METHOD FOR THE PREPARATION OF GREEN PLANT MATERIAL FOR THE EXTRACTION OF JUICES¹

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Introduction

The extraction of plant juices is often desirable or essential for a study of their physico-chemical properties. As methods in general use are laborious and time consuming, it was found desirable in a recent investigation on the nutrition of wheat to develop a rapid, simple, yet accurate method for extracting the plant juice.

A review of the literature indicated that several methods have been devised for the treating of plant tissue to facilitate the expression of its juice. These methods may be divided into two general groups: either the material is ground in a mortar or grinding mill, or it is frozen in a slushy mixture of ice and salt, a cold storage room, liquid air, or solid carbon dioxide. The method in most frequent use is freezing the tissue in a test tube or jar for a number of hours, thawing it rapidly, and subjecting it to pressure. Some excellent and comprehensive reviews of the methods employed have been contributed by MEYER ($\mathbf{6}$), GORTNER ($\mathbf{1}$), SAYRE and MORRIS ($\mathbf{8}$), and GREATHOUSE ($\mathbf{3}$).

In the proposed method the material was heated in an autoclave. Approximately 100 young winter wheat plants were used, some grown in the greenhouse and others under natural field conditions of late winter and early spring. Juices were satisfactorily obtained from the wheat plants at regular intervals during the growing period.

Procedure

The wheat plants were wrapped in damp cloths in the field, placed in a moist, air-tight container, and brought immediately to the laboratory. The wrapped plants were then placed in an autoclave for five minutes at 15 pounds' pressure, although a reasonable variation in the length of time does not affect the results. Material which was not wrapped tightly was often sufficiently cooked in one minute at 15 pounds' pressure, but it was found advisable to leave the tightly wrapped material in the autoclave for five minutes or until the thoroughly cooked plant tissue was light brown in color. After autoclaving, the sample was removed and cooled to room temperature. According to the results of a number of trials, the moisture content of the tissue does not seem to be affected materially by autoclaving.

¹ Published as Scientific Paper no. 278, College of Agriculture and Experiment Station, State College of Washington. The small amount of condensation that may occur during the heating was lost by evaporation while cooling to room temperature. The sap was expressed as soon as the material was cool to prevent further loss of moisture and possible changes in percentage of the plant juice.

A large percentage of the juice might have been expressed by hand, but to be certain of obtaining a representative sample, a hydraulic press was employed, applying uniform pressures of 7000 pounds per square inch and uniform times of draining of four minutes after this pressure was reached. Approximately 90 per cent. of the plant juices was extracted by this method. However, this varies with the type of plant, its age, and the conditions under which it is grown.

The method of freezing the wheat plants in a cold storage room at a temperature of 30° C., and the method of autoclaving them antecedent to expression of the sap were compared for determinations of the freezing point, total solids, total sugars, and nitrate-nitrogen of the expressed plant juice.

The freezing point of the juice was determined in the usual freezing point apparatus with a Beckmann thermometer. The figures reported were the results of four closely agreeing determinations on each sample. The freezing point depression readings were converted into equivalent osmotic values according to the equation of LEWIS (5).

The percentage total solids in the expressed plant juice was obtained by the Abbé refactometer according to GORTNER and HOFFMAN (2).

The total sugar content was determined by the Munson-Walker method, the cuprous oxide being weighed directly according to the recommendation of the Association of Official Agricultural Chemists (7).

The nitrate-nitrogen content was found by the colorimeter method of HoLTZ and LARSON (4).

Results

A comparison of the results for a number of determinations of osmotic pressure and total solids, obtained by the proposed autoclave method and freezing method antecedent to expressing the plant juice, is given in table I.

Samples 1, 2, 3, 4, and 5 were grown in the greenhouse during the winter. Samples 6 and 7 were grown on land continuously cropped by wheat and were sampled in the field in the early spring. Samples 8 and 9 were similar to samples 6 and 7 except that they were grown on summer fallow land.

A comparison of the total sugar and nitrate-nitrogen of the expressed juice as obtained by the two methods is given in table II.

The osmotic pressure, total solids, total sugar, and nitrate-nitrogen of the autoclaved and frozen plant tissue agree closely. The small variation

TABLE I

OSMOTIC PRESSURE AND TOTAL SOLIDS OF PLANT SAP OBTAINED BY AUTOCLAVING AND BY FREEZING THE PLANT MATERIAL ANTECEDENT TO EXPRESSING THE SAP

SAM-	WHEAT	METHOD OF	FREEZING POINT	OSMOTIC	PRESSURE	TOTAL SOLIDS		
PLE NO.	VARIETY	TREATMENT	DEPRES- SION	ATMOS- PHERES	DIFFER- ENCE	PERCENT- AGE	Differ- ence	
			° C.					
1	Hybrid 128	{ Frozen { Autoclaved	$\begin{array}{c} 1.062 \\ 1.054 \end{array}$	$\begin{array}{c} 12.78\\ 12.68\end{array}$	- 0.10	$7.7 \\ 7.5$	- 0.20	
2	White Odessa	{ Frozen { Autoclaved	0.908 0.900	$\begin{array}{c} 10.96 \\ 10.85 \end{array}$	- 0.11	$\begin{array}{c} 5.4 \\ 5.7 \end{array}$	+ 0.30	
3	Coppie	{ Frozen { Autoclaved	$\begin{array}{c} 1.120 \\ 1.099 \end{array}$	$\begin{array}{c} 13.48\\ 13.23\end{array}$	- 0.25	$\substack{6.25\\6.6}$	+0.35	
4	Hussar	{ Frozen { Autoclaved	$\begin{array}{c} 0.940\\ 0.986\end{array}$	$\begin{array}{c} 11.32\\ 11.87 \end{array}$	+ 0.55	$\begin{array}{c} 5.25 \\ 5.05 \end{array}$	- 0.20	
5	Hybrid 128	{ Frozen { Autoclaved	0.953 0.938	11.48 11.29	- 0.19	4.90 5.05	+0.15	
6	Ruddy	{ Frozen { Autoclaved	$\begin{array}{c} 0.861\\ 0.870\end{array}$	$\begin{array}{c} 10.37\\ 10.48\end{array}$	+ 0.11	9.25 9.15	- 0.10	
7	Hybrid 128	{ Frozen { Autoclaved	$\begin{array}{c} 0.800\\ 0.802 \end{array}$	9.60 9.60	0.00	8.80 8.50	- 0.30	
8	Ruddy	{ Frozen { Autoclaved	0.688 0.701	8.28 8.44	+ 0.16	$5.70 \\ 5.75$	+0.05	
9	Hybrid 128	{ Frozen { Autoclaved	$\begin{array}{c} 0.658\\ 0.648\end{array}$	$7.91 \\ 7.80$	- 0.21	$\begin{array}{c} 5.40 \\ 5.40 \end{array}$	0.00	

