Virus diseases on some vegetable fruits from *Solanaceae* family

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Abstract

Fruits of different varieties and types of tomato and pepper are very often with bad market appearance, caused by virus diseases. Viral pathogens put and cover entire tomato, pepper and eggplant plants and damage ripening fruits which become not good for markets, because they are mottled by mild or severe yellow spots, flecks, mosaic, line patterns and manifest deformations and often decreased sizes. The symptoms of virus diseases on the fruits of tomato, pepper and eggplant were observed in 2018 and six viral pathogens: *Tomato spotted wilt virus* (TSWV), *Cucumber mosaic virus* (CMV), *Tomato mosaic virus* (ToMV), *Alfalfa mosaic virus* (AMV) *Potato virus Y* (PVY) and *Potato virus X* (PVX) were analyzed by ELISA method (DAS-ELISA).The indicator method, basing on the reaction of indicator (test) plants was used for three, more wide spread viruses: TSWV, ToMV and CMV. The results have shown: TSWV was established in 30 fruits, CMV – in 19 fruits, ToMV – in 17 fruits, PVY- in 12 fruits, PVX - in 6 fruits and AMV – in 3 fruits, from all 49 tomato, pepper and eggplant fruits, bought from different markets and belonging to different production areas. Severe injuries with viral origin were proven on pink tomatoes, where TSWV and ToMV in mixed infection or only TSWV alone were the basic agents, responsible for the damages. Both viruses: the thrips-transmitted TSWV and the mechanically-transmitted ToMV were in very high viral concentration in pink tomato fruits, whereas CMV, PVY, PVX and AMV were sporadically spread and in more low viral concentration. The major virus, responsible for the injuries on pepper fruits (kapia and kamba types) was the thrips-transmitted TSWV, alone or in mixed infection most often with aphid-transmitted viruses - CMV and PVY. Healthy fruits of tomato, pepper and eggplant (without symptoms of virus diseases) could be produced when the effective control against TSWV, ToMV, CMV and PVY carry out.

Keywords: tomato; pepper; eggplant; viruses; TSWV; ToMV; CMV; PVY

INTRODUCTION

Virus diseases decreased quantitatively the yield and deteriorated quality of the agricultural production. This point of view is in force especially for vegetables, because their fruits are used fresh, in dishes or preserved. The main viral pathogen, causing injuries on pepper fruits in Bulgaria is *Tomato spotted wilt virus* (TSWV) (Dikova, 2018). Injuries deteriorating the market appearance of tomato and pepper production caused by TSWV were established by different researchers (Kostova et al., 2003; Sherwood et al., 2003; Momol et al., 2005; Koike et al., 2017).

One important viral pathogen for vegetables, belonging to *Solanaceae* family is *Tomato mosaic virus* (ToMV). It is established in different areas in the world: USA, Bulgaria, Corea, India, Vietnam (Cerkauskas, 2004b; Dikova, 2014; Petrov, 2014; Chitra et al., 1999; Miah Bae et al., 2018).

Cucumber mosaic virus (CMV) is important viral pathogen for tomato and pepper (Hristova & Maneva, 1999; Yardimci & Eryigit, 2006).

Alfalfa mosaic virus (AMV) is also a pathogen for naturally infected tomato crops (Cerkauskas, 2004a; Zitikaite & Samuitiene, 2008).

Potato virus Y (PVY) is important aphid-transmitted virus. It is most wide spread in some coun-

tries on pepper and tomato plants in some years (Cerkauskas, 2005a; Petrov, 2014; Olawale et al., 2015; Ozdag & Sertkaya 2017).

Potato virus X (PVX) is connected with aphid-transmitted potato viruses, as PVY, frequently in mixed infection (Cerkauskas, 2005b; Olawale et al., 2015).

The objective of the research was the establishment of some of more wide spread viruses on vegetable fruits, belonging to *Solanaceae* family, as tomato, pepper and eggplant.

