

ber plants at the time of inoculation, and cucumber varieties. However, the following symptoms were commonly expressed after inoculation on the cotyledons of young cucumber plants on which the primary leaf was just beginning to unfold and when the plants were kept in an air-temperature range of 20–28° C: From 2 to 4 days after inoculation, small, round, yellow rings appeared on the cotyledons. These rings soon became yellow blotches that coalesced to form a marked mottle. The cotyledons usually persisted as turgid functional organs for many weeks, in contrast to those of normal plants, which soon became functionless, turned brown, and withered. Within 24–48 hrs after symptoms developed on the cotyledons, yellow spots began to appear on the unfolding leaves, beginning at the base of the leaf. The spot symptoms usually were followed by the development of yellow rings, mottle, and crinkle of the affected leaves. Occasionally the primary leaves wilted and died. The apical growing point was killed very quickly, and numerous plants have been maintained for several weeks with only the two cotyledons and the primary leaf. About 30–45 days after inoculation, bud proliferation, without elongation, was apparent in the axis of the killed growing point. Many flowers and dwarfed leaves developed in a very compact rosette. In a few instances, after a prolonged period of high greenhouse temperatures, several of these badly rosetted plants developed weak, spindly shoots.

A limited number of inoculations from cucumber to cherry were made in the greenhouse late in the season of 1948 by placing small pieces of cucumber leaf under the bark of cherry trees. Definite symptoms of necrotic ring spot developed on leaves of one of 6 cherry trees so inoculated. The diseased cucumber plant in this case had been inoculated from a cherry tree known to be affected by both necrotic ring spot and yellows. Ring spot symptoms appeared on one of 3 cherry trees similarly inoculated at the same time with leaf tissue from sour cherry showing necrotic ring spot, and the 3 uninoculated control trees showed no symptoms. The conditions of these experiments were evidently marginal for transmission of necrotic ring spot.

Final conclusions regarding the identity of the virus (or possibly viruses) that incites the disease on cucumber have not yet been reached. The symptoms on cucumber, the single case of transmission from a cherry tree known to be affected by necrotic ring spot but not by yellows, and the single case of apparent transmission of necrotic ring spot from cucumber to sour cherry strongly suggest that the necrotic ring spot virus incites the cucumber disease. However, the possibility is not excluded that another virus (or viruses) from sour cherry may be involved. Since the period of incubation for cherry yellows is long, the cherry trees inoculated from cucumber cannot be read for possible yellows symptoms until 1949. Further work on the identity of the virus (or viruses) that incites the cucumber disease is now in progress.

This, so far as we know, is the first mechanical transmission of a stone fruit virus disease and the first

transmission of a virus disease from sour cherry to an herbaceous host.

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Crystallization of Hypophyseal Growth Hormone¹

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Some time ago we described a method (2, 3) for the preparation of the anterior hypophyseal growth hormone in pure form from ox pituitaries. Although it was not



FIG. 1. Crystalline hypophyseal growth hormone (×125).

a crystalline preparation, physicochemical and biological studies then and since have indicated that it is a pure protein. Recently Fishman, Wilhelmi, and Russell (1) reported that a crystalline pituitary protein with high growth activity may be obtained by alcohol fractiona-

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tion; the crystalline preparations consisted of two components as revealed by electrophoretic analysis. In this communication we wish to report the crystallization of growth hormone from our pure amorphous material by a technique similar to that of Fishman, *et al.*

Approximately 0.1% of the pure growth-hormone solution was adjusted to pH 10 with calcium hydroxide solution and brought to an alcohol concentration of 10% by a slow addition of 1:1 alcohol-water at 2° C. A small amount of the hormone was precipitated out and removed by centrifugation. The supernatant usually has a pH 8.5; if not, it was adjusted to this pH with 0.1 N HCl. Alcohol-water (1:1) was again added very slowly until the alcohol concentration was 15%. On standing at 2° C, crystals appeared as thin plates (Fig. 1). The crystals were highly soluble at room temperature and disappeared quickly during microscopic examination. To obtain a satisfactory crop of crystals, the protein concentration must be low, and the temperature should be below or at 2° C.

When crystalline preparations were assayed by the body growth or tibia test on hypophysectomized rats, there appeared no difference in their activity as compared with that of the starting material, indicating that further concentration or "fractionation" had not been achieved by crystallization. Electrophoretic analysis of the crystals gave results identical with those obtained with the amorphous pure preparation.

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Six-Segment Head Regenerates in an Earthworm, *Eisenia foetida* (Savigny) 1826¹

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In heads regenerated by *E. foetida* after excision of 6 or more segments there are at most, according to Morgan (4), only 4 or 5 segments. A similar limitation of segment number in head regenerates (1) has been assumed to be characteristic of *E. foetida* and other earthworms of the same family. There are in the literature, however, at least two records of greater numbers of segments in head regenerates of this species. Morgan (5) included in tables two 6-segment head regenerates observed 4 and 6 months after operation. In one, the 12 anterior segments had been excised. The number of segments re-

moved from the other was not counted at the time of operation but was estimated later to be 10(3). In 1898 Michel (2) reported one head regenerate of 6 segments after the removal of 8 and one of 7 after the removal of 7. Both were obtained in less than 4 weeks.

In the present study 30 specimens of *E. foetida* from Virginia were operated on in January and February. All were clitellate animals. The presence of male pores on segment xv and the clitellum in normal position were ascertained (5). Worms were anesthetized in dilute chlorotone and the 10 anterior segments amputated with a razor blade exactly at intersegmental furrow 10/11. Animals were examined daily at first and then twice a week. Specimens were fixed after 4-8 weeks.

Twenty-one animals survived the operation, and each regenerated a head. The number of segments in the 10 normal head regenerates was as follows: 4 in 2 specimens; 5 in 5 specimens; and 6 in 3 specimens. The 11 remaining worms had head regenerates of 3-5 normal segments plus one or more partial segments.

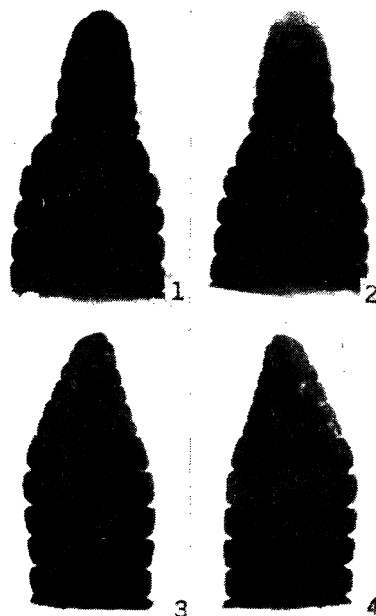


FIG. 1. (1 and 2) Dorsal and ventral views of the same 6-segment head regenerate. (3) Lateral surface view of another 6-segment head regenerate. An additional furrow shallower than the others can be seen in the proximal part of segment ii. (4) Same specimen as (3) but with the focus at the median plane. All specimens were in Cellosolve and photographed $\times 17$.

Among the 10 normal head regenerates, three exceptions were found to the generally accepted statement concerning head regeneration in *E. foetida*. These are the first 6-segment head regenerates to be recorded for this species after amputation exactly at intersegmental furrow 10/11. Dorsal and ventral views of one of these are shown in Fig. 1 (1 and 2). Another specimen, cut through the median plane and photographed in Cellosolve, is shown in Fig. 1 (3 and 4). Segment ii (3) has

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