thiamin in wheat grains,[1, 2, 3, 4] we believe that the fluorescence of treated grains indicates the distribution of thiamin and cocarboxylase in the grain.

By using another set of filters we have obtained fluorescence of a different color which we think is probably riboflavin or some product produced from it by the alkaline ferricyanide solution and, possibly, light. For this purpose we have used Corning filters nos. 511 and 038 on the light source and no. 349 for viewing the fluorescence. This fluorescence does not occur in untreated grains, but probably the concentrated alkali liberates the riboflavin from combination with protein and makes fluorescence possible. Possibly the riboflavin (or some nucleotide of riboflavin) is converted into a more strongly fluorescent compound by the treatemnt. The filter combination used shows the fluorescence of pure riboflavin, both before and after treatment similar to that given the grains. It does not show the fluorescence of oxidized, or unoxidized, thiamin or cocarboxylase. If our interpretation is correct, the results show that the embryonic plant, the scutellum and the aleurone layer are all about equal in riboflavin content. The outer bran layers appear to contain some riboflavin. The endosperm cells, apart from the aleurone cells, appear to contain little or no riboflavin.

Using the above techniques we have observed a fluorescence that indicates a relatively high concentration of thiamin (and/or cocarboxylase) and riboflavin in a region of the grain that hitherto has not been reported. At the base of the "crease" of a wheat grain is a layer one or more cells in thickness just inside the aleurone layer. The cells in this region resemble somewhat the aleurone cells and are continuous with them, but they are larger and more circular in outline. The walls of these cells apparently are rich in thiamin and riboflavin.

The treated grains may be kept for weeks without any apparent change in fluorescence. It is easy to make a photographic record of the fluorescence without using an excessively intense light source. For this purpose we have used a General Electric S–4 lamp, 100 watt, with a large, light-crown glass lens to concentrate the light and a photomicrographic camera with microtessar lenses.

We believe the method described above may prove useful in various studies on the role and distribution of these vitamins. For example, it may prove useful as an aid in the selection of wheat strains high in thiamin and riboflavin. The aim of such selection is twofold. One aim is to produce wheat which has a greater vitamin content. The other aim is to select wheat in which these vitamins are so distributed that they are included in the flour fractions by the ordinary milling procedures. Our method should greatly facilitate the selection of wheat strains which will meet this second aim. Furthermore, once a detailed knowledge is available concerning the distribution of thiamin and riboflavin in wheat, it may be possible to develop milling procedures which will produce flour rich in thiamin and riboflavin and still satisfactory in other ways.

G. FRED SOMERS
MARY H. COOLIDGE

U. S. PLANT, SOIL AND NUTRITION LABORATORY,
ITHACA, N. Y.

A CYLINDER GUIDE FOR USE IN PLATE ASSAY OF PENICILLIN

FOR several months in carrying out the cylinder plate method of assay of penicillin, this laboratory has been using a template or guide to facilitate placing the cylinders on the agar in the desired array. The interest in this device shown by visitors has prompted this brief description of its construction and the manner in which it is used.

The guide is made of Plexiglas but may be made of any similar clear plastic. In form it is simply a circular flanged lid through which six holes are drilled and is constructed to fit into a petri dish, the upper portion or cover of which is 100 mm in diameter and 20 mm high. Fig. 1 shows the specifications for the guide as used in this laboratory.

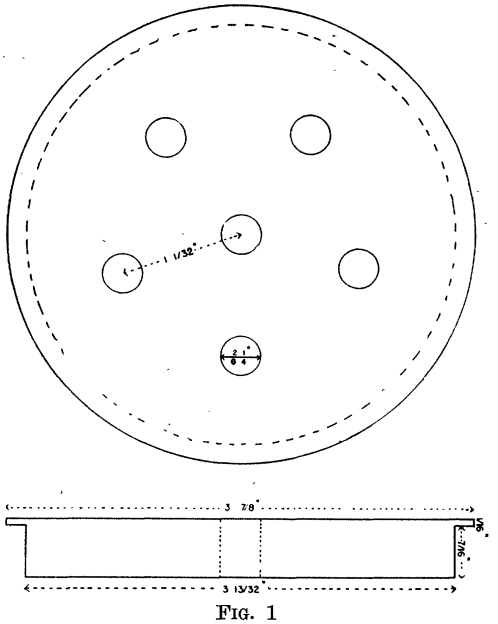

FIG. 1

The Plexiglas was obtained in the form of sheets one foot square and one-half inch thick. In a piece four inches square, cut from the large sheet, the centers for the six holes were marked off and drilled with a 3/16 inch drill. A flat board was placed in

the lathe and centered and to this board the Plexiglas was held by wood screws inserted through the holes previously bored. The Plexiglas was then turned on the lathe to the proper dimensions for the outside diameter and undercut and the holes enlarged to the dimensions given in Fig. 1.

