

Helicella ericetorum, *Helicella obvia*, and *Hellicella bolli*. Future studies will include other species of *Helicella*.

The embryos enter the snail by way of the mucous glands of the foot. Histologic sections of the foot reveal the mode of penetration of the embryos and the transformation into larvae. A small tubercle-like proliferation is formed around the worm as an expression of a reaction to the invasion. The development of the embryo into the larva is quite similar to that of *Mullerius capillaris*. It is completed 12-14 days after infection of the snails. Many changes in shape and size of the parasite characterize the 2 moultings. The cuticula appears separated but it is not shed. The twice moulted embryo represents the infective larva.

The middle sheath is usually attached to the larva; but it may also be completely separated or it may form a tail and a head cap when the worm retracts the head or tail. The non-pigmented, transparent larvae show a distinct intestinal tube. The dimensions are as follows: Greatest width of the finely striated body is 40-45 μ ; the length 560-620 μ ; opening of the excretory duct 100 μ posteriorly to the head; length of the esophagus 200-220 μ , of the intestinal tube 300-350 μ , anus to tip of tail 50 μ . The genital rudiment approximately 160 to 180 μ posteriorly to the esophageal ending.

The development in the mammals is the same as that formerly described for other lungworms. The moult in the mesenteric glands of the host, and the pathological anatomy will be discussed in a consequent paper.

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Poliomyelitis Virus. Results of Treating with Certain Chemicals and with Heat.*

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Vinson and Petre¹ demonstrated that the virus causing mosaic disease of tobacco was "in many ways analogous to that of a chemical substance," since it could be precipitated by certain dyes and chemicals without loss of virulence and could be recovered uninjured

* This study was aided by grants from the anonymous Poliomyelitis Donation of the Hooper Foundation.

¹ Vinson and Petre, *Botan. Gaz.*, 1929, **87**, 14.

after removal of the phosphates, sulphates, pigments and most of the protein contained in the plant juice. It was thought of interest to apply certain of their methods to the virus of poliomyelitis, although the one virus was of plant and the other of animal origin. Certain modifications in technique were necessary, however. Strict asepsis had to be practiced while care had to be taken in neutralizing the end product and in pulverizing any precipitate before inoculation. The necessity of intercranial injection into monkeys required particular attention as to the amount, the character and the toxicity of the substance given.

A Berkefeld V filtrate of a 10% saline suspension of monkey poliomyelitis virus was employed. Definite proportions of the chemicals were added to a standard amount of this filtrate, usually 50 cc. Whenever a precipitate was used, it was repeatedly washed with saline, ground in a sterile mortar with a definite amount of saline and then filtered through sterile gauze before intercranial inoculation. If a supernatant fluid was given the reaction was adjusted to pH 7.0. A control monkey injected with the virus filtrate was always added to each experimental series. Tests for proteins, carbohydrates and occasionally for lipoids were made whenever sufficient material was available. Daily observations, including rectal temperatures were taken on each monkey for 2 to 3 weeks unless the animal developed typical poliomyelitis after the usual incubation of 5-7 days. Histological sections of the cord and brain were examined from all animals succumbing to the disease.

Certain experiments were successfully repeated, while others required numerous attempts before a positive result could be obtained, as indicated by the presence of typical symptoms of poliomyelitis in the monkey: rise in temperature, excitability, tremor, followed by either a partial or complete flaccid paralysis and prostration.

In 2 experiments the poliomyelitis virus was present, both in the supernatant fluid and in the precipitate after treating the filtrate with a solution of 1% aqueous safranin. It was precipitated by the latter after removal of most of the proteins with basic lead acetate.

On 3 occasions the virus was demonstrated in the supernatant fluid after treating the filtrate with basic lead acetate and the further addition of either barium acetate or of sodium bicarbonate. The heat and acetic acid and the ammonium sulphate tests for protein were negative but the ninhydrin was slightly positive in each case.

In 6 experiments the virus was precipitated from the filtrate by

equal parts of acetone (Baker's C. P.), while the monkeys injected with the supernatant fluid did not develop poliomyelitis. The ninhydrin tests for protein were either negative or only very slightly positive with these precipitates.

In one experiment the virus was recovered both in the supernatant fluid and in the washed precipitate after treating the filtrate with acetic acid to pH 5.0 and heating to 55°C., while in another the virus was found only in the supernatant fluid after acidifying to pH 4.4 and heating to 58°C. The washed precipitate from the latter was negative. Heating to 71°C., to 75°C., and to 78°C., destroyed the virus.

After a number of unsuccessful attempts it was most interesting to find the virus in the final precipitate after treating the filtrate with lead acetate and sodium bicarbonate and then leaving it on ice over night following the addition of equal parts of acetone. Monkeys injected both with the supernatant fluid after the removal of the major part of the proteins and with the acetone precipitate developed poliomyelitis. The latter gave a negative ninhydrin test.

In the same experimental series a potent virus was obtained in the precipitate which formed following acidification of the filtrate to pH 4.4, heating to 58°C. and holding it on ice over night with equal parts of acetone. The washed acetone precipitate was ninhydrin negative and produced a typical flaccid paralysis in a monkey.

Several attempts were made to extract a potent virus from the acetone precipitate with 95% alcohol and with ether. Both the evaporated extractive material and the residue failed to infect monkeys. Negative results were also obtained after several attempts both to precipitate the virus with different proportions of absolute alcohol and to recover it in the supernatant fluid after adding varying amounts of phosphotungstic acid.

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Metabolism Studies on the Brucella Group. II. The Fermentation of Monosaccharides.

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It was generally believed that members of the Brucella group lack the power to ferment the common sugars until McAlpine and