

**THE EFFECT OF PROPAMOCARB AND ITS COMBINATIONS WITH THE OTHER ACTIVE INGREDIENTS IN CONTROL OF *PYTHIUM DEBARIANUM* - IN VITRO STUDY**

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**Abstract**

*Pythium debarianum* is the most common causing agent of the damping off disease in the tobacco seedlings production. Propamocarb was a dominant in the suppression of this pathogen, but now, there are fungicides with its different content, but also in combination with other active ingredients.

The aim of this study was to evaluate the efficacy of Propamocarb and its combinations with other active substances, in vitro. These are: Propamocarb 722 g / l (trade name Proplant 722 SL); Propamocarb 530 g / l + Fosetyl-Al 310 g / l (Previcur Energy); Propamocarb 400 g / l + Cymoxanil 50 g / l (Proxanil); Propamocarb 625 g / l + Fluopicolid 62.5 g / l (Infinito). The fungicidal / fungistatic effect (in an appropriate degree) of the tested fungicides was also determined.

The study was performed using Poisoned food technique. The experiment was repeated 3 times, with 5 replications for each variant, with daily measurements of the colony's diameter. The efficacy and the corresponding effect were evaluated on the third but also, on the seventh day.

The fungicide Infinito achieved excellent results in control of *P. debarianum* - 92% effectiveness. The weakest results were achieved by Proxanil, i.e. only 47% effectiveness. Proplant 722 SL and Previcur Energy gave average results (69.00 and 73%, respectively).

No combination of Propamocarb has any in vitro fungicidal action. The fungicide Infinito has a strong fungistatic effect which is only slightly reduced during the test period. Proplant 722 SL and Previcur Energy showed a weak (and very weak) fungistatic effect on this pathogen.

The obtained results would be of particular importance in determining the effect of Propamocarb and / or the most effective combination with other active ingredients in the control of this pathogen. At the same time, it would be in addition to expanding the list of fungicides that can be used to protect tobacco seedlings from the damping off disease caused by *P. debarianum*.

**Keywords:** *Pythium debarianum*, Propamocarb, effectiveness, fungicidal / fungistatic effect

**1. INTRODUCTION**

Healthy and quality seedlings are the basis for successful tobacco production. The protection of tobacco seedlings from diseases is of primary importance for ensuring quality seedling production as well as reducing the problems during the vegetation of the fields.

The most destructive disease that occurs in seedling production is the damping off disease. One of the causing agent of this disease is the pathogenic fungus *Pythium debarianum*.

*Pythium* belongs to the family *Pythiaceae* in the order *Peronosporales* of the class *Oomycetes*-primitive fungi. They can reproduce sexually (in the form of oospores), as well as asexually. The *Pythiaceae* are considered opportunist parasites, preying on weakness and taking advantage of conditions which are not ideal for the plant (Morel Diffusion 2022).

Some species of *Pythium* are found in field soil, sand, pond and stream water and their sediments, and dead roots of previous crops. *Pythium* can cause severe root rot because it has few competitors to check its activity. If *Pythium* infests a cutting bed, large losses occur (Penn State Extension 2017).

Symptoms of *Pythium* infection are variable and can include stem rot, chlorosis, wilting, loss of lower leaves, poor root development and root loss (Chase 2013). Plants are stunted, wilt, and die. The cells of

roots contain round, microscopic, thick-walled spores. *Pythium* also can rot the base of seedlings (Penn State Extension 2017).

In the case of seedlings and young plants, an attack shows up first in the roots, and then displays a glassy rot, chestnut-coloured and wet, at the level of the collar and the infant tuber. The tissue becomes disorganised, loses its firmness and takes on a watery look. The young stalk is weakened and flops, bringing down the whole of the above-ground part of the plant. At this stage it is known as droop, or damping-off disease of seedlings. The disease may go so far as to make the plants literally dissolve and disappear from the surface of the growing medium; so it is also referred to as seedling melt (post-apprition melt). Larger plants can get over an attack thanks to their growth; the symptoms they exhibit are withering or yellowing of a leaf or a few leaves (Morel Diffusion 2022).

Application of a fungicide against *Pythium* is necessary, especially if there is a history of its presence. Certain fungicides, usually systemic fungicides, are said to be 'at risk' to the development of resistance if they are used repeatedly. It is recommended that chemicals at high risk be used sparingly and in rotation or mixed with chemicals with different modes of actions (Penn State Extension 2017).

Aliette Signature and Banol are effective fungicides for all *Pythium* diseases (*Pythium* Blight, *Pythium* Root Rot, *Pythium* Root Dysfunction). There is not resistance up to 20 years (Bayer 2022).

