Załącznik nr 3

AUTHOR'S REVIEW

OF HIS OWN SCIENTIFIC AND SCHOLARLY ACHIEVEMENTS

dr Jarosław Przetakiewicz

Plant Breeding and Acclimatization - Institute - National Research Institute,

Department of Plant Pathology,

Laboratory of Quarantine Organisms,

Radzików

05-870 Błonie

Tel.: 022 733 46 25

e-mail: j.przetakiewicz@ihar.edu.pl

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1. PERSONAL DATA

Name and surname:	Jarosław Przetakiewicz
Date and place of birth:	10 December 1973, Rypin
Place of work:	Plant Breeding and Acclimatization Institute - National Research Institute, Plant Pathology Department, Radzików, 05-870 Błonie, Poland

2. DIPLOMAS AND DEGREES

1993-1998	Nicolaus Copernicus University – Faculty of Biology and Earth Sciences, Department of Biochemistry. (M.Sc.)
2004	Plant Breeding and Acclimatization Institute (PhD in agricultural sciences in agronomy)

3. PROFESSIONAL LIFE

05.10.1998 – 31.03.1999	Internship	Plant Breeding and Acclimatization Institute in Radzików. Plant Transformation and Cell Engineering Laboratory.
01.04.1999 – 31.04.2004	Assistant	Plant Breeding and Acclimatization Institute in Radzików. Plant Transformation and Cell Engineering Laboratory.
01.04.2004 –	Assistant Professor	Plant Breeding and Acclimatization Institute - National Research Institute in Radzików, Department of Plant Pathology, Laboratory of Quarantine Organisms
01.01.2008 – 30-06-2011		Head of Laboratory Laboratory of Quarantine Organisms in Department of Plant Pathology of Plant

Breeding and Acclimatization Institute - National Research Institute in Radzików

4. INDICATION OF THE SCIENTIFIC ACHIEVEMENT resulting from Article 16 sec. 2 of the Act of March 14, 2003. on academic degrees and academic titles, and on degrees and title in the field of art (Journal of Laws of 2016, item 882, as amended in Journal of Laws of 2016, item 1311):

A) TITLE OF SCIENTIFIC ACHIEVEMENT:

"Characterization of the fungus population Synchytrium endobioticum (Schilb.) Perc. occurring in Poland and assessment of potato resistance to its virulent pathotypes"

B) PUBLICATIONS COMPRISING THE SCIENTIFIC ACHIEVEMENT:

H1. **Przetakiewicz J**. 2009. Propozycja zmian w polskiej skali oceny odporności odmian ziemniaka na raka ziemniaka zgodnie z Protokołem Diagnostycznym EPPO PM 7/28. Biul. IHAR 254: 169-177. [Changes in Polish scale of assessment to potato wart disease of breeding lines and cultivars of potato – unification of assessment according to Diagnostic Protocol of EPPO PM 7/28.]. **MNiSW$_{2009}$** = 4 pkt.

H2. **Przetakiewicz J.** 2013. Effects of fungicide treatments of potato sprouts on resistance assessment to *Synchytrium endobioticum* (Schilb.) Perc. using the Glynne-Lemmerzahl method. Bull. OEPP/EPPO Bull. 43(2): 280-284. **MNiSW$_{2008}$** = 5 pkt.

H3. **Przetakiewicz J.** 2015. The Viability of Winter Sporangia of *Synchytrium endobioticum* (Schilb.) Perc. from Poland. Am. J. Pot. Res. 92(6): 704-708.

IF$_{2015}$ = 1,159; **MNiSW$_{2015}$** = 25 pkt.

H4. **Przetakiewicz J.** 2016. A modification of the Potoček's tube test for diagnostic of *Synchytrium endobioticum* (Schilb.) Perc. a causal agent of potato wart disease. Indian Phytopath. 69 (4s): 260-265. **MNiSW$_{2016}$** = 5 pkt.

H5. **Przetakiewicz J.** 2017. Sampling, maintenance and pathotype identification of *Synchytrium endobioticum* (Schilb.) Perc. Plant Breeding and Seed Science 76:29-36. **MNiSW$_{2017}$** = 11 pkt.

H6. Plich J., **Przetakiewicz J.**, Śliwka J., Flis B., Wasilewicz-Flis I., Zimnoch-Guzowska E. 2018. Novel gene *Sen2* conferring broad-spectrum resistance to *Synchytrium endobioticum* mapped to potato chromosome XI. Theor Appl Genet, 131(11): 2321-2331. **IF$_{2017}$** = 3,93; **MNiSW$_{2018}$** = 40 pkt. *Own contribution 30%*

H7. **Przetakiewicz J.** 2014a. First report of *Synchytrium endobioticum* (potato wart disease) pathotype 18(T1) in Poland. Plant Disease 98(5): 688. **IF$_{2015}$** = 3,20

H8. **Przetakiewicz J.** 2015a. First report of new pathotype 39(P1) of *Synchytrium endobioticum* causing potato wart disease in Poland. Plant Disease 99(2): 285.2. **IF$_{2015}$** = 3,192

H9. **Przetakiewicz J. 2010.** Odporność polskich odmian ziemniaka na występujące w kraju wirulentne patotypy 2(Ch1) i 3(M1) grzyba *Synchytrium endobioticum* (Schilb.) Per. Biul. IHAR 257/258: 207-214. [Resistance of Polish cultivars of potato to virulent pathotypes of *Synchytrium endobioticum* (Schilb.) Per.: 2(Ch)1 and 3(M1).**]** **MNiSW$_{2010}$** = 4 pkt.

The total Impact Factor of the publications included in the scientific achievement according to the Journal Citation Reports (JCR) list, according to the year of publication for publications H3, H6, H7 and H8 is **11,481**. Publications H1, H2, H4, H5 and H9 are not on the JCR list and therefore has no IF value. The sum of points for publications included in the scientific achievement, according to the list of scientific journals according to the year of publication for publications H1, H2, H3, H4, H5 and H6 is **94** points.

