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Use of systemic fungicides combined with multisite to control of asian rust and soybean yield

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Abstract

Asian soybean rust is an aggressive disease and chemical control must be handled assertively to minimize productivity damage. The objective was to evaluate systemic fungicides and multisite mancozeb, combined or not, and the effects on disease control and soybean yield, cv. 'BMX Lança'. The work was conducted in the field in the 2016/17 and 2017/18 crops, in a randomized complete block design, with four replications. The following treatments were evaluated: T+P) trifloxystrobin + prothioconazole; A+B) azoxystrobin + benzovindiflupyr; T+P+MB) trifloxystrobin + prothioconazole + mancozeb; A+B+MB) azoxystrobin + benzovindiflupyr + mancozeb; MB) mancozeb; and Control (without fungicide application). After the occurrence of the first symptoms of the disease, the severity (%) was evaluated in five trifolia per plot, every seven days, making it possible to calculate the area under the disease progress curve (AUDPC). The yield components and productivity (kg ha^{-1}) were also evaluated. In both crops, the combination of systemic fungicides and mancozeb reduced the severity (%) and progress of Asian rust, reaching a control above 81%. Treatment with trifloxystrobin + prothioconazole + mancozeb (T+P+MB) obtained the highest percentages of control (94.5% - 2016/17, and 93.7% - 2017/18) of Asian rust, and provided, in the 2016/17 crop, productivity at 15 sc ha^{-1}, in relation to the fungicide applied alone (T+P). The combination of systemic fungicides and mancozeb, as proposed in this work, reduces the severity and progress of Asian rust and ensures the maintenance of the productive potential of the soybean.

Keywords: AUDPC; chemical control; *Glycine max* (L.) Merril; thousand grain weight; *Phakopsora pachyrhizi* (Sydow & Sydow).

Uso de fungicidas sistêmicos combinados a multissítio para o controle da ferrugem asiática e efeitos sobre a produtividade da soja

Resumo

A ferrugem asiática da soja é uma doença agressiva e o controle químico deve ser manejado assertivamente, para minimizar danos sobre a produtividade. Objetivou-se avaliar fungicidas sistêmicos e o multissítio mancozeb, combinados ou não, e os efeitos sobre o controle da doença e a produtividade da soja, cv. 'BMX Lança'. O trabalho foi conduzido à campo nas safras 2016/17 e 2017/18, em delineamento de blocos ao acaso, com quatro repetições. Foram avaliados os tratamentos: T+P) trifloxistrobina + protioconazol; A+B) azoxistrobina + benzovindiflupyr; T+P+MB) trifloxistrobina + protioconazol + mancozeb; A+B+MB) azoxistrobina + benzovindiflupyr + mancozebe; MB) mancozeb; e Testemunha (sem aplicação de fungicidas). Após o surgimento dos primeiros sintomas da doença, a severidade (%) foi avaliada em cinco trifólios por parcela, a cada sete dias, possibilitando o cálculo da área abaixo da curva de progresso da doença (AACPD). Avaliou-se também os componentes de rendimento e a produtividade (kg ha^{-1}). Em ambas as safras, a combinação entre fungicidas sistêmicos e o mancozeb reduziu a severidade (%) e o progresso de ferrugem asiática, atingindo um controle acima de 81%. O tratamento com trifloxistrobina + protioconazol + mancozeb (T+P+MB) obteve os maiores percentuais de controle (94,5% - 2016/17; e 93,7% - 2017/18) da ferrugem asiática, e proporcionou, na safra 2016/17, produtividade superior em 15 sc ha^{-1}, em relação ao fungicida aplicado isoladamente (T+P). A combinação entre fungicidas sistêmicos e mancozeb, conforme proposto nesse trabalho, reduz a severidade e o progresso da ferrugem asiática e

assegura a manutenção do potencial produtivo da soja.
Palavras-chave: AACPD; controle químico; *Glycine max* (L.) Merril; peso de mil grãos; *Phakopsora pachyrhizi* (Sydow & Sydow).

Introduction

Considered the most important soybean disease, Asian rust has as its etiological agent the fungus *Phakopsora pachyrhizi* Sydow & Sydow, whose damage may lead to reductions above 80% of productivity, being this pathogen characterized as of high biological plasticity and easy dissemination (GODOY *et al.*, 2016; FIGUEIREDO *et al.*, 2019). Asian rust reduces the photosynthetic efficiency of soybean to the detriment of coalescence of pustules in the leaf surface (EMBRAPA, 2014), and the control of this disease is essential for the agricultural landscape of soybean (REIS *et al.*, 2019).

