

The control plants also showed "blueing" of the petals after 2 days, in contrast to the relatively unchanged shade of the treated flowers. The treated flowers remained in the loose bud stage for 6 days and, although not wilted appreciably by this time, showed browning of the bases and margins of the petals. Some of the fragrance was lost as a result of treatments.

Azalea, grape hyacinth, iris, carnation and *Spirea Vanhouttei* were also treated, but the results were not outstanding.

As Table 1 indicates, there is a large increase in

TABLE 1
WEIGHT OF CUT FLOWERS BEFORE AND AFTER SUBMERGENCE
IN WATER FOR 20 MINUTES IN PARTIAL VACUUM

Material	Weight (grams)	
	Before treatment	After treatment
Azalea	6.00	16.2
Carnation	25.0	42.0
Grape Hyacinth (<i>Muscari botryoides</i>)	14.1	25.0
Iris	6.6	12.6
Lilac (<i>Syringa vulgaris</i>)	130.8	180.0
<i>Narcissus maximus</i>	18.3	21.3
Rose (Hybrid Tea)	75.1	109.5
<i>Rosa Hugonis</i>	21.1	40.1
<i>Spirea Vanhouttei</i>	60.0	133.8
Tulip (<i>Tulipa Gesneriana</i>)	43.0	58.0

weight following treatment. In some cases it is more than double, depending on the material. During treatment, tissues can be observed to become water-soaked and translucent. This condition disappears rapidly in some plants, as in lilaes, but more slowly in others. Since some plants are capable of taking in more water and holding it longer than others, each kind responds differently to treatment. In general, the best results were obtained with plant materials which have large leaves and stems and large inferior ovaries, capable of serving as reservoirs.

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MODIFIED METHOD OF EXTRACTING CHOLESTEROL*

MANY methods of extracting cholesterol from blood have been devised and most of the methods used for extraction from liver are based on those used for blood. The most common method used is that of Bloor,¹ which involves saponification. An interfering yellow color which reduces the accuracy of the Leibermann-Burchard determination is produced, which may be reduced by absence of heat² or the use of a red filter.³ Ireland⁴ found that the use of a red filter did not eliminate interference and devised a new method of extraction. Schoenheimer and Sperry⁵ purified the cholesterol extract by precipitation with digitonin. Foldes⁶ modified the digitonin precipitation method in order to eliminate the interference of bile. Noyons⁷ using a method similar to Bloor's¹ found that saponification gave consistent but lower values than extraction without saponification. Teeri⁸ states that extraction without saponification produced values 25 per cent. higher than extraction with saponification. Gershberg and Forbes⁹ devised an acetone and alcohol extraction method with saponification for determining cholesterol content of blood.

Most of the above methods are time-consuming and many do not give reproducible results. Therefore, a new method has been devised which reduces time and gives consistent results. The method is as follows: The liver is ground thoroughly with anhydrous sodium sulfate and three portions of 3:1 acetone-alcohol mixture—a ten cc portion followed by two five cc portions. The acetone, alcohol and liver are placed in a centrifuging tube together with 15 cc of anhydrous ether. The mixture is shaken for ten minutes, centrifuged and the supernatant evaporated in a partial vacuum under nitrogen. The cholesterol is determined by means of the Leibermann-Burchard test with the Evelyn photoelectric colorimeter.

The above method produces more consistent results than a modification of the Bloor method.

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DISCUSSION

THE EFFECT OF THIOURACIL ON TISSUE OXIDASE

WE have read with great interest the paper entitled "The Effect of Thiouracil on the Respiration of Bone Marrow and Leucocytes *in vitro*," by Dr. Charles O. Warren.¹

We have recently studied the influence of thiouracil, sulfonamides and a number of other compounds on the cytochrome oxidase (paraphenyldiamine oxidase) of the thyroid gland of the rat.² Thiouracil in

0.002 M solution added to thyroid tissue *in vitro* inhibits the oxidase activity significantly (decrease

* Contributions from the Department of Zoology, Smith College, No. 212.

¹ W. R. Bloor, *Jour. Biol. Chem.*, 17: 377, 1914.

² G. E. Sackett, *Jour. Biol. Chem.*, 64: 203, 1925.

³ W. R. Bloor, *Jour. Biol. Chem.*, 77: 53, 1928.

⁴ J. T. Ireland, *Biochem. Jour.*, 35: 283, 1941.

⁵ R. Schoenheimer and W. Sperry, *Jour. Biol. Chem.*, 106: 745, 1934.

⁶ F. Foldes, *Jour. Lab. Clin. Med.*, 28: 1889, 1943.

⁷ E. C. Noyes, *Biochem. Zeitschr.*, 289: 391, 1938.

⁸ A. E. Teeri, *Jour. Biol. Chem.*, 156: 279, 1944.

⁹ H. Gershberg and J. C. Forbes, *Jour. Lab. Clin. Med.*, 27: 1439, 1942.

