Reduced Asian Soybean Rust Control by Commercial Fungicides Co-formulations in the 2018-2019 Growing Season in Southern Brazil

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Abstract

It has been a growers concen the reduction of Asian soybean rust (ASR) control by commercial fungicide co-formulations in the last growing seasons in southern Brazil. The objective of this work was to assess the ASR control efficacy by the most used co-formulations in the 2018/19 season. In a field experiment, 19 fungicides in commercial formulations to control soybean rust caused by *Phakopsora pachyrhizi*, were evaluated. Chemicals at their recommended doses were sprayed at four soybean growth stages. The first application was performed with 1.82% leaflet incidence and coinciding with R1 phenological stage. The others were performed at 14-18 days intervals. At stage R6, end of the epidemic and coinciding with half of the defoliation in the control plots, the leaf severity was appraised. The experiment was conducted with Ativa soybean cultivar, in 3×6 m plots, four replications and randomized block design. The harvest was made with a plot combine and the yield expressed in grains kg/ha. The means were compared by the Scott-Knott test. The disease control efficacy by 17 fungicide co-formulation showed control less than 57%, one with 78% and none with $\geq 80\%$. The unsprayed treatment severity was 81% and the greatest control of 78% resulted in 3,876 kg/ha yield. Therefore, the hypothesis raised in this work was accepted showing that the site-specific fungicides co-formulations are showing efficacy reduction season after season.

Keywords: fungicide resitance, Glycine max, Phakopsora pachyrhizi

1. Introduction

Soybean [*Glycine max* (L.) Merr.] growing area in Brazil, has increased season-after-season, reaching in 2018/19 35.8 milion hectares (CONAB, 2019).

Asian soybean rust (ASR), caused by *Phakopsora pachyrhizi* (H. Sydow & P. Sydow, 1914), was first reported in March 2001/02 season in Paraguay and in May in Brazil (Yorinori et al., 2001) 18 years ago, beeing since than the major crop disease with the highest damage.

The decision making for disease control with fungicides is based on its amount (Danelli et al., 2015) and on the caused damage, sensu Nutter et al. (1993). The damage caused by ASR can be appraised on commertial farms through the function: y = 1,000 - 6.7 LI, where, y is grain yield normalized to 1,000 kg/ha and LI is leaflet incidence (Danelli et al., 2015).

Aiming at to reduce the damage, among the strategies to control ASR, the most efficient and praticed by growers is fungicides sprayings that, when efficient have potencial to reduce crop damage and loss (Nutter et al., 1993) and cover the application cost (Reis et al., 2019). Nevertheless, their use increase production cost and are showing reduced efficacy due to the fungus sensitivity reduction towards site-specific mode of action (MOA) fungicides (Silva et al., 2008).

The repeated spraying of the same site-specific mecanism of action (MOA) to control a high risk fungus for resistance development, and with a high sporulation potential in a large growing area, with many sprayings per season (up to 10 are still performed), have accelerated the sensitivity reduction of *P. pachyrhizi* towards fungicides (Ishii & Hollomon, 2015; Reis et al., 2017).

In the 2003/2004 growing season, the cooperative experimental fungicides net was implemented with commertial fungicides in several sites of country. The ASR control results have shown a *P. pachyrhizi* sensitivity reduction against the three site-specific fungicides used in its control: DMIs, QoIs, and SDHIs. The early control

efficacy reduction (< 80%) has worried researchers to search for solutions to recover fungicdes efficacy from < 50% to > 80%.

The ASR control failure in Brazil was first reported six seasons after the begginning of rust control with the DMI tebuconazol (Fundação MT em Campo, 2008; Silva et al., 2008; Reis et al., 2017). In the first seasons of its use solo, control was > 80% and in the 2027/18 season, but in the national fungicide trials, its efficacy was reduced to 22% (Reis et al., 2017). Since than on, the evolution of the control reduction season-after-season tor the three MOA site-specific DMIs, QoIs, and SDHIs has been reported and did not stop yet. Co-formulations containing DMI, QoI and SDHI, in double or triple mixes, have been sprayed in a large soybean grown area without the additon of multi-sites, the main reason for the reduction of fungicides efficacy.