In the seasons following the first detection of Asian rust in Brazil, disease control was realized only with systemic action fungicides, mainly with sterol demethylation inhibitors (DMIs - triazoles). However, in 2007, cases of loss of efficiency of this group of fungicides were observed due to its excessive use, which led to the selection of resistant pathogen populations (SCHMITZ *et al.*, 2014; LEMES *et al.*, 2015).

Thus, was started the recommendation of DMIs combined with quinone outside inhibitors (QoIs - strobilurins), but in recent crops these fungicides have demonstrated a gradual loss of efficiency. Some double or triple mixtures, involving fungicides that combine DMIs, QoIs and succinate dehydrogenase inhibitors (SDHIs - carboxamides) have emerged as a possibility of Asian rust control, but the pathogen resistance to this combination of fungicides has been reported (GODOY *et al.*, 2016; KLOSOWSKI *et al.*, 2016; LANGENBACH *et al.*, 2016; FRAC, 2017).

The difficulty in controlling Asian rust with fungicides is becoming increasingly evident, proving the high adaptability of the etiological agent (SILVA *et al.*, 2015). In this sense, the combination between systemic fungicides (site specific) and multisite action arises as a possibility to increase the control efficiency for this plant pathology (KNEBEL *et al.*, 2019).

Multisites act by means of different mechanisms of action, such as inactivation of sulfhydryl groups (-SH) in amino acids and enzymes on fungal cell, resulting in disruption of lipid metabolism, respiration, and ATP production (REIS *et al.*, 2016). In addition, this group of fungicides reduces the severity of the disease and the risk of selection of pathogen-resistant populations to these active ingredients (AZEVEDO, 2015).

In cooperative trials conducted in several Brazilian states in the 2015/16 crop, it was found that systemic fungicides associated with multisites, provided an increase of up to 63% in the control of Asian soybean rust (EMBRAPA, 2016a). Thus, the present work aimed to evaluate the efficiency of the combination of systemic fungicides and multisite on the progress and the control of Asian soybean rust, and the effect of these treatments on crop yield.

Material and Methods

The experiment was conducted in Erechim-RS (27 ° 43' 29.00" S, 52 ° 17' 41.00" W, altitude: 753 m), in the 2016/17 and 2017/18 crops. The soil is a Red Latosol Aluminoferric humic - Oxisol (EMBRAPA, 2018). According to classification system established by Köeppen, the climate is Cfa type (temperate humid with hot summer), with rainfall distributed throughout the year (MATZENAUER *et al.*, 2011). Soil samples were collected at depth of up to 0.10 m and the following chemical properties determined: pH: 5.4; organic matter (OM): 3.6%; P: 5.7 mg dm^{-3}; K: 89 $cmol_c$ dm^{-3}; Al: 0.2 $cmol_c$ dm^{-3}; Ca: 5.6 $cmol_c$ dm^{-3}; Mg: 2.8 $cmol_c$ dm^{-3}; and Cation Exchange Capacity (CEC): 14.2 $cmol_c$ dm^{-3}.

The soybean cultivar used in both crops was 'BMX Lança' (maturity cycle 5.8; undetermined growth). The experiment was conducted in a no-tillage system with wheat as a predecessor crop. The experimental design was randomized blocks (RBD) with 6 treatments and 4 replications, totalizing 24 experimental units, with dimensions of 18 m² (3 m width x 6 m length).

The treatments used were: T + P (trifloxystrobin + prothioconazole; 75 + 87.5 g $a.i.ha^{-1}$); A + B (azoxystrobin + benzovindiflupyr;

90 + 45 g a.i.ha^{-1}); T + P + Mb (trifloxystrobin + prothioconazole + mancozeb; 75+ 87.5 + 1870 g a.i.ha^{-1}); A + B + Mb (azoxystrobin + benzovindiflupyr + mancozeb; 90 + 45 + 1870 g a.i.ha^{-1}), Mb (mancozeb; + 1870 g a.i.ha^{-1}); and Control (absence of fungicidal application). The adjuvant dose followed the manufacturer's recommendation for each fungicide.

The fungicides were applied with the help of a CO_2 pressurized costal sprayer with a constant pressure of 29.0 Psi, resulting in a constant flow rate of 150 L ha^{-1}, with TXA 8002VK conical nozzles spaced 0.5 m apart, and at a constant velocity of approximately 1 m s^{-1}. These applications were realized at stages V6, R1, R5.1 and R6.

