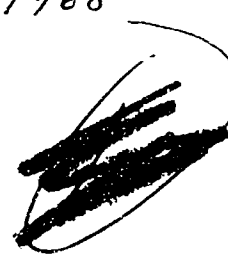


AD 685300

TRANSLATION NO. 611

DATE: July 1968



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THIRTY YEARS OF STUDYING VIRUS DISEASES
IN PLANTS IN USSR

V. L. Ryzhkov

The great October revolution which has transformed all aspects of life in our country, has brought into being new developments in old cities, and new cities in deserts and woods. It has also created new branches of economic and spiritual activity, among which we may list the study of virus diseases in plants. This is correct even though the first experimental investigations of virus disease in tobacco were conducted at the end of the last century by D. I. Ivanovsky. Following the discoveries of this pioneer in the study of filtrating viruses there is a long interval, and published words in the Russian language on the subject of virus diseases in plants begin to appear again only after the October revolution.

The founder of the Russian phytopathology A. A. Yachevsky published in 1925 a pamphlet dealing with the virus diseases in potatoes. The first experimental work which was carried on in the USSR after Ivanovsky deals with infectious chlorosis in the spindle tree and was published in 1927. Somewhat later Ukrainian phytopathologists conducted a large scale investigation of mosaic blight in beets, resulting in a compilation of works published in 1930 under the editorship of V. P. Muraviev. In the same year, under the leadership of the author of this article, at the initiative of T. D. Strakhov, there was organized in the Ukrainian Institute for Plant Protection the first laboratory for the special study

of virus plant diseases. During subsequent development of work in this direction the publication of special monographs acquainting the Russian reader with the literature of the world (1933, 1935) was of great importance, as well as a series of conferences in Kharkov in 1935, also in Moscow in the same year, and then again in Moscow in 1940.

V. A. Riakhovsky took upon himself the task of compiling a Russian bibliography on the virus diseases of plants, and gives the following figures:

<u>Years</u>	<u>Works published</u>
1892-1917	15
1918-1928	20
1929-1933	34
1934-1938	254
1939-1940	122

This table shows that the majority of Russian publications came out since 1933, after appearance of the first monograph devoted to this problem.

The facilities granted by the Government to Soviet scientists made this rapid development possible. During a short time an enormous amount of work has been done in the study of virus diseases occurring in our country, whereby some cultural plants were studied in particular detail.

I. P. Khudyna studied and identified the principal diseases of the tobacco plant, and preventive measures were devised to combat these diseases.

The virus diseases of the tomato plant are discussed for the first time in the works of V. L. Pyzhkov and I. K. Kovachevsky, and simultaneously with Australian authors they published a treatise on the nature of dig-bud.* K. V. Sukhov and A. M. Vovk, V. K. Fazhurilo and G. M. Sytnikov studied some very interesting virus diseases in gramineous plants, which

* Dig-bud is called in Russian "stalbur" and is transmitted by *hyalosthis obsoletus*. Translator's note.

were new to science, rosette blight of oat, and the mosaic disease of wheat. V. A. Kreutzberg has described a disease new to science of the pistachio nut. Here should also be mentioned the works of E. D. Yakimovich on virus diseases of soya, of O. V. Vertogradova, O. B. Vatalina, and others, on the virus diseases of berry plants; of K. S. Rakhlis, on the virus diseases of hops; of K. V. Yatsynin, on the mosaic blight of the cucumber; of E. A. Ositskaya and M. I. Goldina on the tomato blight, etc.

In the various republics of the Soviet Union local centers have arisen for the study of virus diseases. D. D. Verderevsky in Azerbeidjan has discovered the curly leaf of the cotton plant, which has been thoroughly studied by S. V. Moskovets and his collaborators, other virus diseases in this locality having been studied by S. I. Shipinova. Under the leadership of S. A. Konchivelli a serious study of virus diseases in Georgia was conducted by E. A. Eristavi. In Armenia studies along these lines were made by D. V. Babayan, A. V. Kirokasyan; in Bielsrussia, by R. M. Pivovarova, and others.

The first Russian investigations of insects, carriers of virus diseases, can be attributed to A. I. Vavinenko, who ascertained the mosaic blight of beets. K. S. Sukhov and A. M. Vovk found the carriers of the rosette disease, the yellows of Kok-saghyz, and dig-bud. V. K. Fazhurils and G. M. Sytnikova discovered the carriers of the mosaic blight of wheat, V. A. Kreutzberg ascertained trins responsible for the propagation of the rosette disease in the pistachio nut. V. I. Krivik worked with plant lice, carriers of the virus disease of potatoes.

Virus diseases of plants have been studied very diversely by Soviet scientists. Of interest are the pathologic l, anatomical works of E. Ia. Rokhlina, P. V. Mikhailova, and others. The anatomical method makes it possible to diagnose disease of the nature of dig-bud in the tomato, even

in cans and other foodstuffs prepared from diseased tomatoes.

