Agrarian Sciences

Occurrence of the Pathotype 38 of *Synchytrium Endobioticum* in Khulo Municipality of Georgia

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Potato wart caused by the obligate biotrophic, soil-borne fungal pathogen Synchytrium endobioticum (Schilb.) Perc. is one of the important quarantine diseases of cultivated potatoes (Solanum tuberosum L.) worldwide. The pathogen included on A2 quarantine list of EPPO occurs locally in almost all countries in the EPPO region. The disease is widely distributed in Khulo municipality since 2009, which is the main highland potato area production in the Autonomous Republic of Adjara. The pathotype (38 Nevsehir) of the pathogen was first identified in the village of Didajara in 2019. Studies for the identification of the pathotype were conducted in another three villages of Khulo Municipality by using field, pot and Glynne-Lemmerzahl tests. According to the EPPO diagnostic protocol, differential potato cultivars: Gawin, Deodara, Producent, Delcora Belita, Talent, Saphir and Provento were used to see their responses to the isolates pw 14, pw 15 and pw 16. In all three experiments, the cultivars Deodara, Producent, Talent and Saphir were susceptible to isolates of PWD, and the cultivars Gawin, Delcora, Belita and Provento expressed a resistant response. Therefore, when comparing the test results with the results of the Turkish isolate, the differentials showed similar reactions to the Georgian and Turkish isolates. As a result of the similarity of reaction types, the pathotype prevalent in Georgia resembles the pathotype in Turkey 38 (Nevsehir). Based on the results, it is concluded that the same pathotype, which was firstly identified and spread at Didajara village, was also identified in Skvana, Uchkho and Dzirkvadzeebi villages. © 2020 Bull. Georg. Natl. Acad. Sci.

Synchytrium endobioticum, potato wart, pathotype, Khulo municipality

Potato is an important crop which holds promise food to millions of people especially in developing countries. Thus, it is an important consumed commodity in Georgia, where the annual per capita consumption is 55 kg [1], while it is considered by consumers as "second bread". The crop cultivation area covers approximately 20,000 ha. Despite the potato's importance in Georgia the yields still remain

rather low (11 t/ha) due to the fact that the production of potatoes is associated with big challenges [2]. The most crucial of these challenges are the potato diseases and particularly potato warts (PWD). The casual organism is the pathogenic fungus *Synchytrium endobioticum* [3-5], while the disease is under quarantine control. More than 30 pathotypes of the pathogen are identified and widespread in nearly 60

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countries worldwide [6]. Their yield losses range from 50 to 100% [7,8]. The PWD was first reported in Georgia in 2009, without knowing the pathotypes, at the villages of Tabakhmela and Didajara in Khulo Municipality [9]. Furthermore, more infested fields were found in the years 2017 and 2018 in the same Municipality [10]. Thus, the pathotype 38 was reported in 2019 at Didajara village in Khulo [11], it is assumed that this pathotype has been spread to Georgia from Turkey, where it was first reported in 2009 [12].

The aim of the present paper was to identify the pathotypes, isolated in potato cultivated areas located in the three abovementioned villages in the Municipality of Khulo. This study can provide a deeper knowledge of the situation of the infestation of the PWD in southwest of Georgia.

Materials and Methods

To identify the pathotypes present at Skvana, Uchkho and Dzirkvadzeebi villages located in Khulo Municipality, field, pot tests and Glynne–Lemmerzahl tests were carried out according to the EPPO Diagnostic protocols [13,14].

Field tests. For the identification of the pathotype present two field tests were carried out in the

private home gardens of Skvana and Uchkho villages, field tests were carried out according to the EPPO Diagnostic protocol [13].

The first field test was conducted in 2017 at the village Skvana at an altitude of 1291 m above sea level (N41*32.875, E04*29.098). On the 7th of May, 30 tubers of nine differential potato cultivars were chosen according to EPPO diagnostic protocol: Gawin, Deodara, Producent, Delcora Belita, Talent, Saphir, Provento and Marfona. The potato seeds were planted directly into the soil of infested home garden plots covering an area of 100 m² and harvested by hand on the 16th of September 2017. Moreover, another field test was conducted in 2018 at the village Uchkho at an altitude of 1083 m above sea level (N41*40.969, E042*18513). On the 24th of April, the same abovementioned differential cultivars were planted using similar experimental design as in Skvana village, while harvesting was performed on the 27th of July 2018. In both field tests a similar set of eight differential cultivars were used: Gawin, Deodara, Producent, Belita, Talent, Saphir mentioned in the EPPO Diagnostic Protocol [14], and cultivars Delcora and Provento, which were commonly used in the Netherlands and other potato growing countries for pathotype tests in 2000-2015. The susceptibility of the eight cultivars to the PWD

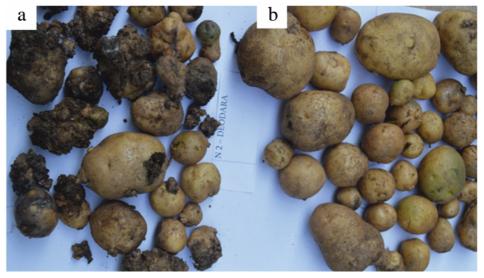


Fig. Potato "Deodara": a) infected, b) healthy.

was evaluated for each individual plant separately, according to standard protocol [13] by measuring the size and the number of warts.