To verify the efficiency of the fungicides in the control of Asian rust, after the onset of the first symptoms, the disease severity was evaluated in five leaf samples (trifoliate leaves) per parcel every seven days, and these were compared with the diagrammatic scale of Godoy *et al.* (2006). From the information obtained through the severity scale, the area under the disease progress curve (AUDPC) was determined, whose values were calculated according to the equation proposed by Campbell and Madden (1990).

Harvesting was performed when all plants no longer had green leaves, considering a useful area of 4.0 m² per parcel. For this, a plot stationary threshing was used. The yield components for each treatment were quantified considering the number of pods per plant, from a sample of ten plants per plot, randomly collected at harvest, and the thousand grains weight. Grain moisture (%) was determined by the oven method at 105 °C (BRASIL, 2009) and, subsequently, the values were corrected to 13%. Then, these values were added to the total grain weight of each treatment, in order to obtain the total yield (kg ha^{-1}).

The data obtained were subjected to analysis of variance by the F test ($p \leq 0.05$) and, when significant, the Tukey test ($p \leq 0.05$) was applied to compare averages. The analyzes were performed with the support of SISVAR version 5.6 statistical *software* (FERREIRA, 2011).

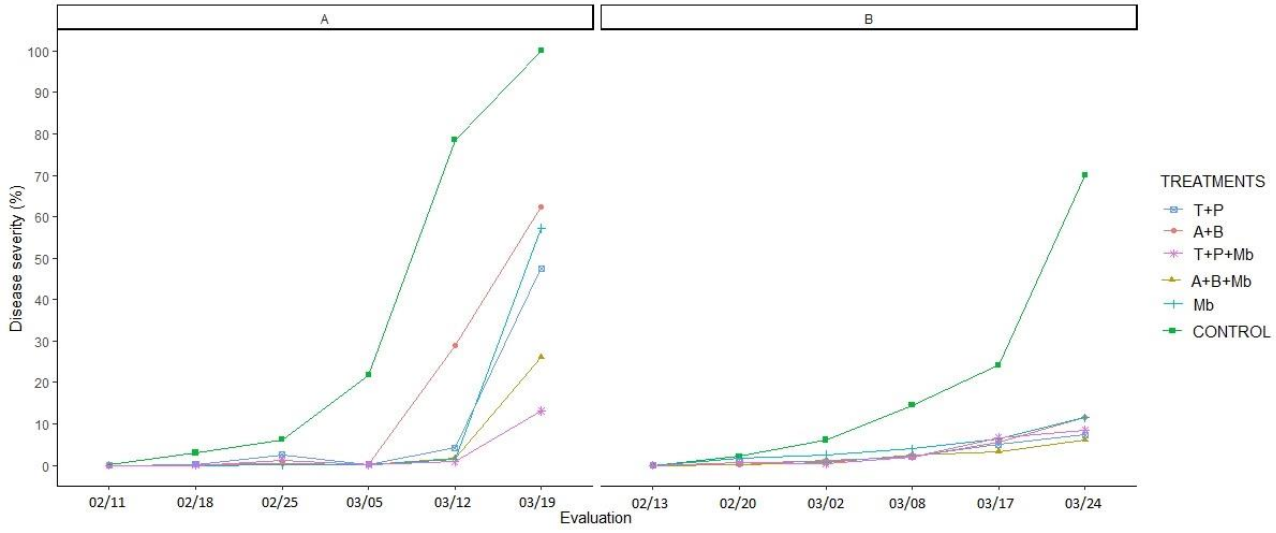
Results and Discussion

All treatments evaluated in this study and that included fungicide application, showed severity of Asian soybean rust lower than the control (Figure 1). For the 2016/17 crop, during the 29 days (03/12/2017) of evaluation, the disease progression in the control treatment reached 80% (Figure 1A), followed by intense defoliation of the plants. However, in the other treatments, there were still trifoliate leaves.

In the 2017/18 crop, the severity of Asian rust was lower, noting that the disease progression in the control treatment reached 70% of severity only at the end of the crop cycle (Figure 1B). In this case, the low initial inoculum pressure may have contributed to a better fungicide efficiency compared to the previous crop.