MATERIAL AND METHODS

Samples from pepper, tomato and eggplant fruits were analyzed by serological ELISA method – DAS-ELISA with kits for the following viruses: *Tomato spotted wilt virus* (TSWV), *Cucumber mosaic virus* (CMV), *Tomato mosaic virus* (ToMV), *Alfalfa mosaic virus* (AMV) *Potato virus Y* (PVY) and *Potato virus X* (PVX). Negative controls were uninfected test plants and positive controls were inoculated by TSWV, ToMV and CMV indicator (test) plants. Negative and positive controls from the kits were used for AMV, PVY and PVX. DAS-ELISA was used by Clark and Adams (1977) with antisera, purchased from the German company LOEWE, Biochemica. The measurements of the optical density and the accounted extinction values were done 30 minutes after the development of ELISA reaction on the spectrophotometer SUMAL PE, Carl Zeiss, Jena, in length of the wave of 405 nm.

Except by DAS-ELISA, some of these viruses, as TSWV, ToMV and CMV, were also identified and by the reaction of the indicator (test) plants: *Nicotiana tabacum*, cv. Samsun NN, *Capsicum annuum*, cv. Sofiyska kapia, *Cucumis sativus* cv. Bistrenski or *Cucurbita pepo* var. giramonia, *Lycopersicon esculentum*, cv. Ideal and *Petunia hybrida* in pepper, tomato and eggplant fruits, bought from the Bulgarian market net. Analyses on indicator (test) plants were done according to Kovachevsky et al. (1995).

RESULTS AND DISCUSSION

The results from DAS-ELISA tests for six virus pathogens, wide spread on pepper, tomato and eggplant fruits, bought from different markets in Sofia, were presented in Table 1. Twelve batches of tomato, pepper and eggplant fruits - all 49 fruits were analyzed and the results showed that TSWV and after that CMV were the most wide spread in the fruits. Both viruses are insect-transmitted viruses. The period from May to September 2018 was very suitable for the appearance of these viruses and their vectors with alternately sun days and rain days. TSWV was established in the most number of fruits - 30 from the total analyzed 49 fruits, while CMV was established in 19 fruits, ToMV – in 17 fruits and PVY – in 12 fruits. TSWV was established in fruits of 10 batches from 12 market batches, originated from different places and CMV – in fruits of 9 market batches and ToMV – in fruits of 7 market batches. TSWV is the major virus causing injuries on the fruits from vegetables in *Solanaceae* family (Table 1), as Dikova (2018) was established from pepper fruits, originating from different markets and areas of production. TSWV, CMV, ToMV and PVY were the most often established viral pathogens in pepper and tomato fruits, while AMV and PVX were sporadically established in the same fruits (Table 1). The data for most spread viruses on tomato and pepper fruits in 2018 were identical with the data from 2012, 2013 and 2014 (Dikova, 2014).

The symptoms caused by the virus diseases on tomato, pepper and eggplant fruits were chlorotic (yellow) spots, ring or irregular, compact or concentric spots and deteriorated more or less the market appearance of the fruits. Such fruits originated from the plants, which were entire put from the viral pathogens and the end the pathogens put of the maturing fruits. The symptoms on the fruits were also dependent on the individual or mixed viral infections, caused from one or several viral pathogens.

The individual infection was established comparatively most rarely in comparison with the mixed infection and in this case only one from five or six analyzed by DAS-ELISA viruses was in high viral concentration, while for the other viruses negative or nearly negative extinction values were accounted. The virus, practically with an individual infection, caused symptoms on the pink tomato from batch of Bankya market is *Tomato spotted wilt virus* (TSWV) and for it a high extinction value – 1,069 OD (Optical Density) was accounted, but for *Tomato mosaic virus* (ToMV) the extinction value for the same pink tomato is near negative -0,349 OD in

Table 1. Established viruses in market batches of fruits from tomato, pepper and eggplant