The collected wart materials from the fields of Skvana, Uchkho and Dzirkvadzeebi villages were used for preparation of composts according to the Spieckermann method [14]. The inoculums were used to conduct the pot tests in 2017 at Glynne Lemmerzahl in order to identify the PWD pathotype. Before planting, the soil samples were collected and examined to measure the density of sporangia per g soil (with a density of 8-10 winter sporangia in the soil of Skvana village, and 5-8 winter sporangia in the soil of Uchko village) [13].

Pot tests. In addition to field, the pot tests were conducted in 2019 to identify the PWD pathotype present in the soils of Skvana, Uchkho and Dzirkvadzeebi villages [13].

Soil samples originating from the infested fields of the abovementioned villages were placed into pots of 5 L size, infected with PWD inoculum. 3-5 tubers of each differential cultivar were planted per pot with three replications including the cultivar Marfona as a positive control (PC). The pots were placed into a glasshouse in the middle of spring with supplementary lighting and manual watering to keep soil moisture at appropriate levels for potato cultivation. The temperature and the relative humidity in the glasshouse ranged from 18 to 20°C and 70-85%, respectively. Afterwards, the grown potato plants were removed from pots and examined for PWD symptoms. The susceptibility of the tested cultivars to the fungus was assessed using the Spieckermann scale [11,15].

Glynne, Lemmerzahl test. The Glynne-Lemmerzahl test was carried out in 2019 in order to identify the kind pathotype present in the fields of Dzirkvadzeebi village, according to the EPPO Diagnostic protocol 2017. Fresh wart materials collected from infected tuber blocks of the cultiavar Deodara were used for pathotype identification. A

set of eight differential cultivars with three replications were inoculated with the fungus and incubated for six weeks in a growth chamber at a constant temperature of 18°C (±1°C). PWD was assessed according to the scale used for the Spieckermann test [4,15].

Results

Field Tests. The results of the field tests of 2017 and 2018 are presented in both field tests, the cultivars Deodara, Producent, Talent and Saphir were susceptible to PWD. Particularly, in "Deodara" 21.8% and 6.6% tubers were susceptible expressing different levels of susceptibility. The predominant wart symptom in the first tests was a single proliferation >5mm (class 2 - S), and 2 or 3 proliferations (<5 mm) or a single large proliferation (5–10 mm) (class 3 - S) in the second test. In "Producent" 20.7%and 16.3% tubers were susceptible. The predominant wart symptom in both tests was a single proliferation >5mm (2 - S). In "Talent" 9.8% and 16.3% tubers were susceptible. In the first test, the predominant wart symptom was very large warts, no normal tubers present (class 9-S), and 2 or 3 proliferations (<5 mm) or a single large proliferation (5–10 mm) (class 3 - S) in the second test. In "Saphir" 10.2%and 7.0% tubers were susceptible. The predominant wart symptom in the first tests were 2 or 3 proliferations (<5 mm) or a single large proliferation (5-10 mm) (class 3 - S), and a single proliferation >5mm (class 2 - S) in the second test. On the contrary, the cultivars Gawin, Delcora, Belita and Provento expressed high resistant response.

Glynne Lemmerzahl test. Table 1 shows the results of the Glynne–Lemmerzahl test in 2019. The cultivars Deodara, Producent, Talent and Saphir expressed similar susceptible response to the isolate pw 15, originating from the village of Uchkho, results which were obtained in the pot tests. However, she ize of wart proliferations varied within the three tests. In contrast, cultivars Gawin, Delcora, Belita and Provento expressed a resistant response.

