CLVI. THE DETERMINATION OF CELLULOSE IN STRAWS.

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For the determination of cellulose in straws the standard method of Cross and Bevan [1918] is the most reliable. However, it suffers from three disadvantages, which are (1) the inconvenience of working with gaseous chlorine, (2) the lengthy extraction with sodium sulphite solution which is required to remove the lignone chloride, and (3) the difficulty of chlorinating a number of samples simultaneously.

A method of determining cellulose in straws is described in this paper, in which the straw is chlorinated by means of sodium hypochlorite in alkaline solution; it is first necessary to boil the straw with dilute acid and alkali in order to bring it into a condition in which it is amenable to hypochlorite treatment. After boiling with acid and alkali the lignin present in the extracted straw is chlorinated and dissolved at the same time while the cellulose remains unattacked owing to its stability in presence of neutral or alkaline hypochlorites in dilute solution. Two such chlorinations completely remove the lignin, leaving a white or cream-coloured product which retains the whole structure of the straw. This product consists of cellulose intimately associated with xylan and in character is identical with the cellulose of Cross and Bevan. One unimportant difference exists between the two methods, viz. the xylan fraction expressed as a percentage of the chlorination product ("cellulose") is less in the case of the hypochlorite method than in that of Cross and Bevan. Actually more "cellulose" is obtained by the latter procedure so that when the xylan content of the chlorination product is taken into account the yields of true cellulose by the two methods are identical. Using hypochlorite it has been found possible for one worker to carry out from 12 to 16 cellulose determinations in a day.

One obvious advantage which the use of hypochlorite offers is the preparation of straw celluloses in a form suitable for structural or chemical examination. To obtain a theoretical yield of cellulose from 200 or 300 g. of straw is quite simple. A preparation of this magnitude involving the use of gaseous chlorine would be tedious and unpleasant.

The suitability of the hypochlorite method for the determination of wood cellulose is now being studied and the results will be given in a later paper.

Experimental.

Solutions required. (1) Sodium hypochlorite, commercial, 15 % free chlorine (Baird and Tatlock, Ltd., London). This should be stored in a cool dark place. The strength of this solution varies from 14 to 19 %, but unless the free chlorine lies outside this range it is not necessary to make allowance for the variation in the volumes of hypochlorite taken for the chlorination. The free chlorine may be determined as follows: 5 cc. of hypochlorite are made up to 500 cc. with water; 10 cc. of the dilute solution are treated with 10 cc. of 5 % acetic acid and 5 cc. of 10 % potassium iodide. The iodine liberated is titrated with $N/20$ sodium thiosulphate using a starch indicator. Then:

$$\% \text{ free chlorine} = \text{cc. of } N/20 \text{ thiosulphate} \times 1{\cdot}775.$$

(2) Sodium hydroxide, 10 % solution.

(3) 10 % hydrochloric acid; 30 cc. conc. hydrochloric acid made up to 100 cc. with water.

(4) 2 % hydrogen peroxide: 10 cc. of 20 vol. hydrogen peroxide made up to 100 cc. with water.

Standard method for the estimation of cellulose in straws. 2 g. of very coarsely pulverised straw in a beaker are brought to the boil with 100 cc. of water and 10 cc. of 10 % NaOH. The beaker is then immediately placed in a boiling water-bath for 5 min. and the solution is poured on to a piece of white poplin cloth stretched over a $2\frac{1}{2}$ inch Büchner funnel. When the liquid has been sucked off the straw is washed with water, the washings being poured on to the cloth each time. The straw is then transferred from the cloth to the beaker by means of a wide jet of water, and the volume is made up to approximately 100 cc. Then 10 cc. of 10 % HCl are added, the mixture is brought to the boil and the beaker placed in a boiling water-bath for 5 min. The straw is filtered and washed as before. The extraction of sugars, starch, and hemicelluloses is completed by a second alkali and acid treatment carried out in exactly the same manner as the first.

The extracted straw is made up to about 100 cc. and 5 cc. of sodium hypochlorite solution are added. This mixture is allowed to stand for 20 min. in a cool place away from bright sunlight. During this time the reaction of the solution must remain alkaline to brilliant yellow or litmus paper: sodium hydroxide solution is added if necessary. The straw is then filtered, washed, transferred to the beaker and chlorinated a second time for 20 min. It is next filtered through cloth, well washed with cold water, then with 100 cc. of 2 % hydrogen peroxide, and then with boiling water. Finally the residual cellulose is washed back into the beaker and then on to a weighed, oven-dried Gooch crucible. It will be found that rapid filtration without loss of cellulose is attained by using a cotton disc of lawn or organdie in place of an asbestos filter. A furfuraldehyde determination is made on the dried and weighed hypochlorite cellulose by the Tollens method [1902] using the Kröber factor [1900] for converting the phloroglucide yield to furfuraldehyde or xylan.

Effect of the hypochlorite method on the yield of cellulose and furfuraldehyde.