that occurred could be accounted for partly by the difference in sampling. The proposed treatment of plant tissue can therefore be satisfactorily used for some phases of plant juice study instead of the freezing method.

SAYRE and MORRIS (8) showed that the sugar content of tissue could be calculated from the sugar content of the expressed juice. A comparison of the percentage sugar and nitrate-nitrogen in the plant juice from the autoclaved material with that of the dried wheat plant is given in table III.

The tissue was dried at a temperature of 60° C. in an air current. There were smaller differences between the duplicate autoclaved samples than between samples of dried tissue. This is especially true of sugar content.

Numerous investigations have shown that drying of green plant material has a tendency to result in a change of the various carbohydrates and proteins. This is well illustrated in table III.

The autoclave method cannot be used in a study of proteins, that is, proteins precipitated by heat, any more than the freezing method can be used in such a study. This method, however, has many advantages for the study of the truly soluble material not affected by heat. For instance, the

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TABLE II

			TOTAL	SUGAR	NITRATE-NITROGEN		
SAMPLE NO.	W HEAT VARIETY	METHOD OF TREATMENT	PERCENT- AGE	DIFFER- ENCE	PERCENT- AGE	DIFFER- ENCE	
			%		%		
3	Coppie	{ Frozen Autoclaved	$\begin{array}{c} 0.41\\ 0.39\end{array}$	- 0.02	$\begin{array}{c} 0.157\\ 0.153\end{array}$	- 0.004	
5	Hybrid 128	{ Frozen { Autoclaved	$\begin{array}{c} 0.41\\ 0.42\end{array}$	+0.01	$\begin{array}{c} 0.141 \\ 0.145 \end{array}$	+ 0.004	
6	Ruddy	{ Frozen { Autoclaved	$3.75 \\ 3.65$	- 0.10	$\begin{array}{c} 0.018\\ 0.019\end{array}$	+ 0.001	
7	Hybrid 128	{ Frozen { Autoclaved	$3.60 \\ 3.65$	+0.05	$\begin{array}{c} 0.009\\ 0.011\end{array}$	+0.002	
8	Ruddy	{ Frozen { Autoclaved	$\begin{array}{c} 0.78\\ 0.82 \end{array}$	+ 0.04	$\begin{array}{c} 0.041\\ 0.038\end{array}$	- 0.003	
9	Hybrid 128	{ Frozen { Autoclaved	$\begin{array}{c} 0.65\\ 0.68\end{array}$	+0.03	$\begin{array}{c} 0.032\\ 0.030\end{array}$	- 0.002	

TOTAL SUGAR AND NITRATE-NITROGEN OF PLANT SAP OBTAINED BY AUTOCLAVING AND BY FREEZING THE PLANT MATERIAL ANTECEDENT TO EXPRESSING THE SAP

enzyme action is almost instantly stopped by the high temperature in the autoclave. A large proportion of the plant juice is extracted and consequently a more representative sample of plant sap is obtained. This is ap-

TABLE III

PLANT SAP AND DRIED WHEAT TISSUE COMPARED FOR DETERMINATIONS OF TOTAL SUGAR AND NITRATE-NITROGEN BASED ON DRY WEIGHT

	PLANT TREATMENT	PERCENTAGE SUGAR			PERCENTAGE NO3-N		
VARIETY		SAMPLE		DIF-	SAMPLE		DIF-
		1	2	FER- ENCE	1	2	FER- ENCE
Ruddy	{ Sap autoclaved { Tissue dried	$\begin{array}{c} 6.02 \\ 6.33 \end{array}$	$\begin{array}{c} 6.11 \\ 5.60 \end{array}$	0.09 0.73	$\begin{array}{c} 1.02\\ 0.88\end{array}$	0.98 0.92	0.04 0.04
Little Club	{ Sap autoclaved { Tissue dried	4.88 4.88	$\begin{array}{c} 4.88\\ 3.17\end{array}$	0.00 1.71	1.04 0.90	1.01 1.10	0.03 0.20
Jones Fife	{ Sap autoclaved { Tissue dried	$\begin{array}{c} 7.35 \\ 6.02 \end{array}$	$\begin{array}{c} 6.93\\ 4.88\end{array}$	$\begin{array}{c} 0.42\\ 1.14\end{array}$	$\begin{array}{c} 0.86 \\ 1.04 \end{array}$	$\begin{array}{c} 0.86\\ 1.02 \end{array}$	$\begin{array}{c} 0.00\\ 0.02 \end{array}$

parently due to a better breaking down of the membranes and cell walls by the heat. The method is rapid as it is possible to autoclave and express ten samples of plant tissue per hour.

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