

### IMMUNIZATION TO INFECTIOUS MYXOMATOSIS

INFECTIOUS myxomatosis (Sanarelli) is known to be a highly specific and an almost invariably fatal virus disease for the domestic rabbit. More than 2,000 rabbits from our inbred colony have been tested with this virus under laboratory conditions in various experiments over a period of ten years. Every normal animal has proved susceptible, not a single one has escaped a fatal issue. Death follows in from 8 to 11 days after proper exposure to the active agent. It was demonstrated by Shope that the rabbit could be protected against this disease by previous treatment with the fibroma virus. We have confirmed and extended this observation. The fibroma virus, by all portals of entry, usually confers a solid and lasting immunity to the otherwise fatal myxoma.

Attempts to immunize the host with the myxoma virus attenuated by heat or chemicals have in the hands of a number of workers proven ineffective. It is in fact the usual experience of all workers with the viruses that little if any immunity results with the use of heat-inactivated virus.

We have found that some resistance to myxoma can be conferred upon the rabbit by previous intradermal injections of heat-inactivated tissue virus (60 C.<sup>o</sup>-30'). This refractory state may be enhanced by the addition of the viable type III *pneumococcus* or *Bact. lepi-*

*septicum* to the heated tissue vaccine, but not by the addition of the vaccine virus or viable tumor cells (Walker's rat carcinoma 256).

That the phenomenon is one of immunity is indicated by the fact that rabbits so treated give a marked allergic response to intradermal injections of the myxoma virus. The disease in many cases is aborted or its course mitigated. The skin lesions are more restricted and circumscribed. Discrete tumor-like nodular formations, instead of spreading oedematous lesions, appear on the ears and eyelids as well as on other parts of the body. Death is delayed. Many animals survive, some fully recover, others show chronic asthmatic symptoms with conjunctivitis. Animals that survive are solidly immune to massive intradermal injections of the myxoma virus except for the local response at the site of injection. Antibodies are present to an appreciable titer as determined by complement fixation. In general the titer of the serum of the treated animals at the time of infection is an index of their degree of resistance. The results suggest that accessory agents may be useful in attempts to establish immunity in other virus diseases where our efforts with heat-inactivated virus have thus far failed. Details of these experiments will be given in a forthcoming series of papers.

ROSCOE R. HYDE

SCHOOL OF HYGIENE AND PUBLIC HEALTH,  
THE JOHNS HOPKINS UNIVERSITY

## SCIENTIFIC APPARATUS AND LABORATORY METHODS

### A MODIFICATION OF THE SET-UP FOR WICK CULTURE

IN the set-up for growing plants by the wick-culture method as previously described by the writer<sup>1</sup> the main wick (the sheet of absorbent material on the surface of which the plant roots develop) is supplied with solution by means of a number of secondary wicks dipping into the solution contained in a pan situated near the upper edge of the main wick. Regulation of solution supply is accomplished by varying the number and width of these secondary wicks as well as the solution level in the pan.

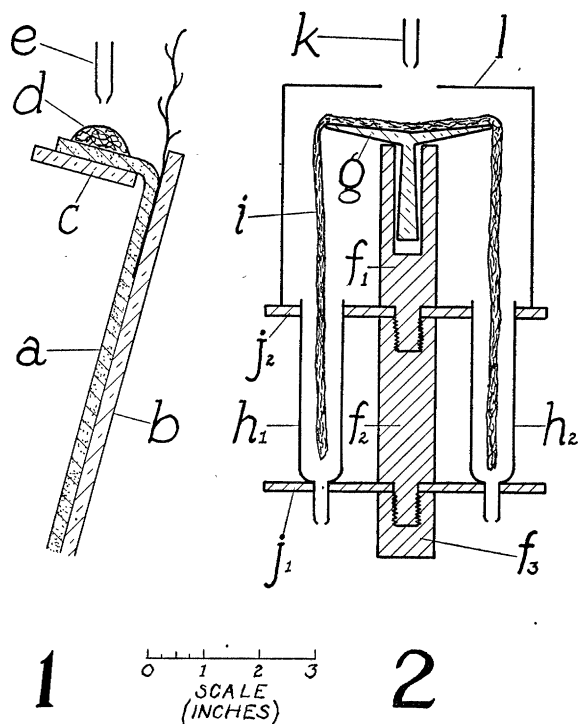
While this method has been used with entire satisfaction by the writer, it has been found almost prohibitively difficult by other workers in this laboratory. Recourse has been had, therefore, to dripping the solution onto the upper edge of the main wick. The modified set-up for this purpose is illustrated diagrammatically in Fig. 1, in which (a) indicates the main wick; (b) the sheet of plate glass against which it rests; (c) is a piece of plate glass about 10 cm wide which is supported in a position perpendicular to (b) near its upper edge, and at a distance of 1 to 1.5 cm

