more Rh-negative individuals might alter present implications.

Although not recorded in the table, intergroup Rh-negative frequencies in Ethiopian troops showed no significant differences.

References

1. WALLER, R. K., and LEVINE, P. *Science,* **100**, 453 (1944).
2. MILLER, E. B., and TAGUCHI, T. *J. Immunol.,* **51**, 227 (1945).
3. CHOWN, B., *et al. Can. J. Research,* **24**, 135 (1946).
4. SCHNEIDER, C. L., and HUGHES, C. H. *Medicine* (Japanese) **4**, 33 (1948).
5. KAKU, M., and NIIMURA, S. *Japan Medicine,* **14**, 3420 (1948).
6. WIENER, A. S. *Blood Groups and Transfusions,* 3rd ed. Springfield, Ill.: Thomas, 297 (1948).

Manuscript received July 7, 1952.

Rooting Lemon Cuttings with Fruits Attached

Louis C. Erickson and Paul DeBach

Divisions of Plant Physiology and Biological Control, University of California Citrus Experiment Station, Riverside

Detached lemon fruits are utilized in many research problems, of both a general and a specialized nature. A major drawback to their use has been the relatively short period during which they would remain turgid and more or less normal. The authors were concerned with prolonging the useful life of lemon fruits for studies of a physiological, biochemical, and entomological nature. A simple solution to the problem seemed to be the production of roots on stems attached to the fruits. Such a technique should not only result in maintaining healthy turgid fruits for long periods under the usual conditions of high humidity but should permit studies involving low relative humidity.

This paper presents the results obtained in an experiment to determine the rooting response of lemon cuttings with fruits attached.

Two hundred and forty-nine medium-sized lemons ranging in color from yellow to light-green were clipped from several Eureka lemon trees on Feb. 7. Stems on the fruits varied from 1 to 2 in. in length, and approximately half of them had one or two leaves attached. The fruits were segregated into three color groups of yellow, silver, and light-green. The silver category is a packing-house designation for yellow fruit which still retains a slight amount of green color, usually at the ends. Each color group was subdivided into cuttings with and without leaves. These groups were further divided into groups to be treated with a rooting preparation (0.2% naphthalenacetic acid on talc, ANA) or left untreated. The cuttings were placed in a rooting bed with sand as a rooting medium and were usually sprinkled two or three times daily during the rooting period.

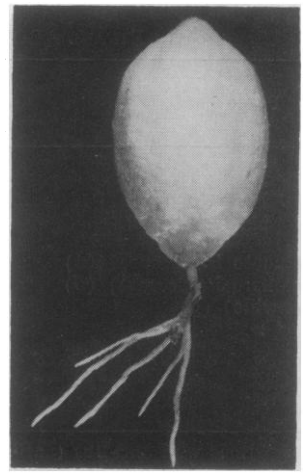

FIG. 1. Rooted lemon cutting without leaf but with light-green lemon attached.

A count of rooted cuttings and roots was made on Mar. 6, 4 weeks after the start of the experiment. The results are presented in Table 1 and Fig. 1. Cuttings with light-green lemons attached rooted most readily, whereas those with yellow lemons rooted least readily. The presence of leaf tissue appeared to be unnecessary in the cuttings with light-green and silver lemons but necessary for root formation in the cuttings with yellow lemons. Naphthalenacetic acid increased the percentage of rooted cuttings in all comparisons.

TABLE 1

ROOTING RESPONSE OF LEMON CUTTINGS

	Light-Green				Silver				Yellow			
	Leaves		No leaves		Leaves		No leaves		Leaves		No leaves	
	No ANA	ANA	No ANA	ANA	No ANA	ANA	No ANA	ANA	No ANA	ANA	No ANA	ANA
No. cuttings	38	37	31	26	21	22	13	12	13	15	10	11
Percentage rooted	31	59	52	81	10	68	15	67	8	67	10	18
Roots per rooted cutting	1.8	3.8	3.3	3.0	1.5	4.4	1.5	2.4	1.0	3.9	5.0	7.5

Leafy lemon cuttings have been reported to root better than leafless ones (*1*), even when treated with a growth regulator such as indolacetic acid (*2*). Cooper (*3*) suggested that the role of indolacetic acid was to mobilize, at the base of the cutting, rhizocaline, a root-forming factor produced in the leaves. On the

other hand, Gregory and van Overbeek (*4*), van Overbeek and Gregory (*5*), and van Overbeek, Gordon, and Gregory (*6*) showed that the leaves of red-flowered hibiscus cuttings could be replaced by a treatment with sucrose and nitrogen, insofar as the number of roots formed was concerned.

In the present study it was found that leaves were not essential for the rooting of leafless cuttings when light-green or silver-colored lemons were attached. It appears, therefore, that immature lemon fruits can supply the same factors as are ordinarily supplied by the leaves. Sugars (*7, 8*) and nitrogen (*9*) are present in both green and yellow lemons. Whether these factors become less available for mobilization to the base of the cutting as the fruit matures or whether other factors for rooting are concerned requires further investigation.

