tube is pulled to a finely drawn-out braking tip. A globule of low-melting-point, transparent, sealing resin[2] is melted onto the capillary near the end opposite the brake. The capillary tube is then dropped into the lumen of the pipette and heated cautiously over a microflame until the resin makes a seal at point

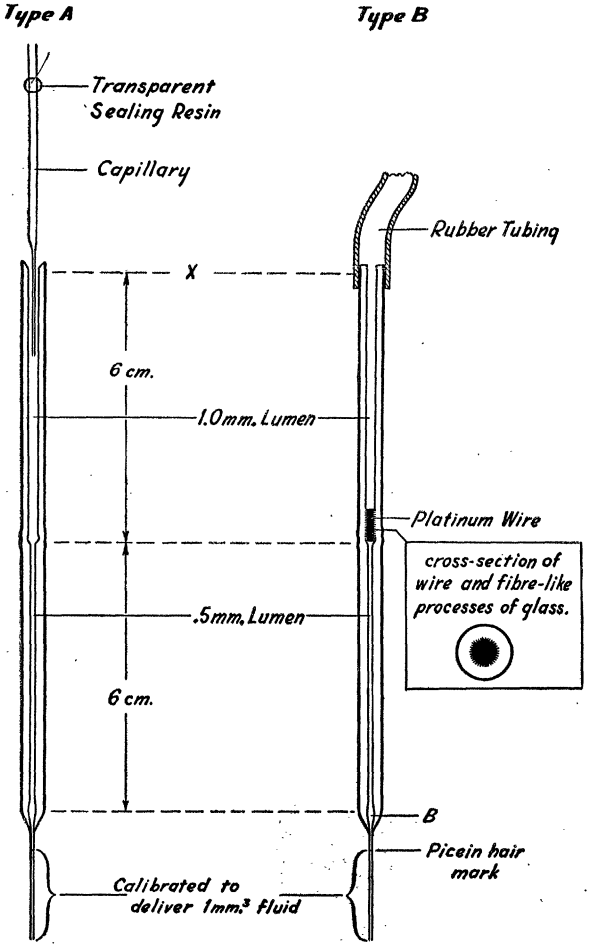

FIG. 1a FIG. 1b

X. The pipette is completed by pulling a tip in the conventional manner. A convenient way to mark the calibration on the tip is to pull a fine thread of heated black picein to a fine hair, winding it in a circle around the capillary tube at the calibration point before it is entirely cool. Then, by bringing the point where it is applied near a microflame, it will be sealed to the glass. The capillary brake may be cut close to the resin seal, and the pipette is ready for attaching the rubber tubing to complete a mouth-suction pipette.

Type B: The rate of air flow is controlled by taking advantage of the slight difference of the coefficient of expansion between platinum wire and pyrex glass. The two main advantages of this type are that (1) any number of pipettes with the same characteristics may be made by using the same length and diameter of platinum wire in the brake, and pyrex capillary tubing from the same lot of glass; and (2) fluid as well as moisture, if drawn into the brake, may be baked out over a microflame, and the pipette dried in a hot-air oven without materially changing the characteristics of the pipette.

A piece of heavy-wall pyrex capillary tubing 6 cm. long, with an internal diameter of 0.5 mm., is fused to a heavy pyrex capillary tubing of the same length, with an internal diameter of 1.0 mm. A piece of platinum wire 8 mm. long, with a diameter of .017 mm. (#25 gage), is inserted at point X (Fig. 1b) and worked down to the point where the two inside-diameter bores meet. The pipette is then heated at that point in an oxygen-air-gas flame until the platinum wire is white hot. The capillary tubing will shrink around the white-hot wire. Care must be taken not to fuse the capillary bore closed at either end of the wire. This can be avoided by adjusting the blowtorch to a rather sharp flame. The pipette should be held in this flame for about a minute after the capillary has shrunk onto the platinum wire, the capillary allowed to cool to a cherry red in an air and gas flame for about another minute, and then the capillary "smoked" in a gas flame and allowed to cool to room temperature. As the pipette cools, the difference in coefficient of expansion of the platinum wire and pyrex glass causes the platinum wire to shrink away from the glass. The space between the wire and the glass will become filled with fiber-like processes of glass, which becomes the air "brake."

The tip is pulled in the ordinary manner, except that bulb B (Fig. 1b) has to be blown from the tip end of the pipette instead of the open end marked X. (The tip may be formed, if desired, before the platinum wire is inserted.)

A pipette made in this manner has a smooth, slow movement of fluid in the tip, even when a maximum of suction is applied by sucking on rubber tubing applied to point X. A slightly faster pipette may be made by using platinum wire 8 mm. long, with a diameter of .026 (#22 gage).

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A New-Type Atomizer for Large-Scale Application of 2,4-D

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If 2,4-D is to be used on a large scale for the eradication of weeds on range land, grainfields, roadsides, and similar situations, a relatively simple and cheap means of application is necessary. Water sprays require 120 gallons of water per acre when used at the rate of 1 to 1,000. Because of the lack of ample water supplies and because of the weight of the material and equipment required for their distribution, water sprays offer difficulties in any extensive operation of weed control. However, by employing the principle of atomization using concentrated oils, oil emulsions, or water solutions of 2,4-D, application can be made at as low a rate as 3 gallons per acre or less. Atomization of the solution is accomplished by passing air at low pressure (2 pounds) over the solution to be atomized. It is broken up into finely divided particles which form a mist.

