Developments in Methods for Microbiological Control in Liquid Sugar

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Representative sampling is a very important factor in obtaining an evaluation of the organism population in a lot of liquid sugar.

A grab sample taken from a storage tank may or may not be representative. A sample taken from the syrup surface of a tank car load may show an excessively high organism count, while a sample taken from the flow during unloading may show that the count is within the acceptable limits.

Continuous sampling during unloading of a tank car or storage tank appears to be the only means of obtaining a representative sample. A satisfactory device for such sampling is still to be thoroughly proven. Figure 1 shows a device for continuous sampling of liquid sugar which has been used in preliminary testing by the Spreckels Sugar Company and which may prove to be satisfactory.

The sampler is permanently attached to the syrup transfer line. The pet cock is adjusted so that the sterile sample bottle will be filled during the period of loading or of unloading. When sampling is completed the sample bottle is removed and closed with a sterile, conventional cap. The sampler is then cleaned and flame sterilized prior to the next sampling.

In attempting an organism count on a large quantity of liquid sugar, a true picture of the problem is desired. Even though continuous sampling is practiced, the chances for getting a representative organism count are better when greater amounts of sample are plated. The ABCB method, in general use by producers and consumers of liquid sugars, specifies the use of only 5 grams of dry sugar equivalent per sample. The result is multiplied by two to obtain the count on a 10 grams basis. Due to the tendency for low populations of yeasts and molds to follow the Poisson distribution (1)[2] throughout a sample of liquid sugar, it becomes quite apparent that a larger sample for plating would be desirable.

Since World War II there has been a steady increase in the use of molecular filter membranes in bacteriological analyses. These filters were originally developed some thirty-five years ago by Professor R. Zsigmondy of Goettingen, Germany. During the

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[2] Numbers in parentheses refer to literature cited.

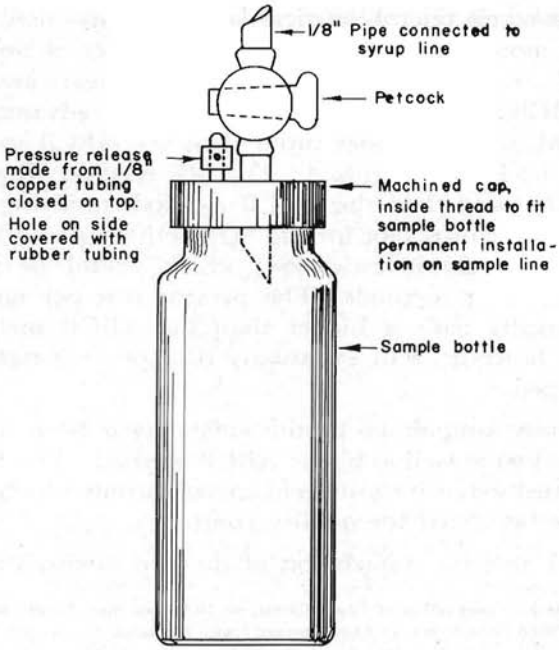

Figure 1.—A continuous liquid sugar sampler.

years, and particularly during the war, the manufacture of these filters reached a very high state of perfection.

When first introduced into this country the filters were principally used in water analyses but they are now being successfully applied to other bacteriological analyses and to the filtration of colloids and proteins, as well as to studies in dialysis and osmosis.

The filters are supplied in this country by at least two manufacturers and are available in several degrees of porosity. Several publications on their use have appeared (2), (3).

The type used by the Spreckels microbiological laboratory is the MF membrane (Millipore Filter Corporation) with a pore diameter of $0.45\mu \pm .02\mu$. The filters are supplied with absorbent pads for impregnation with a nutrient. The membranes with the trapped organisms are placed on these moistened pads during incubation.

One supplier (A. G. Chemical Company) furnishes "dehydrated scheduled nutrient packs," which contain MF membranes and absorbent pads with preimpregnated dehydrated nutrients. These pads need only to be moistened with sterile water prior to use.

The Spreckels microbiological laboratory has used MF membranes for more than two years in the analyses of liquid sugars, water, and air. Routine analyses of liquid sugars are still made by the ABCB method. It is hoped that the advantages of the MF method, which is more rapid than the ABCB method, will eventually be fully recognized. The MF method allows a larger sample to be used than the ABCB method, resulting in greater accuracy. The equipment for the MF method is readily portable, and ideal for field investigations which would be impractical when using other methods. The present cost per sample using prepared media pads is higher than the ABCB method. This difference, however, will eventually disappear as new nutrients are developed.

Numerous samples of liquid sugars have been analyzed by the MF method as well as by the ABCB method. The MF method has been used extensively in field investigations which have been found very beneficial for quality control.

Table 1 shows a comparison of the two methods of analyses.

Table 1.—Comparison of Fungi Counts on 10 Grams (dse) Liquid Sugar.
Samples from factory and consumer storage tanks, including others than Spreckels

Sample No.	Methods Used				Sample No.	Methods Used			
	ABCB		AG			ABCB		AG	
	Yeasts	Molds	Yeasts	Molds		Yeast	Mold	Yeast	Mold
1	74	8	57	9	18	0	2	0	0
2	4	18	2	TNTC	19	0	8	0	0
3	4	166	1	263	20	0	0	0	0
4	24	6	11	7	21	2	4	0	0
5	2	8	0	4	22	0	0	2	0
6	4	10	0	5	23	2	2	5	0
7	0	12	0	3	24	94	2	79	3
8	524	176	226	2	25	8	4	17	0
9	28	24	3	5	26	0	0	0	0
10	398	236	300	14	27	0	2	0	2
11	12	8	12	4	28	0	0	4	0
12	4	10	3	4	29	0	2	3	0
13	0	6	0	1	30	0	2	1	0
14	2510	0	1578	0	31	2	2	450	1
15	0	4	2	1	32	0	0	2	1
16	0	8	0	2	33	4	0	5	0
17	2	8	2	2	34	8	4	7	2

Summary

Conventional sampling methods now employed for microbiological control of liquid sugar are not entirely reliable due to the variation in microorganism distribution within a large volume of syrup.

Spreckels Sugar Company has conducted preliminary testing of a device which may obtain a respresentative composite sample of liquid sugar from a pipe line.

Upon obtaining a representative sample the analytical procedures for microorganism determinations must be relied upon for an evaluation of the organism population.

The accepted ABCB plating method relies on a small sugar sample. It is also slow and requires a large amount of equipment. The membrane filter method is more rapid, larger sample portions may be used, and less equipment is required.

Literature Cited

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