The applicant's contributions to the works above are described in **Appendix 5**. The co-authors' statements describing their contributions to each publication are included in **Appendix 6**. Copies of publications being scientific achievements are attached in **Appendix 4**.

C) REVIEW OF THE SCIENTIFIC PURPOSE AND RESULTS PRESENTED IN THE PAPERS INCLUDED IN THE SCIENTIFIC ACHIEVEMENT, AND DISCUSSION OF THEIR POSSIBLE APPLICATION.

INTRODUCTION

Potato is one of the most important crops in the world and the fourth largest crop species after corn, wheat and rice. In the European Union (EU), potatoes are grown on an area of about 2 million ha. The main growing areas are in Poland, Germany, Romania, France, the Netherlands and the United Kingdom (Flath et al., 2014). In the world, especially in Asia and Africa, its importance is constantly growing, where the area of cultivation has doubled in the last decade, with China and India taking the first two places among the largest producers (FAO, 2017).

The fungus originates from the Andean zones of South America, from where it spread first to Europe in the late of the nineteenth century. Presently, the geographical distribution of this pathogen includes almost all European and Mediterranean Plant Protection Organization (EPPO) countries, Asia, North and South America as well as Oceania (New Zealand). The species was first discovered by Schilberszky in Hungary. In fact, the pathogen was known firstly in Europe. In 1876, potato wart disease was found for the first time in the UK (Hampson 1993). The fungus is a member of genus *Synchytrium* included about 200 species. All of them are parasites but the most important economically and phytosanitary is *S. endobioticum* (Karling, 1964). The main host of *S. endobioticum* is potato, but the fungus is able to infect other species of the genus *Solanum* , including those that occur natively in Poland like *S. nigrum* or *S. dulcamara* (Malec, 1983). In favorable conditions, the pathogen is capable of infecting tubers and their eyes, stolons, shoots, leaves and even potato flowers, but it never develops on the roots (Hampson and Coombes, 1989; Przetakiewicz, 2014b). In the case of tomato, roots are also infected (USDA, 2007). It is one of the most dangerous diseases of potato, and susceptible varieties can be so strongly affected that it can completely destroy or significantly reduce the yield. The fungus can survive without a host for more than 40 years as resting (winter) sporangia (**H3**). Its long persistence in soil and the severe losses it inflicts to potato crops have prompted its inclusion into the A2 quarantine list of EPPO. The EU issued a specific requirement in the Council Directive 69/29/EC of 8 December 1969 on control of Potato wart disease and the Council Directive 2000/29/EC of 8 May 2000 on protective measures against

the introduction into the Community of organisms harmful to plant or plant products and against their spread within the Community (Obidiegwu et al. 2014). As well as Polish regulations and the Polish Minister of Agriculture and Rural Development dated 5 August 2004. on detailed procedures for combating and preventing the spread of the fungus *Synchytrium endobioticum* (Przetakiewicz, 2014b). In USA, the *S. endobioticum* could be taken into consideration as biological weapon as a threat for potato. *S. endobioticum* is included on the list of particularly dangerous plant pathogens listed in the ABPA Act and in the HSDP-9 Directive in the USA (USDA-APHIS 2002). There were about 40 different pathotypes of the fungus in Europe (Baayen et al., 2006). New pathotypes are still detected as 38(N1) in Turkey (Çakir et al., 2009), or 39(P1) in Poland (**H8**). The most widespread pathotype 1(D1) in the world, occurred in Poland in the 1950s and was the serious problem in all country. However, cultivating resistant cultivars and compliance with phytosanitary regulations meant that the pathotype was now less important (Przetakiewicz, 2008a). Currently, it is assumed that the most relevant pathotypes in Europe are: 1 (D1), 2 (G1), 6 (O1), 8 (F1) and 18 (T1) (Ballvora et al., 2011). In Poland in the fifties, two virulent pathotypes 2 (Ch1) and 3 (M1) were detected (Malec, 1981). They are probably only found in the country, but for phytosanitary reasons they are also very important (Przetakiewicz, 2014b). Although the species usually prefers moderate climate, new outbreaks have been observed in countries with a warmer continental climate, such as Turkey, Georgia, Bulgaria or Greece (Çakir, 2005; Dimitrova et al., 2011; Gorgiladze et al., 2014; I. Vloutoglou pers. comm., 2015). This may be evidence of the adaptation of the pathogen to climate change observed in Europe. Another disturbing fact is the spread of virulent pathotypes to countries that were already free of the fungus like Denmark or Sweden. Pathotype identify confirmed that these are virulent pathotypes (Busse et al., 2017; van de Vossenberg et al., 2018b). The pathotype 18 (T1) was also introduced to Poland (**H7**), which so far has occurred in Germany and the Netherlands.

Population characteristics of *S. endobioticum* in Poland, by assessing their virulence and correct pathotype identification, are of great importance for controlling the spread of the pathogen as well as preventing the selection of new more virulent pathotypes. According to EPPO standard PM 7/28 (EPPO, 2004), which was in force until the end of 2017, the Polish pathotype 3(M1) was identical to German 6 (O1) one. In contrast, another pathotype 2 (Ch1) was similar to the pathotype 18 (T1). The use of a few differential cultivars leads to incorrect pathotype classification. This may lead to the cultivation of susceptible or weakly susceptible varieties in safety zones and infected zones (if the number of living spores drops below 5 in 1g

of soil). Consequently, this may lead to the selection of new pathotypes with increased virulence (Malec, 1964 and1974; **H8**; van de Vossenberg et al., 2018b).

