Thesis of Doctoral (Ph.D.) Dissertation

Investigation of the plant protection applicability of paraffin oil against grape powdery mildew (*Erysiphe necator*) and its effect on the physiological parameters of grapes (*Vitis vinifera* L.)

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1. BACKGROUND AND OBJECTIVES OF THE DOCTORAL DISSERTATION

The climate change of the 21st century has resulted in generally occurring problems of plant health and as a consequence an increasing use of plant protection products. Due to these factors, the production of the appropriate quantity and quality of crops is a challenge, which also has cost increases, as well as environmental and food toxicological implications (Pautasso et al., 2010, 2012; Özkara et al., 2011).

One of the most important fungal pathogens of grapevines (*Vitis vinifera* L.) is the *Erysiphe necator*, the causal agent of grape powdery mildew (GPM) and grapevines are one of the most intensively sprayed crops worldwide (Rantsiou et al., 2020). The GPM significantly reduces the photosynthetic leaf area, which leads to a decrease in crop yield and quality, as well as in the number of shoots (Nail and Howell, 2004; Moriondo et al., 2005). A wide range of fungicidal agents can be applied to suppress GPM, but the emergence of fungicide resistance causes a worldwide problem (Vielba-Fernández et al., 2020). Consequently, researches are focused on the possibilities of using environmentally friendly alternatives on their own and/or in combination with synthetic fungicides (Yildirim and Dardeniz, 2010; Rantsiou et al., 2020). In addition, precision viticulture and optimized pest management also play an important role, which takes into account the unique disease susceptibility properties of *V. vinifera* varieties (which are typically sensitive to GPM) and the characteristics of the terroir (Doster and Schnathorst, 1985; Gadoury et al., 2003; Gaforio et al., 2011.; Matese and Di Gennaro, 2015).

Among horticultural oils (HCOs), petroleum-derived spray oils (PDSOs) are also widely used to control pests of many plant crops, which are cost-effective and toxicologically safer than synthetic pesticides (Nile et al., 2019). Due to the infection and colonization characteristics of *E. necator* fungus, PDSOs (e.g. paraffin oil, PFO) have shown good efficacy against GPM as 1) a wash-down spray; 2) a spray additive (due to their adjuvant characteristics); 3) alternatives or rotation partner of DMI- and QoI-fungicide treatments (Dell et al., 1998; Grove et al., 2005; Nita et al., 2007; Janousek et al., 2009). PDSOs exhibited both sporulation-inhibiting and preventive effects against GPM in greenhouse when applied either before or after the appearance of *E. necator*-induced tissue lesions (Northover and Schneider, 1996). However, their efficacy against GPM appears to be primarily indirect under field conditions, likely through the stimulation of the plant's defense system (Northover and Schneider, 1996; Northover and Timmer, 2002). PDSOs may have phytotoxic effects, mainly by altering the stomatal behaviour: impair the gas exchange and transpiration, which can lead to reduced yield and delayed sugar accumulation. PDSO residues have not been reported to negatively affect the fermentation process (Dell et al., 1998; Hodgkinson et al., 2002; Finger et al., 2002; Grove et al., 2005; Nazari et al., 2014). The extent of phytotoxicity may depend on the sensitivity of the plant species, tissue type, and developmental stage (Nile et al., 2019). However, with proper timing, concentration, and application rate, phytotoxic effects can be avoided while maintaining efficacy (Wicks et al., 1999; Martín et al., 2005).

Overall, it can be said that PFO (1; 1.5; 2 v/v%) can show good-moderate effectiveness against GPM alone or when used in combination with synthetic fungicides. This experience based on laboratory, greenhouse and/or field surveys which were conducted with several grapevine varieties (e.g. Chardonnay, Kékfrankos, Tempranillo) in the USA (Northover és Schneider, 1996; Dell et al., 1998; Grove et al., 2005; Janousek et al., 2009), in Spain (Martín et al., 2005) and in Australia (Wicks et al., 1999). However, many factors contribute to successful disease control (e.g. regional and climatic characteristics, infection pressure, cultivation method), which justify the investigation of PFO under domestic conditions.

The focus of our research was to reveal the suitability of paraffin oil (PFO) in the domestic disease management of grapevines. Through field trials and laboratory investigations, we aimed to achieve the following objectives:

1) Evaluation of the disease management efficacy of PFO spraying under field conditions, both in the sole application and and in combined use with fungicides, in the context of controlling grapevine powdery mildew (GPM).
2) Analysis of the effects of field-applied PFO treatments on the phyllosphere and berry-surface microbiota of grapevine, through the characterization of *in vitro* cultivable fungal communities.
3) Monitoring the physiological effects of PFO spraying on grapevine under field conditions using leaf gas exchange measurements, along with the examination of stress physiological parameters under controlled laboratory conditions.
4) Investigation of the direct impact of PFO treatment on the viability of *E. necator* using laboratory viability assays, and microscopic detection of the tissue localization of PFO within grapevine leaves.
5) Assessment of the effects of field-applied PFO spraying on grape yield and on the analytical parameters of must and wine.

<u>During our field and laboratory studies, we aimed to test the following hypotheses:</u>

1) PFO alone possesses plant protection efficacy against GPM, although its effectiveness is lower compared to conventional fungicide treatments. To test this hypothesis, small-plot

field spraying experiments were conducted in Eger using two grapevine cultivars differing in susceptibility to GPM. In the first two-year experiment, we assessed the independent effect of PFO against GPM through visual monitoring of disease symptoms.

2) The combination of PFO with fungicide formulations may exert a synergistic effect due to its adjuvant properties, enhancing efficacy against GPM. To test this assumption, during the second two-year spraying cycle we evaluated the combined effect of PFO and fungicides used in vineyard-scale applications on GPM control.
3) PFO treatment may alter the composition of fungal communities on leaf and berry surfaces. To examine this hypothesis, leaf and berry samples were collected from selected plots of the field experiment to characterize in vitro cultivable fungal communities, complementing the visual assessment of powdery mildew symptoms.
4) PFO treatment may induce phytotoxic effects on grapevine, detectable as a reduction in photosynthetic assimilation and transpiration rates. The possible presence and extent of phytotoxicity were evaluated through leaf gas exchange measurements in the plots of the first field experiment.
5) PFO treatment may affect grape yield and the quality of must and wine produced from the first field trial's harvest. Among the quality parameters, the total soluble solids (TSS) content of must − and consequently the alcohol content of the wine − may be influenced, while the plant protection efficacy of the treatments could also affect the phenolic composition of wines. Based on current knowledge regarding the relationship between PFO phytotoxicity and antifungal efficacy, these effects may act in opposition, depending on the plant's physiological status (e.g. photosynthetic activity), as well as yield and quality. We hypothesized that the balance between these two effects under field conditions may depend on the grapevine cultivar's traits (sensitivity to PFO toxicity and GPM infection) and on vintage-specific factors such as fungal infection pressure, assuming optimal PFO dosage.
6) According to literature, PFO may indirectly affect *E. necator* through physiological changes induced in the host plant. To investigate this, the background of PFO's protective efficacy was studied under laboratory conditions by quantifying stress-related metabolites and enzyme activities. As the mode of action of PDSOs against GPM has not yet been thoroughly characterized, the direct effect of PFO on *E. necator* viability was also examined through laboratory viability assays.

2. MATERIAL AND METHOD

Two two-year small-parcel spraying experiments were carried out under field conditions in three replicates on two grapevine varieties with different sensitivity to GPM (Chardonnay, Kékfrankos). The parcels of each treatment were randomly distributed.

The following treatments were used: three PFO (P1: 1,1; P2: 2,2; P3: 3,3 v/v%) concentration; a positive, traditionally (in the vineyard) used fungicide control (CT) and a negative (untreated) control (C0). The applied PFO treatment dosages were determined based on unofficial results of European field spraying trials conducted by the manufacturer: these proved to be safe from the point of view of plant phytotoxicity and the effectiveness of disease control. The first spraying experiment was between 2013 and 2014 with the settings outlined above (~60 vinestock/treatment/replicate). The changes of the second (2015-2016) spraying experiment (~10-14 vinestock/treatment/replicate): the P1 treatment was abandoned and set up combinations with CT: here the PFO spraying was followed by CT. In the case of P3CT, there was also a modification which received any PFO during the flowering and fruit set period (P3CTm). The CT (and the PFO treatment was adapted to this) was treated according to a conventional spraying protocol depending on vintage characteristics, forecasts and detection of infection and pests, as well as the phenological stage of the grapevine. In summary, it was sprayed eight times in 2013, seven times in 2014, seven times in 2015 and six times in 2016 (*Table 1*). The meteorological data were monitored by an automatic agrometeorological station (Boreas Ltd.) located in the vineyard.

In the parcels of the experimental spraying, the intensity of visible GPM symptoms (% GPM coverage of the plant part) on leaves and clusters (Wicks and Hitch, 2002) were assessed and the occurrence of GPM (%) from these data. In 2013-2014, this survey was only carried out (50 data/parcel/plant part/grape variety) before harvest (BBCH 89). In 2015-2016, the monitoring (smaller parcels; 20-20 data/parcel) was also implemented in the developmental stage of pea-sized berry (BBCH 75) and (BBCH 79) veraison (Pálfi et al., 2016a, 2022). The infection survey was supplemented by a culturable mycobiota (filamentous and yeast colony-forming unit (CFU)) test, for which the leaf and berry samples were taken from the parcels of the P2, P3, CT and C0 treatments in 2014 and 2015, in parallel with the assessment of infection severity. The CFU was related to the surface area of the leaf disc (cm^2) and the weight of the berry samples (g).