Вид зеленчук от пазар/ Kind of vegetable fruits and market	Партиди/ Batches and origin of production	Брой тестирани плодове от една партида/ Number of tested fruits from one batch	Установени вируси / Established viruses					
			TSWV	ToMV	CMV	AMV	PVY	PVX
Розови домати – Женски пазар, София/ Pink tomato – Female Market, Sofia	Сандански/ Sandansky	4	4/4	4/4	0/4	0/4	1/4	0/4
Розови домати – пазар Банкя/ Pink tomato – Bankya Market	Пловдивско/ Near Plovdiv	3	3/3	1/3	0/3	0/3	0/3	0/3
Розови домати – пазар „Д. Петков", София/ Pink tomato "D. Petkov" Market, Sofia	Огняново, Пазарджишко/ Ognyanovo, near Pasardjik	5	5/5	5/5	1/5	0/5	1/5	1/5
Червени домати – тип консерва/ Red tomato, Konserva type	Промаркет, Люлин/ Promarket Lyulin Suburb, near Sofia	4	2/4	¼	¼	1/4	¼	0/4
Червени домати – тип Ръгби, магазин в Костинброд Red tomato Ragby type, Kostinbrod	Гоце Делчевско/ Near Gotse Delchev	6	3/6	4/6	5/6	0/6	0/6	0/6
Зелено-червена капия – Женски пазар, София/ Green-red long fleshy pepper, Female Market, Sofia	Петърч/ Petarch Suburb near Kostinbrod	3	3/3	0/3	3/3	0/3	1/3	0/3
Червена капия, магазин в Костинброд Red long fleshy pepper, Kostinbrod Market	Пазарджишко/ Near Pasardjik	4	4/4	0/4	¼	0/4	0/4	0/4
Червено-зелена капия/ Green-red long fleshy pepper	Промаркет, Костинброд/ Promarket Kostinbrod	5	0/5	0/5	0/5	0/5	0/5	0/5
Червена капия/ Red long fleshy pepper	Промаркет, Люлин/ Promarket Lyulin suburb, near Sofia	5	0/5	0/5	3/5	0/5	3/5	1/5
Червена камба, пазар Банкя/ Red round fleshy pepper, Bankya Market	Пловдивско/ Near Plovdiv	4	2/4	1/4	2/4	0/4	3/4	2/4
Червена камба, пазар Банкя/ Red round fleshy pepper, Bankya Market	Шабла/ Shabla, near Varna	3	3/3	0/3	2/3	2/3	2/3	2/3
Патладжани/ Eggplant	Промаркет, Люлин/ Promarket Lyulin suburb, near Sofia	3	1/3	1/3	1/3	0/3	0/3	0/3
All fruits		49	30	17	19	3	12	6
Tomato fruits		22	17	15	7	1	3	1
Pepper fruits		24	12	1	11	2	9	5
Eggplant fruits		3	1	1	1	0	0	0

comparison with cut off 0,373 OD (Figure 1 and the chart of the Figure 21). Cut off is the border value for each DAS-ELISA test between the negative and positive extinction values.

Mixed viral infection was established on pink tomatoes originated from Ognyanovo village, a batch from Sofia market. High extinction values over 1,0 OD and 2,0 OD were measured for TSWV

Figure 1. Pink tomato fruit with symptoms of fine chlorotic line pattern, caused by TSWV, Bankya market

Figure 2. Pink tomato fruit with severe spotting, caused by mixed infection of TSWV and ToMV, Ognyanovo village, near Pasardjik

Figure 3. A part of a batch pink tomato fruits with spotting, caused by TSWV and ToMV, Ognyanovo village near Pasardjik

Figure 4. Chlorotic spots and line pattern, caused by TSWV on red tomato, type Konserva, Promarket

Figure 5. Chlorotic spots and line pattern, caused by TSWV on red tomato, type Ragby, near Gotse Delchev

Figure 6. Symptoms of severe ring spots, caused by mixed infection of TSWV and CMV on red tomato, Promarket, import of Albania

and ToMV for five spotted tomato fruits from Ognyanovo village (Figures 2, 3 and the chart of the Figure 21).

The symptoms, caused by the TSWV individual infection was moderate fine spots, fine rings and fine line patterns (Figure 1), but the symptoms, caused from the mixed infection of TSWV and ToMV were bright and rough yellow spots (Figures 2 and 3). Red tomato fruit, batch of T-Market, Sofia and import from Albania showed bright yellow-green concentric spots, caused by the TSWV and CMV mixed infection (Figure 10).

Symptoms, as spotting, line patterns and even color changes, caused by virus diseases on different types of tomatoes (pink, red, Konserva, Ragbi) are shown on Figures from 1 to 6; on different types of pepper (long fleshy – kapia and round fleshy – kamba) – on Figures from 7 to 14, and eggplant – on Figures 15 and 16.

Symptoms of individual infection on pepper were shown on Figures 7, 8, 9 and on mixed infection – on Figures 10, 11, 12, 13, 14. Varied and variegated symptoms were caused from the mixed infections of TSWV and CMV on long fleshy pepper – kapia (Figures 11-12).