Over the past years, there are few new fungicides. The most effective product were those that include a.i. Etridiazole (Terrazole, Truban and Banrot) - effective to many *Pythium* species; a.i. Mefenoxam (Subdue and MAXX) which were not develop resistance although both have been used many yaers. Also, the fungicides with phosphonates as active ingredient (Aliette a.i. Fosetyl-Al) and Alude (a.i. Mono and di-basic sodium, potassium and ammonium phosphites).

One of the newest active ingredient for control of Oomycetes (include *Pythium*, *Phytophthora* and the downy mildew is a.i. fluopicolide (fungicideAdorn - fluopicolide 39.5%).

Strobilurins provide varying levels of control The newest strobilurin is pyraclostrobin (the fungicide Empress) which was found to be effective to *Pythium*, *Phytophthora*, as well as *Fusarium* and *Rhizoctonia* in ornamentals (Chase 2013)

There are studies of the fungicides *in vitro* (as the first and quickest way to screen the effect) as well as a guide for their inclusion *in vivo*.

Prasad et al (2014) have investigated ten fungicides on *Pythium debarianum* (and *Rhizoctonia solani*) causing the damping off diseases in tomato. There are active ingredients (and combinations) which shown 100 % inhibition of radial development of the *P. debarianum* - tebuconazole and trifloxystrobin, carbendazin and captan+hexaconazole. The other active ingredients also have shown a high degree of effectiveness.

Mihajlovic et al (2013) investigated several active ingredients against *Pythium aphanidermatum* (propamocarb hydrochloride, fosetyl-Al, mancozeb, azoxystrobin, mefenoxan and tea tree oil. They also made a parallel of results between *in vitro* and greenhouse assays.

From previous experiences, the active substance propamocarb has a dominant place in the suppression of this pathogen. -in our area. Previcur is a standard part of the protective program used by many commercial nurseries to prevent the damping-off of ornamental plants caused by *Pythium* spp. (Bayer 2022 b). Propamocarb is selectively active against Oomycetes. The fungicide is xylem-mobile and can be used as drench, soil incorporation, dip or foliar spray (Kilian and Steiner 2003).

But now, there are some fungicides in which this active substance is in different content, but also in combination with other active substances for example: Fosetyl-Al, Fluopicolide and Cymoxanil.

Previcur Energy is a systemic multi-site fungicide for the contorl of downy mildew and damping off caused by *Pythium* spp. for lettuce, broccoli/calabrese, cauliflower, cabbage, tomato, melon and cucumber seedlings (Bayer 2022). The a.i. Fosetyl-Al has an antifungal effect on Oomycetes. Its efficacy is increased by an indirect effect of enhancing natural plant defenses via release of ethylphosphonate inside the plant tissue, triggering salicylic acid, jasmonic acid and ethylene pathways (Pétré et al 2015).

It is a systemic fungicide which is rapidly absorbed through the plant leaves or roots with translocation both up and down inside the plant. The mode of action is multi-site (Texicon Agriventures corporation 2022).

Fluopicolide has a mode of action which differs from other available fungicides used to control Oomycetes and it can inhibit the growth of strains that are resistant to phenylamides, strobilurin, dimethomorph and iprovalicarb. It has some systemic activity as it moves through the xylem towards the tips of stems, but does not get transported to the roots (Anonymous 2022).

Cymoxanil is a penetrant fungicide with protectant and curative activity. Its post-infection activity stops the development of the fungus during the incubation. Local systemic action: it is able to penetrate the crop leaf and improves the effectiveness of companion fungicides. Therefore, it is used in two-, or in three-way mixtures with protectants and/or systemic fungicides. (Oxon 2022).

The aim of this study was to investigate the efficacy of some fungicides based on the active substance propamocarb and its combinations with other active substances - *in vitro*.

The *in vitro* assay is the first and quickest way was to screen the effectiveness of fungicides and their active ingredients. We can estimate the direct effect on the pathogen, free from environmental influences. This way of study provides significant knowledge of the mode of action and effectiveness of the fungicide / active ingredients, before the application of a commercial product in the field.

## 2. MATERIAL AND METHODS

### 2.1. Poisoned food technique

Pure culture of the fungus is isolated from infected tobacco seedlings by standard laboratory method on the medium potato dextrose agar (PDA).

The same medium - PDA was used for the tests, with the addition of fungicides. An overview of the tested fungicides is given in Table 1.

The medium with the fungicide was poured into petri boxes 110 mm in diameter, and then sown with a fragment of pure fungus culture (about 5 mm). Sown petri boxes with fungicide-free substrate served as a check.

The Petri boxes were incubated for 10 days at a temperature of 25 ° C by daily measuring the diameter of the colony on both opposite sides of the Petrie box. The experiment was set 3 times. The results for the three replications as well as the average values are shown by tables.