It was performed with a portable Ciras-1 infrared gas-analyzer (PP Systems, UK) equipped with a round-shaped (2.5 cm^2) Parkinson's leaf cuvette Pálfi et al., 2022a).

Table 1. Fungicides and their applied dosages in CT (fungicide control) treatments (2013-2016)

	2013		2014		2015		2016	
	Name of pesticide	***Dose/ ha***	***Name of pesticide***	***Dose/ ha***	***Name of pesticide***	***Dose/ ha***	***Name of pesticide***	***Dose/ ha***
1	Kumulus S (WG) Manzate 75 DF (WG) Falcon 460 (EC) Pyranica 20 (WP) Actara 25 (WG) + Nonit (SL)	2 kg 0.4 kg 0.1 l 0.1 kg 0.06 kg 0.1 l	Kumulus S (WG) Manzate 75 DF (WG) Pyranica 20 (WP)	4 kg 1.5 kg 0.3 kg	Kumulus S (WG) Manzate 75 DF (WG)	5 kg 2 kg	Kumulus S (WG) Penncozeb (DG) Tebusha 25 (EW)	3 kg 1.8 kg 0.4 l
2	Kumulus S (WG) Manzate 75 DF (WG) Falcon 460 (EC) Pyranica 20 (WP) Actara 25 (WG) + Nonit (SL)	2 kg 0.4 kg 0.1 l 0.1 kg 0.06 kg 0.1 l	Kumulus S (WG) Falcon 460 (EC) Curzate F (SC) Pyrinex 25 (CS) + Nonit (SL)	3 kg 0.3 l 3 ml 1 l 0.1 l	Kumulus S (WG) Pergado F 45 (WG) Tebusha 25 (EW) Pyrinex 25 (CS) + Nonit (SL)	3.8 kg 2.5 kg 0.5 l 1.25 l 0.06 l	Kumulus S (WG) Penncozeb (DG) Tebusha 25 (EW) Pyrinex 25 (CS)	2.7 kg 1.8 kg 0.4 l 0.9 l
3	Kumulus S (WG) Falcon 460 (EC) Tanos 50 (DP) Pyrinex 25 (CS) + Nonit (SL)	2 kg 0.3 l 0.4 kg 0.8 l 0.1 l	Kumulus S (WG) Curzate F (SC) Dynali (DC) Actara (SC) + Nonit (SL)	3 kg 3 l 0.5 l 0.08 l 0.1 l	Dynali (DC) Kumulus S (WG) Cymbal 45 (WG) Folpan 80 (WDG)	0.6 l 3.5 kg 0.3 kg 1.2 kg	Kumulus S (WG) Folpan 80 (WDG) Cymbal 45 (WG) Actara 25 (WG)	3 kg 1 kg 0.25 kg 0.08 l
4	Kumulus S (WG) Falcon 460 (EC) Tanos 50 (DP) + Nonit (SL)	2.7 kg 0.33 l 0.33 kg 0.16 l	Kumulus S (WG) Curzate F (SC) Dynali (DC) + Nonit (SL)	2.5 kg 2.5 l 0.4 l 0.1 l	Dynali (DC) Karathane Star (EC) Champion (WG) Actara 25 (WG)	0.7 l 1.1 l 2.1 kg 0.09 l	Karathane Star (EC) Rally Q (SC) Champion (WG)	0.9 l 0.9 l 2 kg
5	Kumulus S (WG) Falcon 460 (EC) Tanos 50 (DP) + Nonit (SL)	3.2 kg 0.32 l 0.45 kg 0.2 l	Talendo (EC) Karathane Star (EC) Kocide 2000 (WG) + Spur (LC) + Nonit (SL)	0.3 l 0.9 l 1.8 kg 0.2 l 0.2 l	Karathane Star (EC) Champion (WG): Kékfrankosnál Collis (SC): Chardonnaynál	0.7 l 1.3 kg 0.3 l	Karathane Star (EC) Falcon 460 (EC) Champion (WG) + Nonit (SL)	1 l 0.4 l 2 kg 0.13 ml
6	Kumulus S (WG) Falcon 460 (EC) Folpan 80 (WDG) Pyrinex 25 (CS) + Nonit (SL)	3.2 kg 0.32 l 1.3 kg 1.3 l 0.12 l	Falcon 460 (EC) Folpan 80 (WDG) Champion (WG) + Spur (LC)	0.3 l 1.2 kg 1.8 kg 0.2 l	Vegesol eReS (SE) Kumulus S (WG) Kocide 2000 (WG) Champion (WG)	3.3 l 3.2 kg 1.9 kg 1.9 kg	Vegesol eReS (SE) Kumulus S (WG) Teldor 500 (SC)	2.5 l 3 kg 0.75 l
7	Kumulus S (WG) Folpan 80 (WDG) Tanos 50 (DP) Dynali (DC)	3.2 kg 1.3 kg 0.44 kg 0.65 l	Kumulus S (WG) Kocide 2000 (WG)	3.3 kg 2.2 kg	Kumulus S (WG) Champion (WG)	1.3 kg 2 kg		
8	Kumulus S (WG) Kocide 2000 (WG) Collis (SC)	2.6 kg 2.2 kg 0.44 l						

The following parameters were measured: leaf transpiration rate (*E*, mol H_2O m^{-2} s^{-1}), stomatal conductance (g_s, mmol m^{-2} s^{-1}), leaf assimilation rate (*A*, µmol CO_2 m^{-2} s^{-1}), and intercellular CO_2 concentration (C_i, CO_2 ppm). Instantaneous (extrinsic) water-use efficiency (*WUE*, µmol CO_2 mol^{-1} H_2O) was also calculated (*A*/*E*).

The yield of the treated grapes (cluster weight and number/stock, average cluster weight; Pálfi et al., 2022a-b) and basic analytical parameters were also determined: pH (with Thermo Scientific Orion 3-Star pH meter); Hungarian (MM°) must degree (with e.g. refractometer); titratable acid content by NaOH titration (OIV 2023a-b; Török and Kállay, 2009). Wines were fermented using microvinification (directed; reductive technology for Chardonnay, on skins for Kékfrankos) from the crops of the first experiment (2013-2014). The analytical (pH, alcohol, titratable acidity) and phenolic parameters (total polyphenol, catechin, anthocyanin content) of the wines were also measured (OIV 2023a); the latter with methods based on spectrophotometrical measurements (Singleton and Rossi, 1965; Amerine and Ough, 1980; Ribéreau-Gayon and Stonestreet, 1965).

During the laboratory tests, we worked with the Kékfrankos variety: leaves from the vineyard were used as the source of *E. necator* inoculum and conducted the tests with leaves from greenhouse cuttings (PFO 2 v/v%). The leaf tissue localization of PFO was examined by Nile Red staining (Tan et al., 2005) and fluorescence microscopy. The effect of PFO on *E. necator* was assessed using viability tests based on FDA hydrolysis (Moyer et al., 2010; Schneider et al., 2012; Pálfi et al., 2021). To explore the effect of PFO on the grapevine, the following compounds were measured related to the plant stress response using methods based on spectrophotometric analysis: water-soluble and insoluble (PHEC) phenolics (Singleton and Rossi, 1965), H_2O_2 (Junglee et al., 2014) and pectin content (Miller, 1959). The amount of salicylic acid (SA) was determined using the HPLC-MS/MS method (Pálfi et al., 2021). We also assessed the activity of enzymes linked to the stress response with spectrophotometric methods (Pálfi et al., 2021): superoxide dismutase (SOD; Marklund and Marklund, 1974), catalase (CAT; Aebi, 1984) and guaiacol peroxidase (GPX; Aydin and Kadioglu, 2001), polyphenol oxidase (PPO; Kumar and Khan, 1982).

The statistical evaluation of the results was performed using the GraphPad Prism biostatistics program. Normality tests, then Student t-test or correlation test (Pearson r) or One-Way-ANOVA or Mixed Effect Analysis (similar to Two-Way ANOVA) were carried out. For the latter two, Tukey Multiple Comparison post-test were chosen to analyze the data and compare the treatments.