The established viruses in detail were shown on Figures from 17 to 20. Twelve fruits of pink tomato with yellow spots and figures, bought from three markets and presenting different batches showed different viral concentration for viruses (Figure 17). Pink tomato fruits from Sandansky town (Female market, Sofia – samples 1-4) had high viral concentration for TSWV, moderate viral concentration for ToMV and the other viruses missed with only one exception (a sample 3 with PVY in low viral concentration, Figure 17). A similar situation was for the batch of pink tomato fruits from Bankya (samples 5-7). Pink tomatoes from Ognyanovo village showed TSWV and ToMV in very high viral concentration and per one of the fruits – CMV, PVY and PVX (samples 8-12, Figure 17). Climatic conditions in the growth season 2018 were very susceptible for aphid transmitted viral pathogens, as CMV and PVY. The high viral concentration of ToMV in five tomato fruits from Ognyanovo village can be explain with the specific precondition for superficially-contaminated seeds or seedling contamination by garden tools. PVX is seldom in tomato and pepper fruits: only in one fruit from the all tested 22 tomato fruits and in five pepper fruits from the all tested 24 pepper fruits (Table 1). PVX is mechanically transmitted virus, as ToMV.

TSWV, ToMV, CMV, AMV and PVY were established in red tomato fruits, late subsequent production types Konserva and Ragby (Figure 18). Four fruits of type Konserva from Promarket had TSWV, ToMV, CMV, AMV and PVY(samples 1-4, Fig. 18) with one exception of high viral concentration from TSWV and the other viruses in low viral concentration. The fine chlorotic spottings (Figure 4) on Konserva fruits are probably caused by TSWV (samples 2-3, Figure 18). Six red tomato fruits type Ragby, originated near Goze Delchev town (samples 5-10, Figure 18) showed a mixed infection between several from the tested viruses, as TSWV and ToMV established in 4 fruits, and TSWV and CMV – in five fruits.

The optimum atmospheric conditions for virus diseases development in the growth season of 2018 advantaged an appearance not only of the thrips-transmitted TSWV in high viral concentration, but of the aphid-transmitted viruses as CMV on pepper fruits type long fleshy – kapia (Figure 19). TSWV was established in 9 from total 20 kapia fruits from 4 batches, CMV – in 7 kapia fruits, PVY – in 4 kapia fruits and ToMV and PVX per in 1 fruit (Figure 19). TSWV on kapia fruit (Figure 7) and mixed infection by TSWV and CMV on kapia fruit (Figure 8) were established in Kostinbrod private market.

Individual fruits of red long flashy pepper, bought from Promarket, Kostinbrod, were with severe symptoms of yellow ring spots or concentric spots and chlorotic line pattern and they showed very low extinction values for the viruses, as the negative control (samples 5-14, Figure 19). This situation can be explained with long time staying of the pepper fruits outdoors exposed to the sunshine and the viruses were inactivated. Only one exception in the batch of Promarket pepper fruits – one pepper fruit - sample 12 showed high viral concentration of TSWV, which is a proof for the role of TSWV for the injuries and the bad market appearance of the pepper fruits. Pathogens with other origin (Fungi, Bacteria) or saprophytes can be appeared by such long storage, as red long flashy pepper in Promarket (Figure 10). High viral concentration of aphid-transmitted viruses, as CMV and PVY was established in green-red long flashy pepper fruits, belonging to a batch from Petarch village, near Kostinbrod (samples 15-20, Figure 19).

Symptoms of virus diseases, attended with injuries and bad fruit appearance were seen on round fleshy pepper (type Kamba) too (Figures 13-14). TSWV and CMV and one case of PVX were often established in high viral concentration (over 1,0 OD) in fruits from villages near Plovdiv (samples

Figure 7. Fine chlorotic ring spots, caused by TSWV on red long fleshy pepper, a private market, Kostinbrod

Figure 8. Large chlorotic area, caused by TSWV and CMV on deformed fruit of long fleshy pepper, Promarket, Sofia