<b>Fungicide (Variant)</b>	<b>Active ingredient and its quantity</b>	<b>Concentration in which the preparation is used (%)</b>
<b>Proplant 722 SL</b>	Propamocarb 722 g/l	0,15
<b>Previcur Energy</b>	Propamocarb 530 g/l + Fosetyl-Al 310 g/l	0,15
<b>Proxanil</b>	Propamocarb 400 g/l + Cymoxanil 50 g/l	0,15
<b>Infinito</b>	Propamocarb 625 g/l + Fluopicolid 62,5 g/l	0,15

**Table 1.** Tested fungicides

## 2.2 Estimations

The effect of fungicides was monitored daily, in parallel with the development of control. *P. debarianum* has a very fast development and the maximum development of the air mycelium was achieved on the third day, Therefore, when calculating the efficiency, the value of the third day was taken.

The effectiveness of fungicides in control of *P. debarianum* is calculated according to the average value of the diameter of the colonies (in the appropriate variant, i.e. fungicide and the check) according to the formula of Singh et al (2015).

In order to determine the fungistatic or fungicidal effect of the examined preparations, after seven days of monitoring the development of the colony, a small fragment of the developed colony was transplanted into pure culture, without adding fungicide and the development was monitored. The effect evaluation was performed again (on the 3rd and 7th day), presented according to a scale:

- 0 - no development of the fungus after sowing of pure culture  
(fungicidal action)
- 1-develops outside the disc up to 10 mm (strong fungistatic effect)
- 2-develops up to 20 mm (moderate fungistatic effect)
- 3-develops up to 55 mm (weak fungistatic effect)
- 4- develops from 56 to 110 mm (very weak fungistatic effect)

## 3. RESULTS

The fungus *P. debarianum* has the fastest initial development in the check, ie on a medium without fungicide. The colony continues to develop intensively and already on the third day the petri dishes are completely filled (Table 2).

In Infinito fungicide there is no colony development until the fourth day, when initial development is observed and by the end of the incubation it has a very small diameter.

The first three fungicides show no significant difference in initial development. But on the third day the colony has the largest diameter of medium with the fungicide Proxanil. The difference is almost double that of the Proplant 722 SL and Previcur Energy. At the end of the incubation, it is even more pronounced.

day	1	2	3	4	5	6	7
<b>Fungicide</b>	<b>Diameter in mm</b>						
<b>Proplant 722 SL</b>	32,80	33,80	35,20	37,20	38,40	39,40	40,20
<b>Previcur Energy</b>	29,20	33,00	35,60	35,80	37,00	37,60	38,00
<b>Proxanil</b>	29,40	42,60	67,20	80,80	88,40	94,80	102,00
<b>Infinito</b>	-	-	-	+ -	7,00	10,00	10,40
<b>Check ø</b>	45,20	109,86	110,00	110,00	110,00	110,00	110,00

**Table 2.** Development of the colony of *P. debarianum* (1<sup>st</sup> replication)

In the second replication the situation is the same, except that the initial development of the colony in Infinito begins on the second day and has reached a slightly larger diameter by the seventh day. The greatest development was in Proxanil (Table 3).

day	1	2	3	4	5	6	7
<b>Fungicide</b>	<b>Diameter in mm</b>						
<b>Proplant 722 SL</b>	25,40	29,80	32,80	34,40	35,80	36,80	39,20
<b>Previcur Energy</b>	25,20	28,00	29,60	30,00	30,80	32,40	34,20
<b>Proxanil</b>	32,80	50,40	68,20	80,80	89,40	92,00	97,80
<b>Infinito</b>	-	+ -	6,00	10,00	14,60	16,80	16,80
<b>Check ø</b>	71,60	71,60	110,00	110,00	110,00	110,00	110,00

**Table 3.** Development of the colony of *P. debarianum* (2<sup>nd</sup> replication)

The fungicide Proplant 722 SL in the first two replications gives almost the same results as Previcur Energy. But in the third, the colony has a greater development, or, this fungicide showed poorer results, which is observed from the initial development until the end of the incubation.