The publications included in my scientific achievement concern issues related to the pathogenic, quarantine potato pathogen, *Synchytrium endobioticum*, and include:

- modification of the detection method of *S. endobioticum* from soil with a low density of winter sporangia (**H4**),
- study on the longevity of winter sporangia of *Synchytrium endobioticum* in soil collected from an infested plot 43 years since the last observed infection (**H3**),
- developing a method for collecting, maintaining and identifying *S. endobioticum* pathotype (**H5**),
- identification of new, virulent *S. endobioticum* pathotype in Poland (**H7**, **H8**)
- change in the Polish scale of assessment resistance of potato cultivars to *S. endobioticum* (**H1**),
- study on the effect of fungicide treatments on the results of potato resistance assessment on *S. endobioticum* (**H2**),
- assessment of the resistance of 69 Polish potato varieties to two virulent pathotypes of *S. endobioticum* 2(Ch1) and 3(M1) (**H9**),
- mapping the *Sen2* resistance gene with a broad spectrum of resistance to eight virulent *S. endobioticum* pathotypes present in Europe, pyramiding of two resistance genes, *Sen1* and *Sen2* in potato breeding materials (H6).

S. endobioticum is a soil-born pathogen, an obligatory biotrophe, not producing hyphae. It produces thick-walled winter sporangia that can survive for a long time without the host plant (**H3**). To release zoospores, the presence of water is necessary. As a result of infection of young sprouts of susceptible to *S. endobioticum*, potato wart disease develops. In the galls thin-walled summer sporangia which produce haploid zoospores that are capable of infecting host cells (tubers, stolons). Haploid zoospores form isogamic zygotes, which are capable of infecting host cells and then forming winter sporangia.

The aim of the study was to characterize the Polish population of *S. endobioticum*. Characterization consisted in assessing the viability of winter sporangia from the oldest documented outbreaks, where in the fifties for the first time virulent pathotypes of *S.*

endobioticum were detected in Poland. The aim was also to check whether the living spores are capable of germination and infesting of potato and if so whether there has been a change in the virulence profile after such a long time. Another goal was to check if there is population variability of *S. endobioticum* in these plots in Poland, where the fungus is still present and the symptoms of potato wart disease are still appeared. One of the most important goals of the research was to check whether the fungus spread to areas of economic importance, where there are commercial potato production. This goal is also connected with the question whether after the accession of Poland to the European Union (EU) the risk of introducing European pathotypes of the fungus that have not been present in our country so far increases. The only strategies to confine the disease are strict quarantine and phytosanitary measures as well as the cultivation of resistant cultivars. The aim of the research was to search for Polish potato cultivars resistant to various pathotypes of *S. endobioticum,* as well as the search for R genes of extremely resistance to European pathotypes of *S. endobioticum*.

My scientific work related to the fungus *S. endobioticum* coincided with Poland's accretion to the EU. According to the Accession Treaty, Poland retained the right to authorize [the assessment of the the resistance of potato varieties to pathotype 1 (D1)] resistance of all foreign potato varieties for 10 years. For this purpose, I published a detailed methodology for testing the resistance of potato genotypes to *S. endobioticum* (**H1**).

In the case of assessing the resistance of potato varieties to different pathotypes of S. endobioticum, there was no common methodology in the EU at that time. The same was with pathotype identification. The EU Council Directive 69/464 (EU, 1969) only indicates that resistant varieties are those that respond to infection with a pathogenic agent in such a way that there is no risk of secondary infection. The directive also does not specify which method should be used to determine the absence of secondary infections. Significantly greater refinements are included in the Standard of the European and Mediterranean Plant Protection Organization (EPPO) PM 7/28 (OEPP / EPPO, 2004), where biotests are recommended to assess the resistance and identification of *S. endobioticum* pathotypes: Spieckermann (Spieckermann and Kothoff, 1924) and the Glynne-Lemmerzahl (Glynne, 1925, Lemmerzahl, 1930, Noble and Glynne, 1970) and field tests. Whereas in Standard EPPO PM 3/59 (EPPO, 1999) pot tests or Potock's tube test are also recommended (Potocek, 1977).

As a consequence, in almost every country one of the mentioned methods is used and even methods are used that do not appear in the EPPO Standards. An example of this is the method used in the Scottish Agricultural Science Agency, where zoospores are used to inoculate potato spouts from germinated winter sporangia (Browning, 1995). The Spieckermann method is used in the Netherlands (Spieckermann and Kothoff, 1924), Potock tests (Potocek, 1977) were applied in the Czech Republic, the Glynne-Lemmerzahl method is used in Germany and Poland (Flath et al., 2014; **H1**; Przetakiewicz and Plich, 2017). The German and the Polish version of Glynne-Lemmerzahl method were also differed in some stages of its implementation, which could give different final results under certain conditions.

In 2006, at the request of the European Seed Association (ESA), in Luneburg was the first meeting of experts involved in testing the resistance of potato to *S. endobioticum*. The prepared report stated that only the Glynne-Lemmerzahl method should be used to resistance tests and identify *S. endobioticum* pathotype (Przetakiewicz, 2010a). My research consisting in comparing the most important methods recommended by EPPO, the Glynne-Lemmerzahl and Spieckermann methods, clearly indicated that the use of the Glynne-Lemmerzahl method allows for precise laboratory assessment of the tested potato genotypes and, as a consequence, elimination of susceptible one. However, this method requires much more work and higher costs. Although the use of the Spieckermann method is cheaper and simpler but low infection pressure during inoculation creates the risk of elimination not of all susceptible potato genotypes (Przetakiewicz and Kopera, 2007) and did not allow to distinguish resistant genotypes from slightly susceptible ones.(Przetakiewicz, 2008b).