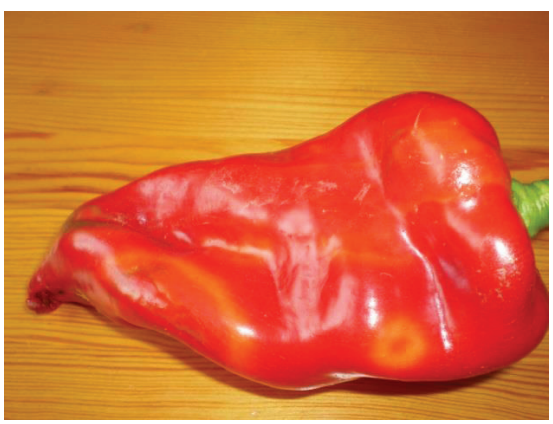

Figure 9. Severe ring spot by TSWV, causing deformations on long fleshy pepper, Promarket, Kostinbrod

Figure 10. Symptoms of TSWV and ToMV, accompanied by necrotic symptoms from saprophytic or other pathogens, Promarket, Kostinbrod

Figure 11. Severe symptoms, caused by mixed infection of TSWV and CMV, Petarch village, near Kostinbrod

Figure 12. Severe symptoms, caused by mixed infection of TSWV and CMV, Petarch village, near Kostinbrod

Figure 13. Large chlorotic areas and spots, caused by TSWV and CMV on round fleshy pepper, type Kamba, a village near Plovdiv

Figure 14. Chlorotic spots, caused by TSWV and CMV on round fleshy pepper, type Kamba, Shabla village near Varna

Figure 15. Eggplant fruits with symptoms of fine spotting and decoloration, caused by TSWV, in the middle-symptomless fruit, Promarket, Sofia

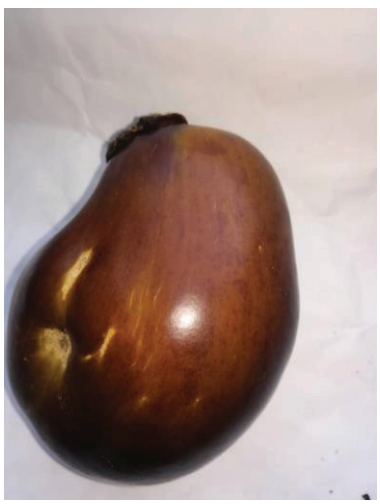

Figure 16. Eggplant fruit with symptoms of fine spotting and decoloration, caused by TSWV, ToMV, CMV, Promarket, Sofia

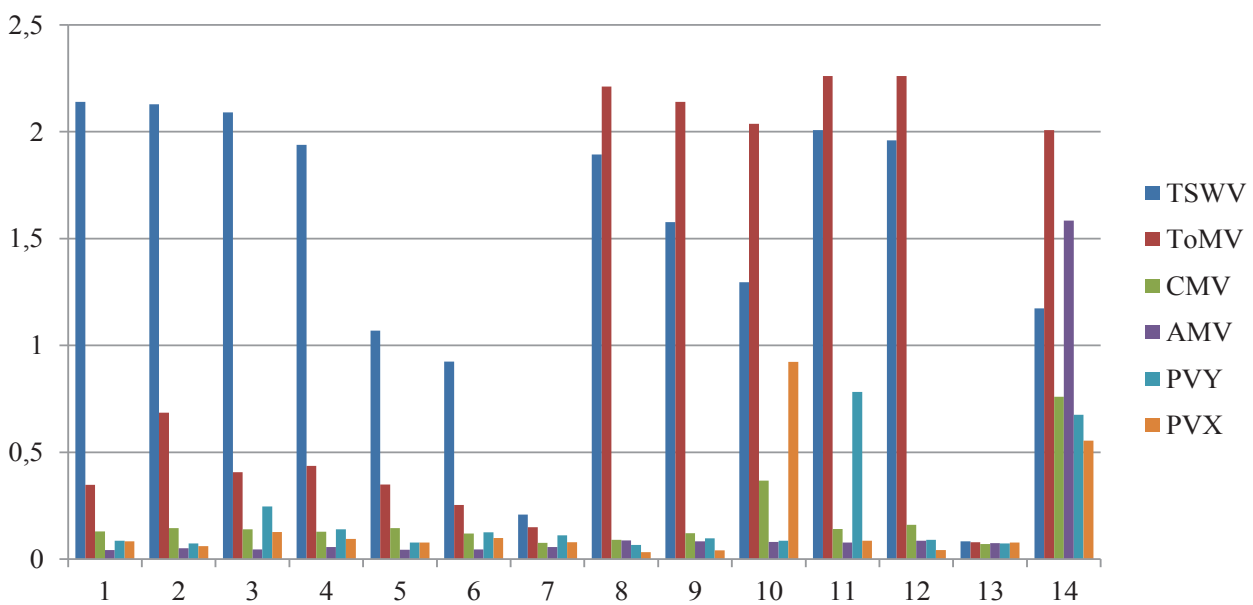

Fig. 17. Viral pathogens on pink tomato fruits, different batches from market net, tested by DAS-ELISA in 2018