3. RESULTS

3.1. Field trials investigating the plant protection efficiency of paraffin oil

3.1.1 Performance of paraffin oil spraying against grapevine powdery mildew

The main goal of the study was to investigate the disease control capabilities of PFO in grapevine, primarily against grapevine powdery mildew. The assessment of disease frequencies and severities was done just before the harvest (BBCH 89) in the case of the first spraying experiment. In the second field experiment these parameters were determined in both veraison (BBHC 79) and pea-sized stage of the berries, while the near-harvest stage (BBCH 89) was examined only in 2015. The climatic characteristics of 2016 resulted in high disease pressure as well as high symptom severity and frequency. Therefore, the last spraying was CT in all parcels and the monitoring of BBHC 89 was omitted this year. The GPM percental symptom severity (BBCH 89 for 2013-2014; BBCH 79 for 2015-2015) were summarized in *Table 2.*

The PFO showed varying effectiveness in 2013-2014 (*Table 2.*). The highest applied PFO dosage (P3, 3.3 v/v%) showed similarly infection percentage values (~4%) as CT (positive control) in 2013 (<6%), a year with low GPM pressure. This effect of P3 was expressed especially on the less GPM-susceptible plant parts (leaves of Chardonnay and bunches of Kékfrankos). The other two PFO treatments (1.1 and 2.2 v/v%) also showed on average 56.8% lower infection levels compared to the untreated control in 2013. Accordingly, the 1 and 2 v/v% PFO were similarly efficient as DMI fungicide on a Chardonnay vineyard (Dell et al., 1998). In our field trial in 2014, the PFO was inefficient (possibly due to the high GPM pressure): each plant part of the two grapevine varieties was on average ~191% more infected compared to the mean infection level observed in the untreated control treatment. The dose-dependent effect of PFO have not been expressed in plant parts with more GPM-susceptibility (Kékfrankos leaves, Chardonnay clusters). In some case (Chardonnay bunches), the negative impact of P3 was also expressed resulting from the combined effect of high GPM infection and the phytotoxicity of PFO. Treatment P1 did not show GPM-inhibiting activity in both years; similar to the results of leaf GPM coverage observed in a Tempranillo vineyard (Martín et al., 2005). However, according to another study, the 1 v/v% PFO treatment moderately inhibited GPM on Chardonnay clusters compared to the untreated control, while it did not differ significantly from the data obtained in the sulfur- and water-treated plots (Janousek et al., 2009). These contradictory results suggested that the field efficiency of the tested PFO dosages is uncertain in the case of their sole application, and raised the necessity of long-term experiments.

Table 2. The effect of the treatments on the intensity of visible grape powdery mildew (GPM) infection of leaves and clusters (%) between 2013 and 2016 under the field spraying experiments. Evaluations were made only before harvest (BBCH 89) between 2013-2014, while in 2015-2016 also under pea-sized berries (BBCH 75) and veraison (BBCH 79) development stages. Since there is no harvest monitoring data from 2016, the results from the veraison of 2015-2016 years were included in the table. Each data represents the mean value ± standard deviation (SD) in the columns and is coloured according to the size of the MV values. ***Treatments:*** C0 (untreated control); CT (vineyard treatment with traditionally used fungicides as positive control); P1(paraffin oil (PFO) 1.1 v/v%); P2 (PFO 2.2 v/v%); P2CT (combined P2 and CT); P3 (PFO 3.3 v/v%); P3CT (combined P3 and CT); P3CTm (modified P3CT: no PFO spraying under blooming and fruit set). Abbreviations of grapevine cultivars: Chardonnay (CH), Kékfrankos (KF).

Year, BBCH	**Plant part**	**Variety (abbr.)**	**The effect of the treatments on the intensity of GPM infection (MV (%) ± SD)**							
			C0	**CT**	**P1**	**P2**	**P2CT**	**P3**	**P3CT**	**P3CTm**
2013	leaf	CH	48.1±3.6	3.8±1.0	12.2±6.5	13.5±5.0	no data	5.6±2.4	no data	no data
BBCH 89		KF	45.6±7.5	5.1±11	27.5±6.6	26.9±10.7	no data	17.0±4.9	no data	no data
	cluster	CH	90.4±8.1	8.8±1.1	68.3±11.8	53.1±9.8	no data	41.7	no data	no data
		KF	55.9±7.1	3.4±0.9	13.8±4.6	7.6±2.1	no data	41.7±14.3	no data	no data
2014	leaf	CH	24.2±23.9	12.6±14.0	18.4±18.9	16.0±20.3	no data	5.0±1.3	no data	no data
BBCH 89		KF	no evaluation data							
	cluster	CH	89.8±18.9	18.7±18.4	69.8±24.4	58.0±18.9	no data	86.7±14.5	no data	no data
		KF	91.2±16.0	25.9±18.9	81.0±25.0	84.3±18.7	no data	69.1±30.9	no data	no data
2015	leaf	CH	36.1±31.3	0.3±1.3	no data	15.7±23.6	0.7±2.2	6.1±15.0	0.0±0.1	0.4±1.2
BBCH 79		KF	46.2±34.3	0.1±0.4	no data	11.2±19.1	0.1±0.4	4.7±11.9	0.0±0.3	0.0±0.3
	cluster	CH	55.7±32.8	6.8±10.4	no data	42.0±30.7	2.5± 3.2	26.8±28.0	2.4± 3.7	15.2±18.8
		KF	8.2±8.6	0.4±1.0	no data	2.6±4.0	0.4±1.1	0.9±2.0	0.1±0.4	0.6±1.8
2016	leaf	CH	67.5±29.7	14.3±19.5	no data	57.9±23.6	9.0±14.3	36.8±28.9	12.0±19.6	12.6±18.2
BBCH 79		KF	63.3±31.4	5.2±6.2	no data	43.7±30.8	2.1±5.5	42.7±29.8	4.0±7.4	3.2±5.5
	cluster	CH	81.3±23.1	54.8±28.1	no data	90.4±10.4	50.5±25.8	78.8±23.5	60.4±27.7	60.9±23.9
		KF	39.6±34.5	10.0±10.8	no data	29.1±26.6	4.8±4.2	29.3±27.9	6.0±5.0	7.0±13.4
		Treatments	**C0**	**CT**	**P1**	**P2**	**P2CT**	**P3**	**P3CT**	**P3CTm**
		Colour scale means	**0%**			**50%**				**100%**

Based on the previous results, a second two-year experiment (2015-2016) was carried out, including the combinations of PFO with CT treatments. Under this survey, sole treatments with PFO in 2.2 (P2) and 3.3 v/v% (P3) dosages showed on average 42.3% lower intensity of GPM infection compared to the untreated control. (*Table 2.*), similarly to the first experiment. During the four-year experiment, the efficacy of PFO fell between the values measured in case of C0 and CT treatments. However, these results were also greatly affected by vintage characteristics and the GPM-susceptibility of the different plant parts of the examined varieties. The effects PFO treatments on GPM infection were very limited in 2016 which year showed similarly high GPM pressure as 2014 in the first field trials. The individual differences in GPM susceptibility of leaves and clusters were manifested in both years and varieties.

Treatment P3CTm occasionally resulted in higher GPM-intensity than CT, P2CT and P3CT treatments in 2015, especially on Chardonnay bunches, at veraison and before harvest and in the case of GPM-frequency on Kékfrankos leaves under pea-sized berry developmental stage. This phenomenon is possibly due to the different spraying strategies. Parcels with P3CTm treatments received only CT treatments during flowering and fruit set, resulting in the loss of the beneficial effects of PFO-CT combinations due to the adjuvant characteristics of PFO on more GPM-susceptible plant parts. The difference between grapevine cultivars and CT-containing treatments did not appear in this form in the more GPM-infected year (2016).

The combination of PFO with synthetic fungicides (CT) resulted in higher protection compared to CT in both years and tested varieties according to our hypothesis. However, this difference was not significant in every comparison and depended on the experimental variables mentioned earlier. Due to these and other factors, studies on the combined use of PFO with fungicides have also produced contradictory findings. Some authors have reported that the combination of 1 or 2 v/v% JSO (PFO) with other agents (e.g., sulfur, potassium, myclobutanil) resulted in improved efficacy (Dell et al., 1998; Janousek et al., 2009). In contrast, other investigations found no evidence or indication of a synergistic interaction between PFO and its combination partners (Martín et al., 2005). Based on the results of these studies and our own experiments, it can be concluded that the efficacy of PFO may be influenced not only by its chemical properties (e.g., viscosity) but also by the morphological characteristics of the plant surface (e.g., the leaf's ability to retain spray solution) (Nail and Howell, 2004; Baudoin et al., 2006). Grove et al. (2005) highlighted the potential impact of vintage-specific conditions (such as the presence or absence of warm and sunny weather) on JSO degradation as an explanatory factor for differences in their results.

In our study, the moderate increase in efficacy observed for the PFO–CT combination may have been caused by one or both of the following factors:

1) PFO may act as an elicitor, positively influencing plant immunity through the induction of various stress-related responses in grapevine, thereby reducing susceptibility to GPM infection, as observed in our experiments (Pálfi et al., 2021).

2) PFO may also function as an adjuvant, enhancing the adhesion and absorption of co-applied fungicides − a phenomenon reported by other authors as well (e.g., Zabkiewicz, 2002; Nita et al., 2007; Santos et al., 2017).

The results of the examination of the culturable yeast and filamentous fungi on leaves and berries in 2014 and 2015 were in accordance with the parallel investigation of GPM intensity. In 2015 with lower fungal infection risk, no differences were observed in the populations of filamentous fungi and yeasts among the treatments for either cultivar or sample type.

Table 3. Climatic characteristics of 2013-2016 vintages: mean monthly temperature (°C), total monthly precipitate (mm) mean air relative humidity (%), according to the report made by Boreas Kft.