Table 1. Reactions of differentials to isolate pw16 originating from the village of Dzirkvadzeebi, Khulo municipality using Glynne Lemmerzahl test, 2019

N	Differentials	Number of tuber eyes recorded	Number of diseased tuber eyes	Resistant			Weakly Suscep- tible	Susceptible -S				Reaction			
				1 2 3			4	5							
				0 *	-	P	F	R	I	II	III	IV	V	X	
1	Gawin	15	0	15											R
2	Deodara	15	15						3	2	8	2			S
3	Producent	15	15						6	6	3				S
4	Delcora	15	0	11	4										R
5	Belita	15	0	15											R
6	Talent	15	15						4	6	5				S
7	Saphir	15	13			1	1		3	5	5				S
8	Provento	15	0	9	6										R
8	Provento	15	0	7	8										R

Table 2. Overall results of susceptibility levels of the differential cultivars to the isolates pw 14, 15 and 16 using field, pot and Glynne–Lemmerzahl tests

N	Differential cultivars	Skvana, 2017 Field Test	Skvana, 2019 Pot test (Isolate pw 14)	Uchkho, 2018 Field Test	Uchkho, 2019 Pot test (Isolate pw 15)	Dzirkvadzeebi, 2019 Pot test (Isolate pw 16)	Dzirkvadzeebi, 2019 Glynne– Lemmerzahl test (Isolate pw 16)
1	Gawin	R	R	R	R	R	R
2	Deodara	S	S	S	S	S	S
3	Producent	S	S	S	S	S	S
4	Delcora	R	R	R	R	R	R
5	Belita	R	R	R	R	R	R
6	Talent	S	S	S	S	S	S
7	Saphir	S	S	S	S	S	S
8	Provento	R	R	R	R	R	R
PC	Marfona	S	S	S	S	S	-

Discussion. Differential cultivars showed the similar levels of susceptibility in the field, pot and Glynne–Lemmerzahl tests (Table 2). The cultivars Deodara, Producent, Talent and Saphir were susceptible to the isolates pw 14, pw 15 and pw 16 originated from the villages of Skvana, Uchkho and Dzirkvadzeebi, respectively. Furthermore, the cultivars Gawin, Delcora, Belita and Provento expressed a resistant response to the abovementioned isolates.

Similar susceptibility levels were obtained by comparing the susceptibility results with the standard differential cultivars set referred in EPPO Diagnostic protocol (EPPO, 2017), while in these

tests we used two extra differential cultivars the "Delcora" and "Provento". These cultivars were used in the study of Turkish [12] and Georgian isolates originating from the village of Didajara, Khulo municipality.

Conclusion. Based on the comparisons of the results presented in the current study with the results obtained recently by the village of Didajara of the Khulo municipality [11], it is concluded that the pathotype found in the villages of Skvana, Uchkho and Dzirkvadzebi, resembles the pathotype 38 in its responses. This pathotype is widespread in Turkey and Khulo municipality of Georgia.

აგრარული მეცნიერებანი

Synchytrium endobioticum-ის პათოტიპ 38-ის გავრცელება ხულოს მუნიციპალიტეტში

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კარტოფილის (Solanum tuberosum L.) ერთ-ერთ მნიშვნელოვან საკარანტინო დაავადებას კარტოფილის კიბო წარმოადგენს, რომელსაც იწვევს ობლიგატური, ბიოტროფული, ნიადაგის სოკო Synchytrium endobioticum (Schilb.) Perc. დაავადების გამომწვევი სოკო ევროპისა და ხმელთაშუაზღვისპირა ქვეყნების მცენარეთა დაცვის ორგანიზაციის საკარანტინო ორგანიზმთა A2 ჩამონათვალშია შეტანილი. 2009 წლიდან კარტოფილის კიბოს გამომწვევი პათოგენი Synchytrium endobioticum ფართოდაა გავრცელებული ხულოს მუნიციპალიტეტში, რომელიც მეტად მნიშვნელოვანი მთიანი ზონაა კარტოფილის წარმოების თვალსაზრისით საქართველოში, განსაკუთრებით კი, აჭარაში. კარტოფილის კიბოს გამომწვევი პათოგენის პათოტიპი (38 Nevşehir) პირველად იდენტიფიცირებულ იქნა ხულოს მუნიციპალიტეტის სოფელ დიდაჭარაში 2019 წელს. პათოტიპების იდენტიფიკაცია განხორციელდა ხულოს მუნიციპლიტეტის კიდევ სამ სოფელში აღებულ ნიმუშებში ე.წ. ქოთნის მინდვრისა და გლინ-ლიმერზალის ცდების გამოყენებით. EPPOს დიაგნოსტიკური პროტოკოლის მიხედვით, კარტოფილის ჯიშ-დიფერენციატორები: გავინი, დეოდორა, პროდუსენტი, დელკორა, ზელიტა, ტალენტი, საფირი და პროვენტო იქნა გამოყენებული კარტოფილის კიბოს იზოლატების w 14, pw 15 და pw 16 მიმართ საპასუხო რეაქციის გამოსაცდელად. სამივე ექსმერიმენტის დროს, ჯიშები: დეოდორა, პროდუსენტი, ტალენტი და საფირი აჩვენებდა მიმღებ რეაქციებს, ხოლო გავინი, დელკორა, ბელიტა და პროვენტო – გამძლე რეაქციას დაავადების მიმართ. ცდის შედეგების შედარებისას მოხდა თანხვედრა თურქული იზოლატის ცდის შედეგებთან, ანუ დიფერენციატორები აჩვენებდა მსგავს რეაქციებს ქართული და თურქული იზოლატების მიმართ. რეაქციის ტიპების მსგავსების შედეგად, საქართველოში გავრცელებული პათოტიპი წააგავს თურქეთში გავრცელებულ პათოტიპს 38(Nevşehir). მიღებული შედეგების საფუძველზე, ხულოს მუნიციპალიტეტის სოფლებში: სკვანაში, ძირკვაძეებსა და უჩხოში გავრცელებულია იგივე პათოტიპი (38 Nevşehir), რაც სოფელ დიდაჭარაში იყო იდენტიფიცირებული.