About 15% of resistant German varieties, after Polish verification, were assessed as susceptible to pathotype 1 (D1) of *S. endobioticum*. However, among these 15% of varieties almost all were slightly susceptible (Przetakiewicz, 2010). The German protocol provides for the treatment of potato sprouts with a fungicide Pencucuron before inoculation, which could result in partial elimination of the zoospores during inoculation. As a consequence, some sligthly susceptible varieties could be assessed as resistant to *S. endobioticum*. Treatment with fungicide of potato sprouts protects them against rotting during the four-week incubation. For this reason, I conducted experiments comparing the treatment with different fungicides before and after inoculation (**H2**). Obtained results clearly indicated that sligthly susceptible potato varieties may appear to be resistant when sprouts are treated before inoculation (**H2**). Partial elimination of zoospores caused the reduction of infection pressure and consequently was too weak to break

down the resistance barrier of slightly susceptible potato genotypes. Pencycuron treated after inoculation did not affect the final results, because the fungal zoospores had already penetrated into the host cells and had no contact with the active ingredient of the fungicide. **H2**'s published work also indicated that potato germination of copper after inoculation not only did not affect the final assessment of resistance, but also was the best to results of protection against rotting of sprouts during incubation.

The Glynne-Lemmerzahl method used in Germany is very similar to the Polish one. Differences in both methods are also small. The main one results from a different approach to assessing the degree of infection of potato sprouts inoculated with *S. endobioticum* zoospores. The Polish assessment primarily takes into account the host's response to pathogen infection. In the case of no necrosis reaction, 2-3 weeks after inoculation, the genotype is assessed as extremely susceptible. However, weakly susceptible genotypes include those that after 2-3 weeks of incubation react to the so-called very late necrotic reactions. At the same time, there are no visible necrosis of the host in other parts of the same shoot or on neighboring shoots subjected to inoculation of the same tuber and epidermal proliferation (**H1**) is noticeable. According to the German methodology, the extremely susceptible genotypes include those that react with strong proliferation, resulting in large wart growth after about 4 weeks of incubation. The slightly susceptible genotypes include those with poor proliferation with visible necrosis, however, the presence in the tissues of the host the resting spores is necessary. In the case of absence the spores, the genotype is classified as weakly resistant.

According to the agreement between the IHAR-PIB and the Inspectorate of Plant Health and Seed Inspection (PIORiN)), all soil samples (containing viable resting sporangia) or potato plants with warted tubers are identified in IHAR-PIB in Radzików. During the implementation of the agreement, I encountered a problem that made it impossible to identify *S. endobioticum* pathotype. Soil samples, especially from newly discovered or from very old outbreaks, contained single viable resting sporangia, which precluded, according to EPPO Standard PM 2/28 and MP 3/59 (2004 and 2005), obtaining fresh warts. Fresh warts are necessary for the Glynne-Lemmerzahl test assess the virulence of *S. endobioticum* isolates on differential cultivars. In my scientific achievement, I developed a special ring method (**H4**), which allowed to obtain fresh warts from single viable spores of the fungus (about 2-3 sporangia per 1 kg of soil). The threshold of sensitivity was higher than PCR based technique (van den Boogert et al., 2015), which was about 10 spores of sporulation in 100 g of soil. The superiority of the method

was based on the fact that the galls can be used to identify *S. endobioticum* pathotype. In the case of the PCR method, DNA product only indicates that the sample contained fungal DNA. My scientific achievement (**H4**) was used to study the oldest outbreak, where in 1965 the second virulent pathotype 3 (M1) of *S. endobioticum* was detected (Malec, 1981). Taken 42 years after the first detection, the soil samples still contained viable winter sporangia of the fungus, of which the next year two galls were separately obtained (**H3**). A positive result of the bioassays was considered as sufficient proof for the ability of spores to germinate and to invade and replicate in the host. Pathotype identification required the use of old potato varieties, which were used for pathotyping by Malec (1981). According to Malec (1981), Polish pathotypes differed in virulence profile from pathotypes found mainly in the former East and West Germany. Finally, I have completed and maintained a set of 23 variations (**H5**). They are used not only to identify and distinguish Polish isolates of *S. endobioticum*, but also to distinguish them from the most important pathotypes present in the EU. The virulence studies of the obtained isolates from Mieroszów confirmed that they are different pathotypes: 1(D1) and 3(M1). According to Malec (1963 and1974), it is possible to increase the virulence of pathotype 1(D1) by passaged on a resistant variety, but characterized by a low degree of resistance, which has recently been proven (van de Vossenberg et al., 2018). From these data, it can be concluded that a single outbreak may contain two different pathotypes, as in the case of Mieroszów, where a virulent pathotype 3(M1) could occur, which despite 43 years retained an identical virulence profile (**H3**). The results clearly indicate that in conditions of the Polish climate, especially in the upland areas, resting sporangia of *S. endobioticum* can survive several times longer compared to Western Europe. Relatively warmer winters in Western Europe promote the germination of the spores, leading in a shorter time to clearing the infested zone. The severe winter of the Polish climate effectively blocks germination of the spores for several months a year, without destroying them by the way. This means that even after 43 years the fungal spores are able to germinate and infect (**H3**). Consequently these results should be taken into consideration when de-scheduling previously infested plots even after 40 years or longer, especially in the mountainous areas of Poland.

Publications included in the scientific achievement (**H1**, **H2**, **H4** and **H5**) allowed to characterize over 50 Polish isolates of *S. endobioticum* (Przetakiewicz, 2014c). All of them except one were virulent. In addition, further pathotypes were detected which were not previously occured. In the Mazowieckie voivodship, a few viable winter spores were detected,

of which due to scientific achievement (**H4** and **H5**), pathotype 18(T1) was identified **(H7**). It was the first detection of the Western European pathotype, after Poland's accession to the EU. These spores were introduced into infected soil, with ornamental plants that are not the host of *S. endobioticum*. This is an example of alternative ways for *S. endobioticum* spreading without potato as a main host. (**H7**; Przetakiewicz, 2014b).