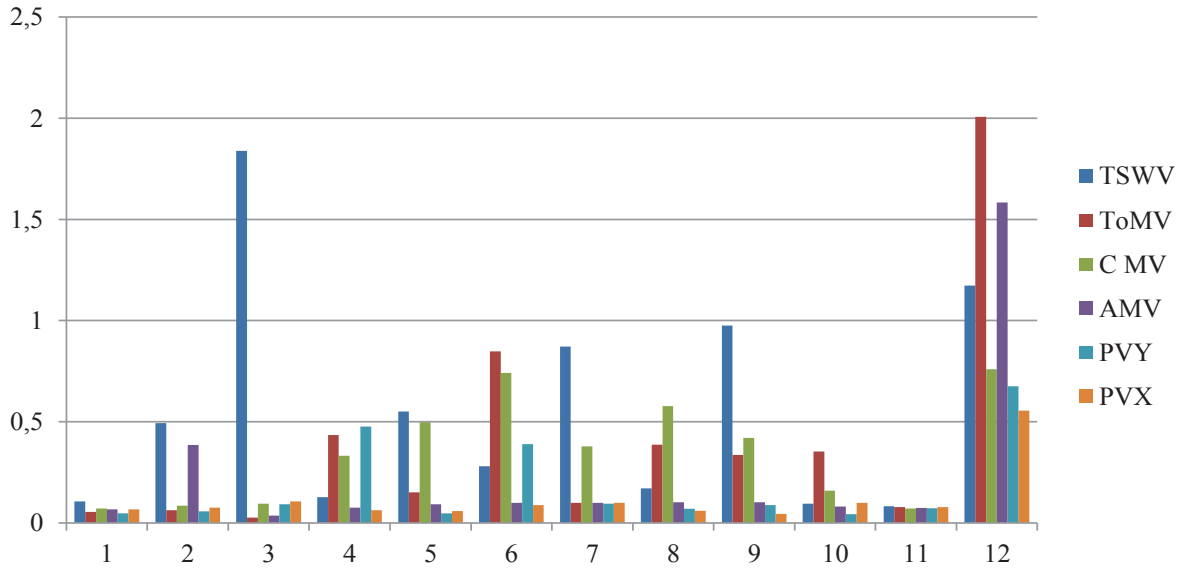

Fig. 18. Viral pathogens on red tomato fruits (late subsequent production for tinned food), different batches from market net, tested by DAS-ELISA in 2018