The hypothesis raised in this work was that the site-specific MOA fungicides co-formulations are showing efficacy reduction each season reaching in the last < 50% control.

Objective of this work was to assess the ASR control efficacy by commertial co-formulations in the 2018/19 season in Southern Brazil.

2. Materials and Methods

The experiment was carried out at Fazenda Carvalho ERS—324, Km 69, Passo Fundo/RS, (28°12'18" latitude, 52°29'45" longitude, and altitude 660 m a.s.l.), during 2018/19 growing season.

Brasmax Ativa RR, soybean cultivar was directed sown under black oats residues on December 10, 2018, with 50 cm row spacing, and 25 seeds/m².

Crop fertilization (300 kg/ha of 02-20-30; $N-P_2O_5-K_2O$), herbicides and insecticides applications were according to technical recommendations.

Fungicides (Table 1) sprayings were performed with a back-pack sprayer CO_2 driven pressure, boom containing six Teejet model XR 110015 nozzles and water volume 150 L/ha.

Fungicide	Abbreviation	Abbreviation Mechanism of action		Dose (mL or kg/ha)	
Difenoconazole + cyproconazole	Dif + cypr	DMI + DMI	75 + 45	0.3	
Azoxystrobin + cyproconazole	Cypr + azox	DMI + QoI	60 + 24	0.3	
Fenpropimorph	Fenp	IBE	225	0.3	
Trifloxystrobin + cyproconazole	Trif + cypr	QoI + DMI	75 + 32	0.2	
Picoxystrobin + cyproconazole	Pico + cypr	QoI + DMI	60 + 24	0.3	
Carbendazim + tebuconazol + kresoxim methyl	Carb + tebu + kres	TSI + DMI + QoI	25 + 125 + 156.25	1.25	
Trifloxystrobin + tebuconazole	Trif + tebu	QoI + DMI	50 + 100	0.5	
Trifloxystrobin + porthioconazole	Trifl + prot	QoI + DMI	60 + 70	0,.4	
Metominostrobin + tebuconazole	Meto + tebu	QoI + DMI	79.75 + 119.63	0.725	
Azoxystrobin + benzovindiflupyr	Azox + benz	QoI + SDHI	60 + 30	0.2	
Picoxystrobin + tebuconazole	Pico + prot	QoI + DMI	60 + 100	0.5	
Trifloxystrobin + prothioconazol + bixafen	Trif + prot + bix	QoI + DMI + SDHI	75 + 87.5 + 62.5	0.5	
Pyraclostrobin _ epoxiconazole + fluxapiroxade	Pyra + epox + flux	QoI + DMI + SDHI	65 + 40 + 40	0.8	
Pyraclostrobin + fluxapiroxade	Pyra + flux	QoI + SDHI	116,55 + 58.45	0.35	
Tebuconazole + chlorothalonil	Tebu + chlo	DMI + multissite	125 + 1,125	2.5	
Azoxystrobin + cyproconazole + mancozeb	Azox + mcz + cypr	QoI + multissite + DMI	90 + 60 + 1.350	2.0	
Picoxystrobin + benzovindiflupyr	Pico + benz	QoI + SDHI	60 + 30	0.6	
Picoxystrobin + tebuconazol + mancozeb	Pico + tebu + mcz	QoI + DMI + multissite	66.5 + 83.33 + 1.000	2.5	

Table 1. Fungicides description and doses

The experimental design was a randomized block with 19 treatments, four replications, with 3.5 m (6 rows) wide and 6.0 m long corresponding to 21.0 m^2 plots.

