and serine.[3] It is apparent that the cells are unable to form protein from these amino acids. The system is thus unique in that it demonstrates protein synthesis to be a more sensitive reaction than photosynthesis. Unfortunately, another physiological malfunction is the rapid decrease in the rate of photosynthesis with the age of the preparation, without visible sign of deterioration. The loss in the ability to assimilate $C^{14}O_2$ is essentially logarithmic, only 20% of the original rate remaining after 1 hr at 23° C (in weak light prior to stronger illumination). It is, of course, possible that the same factors operating in negating protein synthesis are involved in the degradation of the photosynthetic system.

The preparation is intriguing as a possibility for the study of the nutrition and function of a specific type of tissue in leaves, akin to that of a highly specific organ in an animal.

Reference

1. WYLIE, R. D. *Am. J. Botany*, **30**, 273 (1943).

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[3] These are also the only amino acids formed by an intact leaf within the same time period. The fact is that even without the simultaneous formation of all the essential amino acids, protein synthesis (probably partial turnover) occurs in excised leaves. This question will be discussed in a subsequent publication.

Fructomaltose, a Recently Discovered Trisaccharide Isolated from Honeydew

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A trisaccharide, fructomaltose, has been isolated from the honeydew produced by the citrus mealy bug, *Pseudococcus citri* (Risso), while feeding on the sap of etiolated Irish potato sprouts. Fructomaltose was not present in the potato sprout sap. The trisaccharide has also been located chromatographically in the honeydews of the cottony maple scale, *Pulvinaria vitis* (L.), and the spirea aphid, *Aphis spiraecola* Patch, and in honeybee honey. Fructomaltose has been located in the excreta of the black blowfly, *Phormia regina* (Meigen), when it was fed a sucrose solution.

The carbohydrates of honeydew were separated by descending paper partition chromatography, using Whatman No. 1 filter paper and a *n*-butanol, ethanol, acetone, and water (5–4–3–2 v/v) solvent. Benzidine-trichloroacetic acid spray (*1*) was used to locate the carbohydrates which were fructose, glucose, sucrose, fructomaltose, and glucose-1-phosphate. A charcoal-infusorial earth column chromatogram (*2*) was used to isolate the fructomaltose from the other carbohydrates of honeydew.

Fructomaltose is nonreducing to Benedict's copper sulfate solution, chars at 118–124° C but does not melt, is dextrorotatory, and apparently is very hygroscopic. A satisfactory rotation value has not been obtained and the sugar has not been crystallized. Fructomaltose is hydrolyzed to fructose and maltose by either yeast invertase or dilute hydrochloric acid, to fructose, glucose, and sucrose by human saliva, and to glucose and sucrose by pancreatin. Hydrochloric acid eventually hydrolyzes it to fructose and glucose.

TABLE 1

Sample No.	Fructose, μg/mm	Total reducing sugars, μg/mm	Maltose, μg/mm	Weight ratio Fructose/maltose
1	17.5	52.9	34.5	1: 1.97
2	8.2	25.0	16.8	1: 2.05
3	13.2	44.5	30.9	1: 2.27
4	8.0	24.6	16.6	1: 2.07
5	14.0	45.8	31.8	1: 2.27

In order to determine the monosaccharide ratio, fructomaltose was hydrolyzed quantitatively to fructose and maltose by yeast invertase. The sugar-invertase solution was deproteinized with $Ba(OH)_2$ and $ZnSO_4$ (*3*) and the fructose/maltose ratio determined colorimetrically. The fructose was determined by the Roe (*4*) method and the total reducing sugars by the Somogyi (*5*) method. Nelson's (*6*) chromogenic reagent was used in the latter method. The results obtained are shown in Table 1. The data show a fructose/maltose ratio of 1 : 2, by weight; therefore, fructomaltose contains 1 fructose and 2 glucose units.

It is believed that fructomaltose is an intermediate product formed during the action of invertase on sucrose. Sucrose is present in the normal food supply of each of the insects investigated. The presence of invertase in insect digestive systems has been demonstrated by many investigators including Sarin (*7*), Bertholf (*8*), Phillips (*9*), Herford (*10*), and Fraenkel (*11*).

The effects of in e on sucrose have been demonstrated by Blanch nd Albon (*12*), Bacon and Edelman (*13*), A[illegible] and Bacon (*14*), and White and Maher (*15, 16*). These investigators have shown that intermediate products such as trisaccharides, and even tetrasaccharides, are formed during the hydrolysis of sucrose by invertase.

White (*15, 16*) treated sucrose with honey invertase and found that a trisaccharide, which he identified as α-maltosyl-β-D-fructoside was formed as an intermediate product. It was composed of 1 fructose and 2 glucose units and was hydrolyzed to fructose and maltose by either yeast invertase or dilute hydrochloric acid. Honey invertase degraded it to glucose and sucrose.

Judging from the reported reactions of fructomaltose and maltosyl fructoside, it appears that the two sugars may be identical, but this cannot be determined definitely until further studies are made. It is of interest to note that fructomaltose arises as a natural product in the digestive systems of many insects, whereas maltosyl fructoside is a product of an *in vitro* reaction. Present studies indicate that fructomaltose may be expected to arise in the digestive system of any

animal that possesses invertase and utilizes sucrose in its diet.

