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Aggressiveness of Phytopathogenic Viruses: Trace Investigation and Evaluation

Antoniy Stoev

N. Poushkarov Institute for Soil Science, Agrotechnology and Plant Protection,
7 Shosse Bankya Str., 1080 Sofia, Bulgaria

Abstract

The paper deals with the particularities in the manifestation of aggressiveness by phytopathogenic viruses and the methods of its tracing out and evaluation. Aggressiveness is a property of phytopathogens. We present some examples of viral pathogens which cause economically important diseases in the agroecosystems of Bulgaria. We have considered the manifestation of virus aggressiveness in association with other properties which characterize plant pathogens.

Key words: phytopathogenic virus, viral pathogen, aggressiveness

Резюме

Статията представя особеностите в проявата на агресивността на фитопатогенните вируси, както и начините за нейното проследяване и оценяване. Дадени са примери с вирусни патогени, причиняващи стопански важни болести в агроекосистемите на България. Проявата на агресивността е разгледана във връзка с други свойства, характеризиращи растителните патогени.

Introduction

The development of the pathological process depends on the properties of pathogens and the plant immune system. The infectious virus is the virus which is determined by its ability to introduce itself into the living organism (Markov *et al.,* 1989). The viral pathogen is in the closest link with the hospitable cell of the plant organism. Under conditions of viral infection, the cell metabolism is directed towards realizing the ability of the viral genome, and eventually towards producing the pathogen (Goldin *et al*., 1966; Fraenkel-Konrat, 1969).

The pathogen possesses a definite phylogenic specialization. The infection which characterizes the beginning of the pathogenic process spreads in the frames of specific taxonomic units of the hosts (Table 1).

The pathogenic population (if it is possible to use the term of viral population) is not homogeneous. In the population there are smaller systematic units of strains which differ in their infective ability and the ability to cause diseases: for example, necrotic strains causing some necroses in plant tissues, etc. This relative ability is well-known as virulence.

Viruses penetrate plant tissues via different pathways. The tobacco mosaic virus (TMV) penetrates through micro-wounds formed after mechanical damage to the tissue.

Other viruses [such as: plum pox virus (PPV), barley yellow dwarf virus (BYDV), and raspberry ringspot virus (RRSV)] penetrate through the sucking organs of insects and nematodes using the plant sap for feeding (Esau, 1961; Trifonov, 1972; Gibbs and Harrison, 1976; http://www.eppo.int/QUARANTINE/data_sheets/virus/RPRSV0_ds.pdf).

Virulence and aggressiveness depend on the ability of the pathogen to overcome the protecting system of the plant organism. Aggressiveness characterizes the development of infection and the ability of the pathogen to spread into the tissues of the host plant (Gorlenko, 1973, according to Markov *et al.,* 1989).

Methods to follow the spreading of virus

The visual following of viral spread depends on the appearance of disease symptoms. These can

*Corresponding author: e-mail: anton_stoev@yahoo.com

Table 1. Infection ability and aggressiveness of phytopathogenic viruses

Infective ability	
Ability of virus to infect the plants	
It depends on:	
(a) immune reaction of the plant	
(b) phylogenic specialization of the pathogen, which can be:	
Wide – The pathogen infects plant species of different botanical families.	Narrow – The pathogen infects plant species in the frame of one botanical family.
(c) Ontogenetic specialization – The **i**nfectious ability is depending on the plant age (phase of development).	
Aggressiveness	
Ability of virus to disseminate in the plant	
It depends on:	
(a) immune reaction of the plant	
(b) specialization of the pathogen, which can be:	
Histotropic	Organotropic
The virus spreads in some tissues of the plant	The virus spreads in some organs of the plant

appear on certain plant organs under conditions of infection after the expiration of the incubation period. This way is uncertain because the incubation period for the various viruses has different duration which depends on many factors. These factors are not considered in this paper.

It is possible for the infection to remain latent. This is a problem concerning the quarantine and the producing of virus-free propagation material.

The visual observation of the viral spread can be made visible under microscope. It is possible for some viruses which form specific bodies (called viral inclusions) in the plant cell. These viral inclusions vary in form, composition and texture. The differences can help us to determine the systematical belonging taxonomic affiliation? of the virus.

For TMV, these viral inclusions are visible under a light microscope (Goldin *et al.,* 1966). Discovering the real viral particles needs the application of electronic microscope (Matthews, 1970; Protsenko and Legunova, 1960).