Pathotype 2(Ch1) is the most widespread in Poland. It is detected both in the form of winter spores and warted tubers. In addition to numerous outbreaks in the Małopolska region (detected as the spores or infected plants) and Silesia (only winter sporangia), this pathotype was also identified in the province. Kujawsko-pomorskie region (only winter sporangia). In the Lower Vistula Valley of this province, a numerous plots with viable spores have been detected in the floodplains of the river. In most of these cases, the presence of a few viable resting spores of *S. endobioticum* was found. They may come, among other things, from the province Małopolska, which almost entirely belongs to the Upper Vistula basin. This may indicate that sporangia can spread along with the current of the river from the original outbreaks, as a result of, for example, local floods (Przetakiewicz, 2014c). Patotype 2(Ch1) is not only the most widespread in Poland but also the most diverse in terms of virulence. The assessment of the obtained isolates based on the scientific achievement of **H5** showed that pathotype 2(Ch1) is a population of at least three different races. All races of pathotype 2(Ch1) have a common feature because Asche Sämling variety reacts in the same way (identical to pathotype 2(Ch1)). This variety was used to distinguish the pathotype 2(Ch1) from 3(M1) (Malec, 1981). Among population 2(Ch1), a new pathotype 39(P1) was distinguished (**H8**). Its virulence profile differed from other pathotypes. Most of the different isolates were detected in small areas, often in home gardens, in areas where pathotype 2(Ch1) was identified for the first time (Malec, 1981). Long-term cultivation of the same potato varieties with low resistance to pathotype 2(Ch1) could lead to the selection of new populations of pathogens with a different virulence than pathotype 2(Ch1). The exact determination of their virulence is necessary to search sources of resistance in the potato. It is impossible to allow cultivations of varieties that are characterized by a low level of resistance. This situation may lead to further selection and creation of further *S. endobioticum* pathotypes, which often are more virulent.

Despite the passage of several decades, pathotype 2(Ch1) and 3(M1) still occurs in Poland (Przetakiewicz, 2014c). In my scientific achievement **H9** I was searching potato varieties resistant to these pathotypes. Out of several dozens of Polish varieties, only a few were wakly

resistant and only four resistant or extremely resistant (variant Icarus on pathotype 3 (M1)) (**H9**). However, the development of methods for assessing the virulence of *S. endobioticum* pathotypes (**H5**) and the development of an effective method for assessing potato resistance to this pathogen (**H1** and **H2**) enabled searching for new sources of resistance to the most important pathotypes occurring in Europe and Poland in the *Solanum* diploid hybrids. These clones, selected in IHAR-PIB Młochów, are interspecific hybrids, possessed in their pedigree *Solanum acaule, S. chacoense, S. demissum, S. goniocalyx, S. gourlayi, S. microdontum, S. phureja, S. stoloniferum, S. stenotomum, S. tuberosum, S. verrucosum,* and *S. yungasense*. Of the several dozen clones, seven were resistant to all virulent pathotypes of *S. endobioticum*: 1(D1), 2(G1), 2(Ch1), 3(M1), 6(O1), 8(F1), 18(T1) and 39(P1). (Jakuczun et al., 2013). In my scientific achievement **H6**, to characterize and map a new gene for resistance to potato wart, there was used diploid F1 potato population from a cross of potato clone resistant to *S. endobioticum* pathotype 1(D1) and virulent pathotypes: 2(G1), 6(O1), 8(F1), 18(T1), 2(Ch1), 3(M1) and 39(P1) with a potato clone resistant to pathotype 1(D1) only. The studies used a representative set of DArTseq markers, selected from more than 3200 mapped so far in the Solanum species (Śliwka et al. 2012a, Śliwka et al. 2012b,). Resistance to pathotype 1(D1) was additionally conferred by the *Sen1* gene inherited from both parents. *Sen2* was mapped to chromosome XI using DArTseq markers. The genetic and physical distances between *Sen1* and *Sen2* loci were indirectly estimated at 69.1 cM and 32 Mbp, respectively. The new potato cultivars with a broad-spectrum resistance against multiple pathotypes of *S. endobioticum*, will help also to restrict further spreading of virulent pathotypes of the pathogen.

SUMMARY

The six publications constituting my post-doctoral dissertation include the results of research on the detection, identification and resistance to *S. endobioticum*. The results of the research are:

- development of a set of differential cultivars to identify of pathotypes and determine the virulence profile of *S. endobioticum* isolates,
- detection for the first time in Poland of two virulent pathotypes of *S. endobioticum*, 18(T1) and 39(P1),
- discovery a single resistant gene *Sen2* which underlies the extremely resistance to all detected virulent *S. endobioticum* pathotypes, including new virulent pathotypes 18(T1) and 39(P1) detected in Poland,

- pyramiding of two resistance genes, *Sen1* and *Sen2* in potato breeding materials,
- development of a PCR marker for use in the selection of potato breeding materials,
- development of an effective method for assessing potato resistance to various *S. endobioticum* pathotypes,
- change in the Polish scale of potato variety resistance assessment to potato wart dsiease,
- assessment of the resistance of 69 Polish potato cultivars to virulent pathotypes 2(Ch1) and 3(M1) of *S. endobioticum*,
- development of a sensitive detection method of *S. endobioticum* from soil with a low density of winter sporangia,
- demonstrating the ability of *S. endobioticum* to survive in soil in which no potato was cultivated for 43 years, and to preserve the ability of winter sporangia to infect potato after such a long time,

The results of the research presented in this work have cognitive and practical significance. The developed methods may be useful for the preparation of a new EPPO Standard on potato resistance to *S. endobioticum*.