The best colony development was again found in Proxanil, and the weakest in Infinito (Table 4).

day	1	2	3	4	5	6	7
<b>Fungicide</b>	<b>Diameter in mm</b>						
<b>Proplant 722 SL</b>	30,40	32,20	34,00	35,40	37,20	38,40	40,40
<b>Previcur Energy</b>	16,40	21,00	21,60	22,80	23,80	25,20	26,40
<b>Proxanil</b>	25,00	45,40	67,20	82,00	90,40	96,60	102,00
<b>Infinito</b>	-	+ -	6,00	10,00	11,60	13,40	15,80
<b>Check ø</b>	61,60	110,00	110,00	110,00	110,00	110,00	110,00

**Table 4.** Development of the colony of *P. debarianum* (3<sup>th</sup> replication)

The influence of fungicides on colony development in all three replications is confirmed by Table 5 (an average value of replications) and Fig. 1-5. According to the results, the colony on the third day achieves the greatest development in Proxanil (57,33 mm) and the weakest in Infinito (only 8,00 mm), i.e. only a small initial development. Between Proplant 722 SL and Previcur Energy, the first has weaker results. Results are the same on the 7<sup>th</sup> day, too (Table 5).

day	1	2	3	4	5	6	7
	<b>Diameter in mm</b>						
<b>Fungicide</b>							
<b>Proplant 722 SL</b>	29,53	31,93	34,00	35,66	37,20	38,20	39,93
<b>Previcur Energy</b>	23,60	27,33	28,93	29,53	30,53	31,73	32,86
<b>Proxanil</b>	29,06	45,33	57,33	72,46	84,00	91,80	97,93
<b>Infinito</b>	-	+ -	8,00	8,66	11,06	13,40	14,33
<b>Check ø</b>	45,80	109,86	110,00	110,00	110,00	110,00	110,00

Table 5. Development of the colony of *P. debarianum* (the average value)