Additional helping method is the use of dyes for colouring the substances which are accumulated in the plant tissues as a result of the infection. As diagnosis reference point, some intrinsic deformations can be used, for example, tylosis in the xylem (Kovachevski *et al*., 1995).

Another method for visual following of virus spreading is by biological testing. In this method, plants are used which are indicators of viral infection (Matthews, 1970; Gibbs and Harrison, 1976). These plants respond to the supposed carrier of viruses in a specific way with inoculum prepared from different tissues and organs of the carrier. The biological test is reliable for proving and tracing the infection. Its application needs time and availability of vegetation greenhouse or specialized plantations, which is a disadvantage of the method.

The modern methods for determination of plant viruses are based on immunization reactions between virus and antibody. Plants cannot form antibodies. They are produced in an animal organism injected with prepared and purified viral substance. These methods create the possibility for artificial production of viral nucleic acid and give quick and exact evidence of the presence of a virus (Starke, 1968; Clark and Adams, 1977; Wetzel *et al.,* 1991).

Evidence of viral infection can be expressly obtained by applying diagnostic kits out of the laboratory in crop fields and plantations (Stoev and Tomeva, 2005).

Determination of virus aggressiveness depends not only on the chosen method and diagnostic reagents. The researcher has to know the defense reaction of the plant which is in contact with the viral pathogen. The plant might manifest a hypersensitivity reaction. In this case, plant cells die (Corbett and Sisler, 1964; Gibbs and Harrison, 1976; Kegler and Kleinhempel, 1987; Hartmann, 2001).

A necrotic zone can be formed which stops the development of the infection process. The hypersensitivity reaction can lead to perishing of the plant organs and even of the whole plant when the necrosis develops in vital parts of the plant.

All these cases require mutual taking into consideration of the experimental setting and the natural and agrotechnical conditions which exert influence on the plant health.

Features in the manifestation of aggressiveness

Some viruses (TMV) which infect the plant through micro-wounds formed by mechanical impact are disseminated and spread in the whole plant (Samuel, 1934 according to Gibbs and Harrison, 1976).

For the receptive variety of tobacco, the virus is accumulated in the leaves which manifest the specific symptoms of the disease. These symptoms gave the name of mosaic virus and infection of tobacco (Kovachevski *et al.,* 1995). In the sphere of hosts of *Nicotiana* genus, the infection can provoke the appearance of necrotic lesions (Gibbs and Harrison, 1976).

TMV keeps its infection ability in the plant‘s sap. When there is a contact between an infected plant and an uninfected one, the infection can go over to healthy plants. Under natural conditions, the wind can cause the contact. When feeding, the natural carriers of infection can be some gnawing insects, passing from the diseased plant to a healthy one.

Different processes in growing tobacco can cause the opening of micro-wounds, which contribute to spreading the infection into plants of field, where the seedlings are grown, as well as in the future plantation (Kovachevski *et al*., 1995).

For viruses which have natural carriers (as leaf-lice, etc.), the spreading of the infection depends on the attractiveness of the plant hosts for the carriers.

Crop plant species have great diversity of varieties. They differ by many indices. Among these indices is the index of the plant reaction, which is due to the different viruses provoking the diseases.

Having in mind the indicated examples, it can be concluded that aggressiveness is a relative concept, which depends on many factors. Some of these factors are external to the pathogen itself. The human activity is one of these external factors. The creation of new hybrids and varieties should be considered, which are resistant to the attacks of different disease provoking viruses (Kegler *et al.*, 1998).

Resistance is coded in the plant genotype. It can be considered and systematized from different view points. Theoretically, full resistance can be assumed. In this case, there is no existence of infection. The pathogen cannot be introduced and disseminated into the plant organism. The plant possesses immunity. It is obviously healthy and the tests for presence of infection give negative results (Kegler and Kleinhempel, 1987; Vlasov and Larina, 1982).

When the plant is tolerant, the virus can affect the plant tissues and organs of the host completely or partially. In this case, it is not compulsory for the plant to give evidence of disease. From economic point of view, the tolerant property is valuable when the infection does not significantly decrease the quality and quantity of plant production. The details concerning the pathological evidence and the damaging consequences are not considered in this publication. Resistance and tolerance are different categories when characterizing the interactions between the pathogen and the host (Vlasov and Larina, 1982).

