

Original scientific paper

## CONTROL EFFICACY OF A NOVEL SUCCINATE DEHYDROGENASE INHIBITOR FUNGICIDE, PENTHIOPYRAD, AGAINST *BOTRYTIS CINEREA*

Katerina Bandjo Oreshkovikj<sup>1\*</sup>, Biljana Kuzmanovska<sup>2</sup>, Rade Rusevski<sup>2</sup>, Mihajlo Risteski<sup>3</sup>

<sup>1</sup>Institute of Agriculture, Ss. Cyril and Methodius University in Skopje, Republic of North Macedonia

<sup>2</sup>Faculty of Agricultural Sciences and Food-Skopje, Ss. Cyril and Methodius University in Skopje, Republic of North Macedonia

<sup>3</sup>Faculty of Forestry “Hans Em”, Ss. Cyril and Methodius University in Skopje, Republic of North Macedonia

\*corresponding author: kbandzo@yahoo.com

### ABSTRACT

The goal of this experiment was to study the efficiency of the novel active ingredient penthiopyrad (Fontelis SC) in controlling of gray mold disease in tomato (*Botrytis cinerea*), a very destructive disease in our country. As a standard, the well-known active ingredient fenhexamid (Teldor) was used. The experiment was conducted during 2019 in two production regions of tomato in North Macedonia, Bogdanci and Dojran. In the untreated variant in the region of Dojran, a very high level of disease severity (73.2% in the old leaves, 84.8% on the stem) was observed, leading to destructive damages in the tomato production. In the region of Bogdanci, significantly lower level of disease severity was observed in the untreated variant (14% on the leaves, 9.2% on the stem). The tested novel fungicide Fontelis SC demonstrated high level of efficacy. In the region of Dojran, the fungicide Fontelis SC provided efficacy degree of 81.9 to 100%, showing significantly better results than the standard Teldor (29.5-86.6%). In the region of Bogdanci, the new fungicide Fontelis SC proved again to be more effective than the standard Teldor. Fontelis SC demonstrated maximum efficacy of 100% on all plant parts, while for the standard Teldor efficacy performance ranged from 86.9% to 88.5%. These results emphasize the need in expanding the range of novel fungicides, in order to provide better results in gray mold disease management.

**Key words:** *Botrytis cinerea*, fenhexamid, fungicide resistance, penthiopyrad.

### INTRODUCTION

*Botrytis cinerea* Pers.: Fr. (teleomorph *Botryotinia fuckeliana* (de Bary) Whetzel) is one of the most important diseases in commercial greenhouse production of vegetables all over the world, especially in tomato and cucumber production. The pathogen can be found in every part of the tomato plant, including leaves, stems, flowers, fruits and petioles (Yildiz et al., 2007). In North Macedonia, grey mold of tomato is the leading disease in greenhouse production of tomato, with yield losses that can surpass 70% (Kuzmanovska et al., 2012).

Conventional approaches for managing gray mold disease in tomatoes primarily rely on routine fungicide applications. However, the Fungicide Resistance Action Committee (FRAC) classifies *Botrytis cinerea* as a high-risk pathogen due to its ability to develop resistance to fungicides. As a result, effective chemical control of gray mold has been increasingly challenged by the emergence of resistant isolates. Cases of resistance to multiple

site-specific fungicides in *B. cinerea* have been reported in various countries worldwide (Ziogas et al., 2003; Bardas et al., 2010; Weber & Hahn, 2011; Amiri et al., 2013; Leroch et al. 2013; Fernandez-Ortuno et al., 2014).

Penthiopyrad [1-methyl-N-[2-(4-methylpentan-2-yl)-3-thienyl]-3-(trifluoromethyl)pyrazole-4-carboxamide], developed by Mitsui Chemical, Inc., is a novel fungicide classified as a succinate dehydrogenase inhibitor (SDHI) (Liang et al., 2020). Research by Yanase et al. (2013) demonstrated that penthiopyrad exhibited exceptionally high efficacy against various plant pathogens. According to their findings, penthiopyrad had a broad fungicidal spectrum, making it effective in controlling multiple diseases across a wide range of fruits and vegetables. These included gray mold, powdery mildew, tomato leaf mold, cucumber *Corynespora* leaf spot, rust, southern blight, apple and pear scab, apple blossom blight and brown rot in peaches and cherries.

Additionally, penthiopyrad operated through a unique mode of action, distinguishing it from many commercial fungicides used for these diseases. No cross-resistance had been observed with benzimidazole, dicarboximide, anilinopyrimidine, DMI, or strobilurin fungicides. Furthermore, penthiopyrad exhibited low acute toxicity in mammals and birds, making it a relatively safer option for disease management (Yanase et al., 2013).

