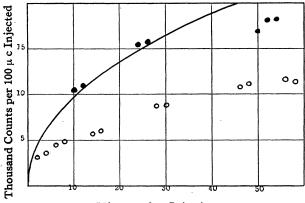
plaints, and may be useful as a guide to treatment. In Fig. 4, the circles are data for a woman with dia-



**Minutes after Injection** 

FIG. 4. Diabetes. Normal curve from Fig. 1. Right foot normal, left gangrenous. Subsequently amputated, with poor wound healing.

betic gangrene in the left foot, no symptoms in the right. The solid circles, for the right foot, fall within normal limits; the open ones for the left, are low. The left foot was subsequently amputated, with poor wound healing. In other cases the region of the leg at which normal counting occurred has been used satisfactorily as a guide to position of amputation.

The interpretation of the type of count which increases more slowly than normal, but eventually, even after several hours, reaches the normal level, is probably concerned with the condition of the circulatory system. In those cases in which the count does not come up to normal after several hours, the explanation may be different and may have to do with actual change in the so-called "sodium space" in the foot.

Sodium space has been determined by Kaltreiter and his associates and by others, by injecting radiosodium as in the present work and then withdrawing blood at a certain time subsequently, and determining the ratio of counts per cu cm of serum to counts for the total material injected, under standard conditions.<sup>4</sup> Further studies along this line are being carried out, for patients with vascular disease.

Up to the present time fifty individuals have been studied, some in more detail than others. In each disease group only a few cases are available; however, the data have already proved of value in diagnosis and prognosis, and it is evident that further work is desirable.

An application of the method which has not as yet been employed to any extent is as an evaluation of therapeutic procedures. If a patient is tested before treatment is instituted and at intervals while it is being carried out, any change either for the better or worse should be indicated. Repetitions of the test at intervals of a few weeks would be entirely safe, since the amount of radiation dose for each test is so small.

The authors wish to express their indebtedness to the Radiation Laboratory of the Department of Physics of Columbia University, for providing the Geiger-Müller counting apparatus and for the preparation of the radioactive sodium.

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## SCIENTIFIC APPARATUS AND LABORATORY METHODS

## A DRY ICE FREEZING UNIT FOR ROTARY MICROTOMES

TISSUE sections cut from the frozen state are being utilized in an increasing number of techniques, but the equipment necessary for cutting such sections is not always available. The methods in common use require either a sliding microtome or a specially designed "freezing" microtome. However, the rotary microtome is the only kind available in many laboratories, and although it is generally considered to be unsuited for cutting frozen sections, it may be quite satisfactorily adapted for this purpose by the use of the device illustrated in Fig. 1.

This consists of a metal box made to conform to the shape of the object clamp of the microtome in which it is to be used, but about 2 mm smaller on all sides to allow for insulation. (The dimensions shown are suitable for No. 818 and No. 820 Spencer microtomes.) One end is made from a piece of sheet copper, to the outer surface of which is soldered a metal object holder about 2 mm in thickness and large enough to accommodate the desired specimens. The face of the object holder is grooved to provide for the firm attachment of the specimen and its entire back surface must be uniformly fused to the end of the box to allow for rapid and efficient heat transport. The sides of the box are made from a single sheet of copper bent to the same size and shape as the end and soldered to it. The other end is made of asbestos board, bakelite or other insulating material. It fits tightly into the open end of the box and should have a small handle to allow easy removal. The entire unit, except the surface of the object holder, is insulated by cementing on a layer of asbestos paper and then covering this with a layer of hard-surfaced paper and several coats of waterproof varnish.

<sup>4</sup> N. L. Kaltreiter, G. R. Meneely, J. R. Allen and W. F. Bale, *Jour. Exper. Medicine*, 74: 569, 1941.

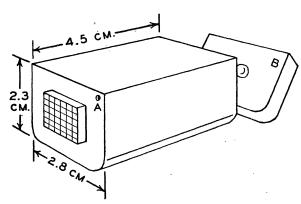


FIG. 1. Freezing unit without insulation. A = Hole for escape of  $CO_2$ , B = Removable end.

In using this unit, the case is filled with dry ice (about 20 g) and a piece of tissue, either fresh or infiltrated with gum or gelatin, is placed on the objectholder, to which it adheres almost instantly. Freezing may be hastened by inverting the unit after the tissue is firmly attached or by placing small pieces of dry ice around the tissue. Pieces 10 mm or more in thickness may be frozen. When the tissue is completely frozen, the dry ice is replenished and the unit inserted in the microtome. The heat transfer within the container is so rapid that it is not necessary to keep the dry ice pressed against the freezing surface. The tissue will remain frozen until all the dry ice is gone, 15–20 min. If more time is required, the unit may be easily refilled before the block has thawed.

This unit provided excellent sections of large feather germs, which are difficult to section by any method, and of such soft tissues as liver, kidney and spleen. Soft tissues could be cut at 20–30 microns without fixing or embedding, and excellent sections as thin as 5 microns could be cut after infiltration with a gum arabic solution. This attachment is easily made from materials available anywhere (other metals might be substituted for copper, although it is important to keep the thermal conductivity as high as possible), it is simple and economical to operate, and it produces sections comparable to those obtained with other freezing devices. It should be adaptable to all work except where very large pieces of tissue must be cut.

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## A DROP METHOD OF PENICILLIN PRO-DUCTION<sup>1</sup>

A NEW method of producing good yield of peni-

<sup>1</sup> From the Department of Pathology and Bacteriology, City Hospital, Welfare Island, Department of Hospitals, New York, N. Y. Preliminary report. cillin consists of culturing *Penicillium notatum* No. 5415 on a solid agar base containing constituents which favor a rapid production of penicillin in drops of sufficient size and quantity to be precipitated like rain upon the opposing side of the container, when inverted. The type of culture is what is sometimes called a still growth and 5 to 6 days are required for production of large drops of clear penicillin. This material is withdrawn by means of sterile pipettes and is preserved anaerobically at 5° C.

Two types of agar medium have been employed: (1) A modified Sabouraud's acid maltose described by Scudder<sup>2</sup> and by Risch<sup>3</sup>; and (2) special synthetic medium modified by Robinson, to which has been added 2 per cent. flaked agar. No adjustment of the media is necessary.

The penicillin thus produced has the same physical properties as were described by Fleming<sup>4</sup> for that which he discovered. It is lighter than chloroform, heavier than ether, soluble in water, transparent, clear and yellow. Penicillin isolated at this institution has been found bactericidal in 1–10 dilution and bacteriostatic in dilutions ranging from 1–200 through 1–600. Its unit value is greater than 6 units per cc.

Preparations of our penicillin have been used for topical application with very satisfactory results and dilutions range from 1-250 to 1-800 in sterile distilled water. The solution keeps satisfactorily in the wards if layered with solid paraffin and kept at ice-box temperature. Sterile oils lighter than the penicillin may also be used.

Acknowledgment is made for the helpful advice of Dr. Charles Thom, formerly mycologist for the U. S. Department of Agriculture, and to Dr. Y. Subba Row, biochemist for the Lederle Laboratories, Pearl River, N. Y. The special synthetic medium was obtained through the courtesy of the Eimer and Amend-Fisher Scientific Co., New York.

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<sup>2</sup> SCIENCE, 79: 16, 1934.

<sup>3</sup> Arch. Otolaryng., 29: 235-251, 1939.

<sup>4</sup> Brit. Jour. Exp. Path., 10: 226-236, 1929.

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