ture and origin. Has the memory of Kapteyn been "vaporized" in the Soviet Union and the great Dutch astronomer become an "unperson"? (See Orwell's *1984*.)

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Mercury as a Casting and a Contrast Medium

THE study of the ear is essentially the study of the interior of the temporal bone, since the labyrinth is completely enclosed by the petrous capsule. A technique for preparing metal casts of the petrous bone was described in a previous issue of this journal (*1*), and this process was later expanded to include the entire temporal bone (*2, 3*). The need for extending this casting method to embrace the limiting membranes became obvious, and a search was made for lower-fusing metals that would not cook or distort the delicate tissue-membranes of the intact temporal bones of human autopsy specimens or the living ears of experimental animals. The search led to the selection of a metal that needs no heating, but is already molten at normal body temperatures. Thus, mercury was instilled through the round or oval windows, and x-ray studies were made with serial stereoscopic views.[1] This not only reveals the contours and outline of the labyrinth itself, but such associate channels as the endolymphatic and perilymphatic ducts. As a result of these studies it was found that, contrary to the generally accepted opinion, the perilymphatic duct does not communicate with the subarachnoid space of the hindbrain but is limited by a terminal sac, similar to the endolymphatic duct and sac (*4*).

The channels by which the perilymphatic fluid normally escapes from the confines of the bony labyrinth have long been debated. Prolonged serial x-ray studies of mercury-filled labyrinths in experimental animals show the escape of this fluid through channels that course in the tegmen tympani and traverse the petrotympanic fissure and continue along the Eustachian tube to the lymph nodes of the nasopharynx. The presence of these channels has been confirmed (*5*) by hard metal casts of the temporal bone and by chemical dye studies (Prussian blue).

When mercury in 2- or 3-ml amounts is injected into the carotid artery of *Macacus rhesus,* the entire carotid system is rendered radiopaque even to the terminal papillary arterioles, but none of the mercury traverses the capillary bed to the venous side, thus giving a true arteriogram. The animals showed no deleterious effects from the mercury instillation during the 3 or 4 hr ensuing before the experiment was finally terminated.

[1] A watertight union may be made with the labyrinth through either of its windows by first inserting a piece of tightly fitting "radio spaghetti" through the window and then forcing the blunted end of the hypodermic needle through the constricted lumen of the spaghetti tube. This may be re-enforced with bone wax, liquid cement, or plaster of Paris, to insure against leakage.

It appears that this cheap and readily available metal offers a superior medium for forming metal casts of various body cavities, as well as a contrast medium for studying fine arterial and lymphatic channels with the aid of x-rays.

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Soxhlet Extraction at Reduced Temperature

IN THE course of a study of alkaloids from Tasmanian plants, it was necessary to extract about 1½ kg of material with chloroform, and since the alkaloids extracted were likely to be heat-sensitive, the extraction had to be carried out at as low a temperature as possible. The simple apparatus illustrated in Fig. 1,

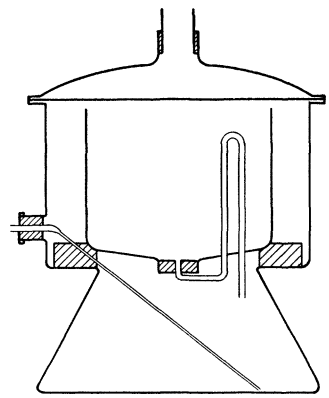

FIG. 1.

which was found to be quite satisfactory for the purpose, consists essentially of three parts. The outer vessel, which contains the boiling chloroform, consists of a large vacuum desiccator, with an opening for evacuation at the side, through which an air or nitrogen leak reaching to the bottom of the vessel can be inserted. The lid of the desiccator is replaced by one from another vacuum desiccator with an opening at the top, on which is mounted an efficient condenser. The two rubber stoppers are protected from the solvent by coating them with a thick paste made from dextrin, mannitol, and glycerol.

The vessel that contains the material to be extracted consists of the upper half of a large bottle with a syphon tube mounted in a cork in the neck. The material is placed in a circular filter paper folded so as to form a large pleated cup fitting inside the extraction vessel.