	2013			**2014**		
Month	**Temperature (°C)**	**Precipitation (mm)**	**Humidiy (%)**	**Temperature (°C)**	**Precipitation (mm)**	**Humidiy (%)**
1	-0.7	56.1	92.6	2.3	34.3	96.5
2	2.4	85.3	91.1	4.1	38.8	92.8
3	3.2	98.8	84.2	9.9	10.4	65.5
4	12.2	30.6	68.3	12.6	56.7	76.0
5	16.4	159.0	79.2	15.8	119.1	74.8
6	20.0	130.5	78.9	20.0	15.8	63.0
7	22.8	6.5	62.2	22.0	114.3	74.1
8	22.8	27.1	60.0	20.0	114.3	77.5
9	14.8	34.4	63.9	17.0	153.5	83.3
10	12.6	53.4	75.9	11.7	60.0	91.3
11	7.5	53.1	88.1	7.2	13.5	94.5
12	0.8	2.8	92.5	3.0	36.9	88.4
	2015			**2016**		
Month	**Temperature (°C)**	**Precipitation (mm)**	**Humidity (%)**	**Temperature (°C)**	**Precipitation (mm)**	**Humidiy (%)**
1	1.3	69.2	92.6	-2.0	71.4	96.3
2	1.8	11.7	91.1	4.9	95.8	92.8
3	6.9	20.8	84.2	6.7	59.2	65.5
4	11.1	10.0	68.3	12.6	32.1	76.0
5	16.2	74.8	79.2	16.6	43.2	74.8
6	20.5	32.4	78.9	21.1	70.3	63.0
7	23.7	51.0	62.2	22.2	124.7	74.1
8	23.7	139.3	60.0	20.8	26.8	77.5
9	17.6	66.6	63.9	18.3	7.6	83.3
10	10.1	55.6	75.9	9.1	57.4	91.3
11	6.2	31.5	88.1	4.5	87.4	94.5
12	2.1	13.0	92.5	-1.9	0.6	88.4

In 2014, PFO treatments decreased the CFU of filamentous fungi on Chardonnay berries similarly to CT. On the leaves of Kékfrankos, PFO treatments increased the portion of filamentous fungi in the whole culturable mycobiota compared to the control. This latter may in connection with the higher susceptibility of Kékfrankos leaves to fungal infections. Treated Kékfrankos berries showed higher CFU for filamentous fungi compared to C0.

The CT treatment resulted in the highest filamentous/yeast fungi CFU rate, while this parameter was the lowest in C0 samples. The values for PFO-treated berries fell between C0 and CT. This may be due to the late harvest and sampling time (this carried out with the GPM monitoring) in a rainy season (*Table 3* on page 10) resulting in the increased growth of saprophytes. Therefore, the differences between the treatments lesser expressed. In general, PFO treatments no or lesser affected the yeast population than CT in 2014: the percentage of yeasts within the culturable mycobiota of PFO treatments averaged 61.5% in Kékfrankos (compared to 27% in the CT treatment) and 80% in Chardonnay (compared to 78% in the CT treatment). This may be important as wild yeast populations can also exert a significant influence on the aromatic complexity of wines. The rate of filamentous/yeast fungi was more balanced in 2015 than in 2014, in both varieties and examined plant parts.

3.1.2. The effect of paraffin oil on the gas exchange of grapevine leaves

The application of PFO showed a positive (but lower than CT) impact on grapevine physiology according to the photosynthetic parameters (g_s, A, E) of leaves. A decline in gas exchange associated with oil treatments has also been reported by other studies (Hodgkinson et al., 2002; Finger et al., 2002). The positive effect of PFO was primarily observed under climatic conditions characterized by low GPM pressure, in contrast to the generally reduced photosynthetic performance recorded in the 2014 season with high GPM infection levels. This vintage-dependent phenomenon was expressed to a higher extent on Kékfrankos leaves, which are more susceptible to GPM than Chardonnay leaves. This suggests that the applied PFO dosage should be specified according to both vintage, as well as variety characteristics. For instance, other studies have also highlighted the role of morphological differences between leaves in determining spray coverage and retention capacity (Mullins et al., 1995; Baudoin et al., 2005). Since the experiment was conducted under field conditions, the potential direct phytotoxic effect of PFO could not be measured directly. However, the absence of a PFO dose-dependent decrease in photosynthetic parameters, regardless of cultivar or vintage, suggests that the PFO formulation used in this study does not exert a significant phytotoxic effect on grapevines, at least relative to the negative impact of GPM infection. This assumption is partially supported by the observation that leaves treated with 3.3 v/v% PFO, which provided the best control against GPM, showed no significant differences in g_s, A, or E compared to the CT during the first gas exchange measurement following the fourth spraying, in any year or cultivar. Nevertheless, based on disease assessments, the 3 v/v% PFO-treated Chardonnay leaves exhibited on average 47.2% higher infection intensity than the CT in both years, while the corresponding difference for Kékfrankos exceeded 200%.

Analysis of the correlation between g_s and E values (*Table 4.*) supported that GPM has a more negative impact on E, relative to the PFO treatments, through affecting g_s values (Moriondo et al., 2005). Control treatments did not show a significant g_s-E correlation in 2013, while C0 showed a significant correlation in 2014, possibly due to the high GPM pressure in the latter year (Pálfi et al., 2016a). The difference between the GPM-inhibitory efficiency of P1 and P2 dosages was expressed in the g_s-E correlations in both years and in both varieties (*Table 4.*), while generally there was no correlation in the case of P3. The connection between g_s and E values suggests, that the GPM-inhibitory effect of P3 has a higher impact on the gas exchange of grapevine leaves, than the phytotoxicity of PFO described in previous papers (Hodgkinson et al., 2002; Finger et al., 2002).

Table 4. Results of the correlation analysis between mean stomatal conductance (gs) and leaf assimilation rate (A) data or leaf transpiration rate (E) data in the case of each treatment, variety, and year (5-5 measurement dates/year). Asterisks mark the significance of differences (* $p < 0.05$, ** $p < 0.005$), ns: not significant. Treatments: C0 (untreated control); CT (vineyard treatment with traditionally used fungicides as positive control); P1(paraffin oil (PFO) 1.1 v/v%); P2 (PFO 2.2 v/v%); P3 (PFO 3.3 v/v%).

Chardonnay 2013	**mean g_s vs. A data**					**mean g_s vs. E data**				
	C0	**CT**	**P1**	**P2**	**P3**	**C0**	**CT**	**P1**	**P2**	**P3**
Pearson r	0.921	−0.114	0.787	0.940	0.821	0.730	0.833	0.870	0.930	0.806
P-value	0.026	0.854	0.114	0.017	0.088	0.161	0.079	0.055	0.020	0.099
Significance	*	ns	ns	*	ns	ns	ns	ns	*	ns
Kékfrankos 2013	**mean g_s vs. A data**					**mean g_s vs. E data**				
	C0	**CT**	**P1**	**P2**	**P3**	**C0**	**CT**	**P1**	**P2**	**P3**
Pearson r	0.793	0.208	0.834	−0.651	0.467	0.775	0.677	0.970	0.761	0.807
P-value	0.109	0.737	0.079	0.234	0.428	0.123	0.209	0.006	0.135	0.090
Significance	ns	ns	ns	ns	ns	ns	ns	**	ns	ns
Chardonnay 2014	**mean g_s vs. A data**					**mean g_s vs. E data**				
	C0	**CT**	**P1**	**P2**	**P3**	**C0**	**CT**	**P1**	**P2**	**P3**
Pearson r	0.987	0.606	0.955	0.815	0.909	0.899	−0.210	0.969	0.985	0.865
P-value	0.001	0.278	0.011	0.092	0.032	0.038	0.734	0.006	0.002	0.058
Significance	**	ns	*	ns	*	*	ns	**	**	ns
Kékfrankos 2014	**mean g_s vs. A data**					**mean g_s vs. E data**				
	C0	**CT**	**P1**	**P2**	**P3**	**C0**	**CT**	**P1**	**P2**	**P3**
Pearson r	0.859	0.864	0.833	0.498	0.442	0.979	0.760	0.929	0.957	0.948
P-value	0.062	0.058	0.080	0.393	0.455	0.003	0.135	0.022	0.010	0.014
Significance	ns	ns	ns	ns	ns	**	ns	*	*	*

Kékfrankos leaves did not show a positive correlation between g_s and A, regardless of experimental year, or applied treatments, while correlation occurred in some cases of Chardonnay in both years (*Table 2.*). This difference is possibly based on the morphological differences of the leaves of these varieties. According to Baudoin et al. (2006) PFO coverage and retention determine its negative effect on photosynthesis, which parameters are affected by the characteristics of the treated leaves, as well as the physico-chemical parameters of the applied PFO. Oils with a lower viscosity can penetrate into the tissues causing photosynthetic inhibition. In contrast, oils with a higher viscosity remain on the leaf surface (Nail and Howel, 2004; Gudin et al., 1976) with negligible negative impacts on leaves. Our examined PFO product can not diffuse deeper than the first layer of epidermal cells, therefore its phytotoxicity would be limited (Pálfi et al. 2021).

3.1.2. The effect of paraffin oil on harvest yields and analytical parameters of musts and wines

The results of the estimation of gas exchange parameters, harvest yields, as well as must and wine analytical parameters were in accordance with the GPM-inhibitory efficiency of the treatments according to our hypothesis. The effectivity of PFO in GPM inhibition and prevention of yield losses was generally lower compared to CT. The observed higher yield losses are possibly the result of the GPM-induced damage of photosynthetic leaf area. The earlier hypothesized direct (either positive or negative) effect of PFO on harvest yield could not be observed, neither in the case of P3CT and P3CTm treatments. There were no notable differences between the treatments in 2015-2016, possibly due to the smaller treated parcels, as well as the low number of samples. In general, the CT- and P2CT-treated plots (and in some cases the P3CT-treated plots) produced higher yield per vine and larger clusters in both years compared to the C0 and PFO-treated plots.