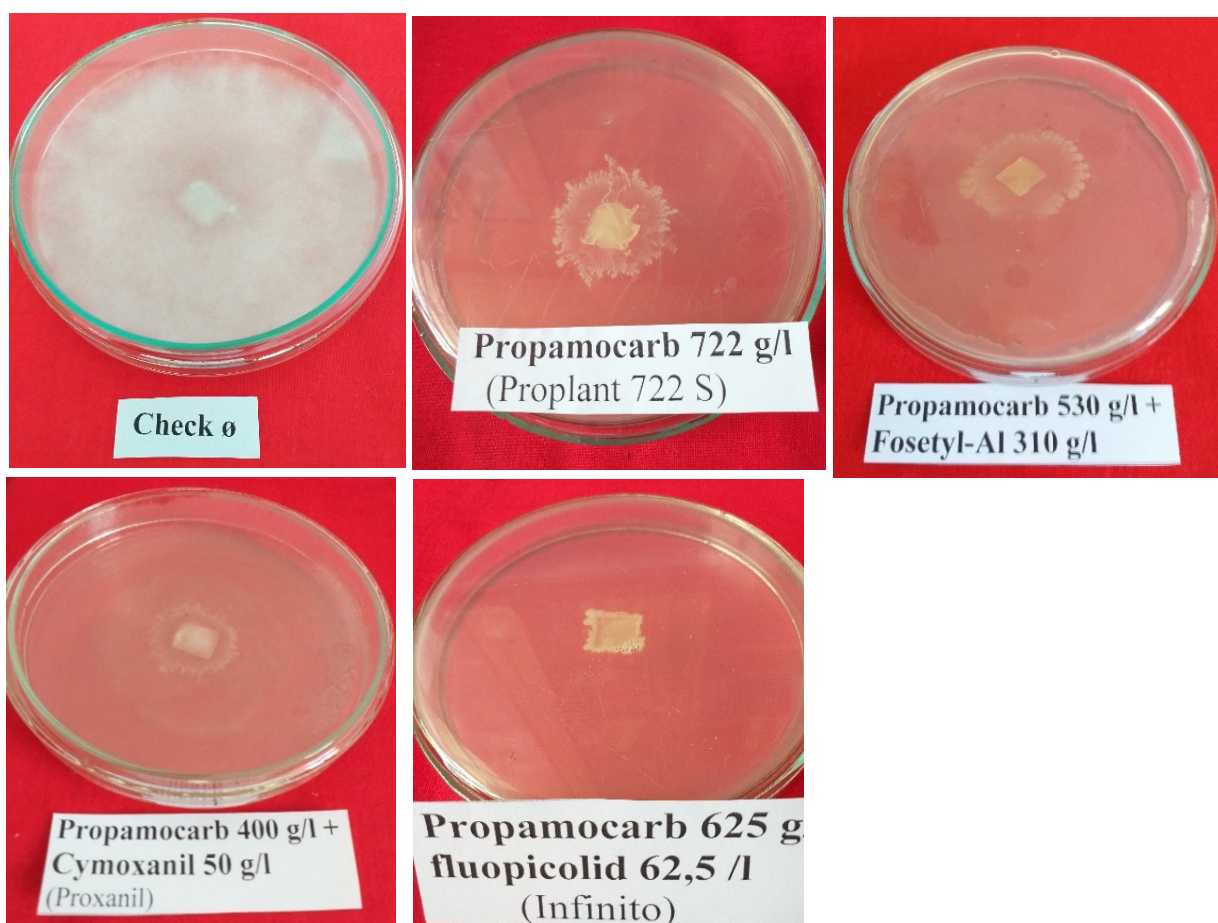


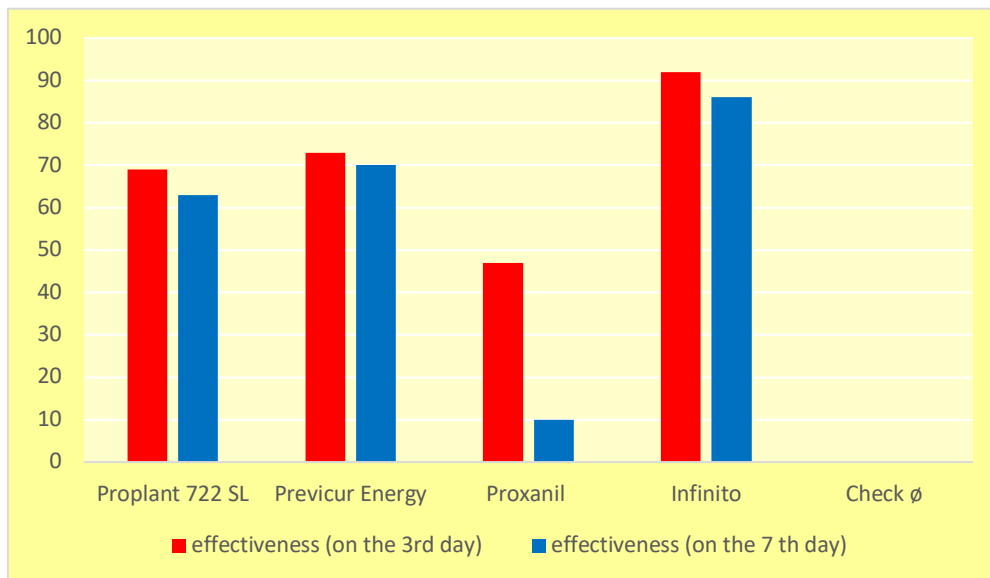
Fig 1-5. Development of a colony of *P. debarianum* depending on the tested fungicides (3<sup>th</sup> day)

The efficacy of the tested fungicides in control of the pathogen *P. debarianum* is presented in Table 6 and Fig 6. The value is ranged from 47.00% in Proxanil (Propamocarb 400 g / l + Cymoxanil 50 g / l) to 92% in Infinito (Propamocarb 625 g / l + Fluopicolid 62.5 g / l). The other two fungicides - Proplant 722 SL (Propamocarb 722 g / l) and Previcur Energy (Propamocarb 632 g / l + Fosetyl-Al 310 g / l) showed similar results (69,00 and 73,00%). Therefore, Infinito (Propamocarb 625 g / l + Fluopicolid 62.5 g / l) achieved the best result in control of *P. debarianum*.

Passing the incubation period, the expected reduction in efficiency has been determined. There is a very small decrease in effectiveness after 7 days in the fungicide Previcur Energy (only 3%) while the largest decrease is observed in the fungicide with the lowest effectiveness - Proxanil (37%).

Variant	Effectiveness (%)	
	3 <sup>th</sup> day	7 <sup>th</sup> day
Check Ø	-	-
Proplant 722 SL	69,00	63,00
Previcur Energy	73,00	70,00
Proxanil	47,00	10,00
Infinito	92,00	86,00

**Table 6.** Effectiveness of the fungicides



**Fig. 6.** Effectiveness of the fungicides in control of *P. debarianum*

Regarding the determination of the action of the examined fungicides, more precisely whether they have a fungicidal action or a fungistatic (and to what extent) it was determined that none of the fungicides has a fungicidal action. The expectation that there will be no development in Revurur Energy is not fulfilled because the fungus starts to develop intensively from the second day (Table 7, Fig 8, 9, 10).

In Infinito, the fungus also begins to grow.

Passing the incubation period, a strong fungistatic effect in Infinito is only slightly reduced (from strong to moderate).