For rust assessment two pathometric methods were used: (a) For rust detection in the experimental area, weekly from V5 GS, five plants from the experiment borders were collected. Only central leaflets with leaves petioles inserted in the main plant stem were removed and assessed according to Ogle et al. (1979) and rust incidence was assessed under stereo microscope (30-50x); (b) The treatments effects on rust control were assessed based on leaflet severity. Severity notes, considering the percent leaflet area with symptoms/signs, were assigned in the central row according to Godoy et al. (2005).

At plant ripening, 12 m² area/plot was harvested with a Massey Ferguson adapted plot combine, grains were cleaned, moisture content adjusted to 13%, and grain yield appraised to kg/ha.

Collected data were submitted to test of normality, analyses of variance, regression and means compared Scott & Knott test.

3. Results and Discussion

Regarting the 2018/19 season, ASR was detected on January 16th, at V5 GS, 37 days after sowing on December 10th, 2018. In 43 days epidemics reached 100% leaflet incidence at R4 GS (Figure 1).

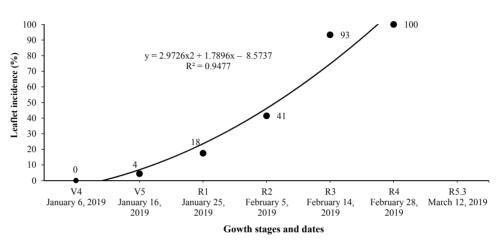


Figure 1. Progress curve of Asian soybean rust rated by leaflet incidence

The data related to the effect of the treatments on leaflet severity, means compared by the Scott & Knott test and with coefficient of variation of 5.91% are shown in (Figure 2). The epidemic intensity in unsprayed plots reached 81% considered severe but among the means registered in previous sesons. The 18%, the lowest severity, was quantified in the treament with picoxystrobin + tebuconazol + mancozeb a multissite fungicide. On the other hand, in treatment with just picoxystrobin + tebuconazol without mancozeb control was 49% which shows the increasing fungitoxicity by the multissite addition. This finding shows the benefical effect of the multissite mancozeb to fight ASR resistance (Figure 3). Among the tested fungicides only three have multi-site in their co-formulation: chlorothalonil + tebuconazole, azoxystrobin + cyproconazole + mancozeb, and picoxystrobin + tuboconazole + mancozeb.

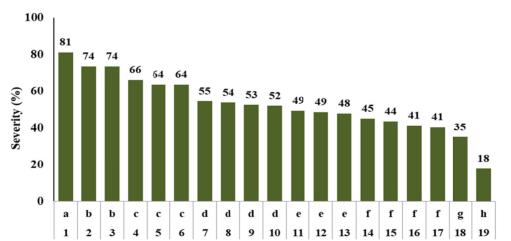


Figure 2. Effect of treatments on Asian soybean rust leaflet severity at R6 GS. Means (letters at columns base) compared by Scott & Knott test. Treatments: (1) Unsprayed; (2) dife + cypr; (3) azox + cypr; (4) fenp.; (5) trifl + cypr; (6) carb + tebu + kres; (7) trifl + tebu; (8) trif+ prot + bixa; (9) meto + tebu; (10) azox + cypr; (11) azox + benz; (12) pico + tebu; (13) trifl + prot; (14) pyra + epox + flux; (15) pyra + fluxa; (16) chlo + tebu: (17) azox + cypr + mcz; (18) pico + benz; (19) pico + tebu + mcz

Only three fungicides showed control higher than 50%, five between 40 to 49%, five between 33 to 39%, two with 22%, and two with 9%.

The evolution of the control reduction was analysed by Reis et al. (2017). In the 2005/05 seasons the control by 'tebu' was 90-91%, in the 2007/08 by 'cyp' + 'azox' was 86%, in the 2009/10 by 'epox' + 'pyra' was 79% and, in the 2009/10 by 'azox' was 79%. Therefore, comparing those control with the obtained in the present study, the reduced control in the last season was clearly shown (Figure 3).