Because of the positive characteristics of this novel active ingredient and the tendency of gray mold in creating resistant isolates, this study aimed to explore the potential of controlling *Botrytis cinerea* using other innovative active ingredients, such as penthiopyrad. The goal was to expand the range of effective fungicides available for tomato production in North Macedonia.

## MATERIAL AND METHODS

The research was conducted during the vegetative season in 2022 on tomato plants grown in greenhouses, in two regions: Dojran and Bogdanci. In the region of Dojran, the trial was conducted on the tomato hybrid Kannu, grown on alluvial soil, while in the region of Bogdanci, the test was performed on the tomato hybrid Mamston, on hydroponics. The treatments were preventive, starting from the stage of first inflorescences visible until the stage of full maturity (BBCH 51-89). Each trial consisted of three different variants, the first with the novel fungicide FONTELIS SC (penthiopyrad), the second with the standard TELDOR (fenhexamid), and the third was an untreated control (Table 1). The experiment was set in a randomized block design with three replications. In both regions, each replicate consisted of 30 tomato plants, on an experimental block of 10 m<sup>2</sup>. The application of pesticides was foliar, performed with the use of a hand-compression sprayer with a volume of 10 L.

Table 1. Tested variants represented in the regions of Dojran and Bogdanci in 2022

No.	Fungicides	Active substance	Content of active substance	Producer	Rate of application
1.	<b>FONTELIS SC</b>	Penthiopyrad	200 g/L	DuPont, Switzerland	2 L/ha
2.	<b>TELDOR</b>	Fenhexamid	500 g/L	Bayer, Germany	1.5 L/ha
3.	<b>CONTROL</b>		Untreated		

A total of 3 foliar applications with the tested fungicides were performed in an interval of 7 days, on the following dates: I treatment - 30.11.2022, II treatment - 07.12.2022 and III treatment - 14.12.2022. Efficacy assessment of the tested fungicides was performed twice during the trials. First assessment was performed 7 days after the second treatment

(14.12.2022) and the second evaluation was completed after the third (last) treatment, on 20.12.2022.

In both tested regions, disease incidence was calculated on infected leaves, stems and fruits. Stem and fruit disease incidences were expressed as the percentage of infected stems (proportion of infected stems in relation to the total number of examined plants) and as the percentage of infected fruits (proportion of fruits that are infected in relation to the total number of fruits examined in 30 plants). The leaves infection was assessed following the 0-5 scale (percentage of diseased area or sporulation intensity) based on Mouria et al. (2015): 0 - 0 decayed leaf area (DLA), 1 - 1-5 DLA, 2 - 6-15 DLA, 3 - 16-50 DLA, 4 - 51-95 DLA and 5 - 100 DLA.

The disease severity index was, after that, calculated according to the Townsend-Heuberger's formula (Townsend-Heuberger, 1943). The efficacy of the tested fungicides was calculated according to the Abbott's formula (Abbott, 1925).

## RESULTS AND DISCUSSION

During the vegetation in 2022, in the region of Bogdanci in the untreated control variant, disease symptoms were observed on the leaves and stems of the tomato plants, while signs of the disease were not detected on the fruits. The measured disease incidence and disease severity of gray mold on the examined leaves was 70 % and 14 %, respectively. On the stems, the disease incidence was 50 %, while disease severity 9.2 %. In these disease conditions, the observed efficacy of the standard fungicide TELDOR was below 90% on the leaves and stems of the inspected tomato plants, while the novel fungicide FONTELIS SC managed to provide total plant protection of the disease (100 % efficacy) (Table 2).

Table 2. Efficacy of the fungicide FONTELIS SC in controlling of *Botrytis cinerea* on tomato in Bogdanci region after the second assessment (20.12.2022)

<b>CONTROL</b>	<b>Leaves</b>	<b>Stem</b>	<b>Fruits</b>
Disease severity (%)	14	9.2	0
Disease incidence (%)	70	50	0
<b>TELDOR</b>	<b>Leaves</b>	<b>Stem</b>	<b>Fruits</b>
Disease severity (%)	1.6	1.2	0
Disease incidence (%)	20	30	0
Efficacy (%)	88.5	86.9	/
<b>FONTELIS SC</b>	<b>Leaves</b>	<b>Stem</b>	<b>Fruits</b>
Disease severity (%)	0	0	0
Disease incidence (%)	0	0	0
Efficacy (%)	100	100	/

In the region of Dojran, more severe disease conditions were observed. The disease severity detected in the untreated control variant on the leaves was 73.2 % on the old leaves and 9.6 % on the young ones. The estimated disease severity on the stems was 84.8 %. Furthermore, unlike Bogdanci, in the region of Dojran disease symptoms were observed also on the tomato fruits. The measured disease severity was 6 %. In these destructive disease conditions, the standard fungicide TELDOR performed very inconsistent efficacy, varying from 29.5 % on the old leaves, to 86.6 % on the fruits. The efficacy of FONTELIS SC was far more stable and effective, providing protection to the tomato plants from 81.9 % for the old leaves to a total of 100 % for the fruits (Table 3). During the trials, no negative effects were detected during the usage of the fungicides on the tested tomato plants in the proposed rates.

