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DISTRIBUTION OF SOYBEAN MOSAIC VIRUS WITHIN SOYBEAN EMBRYO

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It has been found by ELISA-test that soybean mosaic virus (SMV) occurs in equal concentrations in all parts of embryo of seed produced in infected soybean cultivars NS-9 and NS-16 which were grown in southeastern part of Yugoslavia (the Province of Kosovo). The correlation between percentage of seed coat mottling symptoms and SMV infection was significant in both cultivars.

Introduction

Soybean mosaic virus (SMV), a member of the potyvirus, occurs in soybean all over the world. It is spread in Yugoslavia as well, causing significant yield losses in soybean production. This virus belongs to seedborne viruses. About 30% or more of the seeds of diseased plants contain the virus, depending on cultivar and duration of infection before flowering (Bos 1972).

Viruses transmitted by seed can, generally, invade various parts of it. They can be found on the testa, in the endosperm and many of viruses are carried in the embryo of the seed and may survive in it for several years (Gold et al. 1954, Shepard 1972).

Virus concentrations in soybean seed are usually lower than in the leaves of infected plant. Because of that the detection of viruses in seeds is practicable by ELISA technique (Chen et al. 1982, Lister 1978)

or by some other sensitive techniques such as specific electron microscopy (Lange et al. 1983, Brlansky and Derrick 1979) and solid-phase radioimmunoassay (Bryant et al. 1982). Exceptionally, the virus in the seed can be detected by agargel double diffusion technique; this is possible only in the cases when the virus concentration is very high (Gibbs and Harrison 1976).

The results of the testing of soybean seed produced in the south-eastern region of Yugoslavia for distribution of SMV in embryo will be presented herein.

Material and Methods

The seed of infected soybean plants (*Glicine max* cv. NS-9 and cv. NS-16) with seed coat mottling was tested for virus content. The seed was produced by plants cultivated in the vicinity of Peć in the Province of Kosovo. Two parts of embryo were tested: cotyledons and left over part consisting of epicotyl, hypocotyl and radicle.

The embryo parts were separated in the following way: the seed was put in distilled water and after 24 hr seed coat was stripped off and after that cotyledons were separated from the remaining part of embryo (consisting of epicotyl, hypocotyl and radicle). Both parts of embryo were homogenized separately and filtered through filter paper. The filtrates obtained were then tested by ELISA. Antiserum to SMV (titer 1:64) was kindly suplied by Dr. Sue A. Tolin. Concentration of gamma-globulin used was at 1 or 2 mg/ml and alkaline phosphatase conjugated globulin was used in dilution 1:1000, 1:1500 and 1:2000. Incubation times in the ELISA tests were 1 and 2 hr at 30°C. The samples were prepared either by 30 cotyledons or 50 embryo structures consisting of epicotyl, hypocotyl and radicle. The control tests were carried out by virus-free seed, virus infected leaves and phosphate buffered saline.

Results

From 400 seeds obtained by infected soybean plants cv. NS-9, 112 seeds (28%) contained SMV. This was stated by ELISA-test. Similar results were obtained with cv. NS-16 as well. The tested seed was 6 months old and it was rather dried. Most of the infected seeds showed mottling on their surface (82% for cv. NS-9 and 78% for cv. NS-16). Consequently, in our case there was a correlation between the percentage of seed coat mottling and virus infection of seed.

Detection of SMV in cotyledons and the left over part of embryo by agargel diffusion technique was unsuccessful. By contrast, the virus was readily detected in all samples tested by ELISA. The relative concentrations of SMV in cotyledons and the left over embryo part of cv. NS-9 are presented in the Table 1. The results obtained with cv. NL-16 were not included in the Table because they were equal to the results obtained with cv. NS-9. The data presented show that there is no significant difference between the virus concentration in the cotyledons and the virus concentration in the left over part of embryo of infected soybean seed. However, the concentrations of the virus in whole embryos were significantly lower than the ones in the leaves.