To test the percentage recovery of cellulose in the method described above and the amount of oxycellulose formed, a pure cotton cellulose was prepared in the following manner. A half-pound hank of the best American cotton was gently boiled with successive quantities of 1 % sodium carbonate for 4 hours, acidifying between each boil with 0·5 % HCl. After four such treatments the cotton was well washed with water and then bleached twice with 0·25 % sodium hypochlorite solution. The cotton was kept out of contact with air during the bleaching. It was thoroughly washed with water, then with 0·1 % HCl and again with water until the p_H of the washings and of the water used was identical. After the excess of water had been squeezed out of the cotton it was dried at 100°. The purified cellulose gave a yield of 0·29 % furfuraldehyde which corresponds to 0·46 % of anhydroxylose. It is not suggested that the furfuraldehyde-yielding substances in cotton cellulose consist entirely of xylose units but the figure is given for comparison with the anhydroxylose in straw cellulose.

Table I shows the effect on the pure cellulose of (*a*) two chlorinations with 5 cc. NaOCl and 100 cc. of water, each of 20 min.; (*b*) two extractions with 1 % NaOH and 1 % HCl, followed by two chlorinations for 20 min. as in the method described for straws.

Table I.

Treatment	(*a*)	(*b*)
Recovery of cellulose %	97·28	97·01
Anhydroxylose in cellulose %	0·59	0·43

It may therefore be concluded that neither treatment has an appreciable oxidising effect on the cellulose. Figures of a similar order are obtained when pure cellulose is chlorinated according to the Cross and Bevan method.

The yield of cellulose obtained by the Cross and Bevan method and by the hypochlorite method will now be compared. The latter procedure, as has already been pointed out, always seems to give about 2 or 3 % less of the chlorination product than the standard method. But when allowance is made for the larger amount of xylan (shown by the higher furfuraldehyde yield) invariably present in the Cross and Bevan cellulose it is at once seen from the figures in Table II that the amount of true xylan-free cellulose obtained is the same by both methods.

Table II.

Method	"Cellulose" in 100 g. oat straw g.	Furfurald. yielded by 100 g. "cellulose" g.	Furfurald. yielded by "cellulose" in 100 g. straw	Xylan equivalent of furfurald. in "cellulose"	Pure cellulose in straw %
Cross and Bevan	48·92	12·30	6·02	9·33	39·59
Hypochlorite	47·50	11·07	5·26	8·15	39·59

Further examples showing the results of cellulose estimations by the hypochlorite method are given in Table III.

Table III.

Straw	Hypochlorite "cellulose" %		Furfurald. in hypochlorite cellulose %		Anhydro-xylose equivalent to furfurald. %		Anhydro-xylose in hypochlorite cellulose in 100 g. straw		Pure cellulose in straw %	
Rye	56·36	55·66	14·56	14·20	22·55	22·01	12·71	11·3	43·65	43·36
Barley	50·45	49·25	11·05	11·24	18·33	18·05	9·25	8·89	41·20	40·36
Oat from straw filter fed with nitrogenous soln. for 23 days	51·10	51·21	10·94	11·22	16·95	17·39	8·49	8·91	41·61	42·20
Oat from straw filter fed with water alone for 53 days	54·01	52·45	12·77	10·53	19·78	16·21	10·33	8·50	43·33	43·95

Preparation of cellulose from straws. The examination of reasonably pure celluloses from straws is obviously of importance in connection with X-ray analysis, the morphology of the plant, and its chemical constitution. Hitherto such examination has been somewhat restricted owing to the difficulty of preparing large samples of the cellulose in a reasonably pure and unchanged condition. However, with the hypochlorite method very little modification of the proportions of the reagents used in the estimation is required for the isolation of larger quantities of cellulose. The following method has proved satisfactory for oat straw. 178·5 g. of dry straw were mixed with 7 litres of boiling water containing 100 cc. of concentrated HCl and left in a boiling water-bath for 10 min. The mixture was then strained through a cloth and the residual straw added to 7 litres of boiling water containing 15 g. of NaOH. After standing in the boiling water-bath for 10 min. the aqueous portion was poured off through a cloth and then chlorinated for 20 min. with 200 cc. of 15 % NaOCl. The straw was filtered off, washed and again chlorinated with the same quantity of hypochlorite. After washing the cellulose first with cold, then with hot water, it was washed with 2 litres of 0·5 % hydrogen peroxide. The use of hydrogen peroxide ensures complete decomposition of the hypochlorite. Finally the cellulose was washed free from hydrogen peroxide with cold and hot water. In this manner 93 g. of cellulose were obtained, equivalent to a 52·1 % yield on the weight of straw taken. An analysis of the sample of oat straw showed that it contained 52·2 % of cellulose, which gave a furfuraldehyde yield of 11·1 %.

Summary.

1. The cellulose in straw is readily determined by treating the straw with hot dilute alkali and acid and then with cold hypochlorite solution.

2. The product obtained is practically identical in character with the Cross and Bevan chlorination product, except that it contains slightly less xylan than the latter. When due allowance is made for the xylan present the

percentage of pure cellulose found in a straw is the same by the Cross and Bevan and the hypochlorite method.

3. The treatment referred to in 1 has a negligible effect on pure cellulose.

4. The hypochlorite method of chlorinating has the following points in its favour.

(*a*) From 12 to 16 cellulose determinations can be carried out in a day by one worker.

(*b*) Large scale preparations of straw cellulose are possible without inconvenience.

(*c*) The cellulose can be prepared in quantitative yield.

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