

PROPERTIES OF TWO VIRUS ISOLATES FROM CLOVERS IN CROATIA

NYSRET TARAKU, NIKOLA JURETIĆ and DAVOR MILIČIĆ

(Institute of Botany, Faculty of Science, University of Zagreb)

Received February 1, 1977

Introduction

Virus diseases of clovers have been the subject of intensive investigations for 40 years. Numerous viruses belonging to various virus groups have been isolated from clovers so far. Among them cucumber mosaic virus (CMV) and been yellow mosaic virus (BYMV) have been isolated, not only abroad but also in Yugoslavia (Grbelja 1974; Malak 1974; Šutić and Babović 1966).

In 1973 two virus isolates were found on clovers in the vicinity of Zagreb. Preliminary investigations indicated that the isolate from white clover (*Trifolium repens* L.) could belong to CMV, and the other, from red clover (*Trifolium pratense* L.), to BYMV. Identification of the two isolates on the basis of differential hosts, physical properties, serological relationship, and light and electron microscopic inclusion bodies is presented here.

Material and Methods

Samples of white and red clover, from which the investigated viruses were isolated, grew spontaneously north of Zagreb. The isolate found on *T. repens* was marked CMV-C, and the isolate from *T. pratense* BYMV-C.

The isolates were cleaned before further investigations by means of »one lesion method«. Specimens of *C. quinoa* for CMV-C and specimens of *C. amaranticolor* for BYMV-C were used for that purpose. Physical properties were determined in the standard manner.

Antiserum against cucumber mosaic virus (CMV) was employed in serological experiments with CMV-C. The antiserum was obtained through the courtesy of Dr. E. Luisoni (Torino). Serological tests were

carried out in 0.9% agarose gel prepared in distilled water without the addition of any preservative. BYMV-C was serologically investigated by an antiserum to BYMV that was kindly supplied by Dr. D. Z. Maat (Wageningen). Serological tests with BYMV-C were performed by "drop method" on microscope slides. The infectious plant sap was clarified by heating at the temperature of 30° C for 10 min and then centrifuged at low speed.

CMV-C was examined by means of a plant-protection test using the "white" strain of CMV (CMV-W) which was sent us by courtesy of Dr. J. Horváth (Budapest).

Aphid transmission of CMV-C was investigated by *Myzus persicae*. Before acquisition feeding (30 min) the aphids were starved 120 min. The infection feeding lasted 120 min. Seventeen aphids were employed per a plant. To find out whether CMV-C was transmissible by soil, the healthy plants were planted into soil in which previously the plants infected with CMV-C had been grown.

The inclusion bodies of the isolate BYMV-C were observed by light microscope and electron microscope. For electron microscope investigations ultrathin sections of *Nicotiana clevelandii* tissue were used. Strips of leaf tissue from infected plants were fixed for 30 min in 1% (v/v) glutaraldehyde (in cacodylate buffer pH 7.2) and postfixed for 2 hr in 1% (w/v) osmium tetroxide. After fixation, samples of tissue were dehydrated in ethanol series and embedded in Araldite. Ultrathin sections were examined in a Siemens Elmiskop I.

Results

A. Investigation of CMV-C isolate

1. Symptoms on test plants

Since the preliminary investigations showed that CMV-C isolate probably belonged to CMV, the test plants which reacted characteristically to CMV and which served as differential hosts for distinguishing CMV from some other viruses were primarily used. They were *C. amaranticolor*, *C. quinoa* and *C. murale* which are known to react to CMV exclusively with local symptoms, and *N. glutinosa* and *Datura stramonium* which react with systematic symptoms. CMV-C was transmitted on 13 test plants in all (Tab. 1). Symptoms on plants in Table 1 indicated that isolate CMV-C could belong to CMV. It may be concluded primarily on the basis of the reactions on *C. quinoa*, *Cucumis sativus*, *N. glutinosa* and *Datura stramonium* (cf. Uschdraweit 1955; Gibbs and Harrison 1970).

2. Physical properties

For CMV-C, thermal inactivation point (TIP) at 70—72° C, and dilution end point (DEP) at 1 : 20 000 were established. Longevity in vitro (LIV) was about 6 and 3 days at 20° C and 25° C, respectively. Samples of *C. amaranticolor*, *C. quinoa* and *N. megalosiphon* were employed for determination of TIP. Physical properties of CMV-C corresponded to those of CMV (cf. Simons 1957; Hein 1957; Lovisolò and Benetti

