

would be longitude 100; actually it had been found on longitude 107. Lowell's work was worthy of considerable attention. He thought the discovery would explain a good deal of the perturbation of the planets.

Professor A. S. Eddington, of Cambridge, remarking on the closeness of the prediction, said that if one had had faith and had given a long exposure with a reasonably large lens, the planet must have been picked up. Many of them had been much impressed with the thoroughness, caution and honest work at Lowell Observatory.

Captain Ainslie, president of the British Astronomical Association, asked if the planet had been seen or only photographed.

The chairman said that as the size was given he assumed it had been seen visually.

Professor Turner thought this might rather be a calculation from its brightness.

Dr. Jackson said they were going to make a search

of the Franklin Adams plates at Greenwich, but if the planet were there it would be very near the edges of the plates.

Summing up, the chairman said this planet would probably have an effect on Halley's Comet that would alter the period of each return by as much as a couple of days, so that some of the unaccounted for days in the last two returns might meet with an explanation. In the course of time, when they had got a good enough orbit, it would involve the preparing of new tables of Uranus and Neptune. In the case of Neptune, the mass was very quickly got by the discovery of a satellite; there was very little chance of discovering a satellite of this body, and the only way to get its mass would be by getting the perturbations of Uranus and Neptune, and that would only give a rough result. Finally, every book of descriptive astronomy from that day was out of date.

SCIENTIFIC APPARATUS AND LABORATORY METHODS

A CONVENIENT METHOD OF REDUCING DESICCATION IN SLANT CULTURES

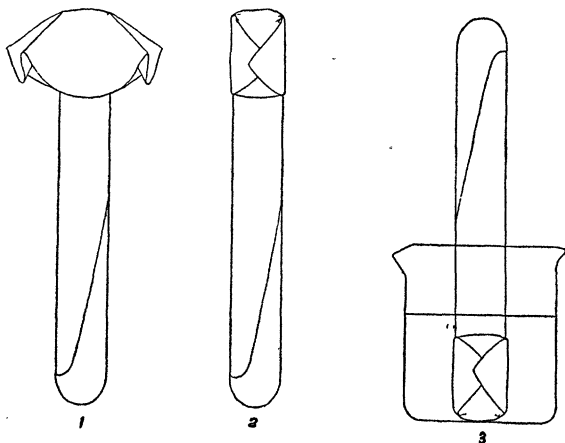
It is often desirable to reduce desiccation in slant cultures of fungi or bacteria, or in tubed media. The time-honored method of sealing cultures by dipping the stoppered end of the tube in paraffin presents some unsatisfactory features. When cotton stoppers are permeated with the wax they are difficult to remove and somewhat unwieldy. When replaced they do not fit tightly, and are not then proof against contamination. When the cotton is filled with wax the exchange of gases through the stoppers may be so reduced as to limit the growth of the culture, or alter the appearance of the organism.

Folding a piece of tinfoil over the mouth of culture tubes is a convenient means of reducing desiccation and has the advantage that the tubes of culture media may be sterilized after the foil caps are in place. Since vapor may readily escape between the foil cap

and outer wall of the culture tube a greater reduction in the rate of desiccation is often desirable. In this paper is described a method of reducing desiccation in slant cultures that seems to possess certain advantages.

Paper disks of a convenient size for folding over the end of culture tubes are cut from thin, tough and pliable paper. These disks are stacked up and submerged in melted paraffin just deep enough to cover them. After the papers are thoroughly saturated with the wax they are lifted out of it one at a time with needles or fine-pointed tweezers. They are held vertically for a few seconds to allow the excess of wax to drain off and are then dropped into cold water. The wax should be kept hot enough to allow the excess to drain from the paper before solidification occurs. The waxed paper disks may be used as soon as they are removed from the water or may be stored until needed.

Before a tube is sealed the open end is warmed slightly and the end of the cotton stopper is singed. A waxed paper disk is warmed until the wax becomes plastic, a condition that is obtained just before the melting-point is reached. It is then folded over the end of the tube (Fig. 1) and pressed firmly against the outer wall, and the folded edges are pressed down firmly. This procedure gives a waxed paper cap (Fig. 2) that permits an exchange of gases along the line of the folds, reduces the desiccation materially and keeps the cotton stoppers free from wax. The caps may be removed by giving them a firm twist or by unfolding the paper. When a perfect seal is desired, the tubes are inverted and dipped one or more times into paraffin, to a point just above the waxed paper cap (Fig. 3). For this purpose the tempera-



ture of the wax should be but slightly above its melting-point.