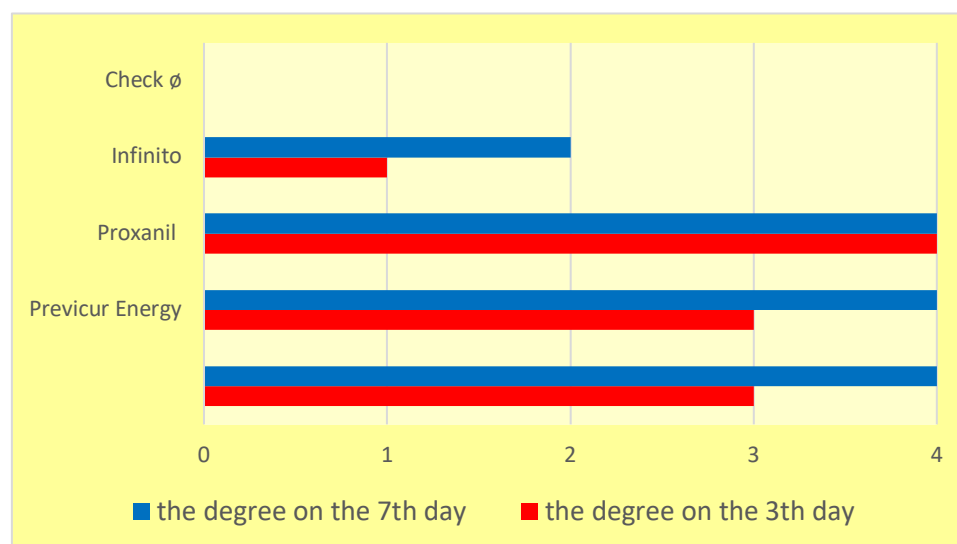
The fungicides Proplant 722 SL and Previcur Energy showed a weak fungistatic effect which has turned to very weak fungistatic effect. and the colony has been developing intensively since the third day, ie these fungicides have a very weak fungistatic effect on the pathogen *P. debarianum*. The weakest effect on the pathogen *P. debarianum* has the fungicide Proxanil (Table 8, Fig 7).

day	1	2	3	4	5	6	7
<b>Fungicide</b>	<b>Diameter in mm</b>						
<b>Proplant 722 SL</b>	30,66	40,33	<b>53,33</b>	82,60	100,06	102,33	110,00
<b>Previcur Energy</b>	-	24,00	<b>53,33</b>	76,33	94,33	101,66	110,00
<b>Proxanil</b>	26,66	55,66	<b>83,07</b>	100,00	105,00	110,00	110,00
<b>Infinito</b>	-	- +	<b>8,00</b>	8,80	11,20	13,10	16,60
<b>Check ø</b>	66,05	109,93	<b>110,00</b>	110,00	110,00	110,00	110,00

**Table 7.** Fungicidal / fungistatic effect of the investigated fungicides on the *P. debarianum*

<b>Variant</b>	<b>The degree of the fungicidal / fungistatic effect</b>	
	3 <sup>th</sup> day	7 <sup>th</sup> day
<b>Proplant 722 SL</b>	3	4
<b>Previcur Energy</b>	3	4
<b>Proxanil</b>	4	4
<b>Infinito</b>	1	2
<b>Check ø</b>	-	-

**Table 8.** The degree of fungicidal / fungistatic effect of the tested fungicides on *P. debarianum*

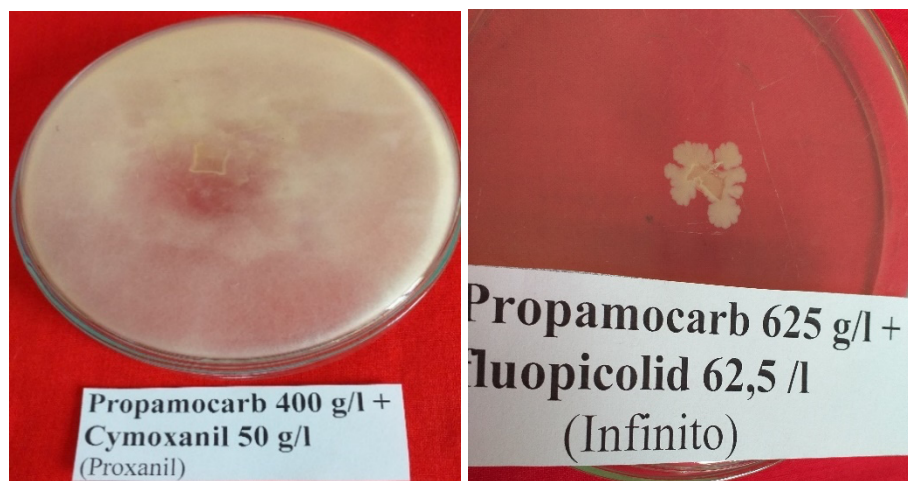


**Fig. 7.** Change in the degree of fungicidal / fungistatic effect





**Fig 8.** Fungicidal / fungistatic effect - development of *P. debarianum* (3<sup>th</sup> day)



**Fig 9, 10.** Fungicidal / fungistatic effect - development of *P. debarianum* (7<sup>th</sup> day)

#### 4. DISCUSSION

All tested fungicides, although containing the active substance Propamocarb (but with different content), gave different results. Therefore, the content of Propamocarb has the effect.