Table 1. Reactions of plants infected with the CMV-C isolate*

Hosts	Symptoms
<i>CHENOPODIACEAE</i>	
<i>Chenopodium amaranticolor</i> Coste et Reyn.	L chlorotic lesions
<i>C. quinoa</i> Willd.	L chlorotic lesions
<i>C. murale</i> L.	L chlorotic and necrotic lesions
<i>CUCURBITACEAE</i>	
<i>Cucumis sativus</i> L.	L chlorotic spots; S vein clearing, mottling, stunting (Fig. 1a)
<i>LEGUMINOSAE</i>	
<i>Phaseolus vulgaris</i> L.	L chlorotic spots, necrosis; S vein clearing, mosaic, blistering, shoestring symptom (Fig. 1b, c)
<i>Pisum sativum</i> L.	S mottling, leaf deformation, stunting
<i>Trifolium incarnatum</i> L.	S vein clearing
<i>SOLANACEAE</i>	
<i>Datura stramonium</i> L.	L chlorotic spots; S mottling
<i>Nicotiana clevelandii</i> Gray	L chlorotic spots; S vein clearing, mosaic, curving of leaf, shoe-string symptom
<i>N. glutinosa</i> L.	S chlorotic spots
<i>N. megalosiphon</i> Heurck et Muell.	L chlorotic and necrotic spots, variegation; S chlorosis, necrosis, shoe-string symptom (Fig. 1d)
<i>N. tabacum</i> L., cv. Samsun	L chlorotic spots; S vein clearing, mild variegation
<i>N. tabacum</i> L., cv. White Burley	L chlorotic spots; S mottling, mild variegation

*L local symptoms; S systemic symptoms

1961). It must be stressed that the DEP of our isolate corresponds to the same property of the CMV isolate found on some leguminous plants earlier in the Eastern part of Yugoslavia (B a b o v i ć 1968; M a l a k 1974).

3. Plant-protection test

To identify our isolate, the plant-protection test was performed. The investigated isolate CMV-C was compared by means of this test with a known strain of CMV, i. e. with the "white" strain CMV-W. Since the symptoms on *N. megalosiphon* caused by CMV-W were different from the symptoms caused by CMV-C on the same plant, it was possible to carry out the plant-protection test. CMV-C produced a "green" mosaic on *N. megalosiphon* whereas CMV-W provoked a "white mosaic".

The plant-protection test was performed in the following way. First 10 specimens of the plant were inoculated with CMV-C (set No 1) and 5 samples with CMV-W (set No 2). Symptoms appeared on both sets of plants about twenty days later. Samples of the set No 1 which showed symptoms of CMV-C were superinoculated with CMV-W (set No 3). At the same time 5 healthy samples were inoculated with CMV-W as a control (set No 4). Finally, results of the test were recorded 30 days after the latter inoculation. The results were: plants of the set No 1 showed "green" symptoms, plants of the set No 2 "white" symptoms, plants of the set 3 "green" symptoms and plants of the set No 4 also "white" symptoms. The following conclusion may be postulated: the plants of the set No 3 which had symptoms characteristic of CMV-C ("green" symptoms) did not show the "white" symptoms which are a characteristic feature of CMV-W after superinfection with CMV-W. Consequently, this means that CMV-W could not infect the plants because they were protected by CMV-C. This proves that CMV-C really belongs to CMV.

4. Serological investigations

Our isolate reacted positively with the antiserum supplied by Dr. E. Luisoni (Fig. 1e, f). The antiserum used to CMV was previously absorbed with the sap of a healthy plant. The serum absorption was suggested to us by Dr. E. Luisoni; it was performed according to the method of serum absorption in agar gel by van Regenmortel (1967).

On the basis of serological tests it has been concluded that CMV-C indeed belongs to CMV, i. e. the tests confirmed the results of the plant-protection test.

5. Aphid and soil transmission

It was established that CMV-C could be transmitted from infected *N. megalosiphon* to healthy plants by *Myzus persicae*. Incubation was about 30 days, thus it was much longer than in the case when the virus was mechanically inoculated. The successful transmission of CMV-C by aphids showed that our virus did not differ in this property from CMV.

When young healthy plants of *N. megalosiphon* were planted into soil in which plants infected with CMV-C had been grown, it was found that all 15 plants remained healthy. This was checked by back inoculation tests. Therefore, CMV-C is not soil transmissible.

B. Investigation of BYMV-C isolate

1. Symptoms on host plants

Isolate BYMV-C which was found on *Trifolium repens* was transmitted into 6 test plants belonging to three plant families. The symptoms observed on them are presented in Table 2.

According to symptoms on test plants it was obvious that BYMV-C did not represent either CMV or alfalfa mosaic virus which often attack clovers (Hanson and Hagedorn 1952; Grbelja 1974). Although typical BYMV, unlike our isolate, causes both local and systemic infection on *C. amaranticolor*, it is not improbable that BYMV-C belongs to BYMV.