This method of reducing desiccation in slant cultures and in tubed media, as used for several years by the writer, has been uniformly successful. The method is very useful when a long period of incubation is required. Cultures can be kept in a suitable condition for study for several weeks, and, by completely sealing, sterile tubed media may be kept ready for use during a long period.

The method has a decided disadvantage. When the tubes are sealed the cotton stoppers are kept moist by water of condensation, permitting fungi to grow through them and contaminate the cultures. Contamination in this way is almost entirely avoided, however, when the work of preparing the waxed paper disks, the storing of them and the sealing of the tubes is done under aseptic conditions.

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A METHOD FOR DETECTING ACID-FAST BACTERIA IN THE SOIL

It has been assumed that saprophytic acid-fast bacteria are rather wide spread in nature and that their appearance in soil is not an uncommon occurrence. There has been, however, no definite evidence to substantiate this assumption.

In view of these facts experiments have been under way for several months attempting to identify these organisms under various natural conditions and more especially in the soil.

The technique used is as follows. Soil samples in approximately one gram portions are mixed with about 50 to 60 cc of modified Büttner's¹ medium in 200-cc flasks. This medium which has previously been sterilized in the flasks has the following composition:

Tap water	1000 cc
K ₂ HPO ₄	0.5 gm
NH ₄ Cl	0.5 gm
Mg SO ₄	0.2 gm
CaCO ₃	0.2 gm

It will be noticed that this medium contains no available carbon. To supply this carbon, paraffin-coated pebbles are placed in the flasks. These pebbles are large enough so that they will extend above the surface of the medium. Supplying carbon in this way seems to be quite effective in keeping down contamination since a great many organisms can not thrive under these conditions.

¹ Hans Büttner, "Zur Kenntnis der Mykobakterien insbesondere ihres quantitativen Stoffwechsels auf Paraffinnährboden," *Arch. f. Hyg.*, 97: 12, 1926.

Incubation is carried out at 47.5° C. It was found that at lower temperature there were gross contaminations by molds so that the acid-fast bacteria were so completely covered as to render detection of the organism very difficult and their isolation virtually impossible. When the high temperature was used there was no evidence of contamination by molds.

Up to the present time this technique has been applied to some thirty soil samples, and acid-fast bacteria have been found in every instance after from two to seven days' incubation.

In the incubation process some of the paraffin melts from the coated pebble and forms a thin pellicle on the fluid. The organisms are found on the under side of this pellicle and on the paraffined surface of the pebble. They can also be found, in many cases, adhering to the sides of the flask just above the surface of the medium. Under this latter condition we frequently get large masses of the organism which are comparatively free from contamination. After prolonged incubation it is usually possible to detect masses of the organisms with the naked eye. These have the appearance of opaque bodies on the surface layer of paraffin, on the pebble or on the side of the flask. In chromogenic species the color can be detected in these areas.

The organisms when isolated show numerous variations both morphologically and culturally. A discussion of these variations does not seem necessary at this time. When the organisms are stained with hot carbol fuchsin all are highly resistant to decolorization by 3 per cent. HCl in 95 per cent. alcohol.

A number of these organisms have been isolated by making dilution plates using Conn's² medium to which is added gentian violet in a dilution of 1 to 10,000. The composition of this medium is as follows:

Water	1000 cc
Agar	15 gm
Glycerin	10 gm
K ₂ HPO ₄	1 gm
Sodium Asparaginate	1 gm

While the number of soil samples examined is rather small, the writer feels that the method described is a relatively simple way of detecting these organisms in soil. The collecting of samples from various parts of the United States is now under way, and these will be subjected to this technique. A more detailed account of this problem will appear at a later date.

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² H. J. Conn, "The Use of Various Culture Media in Characterizing Actinomycetes," *N. Y. Ag. Exp. Sta. Tech. Bul.* 83, April, 1921.