In cytological works special attention was paid to crystalline inclusions. M. I. Goldin described new types of crystals in the mosaic disease of the tobacco plant, using a micromanipulator in studying them. K. S. Sukhov and A. M. Vovk studied virus crystals in the mosaic blight of wheat in rosette and breaking disease of the tulip. Many facts have been found facilitating the diagnosis of virus diseases from crystals.

Pathophysiological studies have always occupied an outstanding place in Soviet investigations of virus diseases. The first writings along these lines dealt with the transformation of matter in dig-bud, (I. K. Korachevsky) and catalysis in virus diseases of the tobacco plant and potato (V. L. Ryzhikov and I. K. Korachevsky.) Mention must be made of the extensive studies of V. F. Kuprevich, and also of A. I. Kokin. L. Kh. Kara-Murza has studied systematically the physiology of curly leaf in the cotton plant. The phosphorus metabolism in virus diseases of plants was first studied in the USSR, and here too the infiltration method for the study of the fermentative activity of diseased plants was first applied. Likewise experiments were conducted for studying diseased plants under conditions of starvation (V. L. Ryzhikov and collaborators.) As a result of these works it was not only possible to define from a pathophysiological point of view a whole series of virus diseases, but likewise to obtain general results. Thus it was established that with the mosaic disease in tobacco plants there is a sharp increase in the relationship of albumin nitrogen to albumin phosphorus, which points to a decrease of phosphorus in the diseased plant. It has been shown that with a lack of nitrogen, plants affected with mosaic disease expend nitrogen more economically than healthy plants, etc. Interesting data obtained by L. H. Kara-Murza confirm the origin of tonic

substances from hydrocarbon. The examples mentioned show the importance of the Soviet school in the blazing of new trails for the pathophysiology of virus diseases in plants.

Sereological studies were first conducted by B. P. Matsulevich in the laboratory of T. I. Fedotov. Upon organization of a virus laboratory in the Moscow Station for Plant Protection, its manager, Prof. M. S. Dunin has paid much attention to the development and improvement of the sereological method. The drop method of this author permits to apply serodiagnosis under field conditions. P. A. Gerasimov, T. L. Kudriavtsev, E. P. Shatova have worked on improved preparation of various serums.

A characteristic feature of Soviet works dealing with virus diseases is the emphasis placed on attempts to combat them. Previously it has been mentioned that I. P. Khudyna has devised a system of measures contributing to a drastic decrease of virus diseases in the tobacco plant. M. F. Ternovsky and I. P. Khudyna, by means of interspecies hybridization, have developed a clever method permitting to remove the virus of mosaic disease from the surface of tomato seeds. Works of the same authors have brought about a system of methods for combatting virus diseases of tomatoes under conditions of protected ground. The curly leaf of cotton plants was a serious threat to crops in Azerbeidjan in certain years, but thanks to the works of S. M. Moskoveto and collaborators it was possible to reduce the disease of cotton plants to insignificant proportions.

To Soviet scientists may be ascribed a great many new ideas in the combatting of virus diseases. The feature which all these ideas have in common was the attempt to act on the virus diseases through changes in ecology. In this connection mention should be made of the work of I. K. Korachevsky who proposed dense plantings in order to combat dig bud, a

definite watering regime, a mulching of soil, etc.; also of the works dealing with rosette disease by S. D. Grobennikov, Sh. Sh. Khairullin, and others.

Aside from work of a phytopathological character there also was developed in the USSR an original slant in investigations of filtrating viruses. Soon after Stanley segregated the virus of the tobacco mosaic disease in the form of a pure albumin preparation, we obtained this virus by original method, which as the works of E. P. Shvetova have shown, may also be utilized in obtaining the virus of the mosaic disease in potatoes. World science, including the Soviet scientists, were faced by the fact that some of the filtrating viruses were nucleoproteids capable of forming real crystals or paracrystals. The filtrating viruses which in the course of a great many years would escape us through the pores of the chamberlain candle, and which gave rise to many suppositions, were now in our hands. The widest possibilities opened before science, and a serious responsibility rested on the shoulders of investigators. What means to chose in order to make these new unusual facts understandable and reconcile them with our existing concept of life?

The work of foreign investigators was conducted principally along the lines of studying physical and chemical properties of virus protein. Because all efforts were concentrated in this direction in a short time virus albumins were studied chemically and physically not less thoroughly than accessible albumin matter. However, very little attention was paid to the biological activity of virus albumin and to the problem of self-reproduction, while these very questions were given the center of attention by Soviet scientists, who deserve credit for devising plans in conducting research, for original methods of research and for obtaining the first significant results.

According to the plans providing for these investigations, it was necessary to find out first of all whether some biological or biochemical activity could not be ascertained in vitro. The first investigations conducted by V. I. Ryzhkov and K. S. Sukhov revealed no fermentative activity in preparations of virus albumin, which apparently remains true to this day, because at times indications of the fermentation of certain proteins may be conditioned by the addition of corresponding ferments. Another series of experiments was devoted to attempts at binding active groups of virus albumin and at ascertaining upon what fundamentals the biological activity of the virus rests. These investigations conducted by P. A. Agatov produced results similar to those obtained by foreign scientists which were published about the same time.

