of the plants were covered with tar paper, and uncovered next morning between 7:00 and 8:00 A.M. The first ripe fruits were harvested from the covered plants on April 1, 1944, and from then on they continued to produce. The non-covered plants did not produce fruits at all.

A more extensive experiment was started on March 17, 1944. Eight plots of 9′ × 9′ were each planted to tomatoes, beets and a few other vegetables. Two plots served as controls, and were not covered, but the other plots were covered at different hours in the afternoon. On July 6, after an exceptionally cool spring, the plants were harvested. Some of the results are shown in Table 1 (fresh weight in g. per plant).

TABLE 1

	Stone tomato		Earliana tomato		Beets	
	Whole plant	Fruit only	Whole plant	Fruit only	Whole plant	Root only
Control, not covered	304	10	420	37	126	61
Covered 2 P.M.–8 A.M.	433	0	545	7	62	9
Covered 3 P.M.–8 A.M.	1264	256	1054	272	122	37
Covered 4 P.M.–8 A.M.	719	101	677	121	149	67

From these data it appears that the development of tomato plants is not limited by photosynthesis, but that the use of the photosynthates is regulated by processes occurring in darkness at temperatures between 15° and 20°. This is not true for all plants, since the covering of beets produced only a slight and insignificant increase in weight. These experiments show that under certain conditions by the proper treatment the apparent efficiency of photosynthesis can be increased considerably in tomatoes.

The effects of the covering in these experiments were not due to photoperiodicity, but to thermoperiodicity.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

LOCATION OF THIAMIN AND RIBOFLAVIN IN WHEAT GRAINS

WE were interested in determining the distribution of thiamin in wheat grains in conjunction with analytical data on the thiamin content of various hybrid and inbred selections of wheat. We have developed a simple and rapid method that apparently gives results which are, in general, consistent with the milling data (see Bailey[1] and Andrews[2] for reviews) and with dissection studies previously reported.[3, 4]

Briefly, the technique is as follows: The wheat grains are dropped into molten paraffin. After the paraffin has hardened, the grain is cut on a rotary microtome. No attempt is made to cut entire sections; rather, 20-micron slices are cut from the grain until approximately the desired region is reached. Then one-micron slices are cut. This latter step gives a portion of a grain with a smooth surface. The cut surface of the grain is next treated with an alkaline solution of potassium ferricyanide similar to that used in oxidizing thiamin to thiochrome for fluorometric assay of extracts. The grains may be treated directly while still mounted in the paraffin, or the paraffin can be removed, using anhydrous benzene, if desired, and the grain mounted in modeling clay. The ferricyanide is applied by gently pressing the cut surface of the grain against a piece of Cellophane that has been soaked in the ferricyanide solution. Before the Cellophane is used, the excess moisture is blotted from it with filter paper.

The treated surface of the grain is then illuminated with a mercury vapor lamp using a suitable filter or filters. Various filters have been tried, but for thiochrome we have used mostly Corning filter no. 597. The fluorescence of the illuminated grain can be observed directly, with or without additional filters. We have found that viewing the fluorescence of the grain through a combination consisting of Corning filters nos. 338 and 428 gives the best results for thiochrome. Filters more specific for thiochrome fluorescence have been used. They show essentially the same picture. Cocarboxylase has been found to fluoresce the same way as thiamin after oxidation with the alkaline ferricyanide solution.

When treated grains are viewed using such combinations of filters, it appears that the cell walls of the aleurone layer fluoresce brightly. This is particularly true of the inner cell walls of the aleurone layer. Likewise, the scutellum fluoresces, but not as brightly as the epithelium and/or the adjacent crushed cells of the endosperm. The embryo, exclusive of the scutellum, shows little or no fluorescence. The endosperm shows some fluorescence, but this is apparently restricted to the walls of the endosperm cells. In fact, in all cases where cell structure can be made out, the fluorescence appears to be primarily in the cell walls. The untreated grains show no fluorescence except for a very weak fluorescence in the walls of the aleurone cells. In view of the previously reported distribution of

[1] C. H. Bailey, "Constituents of Wheat and Wheat Products," pp. 280–317, Reinhold, 1944.
[2] J. S. Andrews, *Food Industries*, 15: 78, August, 1943.
[3] J. J. C. Hinton, *Jour. Soc. Chem. Ind.*, 61: 143, 1942.
[4] A. H. Ward, *Chem. Ind.*, 62: 11, 1943.

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.

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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.

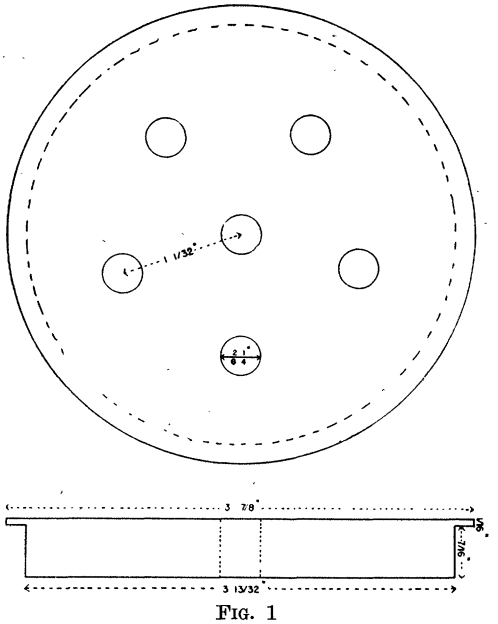

FIG. 1

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