Summa Phytopathologica
The Official Journal of São Paulo State Plant Pathology Association
ISSN-1980-5454
Vol 51 - 2025
artigo e281570

ORIGINAL ARTICLE

Effectiveness of benzimidazoles and strobilurin-triazole fungicide mixtures against *Cercospora sojina*

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Data de recebimento: 18/12/2023. Aceito para publicação em: 09/06/2025

https://doi.org/10.1590/0100-5405/281570

ABSTRACT

Bravo, M.B.; Luque, A.; Sautua, F.; Pérez-Pizá, M.C.; Scandiani, M.M.; Alvarez, D.M.; Reis, E.M.; Carmona, M.A. Effectiveness of benzimidazoles and strobilurin-triazole fungicide mixtures against *Cercospora sojina*. ***Summa Phytopathologica***, v.51, p.1-3, 2025.

Frogeye leaf spot (FLS), caused by *Cercospora sojina*, is an important soybean disease. The present study evaluated six Argentine isolates for their sensitivity to benzimidazoles (BZMs) and mixtures of quinone outside inhibitors (QoIs) with demethylation inhibitors (DMIs). *In vitro*, trifloxystrobin + cyproconazole and pyraclostrobin + epoxiconazole showed the lowest EC_{50} values and were more effective than carbendazim. While the responses of isolates varied, the differences in sensitivity were not statistically significant. *In vivo*, the most and least sensitive isolates had distinct responses. Tri+Cyp effectively reduced the symptoms, which is consistent with *in vitro* data. The present findings reinforce the importance of monitoring *C. sojina* sensitivity to optimize its chemical control.

Keywords: *Cercospora sojina*, fungicides, sensitivity, frogeye leaf spot

RESUMO

Bravo, M.B.; Luque, A.; Sautua, F.; Pérez-Pizá, M.C.; Scandiani, M.M.; Alvarez, D.M.; Reis, E.M.; Carmona, M.A. Eficácia de benzimidazóis e misturas de fungicidas estrobilurina-triazol contra *Cercospora sojina*. ***Summa Phytopathologica***, v.51, p.1-3, 2025."

A mancha olho-de-rã (FLS), causada por *Cercospora sojina*, é uma doença importante da soja. Este estudo avaliou seis isolados argentinos quanto à sensibilidade a benzimidazóis (BZMs) e a misturas de inibidores do sítio Qo (QoIs) com inibidores da desmetilação (DMIs). *In vitro*, trifloxistrobina + ciprocônazol e piraclostrobina + epoxiconazol apresentaram os menores valores de EC_{50} e foram mais eficazes que o carbendazim. Embora as respostas dos isolados tenham variado, as diferenças na sensibilidade não foram estatisticamente significativas. *In vivo*, os isolados mais e menos sensíveis apresentaram respostas distintas. Tri+Cyp reduziu os sintomas de forma eficaz, em concordância com os dados *in vitro*. Os resultados reforçam a importância de monitorar a sensibilidade de *C. sojina* para otimizar seu controle químico.

Palavras-chave: *Cercospora sojina*, fungicidas, sensibilidade, mancha olho-de-rã

Frogeye leaf spot (FLS), caused by *Cercospora sojina*, can reduce soybean yields by 31–60% (3) and has caused major outbreaks in Argentina since its first report in 1999/2000 (5). Although severity has declined with resistant cultivars, the high genetic variability of *C. sojina* remains a threat (1), with two races identified in key regions (11). Benzimidazoles (BZM) and mixtures of quinone outside inhibitors (QoI) with demethylation inhibitors (DMI) are widely used (2), although both groups carry a moderate to high risk of resistance development (7). QoI-resistant isolates were first reported in 2010, and the G143A mutation linked to resistance is now widespread (9). In Argentina, fungicide efficacy and pathogen sensitivity data are lacking. This study evaluated the *in vitro* efficacy of commonly used BZM and QoI+DMI fungicides, assessed *C. sojina* sensitivity, and validated results *in vivo*.

Six *C. sojina* isolates were selected from a previously characterized collection at CEREMIC (11). Five fungicides were tested *in vitro*: i) azoxystrobin + cyproconazole (Azo+Cyp), ii) picoxystrobin + cyproconazole (Pic+Cyp), iii) trifloxystrobin + cyproconazole (Tri+Cyp), iv) pyraclostrobin + epoxiconazole (Pyr+Epo), and v) carbendazim (Car). *In vitro* procedures followed Carmona et al. (4). After 15 days, colony diameters were measured and EC_{50} values estimated by non-linear regression in R (10), using log-transformed data. For the *in vivo* assay, Tri+Cyp and the isolates with the lowest and highest EC_{50} were selected. Soybean genotype DM 4670 was grown in pots with tyndallized substrate and inoculated at V2–V3. Treatments included both isolates with and without fungicide; Tri+Cyp was applied 6 h post-inoculation. Plants were covered with polyethylene for 48 h. Incidence and severity were assessed on day 21, following Carmona et al. (2). The trial followed a completely randomized design with eight replicates. Data from both assays were analyzed with InfoStat (8).