Table 2. Reactions of plants infected with the isolate BYMV-C*

Plants	Symptoms
<i>CHENOPODIACEAE</i>	
<i>Chenopodium amaranticolor</i> Coste et Reyn.	L chlorotic and necrotic lesions (Fig. 2b)
<i>C. quinoa</i> Willd.	L chlorotic and necrotic lesions
<i>C. murale</i> L.	L chlorotic lesions
<i>LEGUMINOSAE</i>	
<i>Pisum sativum</i> L.	L chlorosis, necrosis; S mosaic
<i>Vicia faba</i> L.	S blistering, mild mottling (Fig. 2a)
<i>SOLANACEAE</i>	
<i>Nicotiana clevelandii</i> Gray	L chlorotic spots; S vein clearing, chlorosis, blistering, stunting

*L local symptoms; S systemic symptoms

2. Physical properties

BYMV-C had TIP 60°—65° C, DEP about 10⁻⁴ and LIV 3—4 days. TIP and DEP were determined by means of *C. amaranticolor* and LIV was investigated with the exception of the two plants mentioned above, also with *N. megalosiphon*. Physical properties of BYMV-C correspond to the data found for BYMV (Bos 1970; Šutić and Babović 1966; Kovachevsky 1973).

3. Serological data

Our isolate reacted positively with antiserum against BYMV supplied by Dr. D. Z. Maat. The experiments were carried out by "drop test" on microscope slides. Homologous titre of the serum was 1:512 and our isolate reacted with it to the dilution 1:32. The positive serological reaction and the heterologous titre suggest that our isolate is only remotely related to BYMV.

4. Inclusions bodies

Since our isolate BYMV-C produces inclusions bodies, it was possible to use that property in its identification. The inclusions were studied by means of light and electron microscopes. One month after inoculation amorphous and granular bodies in the form of x-bodies were found in leaf hair cells of *N. megalosiphon* and in epidermal cells of *V. faba*. The bodies, usually oval, were often in contact with the nucleus and their size was usually smaller than that of the nucleus (Fig. 2c, d).

The examined ultrathin sections of infected tissue revealed the presence of pin-wheel structures and laminated aggregates. Since our isolate produces pin-wheel structures with laminated aggregates, it belongs to the second subgroup of potyviruses according to Edw ar d s o n et al. (1974) who classified potyviruses into three subgroups on the basis of the type of pin-wheel structures.

Figs. 3 and 4 show pin-wheel structures with laminated aggregates. The aggregates are often very long and similar to bands. Laminated aggregates sometimes form a cluster in which they are oriented parallelly (Fig. 4). This seems to be the case when the laminated aggregates of pin-wheel structures are longitudinally sectioned. Besides pin-wheel structures, small aggregates of elongated virus particles can be seen (Fig. 4). In some of the ultrathin sections, parts of chloroplasts and tubular structures which are similar to endoplasmic reticulum can be observed. The parts of cytoplasm containing the above structures are fairly compact and differ significantly from normal cytoplasm parts (Fig. 5). Such a region of the cytoplasm may also contain large lipid globules (Fig. 4) and is likely to belong to the x-bodies. As can be seen in Figs. 3, 4, 5, mitochondria and sometimes peroxisome crystals always enveloped by a special membrane are common in this region.

Discussion

Isolate CMV-C produces a shoe-string leaf deformation of *N. cleve-landii*, *N. megalosiphon* and *Phaseolus vulgaris*. The shoe-string symptom is provoked by many CMV strains (Mogendorff 1930; Kovachevsky 1965; Juretić 1968). It is interesting that the shoe-string symptom was caused also by CMV isolated from *Leguminosae* (Babović 1968; Malak 1974). This symptom usually appeared in our tests when the temperature in the greenhouse was above 25° C. These observations correspond to the statement by Mogendorff (1930) who found that the appearance of the shoe-string symptom depended on external conditions.

The symptoms provoked by CMV-C on *N. megalosiphon* are different from the symptoms produced on this plant by CMV-W. It must be stressed that these differences were much more pronounced when inoculated plants were maintained at 22° C to 25° C. When the temperature was above 28° C the differences mentioned above were less significant. Therefore we performed the plant-protection test during autumn period when the temperature in the greenhouse was about 24° C.

Our finding that CMV-C is not transmissible by soil corresponds to the results of Malak (1974) who also investigated a CMV isolate from leguminous plants for the same purpose.

It must be mentioned that CMV was also found, except in clovers, in some other leguminous plants. This virus was, for instance, isolated from alfalfa (Doolittle and Zaumeyer 1953) and from bean (Bos and Mat 1974).

