

The results of analyses of four composited samples of representative fruit from each of fifteen different trees from three experimental plots in Yuma are shown in Fig. 1.

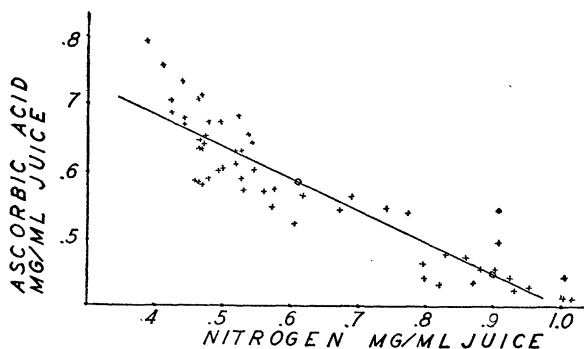


FIG. 1. Regression of ascorbic acid on nitrogen in grapefruit juice.

The data of the figure show a negative correlation coefficient of 0.91 between the nitrogen and ascorbic acid content of the juice of the grapefruit. Just why this relation between nitrogen and ascorbic acid exists in citrus juice is not clear.

As far as we are aware the high values for ascorbic acid are higher than any reported for grapefruit juice. At the same time, however, the nitrogen values are extremely low for those samples that show a high ascorbic acid content.

Perhaps it is associated with the ascorbic acid—oxidase respiratory system as described by Szent-Györgyi.<sup>2</sup>

It would be of interest to know if this relation between ascorbic acid and nitrogen, which has been found in Arizona grapefruit, holds for other citrus-producing areas and for other fruits or vegetables.

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# THIOUREA AND RESISTANCE TO LOW ATMOSPHERIC PRESSURES ("HIGH ALTITUDES")\*

It has been reported that thyroidectomized rats are

<sup>2</sup> A. V. Szent-Györgyi, "On Oxidation, etc." Williams and Wilkins Co., Baltimore, 1939.

\* This investigation was supported by a grant from The Commonwealth Fund.

better able to tolerate exposures to anoxia and reduced atmospheric pressures<sup>1, 2, 3</sup> than normal animals. Administration of thyroid and injections of anterior pituitary substance have been found to increase the sensitivity of animals to oxygen lack.<sup>1, 2, 4, 5</sup>

Mackenzie and Mackenzie<sup>6</sup> and Astwood *et al.*<sup>7</sup> have shown recently that treatment of rats with thiourea and the sulfonamides results in the development of a state of functional hypothyroidism, characterized by an enlarged, hyperplastic thyroid gland and a drop in the B.M.R. Withdrawal of the drugs results in a return of the animal to a normal condition.<sup>7</sup> The administration of thiouracil to the human suffering from hyperthyroidism results in a reduction of the basal metabolic rate.<sup>8, 9</sup> It has been demonstrated that thiourea produces these effects by interfering with the production of normal thyroid hormone by the thyroid gland.<sup>10</sup>

In view of these results it became of interest to test the effects of thiourea upon the resistance of animals to low atmospheric pressures. One hundred and two adult female rats were employed in these experiments. Thirty-six of these were fed a diet containing 0.5 per cent. thiourea for periods of time varying

TABLE 1  
EFFECT OF THIOUREA ON RESISTANCE TO LOW ATMOSPHERIC PRESSURES

Days on thiourea	No. of animals	Percentage mortality	Mean thyroid weight (mg)
0 (Controls) . .	36	75	14.0
0 (Thiourea injections) . . . .	30	60	13.9
4-8 . . . . .	18	50	18.0
12 . . . . .	6	0	29.3
14 . . . . .	6	0	30.0
30 . . . . .	6	0	37.4

from 4 to 30 days. Thirty others were injected with 200 mg of thiourea in distilled water 5 hours prior to initiation of the low pressure treatment. Thirty-six animals served as untreated controls. All animals were then exposed to pressures of 200 mm Hg (32,000 feet), for 2 hours in a specially constructed low pres-

<sup>1</sup> H. Streuli, *Biochem. Ztschr.*, 86: 357, 1918.

<sup>2</sup> M. Duran, *Biochem. Ztschr.*, 106: 254, 1920.

<sup>3</sup> A. L. Barach, M. Eckman and N. Molomut, *Am. Jour. Med. Sci.*, 202: 336, 1941.

<sup>4</sup> B. Houssay and C. Riatti, *Compt. Rend. Soc. de Biol.*, 110: 144, 1932.

<sup>5</sup> J. A. Campbell, *Quart. Jour. Exp. Physiol.*, 24: 271, 1935.

<sup>6</sup> C. G. Mackenzie and J. B. Mackenzie, *Endocrinology*, 32: 185, 1943.

<sup>7</sup> E. B. Astwood, J. Sullivan, A. Bissell and R. Tyslowitz, *Endocrinology*, 32: 210, 1943.