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REFERENCES

- Geostat. (2019) Food balance sheets. Balance sheet for potato. National Statistical Office for Georgia, Tbilisi, Georgia.
- 2. CIP (2019). Plan for improving seed potato in Georgia. RTB Working Paper. CIP, Lima, Peru.
- 3. Smith IM., McNamara DG., Scott PR., Holderness M., Burger B. (1997) Qurantine pests for Europe. Data sheets on quarantine pests for European Union and for the European and Medterranean Plant Protection Organisation, 2nd ed. CAB International, Wallingford.
- 4. Baayen HP., Bonthuis H., withagen JCM., wander JGN., Lamers JL., Meffert JL., Cochius G. et al. (2005) Resistance of potato cultivars to *Synchytrium endobioticum* in field and laboratory tests, risk of secondary infection, and implications for phytosanitary regulations. *ePPO Bull.*, **35**(1):9–23.
- 5. van de Vossenberg B., Westenberg M., Adams I., Afanasenko O., Besheva A., Boerma M. et al. (2018) Euphresco Sendo: an international laboratory comparison study of molecular tests for *Synchytrium endobioticum* detection and identification. *European Journal of Plant Pathology*, 151:757–766.
- 6. CABI (2013) *Synchytrium endobioticum* (wart disease of potato). Crop Protection Compendium. Last accessed November 4, 2013 from: www.cabi.org/cpc.
- Hampson MC. (1993) History, biology and control of potato wart disease in Canada. Can. J. Plant Pathol., 15:223–244.
- 8. Melnik PA. (1998) wart disease of potato, *Synchytrium endobioticum* (Schilbersky) Percival. EPPO Technical documents 1032, Paris.
- 9. Gorgiladze L., Meparishvili G., Sikharulidze Z., Natsarishvili K., Meparishvili S. (2014) First report of *Synchytrium endobioticum* causing potato wart in Georgia. *New Disease Reports*, 30:4.
- Ghoghoberidze S., Sikharulidze Z., Meparishvili G., Gorgiladze L., Meparishvili S. (2018) Occurrence of potato wart in Georgia. *Proceedings of IX International Scientific Agricultural Symposium*. Bosnia and Herzegovina, p. 876–880. http://agrosym.ues.rs.ba/agrosym/agrosym_2018/BOOK_OF_PROCEEDINGS_2018_FINAL.pdf [accessed on 04 October 2018]
- 11. Sikharulidze Z.V., Ghoghoberidze S.Y., Mentink N.M., Meparishvili G.V., Tsetskhladze Ts.M., Leeuwen G.C.M. (2019) Identification of the pathotype of *Synchytrium endobioticum*, causal agent of potato wart disease, present in Georgia. *EPPO Bulletin*, 49: 314–320.
- 12. Cakir E., van Leeuwen G.C.M., Flath K., Meffert J.P., Janssen W.A., Maden S. (2009) Identification of pathotypes of *Synchytrium endobioticum* found in infested fields in Turkey. *Bulletin OEPP/ EPPO Bulletin*, 39: 175–178.
- EPPO (2004) EPPO Standard PM 7/28 (1) Synchytrium endobioticum. Bulletin OEPP/EPPO Bulletin, 34:213–218.
- EPPO (2017) EPPO Standard PM 7/28 (2) Synchytrium endobioticum. Bulletin OEPP/EPPO Bulletin, 47(3), 420–440.
- 15. Dimitrova L., Laginova M., Becheva A., van Leeuwen G. C. M. (2011) Occurrence of potato wart disease (*Synchytrium endobioticum*) in Bulgaria: identification of pathotypes(s) present. *Bulletin OEPP/EPPO Bulletin*, 41:195–202.

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