As regards BYMV-C isolate, our results indicate that it is possible for this isolate to belong to BYMV. Some data show that BYMV-C may be a distinct strain of BYMV. BYMV-C provoked only local symptoms on *C. amaranticolor*, and not both local and systemic symptoms as the typical BYMV would. Moreover, our isolate reacted with antiserum against the typical BYMV (homologous titre 1/512) only to a dilution 1 : 32 and this fact also supports the opinion that BYMV-C is serologically related to the typical BYMV. Edwardson et al. (1974) investigated serologically the properties of potyviruses and found that about 17 potyviruses of a total of 69 investigated, were serologically related to BYMV.

Fig. 1. A—D symptoms caused by CMV-C on test plants, E, F serological reactions in agar gel diffusion tests; A *Cucumis sativus*; B, C *Phaseolus vulgaris*; D *Nicotiana megalosiphon*; E, F reactions of CMV-C (D) in agar gel diffusion test with CMV antiserum (sL) which was absorbed with healthy plant sap; Well Z contained healthy plant sap.

Sl. 1. A—D simptomi na pokusnim biljkama koje izaziva CMV-C, E, F serološke reakcije u agarском gelu; A *Cucumis sativus*, B C *Phaseolus vulgaris*; D *Nicotiana megalosiphon*; E, F reakcije CMV-C izolata (D) u agarском gelu s imunim serumom protiv virusa mozaika krastavca (sL) koji je bio apsorbiran sa sokom zdrave biljke; bazen Z sadržavao je sok zdrave biljke.

Fig. 2. Symptoms and inclusion bodies provoked by BYMV-C: systemic spotting on *Vicia faba* (A) and local lesions on *Chenopodium amaranticolor* (B); C, D x-bodies in hair cells of *Nicotiana clevelandii* (X — x-body, n — nucleus).

Sl. 2. Simptomi i stanične inkluzije izazvani izolatom BYMV-C: Sistemična pjegavost na vrsti *Vicia faba* (A) i lokalne lezije na vrsti *Chenopodium amaranticolor* (B); C, D x-tijela u dlačnim stanicama vrste *Nicotiana clevelandii* (X — x-tijelo, n — jezgra).

Fig. 3—5. Submicroscopical view of *Nicotiana clevelandii* leaf cells infected with BYMV-C: pw pin-wheel structures, m mitochondrion, er endoplasmic reticulum, lg lipid globule, la laminated aggregate, ct tubular structure, cl chloroplast, p peroxisome crystal, v aggregate of virus particles. Magnification: 39000× (Fig. 3), 40000× (Fig. 4), 42000× (Fig. 5).

Sl. 3—5. Submikroskopske promjene u listu vrste *Nicotiana clevelandii* inficiranom izolatom BYMV-C: pw strukture pin-wheel, m mitohondrij, er endoplazmatski retikulum, lg lipoidna globula, la laminarne strukture, ct cjevaste strukture, cl kloroplast, p peroksisomalni kristal, v nakupina virusnih čestica. Povećanje: 39000× (sl. 3), 40000× (sl. 4), 42000× (sl. 5).