The apparatus, heated in a water bath, is used under reduced pressure in conjunction with a modified form

of Cartesian diver manostat (*1, 2*), which maintains the system at any desired constant pressure. The extractor was found to work satisfactorily over extended periods when the pressure was adjusted to give an internal temperature of about 30° C.

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Microsporum gypseum and *Histoplasma capsulatum* Spores in Soil and Water[1]

THE use of a membrane filter for recovery of bacteria from water has been described recently by Clark *et al.* (*1*). The method entails incubation of an exposed membrane on a sterile absorbent pad saturated with culture medium. We have found that, following exposure to liquids or gases, the membrane may be stained so as to render feasible direct microscopic examination for such relatively large structures as fungus spores and pollen. Such a procedure is, of course, effective only for those organisms producing distinctive and identifiable microscopic structures.

Following filtration of air, water, or other fluids, the membrane is removed to a Petri dish, and an adhesive, such as Haupt's gelatin, is added by dropper to the exposed surface area in order to prevent washing off the spores during the staining process. The membrane is then processed by a modification of the Hotchkiss-McManus (periodic acid–Schiff) technique (*2*), which stains hyaline fungus spores and mycelium a dark-red against a colorless background. The stained, moist membrane is mounted on a glass slide, under a cover glass, and examined under the microscope.

Application of this technique to aqueous soil extracts has resulted in the finding of numerous tuberculate spores typical of *Histoplasma capsulatum* (Fig. 1, *A*) in four of four soil samples from Williamson Co., Tenn., an area in which there is an extraordinarily high prevalence of histoplasmin sensitivity among the human population. All the samples tested had previously yielded *H. capsulatum* in culture by means of the mouse-inoculation test (*3*). That the tuberculate spores occur in soil had been demonstrated earlier by Emmons (*4*) in Virginia.

Also, by means of the filter method, a tuberculate spore morphologically identical with those obtained from soil and from artificial cultures of *H. capsulatum* has been recovered from a single 1-liter specimen of river water from Williamson Co. This is the first report of the natural occurrence of this organism in water.

[1] Grateful acknowledgment is made to Lawrence B. Hall, of the Communicable Disease Center, for having made available some of the equipment used in this study.

These results suggested the advisability of comparing the membrane filter method with the mouse-inoculation technique for effectiveness in the recovery of *H. capsulatum* from soil. Such a study is now under way.

From one of the above soils there were recovered, in addition, upon the membrane filter, several multiseptate spores typical in all respects of the macroconidia of *Microsporum gypseum* (Fig. 1, *B*). Two strains of *M. gypseum* subsequently were isolated in pure culture from this soil specimen. This dermatophyte had been cultured previously by Mandels *et al.* (*5*) from wool cloth that had been buried in potted soil in an attempt to isolate keratinophilic fungi from the soil. Whether the isolate of *M. gypseum* had actually originated or was growing in the soil is not certain. According to information received from the laboratories in which the isolation was made,[2] the soil was composed of clay loam, sandy silt, humus, and manure, and had been used in indoor tests for several years. There is no record that the wool had been sterilized prior to burial.

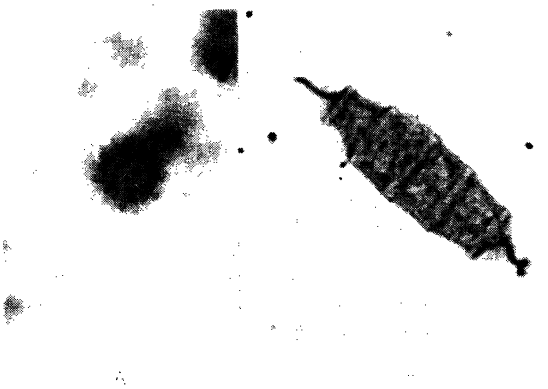

FIG. 1. *A*, tuberculate spore of *H. capsulatum* recovered from soil; stained on filter membrane. × 760. *B*, macroconidium of *M. gypseum* recovered from water; stained on filter membrane. × 760.

We know of no report prior to the present one of the occurrence of macroconidia of *M. gypseum* in nature. Details of cultural procedures and of the use of filters in spore detection will be reported shortly.

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