

observing the thryxotropy. The crystals stain with methyl violet and are fairly soluble in water. Their dilute solution is yellow. Their concentrated solution is brown and gives an absorption band in the red at 627 m μ and a fainter band in the green at 536 m μ .

Catalase can be recrystallized easily by dissolving the crystals in dilute phosphate buffer of pH 7.3, bringing the pH of the solution to approximately pH 5.4 through the addition of acid potassium phosphate and then adding ammonium sulfate slowly with cooling. The crystals form very rapidly.

One sample of twice recrystallized catalase, after dissolving in phosphate buffer and dialysing until free

from ammonium sulfate, was found to possess a "Kat. f" of 43,000 and an iron content of approximately 0.10 per cent. Crystalline catalase coagulates upon heating and gives many of the usual protein tests. A strong odor of burnt hair is produced on ashing. The pyridine hemochromogen test is readily obtained.

The properties of our crystalline catalase are in complete agreement with the properties of the catalase preparations of von Euler and Josephson,² and Zeile and Hellström.³

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

THE USE OF SYNTHETIC RESINS IN THE PREPARATION OF PERMANENT BACTERIAL MOUNTS

THE most common method employed in making permanent preparations of bacterial mounts entails the use of Canada balsam, a naturally occurring resinous substance. It serves as a cementing agent and, when spread upon a glass slide, seals the thin cover slip placed over the mount to protect and preserve the latter for subsequent observation. When permanent mounts are prepared in this manner, several days may be required before the solvent has completely evaporated, leaving a hard cement. In recent years, several synthetic resins have been prepared commercially, solutions of which harden rapidly on exposure to air, giving a hard, clear, colorless layer possessing a refractive index very close to that of glass. Coatings prepared by these resins adhere tenaciously to glass surfaces.

In determining the applicability of these resins, bacterial smears were made on glass slides and stained with dyes commonly used, including crystal violet, carbol fuchsin, methylene blue and the Gram stain. Preliminary investigations indicated that butyl acetate, free of acetic acid, was the most desirable of the organic solvents used. With one resin ("Pontalite"), xylol was substituted for butyl acetate with success. The solutions employed were between 15 and 20 per cent.

Mounts were made in two ways. First, solutions of the synthetic resins were substituted for Canada balsam as the cementing medium. The resin was used in exactly the same manner as the balsam, that is, a drop or two of the solution was placed upon the smear and a glass cover slip placed over it, care being exercised not to include air bubbles. The cover glass was pressed down lightly and the preparation ready for use after ten minutes of air drying.

Secondly, a solution of the resin was applied by tilting the glass slide bearing the mount, lengthwise, at a 45° angle, flooding by means of a dropping pipette and permitting the excess solution to drain off, thus leaving a thin, smooth, glass-like layer of uniform thickness. This may air dry for thirty minutes or more, before being used for observation or, if required at once, drying can be forced by baking the slide at 135° C. for five minutes with no apparent damage to the mount. No cover slip is used, the thin film of resin serving in its stead. When observing mounts prepared in this manner with the aid of the oil immersion objective, xylol can not be used to remove the cedar oil from the slide because of its solvent effect on the resin. The oil can be removed by washing off with ligroin, gasoline or mechanically by merely wiping off with lens paper. Mineral oil, which is often employed as the immersion medium, is much easier to remove from the slides than is cedar oil. If used a great deal, the thin resin layer will eventually become scratched. These scratches can be erased by covering with another film of the resin.

Slides were prepared in February, 1936, in the manner indicated above, using two commercial resins.¹ Each slide was divided into four portions consisting of mounts covered with (a) Canada balsam and a cover slip, (b) the synthetic resin and a cover slip, (c) the synthetic resin alone and (d) an uncovered portion. Over an eleven-month period there has been no noticeable change, such as fading due to the solvent action of the butyl acetate, in any of the covered preparations. Those under the synthetic resin appear as stable as those preserved under Canada balsam.

² H. von Euler and K. Josephson, *Liebig's Ann.*, 452: 158, 1927.

³ K. Zeile and H. Hellström, *Zeit. physiol. Chem.*, 192: 171, 1930.

¹ "Vinylite" (Series A Resin), Carbide and Carbon Chemicals Corporation, New York; "Pontalite," du Pont de Nemours and Company, Wilmington, Del.