GPM infection had a great impact on the harvest yield and quality in our experiment. This effect was observed as the inhibition of assimilation, delayed ripening, lower soluble sugar (TSS) content (and lower alcohol content in wine) in must, as well as the increased titratable acidity (TA); similar to other studies (Finger et al, 2002). Based on our results, the basic and phenolic analytical parameters of wines produced from the 2013–2014 experimental plots reflected the efficacy of the respective treatments − as hypothesized − while also capturing the effects of the specific vintage and the GPM susceptibility of the studied cultivar.

3.2. The effect of paraffin oil on the causal agent of grapevine powdery mildew

The quantitative assessment of the viability of *E. necator* conidia -using FDA-hydrolysis assay- did not show a difference between water and 2 v/v% PFO treatments (*Figure 1A*).

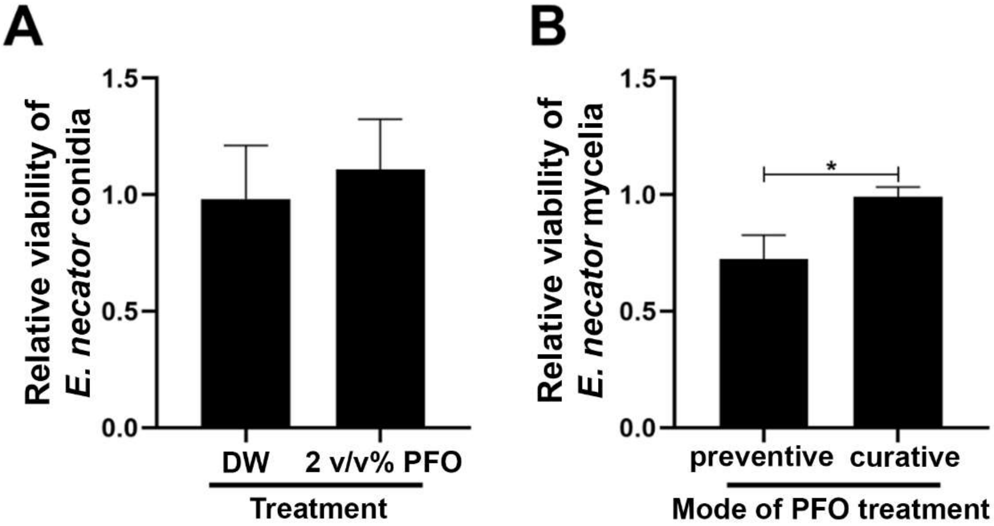


Figure 1. (A-B): Effect of 2 v/v% PFO and distilled water (DW) on the viability of *E. necator* conidia (A), and the effect of preventive or curative application of 2 v/v% PFO on the viability of *E. necator* mycelia (B) assessed by FDA hydrolysis assay. Mean values and standard deviations were shown. Asterisk marks significant ($p<0.05$) difference.

According to the examination of artificially infected grapevine foliar disks, the curative application of PFO did not affect the viability of *E. necator* mycelia, while its preventive application resulted in a mild but significant decrease (*Figure 1B*), in accordance with the results of Northover and Schneider (1996). Our microscopic examinations of PFO-treated, naturally infected grapevine leaves also did not show any direct antifungal effect of PFO against *E. necator*. In accordance with previous studies, these results suggest that PFO affects the host plant rather than *E. necator* itself (Calpouzos, 1966; Northover és Schneider, 1996).

3.3. The localization of paraffin oil in grapevine leaf tissues

Epifluorescent microscopic examination of the localization of Nile Red-stained PFO on leaves, showed the low coverage of leaf surface by the oil, which later can be detected in small, separate spots (*Figure 2A*). These were absent on leaves treated with distilled water (*Figure 2D*). At higher magnification, the PFO seemed to be bound to the walls of the epidermal cells (*Figure 2B*), and can not diffuse deeper than the first layer of cells, detected on the leaf cross sections (*Figure 2C*). This latter phenomenon can be observed after an additional 24 h incubation. These results are in accordance with the indirect antifungal mode of action of PDSOs, previously proposed by Northover and Schneider (1996).

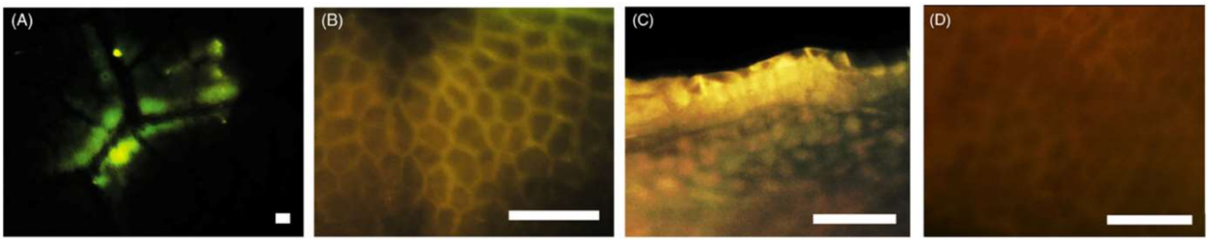


Figure 2. (A-D): Epifluorescent photomicrographs of grapevine leaf surface (A, B, D), or cross-section (C) 24 h after treatment with distilled water (control, D), or 2 v/v% PFO amended with Nile Red dye (A, B, C). Scalebars represent: 50 µm (own figure).

3.4. The effect of paraffin oil on grapevine

The PFO possibly causes local stress in grapevine leaves (*Figure 3.)* which is expressed in the elevated concentration of H_2O_2, possibly a result of the measured elevated SOD activity accompanied by an unaffected CAT activity. H_2O_2 is a well-known intracellular signal molecule (Levine et al., 1994) promoting plant defense reactions like the accumulation of SA (Leon et al., 1995), which latter was also measured in PFO-treated leaves. This phytohormone – as a long-range signal – systemizes stress and defense reactions in the whole plant (Gao et al., 2015). This process results in the elevated synthesis of phenolics (Blanch et al., 2020) through the induction of e.g. GPX and PPO enzyme activities, which phenomena were also detected in PFO-treated grapevine leaves. The deposition of lignin leads to the secondary thickening of the plant cell wall providing mechanical defense against the penetration of pathogens (Bhuiyan et al., 2009). Water-soluble phenolics (e.g. phytoalexins) can possess antifungal activity (Jeandet et al., 2002), resulting in the decreased growth of pathogens (e.g. *E. necator*).

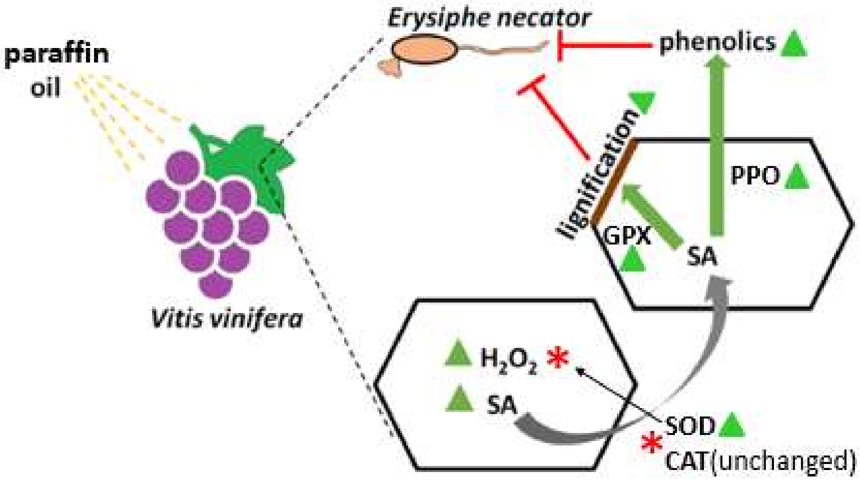


Figure 3.: Graphical abstract of possible mode of action of PFO (own figure).
Abbreviations: H_2O_2 (hydrogen peroxide); SA (salicylic acid); SOD (superoxide dismutase); CAT (catalase); GPX (guaiacol peroxidase); PPO (polyphenol oxidase). Green triangles: detected increasement in PFO-treated leaves; green arrows: positive correlation according to literature; red lines: inhibition according to literature

According to our result (Pálfi et al. 2021) PFO may induce systemic acquired resistance (SAR). The PFO-induced SAR may decrease the susceptibility of treated grapevine leaves towards GPM infection, which was also observed in the field experiments. This phenomenon may explain the mode of action of PFO against phytopathogens.