Literature

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5. DISCUSSION OF THE REMAINING SCIENTIFIC ACHIEVEMENTS.

In the years 1997-1998, I participated as an MA student in immunochemical identification of amino acids phosphorylated in phytochrome protein of blanched coleoptiles (*Avena sativa* L.) in the Institute of general and molecular biology, in the Holocaust of Biochemistry, Faculty of Biology and Earth Sciences, UMK in Torun. The supervisor of my master's thesis was Prof. Dr hab. Stanisław Kowalczyk. The aim of the study was to compare the immunological methods and the identification of antigenic proteins in the raw oat germ extract and a test of monoclonal antibodies against phosphorylated to amino acids for the study of changes in phosphorylated phytochrome A (Phy A). As part of my work by immunprecipitation, using lyophilized bacterial walls containing Protein A or Protein A, I received a protein of 120 kDa. This protein showed an immune response with polyclonal rabbit antibodies directed against PhyA. Exposure of PhyA extracts with red light (λ650 nm) produced reactions with monoclonal antibodies directed against phosphorylated serine and tyrosine, which was not observed in the absence of irradiation. I described the results in my master's thesis, which I defended in July 1998.

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In the period from October 5, 1998 to October 21, 2004, I worked as an engineer, assistant and then as an assistant professor at the Plant Transformation and Cell Engineering Department, Plant Breeding and Acclimatization Institute - National Research Institute in Radzików. The head of the Laboratory was doc. dr hab. Wacław Orczyk. My scientific work consisted in developing a method of isolation, culture and regeneration of diploid potato protoplasts (Solanum tuberosum L.). The donor material was sixteen dipoid potato clones obtained from the IHAR Młochów Research Center. The plant material, for the isolation of protoplasts, was obtained from plants *in vitro*. Plants obtained on medium with norflurazone were completely free of chlorophyll. Green and chlorine-free protoplasts were used to perform protoplast fusion. Thanks to such experiments, hybrids were distinguished from other protoplasts, which could be caught using a micromanipulator. Shoot regeneration from captured hibrids was induced in 10 different combinations and as a result more than 300 plants were obtained.

Molecular analyzes using RAPD and semi-random markers confirmed plant hybridity in 6 different combinations. DNA analysis using RFLP and mtDNA specific probes allowed to identify three different types of mitochondria in the tested lines: type α, β and ε. These analyzes also showed that all tested hybrids contain only the β type mitochondria. Using probes prepared on the basis of rape cpDNA, the chloroplast DNA of the starting lines and hybrids obtained from them was differentiated. Based on the results obtained, the lines were divided into five groups. Analyzes carried out on fusion components (with different chloroplasts) and their hybrids showed a random segregation of these organelles. The hybrids DW 84-1920 (+) DG 82-199, DW 84-1920 (+) DG 88-596 and DW 84-1920 (+) DG 82-258 were carried out on resistance to potato leaf-rolling virus (PLRV), where the components of the fusion were resistant clones: DG 82-199, DW 84-1920 and susceptible clones: DG 88-596, DG 82-258. The results of the DAS-ELISA test showed that most of the tested hybrids were resistant to PLRV.

The assessment of resistance to potato M virus (PVM) determined by Gm gene was performed in hybrids: DW 84-1920 (+) DG 88-596 and DW 84-1920 (+) DG 82-258, where the first component of DW 84-1920 was resistant the other (DG 88-596 or DG 82-258) - susceptible. ELISA results showed susceptibility to PVM of all hybrids. The test of resistance of tubers to *Erwinia carotovora* subsp. *atroseptica*, carried out in hybrids DW 84-1920 (+) DG 82-199, where the components of the fusion were: DG 84-1920 - susceptible and DG 82-199 - medium-resistant. The bacterial infection of hybrids was indirect in relation to the paralysis of both fusion components. The work ended with the defense of the doctoral thesis: "Tetraploid somatic hybrids of potato (*Solanum tuberosum* l.) obtained from diploid breeding lines." -Promotor: doc. dr hab. Wacław Orczyk

PUBLICATIONS:

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PROJECT:

1. 3 PO6A 041 24 pt.: „Fuzja protoplastów z diploidalnych linii ziemniaka (Solanum tuberosum L.) w celu otrzymania roślin tetraploidalnych łączących cechy komponentów rodzicielskich." Finansowany przez KBN.

From October 21. 2004, I work as professor assistant at the Plant Pathology Department at the IHAR-PIB in Laboratory of Quarantine Organisms. My entire scientific activity is related to *S. endobioticum*. As part of the statutory activities, I carried out the topic titled " The improvement of detection and test methods of potato wart resistance caused by Synchytrium endobioticum in genotypes of potato." As part of this topic, I developed a detailed methodology for assessing potato genotypes for various fungal patotypes. I also compared various methods that are used in the EU. In the years 2008-2013, I carried out task 4 in the project: "Developing methods for distinguishing potato forms combining different uses with resistance to important

potato pathogens." - Biological Progress in Plant Production (PBwPR), financed by the Ministry of Agriculture and Rural Development. The project's head was prof. dr. bab. Ewa Zimnoch-Guzowska. In my task titled: Improvement of methods for identification of potato genetic pool including resistance to various pathotypes of *Synchytrium endobioticum*." I developed a set of potato varieties identifying and differentiating the pathotypes of *S. endobioticum*. I checked the potato inheritance in potato pathotype (D1) inheritance by analyzing the immunity of selected parental forms and their progeny clones. I also distinguished the potato genotype pool from varieties, 2x and 4x clones that resist comprehensively on the most important, virulent pathotypes of *S. endobioticum*.