The increase in severity of Asian rust during the crop is determined by environmental factors, including night temperature (between 18 and 26.5 ^{o}C), continuous leaf wetness and constant rainfall (EMBRAPA, 2013b; MINCHIO *et al.*, 2018). In the present study, Asian rust was late diagnosed in both seasons, that is, in the first half and at the end of February for the 2016/17 and 2017/18 crops, respectively (Figure 1).

Figure 1. Severity (%) of Asian soybean rust, cv. 'BMX Lança', according to the application of systemic fungicides, combined or not with the multisite mancozeb, in the 2016/17 (A) and 2017/18 (B) crop seasons.



On treatments with fungicides combined with mancozeb (T+P+Mb and A+B+Mb) showed the lowest disease severity. Similar to this result, the use of systemic fungicides combined with the multisite mancozeb provided lower severity of Asian rust (27% to 30%), plus an increase in control of up to 63% (EMBRAPA, 2016a). The sporulation of *Phakopsora pachyrhizi* occurs daily when in contact with the host and thus, according to prevailing crop environmental conditions, fungicide applications tend to slow the progress of Asian rust. However, there may be variations depending on the protective or curative effect of each fungicide, beyond the time of application (EMBRAPA, 2016b).

For the area under the disease progress curve (AUDPC) and the control (%), systemic fungicides combined or not with mancozeb multisite achieved greater efficiency in Asian rust control, differing of the control treatment in both seasons (Table 1).

Table 1. Area under the disease progress curve (AUDPC) and control (%) of Asian soybean rust, cv. 'BMX Lança', according to the application of systemic fungicides, combined or not with multisite mancozeb, in the 2016/17 and 2017/18 crop seasons.

Treatments	Crop Season			
	2016/17	2017/18	2016/17	2017/18
	AUDPC		Control (%)	
Trifloxystrobin + prothioconazole	218.1 cA[1]	57.9 bB	80.4 abA	89.7 aA
Azoxystrobin + benzovindiflupyr	426.1 bA	103.2 bB	61.8 bB	81.7 aA
Trifloxistrobin + prothioconazole + mancozeb	60.9 cA	35.3 bA	94.5 aA	93.7 aA
Azoxystrobin + benzovindiflupyr + mancozeb	111.9 cA	84.4 bA	89.9 aA	85.0 aA
Mancozeb	211.5 cA	137.5 bA	81.0 abA	75.7 aA
Control	1116.3 aA	566.7 aB	0.0 cA	0.0 bA
C.V. (%)[2]	36.9		15.9	

[1] Averages followed by the same lowercase letter in the column (seasons) and uppercase letter in the row (between seasons) do not differ significantly from each other by the Tukey test ($p \leq 0.05$). [2] Coefficient of variation.

In terms of the control of Asian rust, in the 2017/18 crop there was a more pronounced effect of the treatments, which showed greater control than in the previous crop (Table 1). This indicates the relation of the progress of the disease with the different environmental conditions observed in both harvests. In the treatment with trifloxystrobin + prothioconazole

+ mancozeb (T+P+Mb) there was a control increase of 14.1% when compared to its use separately (T+P) in the 2016/17 crop (Table 1). These results reinforce the importance of adopting practices involving the combination of systemic and multisite fungicides in the preventive treatment of this pathology (KNEBEL *et al.*, 2019).

After verifying differences in the sensitivity of two populations of *Phakopsora pachyrhizi* to systemic fungicides, Juliatti *et al.* (2017) concluded that the combination of them with multisites, such as mancozeb, should be considered as essential in the management of Asian soybean rust, given the reduced efficiency of active ingredients such as strobilurins, triazoles and carboxamides. It is noteworthy that DMIs and QoIs are fungicides that have high risk for the development of resistance in phytopathogens and, therefore, their use alone is not recommended, as occurred with the fungicide tebuconazole, whose efficiency was gradually reduced from 90% to 24%, in ten harvests (EMBRAPA, 2013a).

When the pathogen enters in the host, plant metabolism is programmed to activate defense mechanisms, thus, decreasing photosynthetic activity and CO_2 absorption, which can lead to flower and pod abortion (TAIZ; ZEIGER, 2013). In this context, it was verified the effect of harvests and of the severity of Asian soybean rust, about the variables related to yield and yield components, whereby, for both harvests, no statistical difference was observed for the number of pods per plant (Table 2).

Similarly, for the thousand grains weight (g), in the 2016/17 crop, there was no difference between treatments with fungicide application (systemic, with and without mancozeb) and the control (Table 2). However, all treatments with fungicides presented above average values for cv. 'BMX Lança', which is approximately 177 g (BRASMAX, 2018), in comparison with the control (176.2 g), thus evidencing the beneficial effect of fungicide application on the thousand grains weight.