Table 3. Efficacy of the fungicide FONTELIS SC in controlling of *Botrytis cinerea* on tomato in Dojran region after the second assessment (20.12.2022)

<b>CONTROL</b>	<b>Old</b>	<b>Young leaves</b>	<b>Stem</b>	<b>Fruits</b>
<b>leaves</b>				
Disease severity (%)	73.2	9.6	84.8	6
<b>TELDOR</b>	<b>Old</b>	<b>Young leaves</b>	<b>Stem</b>	<b>Fruits</b>
<b>leaves</b>				
Disease severity (%)	51.6	4.8	50.8	0.8
Efficacy (%)	29.5	50	40	86.6
<b>FONTELIS SC</b>	<b>Old</b>	<b>Young leaves</b>	<b>Stem</b>	<b>Fruits</b>
<b>leaves</b>				
Disease severity (%)	13.2	0	2.8	0
Efficacy (%)	81.9	100	96.6	100

After the assessment of the fungicide efficacy of both TELDOR and FONTELIS SC, a noticeable difference between the efficacy of the fungicides was observed. That was due to the capability of *B. cinerea* in creating resistant isolates to various fungicidal active ingredients in relatively short period of time (FRAC). A recent study conducted in Spain reported the presence of *Botrytis cinerea* isolates resistant to six different classes of site-specific fungicides approved for gray mold management in strawberries (Fernandez-Ortuno et al., 2016). The findings revealed high resistance levels and reduced effectiveness of several fungicides, including pyraclostrobin, boscalid, cyprodinil, and fenhexamid, highlighting the urgent need for the development and use of new, more effective fungicidal options.

The capacity of *B. cinerea* to develop resistance to various active ingredients has also been investigated in other crops. Findings by Jelenic et al. (2024) revealed that *Botrytis cinerea* resistance presents a serious concern, particularly due to the limited number of registered fungicidal products available in Croatia for managing this pathogen in grape production. Their study demonstrated that even at the highest tested concentrations, active ingredients such as pyrimethanil, boscalid, and fenhexamid inhibited fungal growth at rates well below 90%, indicating reduced effectiveness. These results corresponded to our findings about the efficacy of the standard fungicide TELDOR (fenhexamid) on tomato plants. Gabriolotto et al. (2009) also reported that the effectiveness of fenhexamid and iprodione in controlling gray mold disease was unsatisfactory.

All these findings about resistant isolates of *B. cinerea* against various fungicides highlights the need in widening the spectrum of available registered active ingredients. One of these novel and prosperous compounds on the market is penthiopyrad.

In order to effectively manage gray mold in tomatoes and slow down the development of resistant isolates, Bi et al. (2022) used penthiopyrad in rotation or combined with fungicides that have different modes of action. Moreover, the authors limited the usage of penthiopyrad to no more than two applications per season.

Penthiopyrad and its mode of action has been examined by Yanase et al. (2013). In their findings, penthiopyrad had demonstrated a stronger inhibitory effect on spore germination in various plant pathogens, compared to its impact on mycelial growth. Specifically, they found greater antifungal activity against the spore germination of *Botrytis cinerea*, than against its mycelial development. Field trials performed in this study further revealed that penthiopyrad was more effective in a preventive treatment, rather than in a curative one for managing gray mold. The pronounced inhibition of spore germination was likely a key factor contributing to its superior performance when applied preventively, rather than after infection had occurred.

These findings are in accordance with our results, where performed efficacy of the fungicide FONTELIS SC was due to its preventive application in the field trials.

## CONCLUSIONS

In our study, the observed efficacy results of the tested active ingredients (fenhexamid and penthiopyrad) could be explained by the tendency of *Botrytis cinerea* in developing resistant isolates to fungicides. This is emphasizing the need in broadening the range of fungicides effective against gray mold, by introducing new active ingredients. The efficacy of the novel fungicide FONTELIS SC (penthiopyrad) was achieved by its proper application, underlining its preventive mode of action against gray mold disease on tomato plants.