The desire to ascertain the place occupied by virus albumin among other plant albumins has made us search for ways of dividing these plant proteins. We were not able to ascertain that to the extent that virus albumin would accumulate in a plant the quantity of other albumins would decrease. Later however, as mentioned above, it was possible to demonstrate that with the accumulation of virus albumin, the plant proteins are subjected to a far-reaching qualitative change.

The study of the pathophysiology of the diseased plant helps but little in ascertaining the biochemical shifts which cause an accumulation of virus albumin, because it is impossible to exclude at the same time the occurrence of deep secondary changes resulting from the diseased condition of the assimilatory apparatus in the plant, and other symptoms.

In order to study the physiological conditions of virus accumulations we suggested a method which has been called "method of halves." The isolated tobacco leaves are rubbed with juice containing virus and are then cut along the main stem, one half of the leaf being used for experi-

ments, and the second for control.

There have also been conducted planned investigations of the physiological conditions of virus accumulation. First of all this means studying virus accumulation under loss of nitrogen and phosphorus. These experiments by Ryzhkov and Smirnova have definitely shown that under conditions of starvation the virus albumin is not subjected to hydrolysis, the virus titre being as high as in normally feeding plants.

The latter conclusions however have been contested by American authors, and at the present time the question has been greatly complicated by the fact that the existence has been proven of a plant virus in bound form, whereas previous experiments have shown only a soluble virus. Virus in plastids was found by Ryzhkov and Smirnova, was later confirmed in English investigations, which have revealed a great quantity of virus in plant fibers.

When we started this work it was necessary to ascertain the most elementary questions, because we were working on virgin territory. As a result it was possible to determine the temperature limits of virus accumulation, the interrelationship between the accumulation of virus and partial pressure of oxygen, accumulation of virus under conditions of narcosis, etc. Systematic investigations were made in order to determine to what extent the accumulation of virus depended upon various fermentative systems. We used inhibitors of fermentative systems and obtained a whole series of negative results; for instance we determined the independence of the selfreproduction of virus from glycolytic processes, from ferments acting with sulphuric combinations and pyrophosphate, etc. At the same time substances were discovered capable of suppressing the accumulation of virus, such as, thiamin, dinitrophenol, and some acridinic preparations. The study of the relationship of virus accumulation from cations has led

to an unexpected discovery that some cations suppress the accumulation of virus, while others do not have these functions. There was nothing analogous in any foreign literature, not counting works dealing with bacteriophages.

Filtrating viruses are of scientific interest not only as such, but as a model for the study of various general biological problems. It is from this point of view that the liquid virus crystals have been studied. It had been found that the hydrophilic colloids cause a segregation of virus in the form of liquid crystals which, together with other similar ones, were studied as a model for paracrystalline structures of cytoplasm. For a long time it was not clear how crystalline virus deposits are formed in the cell of the diseased plant, because outside of cells it was possible to obtain virus of a crystalline formation only in abiological conditions (low pH, great concentrations of ammonia sulphate.)

In original phase in the study about viruses is the influence exercised by viruses on the formation process in plants. A whole series of experiments have shown the possibility of obtaining antolyses by means of viruses, and on hand of the material affected by virus it became possible to study various regular processes in the flower formation. Lately it has become fashionable to juxtapose virus and genes, as a result of which some American investigators arrived at the most extreme conclusions. Thus Farlington maintains that there is no fundamental difference between infection and heredity, while Wood assumes that the genetically conditioned heterogeneous form may become infectious. In our Soviet literature questions of this sort had been discussed in detail, even before the discovery of the albumin nature of virus, witness the monograph "Mutation and Diseases of the Chlorophyll Grain" (Moscow, 1933.)

We believe that these works have demonstrated the futility of doing away with the principal difference between heredity and infection. There is no necessity of adding anything at all to these facts obtained more than 10 years ago. The conclusions of Darlington and Wood remind of the pre-Pasteur teaching of self-generation, and show a deterioration of scientific thought.

In making general conclusions and wishing to define the most significant points for the comprehension of viruses by Soviet scientists, it must be stressed that our investigators had no part in the separation between viruses and the biological medium in which they circulate. Although many filtrating viruses were capable of crystallizing with albumin, we considered it incorrect to regard the study of viruses as a chapter in biochemistry. Viruses are not only matter, but matter of a special order, capable of parasitism, equipped with properties having arisen in the process of the struggle for survival and selection. This permits to speak of the divergence of viruses and to demand a natural phylogenetic system for them. It shows that viruses, owing to the very methods of studying them, are on the harder line between living and non-living.

↘ Thirty years have gone by, during which Soviet investigators have classified the majority of virus diseases, have described many new virus diseases in plants, studying their propagation in nature, prescribing new and original means of combatting them, using simple and effective methods of investigation; finally they have taken their place in world science by means of their own progressive concepts on the nature of virus, which enables to express all qualitative peculiarities of these remarkable albuminous, crystallizing parasites which autonomously circulate in nature and enter into most complex relationships with various insects and plants.

Institute of Microbiology of the USSR Academy of Sciences,

Moscow, 17 July 1947