Table 1. ELISA values for cotyledons and the left over part of soybean embryo (cultivar NS-9) of infected seed tested for soybean mosaic virus

	Absorbance values at 405 nm			
	Cotyledons		Left over part of embryo	
Readings after	1 hr	2 hr	1 hr	2 hr
Sample No		Infected seed		
1	0.226	0.362	0.163	0.298
2	0.301	0.524	0.187	0.487
3	0.212	0.456	0.224	0.459
4	0.233	0.472	0.281	0.537
5	0.182	0.300	0.192	0.431
6	0.143	0.782	0.211	0.382
7	0.312	0.730	0.350	0.422
8	0.289	0.384	0.398	0.689
9	0.291	0.526	0.157	0.789
10	0.200	0.446	0.172	0.345
11	0.135	0.431	0.200	0.466
12	0.227	0.479	0.176	0.369
13	0.199	0.614	0.158	0.368
14	0.187	0.521	0.142	0.399
15	0.254	0.489	0.174	0.489
Average:	0.226	0.501	0.212	0.463
		Virus free seed		
1	0.099	0.199	0.089	0.066
2	0.138	0.189	0.069	0.052
3	0.177	0.177	0.035	0.073
4	0.175	0.154	0.074	0.101
5	0.125	0.200	0.062	0.087
6	0.129	0.157	0.051	0.080
7	0.100	0.072	0.044	0.071
8	0.052	0.082	0.195	0.044
9	0.104	0.137	0.084	0.059
10	0.029	0.084	0.093	0.097
11	0.095	0.072	0.102	0.062
12	0.086	0.125	0.075	0.090
13	0.145	0.044	0.171	0.054
14	0.138	0.032	0.062	0.086
15	0.120	0.094	0.042	0.084
Average:	0.114	0.121	0.083	0.074
	SMV-infected leaf		Phosphate buffered saline	
1	0.299	0.562	0.036	0.080
2	0.325	0.673	0.102	0.125
3	0.392	0.570	0.044	0.099
4	0.267	0.425	0.069	0.161
5	0.330	0.552	0.045	0.102
6	0.249	0.429	0.086	0.104
Average:	0.310	0.535	0.064	0.112

Discussion

In our case about 30% of seeds produced in infected plants contained the virus. These data are in concordance with those noted in the literature (Bos 1972). Our observations have shown that the association of seed coat mottling with seed infection is rather consistent. Based on that it seems that seed mottling could be in our case a good indicator of the presence of SMV in seed. However, since this association depends on environmental conditions and maturity (Bryant et al. 1982) this correlation should be studied during several seasons.

The concentrations of SMV in cotyledons and in the left over part of embryo of two soybean cultivars were similar to the concentrations stated earlier in the embryo of the one not determined soybean cultivar which was also grown in the Province of Kosovo (Taraku et al. 1987).

Seed transmission of SMV in this region is probably the most important means for the transmission of the virus from one season to the next and as a primary source of virus inoculum for vector (Brlansky and Derrick 1978). Detection of pathogenic strains of SMV in soybean seed in this region is very important, since most soybean cultivars now grown in the Province of Kosovo are susceptible to SMV. The best control measure is to have available SMV-free seed (comp. Phatak 1974, Chen et al. 1982, Bryant et al. 1982).

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SAŽETAK

RASPROSTRANJENOST VIRUSA MOZAIKA SOJE UNUTAR EMBRIJA SOJE

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Utvrđeno je s pomoću metode ELISA da virus mozaika soje (SMV) dolazi unutar embrija soje u podjednakim koncentracijama u kotiledonima i u onom dijelu koji čine epikotil, hipokotil i radikula. To je ustanovljeno na sjemenu soje (sorte NS-9 i NS-16) koja se uzgaja na području Kosova. Istraživanje je pokazalo da postoji značajna korelacija između pjegavosti koja se opaža na testi sjemenaka soje i inficiranosti sjemenaka SMV-om.

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