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# THE STRAINS OF SOYBEAN MOSAIC VIRUS OCCURRING IN SOYBEAN BREEDING MATERIAL IN CROATIA

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From different genotypes of soybean included in the breeding program of the Faculty of Agricultural Sciences in Zagreb seventeen isolates of soybean mosaic potyvirus (SMV) were isolated. Their identity was established by host range study, morphology of virus particles and serology. On the basis of reactions provoked in specific soybean differential cultivars the isolates were classified in respective SMV strains established by Cho and Goodman (1979). Most of the isolates belong to the least (Gl) or less virulent (G2) strains indicating a low degree of resistance to SMV of the soybean cultivars investigated. From the introduced genotypes three highly virulent SMV isolates were also obtained, which belong to G7 and G5 virulent strains. It is suggested that in soybean breeding programme more attention should be paid to resistance to SMV.

## Introduction

Soybean plays an important role among the crops in the agriculture of Croatia, as a source of high quality protein and oil for use in human food.

From the viruses infecting soybean, soybean mosaic potyvirus (SMV) deserves special attention because it may cause significant seed yield losses, affects the nodulation (T u et al. 1970) and delays maturity (T u 1989). Owing to its high transmission rate in many genotypes and transmission by aphids in non-persistent manner, SMV is widespread all over the world. It occurs in a variety of symptoms, from mild to severe (Fig. 1), depending on the virulence of strains and on soybean cultivars. C h o

and  $G \circ o d m an$  (1979) established seven SMV strains (SMV G1-G7) according to their virulence expressed in the reaction induced in inoculated specific differential soybean cultivars.

In the soybean breeding programme performed at the Faculty of Agricultural Sciences in Zagreb (Maksimir), attention has been paid to resistance of soybean to virus diseases. The objective of the present investigation conducted from 1986—1989 was primarily to detect the presence of SMV strains in breeding material as well as the presence of other viruses infecting soybean.

# Material and Methods

The material for investigation originated from breeder's plots and from the collection of different genotypes, domestic and introduced in Zagreb (Maksimir).

The trifoliolate leaves collected from plants showing symptoms served to prepare inoculum, by homogenizing in 0.01M sodium phosphate buffer pH 7.0, for mechanical transmission to primary leaves of soybean cultivar Rampage. The same inoculum was used for the local lesion assay on the detached leaves of Phaseolus vulgaris Top crop (Milbrath and Song 1946) to confirm the presence of SMV and also to inoculate the herbaceous plants for the isolation of other viruses infecting soybean. Only the isolates passed through two single lesions transfers in Top Crop were used to inoculate the soybean differentials: cvs. Rampage, York, Marshall, Ogden and Kwanggyo, and herbaceous plants for host range detection: Phaseolus vulgaris cvs. Top crop, Blue Lakes, Niagara, P. lunatus, Pisum sativum cvs. Bonnerville and Ranger, Chenopodium amaranticolor, C. quinoa, Nicotiana tabacum White Burley, N. glutinosa, and Vigna unguiculata Midget and Iron clay. Each confirmed SMV isolate was inoculated to 8-10 plants of each differential cultivar. The experiments were replicated four times and conducted in greenhouse at 25-30° C in clay pots with autoclayed soil.

The serological tests were carried out by Ouchterlony double diffusion tests in  $0.6^{\circ}/_{0}$  Ionagar No. 2 containing  $0.2^{\circ}/_{0}$  sodium dodecyl sulphate (SDS) and  $0.7^{\circ}/_{0}$  sodium chloride (Hunst and Tolin 1982) with crude sap extraxted from leaves and antiserum to typical strain of SMV kindly supplied by S. A. Tolin.

Particle size and morphology were determined by electron microscopy from leaf dip preparations negatively stained with uranyl acetate.

## Results and Discussion

From 50 samples of different genotypes displaying various symptoms 22 isolates were obtained. After the passage through the local lesions on detached leaves of Top crop, 17 isolates were identified to be SMV (cf. Table 1). The presence of SMV was established by positive reaction with antiserum to SMV in immunodiffusion gel. The isolates obtained did not differ markedly in host range assays. None of the isolates infected Nicotiana tabacum, Pisum sativum cvs., Vicia faba and Chenopodium quinoa. In Glycine max (L.) Merr. cvs. Rampage and Hodgson they

### STRAINS OF SOYBEAN MOSAIK VIRUS

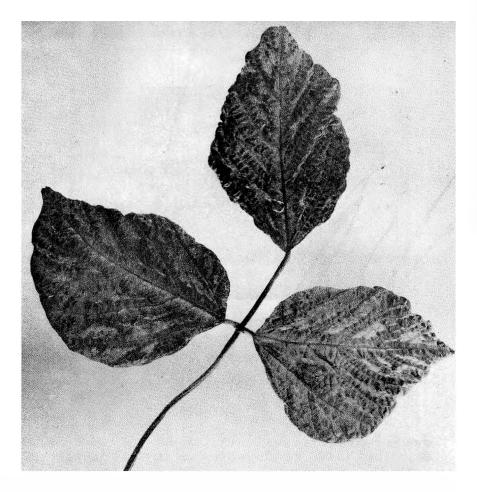


Fig. 1. Severe symptoms of soybean mosaic potyvirus (SMV) on trifoliate leaf of soybean PI 290—151 seedling infected through the seed.



Fig. 2. Flexuous filaments of SMV in leaf dip preparations.