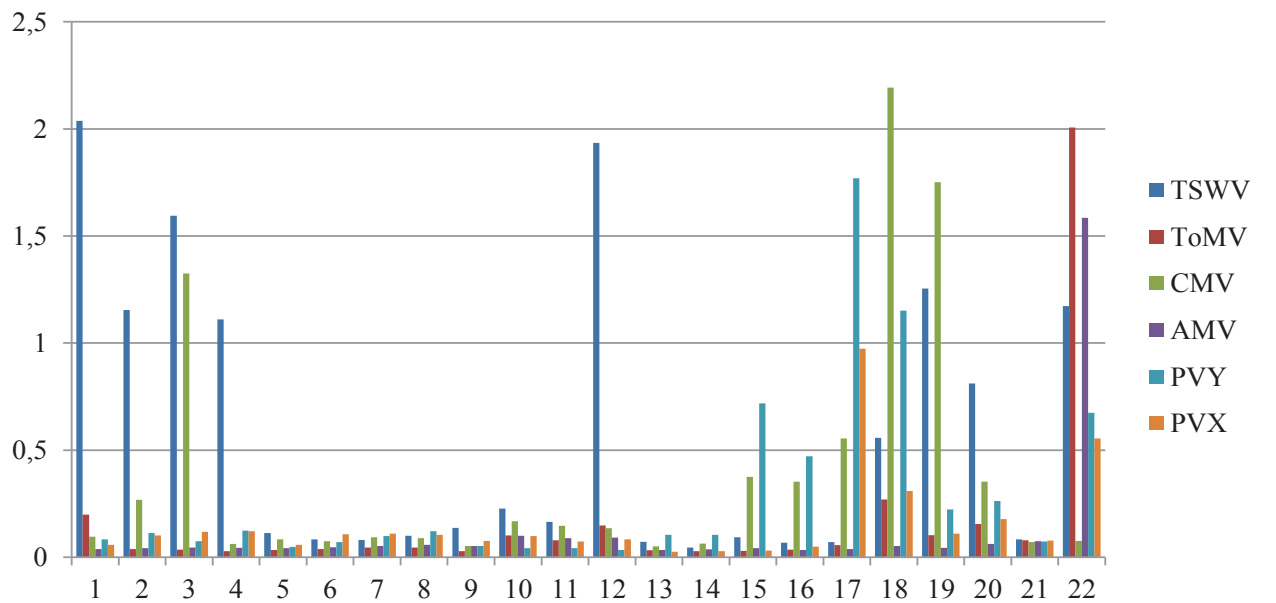

Fig. 19. Viral pathogens on red long fleshy pepper fruits (kapia), different batches from market net, tested by DAS-ELISA in 2018

1-4, Figure 20) and in fruits from Shabla, North Bulgaria (samples 5-8, Figure 20). Moderate viral concentration was established for ToMV, PVV and PVX in the round fleshy pepper from both batches. Sample number 5 is symptomless round fleshy pepper fruit with very low viral concentration.

TSWV is the main virus, isolated from pepper fruits in Bulgaria and it is wide spread in pepper fields (Dikova, 2014). TSWV was established in 12 pepper fruits (50 %), CMV – in 11 pepper fruits (46 %) and PVY – in 9 pepper fruits (38 %) from all 24 pepper fruits, tested in the present study - 2018. For comparison with our data *Potato virus Y* (PVY) was in the highest percentage (79%) and CMV (61%) and ToMV (23%) in pepper crops of Nigeria (Olawale et al., 2015). Similar situation is in Turkey,

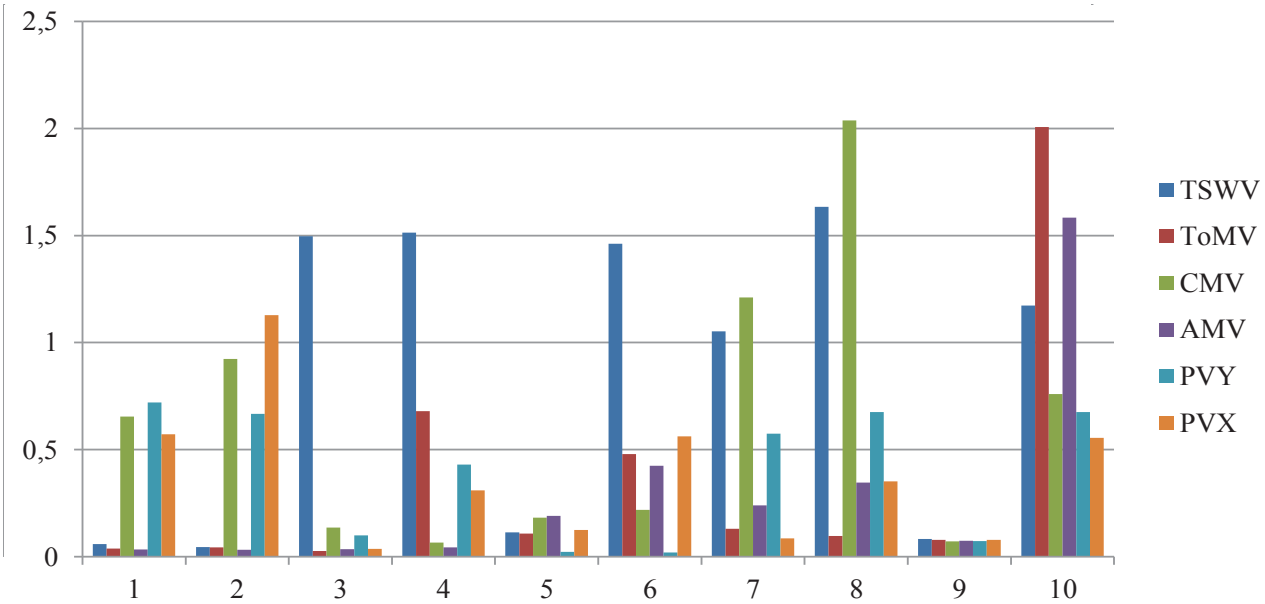

Fig. 20. Viral pathogens on red round fleshy pepper fruits (kamba), different baches from market net, tested by DAS-ELISA in 2018

where PVY was determined as the most common virus in pepper growing areas in Hatay, followed by CMV (Ozdag & Sertkaya (2017). Therefore aphid-transmitted viruses CMV and PVY are economically important for the pepper culture in different countries.