4. NEW SCIENTIFIC RESULTS OF THE DOCTORAL DISSERTATION

1) First direct proof of the lack of fungicide activity of PFO against *E. necator*, determined by FDA-hydrolysis assay.
2) Examination of the leaf tissue localization of paraffin oil revealed that, on treated leaves, the oil separates into small droplets on the surface and does not diffuse beyond the first epidermal cell layer; however, it is capable of binding to the cell walls of these cells. This property may make paraffin oil suitable for use as a spray adjuvant or efficacy-enhancing additive.
3) Paraffin oil treatment induced the following physiological responses in grapevine: a moderate accumulation of H_2O_2 (<50%) occurred in the leaves. Through the modulation of stress-related enzymes, this induced salicylic acid biosynthesis, resulting in lignin accumulation and enhanced phenolic biosynthesis. Based on these observations, paraffin oil is likely to trigger systemically acquired resistance (SAR), thereby reducing grapevine susceptibility to *E. necator* infection. In our field spraying experiments, under years with low to moderate powdery mildew pressure, the 2.2 v/v% paraffin oil treatment showed an average infection intensity 58.2% lower than the untreated control, while the 3.3 v/v% treatment showed a 76.2% reduction; the fungicide-treated control exhibited a 93.4% reduction
4) The analysis of culturable fungal communities from leaf and berry samples collected simultaneously with field powdery mildew monitoring represented a novel approach. Our results indicated that, unlike the fungicide-treated control (CT), paraffin oil treatments did not or only slightly affected the yeast population on grape berries during the year of higher fungal infection pressure (2014). The proportion of yeasts within the culturable mycobiota was on average 61.5% for paraffin oil treatments in Kékfrankos (CT: 27%) and 80% in Chardonnay (CT: 78%).
5) Results of leaf gas exchange measurements in the small-plot spraying experiment confirmed that no clearly defined paraffin oil concentration could be identified as phytotoxic. Leaves treated with the 3.3 v/v% paraffin oil (P3) − which provided the best control against powdery mildew − showed no significant differences in stomatal conductance (g_s), assimilation rate (A), or transpiration rate (E) compared to the fungicide-treated control (CT), for either cultivar or year, even during the first gas exchange measurement (late June–early July) following four treatments. Based on these findings, we conclude that the phytotoxicity caused by paraffin oil is minimal when compared with the negative effects of *E. necator*

infection on photosynthesis. Compared to CT, P3-treated Chardonnay leaves exhibited, on average, 47.2% higher infection intensity in both years, while in Kékfrankos this difference exceeded 200%.

6) The observed plant protection efficacy of paraffin oil against powdery mildew, along with its long-recognized potential phytotoxicity, is associated with its role as an elicitor in plant stress responses. This is closely related to the leaf tissue localization capacity determined by the physical properties of the paraffin oil formulation (e.g. viscosity) and to the morphological characteristics of the tested grapevine cultivars that influence spray retention (e.g. dense leaf trichomes and thick, waxy berry cuticles improve adherence). This relationship highlights the need to specifically consider these factors (e.g. adjusting the applied concentration) when using paraffin oil in plant protection, to maintain a balance between its adverse (phytotoxic, concentration-dependent) and beneficial (protective) effects.

5. RESULTS THAT CAN BE USED IN PRACTICE

1) As a result of our spraying experiments, we obtained data and observations on powdery mildew infection for several paraffin oil concentrations applied alone on the grapevine cultivars Chardonnay and Kékfrankos, covering two (1.1 v/v%) or four (2.2; 3.3 v/v%) growing seasons. Based on our results, paraffin oil at 2.2 and 3.3 v/v% concentrations can be successfully applied on its own under low to moderate infection risk conditions: compared to the untreated control, these paraffin oil treatments exhibited an average reduction of 67.2% in infection intensity.
2) Since the mechanism of action of paraffin oil against grapevine powdery mildew is based on the induction of the plant's stress response, its inhibitory effect on Erysiphe necator is presumably only slightly influenced by the extent of the exposed leaf surface; rather, it appears to correlate positively with the applied concentration of paraffin oil. It may therefore be effective even at lower application doses when used at higher concentrations, while maintaining negligible phytotoxicity.
3) Our study evaluated higher paraffin oil concentrations (2.2 and 3.3 v/v%) than those used in previous grapevine-related research, as well as their combinations with fungicides exhibiting multiple modes of action (not only DMI or QoI types) applied as the control treatment (CT), from a plant protection perspective against powdery mildew. Furthermore, in our experiment, paraffin oil was not applied as a complementary partner to fungicides or as part of their gradual substitution. Instead, a sequential spraying protocol was used within the same day, where paraffin oil was applied first, followed by the CT mixture adjusted to the given year's characteristics. This approach eliminated the potential phytotoxicity that could arise from mixing spray liquids.
4) The P3CTm plots received only the fungicide treatment (CT) used in the vineyard during the powdery mildew–susceptible flowering and fruit set stages, without paraffin oil (3.3 v/v%). Consequently, the potential beneficial effect of paraffin oil as an efficacy-enhancing adjuvant could not manifest effectively: in some cases (e.g. 2015), the P3CTm treatment showed on average 18.2% higher powdery mildew infection indices compared to other CT-containing treatments. This represents an important observation from a spray technology perspective for the grapevine cultivars studied.
5) Our research findings contribute to one of today's key objectives — the advancement of precision viticulture and crop protection — through the assessment of paraffin oil's applicability in plant protection on Chardonnay and Kékfrankos cultivars.

6. LIST OF REFERENCES

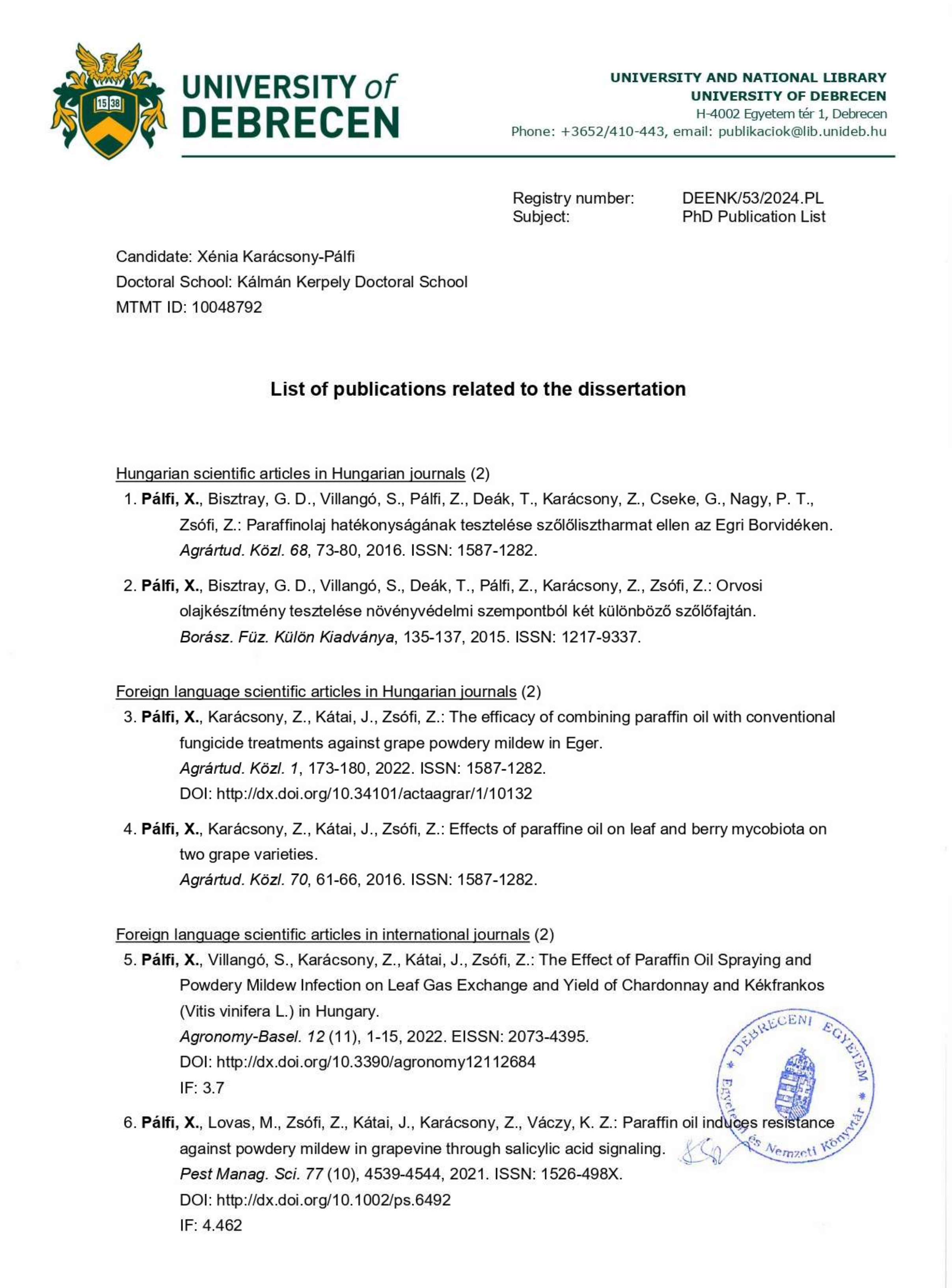
1. Aebi, H.: 1984. Catalase *in vitro*. [In: William, B. J. (Ed) Methods in Enzymology.]. Academic Press, New York, pp. 121–126.
2. Amerine, M. A. - Ough, C. S.: 1980. Phenolic compounds. [In: Methods for analysis of musts and wines.]. Wiley, New York, pp. 175-199.
3. Aydin, N. – Kadioglu, A.: 2001. Changes in the chemical composition polyphenol oxidase and peroxidase activities during development and ripening of medlar fruits (*Mespilus germanica* L.). *Bulgarian Journal of Plant Physiology*. 27:85–92.
4. Baudoin, A. - Finger, S. M. - Wolf, T. K.: 2006. Factors affecting reductions in photosynthesis caused by applying horticultural oil to grapevine leaves. *HortScience*. 41(2):346-351.
5. Bhuiyan, N. H. - Selvaraj, G. – Wei, Y. – King, J.: 2009. Role of lignification in plant defense. *Plant Signaling & Behavior*. 4:158–159.
6. Blanch, G. P. - Gómez-Jiménez, M. C. - Del Castillo, M. L. R.: 2020. Exogenous salicylic acid improves phenolic content and antioxidant activity in table grapes. *Plant Foods for Human Nutrition.* 75:177–183.
7. Calpouzos, L.: 1966. Action of oil in the control of plant disease. *Annual Review of Phytopathology*. 4:396-390.
8. Dell, K. J. - Gubler, W. D. - Krueger, R. - Sanger, M. - Bettiga, L. J.: 1998. The efficacy of JMS Stylet-Oil on grape powdery mildew and bunch rot and effects on fermentation. *American Journal of Enology and Viticulture.* 49:11-16.
9. Doster, M. A. - Schnathorst, W. C.: 1985 Compare susceptibility of various grapevine cultivars to the powdery mildew fungus *Uncinula necator*. *American Journal of Enology and Viticulture.* 36(2):101-104.
10. Finger, S. A - Wolf, T. K. - Baudoin, A. B.: 2002. Effects of horticultural oils on thephotosynthesis, fruit maturity, and crop yield of winegrapes. *American Journal of Enology and Viticulture.* 53:116-124.
11. Gadoury, D. M., Seem, R. C., Ficke, A., and Wilcox, W. F. 2003. Ontogenic resistance to powdery mildew in grape berries. *Phytopathology.* 93:547-555.
12. Gaforio, L. - García-Muñoz, S. - Cabello, F. - Muñoz-Organero, G.: 2011. Evaluation of susceptibility to powdery mildew (*Erysiphe necator*) in *Vitis vinifera* varieties. *Vitis*. 50(3):123–126.
13. Gao, Q. M. – Zhu, S. – Kachroo, P. – Kachroo, A.: 2015. Signal regulators of systemic acquired resistance. *Frontiers in Plant Science*. 6:228.
14. Grove, G. G. - Lunden, J. - Spayd, S.: 2005. Use of petroleum delivered spray oils in Washington grapevine powdery mildew management programs. *Plant health progress.* 6(1):14.
15. Gudin, C. - Syratt, W. J. - Boize, L.: 1976. The mechanism of photosynthetic inhibition and the development of scorch in tomato plants treated with spray oils. *Annals of Applied Biology.* 84:213-219.
16. Hodgkinson, M. C. - Johnson, D. - Smith, G.: 2002. Causes of phytotoxicity induced by petroleum-derived spray oils. [In: Beattie, G. A. C., Watson, D. M., et al. (Eds) Spray Oils Beyond 2000.]. University of Western Sydney, NSW, Australia, pp. 170-178.

