THE USE OF SEMI-PERMANENT MOUNTS IN SANITATION COLLABORATIVE WORK

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Due to the failure of technicians to agree on their results, it is very difficult to obtain significant answers to problems of sanitation analysis through collaborative tests. Much of the reason for disagreement is individual differences in ability to count fragments and rodent hairs.

The paper describes a technique whereby these differences are smoothed out. Filter papers bearing residues from sanitation tests are embedded in a thin layer of paraffin and read collaboratively. The paraffin is melted while the filter paper is being read, and then solidifies, anchoring all particles in place.

In addition to giving more reliable results, the paraffin-coated slide technique has shown itself to be a good training medium.

A serious obstacle in the path of anyone attempting to evaluate a method of sanitation analysis is the difficulty of obtaining agreement between collaborators. This is particularly true when working with flour and other cereal products.

In this type of material, any insect tissue or rodent filth that may be present are so pulverized that many particles are not detected, and of those located, many cannot be recognized. This difficulty is further complicated by the fact that most of the persons doing sanitation analyses in cereal laboratories are chemists who have had little or no formal training in microscopy or insect morphology.

The results obtained on the 1953-54 American Association of Cereal Chemists Sanitation Check-Sample series are typical of what may be expected. On the sample of this series where the agreement was best, the highest number of fragments reported was five times that of the lowest. On the sample where the agreement was poorest, the ratio between high and low was 63:1. The mean of the high-to-low ratios for 10 samples of the series was 26:1 (1). These differences can be caused by; (a) lack of uniformity of sample, (b) differences in methods, (c) differences in ability to find and recognize fragments and rodent hairs, and (d) differences in manipulative skill.

Since the fall of 1953 much of the work of the Sanitation Methods Committee, A.A.C.C. has been pointed toward finding ways to overcome, eliminate, or cancel out these causes for differences. The most effective technic developed has been the use of semi-permanent mounts for filter papers containing fragment and filth bearing residues.

**METHOD**

The papers containing the residues are embedded in a thin layer of paraffin. While the slides are being read, the paraffin is melted so that the effect is the same as though the filter paper has been cleared with mineral oil. When the reading is completed, the resolidified paraffin anchors all particles in place, allowing the slide to be sent to another person to be read collaboratively. One type of light in general use (2) furnishes enough heat to melt the paraffin while the slide is being read. Those who do not use this light may use a substage warmer consisting of a 100 watt light bulb enclosed in a box.

Special filter papers are used. They are marked off in 5.0 mm. cross sections. The squares are identi-
ified by letter and number, allowing the collaborators to report not only the numbers of insect fragments and rodent hairs found, but also their exact locations. The filter papers are ruled with a rubber stamp and green ink (3). Similar cross-sectioned filter papers are now available commercially (4). They are excellent for routine work; however, if they are to be used for collaborative work it is necessary to print the identifying numbers and letters on them by hand.

In practice, the slides are routed in pairs to groups of collaborators ranging in size from five to ten persons. Depending upon the information wanted, these collaborators report either the exact locations (by squares) or the numbers of insect fragments and rodent hairs found on each line. After the slides have been read, a summary of the reports is prepared, and distributed to the collaborators. The slides are re-routed to them in order to give them an opportunity to review the slides in the light of the report.

**Advantages of the Technic**

The use of these semi-permanently mounted filter papers has many advantages in the study of sanitation methods. They are as follows:

1. One of the slides of the pair being circulated always contains the residue from a test by a “standard” method. The other one contains the residue from a test on the same flour by some variation of this “standard” method. A comparison of the averages reported by those who read the slides collaboratively gives a much better indication of the relative numbers of fragments and rodent hairs present than would be obtained if they were read by only one person.

2. A method has been devised to analyze, statistically the composite reports of a group of collaborators (5) and assign a numerical accuracy rating to each collaborator. Some of the factors having an influence on an individual’s accuracy have also been measured and correlated with the accuracy ratings. This has given some insight into the reasons why a person may or may not be an accurate worker. Being able to promise and perform this service has helped very much in the recruitment of collaborators to work on committee projects, as this is something that will be of immediate and practical benefit to them.

3. It has made possible the measurement of the consistency of performance of individuals when reading slides.

4. The collaborative reading of these slides has helped to improve the recognition skills of those taking part in the work. The calculation and assignment of accuracy ratings (5) has given each one an indication of how he stands, in comparison to the group. The figures are broken down, also, to show if he is lacking in visual acuity, is reckless or over-cautious in naming a fragment or rodent hair, and whether, compared to the group, he is a rapid or slow worker. At the end of last year’s work (1954-55 season) each collaborator was asked if his participation in the work had helped him to improve his recognition skills. The answer was an almost unanimous “yes”.

This improvement is expected to result in better agreement and therefore more significant information to be obtained from the projects of the Sanitation Methods Committee.

**Technic Precautions**

There are some precautions that must be observed when the paraffin coated slide technic is used. Metal dishes are much more satisfactory than the usual petri dishes. It has been found that the flat, metal lids for friction-top cans, known to the can trade as “plugs”, make ideal dishes for this purpose. The manner in which the edge is formed stiffens and re-inforces them so that they may be mailed with only slight danger of their being damaged. When a sub-stage warmer is used, the bottom light would be distracting if a petri dish were used. Use of a metal dish avoids this. The minimum possible amount of paraffin should be used. If the filter paper is covered with a thick film of paraffin, the heat necessary to melt it may set up convection currents which will cause lighter particles to shift their positions. It has been found that when a 7 cm. filter paper is used, about 0.7 gram of paraffin is satisfactory. The filter paper should not be allowed to dry before it is embedded in the melted paraffin. It will curl as it dries and then cannot be made to lie flat in the dish. Also, if the paper is moist and flexible, it can be rolled into the melted paraffin in such a way that the minimum amount of air will be entrapped under it. A dry paper has a further disadvantage in that more paraffin will be required to impregnate it.

Collaborators should be warned that the paraffin must be held at a high enough temperature so that a ball will not form on the tip of a cold probe. If this happens, fragments may become shifted or lost.

**References**

1. Unpublished reports of Check-Sample Committee, A.A. C.C. to subscribing members.
2. Bausch and Lomb Reflector – Illuminator.
3. Furnished through the courtesy of Carl Schliecher & Schuell Co.
4. S and S “Quadrant Ruled” filter papers.