References

1. HALMA, F. F. *Hilgardia,* **6**, 131 (1931).
2. COOPER, W. C. *Plant Physiol.,* **10**, 789 (1935).
3. ———. *Botan. Gaz.,* **99**, 599 (1938).
4. GREGORY, L. E., and VAN OVERBEEK, J. *Proc. Am. Soc. Hort. Sci.,* **46**, 427 (1945).
5. VAN OVERBEEK, J., and GREGORY, L. E. *Am. J. Botany,* **32**, 336 (1945).
6. *Ibid.,* **33**, 100 (1946).
7. BARTHOLOMEW, E. T., and SINCLAIR, W. B. *The Lemon Fruit.* Berkeley: Univ. Calif. Press (1951).
8. SINCLAIR, W. B., and CRANDALL, P. R. *Plant Physiol.,* **24**, 681 (1949).
9. CAMERON, H. S., APPLEMAN, D., and BIALAGLOWSKI, J. *Proc. Am. Soc. Hort. Sci.,* **33**, 87 (1935).

Manuscript received July 8, 1952.

The Stimulation *in Vitro* of Phospholipid Synthesis in Thyroid Tissue by Thyrotrophic Hormone

M. E. Morton and J. R. Schwartz

Radioisotope Unit, Veterans Administration Hospital, Long Beach, California,* and *The School of Medicine, University of California, Los Angeles

The tissue slice technique has proved a most useful tool in investigations of the metabolism of the thyroid gland. Studies in Chaikoff's laboratories (*1, 2*) showed the ability of surviving thyroid slices to synthesize diiodotyrosine and thyroxine from inorganic iodine. With the same technique, the inhibition of thyroxine and diiodotyrosine synthesis by large amounts of iodide (*3*) and the effects of goitrogens (*4*) on the organic binding of iodine were demonstrated.

The present results appeared during the course of a study (*5*) of the possible correlation between phosphorus and iodine metabolism in a similar system of thyroid slices. A striking stimulation of the rate of incorporation of phosphorus into the phospholipids of thyroid tissue was found when thyroid slices were incubated in a medium containing radioactive orthophosphate in the presence of the thyroid-stimulating hormone (thyrotrophin, TSH) of the anterior pituitary gland. No such effect was observed in either the trichloracetic acid soluble or insoluble fractions.

Beef thyroid[1] was used. The procedure of slicing and incubation was as previously described (*1*). A total of 300 mg of tissue slices was incubated in 3.00 ml of Krebs-Ringer bicarbonate medium at pH 7.4, 37° C, under 95% oxygen and 5% CO_2 for a 3-hr period. Approximately 1 μc P^{32} as orthophosphate[2] was used in each beaker. Thyrotrophin,[3] dissolved in the buffered medium, was used in the concentrations noted in Table 1. The analytical procedure used will appear elsewhere (*5*).

The data of Table 1 demonstrate the marked stimulation of thyrotrophin (TSH) on the incorporation of radioactive orthophosphate into the lipid fraction of surviving thyroid slices. An amount of TSH as low as 0.3 J.S.[4] units produced phospholipid synthesis of the order of 181% that of controls. The maximum stimulation observed was 254% of the control, using 6–8 units of TSH (1 mg) in the bath. These findings suggest that this system may well be at least as sensitive as that of Borell and Holmgren (*6*) in the assay of pituitary thyrotrophin.

Dialysis of the protein hormone against 50 volumes of distilled water did not decrease its activity in promoting the incorporation of P^{32} into the lipid fraction. This would seem to rule out any effect from small molecule contaminants such as choline. A further test of this possibility showed that 1 mg choline/3 ml medium had no effect.

The specificity of this action is shown by a study with liver and kidney slices using 3 times as much TSH as was used with thyroid tissue. There was no evidence of any increase in P^{32} incorporation in the

[1] Grateful acknowledgment is made to Don Sherman, of the Alpha Beta Meat Packing Company, Wintersburg, Calif., who made available the beef thyroids used in this study.

[2] The radiophosphorus used in this investigation was supplied by Oak Ridge National Laboratory on authorization from the Isotopes Division, U. S. Atomic Energy Commission.

[3] Grateful acknowledgment is made to Wayne Donaldson, of Parke-Davis & Co., for the thyrotrophin used in this study.

[4] Junkmann-Schoeller.

TABLE 1

EFFECT OF THYROTROPHIN (TSH)* ON UPTAKE OF RADIOPHOSPHATE BY PHOSPHOLIPIDS OF BEEF THYROID SLICES

Expt. No.	S-3-125	S-5-23	S-3-125†	S-3-119	S-3-115	S-3-115	S-3-115
Mg TSH in medium	0.01	0.038	0.1	1.0	5	10	15
% increase P^{32} over control	105 ± 11	181 ± 16‡	180 ± 11	254 ± 12	217 ± 9.9	208 ± 9.6	210 ± 8.4

* In all studies Parke-Davis TSH Rx 099802, estimated to contain 6–8 u/mg, was used.
† Thyrotrophin treated by dialysis against 50 vol distilled water for 48 hr.
‡ σ for $n = 4$; in all other studies $n = 2$.