17. Janousek, C. N. - Bay, I. S. - Gubler, W. D.: 2009. Control of grape powdery mildew with synthetic, biological, and organic fungicides: 2009 filed trials. UC Davis: Department of Plant Pathology, CA, USA. https://escholarship.org/uc/item/8fz3p4vc
18. Jeandet, P. - Douillet-Breuil, A. C. – Bessis, R. – Debord, S-. – Sbaghi, M. – Adrian, M.: 2002. Phytoalexins from the Vitaceae: biosynthesis phytoalexin gene expression in transgenic plants antifungal activity and metabolism. *Journal of Agricultural and Food Chemistry*. 50:2731–2741.
19. Junglee, S. – Urban, L. – Sallanon, H. - Lopez-Lauri, F.: 2014. Optimized assay for hydrogen peroxide determination in plant tissue using potassium iodide. *American Journal of Analytical Chemistry*. 5:730–736.
20. Kumar, K. - Khan P.: 1982. Peroxidase and polyphenol oxidase in excised ragi (*Eleusine corocana* cv PR 202) leaves during senescence. *Indian Journal of Experimental Biology*. 20:412–416.
21. Leon, J. – Lawton, M. A. – Raskin, I.: 1995. Hydrogen peroxide stimulates salicylic acid biosynthesis in tobacco. *Plant Physiology*. 108:1673–1678.
22. Levine, A. – Tenhaken, R. – Dixon, R. – Lamb, C.: 1994. H_2O_2 from the oxidative burst orchestrates the plant hypersensitive disease resistance response. *Cell*. 79:583–593.
23. Marklund, S. – Marklund, G.: 1974. Involvement of the superoxide anion rad-ical in the autoxidation of pyrogallol and a convenient assay for superoxide dismutase. *European Journal of Chemistry*. 47:469–474.
24. Martín, B. - Hernández, S. - Silvarrey, C. - Jacay, J. A. - Cabaleiro, C.: 2005. Vegetable, fish and mineral oils control grapevine powdery mildew. *Phytopathologia Mediterranea*. 44:169-179.
25. Matese, A. - Di Gennaro, S. F.: 2015. Technology in precision viticulture: a state of the art review. *International Journal of Wine Research*. 7:69–81.
26. Miller, G. L.: 1959. Use of dinitrosalicylic acid reagent for determination of reducing sugar. *Analytical Chemistry*. 31:426-428.
27. Moriondo, M. - Orlandini, S. - Giuntoli, A. - Bindi, M.: 2005. The effect of downy and powdery mildew on grapevine (*Vitis vinifera* L.) leaf gas exchange. *Journal of Phytopathology*. 153:350–357.
28. Moyer, M. M. – Gadoury, D. M. - Cadle-Davidson, L. – Dry, I. B. – Magarey, P. A.- Wilcox, W. F. - Seem, R. C.: 2010. Effects of acute low-temperature events on development of *Erysiphe necator* and susceptibility of *Vitis vinifera*. *Phytopathology*. 100:1240–1249.
29. Nail, W. R. - Howell, G. S.: 2004. Effects of powdery mildew of grape on carbon assimilation mechanisms of potted ‘Chardonnay’ grapevines. *HortScience*. 39(7):1670-1673.
30. Nazari, M. - Dada, A. - Asgharzadeh, A.: 2014. Effects of spray volck oil in different times on the cluster characters of grape (Kolahdari var) in North-Khorasan Condition. *Indian Journal of Fundamental and Applied Life Sciences*. 4(2):2231-2345.
31. Nile, A. S. - Kwon, Y. D. - Nile, S. H.: 2019. Horticultural oils: possible alternatives to chemical pesticides and insecticides. *Environmental Science and Pollution Research*. 26:21127-21139.

32. Nita, M. - Ellis, M.A. -. Wilson, L.L. - Madden, L.V.: 2007. Evaluation of the curative and protectant activity of fungicides and fungicide–adjuvant mixtures on Phomopsis cane and leaf spot of grape: A controlled environment study. *Crop Protection*. 26(9):1377-1384.
33. Northover, J. - Schneider, K. E.: 1996. Physical modes of action of petroleum and plant oils on powdery and downy mildews of grapevines. *Plant Disease.* 77:152-157.
34. Northover, J., and Timmer, L. W. 2002. Control of plant diseases with petroleum- and plant-dervived oils. [In: Beattie, G. A. C., Watson, et al. (Eds) Spray Oils Beyond 2000.]. University of Western Sydney, NSW, Australia, pp. 512-526.
35. OIV, 2023a.: Total acidity (Method OIV-MA-AS313-01). [In: OIV (ed) Compendium of international methods of wine and must analysis. Vol 1. Edition 2023.]. Dijon, France, pp. 398-401.
36. OIV, 2023b.: Evaluation by refractometry of the sugar concentration in grape musts (Method OIV-MA-AS2-02). [In: OIV (ed) Compendium of international methods of wine and must analysis. Vol 1. Edition 2023.]. Dijon, France, pp. 64-66.
37. Özkara, A. - Akyil, D. - Konuk, M.: 2011. Pesticides, environmental pollution, and health. [In: Larramendy, M. L., Soloneski, S. (szerk.) Environmental Health Risk - Hazardous Factors to Living Species.]. IntechOpen, London, US. doi: 10.5772/63094.
38. Pautasso, M. - Dehnen-Schmutz, K. - Holdenrieder, O. - Pietravalle, S. - Salama, N. - Jeger, M. J. - Lange, E. - Hehl-Lange, S.: 2010. Plant health and global change – some implications for landscape management. *Biological Reviews.* 85:728-755.
39. Pautasso, M. – Döring, T. F. – Garbelotto, M. – Pellis, L. – Jeger, M. J.: 2012. Impacts of climate change on plant diseases – opinions and trends. *European Journal of Plant Pathology*. 133:295–313.
40. Pálfi X. – Bisztray Gy. D. – Villangó Sz. – Pálfi Z. – Deák T. – Karácsony Z. – Cseke G. - Nagy P. T. – Zsófi Zs.: 2016a. Paraffinolaj hatékonyságának tesztelése szőlőlisztharmat ellen az Egri Borvidéken. *Acta Agraria. Debreceniensis.* 68:73-80.
41. Pálfi, X. - Karácsony, Z. - Kátai, J. - Zsófi, Z.: 2016b. Effects of paraffine oil on leaf and berry mycobiota on two grape varieties. *Acta Agraria Debreceniensis*. 70:61–66.
42. Pálfi, X. - Lovas, M. - Zsófi, Zs. - Kátai, J. - Karácsony, Z. - Váczy, K. Z.: 2021. Paraffin oil induces resistance against powdery mildew in grapevine through salicylic acid signaling. *Pest Management Science*. 77(7):6492.
43. Pálfi, X. – Karácsony, Z. – Kátai, J. – Zsófi Zs.: 2022a. The efficacy of combining paraffin oil with the conventional fungicide treatments against grape powdery mildew in Eger. *Acta Agraria Debreceniensis.* 1:173-180.
44. Pálfi, X. – Karácsony, Z. – Kátai, J. – Zsófi Zs.: 2022b. The Effect of Paraffin Oil Spraying and Powdery Mildew Infection on Leaf Gas Exchange and Yield of Chardonnay and Kékfrankos (*Vitis vinifera* L.) in Hungary. *MDPI Agronomy.* 12(11), 2684.
45. Rantsiou, K. - Giacosa, S. - Pugliese, M. - Englezos, V. - Ferrocino, I. - Río Segade, S. - Monchiero, M. - Gribaudo, I. - Gambino, G. - Gullino, M. L. - Rolle, L.: 2020. Impact of chemical and alternative fungicides applied to grapevine cv Nebbiolo on microbial ecology and chemical-physical grape characteristics at harvest. *Frontiers in Plant Science.* 11, 700.
46. Ribéreau-Gayon, P. – Stonestreet, E.: 1965. Le dosage des anthocyanes dans le vin rouge. *Bulletin de la Société Chimique de France.* 9:2649-2652.