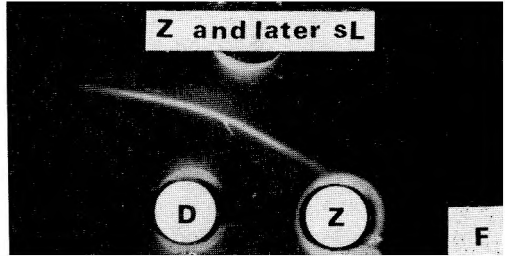
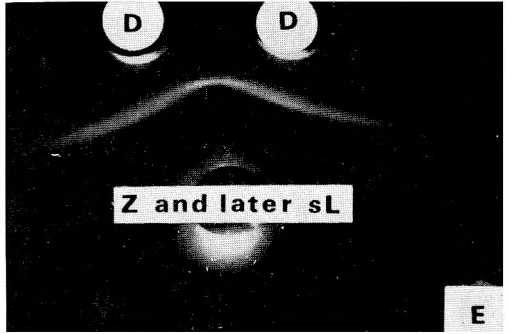
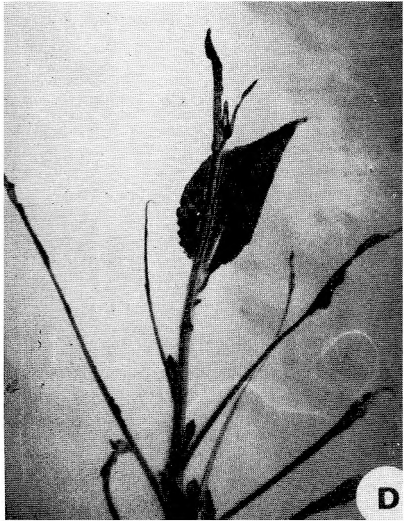
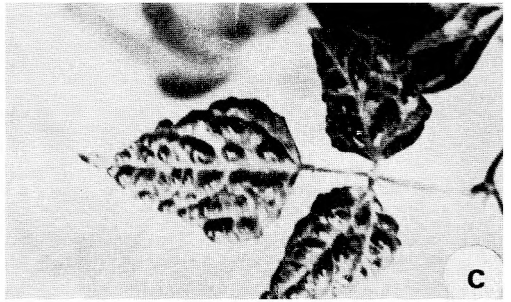
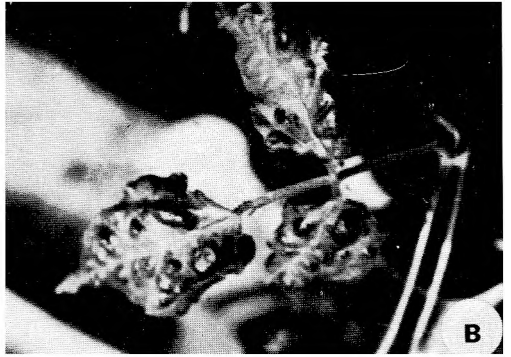
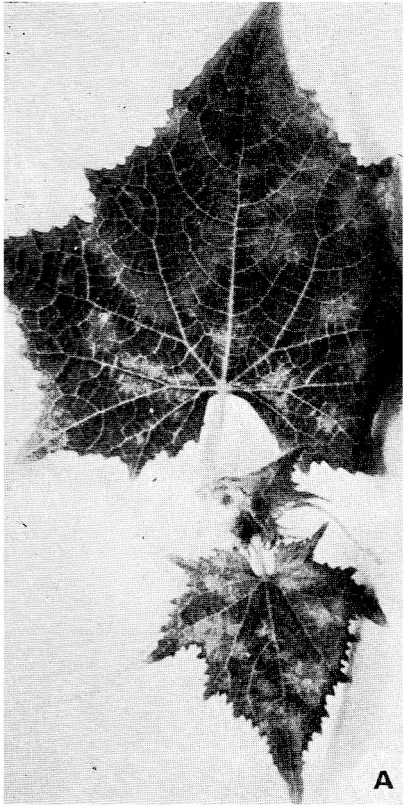


Fig. 1. — SL. 1.

