

coincident for a relatively short period of time only, a few white "spottings" at a distal end of the leaflets may result; if the time element is lengthened, all the cells of the leaflet may suffer a water loss much below the wilting coefficient, and instead of a "spotted" appearance the entire leaflet will bleach white. This is exactly what happens under both experimental and field conditions. In the intermountain country where a very large number of observations have been made, it has been noted that fields showing a considerable incrustation of alkali when irrigated exhibited white spot in more or less amount, depending upon the other environmental factors above mentioned. Also, a sudden rise of the water table in irrigated districts has brought about the same appearance of the plants in the fields. Some very interesting observations have been made on fields adjacent to each other, with plants of the same age and all conditions the same excepting the application of water. The irrigated fields showed extensive white-spot trouble, while the non-irrigated fields showed none.

It has been noted by eastern pathologists who have made observations on this disease that it occurs mainly in the spring of the year. However, the writer has observed it in the intermountain districts during the early spring, during mid-summer and during the late fall; in short, throughout the entire growing season.

Specimens of artificially produced white spot of alfalfa were submitted to several plant pathologists who reported that these specimens were identical with diseased alfalfa plants which they had themselves collected.

An extended report will be published in due time after the completion of certain experiments which are now in progress.

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THE POLYHEDRAL VIRUS OF INSECTS WITH A  
THEORETICAL CONSIDERATION OF FIL-  
TERABLE VIRUSES GENERALLY

IN a previous paper<sup>1</sup> J. W. Chapman and I called attention to the fact that the wilt or

<sup>1</sup> *Biol. Bull.*, Vol. XXX., No. 5, pp. 367-390.

polyhedral disease affects many different species of insects. We also showed that the disease is not produced by bacteria, but is caused by minute organisms capable of passing through diatomaceous filters, and further attempted to demonstrate that the polyhedral bodies always found associated with the disease are not organisms, as supposed by Bolle, Fischer, Marzocchi and Knoche, but reaction bodies—simply nucleoprotein by-products.

In order to satisfy myself that we were really dealing with an organism and not merely with an enzyme, toxin or other material a large series of passage infections were instituted. Twenty-five gipsy moth caterpillars were infected at a dilution of 1:1,000 with material obtained from a caterpillar previously dead of wilt. The animal was merely ground up, sterile water added and the whole filtered through a sterile Berkefeld grade "N" candle. All twenty-five caterpillars fed with the filtrate died typically of wilt during the course of three weeks, whereas twenty-five controls fed with the autoclaved filtrate lived, pupated and transformed into moths. One animal dead in this first series was prepared, the material diluted as before (1:1,000), filtered and fed to another series of twenty-five caterpillars. The experimental animals all succumbed, whereas the controls did not. Third and fourth passage infections were performed and the results were similar with the exception that the period from infection to death was considerably shorter at the fourth passage than at the first three. This shortening of the time between infection and death seems to point towards an increase in virulence with successive passages.

There are certain autocatalytic substances like chromatin that increase progressively, so to the physiologists my passage infections may not necessarily be proof for the contention that I am dealing with parasitic ultra-microscopic organisms. However, if one reviews the field of the filterable viruses<sup>2</sup> and compares all of the results obtained by other workers with my results, one can not but feel inclined to

<sup>2</sup> Thirty-two diseases are now known to be caused by filterable viruses.

adopt the view that one is dealing with minute parasitic forms. Some of the filterable viruses (pleuro-pneumonia of cattle, fowl pest, fowl diphtheria, epithelioma contagiosum and Novy's rat disease) have been cultivated, so that the question as to whether we are dealing

realize Osborn's<sup>3</sup> "hypothetical chemical pre-cellular stages"; they lie somewhere in the scheme between simple colloidal and more complex cellular states like bacteria. Some thirty-two or thirty-three disease-producing filterable viruses are now known to exist, so it

TABLE SUMMARIZING CHARACTERS OF POLYHEDRAL VIRUS OF INSECTS

1. Cultivation of virus	Has not been cultivated
2. Filtration of virus	Passes through Berkefeld "N" but not through Pasteur-Chamberland filter
3. Examination of virus with ultra-microscope.	Nothing visible that could be interpreted as being different from minute protein or pigment particles
4. Effect of heating on virus when suspended in water	Destroyed at 60° C. in 20 minutes
5. Effect of dry heat on virus	Destroyed at 70° C. to 80° C. in 20 minutes
6. Effect of drying on virus at room temperature.	Resistant for 2 years
7. Effect of glycerine on virus	Resists 98 per cent. for 6 months
8. Effect of direct sunlight on virus when dry	Resistant for 12 hours
9. Effect of putrefaction on virus	Resistant for an indefinite time
10. Effect of alcohol on virus	Destroyed by 80 per cent. in 15 minutes
11. Effect of carbolie acid on virus	Destroyed by 5 per cent. in 3 weeks.
12. Effect of virus on 1 per cent. sugar solutions	No growth, no fermentation
13. Effect of virus on methylene blue and sodium nitrate solutions	No growth, no reduction
14. Effect of virus on gelatin and casein	No growth, no liquefaction

with organisms or not is solely an academic one. We are justified at present, however, in not classifying such viruses either with the plants or animals.

The table gives a summary of the chief characters of the wilt virus. The virus used in these tests was prepared from diseased gipsy moth, army worm and tent caterpillars. That proteins like gelatin and casein are not affected when treated with the filtrate in which the virus has been concentrated by centrifuging is curious because insect tissue is completely emulsified through the action of the wilt virus. This action is therefore probably a cytolytic one due to the action of toxins and is not caused by the elaboration of a proteolytic enzyme on the part of the virus.

In a physico-chemical explanation of the origin of organisms on our planet the filterable viruses seem to be of considerable interest and I do not understand why they seem to be so persistently neglected by all writers on the evolution of life. The filterable viruses probably

is reasonable to assume that the earth, water and atmosphere are full of non-parasitic forms which we have no means of recognizing at present.

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<sup>3</sup> "The Origin and Evolution of Life," by Henry Fairfield Osborn, Charles Scribner's Sons, New York, 1917, p. 80.

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