References

1. BACON, J. S. D., and EDELMAN, J. *Biochem. J.*, **48**, 114 (1951).
2. WHISTLER, R. L., and DURSO, D. F. *J. Am. Chem. Soc.*, **72**, 677 (1950).
3. SOMOGYI, M. *J. Biol. Chem.*, **160**, 69 (1945).
4. ROE, J. H. *Ibid.*, **107**, 15 (1934).
5. SOMOGYI, M. *Ibid.*, **195**, 19 (1952).
6. NELSON, N. *Ibid.*, **153**, 375 (1944).
7. SARIN, E. *Biochem. Z.*, **120**, 250 (1921).
8. BERTHOLD, L. M. *J. Agr. Research*, **35**, 429 (1927).
9. PHILLIPS, E. F. *Ibid.*, **35**, 385 (1927).
10. HERFORD, G. V. B. *Ann. Appl. Biol.*, **22**, 301 (1935).
11. FRAENKEL, G. *J. Exptl. Biol.*, **17**, 18 (1940).
12. BLANCHARD, P. H., and ALBON, N. *Arch. Biochem.*, **29**, 220 (1950).
13. BACON, J. S. D., and EDELMAN, J. *Ibid.*, **28**, 467 (1950).
14. ARONOFF, S., and BACON, J. S. D. *Arch. Biochem. and Biophys.*, **41**, 476 (1952).
15. WHITE, J. W., JR., and MAHER, J. *Ibid.*, **42**, 360 (1953).
16. ———. *J. Am. Chem. Soc.*, **73**, 1259 (1953).

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Malignant Tumors Resulting from Embedding Plastics in Rodents[1]

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In two previous communications (*1*, *2*) we have described various types of sarcomas which were induced in rats and mice by embedding certain plastic films in the anterior abdominal wall just ventral to the fascia. The initial observations were made on rats in which one kidney had been wrapped in cellophane to produce hypertension. Seven of these rats, autopsied after nearly 2 yr, were found to have developed sarcomas around the wrapped kidney. Later experiments showed that subcutaneous embedding produced similar results and the abdominal wall technique is now generally used by us.

In addition to cellophane (regenerated cellulose) we have embedded a number of other plastics and have produced malignant tumors in a considerable percentage of the animals. These are all long-term experiments lasting usually 1–2 yr before the appearance, if at all, of a sarcoma. The final results of some of these experiments cannot be reported as yet, since in many cases the time elapsed after embedding has not been sufficient for the appearance of tumors.

Nevertheless there are practical reasons for publishing further results now, as plastics are being used more and more extensively on humans by surgeons and surgical specialists. It is, however, very important to note that so far there is no proven instance in the literature of a malignant tumor induced in man by embedding a plastic. (Paraffinomas are foreign-body reactions, not malignant growths.) On the other hand, oncologists have reminded us that if it takes 1–2 yr for a malignant tumor to appear in a rodent, it may take 10–15 yr for a similar result in a human being.

TABLE 1

TUMORS OBTAINED BY EMBEDDING PLASTICS SUBCUTANEOUSLY

Completed Experiments

Material	Animals	Malignant tumors produced	
		No.	%
Cellophane A	Rats	15/42	35.7
Cellophane A	Mice	8/35	22.8
Cellophane A	Mice (black)	1/22	
Cellophane B	Rats	20/44	45.4
Polyethylene A	Rats	10/80	12.5
Pure polyethylene	Rats	7/38	18.4
Pure polyethylene	Mice	3/29	10.3
Polyvinyl chloride	Rats	17/44	38.6
Glass coverslip	Rats	1/50	

Experiments Still in Progress

Material	Animals	Malignant tumors produced	Animals still alive
Cellophane C	Rats	11	16
Pure polyethylene perforated	Rats	1	30
Pure polyethylene textile	Rats	1	31
Silastic	Rats	12	3
Teflon	Rats	4	15
Nylon	Rats	1[2]	21
Dacron	Rats	3	29
Dacron perforated	Rats	1	30
Polystyrene	Rats	2	22

Malignant tumors, adjacent to or actually surrounding the film, have been produced in rats or mice or both with the following plastics: (1) commercial cellophane film (regenerated cellulose), for convenience called by us Cellophane A; (2) the same cellophane film after it had been subjected to intensive extraction by methyl alcohol, called Cellophane B; (3) the same cellophane subjected first to alcohol and subsequently to benzene extraction, called Cellophane C; (4) polyethylene film, called Polyethylene A; (5) a pure polyethylene film, specially prepared for these experiments; (6) polyvinyl chloride film; (7) silastic, a silicone product; (8) Teflon film; (9) Dacron film; (10) polystyrene film; (11) with nylon film, so far only one tumor, a reticulum cell sarcoma surrounding the nylon, has appeared, 441 days after insertion. Successful transplantation of this tumor was made, producing reticulum cell sarcomas to the second generation. The remaining rats embedded with nylon are still under observation.[2]

To date the highest percentage of positive results (45.4%) was obtained by embedding cellophane B. Up to the present we have obtained a total of at least 126 primary tumors, including those from kidney wrappings, and many successful transplantations.

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[2] Since the above was written 3 more sarcomas have appeared at the site of embedding nylon film.