Propamocarb is selectively active against Oomycetes (Kilian and Steiner 2003). The action of Propamocarb is related to cell membrane permeability, causing an efflux of cell compounds. Leakage out of the mycelium can be inhibited by the addition of sterols. Production of an unusual fatty acid was later reported in mycelial cells of Oomycetes (Kilian and Steiner 2003, Extension 2022).

The fungicides Proplant 722 SL (Propamocarb 722 g / l) and Previcur Energy (Propamocarb 530 g / l + Fosetyl-Al 310 g / l) do not differ much in content (722 and 632 g / l) and have similar results, in contrast of Proxanil (with 400 g / l Propamocarb). That is, preparations with a lower content are less effective.

Other active ingredients of the tested fungicides also have an effect on the effectiveness of the preparation. It does not depend on their content, but on the nature of that active substance, ie the reducing effect on the pathogen.

An antifungal effect of Cymoxanil on Oomycetes as it acts by preventing sporulation (AgChem Access 2022). The fungicide inhibits nucleic acid and protein synthesis (Kilian and Steiner 2003).

Fosetyl-Al has a direct effect on the inhibition of the growth of mycelium by competing for the assimilation of phosphates (Petré et al 2015). It inhibits spore germination and penetration of pathogen into the plant by blocking its mycelial growth and spore production. The mode of action is multi-site (Texicon Agriventures corporation 2022).

On the contrary, the mode of action in cyano-acetamide oximes (cymoxanil) is unknown but not multi-site (Gisi and Sierotzki 2008, Extension 2022). Thus, the colony reached the largest diameter on the medium with the fungicide Proxanil (Propamocarb 400 g / l + Cymoxanil 50 g / l). The difference is almost double that of Proplant 722 SL and Previcur Energy and by the end of the incubation, it is even more pronounced. This means that Proxanil has the weakest reducing effect. This is not according to Kilian and Steiner (2003) that mycelial growth is more sensitive to Cymoxanil than early growth phases, including the release of zoospores from sporangia and their germination. Cymoxanil is a systemic product with protectant and curative activity against Oomycetes.

The best reducing effect on the development of *P. debarianum* is found in the fungicide Infinito - there is no development of the colony until the fourth day, when the initial development is observed and by the end of the incubation it has a very small diameter. This result is according to these of Wu et al (2020) that Fluopicolide had strong inhibitory activity on the mycelium growth *in vitro*. Also, the inhibitory activity of Fluopicolide on zoospore release was significant. The lysing zoospores increased with higher concentrations of this a.i. At a concentration of 10 mg/ml, all zoospores lost motility.

The precise mode of action of Fluopicolide is not known, but it is thought to act by affecting spectrin-like proteins in the cytoskeleton of oomycetes. It affects the motility of zoospores, the germination of cysts, the growth of the mycelium and sporulation. (Anonymous 2022). The intracellular substances of hyphae or zoospores are leaked after Fluopicolide treatment and the potential target protein of Fluopicolide is speculated to be a spectrin-like protein but this remains unclear (Wu et al 2020).

According to EPA (2007), even that Fluopicolide's mode of action has not been determined, however, it is a mode of action unlike the known modes of action of other registered fungicides.

The fungicide Infinito (Propamocarb 625 g/l + Fluopicolide 62,5 g/l) achieved really great results - 92%. Thus, Fluopicolide (as one of the new fungicidal active substances), besides Procamocarb, confirms its importance.

According to the effectiveness of the tested fungicides on the third day (when achieving full development of the colony in control), the fungicide Infinito achieved really great results - 92%. Thus, Fluopicolide (as one of the new fungicidal active substances), confirms its importance.

Fluopicolide is a fungicide used in agriculture to control diseases caused by oomycetes such as late blight of potato. It is classed as an acylpicolide and its chemical name is 2,6-dichloro-*N*-{[3-chloro-5-(trifluoromethyl)pyridin-2-yl]methyl}benzamide (Anonymous 2022).

Fluopicolide controls a wide range of Oomycete (Phycomycete) diseases including downy mildews (Plasmopara, Pseudoperonospora, Peronospora, Bremia), late blight (Phytophthora), and some Pythium species (EPA 2007).

Fluopicolide is a benzamide plant fungicide discovered and patented by AgrEvo UK Limited (since 2002 being a part of Bayer Crop Science) in 1999. It has been widely used for controlling a variety of oomycetes such as *Plasmopara viticola*, *Phytophthora infestans*, *Pseudoperonospora cubensis*, and *Bremia lactucae*. It provides excellent efficacy on several pathogen developmental stages such as mycelial growth, sporangial production, zoospore release and motility, and cyst germination (Wu et al 2020).