While glass cylinders have been and are to some extent still used, most of the cylinders now being used in this laboratory are made of high-strength aluminum alloy tubing, No. 24ST, outside diameter 5/16 inch, wall thickness 0.035 inch, inside diameter 0.242 inch. This tubing is cut in 1 cm lengths, beveled at one end on the outside and the beveled end ground smooth. It has recently come to our attention that stainless steel cylinders are now available. These should prove to be more durable than aluminum.

The arrangement of the six apertures, which allow free passage of the cylinders and at the same time direct them to positions properly spaced, permits the latitude desired in setting up various forms of the assay. In one type of assay procedure used in this laboratory five cylinders are filled on each plate, using four plates for each assay. On each plate two cylinders are filled with standard penicillin diluted to one unit per cc with 1 per cent. phosphate buffer and to three cups is added the unknown solution diluted with phosphate buffer to approximately 1 unit per cc. The ampuls or vials submitted to this laboratory are quite consistently labeled as containing 100,000 units. A primary dilution to 40 units per cc is made with pyrogen-free distilled water, after which further dilution is made with phosphate buffer to the 1 unit per cc level. In this procedure the potency is calculated using a standard curve. Numerous assays have been conducted in which the potency is interpreted statistically.[1] In the latter method four cups are utilized, two dose levels of standard and unknown being placed on each plate. When the guide is used in this procedure one of the periphery cups is simply omitted.

When large numbers of assays are to be calculated, some time may elapse before the results can be tabulated. In order to obtain a potency figure while the statistical results are being calculated, it has been the practice to use five cups on each plate in setting up the assay. Two dose levels, one at 0.25 unit per cc and another at 1 unit per cc are employed and in the fifth cup is placed an additional unknown at a concentration level of 1 unit per cc. In a four-plate assay this gives four standards and eight unknowns. The potency can be calculated by using a standard curve and the value corrected, if necessary, when the result of the statistical analysis is available.

The center aperture is seldom used, but there are occasions which call for the assay of crude extracts, filtrates, etc., when a rough estimate of activity suffices. In such instances a one-cup assay may be run, permitting five assays on one plate with the standard in the center cup.

The use of the guide offers several advantages. It is simple to make, easy to clean and virtually indestructible. The cylinders fall the same distance onto the agar, insuring a good seal between the agar and all cups and providing for accurate and uniform spacing on all the plates. The guide should prove helpful for inexperienced operators who often have difficulty in evenly spacing the cylinders and who may drop them in such manner that they fail to remain upright. Assayists who have had nearly a year's experience in testing penicillin have found that the guide expedites placing the cylinders in position and that the operation requires less concentration on the part of the assayist.

[1] The statistical method used in calculating the potency and its error has been submitted for publication.

SUMMARY

A guide or template is described which facilitates placing the cylinders on the agar surface in the plate assay of penicillin.

The general plan of conducting the assay is discussed and several advantages of the use of the guide are noted.

We wish to thank Mr. Albert G. Sterling, instrument maker, for fabricating the guide and the aluminum cylinders.

ELIZABETH J. OSWALD
WILLIAM A. RANDALL

FOOD AND DRUG ADMINISTRATION,
WASHINGTON, D. C.

BOOKS RECEIVED

ANDREWS, EDMUND and IRENE D. ANDREWS. *A Comparative Dictionary of the Tahitian Language.* Pp. xvi + 253. The Chicago Academy of Sciences. 1944.

BLOCK, RICHARD J. and DIANA BOLLING. *The Amino Acid Composition of Proteins and Foods.* Illustrated. Pp. xiv + 396. Charles C Thomas. $6.50. 1945.

JORDAN, PASCUAL. *Physics of the Twentieth Century.* Pp. xii + 185. Philosophical Library. 1944.

KROEBER, A. L. *Configurations of Culture Growth.* Pp. x + 882. University of California Press. $7.50. 1944.

LANDE, ALFRED. *The Physics of Flight.* Illustrated. Pp. vi + 122. Reinhold Publishing Corporation. $2.50. 1945.

LONGYEAR, JOHN M. *Archaeological Investigations in El Salvador.* Illustrated. Pp. xi + 90. Peabody Museum of Archaeology and Ethnology. $3.75. 1944.

MANTELL, C. L. *Adsorption.* Illustrated. Pp. viii + 386. McGraw-Hill Book Company. $4.50. 1945.

MORRIS, C. L. "LES." *Pioneering the Helicopter.* Illustrated. Pp. xiv + 161. $2.75. 1944.

ROWE, JOHN H. *An Introduction to the Archaeology of Cuzco.* (Research project No. 7 of the Institute of Andean Research under the sponsorship of the coordinator of Inter-American Affairs.) Illustrated. Pp. xii + 70. Peabody Museum of Archaeology and Ethnology. $2.50. 1944.

UMBREIT, W. W. and OTHERS. *Manometric Techniques and Related Methods for the Study of Tissue Metabolism.* Illustrated. Pp. 198. Burgess Publishing Company. $3.50. 1945.