from it, on which the upper edge of the main wick (a) rests. The solution dripping from the tube (e) falls on a wad (d) of absorbent cotton or glass wool placed along the upper edge of the main wick and serving to distribute the solution laterally across its width. We have found dripping the solution on at two points along its width entirely satisfactory for a main wick of a width of 45-60 cm.

### A WICK ARRANGEMENT FOR SUBDIVISION OF THE SOLUTION STREAM

A slow drip of solution of constant rate is readily obtainable by the use of either a capillary resistance device such as described by Trelease and Livingston,<sup>2</sup> by Shive and Stahl<sup>3</sup> and by Zinzadze<sup>4</sup>; or a small-volume metering pump such as has recently been devised by the writer (shortly to be described). For subdivision of the solution stream (for dripping on at several points of the upper edge of the main wick) the wick arrangement illustrated diagrammatically in Fig. 2 has proved very satisfactory. It will be noted that the device contains no moving parts and no small orifices, and that the solution passing through it need not come into contact with any material other than glass.

<sup>1</sup> *Am. Jour. Bot.*, 24: 185-187, 1937.



FIGS. 1 and 2 (the scale applies only to Fig. 2.)

Referring to the figure, ( $f_1$ ), ( $f_2$ ) and ( $f_3$ ) are sections of brass rod which screw together to form a single central column. At the two points of junction of the sections are mounted the circular brackets ( $j_1$ ) and ( $j_2$ ), in which are holes for holding the funnel tubes ( $h_1$ ) and ( $h_2$ ). On the top of the brass column is mounted a shallow glass dish ( $g$ )—made from the inverted foot of a wine glass or sherbet glass, with the stem suspended in a deep recess bored in the top of the column. A wick ( $i$ ), consisting of a strip of glass cloth or tape,<sup>5</sup> is placed with its central portion resting in the shallow glass dish ( $g$ ) and its ends suspended in the funnel tubes ( $h_1$ ) and ( $h_2$ ). ( $l$ ) is a glass cover for maintaining the humidity around the upper portion of the wick ( $i$ ). It is made from a small battery jar, inverted, with a hole bored in the bottom for the inflow tube ( $k$ ). The apparatus is mounted by means of burette clamps (not shown) on the half-inch rod of a laboratory stand.

In operation of the device, the incoming solution stream, dripping from the inflow tube ( $k$ ) onto the central portion of the wick ( $i$ ), is subdivided by the capillary action of this into two outgoing streams dripping from the ends suspended in the funnel tubes

( $h_1$ ) and ( $h_2$ ). From these the solution flows to the wick-culture apparatus below. If the two portions of the wick are closely equal in width and length, then the solution streams dripping from them will also be found to be closely equal (readily within 5 per cent., which is adequate for the purpose in hand). Simultaneous subdivision into four streams has been accomplished without difficulty in this manner.

With regard to the mode of operation of the device, it may be interesting to note that there is a tendency to a pulsation or rhythm in its action. Liquid tends to accumulate in the shallow glass dish, and then to drain out through the ends of the wick. It would seem therefore that, at least in part, the mode of action of the device is that of an intermittent siphon with multiple outlet tubes.

M. A. RAINES

HOWARD UNIVERSITY

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<sup>2</sup> SCIENCE, 55: 483–486, 1922.

<sup>3</sup> Bot. Gaz., 84: 317–323, 1927.

<sup>4</sup> SCIENCE, 81: 540–542, 1935.

<sup>5</sup> Cloths and tapes woven from spun glass may be obtained from the Fibre Products Division of the Corning Glass Works, Corning, N. Y.