Discrepancies in the results of assessing the resistance of potato varieties to potato wart disease in connection with the use of other methods have been tried in cooperation with partners from other EU teams. I took an active part in organizing and conducting experiments with other research Institutes from the Netherlands: Plant Protection Service, National Reference Laboratory (Wageningen),AGRICO Cooperatie (Emmeloord), AVERIS Seeds B.V. (Valthermond), HLB, Hilbrands Laboratory (Wijster); Germany: JKI, Julius-Kühn Institut, Federal Research Center for Cultivated Plants (Kleinmachnow) and IHAR-PIB. Reaction of differential cultivars (cultivars used to identify *S. endobioticum* pathotypes) on pathotypes; 1(D1), 2(G1), 6(O1) and 18(T1) were consistent in all partners only for the pathotype 1(D1) and partly for the pathotype 18(T1) (Flath et al., 2014). Harmonization of resistance assessment and identification of *S. endobioticum* pathotypes has become an important element of my cooperation with many scientific Institutes in the EU as well as the so-called third countries:

1. Dutch National Plant Protection Organization, National Reference Centre, Geertjesweg 15, 6700HC, Wageningen, the Netherlands.
2. Fera Science Ltd., Sand Hutton, YO41 1LZ York, United Kingdom.
3. All Russian Research Institute for Plant Protection, Podbelsky sh. 3, Pushkin, Saint Petersburg, Russia.
4. The State Planting Service, Ministry of Agriculture, Sukileliu str. 9, LT - 11352, Vilnius, Lithuania.

5. Central laboratory for plant quarantine, 120, N. Moushanov Blvd., 1330 Sofia, Bulgaria.
6. Hilbrands Laboratorium BV, Kampsweg 27, 9418 PD Wijster, the Netherlands
7. Wageningen UR, PRI, Droevendaalsesteeg 1, 6708 PB Wageningen, the Netherlands.
8. Department of Agriculture, Food and the Marine, Backweston Campus, Celbridge, Co. Kildare, Ireland.
9. NAK, Randweg 14, 8304 AS, Emmeloord, the Netherlands.
10. Julius Kühn-Institut, Stahnsdorfer Damm 81, 14532 Kleinmachnow, Germany.
11. Institute for Agricultural and Fisheries Research, Plant Unit, Burg. Van Gansberghelaan 96, 9820 Merelbeke, Belgium, kurt.heungens@ilvo.vlaanderen.be
12. Institute of Plant Protection, 33 Vasylkivska Str., 3022, Kiev, Ukraine.
13. Science and Advice for Scottish Agriculture, 1 Roddinglaw Road, EH12 9FJ Edinburgh, United Kingdom.

All-Russian Plant Quarantine Center, Pogranichnaya 32, Bykovo 140150, Ramenskoe region, Moscow oblast, Russia.

As part of this cooperation, I carried out two international projects: EUPHRESCO 2, the acronym SENDO ("Diagnostic methods for *Synchytrium endobioticum*, especially for pathotype identification.") and CORNET, the acronym SynTest ("Establishment of a harmonised methodology for testing the resistance of potato cultivars to potato disease (*Synchytrium endobioticum*) in the EU. ").

The research work in these projects also included the use of molecular techniques for the detection of *S. endobioticum* and the identification of some pathotypes (Vossenberg et al., 2018) and the use of molecular markers for the selection of potato genotypes for pathotype 1(D1) (Przetakiewicz and Plich, 2017).

In the years 2008 - 2013 I was head of the project: "Monitoring and the occurrence of new, aggressive pathotypes of *Synchytrium endobioticum*, including the detection of possible new virulence factors in populations occurring in Poland." The topic was implemented under the Multiannual Program: "Plant Improvement" for Sustainable AgroEcoSystems, High Quality Food and Plant Production for Non-Food Objects. ", financed by the Ministry of Agriculture and Rural Development. W the years 2015 – 2020 I am carrying out the task: “Monitoring in the virulence changes of pathotypes

in the population of *Synchytrium endobioticum* (Schilb.) Perc. and populations of nematodes (*Globodera rostochiensis, Globodera pallida*) in Poland." within the framework of Multiannual Program under the common title: "Creating the scientific basis of biological progress and protecting plant genetic resources as a source of innovation in supporting sustainable agriculture and food security of the country."

As part of the current statutory activity (2018 - 2019), I perform HRM (high-resolution melting analysis) to differentiate *S. endobioticum* pathotypes from my collection. The analyzes are carried out on amplicons obtained for *S. endobioticum*-specific SNP (Single Nucleotide Polymorphism) markers.

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1. Busse, F., Bartkiewicz A., Terefe-Ayana D., Niepold F., Schleusner Y., Flath K., Sommerfeldt-Impe N., Lübeck J., Strahwald J., Tacke E., Hofferbert H.-R., Linde M., **Przetakiewicz J.**, Debener T. 2017. Genomic and transcriptomic resources for marker development in *Synchytrium endobioticum*, an elusive but severe potato pathogen. Phytopathology. 107: 322-328.
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endobioticum. EAPR 20th Triennial Cnference Potato Facing Global Challenges, Versailles, Francja, oral presentation.

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MONGRAPHS:

1. Przetakiewicz J. 2014. Manual of Security Sensitive Microbes and Toxins. Chapter 73: *Synchytrium endobioticum*. ss.823-829. ed. Dong Liu, ISBN 13: 9781466553965, CRC Press.
2. Przetakiewicz J. 2014. Monitoring występowania nowych, agresywnych patotypów *Synchytrium endobioticum* z uwzględnieniem wykrycia ewentualnego pojawienia się nowych czynników wirulencji w populacjach patogena występujących w Polsce. ss. 263-271. Praca zbiorowa pod red. Prof. dr hab. E. Arseniuka, MONOGRAFIE I ROZPRAWY NAUKOWE IHAR-PIB 48/2014.
3. Gawińska-Urbanowicz H., Michalak K., Przetakiewicz A., Przetakiewicz J., Śliwka J., Sobkowiak S., Węgierek A. i Yin Z. 2014. Gromadzenie, charakterystyka w zakresie biologii oraz przechowywanie ras i patotypów najważniejszych patogenów ziemniaka. ss. 120-126. Praca zbiorowa pod red. Prof. dr hab. E. Arseniuka, MONOGRAFIE I ROZPRAWY NAUKOWE IHAR-PIB 48/2014.