In vitro, Tri+Cyp (0.000796 µg ml^{-1}) and Pyr+Epo (0.001202 µg ml^{-1}) exhibited the lowest EC_{50} values, both significantly lower than those of Azo+Cyp and Car ($p = 0.0077$). Car had the highest EC_{50} (0.020231 µg ml^{-1}), significantly different only from Tri+Cyp and Pyr+Epo. Pic+Cyp (0.004080 µg ml^{-1}) did not differ significantly from any other fungicide (Table 1). These results confirm the strong fungitoxicity of QoI+DMI mixtures, with Tri+Cyp and Pyr+Epo being the most effective. Regarding isolate sensitivity, although no significant differences were observed among isolates ($p = 0.0831$), five out of six showed mean EC_{50} values below 0.035 µg ml^{-1}, indicating increased sensitivity according to the threshold proposed by Edgington et al. (6). In contrast, isolate 172-09 showed a mean EC_{50} value of 0.040 µg ml^{-1}, exceeding this threshold (Table 1). These findings suggest that most isolates are highly sensitive to the evaluated fungicides, although variability in EC_{50} values highlights the need for ongoing monitoring. Previous reports of reduced sensitivity in *C. sojina* and related species (9) further support the importance of continued regional surveillance to ensure sustained fungicide efficacy.

Table 1. EC_{50} values (mean ± SE) of fungicides and *Cercospora sojina* isolates. Fungicide values represent means across six isolates; isolate values across five fungicides. Different letters indicate significant differences (Tukey test, $p<0.05$). Isolate codes refer to strains from CEREMIC.

Average EC_{50} (µg ml^{-1})	
Fungicides	
Tri + Cyp	0.000796 ± 0.000451[a]
Pyr + Epo	0.001202 ± 0.000682[a]
Pic + Cyp	0.004080 ± 0.001772[ab]
Azo + Cyp	0.024656 ± 0.013879[b]
Car	0.020231 ± 0.014113[b]
Isolates	
232-10	0.002500 ± 0.001600[a]
278-10	0.002800 ± 0.001900[a]
234-10	0.010000 ± 0.002100[a]
269-10	0.010000 ± 0.002300[a]
243-10	0.010000 ± 0.005100[a]
172-09	0.040000 ± 0.021600[a]

In vivo, treatment T3 (isolate 232-10 + Tri+Cyp; I% = 8.57 ± 1.09) significantly reduced disease incidence compared to T4 (isolate 172-09 + Tri+Cyp; I% = 40.00 ± 0.36) ($p = 0.0091$). Treatments T1 (isolate 232-10 without fungicide; I% = 17.50 ± 2.64) and T2 (isolate 172-09 without fungicide; I% = 25.00 ± 4.77) exhibited intermediate values and did not differ significantly from either T3 or T4. Disease severity did not differ significantly among treatments ($p = 0.0573$), with values ranging from 2.5 to 3.8% across all treatments. Characteristic FLS symptoms on inoculated plants confirmed effective inoculation (Figure 1). The contrasting EC_{50} values of the two isolates allowed detection of differential responses to Tri+Cyp *in vivo*. Lower FLS incidence was observed in plants inoculated with the lowest EC_{50} isolate and treated with Tri+Cyp, supporting the trends observed *in vitro*. Although severity differences were not significant, incidence trends reflected *in vitro* sensitivity. Further studies are needed to confirm this relationship under field conditions.

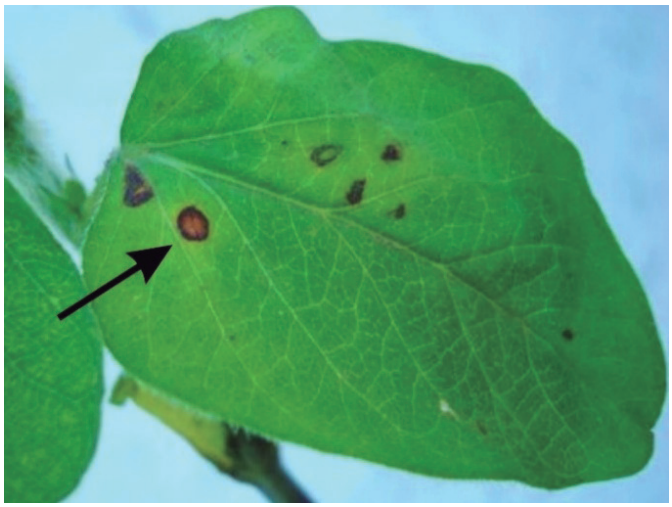

Figure 1. Representative soybean leaf from the *in vivo* assay showing characteristic Frogeye Leaf Spot (FLS) symptoms caused by *Cercospora sojina*. Arrow indicates symptomatic lesions, confirming successful inoculation.

Overall, the results indicate that most *C. sojina* isolates tested were highly sensitive to the evaluated fungicides, particularly QoI+DMI mixtures. Nonetheless, the variability in EC_{50} values (reflected also in the *in vivo* response) underscores the importance of implementing a broader fungicide sensitivity monitoring program in Argentina, with more isolates and active ingredients.

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Data availability statement
Research data is available in the body of the article.

EDITOR CIENTÍFICO: Edson Luiz Furtado

EDITOR ASSOCIADO: José Otávio Machado Menten

CONFLITO DE INTERESSES: Nada a declarar.