmann, Pasteur, Williamson, Frankland, Kekulé, Mendeléeff, Perkin, and V. Meyer. The author says in the preface: "The object of this little book is to give an account of the chief work of the most famous chemists, and to indicate briefly the part played by each in the development of the science." While the names of a few of "the most famous chemists" are lacking in the list, the accounts which are given are in the main satisfactory. However, the book contains a few errors, the most serious of which are listed below.

P. 52, "Lavoisier showed that respiration is analogous to combustion." This fact had long been known. P. 59, "Chemists, up to the beginning of the nineteenth century, considered it a self-evident fact that sub-