According to Almeida *et al.* (2017) an increase of up to 10% in thousand grains weight (g) on soybean genotypes (A 4910 RG; BMX Apolo RR; A 6001 RR; Fundacep 55 RR; and Coodetec 214 RR) that received systemic fungicides (azoxystrobin + cyproconazole; or piraclostrobin + epoxiconazole), realized at: *i)* pre-closing between lines and at 21 days after closing; *ii)* pre-closing between lines and at stage R5.1; and *iii)* at stages R1 and R5.1.

Still regarding the thousand grains weight, in the 2017/18 crop, it can be observed that there was a reduction when compared to the previous crop season (Table 2). This can be explained by the period of water stress at the beginning of the establishment of the crop (stage V2), as it is at this moment that its yield components begin to be defined. It is noteworthy that soybean is sensitive to drought at the beginning of the cycle. This causes a reduction in the emission of new branches and, consequently, reflects on the number of nodes that would produce pods, affecting crop yield (GAVA *et al.*, 2016).

For the grain yield, in the 2016/17 crop, treatments A+B, T+P+Mb, A+B+Mb and Mb differed statistically from the 2017/18 crop (Table 2), corroborating with Silva *et al.* (2015), in which the multisite mancozeb, applied four and five times, at doses of 2.0 and 1.5 kg ha^{-1}, provided better control of Asian soybean rust.

Table 2. Number of pods per plant, weight of thousand grains (g^{-1}) and yield (kg ha^{-1}), according to the application of systemic fungicides, combined or not with multisite mancozeb, for the control of Asian soybean rust, cv. 'BMX Lança', in the 2016/17 and 2017/18 crop seasons.

Treatments	Crop Seasons					
	2016/17	2017/18	2016/17	2017/18	2016/17	2017/18
	Number of pods per plant		Thousand grain weight (g^{-1})		Yield (kg ha^{-1})	
Trifloxystrobin + prothioconazole	46.9 ^{ns}B[1]	71.4 ^{ns}A	190.6 aA	140.0 abB	3589.2 bcA	3643.4 abA
Azoxystrobin + benzovindiflupyr	55.0 A	69.0 A	187.7 abA	137.6 abB	4319.2 abA	3356.6 abcB
Trifloxistrobin + prothioconazole + mancozeb	43.0 B	68.3 A	199.8 aA	144.6 aB	4490.2 aA	3874.4 aB
Azoxystrobin + benzovindiflupyr + mancozeb	43.3 B	60.5 A	198.7 aA	138.5 abB	3994.6 abA	3412.3 abcB
Mancozeb	45.9 B	76.3 A	186.6 abA	129.2 bB	4129.1 abA	2858.2 cB
Control	39.9 B	58.3 A	176.2 bA	126.5 bB	2869.4 cA	3098.5 bcA
C.V. (%)[2]	18.3		3.9		9.9	

[1] Averages followed by the same lowercase letter in the column (season) and uppercase letter in the row (between season) do not differ significantly from each other by the Tukey test ($p \leq 0.05$). ^{ns} not significative. [2] Coefficient of variation.

Additionally, the treatment with trifloxystrobin + prothioconazole + mancozeb (T+P+Mb) stood out for yield in the two crop seasons in which the work was conducted (Table 2). In 2016/17 crop, the addition of mancozeb to the systemic fungicide resulted in a productivity increase of 900 kg ha^{-1} when compared to the systemic fungicide applied isolate (Table 2).

This reinforces the importance of the use of systemic fungicides combined with multisite, in the control of Asian soybean rust, as this practice is a tool for the management of *Phakopsora pachyrhizi* resistance, preserving the efficacy and extending the useful life of fungicides acting on specific sites (DMIs, QoIs and SDHIs) (SILVA *et al.*, 2015).

Conclusions

The combination of systemic fungicides and mancozeb (T+P+Mb - trifloxystrobin + prothioconazole + mancozeb; and A+B+Mb - azoxystrobin + benzovindiflupyr + mancozeb), causes reduction in the severity and the progress of Asian soybean rust, cv. 'BMX Lança', with control over 81%.

The treatment with trifloxystrobin + prothioconazole + mancozeb is effective in controlling Asian rust and increases soybean yield by up to 900 kg ha^{-1} compared to systemic fungicide applied alone.

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