

In the first place, it was found that *Phialophora* is not limited to North America, as MacKinnon was able to point out in Uruguay. One of the original cultures of Pedroso and Gomes, described as *Acrotheca* by Fonseca, we found to be *Phialophora*. A fungus recently described from Buenos Aires presents characteristics which are clearly those of *Phialophora*. In the second place, a careful examination of fungi described as *Acrotheca* revealed properties, such as branching, successive conidiophores and conidia in head formation, which are definitely not those of the genus, but more related to the large heterogeneous *Botrytis*. Due to additional characteristics which are those of the Dematiaceae and not of the Mucedinaceae, a new genus is proposed, namely, *Botrytoides* Moore and Almeida, to replace *Acrotheca* and *Trichosporium* for the causative organism of chromomycosis. Thirdly, the genus *Hormodendrum* has been isolated from authentic cases of this disease. Finally, a microbe isolated from a recent case in São Paulo has characteristics common to all the above-named genera.

When first grown, the new fungus appears much like *Trichosporium* as described by Langeron, a characteristic which, if interpreted correctly, however, is not consistent with that genus. When examined carefully on several media, the conidiophore of *Botrytoides* is clearly visible, and further, the type of spore formation of *Hormodendrum* is discernible. On several media, particularly Czapek's, in addition to these facts, the cup formation of conidia production is quite marked, appearing on approximately the twelfth day. This is distinctly a property of *Phialophora*. On Sabouraud's maltose agar the cup formation is seen almost exclusively, while on still other media, no cups, but branching conidia or conidiophores similar to those of the genus *Acrotheca* are found. For this complicated new organism, which apparently suggests itself as a missing link that binds *Botrytoides*, *Phialophora* and *Hormodendrum* in close relationship, the name *Phialoconidiophora Guggenheimia* Moore and Almeida, new genus and new species, is given. The species is gratefully dedicated to the John Simon Guggenheim Memorial Foundation for making this study possible.

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DIFFERENTIAL SUSCEPTIBILITY OF LIVING ORGANISMS TO SUPERSONIC VIBRATIONS

GRADED differentials in susceptibility to a large number of physical and chemical agents, not differing specifically with different agents, are very generally characteristic of the polar axes of animals at least during the earlier stages of development and in some of the

simpler forms throughout life. A respiratory gradient and a gradient in rate of reduction of vital dyes, as well as various other differentials have been found to parallel closely the susceptibility gradient in animals from which data have been obtained. In view of these facts, it is of interest to determine whether a differential susceptibility to supersonic waves, a primarily mechanical factor, exists.

In the experiments described below a modified Hartley oscillator circuit employing a 100-watt tube operated by raw alternating current was used to drive a large quartz crystal at its natural frequency of 612 kilocycles per second. The crystal was mounted between two heavy brass electrodes and served as a microscopic slide on the insulated stage of a grounded microscope. The lower electrode had a circular opening to permit a beam of light to pass through. The upper grounded electrode, when placed on the crystal, formed the wall of a cavity or cell for holding the experimental animals in water. The arrangement was held in place by a wooden frame soaked in paraffin. The intensity of the supersonic field was changed by modifying the input of the oscillator. A radio frequency milliammeter was used to give a rough measure of intensity.¹

The temperature of water in the electrode cell rose approximately 6° C. during the average exposure required, but this rise was decreased to approximately 2° by introduction of a glass capillary tube cooling system. This rise in temperature during the few minutes of exposure which was required to observe the visible changes was not a factor in results. Control experiments with a similar rise in temperature and a period of exposure three times the usual experimental exposure showed no effects of temperature.

When *Euplanaria dorotocephala* is exposed to supersonic vibration the posterior tip and margins of the posterior zooid or zooids show disintegration first, and as the posterior zooid region disintegrates, disintegration begins in the head region by the cytolysis of the lateral margins and the auricles. Next the ganglionic region disintegrates, leaving the optic pigment visible for some time in the disintegrated mass of the head. Intact parts show strong muscular stimulation and profuse mucus secretion. Intense stimulation of the pharynx causes it to be extruded through the dorsal body wall and complete separation occurs, as with irritating chemical agents. The pharynx does not disintegrate, because it is thrown about the field by the wave patterns, and probably is not subjected to definite waves in one position long enough to show any effects.

The time required for disintegration varies with the intensity of the supersonic field and the condition of

¹ The cooperation of Dr. J. Barton Hoag, of the department of physics, University of Chicago, in construction of the apparatus is gratefully acknowledged.

the animal. With medium intensity the first signs of cytolysis appear in one minute and in three to four minutes disintegration is far advanced. Disintegration ceases after cessation of the vibration, intact parts remain alive and may reconstitute whole animals.