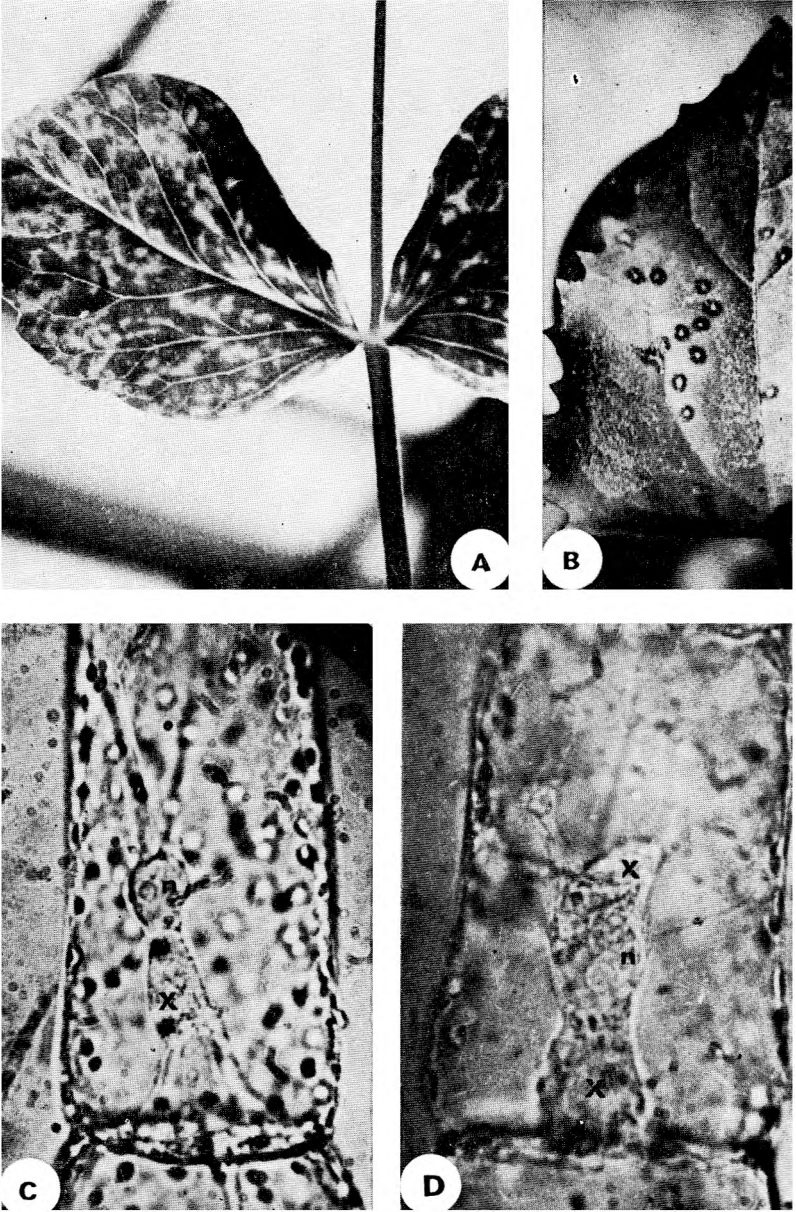


Fig. 2. — Sl. 2.

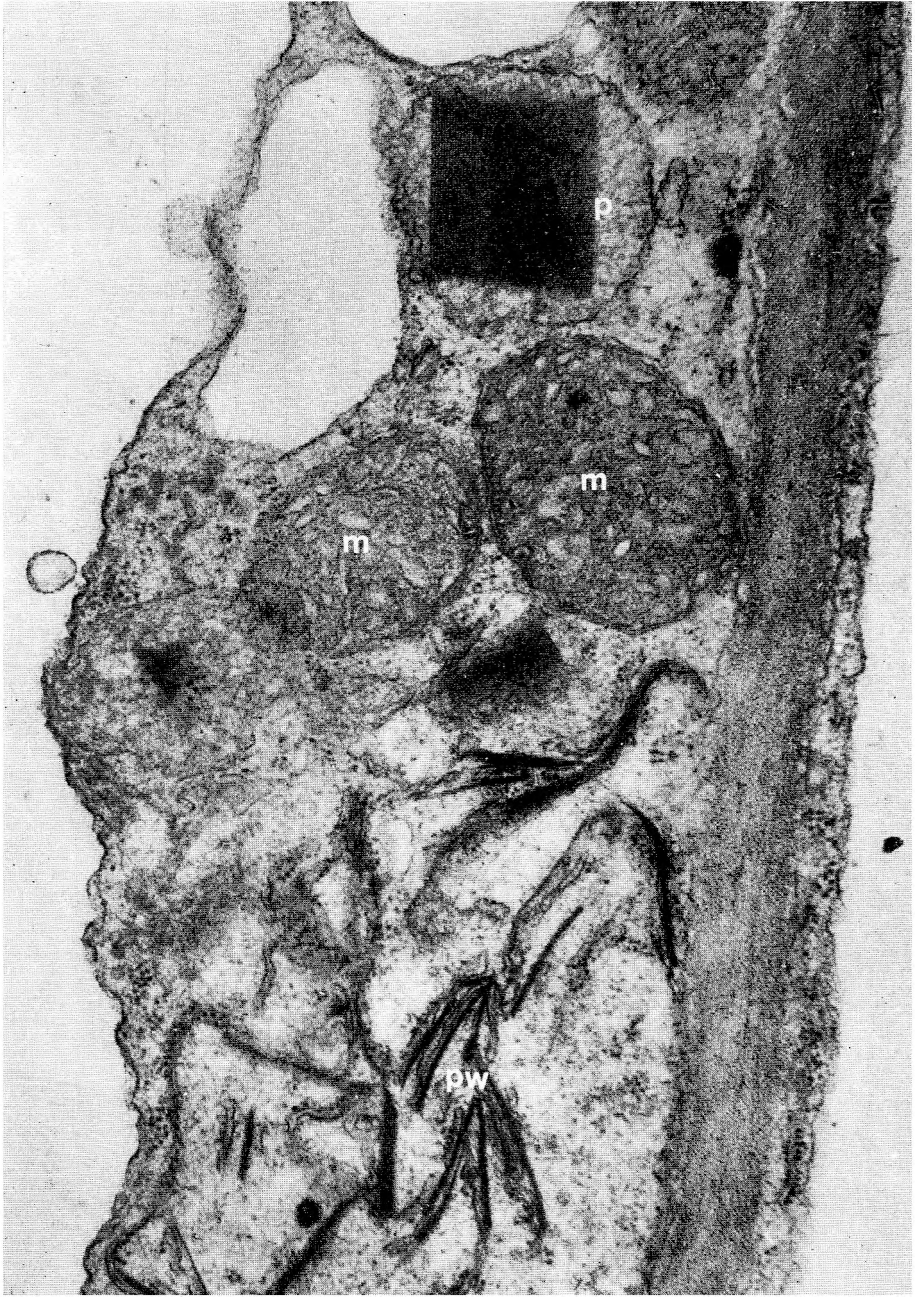


Fig. 3. — Sl. 3.