PROJCTS:

1. 2012-2014. Euphresco II, Euphresco Phytosanitary Era-Net. Diagnostic methods for *Synchytrium endobioticum*, especially for pathotype identification (SENDO). Principal Investigator

2. 2013-2015. CORNET/2/13/2012. Harmonizacja w ramach Unii Europejskiej oceny odporności odmian ziemniaka na *Synchytrium endobioticum*, sprawcy raka ziemniaka (SynTest). Projekt CORNET 13, NCBiR. Principal Investigator.

3. 2015-2017- UMO 2013/11/NZ9/01959. Poznanie genetycznych podstaw odporności ziemniaka na różne patotypy *Synchytrium endobioticum* sprawcy raka ziemniaka Projekt OPUS6, NCN. Investigator

6. SUMMARY OF SCIENTIFIC AND RESEARCH ACHIEVEMENTS

My postdoctoral output includes twenty-four publications, including nine scientific achievements. Among them, tenty-one are original scientific papers, including eight published in journals with the IF impact factor, highlighted in the Journal Citation Reports (JCR). I am also the author and co-author of three chapters of the monograph, one of which is indexed on Web of Science. I also disseminated the results of my doctoral dissertations in the form of sixteen reports presented at twenty-two international (11) and national (5) conferences, 9 of which were presentations or co-authored by me.

After PhD I participated as an expert in the qualification of quarantine organisms for ESA or regarding only S. endobioticum for EPPO. I took an active part in the preparation of the New EPPO Standard 2/28 (2) on the identification of pathotypes, where my scientific achievement (**H3**) was introduced as one of the methods of obtaining fresh cancerous growths from spores of S. endobioticum.

At the request of PIORiN, I participated as an expert on many EPPO panels on the harmonization of resistance assessment for S. endobioticum and the identification of fungal patches.

In accordance with the Accession Treaty of Poland's accession to the EU, for 10 years from this entry I was responsible for the authorization of pathotype 1 (D1) resistance to all foreign potato varieties to be cultivated in Poland. All publications from my scientific achievement have allowed me to implement this knowledge into practice, i.e. perform other scientific activities. As part of this activity, I cooperate with over thirty seed and breeding companies from Poland and abroad (Ireland, Belgium, Denmark, the Netherlands, Germany, Hungary, Bulgaria, Greece, Sweden, Lithuania, Latvia and the Czech Republic). During this time, I evaluated over 200 potato varieties in terms of resistance to various S. endobioticum pathotypes and issued 241 certificates of resistance for the Central Research Center for Cultivar Testing (COBORU) and other foreign companies. I also performed pathotype identification for isolates detected in other EU countries: Bulgaria (Laboratory for Potatoes, Central Laboratory for Plant Quarantine POB 55Samokov 2000 Samokov), Greece (Laboratory of Mycology, Department of Plant Pathology, Benaki Phytopathological Institute, 8, St. Delta Street , 145 61 Kifissia (Athens)), Denmark (Ministry of Food, Agriculture and Fisheries, Danish Veterinary and Food Administration, Plant Diagnostic Section).

Due to my employment in a research institute, I do not have much achievement in didactic work. Nevertheless, I had the opportunity to repeatedly present my research in the laboratory to numerous student and student trips.

The data in numbers for the achievements mentioned above are summarized below in Tables 1.

Table 1. Numerical list of achievements before and after obtaining the doctoral degree.

Title of the scientific achievements	**Punkty za publikacje wchodzące w skład osiągnięcia**		
	IF	**Points by MNiSzW**	**Hirsch Index**
Characterization of the fungus population Synchytrium endobioticum (Schilb.) Perc. occurring in Poland and assessment of potato resistance to its virulent pathotypes.	**11,481**	**94**	**5**

SUMMARY OF THE SCIENTIFIC ACHIEVEMENTS

Wyszczególnienie	**Number**		
	Before doctorate	**After the doctorate**	**Total**
Original cientific publication in journals (including publications that create an achievement with the IF impact factor)	**1**	**23**	**24**
Monographs	**0**	**3**	**3**
Review and popular-scientific-dissemination papers	**1**	**2**	**3**
Points for publications by MNiSzW	**21**	**254**	**275**

The total impact factor (if) of publication, according to JCR	***1,506***	***22,192***	***25,228***
Number of cites , according to WoS		**78**	
Hirsh index		**5**	
Research works and reports from congresses, conferences, including: • reviewed • placed in journal supplements • summaries	 5	 2 3 17	 **2** **3** **22**
Lectures and seminars on conferences: • Invited lector (including international conf.) • international • Polish • at universities and other scientific units • seminars	 1 4	 2 5 1 6 2	 **2** **6** **5** **6** **2**
Membership in editorial committees of magazines: • national, including IHAR Monographs and Dissertations • international			
Membership in scientific committees of Scientific Conferences - international			
Membership in organizational committees Scientific - international conferences			
Chairing sessions at conferences and seminars: • international • national			
EU grants (Polish partner and co-operator)		**1**	**1**
SPUB-M or SPUB - aid program MNiSzW			

National grants MNiSzW (as manager and/or cooperator)	1	2	**3**
Participation in expert and competition teams: Reviewing EU projects Reviewing national research projects **Reviewing the publication:** • **in international journals with IF** • **in national magazines** • **in materials from international conferences**		1	**1**
Promoter: • Master's theses • doctoral thesis completed • doctoral thesis open			

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Applicant signature