Viral pathogens, caused symptoms of spotting and color changes were established in eggplant fruits too (Figures 15-16). TSWV, CMV and ToMV were established in one of eggplant fruits (a batch from Promarket, Lyulin suburb, near Sofia).

More spread plant viral pathogens mainly TSWV, but also CMV and ToMV on tomato, pepper and eggplant fruits were identified and by indicator (test) plants (Figures 21- 26). Indicator (test) plants for TSWV are shown on Figures 21-24. Some indicator plants were used for ToMV – *Nicotiana tabacum* cv. Samsun NN (Figure 25) and for CMV – *Cucurbita pepo* var giramonia (Figure 26). Systemic symptoms on *Nicotiana tabacum* cv. Samsun NN, *Capsicum annuum* cv. Sofiyska kapia and *Lycopersicon esculenthum* cv. Ideal, caused by red long fleshy pepper fruit, in which only TSWV in high viral concentration was established by DAS-ELISA, were shown on Figures 21-23. Local lesions, caused by TSWV were formed after the treatment with inoculum of eggplant fruit in which TSWV was established in high viral concentration by DAS-ELISA (Figure 24). ToMV from pink tomato fruit was identified on *Nicotiana tabacum* cv. Samsun NN with only local lesions without symptoms of systemic infection (Figure 25). CMV from round fleshy pepper (type Kamba) was identified on *Cucurbita pepo* var. *giramonia*, cv. Izobilna (Figure 26).

CONCLUSION

The results from the studies of the symptoms on virus diseases on tomato, pepper and eggplant fruits showed that the viral pathogens cause serious injuries on the fruits that worsen their market appearance qualitatively. ELISA method (DAS-ELISA) permitted an analyzing of a large number of fruit samples, collected by different markets and originated by different production areas simultaneously for several viral pathogens. The indicator method by using some differential indicator (test) plants, as *Nicotiana tabacum* cv. Samsun NN, which was differential indicator for TSWV, ToMV and CMV is very suitable for visually accounting of each of these plant viruses (Dikova, 2016). The present monitoring of viral diseases on fruits of vegetable plants, belonging to *Solanaceae* fam-

Figure 21. Systemic symptoms on *Nicotiana tabacum* cv. Samsun NN, caused by TSWV

Figure 22. Chlorotic systemic ring spots on *Capsicum annuum* cv. Sofiyska kapia, caused by TSWV

Figure 23. Chlorotic line pattern, turned to necrotic, caused by TSWV on *Lycopersicon esculenthum*, cv. Ideal

Figure 24. Local lesions, caused by TSWV on *Petunia hybrida*

Figure 25. Local lesions, caused by ToMV on *Nicotiana tabacum*, cv. Samsun NN

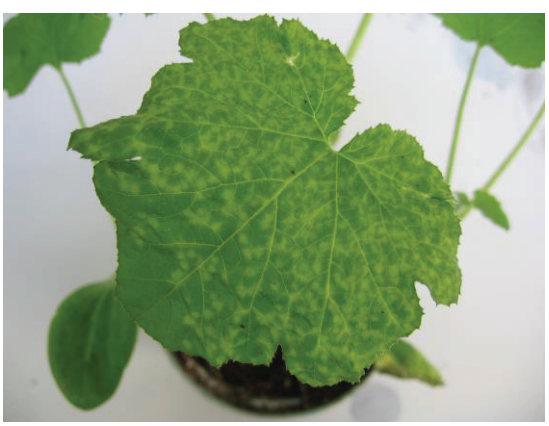

Figure 26. Systemic symptoms, caused by CMV on *Cucurbita pepo* var. *giramonia*

ily showed that the control of virus diseases on these vegetable plants would be begun as prophylaxis against them, therefore against their vectors and reservoirs.

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