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BRIEF COMMUNICATION

A Leaf Dip Method for Routine Identification of Plant Viruses Using the Latex Agglutination Test

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Abstract. A description is presented of a simplified procedure for the serological diagnosis of plant viruses which utilizes the high sensitivity of the latex test. Small pieces of the plant examined are soaked in a drop of water, buffer, or directly in the sensibilized antiserum. Following the reaction with the antiserum the virus is evidenced by visual evaluation of the presence of agglutination. The procedure is applicable for routine serological diagnosis; it demonstrates even small virus concentrations and is as reliable as the normal procedure of the latex test. Using this simplified procedure the following viruses were demonstrated in various host plants: X-, Y-, S-, and M-virus of potato, the carnation mottle virus, and the tomato bushy stunt virus.

The procedures so far applied for routine serological diagnosis of plant viruses, are based on obtaining cell sap by homogenization of the plant to be examined or of its part. In the crude or centrifuged sap the presence of the virus is then demonstrated by means of the antiserum. The procedures of serodiagnosis are specific, rapid and less laborious than the biological tests, but in spite of this they are exacting and the centrifuging of plant extracts requires up a certain amount of electric energy. Some viruses occurring at high concentrations in host plants, may be proved by the precipitin test, which comprises the soaking of small leaf pieces in the antiserum (BALL and BRAKKE 1968, RICHTER 1971), or using the test of simple radial diffusion in agar, consisting in placing small leaf pieces into agar mixed with the antiserum in a Petri dish (RICHTER and POLÁK 1975). However, these simple methods cannot be applied practically for routine serological diagnosis, since hardly any virus reaches the necessary concentration in some of the host plants.

During the last decade the methods enhancing the sensitivity of serological reactions have been developed and applied in plant virus serology.

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For routine diagnosis the test of latex agglutination developed by BERCKS (1967) is of special importance. The latex test is hundred to thousand times more sensitive than the precipitin tests (*e.g.* YOKOYAMA 1975, OERTEL 1977, VAN REGENMORTEL 1978). The high sensitivity of the latex test was utilized in our laboratory for developing a simple serodiagnostic method for plant viruses which removes the imperfections of previous procedures.

Pieces of the plant examined (leaf or stem cuttings, *etc.*) are soaked in a drop of water, buffer, or directly in a drop of sensibilized antiserum. The virus particles get out of the damaged cells into the drop, and after the reaction with the antiserum their presence is demonstrated by (the evaluation of) agglutination. Two to four plant pieces in one drop are sufficient to prove the presence of most viruses by the latex test even in host plants with a lower virus concentration.

In contrast to the procedure of the latex test so far used (BERCKS 1967) the homogenization of the plant is omitted, as well as the centrifugation of the crude extract and application of various stabilizers. The drops containing the antigen and sensibilized antiserum are immediately shaken in a rotary shaker, and after 10—30 min (the shaking period depends on the virus and its concentration) the agglutination is visualized on a black background.

Using this simplified procedure the diagnosis was tested for X-, Y-, S-, and M-potato virus in potato plants, X-, and Y-potato virus in tobacco plants, the carnation mottle virus in carnation, and the tomato bushy stunt virus in sweet cherry with the symptoms of detrimental cancer of sweet cherry. The results were compared with those obtained using the non-modified procedure of the latex test. The reliability of diagnosis was identical, and in some cases in M- and Y-potato virus it was even higher than in the control procedure. More detailed results will be given in papers published elsewhere.

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