Between Proplant 722 SL and Previcur Energy, the first has poorer results, despite the results of Kilian and Steiner (2003) according to which Aluminium tris-*O*-ethylphosphonate (Fosetyl-Al) has a low degree of *in vitro* inhibition that has been interpreted as an indication that antifungal activity is indirect and host defense reactions are stimulated. The application enhances the accumulation of phenols and antifungal stilbenes and flavonoids, resulting in a reduced symptom expression (Kilian and Steiner 2003).

But, Propamocarb although has little or no inhibition of mycelial growth nor any control of mycelial infection (in *Phytophthora nicotiana*), it consistently suppressed all other growth stages assessed *in vitro* and reduced zoospore infection. These results have several significant practical implications.

The weakest results were achieved by Proxanil, ie only 47% efficiency. Cymoxanil is a penetrant fungicide with protectant and curative activity. Its post-infection activity stops the development of the fungus during the incubation. Local systemic action: it is able to penetrate the crop leaf and improves the effectiveness of companion fungicides (alone, it has a very short period of activity), especially during periods of intensive disease pressure (Oxon 2022).

The fungicides Proplant 722 SL and Previcur Energy gave not very great results in the suppression of the pathogen *P. debarianum* (69.00 and 73.00%) in *in vitro* tests. It differs than *in vivo* studies because Fosetyl-AI has enhanced antifungal effect on Oomycetes by an indirect effect of enhancing natural plant defenses via release of ethylphosphonate inside the plant tissue, triggering salicylic acid, jasmonic acid and ethylene pathways (Petre et al 2015).

Also, although the Propamocarb has little or no inhibition of mycelial growth nor any control of mycelial infection (in *Phytophthora nicotiana*), it consistently suppressed all other growth stages assessed *in vitro* and reduced zoospore infection. These results have several significant practical implications (Wu et al 2020).

Efficiency has a lower value over the incubation period than expected. There is a very small decrease in effectiveness after 7 days with the fungicide Previcur Energy (only 3%) while the largest decrease (37%) is observed with the fungicide with the lowest efficiency - Proxanil (Fig 6).

Regarding the determination of the action of the examined fungicides, more precisely whether they have a fungicidal action or a fungistatic (and to what extent) it was determined that none of the fungicides has a fungicidal action. The expectation that there will be no development in Revurur Energy is not fulfilled because immediately, from the second day, the fungus starts to develop intensively (Table 6).

The situation is similar with Infinito. However, at the end of the follow-up interval, in this variant the colony has a very small diameter value, unlike the others. This means that this fungicide has a strong fungistatic effect on the pathogen, which is only slightly reduced (from strong to moderate) with the passage of the incubation period. The strongly reducing effect has a negative impact on the "reconstruction" of the colony.

In the variants / fungicides Proplant 722 SL and Previcur Energy the colony develops intensively from the third day. Therefore, they showed a weak to very weak fungistatic effect on the pathogen *P. debarianum*.

The weakest effect has the fungicide Proxanil, which corresponds to the small reducing effect of the fungicide on the development of the colony. This can be explained by the fact that Cymoxanil, as it is local acting however, it is not the most persistent of fungicides (so it is usually applied in combination with a protective translocatable systemic fungicide). Cymoxanil acts by preventing sporulation (AgChem Access 2022).

Cymoxanil alone, has a very short period of activity: two days at most. Therefore, it is used in two-or in three-way mixtures with protectants and / or systemic fungicides (Oxon 2022).

## 5. CONCLUSIONS

- The tested fungicides showed a difference in effectiveness in control of the pathogenic fungus *P. debarianum*.
- The effectiveness ranges from 47.00% to 92%.
- The weakest efficacy was shown by the fungicide Proxanil (Propamocarb 400 g / l + Cymoxanil 50 g / l).

- Fungicide Infinito (Propamocarb 625 g / l + Fluopicolid 62.5 g / l) showed the highest efficiency (92%)
- The content of the active substance Propamocarb has an effect on the effectiveness of the preparations. Fungicides with greater its content - Proplant 722 SL, Infinito and Previcur Energy showed better results compared to Proxanil.
- Other active ingredients of the tested fungicides also affect the effectiveness of the fungicides. It does not depend on their content, but on the nature of that active substance, ie the reducing effect on the pathogen.
- When applying fungicides with the active substance Propamocarb to control the pathogen *P. debarianum*, it is recommended to choose the preparation correctly, depending on its content and the combination with another active substance.
- The results of these studies contribute to increasing the list of fungicides with a.m. substance Propamocarb and inclusion of new ie combinations with other active substances for control of *P. debarianum*.

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