¹ SCIENCE, 102: 174, August 17, 1945.

51 per cent. ± 6.3). A significant decrease of oxidase activity was also observed in the thyroid glands of rats treated for 11 to 23 days with thiouracil (0.05 per cent. drinking solution) as compared with the oxidase activity of thyroids of normal controls. Inhibition of the cytochrome oxidase system in the thyroid gland may possibly form the basis for the "anti-thyroid" effect of thiouracil and sulfonamides. In view of the ubiquity of cytochrome oxidase it would be expected that the antithyroid drugs would exert some influence on a number of tissues. However, approximately 90 per cent. of patients treated with thiouracil show no effects other than suppression of formation of thyroid hormone, and only 10 per cent. show untoward reactions indicative of an influence of the drug on other tissues. In experimental animals most investigators have found little effect from thiouracil on tissues other than the thyroid unless very large doses were employed.

In unpublished experiments we have observed that the cytochrome oxidase activity (paraphenylenediamine oxidase activity) of the bone marrow and of the kidney, which is of the same magnitude as that of the thyroid gland, can not be inhibited *in vitro* by thiouracil in 0.002 M concentrations which significantly inhibit the oxidase system of the thyroid gland. Thiourea in concentrations up to 0.01 M is equally ineffective. Experiments are also now under way to determine the nature of this protective mechanism. The data presented by Warren indicate that the protection of the bone marrow is not absolute but can be overcome by the use of higher concentrations of thiouracil.

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TEMPERATURES IN SOME DEEP WELLS IN PENNSYLVANIA AND WEST VIRGINIA

THE average temperature gradient, as given by some text-books in geology, is one degree Fahrenheit per 100 feet. The rate of increase varies within wide limits in different parts of the globe. In some places, it is as high as one degree in 30 feet. In the deep gold mines of the Transvaal in South Africa, it is only one degree Fahrenheit in 250 feet. The average for all observations is believed to be about one degree in 60 feet. In the copper mines at Butte, Montana, which are several thousand feet deep, it is uncomfortably warm.

In some wells, a mile and a half deep, the difference

² Preliminary report: *Fed. Proc.*, 4: 55: 1945, final paper in press.

in temperature, between top and bottom, is 125 degrees Fahrenheit, or more. The oil well drilled by the Continental Oil Company, known as K.C.L. A.-2., located four miles west of Wasco, near the middle of the San Joaquin valley in California, showed a temperature of 268 degrees Fahrenheit at the bottom, which is 15,004 feet, or nearly three miles below the surface. This is 6,000 feet deeper than the deepest mine in the Rand in South Africa.

The writer has taken the figures for temperature at various depths from deep wells in Pennsylvania and West Virginia, and compiled the results as indicated in Tables 1 and 2. The data were obtained from

TABLE 1

Location of well	Depth (feet)	Temp. Fahr. at bottom
Volcano Oil and Coal Co., Wood Co., W. Va.	4,250	113.1
Slaughter Creek Coal and Land Co., Kanawha Co., W. Va., No. 1 Well	4,730	113
Lehigh Portland Cement Co., Lawrence Co., W. Va., No. 1 Well	5,000	126.6
Slaughter Creek Coal and Land Co., Kanawha Co., W. Va., No. 1 Well	5,230	129
W. T. Beidenbach, Armstrong Co., Pa., No. 1 Well	5,965	146.2
J. S. Lightcap, Indiana Co., Pa., No. 1 Well	6,460	153
R. A. Geary (now Sarah Cummings), Washington Co., Pa., No. 770 Well	6,975	144.9
Martha Goff, Harrison Co., W. Va., No. 4190 Well	7,310	158.3
Jacobs Creek Oil Co., Westmoreland Co., Pa., No. 1(3297) Well	8,080	211

TABLE 2

Depth (feet)	Temp. Fahr.	Temp. difference	Increase in temp. per 100 feet in depth
(1) 8,080	211		
4,250	113.1	97.9	2.55
(2) 5,000	126.6		
4,250	113.1	13.5	1.8
(3) 5,230	129		
4,730	113	16	3
(4) 5,965	146.2		
5,000	126.6	19.6	2
(5) 6,460	153		
5,000	126.6	26.4	1.8
(6) 6,460	153		
5,965	146.2	6.8	1.37
(7) 7,310	158.3		
6,975	144.9	13.4	4
(8) 7,310	158.3		
6,460	153	5.3	.62
(9) 8,080	211		
7,310	158.3	52.7	6.8

"Summarized Records of Deep Wells," West Virginia Geological Survey, Vol. XVI, 1944, by R. C. Tucker. The methods used for obtaining these temperatures is not known to the writer. One important fact brought out by a study of the well records is that the average increase in temperature per 100 feet in depth, from the 4,250 to 8,080 foot level, is 2.77 degrees Fahrenheit. Another interesting feature is the high tempera-