47. Schneider, C. A. - Rasband, W. S. – Eliceiri, K. W.: 2012. NIH Image to ImageJ: 25 years of image analysis. *Nature Methods*. 9:671–675.
48. Singleton, V. L. - Rossi, J. A.: 1965. Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. *American Journal of Enology and Viticulture*. 16:144-158.
49. Tan, B. L. – Sarafis, V. – Beattie, G. A. C. – White, R. – Darley, E. M. -Spooner-Hart, R.: 2005. Localization and movement of mineral oil in plants by fluorescence and confocal microscopy. *Journal of Experimental Botany*. 56:2755–2763.
50. Török, S. – Kállay, M.: 2009. Must- és borvizsgálatok. [In: Török, S. (szerző) és Récsey, A. – Berkes, T. (szerk) Borászok zsebkönyve.]. Mezőgazda Kiadó, Budapest, pp. 165-173.
51. Vielba-Fernández, A. - Polonio, Á. - Ruiz-Jiménez, L. - de Vicente, A. - Pérez-García, A. - Fernández-Ortuño, D.: 2020. *MDPI Microorganisms.* 8, 1431.
52. Wicks, T. J. - Hitch, C. - Campbell, K. - Hall, B.: 1999. Control of grapevine powdery mildew with mineral oil: an assessment of oil concentration and spray volume. *Australian Journal of Grape and Wine Research.* 5(2):61-65.
53. Wicks, T. J. - Hitch, C. J.: 2002. Integration of strobilurins and other fungicides for the control of powdery mildew on grapes. *Australian Journal of Grape and Wine Research.* 8(2):132-139.
54. Yildirim, İ. – Dardeniz, A.: 2010. Effects of alternative spray programs and various combinations of green pruning on powdery mildew [*Uncinula necator* (Schw.) Burr.] in Karasakız (Kuntra) grape cultivar. *Turkish Journal of Agriculture and Forestry.* 3:213-223.

7. PUBLICATIONS ON THE SUBJECT OF THE DISSERTATION



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Registry number:	DEENK/53/2024.PL
Subject:	PhD Publication List

Candidate: Xénia Karácsony-Pálfi
Doctoral School: Kálmán Kerpely Doctoral School
MTMT ID: 10048792

List of publications related to the dissertation

Hungarian scientific articles in Hungarian journals (2)

1. **Pálfi, X.**, Bisztray, G. D., Villangó, S., Pálfi, Z., Deák, T., Karácsony, Z., Cseke, G., Nagy, P. T., Zsófi, Z.: Paraffinolaj hatékonyságának tesztelése szőlőlisztharmat ellen az Egri Borvidéken. *Agrártud. Közl. 68*, 73-80, 2016. ISSN: 1587-1282.

2. **Pálfi, X.**, Bisztray, G. D., Villangó, S., Deák, T., Pálfi, Z., Karácsony, Z., Zsófi, Z.: Orvosi olajkészítmény tesztelése növényvédelmi szempontból két különböző szőlőfajtán. *Borász. Füz. Külön Kiadványa*, 135-137, 2015. ISSN: 1217-9337.

Foreign language scientific articles in Hungarian journals (2)

3. **Pálfi, X.**, Karácsony, Z., Kátai, J., Zsófi, Z.: The efficacy of combining paraffin oil with conventional fungicide treatments against grape powdery mildew in Eger.
   Agrártud. Közl. 1, 173-180, 2022. ISSN: 1587-1282.
   DOI: http://dx.doi.org/10.34101/actaagrar/1/10132

4. **Pálfi, X.**, Karácsony, Z., Kátai, J., Zsófi, Z.: Effects of paraffine oil on leaf and berry mycobiota on two grape varieties.
   Agrártud. Közl. 70, 61-66, 2016. ISSN: 1587-1282.

Foreign language scientific articles in international journals (2)

5. **Pálfi, X.**, Villangó, S., Karácsony, Z., Kátai, J., Zsófi, Z.: The Effect of Paraffin Oil Spraying and Powdery Mildew Infection on Leaf Gas Exchange and Yield of Chardonnay and Kékfrankos (Vitis vinifera L.) in Hungary.
   Agronomy-Basel. 12 (11), 1-15, 2022. EISSN: 2073-4395.
   DOI: http://dx.doi.org/10.3390/agronomy12112684
   IF: 3.7

6. **Pálfi, X.**, Lovas, M., Zsófi, Z., Kátai, J., Karácsony, Z., Váczy, K. Z.: Paraffin oil induces resistance against powdery mildew in grapevine through salicylic acid signaling.
   Pest Manag. Sci. 77 (10), 4539-4544, 2021. ISSN: 1526-498X.
   DOI: http://dx.doi.org/10.1002/ps.6492
   IF: 4.462

Foreign language conference proceedings (1)

7. **Pálfi, X.**, Karácsony, Z., Villangó, S., Zsófi, Z.: Possible mode of action of paraffin oil as a spray agent in the control of powdery mildew of grapevine.
   In: 20th GiESCO International Meeting Mendoza, Universidad Nacional de Cuyo, Mendoza, 970-974, 2017.

Hungarian abstracts (5)

8. **Pálfi, X.**, Karácsony, Z., Villangó, S., Zsófi, Z.: A paraffinolaj hatásmechanizmusának vizsgálata szőlőlisztharmat ellen.
   In: Kutatás-Fejlesztés és Innováció Workshop. Szerk.: Szalontai Helga, Váczy Kálmán Zoltán, EKE Líceum Kiadó, Eger, 12, 2017. ISBN: 9786155621635
9. **Pálfi, X.**, Karácsony, Z., Zsófi, Z., Kátai, J.: Paraffinolaj stressz élettani hatásainak vizsgálata Kékfrankos szőlőfajtán.
   In: XVIII. Szőlészeti és Borászati Konferencia. Szerk.: Pál Károly, Váczy Kálmán Zoltán, EKE Líceum K., Eger, 48, 2017. ISBN: 9786155621376
10. **Pálfi, X.**, Bisztray, G. D., Villangó, S., Deák, T., Pálfi, Z., Karácsony, Z., Zsófi, Z.: Orvosi olajkészítmény tesztelés Chardonnay és Kékfrankos fajtákon két különböző évjáratban.
    In: Szőlő és Klíma Konferencia. Szerk.: Puskás János, [s.n.], Kőszeg, 21-21, 2015.
11. **Pálfi, X.**, Bisztray, G. D., Villangó, S., Deák, T., Pálfi, Z., Karácsony, Z., Zsófi, Z.: Orvosi olajkészítmény tesztelése a szőlő (Vitis vinifera L.) gombakórokozói elleni védekezés céljából.
    In: Tavaszi Szél : Absztraktkötet 2015. Szerk.: Keresztes Gábor, Publio Kiadó, Budapest, 39-39, 2015. ISBN: 9789633977026
12. **Pálfi, X.**, Bisztray, G. D., Villangó, S., Deák, T., Pálfi, Z., Karácsony, Z., Zsófi, Z.: Új szer használata a szőlő (Vitis vinifera L.) növényvédelmi munkálataiban.
    In: PSAK XII. Pro Scientia Aranyérmesek XII. Konferenciája. Szerk.: Harsányi Szabolcs Gergő, Szabó Beáta, EKF Líceum Kiadó, Eger, 154-155, 2015. ISBN: 9786155509421

Foreign language abstracts (1)

13. **Pálfi, X.**, Lovas, M., Karácsony, Z., Kátai, J., Váczy, K. Z., Zsófi, Z.: Mineral oils against powdery mildew: paraffin oil induces resistance in grapevine against Erysiphe necator, and is applicable in disease management.
    Phytopathol. Mediterr. 61 (1), 201, 2022. ISSN: 0031-9465.
    DOI: http://dx.doi.org/10.36253/phyto-13613

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List of other publications

Foreign language scientific articles in international journals (1)

14. **Pálfi, X.**, Karácsony, Z., Csikós, A., Bencsik, O., Szekeres, A., Zsófi, Z., Váczy, K. Z.: The potential use of the culture filtrate of an Aspergillus niger strain in the management of fungal diseases of grapevine.
    J. Cent. Eur. Agric. 21 (4), 839-850, 2020. EISSN: 1332-9049.
    DOI: http://dx.doi.org/10.5513/JCEA01/21.4.2942

Total IF of journals (all publications): 8,162
Total IF of journals (publications related to the dissertation): 8,162

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