The most important feature of the apparatus is the atomizing nozzle, a pattern for which was furnished by the Shell Oil Company of New York City through its representative, C. S. Harris. The nozzle is of brass, 1 $\frac{11}{16}$ inches long, $\frac{7}{8}$ inch wide, and

[2] Transparent sealing resin (see *Chemistry and physics hand book* (23rd ed.), p. 2002.

1¾ inches deep, with separate openings on one edge for an air stream and for the spray material. A chamber inside the nozzle is so constructed that the stream of air flows over a well of material and is blown out from the opposite side in a flat, fan-shaped spray in an arc of about 40°. The openings for the ejection of the spray are on the edge of the nozzle opposite to the openings for entry.

The nozzles are connected on a spray boom 12 inches apart in 6-foot sections. The support for the boom is furnished by the air line, which is a steel pipe ½ inch in diameter, and by an oil line ⅜ inch in diameter. The oil line is under slight pressure (1–5 pounds), and the air line delivers 2 pounds. The delivery rate of the nozzle is easily adjusted by controlling the feed of the solution with a valve. Adjustments can be made to distribute as little as 1 pint over an acre.

A ¼-horsepower gasoline engine, to operate a small air compressor, can be placed on a ½-ton truck.

The apparatus here described makes feasible the return of range land to a grass dominance, which, due to overgrazing, has been shifted to other plants. Also, on roadsides, along fence rows, and on right-of-way of such utilities as power companies and railways, the dominance may be changed to grass by the elimination of woody plants, climbers, and tall, undesired vegetation.

Detection of Serum Agglutinins for *Monilia* and Other Yeast-like Organisms

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In the detection of serum antibodies for *Monilia* (*Candida*) *albicans* and other yeast-like organisms, the agglutination test is not widely used in clinical laboratories. When the agglutination reactions of these organisms have been studied in man and experimentally immunized animals, discrepancies in the results have cast doubt on the value of the procedure for diagnosis. Since in most of the reports the method employed has not been given in detail, this difficulty may be due in part to differences in technique. Another objection has been the reported frequency of spontaneous agglutination of the antigen (*3, 5, 6*). Consequently, there is need for a standardized, reliable, agglutination procedure.

For some time we have employed an agglutination technique which differs only slightly from the usual methods of determining bacterial agglutinins and which in our hands has proved consistently reliable.

The *Monilia* or other yeast-like organism to be used as an antigen is heavily inoculated on Sabouraud's glucose agar, which is incubated at room temperature (approximately 20°C.) for not more than 24 hours. At the end of this period there is usually sufficient growth to make a heavy suspension in physiological salt solution. The organisms, suspended as an antigen, are then killed by heat at 60°C. for 1 hour.

The concentration by volume of the original suspension is determined by centrifugation in a graduated tube, and the suspension is then diluted with physiological salt solution to the optimal concentration.

Serial dilutions of the test serum, previously inactivated at 56°C. for 30 minutes, are made up, and equal parts of the antigen suspension are added to each tube. For each group of tests, known negative and positive sera are set up as controls in the lowest dilution to be employed in the tests. In addition, we customarily use a saline control.

The tubes are incubated at 56°C. for 2 hours, after which they are placed in the refrigerator overnight and then read. For this purpose individual tubes are held in a nearly horizontal position over the concave surface of a microscope substage mirror placed on a table top before a window or a strong light. The tube is tapped gently with the finger to agitate the contained particles. The images of the particles reflected in the mirror are slightly magnified and can be readily observed.

In a negative test there is a uniform suspension of small, translucent, gray particles which are not agglutinations since they have one or more buds.

In a positive test complete agglutination is shown by a dense, opaque, white, flocculent precipitate which is broken up into large, irregular clumps on agitation. This reaction is considered to be 4+; 3+ is indicated when a flocculent precipitate breaks up into smaller and more uniform coarse white particles; a sediment of easily suspended, large white granules is called 2+; a suspension slightly larger and more opaque than the negative controls is called 1+.

The optimal concentration of the antigen is determined by titrating serial dilutions of an antigen suspension against serial dilutions of an immune serum. Immune serum is prepared by injecting rabbits intravenously with heat-killed, 24-hour saline suspensions of organisms by the method of Benham (*1*).

The use of a 24-hour culture of the yeast-like organism, as employed by Kesten, *et al.* (*4*), is highly important to avoid the presence of mycelia, which in many instances appear to be the cause of spontaneous agglutination.

By employing optimal concentrations of antigen, agglutinin titers are more apt to be constant and, presumably, maximal.

The use of the substage microscope mirror for reading the tests eliminates the uncertainty of end-points and requires much less time than do microscopic readings.

In repeated microscopic checks of our macroscopic readings, we have found that, in 1 +, there are clumps of 10–100 organisms; in 2 +, clumps average about one-third the diameter of an oil immersion field; and in 3 and 4 +, there are gross clumps.

In routine tests of human and rabbit sera there has been no evidence of spontaneous agglutination, and zone reactions have not occurred when previously inactivated sera were used (*2*).

The titers of antibodies in the same individual, when repeated at short intervals, are constant, and the majority of different strains of a species are agglutinated in the same titer by an individual serum. Among different strains of any one species, variations greater than one tube in titer have not been observed.

The method has been found to be reliable in the study of several species of *Monilia* and *Saccharomyces* and in the study of *Willia anomala*. It has not been found reliable in the study of organisms from which mycelial filaments cannot be eliminated.

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