In the case of infection the following different cases are possible**:**

- duration of incubation period;
- appearance of symptoms which can disappear at a later stage and the plant is restored and looks healthy;
- manifestation of symptoms which are common pnenomena concerning some viral infections in tree species;
- manifestation of hypersensitivity, which leads to premature perishing of the infected plant;
- availability of symptomswhich vary by the strength of manifestation but the plant-host does not perish.

In the above cases, depending on its aggressiveness, the pathogen can be established in the whole organism of the plant. Its determination depends on the specificity of the chosen method for identification. The visual diagnosis of viral diseases is of secondary importance only.

Methods for evaluation of aggressiveness

Aggressiveness of viruses can be evaluated through their organotropic and histotropic specializations (Table 2). The presence of the virus in one or another organ or in the issues of the plant-host is a qualitative index (Petrov, 2014; Stoev and Kamenova, 1995).

Table 2. Estimation of virus aggressiveness

Qualitative estimation
1. According to the availability or the absence of pathogen
(a) investigation of different plant tissues, organs and parts
(б) following and measuring of the spreading velocity
Quantitative estimation
1. Taking into account the affected area (volume) of the infected organ
2. Taking into account the number of the affected organs (fruits, leaves, blooms etc.)

The virus concentration in the infected tissue is an additional quantitative indicator, which characterizes the infection (Gibbs and Harrison, 1976). The measurement of concentration is performed by applying physical and chemical methods. The number of local lesions gives an idea on the infection degree of the crude sap, which is used as inoculum (Nordam, 1973).

An indirect indicator of the virus concentration is the optical density, which is measured at the end of the enzyme-linked assay of the immunosorbent (Clark and Adams, 1977).

The movement velocity of the virus in the plant organism is another indicator of its aggressiveness (Samuel, 1934, according to Gibbs and Harrison, 1976). The formed lesions can serve as a reference point for orientating, which should not abolish the check-up concerning the presence of virus in the rest of plant tissues (Dikova, 2009, 2011, 2014; Milusheva, 2014).

According to Van der Plank (1963), described and cited by Pariaud *et al.* (2009), aggressiveness is a non-specific component of the pathogen phenomenon. From the quantitative point of view, the strength of the disease manifestation (or the pathological activities) can be characterized through the size of the lesions, according to Markov *et al.* (1989).

The aggressiveness of a pathogen has to be considered also in relation to the whole crop fields or plantation. In the areas with crop plants, the infection can be general or local. For example, in the fields with cereals, having a combined homogeneous distribution in the area (such as wheat, barley and oats), the plants can be infected in separate land parcels, looking like threshing-floor or halos (Kovachevski *et al*., 1995). In other cases, the periphery of the crop field can be affected, etc.

In plum (*Prunus domestica*) plantations created with material of saplings free of viruses, the PPV infection can be localized in separate trees. These have to be eradicated. In these plantations, sprinklings with insecticides are carried out against leaf lice vectors of the mentioned pathogen as a protecting measure (Dragoiski *et al*., 1990).

In the nursery gardens, the trees infected by viruses which can be spread through the pollen, have to be eradicated. This measure ensures the production and distribution of healthy propagative material (Trifonov, 1972).

Significance of aggressiveness for the practices

Cultivation of crop plants can be developed in different aspects. Obtaining food for humans and animals is of primary significance. Furthermore, plant production can be basic material for obtaining certain goods which are necessities of life or raw materials for different industrial branches.

Other important aspects of growing crop plants are:

- reproduction of the crop species and varieties (production of seeds and sapling material);
- selection (plant breeding), creating new hybrids and cultivars;
- variety testing and cultivar maintenance;
- domestication of new plant species.

When all these plant-growing aspects are implemented, the plants grow and develop under conditions of a fixed background of infection. The plants interact with the pathogens available in this background.

The distribution of the infection inside the agroecosystems depends on certain conditions. It has its own characteristics. These conditions and characteristics are subjects for investigation in the field of epidemiology. The possibility of mixed infections should be taken into account (Petrov, 2015).

The ability of plants to limit the spreading of viruses is of important significance for obtaining generative (sexual) and vegetative non-infected posterity. The availability of seeds, tubers and bulb which are free of viruses is an important prerequisite for obtaining a good crop yield in amount and quality.

The hypersensitivity reaction does not allow the survival of the virus in the plant organism. In this case, there is no appearance or retention of infected areas in the crop fields.

The knowledge on genetical determination of the relationships between the virus and its host enables the creation of new plant cultivars which possess not only valuable economic and market qualities of the production, but also high resistance to the virus agents of dangerous diseases in crop plants.

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