<sup>8</sup> E. B. Astwood, *Jour. Am. Med. Assn.*, 122: 78, 1943.

<sup>9</sup> R. H. Williams and G. W. Bissell, *SCIENCE*, 98: 156, 1943.

<sup>10</sup> A. S. Keston, E. D. Goldsmith, A. S. Gordon and H. A. Charipper, *Jour. Biol. Chem.*, in press.

sure chamber.<sup>11</sup> At the end of this period, the mortality percentages were recorded and the thyroid glands of all animals were dissected and weighed. The results are shown in Table 1.

The data indicate that treatment with thiourea for 12 or more days enables the rats to survive a reduction of the atmospheric pressure to 200 mm of Hg, whereas the majority of the untreated controls succumb. Thiourea injections made a short time (5 hours) prior to exposure to low pressures have no apparent beneficial effect. Preliminary experiments performed with males have shown that these are also benefited by thiourea provided the treatment is applied for at least 12 to 14 days. It is to be observed that the animals on a thiourea ration for 12 or more days possess heavier thyroids than do the untreated rats or those

given the drug for shorter periods of time. This enlargement of the thyroid gland, a condition which is readily reversible when thiourea is withdrawn, is indicative of the development of a hypothyroid condition.<sup>6,7</sup> We are of the opinion that this state of functional hypothyroidism (chemical thyroidectomy) is responsible for the increased tolerance to reduced atmospheric pressures shown by rats treated with thiourea.

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## SCIENTIFIC APPARATUS AND LABORATORY METHODS

### A NEW METHOD FOR PRESERVATION OF HUMAN AND ANIMAL TISSUES BY THE USE OF A TRANSPARENT PLASTIC

A GOOD preservation of form of human and animal organs can be obtained by imbedding them in a transparent plastic, "Plexiglas." A method for imbedding insects and similar structures in "Plexiglas" has been proposed by Dr. Charles Sando,<sup>1</sup> but this procedure could not be applied successfully to fleshy animals or organs of animals and humans without considerable loss of form and color. The present process has been used so far in the case of relatively small organs (approximately 8×5×5 cm) and to slices of larger organs (10×10×2 cm). There appears to be, however, no difficulty in applying the method to considerably larger objects.

The advantages of this method of tissue preservation are: (1) the remarkable preservation of size, shape and texture and the satisfactory retention of color; (2) the ease of handling of the finished specimen, which is permanent and practically unbreakable; (3) the avoidance of distortion of vision due to the curvature of the surface of containers and the preserving fluids generally employed.

Preliminary to the imbedding, it is necessary to dehydrate the tissue. This is accomplished by placing the fresh organ or piece of tissue on a layer of frozen water (5 to 10 mm in thickness) contained in a suitable receptacle, and cooled to -20° C. or below, by covering the organ with cold distilled water and freezing the whole mass as rapidly as possible at a temperature of -20° C. or below. The mass of ice without and within the organ is then removed by condensation of water vapor at low temperature *in vacuo*.<sup>2</sup>

<sup>11</sup> M. Dubin, *Quart. Jour. Exp. Physiol.*, 24: 31, 1933.

<sup>1</sup> H. G. Knight, *SCIENCE*, 86: 333, 1937.

The tissues thus obtained are very light and porous and appear to have lost a great deal of the color and texture. At this point the specimen may be readily trimmed with a sharp scalpel. The dried tissues should be promptly sealed or processed so as to avoid absorption of moisture. They should be carefully brushed with a camel's hair brush, to remove any loose particles or dust and similar foreign material.

The next step in the process is the saturation of this tissue with the liquid acrylic ester (monomeric ethyl methacrylate).<sup>3</sup> This is attained by pouring the ethyl methacrylate in a large-mouthed receptacle, placing the tissue in it and then producing a vacuum (about 700 mm of Hg.). This is easily accomplished by setting the open receptacle containing the specimen in a vacuum-desiccating jar with stout walls. In a period of one half hour to one hour, the air will be entirely replaced by the ethyl methacrylate, and upon readmitting air into the jar, the tissue will sink in the fluid. In the case of highly porous materials, it may be necessary to repeat this vacuum treatment several times before all trapped air has been replaced by the monomer. The colors of the tissue appear to have returned, but they are somewhat duller than normal and the natural texture is still lacking at this point of the process.

The subsequent steps have to deal with the imbedding of the tissue by polymerization of the monomer. In view of the fact that prolonged immersion of the tissue in the liquid ethyl methacrylate causes bleaching, it is desirable to hasten the polymerization by the addition of a catalyst, such as benzoyl peroxide, and

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<sup>2</sup> Max M. Strumia and John J. McGraw, *Jour. of Lab. and Clin. Med.*, 28: 9, 1140-1153, June, 1943.

<sup>3</sup> Obtained from Rohm and Haas Company, Philadelphia 5, Pa.