Fig. 4. — Sl. 4.



Fig. 5. — Sl. 5.

Therefore, our isolate BYMV-C may well belong to some other potyvirus, but not to BYMV. There is one more difference between our isolate and the typical BYMV. Unlike BYMV our isolate has never produced crystals in the x-bodies or in the nucleus of infected cells.

Summary

Two viruses have been isolated from *Trifolium repens* and *Trifolium pratense*. The isolate found in *T. repens* (CMV-C) was identified as an isolate of cucumber mosaic virus (CMV). It was readily sap transmissible by sap and also by *Myzus persicae* Sulz., but was not soil-borne. Symptoms of test plants were similar to or identical with the symptoms common to CMV. CMV-C provoked leaf deformations and shoe-string symptoms of leaves on *N. clevelandii*, and *Phaseolus vulgaris*. Physical properties were well in concordance with those found for CMV. In serological investigations, performed by means of agar gel double diffusion test and absorption test in agar, CMV-C reacted positively with CMV antiserum. Plant-protection test have confirmed that CMV-C belongs to CMV.

The investigations of the isolate from *T. pratense* (BYMV-C) showed that it probably belonged to bean yellow mosaic virus (BYMV). The physical properties of BYMV-C correspond to the properties of BYMV isolates. The statement that BYMV-C represented an isolate of BYMV was supported by investigations of cell inclusions. Granular cytoplasmic inclusions in the form of x-bodies occurred in the infected cells, but they were smaller than in the typical BYMV. The examined ultrathin sections of infected tissue revealed small aggregates of elongated virus particles, pin-wheel structures, and laminated aggregates. Another indication in favour of our conclusion that BYMV-C belongs to BYMV, is the positive serological reaction and moderately high heterologous titre (1:32) of BYMV antiserum (homologous titre 1:512) when tested with BYMV-C. It seems that BYMV-C represents a distinct strain of BYMV.

*

Acknowledgment. We are grateful to Dr. Mercedes Wrischer for electron microscope analysis of inclusion bodies. The technical assistance of Mrs. Slava Petrović is gratefully acknowledged.

References

- Babović, M., 1968: Viroze lucerke u Jugoslaviji. *Zaštita bilja* 102, 335—410.
- Bos, L., 1970: Bean yellow mosaic virus. C. M. I. / A. A. B. Descriptions of plant viruses 40.
- Doolittle, S. D., W. J. Zaumeyer, 1953: A pepper ringspot caused by strains of cucumber mosaic virus from pepper and alfalfa. *Phytopathology* 43, 333—337.
- Edwardson, J. R., 1974: Some properties of the potato virus Y-group. Florida Agricultural Experiment Station Monographs 4. Gainesville.
- Gibbs, A. J., B. D. Harrison, 1970: Cucumber mosaic virus. C. M. I. / A. A. B. Descriptions of plant viruses 1.
- Grbelja, J., 1974: Istraživanje virusnih izolata iz djeteline. IVth Congr. Yugoslav Biol., Sarajevo, p. 133.
- Hanson, E. W., D. J. Hagedorn, 1952: Red clover reservoir of legume viruses in Wisconsin. *Phytopathology* 42, 467.

- Hein, A., 1957: Beiträge zur Kenntnis der Viruskrankheiten an Unkräutern. *Phytopath. Z.* 29, 204—229.
- Juretić, N., 1968: Deformacije na listu i cvijetu kužnjaka (*Datura stramonium* L.) inficiranog virusom mozaika krastavca. *Acta Bot. Croat.* 26/27, 117—144.
- Kovachevsky, I., 1965: Krastavično mozaičnata viroza v Blgaria. Izdatelstvo na Blgarskata akademija na naukite, Sofija.
- Kovachevsky, I., 1973: Proučvanija vrhu pričinitelja na žltata mozaika po fasula. *Rastenievedni nauki* 6, 129—135.
- Lovisolò, O., M. P. Benetti, 1961: Su di ceppo del virus del mosaico del cetriolo di tipo alloiofillia isolato da pomodoro. *Boll. Staz. Pat. Veget.* 19, 35—50.
- Mogendorff, N., 1930: Fern-leaf of tomato. *Phytopathology* 20, 25—46.
- Malak, J., 1974: Crvena detelina kao domaćin virusa mozaika krastavca. *Zaštita bilja* 25, 219—226.
- Regenmortel, M. H. V. van, 1967: Serological studies on naturally occurring strains and chemically induced mutants of tobacco mosaic virus. *Virology* 31, 467—480.
- Simons, J. N., 1957: Three strains of cucumber mosaic virus affecting bell pepper in the Everglades Area of South Florida. *Phytopathology* 47, 145—150.
- Sutić, D., M. Babović, 1966: Red clover, a host plant of bean yellow mosaic virus. *Rev. Roum. Biol. Botanique* 11, 225—229.
- Ushdraweit, H. A., 1955: *Chenopodium quinoa* als Testpflanze für das Gurkenmosaik. *Nach. — Bl. dtsch. Pflanzenschutzdienst (Braunschweig)* 7, 151—152.