		Soybean differential cultivars					
No.	SMV isolates from soybean cvs./lines	Rampage	York	Marshall	Ogden	Kwanggyo	SMV strains
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15	EXL 79-1 EXL 517 EXL-131-82 EXL-668 EXL-324A EXL-223 PI 822 PI 820 PI 786 PI 290-131 PI 159-764 ZVIJEZDA MAKSIMIRKA SREĆKA B 36/60	-/M -/M -/M -/M N/M -/M N/M -/M -/M -/M -/M -/M	N/N -/- N/N -/M -/M -/M -/- N/M -/- -/- -/- -/-	N/- N/N N/N -/N N/N -/- N/- -/- N/- -/- N/N LNL/- N/N	N/- -/- N/- N/N -/- -/- N/- -/- -/- LNL/- LNL/- -/-	-/ N/ N/N N/N N/ N/N -/ N/N -/ -/ -/ -/	$\begin{array}{c} -G 4^{*} \\ -G 2 \\ -G 2 \\ -G 4^{*} \\ -G 5 \\ -G 1 \\ -G 4^{*} \\ -G 5 \\ -G 1 \\ -G 1 \\ -G 2 \\ -G 1 \\ -G 2 \end{array}$
16 17	NS/NM ŽUPANJKA	-/M -/M	-/- -/- -/-	N/N -/-	-/- N/N -/-	-/- -/-	G3 G1

Table 1. Reactions of soybean (Glycine max) differential cultivars to inoculation with seventeen soybean mosaic virus (SMV) isolates

-/-: reaction on inoculated leaves/reaction on noninoculated trifoliolate leaves M : mosaic symptoms

N : necrosis

LNL: local necrotic lesions

- : symptomless, no virus detected by Top Crop bean indexing

G\* : tentative classification

induced local lesions and systemic mosaic. *Phaseolus vulgaris* Top crop was infected locally displaying small, distinct, brown lesions; *cvs.* Niagara and Blue Lake as well as *P. lunatus* were infected locally. *Vigna unguiculata* (L.) Walp. *cvs.* Midget and Iron clay showed mild systemic mottling.

Flexuous rods 750—760 nm long were observed in leaf dip preparations of the isolates investigated (cf. Fig. 2).

On the basis of symptoms provoked in soybean differential cultivars an attempt has been made to classify the SMV isolates obtained into seven SMV strains (G1—G7) established by Cho and Goodman (1979; cf. Chen *et al.* 1982). Strain G1 presents the least virulent group of isolates; G2, G3 and G4 represent isolates of intermediate virulence; G5, G6 and G7 are highly virulent. Our isolates Nos. 1, 4 and 8 were tentiatively classified in G4 strain group on the basis of local and systemic necrosis induced in cv. York. The classification of isolate No. 5 obtained from EXL 324A (USA blend) is tentative as it was not assayed on cv. Buffalo. However, the local and systemic necroses in three resistant cvs. (Marshall, Ogden and Kwanggyo) which produced only strain G7, support its ranging into G7 strain group.

The results summarized in Table 1 show that more than half of SMV isolates investigated belong to the least (G1) or less (G2) virulent SMV strains. The least virulent G1 strain is able to infect only very susceptible

#### ANA ŠARIĆ

cvs. like Rampage and Clark. Among the isolates obtained from domestic released cvs., those from Maksimirka, Srečka and Županjka belong to G1 strain group, from Zvijezda to G2 strain, and only those from the NS/NM from Novi Sad to G3 strain.

From three introduced cvs. EXL 324A (USA blend), EXL 223 (USA high protein content) and PI 786 (Belgium), highly virulent SMV isolates were obtained, one belonging to G7 and two to G5 strains. The G1 strain was also identified by Taraku *et al.* (1987) in soybean local cultivars from the Province of Kosovo.

According to the study of Tu (1989), the SMV strains G1, G2, G3 as well as G6 and G7 significantly delayed maturity in susceptible cultivars producing smaller plants with delayed maturity and the reduced seed yield.

The increased seriousness of the SMV problem in soybean nurseries is presented by the breeding material with high seed transmission rates and the low degree of resistance to SMV of soybean cultivars (Rossel and Thottappilly 1985).

The presence of isolates of very low virulence in domestic cultivars indicates that the domestic cultivars possess a very low resistance to SMV and that the breeding strategy must be adjusted to put more emphasis on resistance to SMV.

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## SAŽETAK

#### SOJEVI VIRUSA MOZAIKA SOJE U SELEKCIJSKOM MATERIJALU SOJE U HRVATSKOJ

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Iz različitih genotipova soje. uključenih u program selekcije na Fakultetu poljoprivrednih znanosti u Zagrebu, iz biljaka s izraženim simptomima virusne zaraze izdvojeno je sedamnaest virusnih izolata za koje je utvrđeno da prema krugu domaćina, morfologiji virusnih čestica i serološkim reakcijama pripadaju potyvirusu mozaika soje (SMV). Na osnovi reakcije specifičnih diferencijalnih kultivara soje, virusni izolati klasificirani su u sojeve različite virulencije po sistemu Cho i Goodman (1974). Većina istraženih izolata pripada sojevima G1 i G2 vrlo slabe virulentnosti što ukazuje na to da kultivari iz kojih su ti sojevi izolirani posjeduju vrlo slabu rezistentnost prema SMV. Preporučuje se da u programu selekcije soje više pozornosti treba pokloniti rezistentnosti sjemenskog materijala prema tom virusu.

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