Solutions of these resins were also used in making permanent mounts of moulds. The slide was first covered with a uniform layer of the resin in precisely the same manner as described above. The film was then air dried for two to three minutes, at the end of which time it still presented a slightly sticky surface. This surface was then impregnated with the fungus in one of two ways. Either the resin side of the slide was laid gently on the colony, removed and air dried, or a portion of the colony was "fished out" with a platinum loop and these fragments placed on the partially solidified resinous layer, allowing the latter to air dry. Then, employing the methods of the mycologist, the organism was fixed, using any solution which has as a solvent, water, *e.g.*, mercuric chloride—formaldehyde solution. A fixing agent having an organic solvent can not be used because of its effect upon the resin. The preparation was then stained, applying any of the dyes used in aqueous solution, such as safranin, erythrosin or fuchsin.

It is believed that the application of the synthetic resin is superior to the use of Canada balsam from the standpoint of ease of manipulation, simplicity, rapidity and cost.

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A PRACTICAL DEVICE FOR THE RAPID QUANTITATIVE DETERMINATION OF PLANT PIGMENTS

If the wave-length of the light employed in measuring the absorption of light by a solution is restricted to one of the absorption bands of the solution, the specific transmissive index, symbolized by k , is expressed closely by the equation,

$$k = -\log_{10} T,$$

in which T is the transmittancy.

If, for practical reasons, the wave-lengths of the light employed can not be restricted to a single absorption band, the relation between the light absorbed and the concentration can not be expressed in so simple a manner. The relative transmission can, however, be plotted against the relative concentration; and the concentration of an unknown solution can be determined from the known relative transmission. Evidently, the ideal conditions should be approached as nearly as possible.

In the device we employed, a filter was used, that permitted the passage of only the light having wave-lengths ranging from 4,000 to 5,000 Angstrom units. All three plant pigments have absorption bands in this region. The filter was very dense, so that it was necessary to use a powerful light source.

A standard projection lantern having a 500-watt lamp was used. The condenser lenses and the projection lens were set so that the beam of light that fell upon the absorption cell containing the solution was plane parallel. The absorption cell was the kind used in spectrometry. It was in the form of a parallelepipedon and was closed with a glass stopper. Two such cells were mounted in such a manner that the one could quickly be interchanged for the other. The transmitted light was registered by means of a microammeter which recorded the current produced by a photronic cell after being excited by the transmitted light. When the two absorption cells were filled with water, they registered equally 50 arbitrary units when they stood in the same relative position with respect to the optical system and the photo-electric cell.

In practice, one cell was filled with water and the other with the solution to be studied. By means of a shutter device, the light fell upon the absorption cell for only a short time while a test was being made. The light source was kept constant by properly balancing the electrical system. The water reading was made before and after each solution reading. When the solution reading was multiplied by 2 the percentage transmission was obtained when referred to water, since the water was 50.

Standard solutions were prepared for all three plant pigments, using the pure chlorophyll, xanthophyll and carotene. The solutions ranged in intervals of 2.5 per cent. from 0 to 10 per cent., and in 5 per cent. intervals from 10 per cent. to 100 per cent. The 100 per cent. chlorophyll solution represented 5 milligrams per 100 cc of solution, while the 100 per cent. for the other two pigments represented 0.5 milligrams per 100 cc of solution. The values obtained were plotted against the known concentration, and a graph for each pigment was drawn. From the graphs, tables were made that made it possible to read quickly the concentration of any unknown solution from the relative per cent. transmission. Care was taken that all parts of the instrument were constantly in the same position.

The probable error was calculated and was found to be less than 2 per cent.

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BOOKS RECEIVED

- Commercial Shipyards and the Navy.* Pp. 105. National Council of American Shipbuilders.
DUNLAP, ORRIN E., JR. *Marconi the Man and His Wireless.* Pp. xxi + 360. 16 plates. Macmillan. \$3.50.
HILL, DOUGLAS G. and others. *Elementary Chemistry.* Pp. vi + 473. 68 figures. Henry Holt. \$2.80.
SIDGWICK, N. V. *The Organic Chemistry of Nitrogen.* Second edition, revised by T. W. J. TAYLOR and WILSON BAKER. Pp. xix + 590. Oxford University Press. \$8.50.
STRONG, EDWARD W. *Procedures and Metaphysics.* Pp. vii + 301. University of California Press. \$2.50.