SADRŽAJ

OSOBI NE DVAJU VIRUSNIH IZOLATA NAĐENIH NA DJETELINI U HRVATSKOJ

Nysret Taraku, Nikola Juretić i Davor Miličić

(Botanički zavod Prirodoslovno-matematičkog fakulteta u Zagrebu)

Istraživana su dva virusna izolata nađena u jesen 1973. godine na primjercima djeteline (*T. repens* i *T. pratense*) u okolici Zagreba. Izolat nađen na vrsti *T. repens* označili smo kraticom CMV-C, a izolat nađen na vrsti *T. pratense* oznakom BYMV-C.

Izolati CMV-C identificirali smo kao virus mozaika krastavca (CMV). To smo ustanovili na osnovi sljedećih podataka. Izolat se lako prenosi sokom zaražene biljke, te lisnim ušima vrste *Myzus persicae* Sulz. Međutim, taj se izolati ne prenosi zemljom. Simptomi koje uzrokuje CMV-C na pokusnim biljkama koje karakteristično reagiraju na CMV bili su slični ili identični sa simptomima koje uzrokuju drugi poznati sojevi CMV. Na vrstama *N. megalosiphon*, *N. clevelandii* i *Phaseolus vulgaris* istraživani CMV-C izolati uzrokuju nitavost lista. Termalna točka inaktivacije, krajnja točka razređenja i postojanost in vitro izolata CMV-C kreću se u granicama svojstava koja su utvrđena za CMV. Naš izolati CMV-C je pozitivno serološki reagirao s antiserumom protiv CMV u pokusima izvedenim u gelu agara (sl. 1e. f). Premunitetni pokus s izolatom CMV-C, u kojem je naš istraživani izolati bio uspoređen s dobro karakteriziranim »bijelim« sojem CMV, bili su pozitivni.

Drugi istraživani izolati (BYMV-C) koji je nađen na vrsti *T. pratense* čini se da predstavlja soj virusa žutog mozaika graha (BYMV). To je zaključeno na osnovi sljedećih rezultata. Na vrsti *Pisum sativum* on

izaziva simptom u obliku žutog mozaika. Fizička svojstva BYMV-C (termalna točka inaktivacije 60° C, krajnja točka razređenja 10⁻⁴, postojanost in vitro 3 do 4 dana) odgovaraju fizičkim osobinama izolata BYMV. Naše mišljenje da BYMV-C pripada BYMV potkrijepljeno je istraživanjima staničnih inkluzija. U inficiranim stanicama istraživani izolat stvara zrnaste citoplazmatske inkluzije u obliku x-tijela (sl. 2c; d). Treba istaći da su te inkluzije bile sitnije od inkluzija koje uzrokuje tipični BYMV. Istraživanja ultratankih presjeka inficiranog tkiva pokazala su da BYMV-C stvara sitne agregate izgrađene od produženih virusnih čestica, te strukture pin-wheel i laminarne agregate (sl. 3—5). Činjenica koja također govori u prilog našem zaključku da BYMV-C pripada BYMV je pozitivna serološka reakcija s antiserumom protiv BYMV. Međutim treba istaći da je naš izolat reagirao s tim antiserumom (homologi titar 1 : 512) samo do razređenja 1 : 32. To pokazuje da se naš izolat serološki dosta razlikuje od tipičnog BYMV.

Da se naš izolat dosta razlikuje od tipičnog BYMV, pokazuje i reakcija vrste *C. amaranticolor*. Naime, naš izolat izaziva na vrsti *C. amaranticolor* samo primarne simptome, dok tipični BYMV izaziva i primarne i sekundarne simptome. Osim toga BYMV-C se od tipičnog BYMV razlikuje i po tome što ne stvara kristalične inkluzije, i to niti u x-tijelima niti u jezgri. Na osnovi ovih razlika zaključili smo da naš izolat BYMV-C predstavlja naročit izolat BYMV-a.

Nysret Taraku, mr biol.
doc. dr Nikola Juretić
prof. dr Davor Miličić
Botanički zavod
Prirodoslovno-matematičkog fakulteta
Marulićev trg 20/II
Yu 41000 Zagreb (Jugoslavija)