embryonated eggs, possibly in the presence of living tumor cells.

In a second group of experiments, further studies were made on the survival of the milk influence in embryonated eggs. These experiments were performed with a filtrate made from lactating mammary tissue of mice which carried the active agent. Lactating mammary tissue was macerated, suspended in broth (1:10) and centrifuged. The supernatant liquid was then filtered through a tested Berkefeld N filter, was tested for the presence of the active agent, and used to inoculate chick embryos. The test to ascertain whether the filtrate contained the active milk agent was performed by injecting 14 mice with 1 ml of the material intraperitoneally. Mammary tumors have resulted in 6 of the test mice, demonstrating the presence of the active agent. The remaining mice are alive and free from tumors at 13 months of age.

Eggs containing 5-day-old chick embryos were injected with 0.25 ml of the filtrate of mammary tissue. Two eggs were incubated for 1 hour, after which the yolks were collected and pooled, and 1 ml of the unfiltered yolk was injected intraperitoneally into each of 8 mice. Of these, 5 have developed spontaneous mammary tumors while the others are still under observation.

Other eggs of 5 days' embryonation that had received the filtrate were incubated for 12 days, after which time the yolks were pooled. Part of the yolk was centrifuged undiluted at low speed. Of 16 mice that received 1 ml of the unfiltered yolk intraperitoneally, 6 have developed mammary cancer while the others are still under observation. The balance of the yolk from eggs was extracted with Locke's solution (1:3) and centrifuged. The supernatant liquid was filtered through a Berkefeld V filter. One milliliter of the filtrate was injected intraperitoneally into each of 18 mice. Of these, 6 have shown spontaneous mammary tumors and the 12 others are living without growths at 13 months of age.

In the second group of experiments, a filtrate of mammary tissue, proved to contain the active tumor milk agent, did not produce grossly demonstrable tumors in 5-day embryonated eggs after 12 days of incubation. However, both unfiltered and filtered egg yolks, after 12 days' incubation, were found to contain the active mammary tumor milk agent. Therefore, our results, although adduced from a small number of mice, are interpreted to mean that the milk agent survived 12 days in the yolk sac in the absence of living mouse cells.

SUMMARY

The mammary tumor milk agent has been recovered from a transplanted mammary carcinoma that was carried for 10 passages in mice that did not themselves originally have the agent. It is possible that the agent, carried in the transplanted mammary tumor cells, may be responsible for the genetic mutations which have been detected in transplantable tumors.

Preliminary results show that the agent can be recovered from the yolk sac of chick embryos 12 days after the injection into eggs of either tumor suspensions or cell-free filtrates of tissues containing the active agent.

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SIMULATION OF PHOTOPERIODICITY BY THERMOPERIODICITY

A TOMATO plant is photoperiodically indifferent, since under properly controlled temperatures the daily length of illumination has practically no effect on its development. For best growth and fruit set, tomatoes have to be kept warm during day (26° C) and cool during night (15–18° C), which has been called thermoperiodicity.[1] The cool period for optimal development is only effective in darkness or in at least greatly reduced light, so that plants subjected to the proper temperature sequence in continuous light do not set fruit. Since no fruit set is possible above 22° and below 10° night temperature, tomatoes do not bear fruit in winter or spring nor during hot spells in summer, even though day temperatures are within the rather wide range of possible growth (15–35° C). It also has been established that each day sugar production by assimilation in tomato leaves continues only until early afternoon, when a maximum sugar content is reached.[2]

In Southern California winter and early spring night temperatures are usually below 10°, but the afternoon temperatures range between 15° and 20°, optimal for growth and fruit set. Therefore, if part of the afternoon were changed into a functional night, by daily covering tomatoes from 3:00 P.M. (war time) on, no loss of photosynthesis would occur, and an optimal night temperature would exist for a few hours.

To find out whether by these means tomato plants, growing outside, could be made to produce fruits out of season, some were planted in the field in the middle of November, 1943. Each afternoon at 3:00 P.M. half

[1] F. W. Went, *Am. Jour. Bot.*, 31: 135–150, 1944.
[2] F. W. Went, *Am. Jour. Bot.*, 31: in press.

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.