Tubifex tubifex, subjected to supersonic vibration, is very active showing intense stimulation. High intensity results in almost instantaneous death and disintegration, but with lower intensity the posterior growing segments disintegrate first, and disintegration progresses anteriorly. Simultaneously, a short gradient appears in the anterior region of the body.

Observations on hydra show that the tentacles dis-

integrate basipetally, but the body contracts so strongly that it is difficult to determine the course of disintegration there.

These supersonic disintegration gradients are the same as those observed with other agents and, so far as data are available, the same as the respiratory gradients and gradients in reduction of vital dyes. Evidence of alteration of reconstitution and of head frequency in *Euplanaria* by supersonics has already been obtained and work is being continued.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

ARTIFICIAL MEDIA FOR THE CULTIVATION OF FIBROBLASTS, EPITHELIAL CELLS AND MONOCYTES

THE object of the present communication is to describe two synthetic media that have been developed for the cultivation of cells outside the body. One was designed to promote the rapid proliferation of fibroblasts and epithelial cells; the other, that of monocytes. These media contain practically the same ingredients; they differ in the concentration of these ingredients.

Two synthetic media for fibroblasts, less complete than the one to be described here, have already been devised. The first of these contained a peptic digest of casein, glycocholic acid, nucleic acid, glutathione, hemoglobin, glucose and some ash of liver. A pure strain of fibroblasts from Crocker 10 sarcoma proliferated in this medium for 40 days as rapidly as did control tissues that were cultivated in embryo juice.¹ This medium did not suffice, however, for the cultivation of normal fibroblasts. The second medium, one devised by Vogelaar and Erlichman,² has given better results with the normal cells. It contained Witte's peptone, hemin, cysteine, insulin, thyroxine, glucose and the usual salts. Fibroblasts emanating from human thyroid tissue that was embedded in irradiated cow plasma proliferated in this medium for three months. The medium failed, however, to nourish chicken heart fibroblasts that were embedded in coagula made of horse plasma.³

The new medium to be described here contains all the substances used by Vogelaar and Erlichman, and in addition a number of others that greatly enhance its power to promote growth and maintain cell life. Its composition is as follows:

	Per 100 cc
Witte's peptone	675.00 mg
Cysteine hydrochloride	9.00 "
Hemin	0.0036 "
Insulin	0.09 units
Thyroxine	0.0009 mg
Glucose	100.00 "
Serum homologous to the tissue	10.00 cc
Vitamin A ⁴	900.00 to 1800.00 units
Vitamin D ⁴	about 15.00 to 30.00 "
Vitamin C (crystalline ascorbic acid)	0.25 mg
Glutathione	1.00 "
Phenol red	5.00 "
Sodium chloride	720.00 "
Potassium chloride	18.00 "
Calcium chloride, anhydrous	18.00 "
Magnesium chloride, 6 H ₂ O	9.00 "
Sodium dihydrogen phosphate	4.50 "
Sodium bicarbonate, anhydrous	100.00 "

When the solution is used for the cultivation of organs, the glucose is increased to 300 mg per cent., and the sodium chloride reduced sufficiently to keep the solution isotonic. A small amount of iodine is added when the medium is used for the cultivation of the thyroid gland.

This new medium promotes more rapid and more prolonged growth of fibroblasts than does any artificial medium previously devised. Chicken heart fibroblasts that were embedded in horse plasma proliferated in this medium two or three times as rapidly as did control tissues that were cultivated in the feeding solution of Vogelaar and Erlichman. A pure strain of these cells multiplied actively for six weeks without showing any deterioration or decrease in growth rate. Control fibroblasts, cultivated under the same conditions in the Vogelaar feeding solution, underwent fatty degeneration and died in 12 or 14 days. The new medium causes fibroblasts to proliferate for a time fully as rapidly as they do in embryo juice. In one experiment, two tiny fragments of heart fibroblast that were in their sixth passage *in vitro* increased in size so fast that they completely covered the coagulum in a D-3 flask in 11 days. This rapid growth does not continue indefinitely, of course, for the medium is still incomplete.

⁴ Vitamins A and D were supplied together by using a concentrate prepared from halibut liver oil.

¹ L. E. Baker, *Jour. Exp. Med.*, 49: 163, 1929.

² J. R. M. Vogelaar and E. Erlichman, *Am. Jour. Cancer*, 18: 28, 1933.

³ Unpublished experiments of the author's.