## REFERENCES

- Abbott, W.S.A. (1925). Method of Computing the Effectiveness of an Insecticide. *Journal of Economic Entomology*, 18(2), 265-267.
- Amiri, A., Heath, S.M., & Peres, N.A. (2013). Phenotypic characterization of multifungicide resistance in *Botrytis cinerea* isolates from strawberry fields in Florida. *Plant Dis.*, 97, 393-401.
- Bardas, G.A., Veloukas, T., Koutita, O., & Karaoglanidis, G.S. (2010). Multiple resistance of *Botrytis cinerea* from kiwifruit to SDHIs, QoIs and fungicides of other chemical groups. *Pest Manage. Sci.*, 66, 967-973.
- Bi, Q., Lu, F., Yang, K., Wu, J., Zhang, S., Han, X., Wang, W., & Zhao, J. (2022). Baseline Sensitivity and Resistance of *Botrytis cinerea* to Penthiopyrad in Hebei Province, China. *Horticulturae*, 8, 686.
- Fernandez-Ortuno, D., Grabke, A., Bryson, P.K., Amiri, A., Peres, N.A., & Schnabel, G. (2014). Fungicide resistance profiles in *Botrytis cinerea* from strawberry fields of seven southern U. S. states. *Plant Dis.*, 98, 825-833.
- Fernandez-Ortuno, D., Tores, J.A., Chamorro, M., Perez-Garcia, A., & de Vicente, A. (2016). Characterization of resistance to six chemical classes of site-specific fungicides registered for gray mold control on strawberry in Spain. *Plant Dis.*, 100, 2234-2239.
- FRAC (Fungicide Resistance Action Committee). [www.frac.info](http://www.frac.info)
- Gabriolotto, C., Monchiero, M., Nègre, M., Spadaro, D., & Gullino, M.L. (2009). Effectiveness of control strategies against *Botrytis cinerea* in vineyard and evaluation of the residual fungicide concentrations. *Journal of Environmental Science and Health Part B*, 44(4), 389-396.
- Jelenic, J., Ilic, J., Cosic, J., Vrandečić, K., & Velki, M. (2024) Growing our own poison-a vicious circle of more fungicides and more resistant *Botrytis cinerea* isolates. *J Plant Pathol*, 107, 53-65.
- Kuzmanovska, B., Rusevski, R., Jankuloski, L., Jankulovska, M., Ivic, D., & Bandzo, K. (2012). Phenotypic and genetic characterization of *Botrytis cinerea* isolates from tomato. *Genetika*, 44(3), 633-647.
- Leroch, M., Plesken, C., Weber, R.W.S., Kauff, F., Scalliet, G., & Hahn, M. (2013). Gray mould populations in German strawberry fields are resistant to multiple fungicides and dominated by a novel clade closely related to *Botrytis cinerea*. *Appl. Environ. Microbiol.*, 79, 159-167.
- Liang, X., Peng, Y., Zou, L., Wang, M., Yang, Y., & Zhang, Y. (2020). Baseline sensitivity of penthiopyrad against *Colletotrichum gloeosporioides* species complex and its efficacy for the control of *Colletotrichum* leaf disease in rubber tree. *Eur. J. Plant Pathol.*, 158, 965-974.

- Mouria, B., Ouazzani-Touhami, A., Mouria, A., Benkirane, R., & Douria, A. (2015). Effect of compost and antagonistic fungi on suppression of Tomato Grey Mold. *Biolife*, 3(2), 378-390.
- Townsend, G.R., & Heuberger., J.W. (1943). Methods for estimating losses caused by diseases in fungicides experiments. *Plant Disease Reporter*, 27, 340-343.
- Weber, R.W.S., & Hahn, M. (2011) A rapid and simple method for determining fungicide resistance in *Botrytis*. *J. Plant Dis. Prot.*, 118, 17-25.
- Yanase, Y., Katsuta, H., Tomiya, K., Enomoto, M., & Sakamoto, O. (2013). Development of a novel fungicide, penthiopyrad#. *J. Pestic. Sci.*, 38(3), 167-168.
- Yildiz, F., Yildiz, M., Delen, N., Coskuntuna, A., Kinay, P., & Turkusay, H. (2007). The effects of biological and chemical treatment on gray mold disease in tomatoes grown under greenhouse conditions. *Turk. J. Agric. For.*, 31, 319-325.
- Ziogas, B.N., Markoglou, A.N., & Malandrakis, A.A. (2003). Studies on the inherent resistance risk to fenhexamid in *Botrytis cinerea*. *European Journal of Plant Pathology*, 109, 311-317.