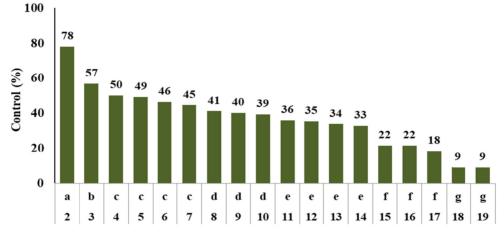


Figure 3. Treatments effect on ASR final leaflet severity control at R6 GS. Means (letters at columns base) compared by Scott & Knott test. (2) pico + tebu + mcz; (3) pico + benz; (4) azox + cypr + mcz; (5) chlo + tebu; (6) pyra + flux: (7) pyra + epox + flux; (8) trifl + prot: (9) pico + tebu; (10) azox + benz; (11) pico + cypr; (12) meto + tebu; (13) trifl + prot + bixa; (14) trifl + tebu; (15) trifl + cypr; (16) carb + tebu + kres; (17) femp; (18) dife + cypr; (19) azox + cypr

The quality and timing of fungicides spraying in field experiments are more precise than in commercial crops, therefore, the results obtained in the field may be inferior to those obtained here. The low control obtained in commercial farms, even where multisite is applied, presents low efficacy by not using the recommended dose (*i.e.*, mancozeb 1.5 kg/ha), not in all sprayings (national average of 2.6/ha). The maximum yield control is achieved with > 80% efficacy (Reis et al., 2019). Spraying site-specifics in double or triple co-cormulations, with cross and mutiple resistance, in all sprayings/area, and in most of the growing area is increasing the *P. pachyrhizi* sensitivity reduction to these fungicides threatening the economically sustainable control.

In the experiment, the relationship between grain yield (y) and ASR leaflet severity (x) was represented by the equation y = -19.46x + 4,421.1 and $R^2 = 0.8244$ (Figure 4). Therefore, for each severity percent point there was a grain yield reduction of 19.46 kg for 4,421.1 kg/ha potential yield. With this equation one can calculate the economic damage threshold according to Munford and Norton (1984) for any grain yield and rust severity, to time the first fungicide spraying as performed in the present work (Danelli et al., 2015).

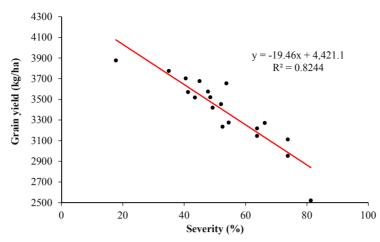


Figure 4. Negative relationship between soybean grain yield and Asian soybean rust leaflet severity

ASR damage, sensu Nutter et al. (1993), was calculated by the difference between the highest yield (3,876 kg/ha) and the yield in each treatment. The negative relationship between damage (kg/ha) and ASR control (%) was represented by the function y = -13.45x + 931.12 (R² = 0.819). For each percent point control reduction there was a 13.45 kg grain reduction for a maximum 931.12 kg/ha damage (Figure 5). The highest yield 3,826 kg/ha was achieved with 78% control and considered without damage (Table 2). Thus, taking for example the lowest and the highest fungicides control in Figure 3, damage can be appraised with the generated function: for 9% control, 810.07 kg damage, and for 57% control, 164.47 kg reduction.

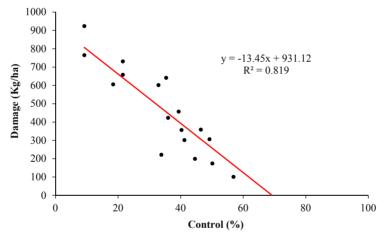


Figure 5. Negative relationship between Asian soybean rust damage and control

Related to the damage caused by plant diseases most of the published papers quantified the yield reduction only by adjectives instead of numbers and the used methodology for damage quantification is not described. The damage can be appraised knowing the relationship between different disease intensities and the resulting damage (Sah & MacKenzie, 1987). In our study, considering the maximum ASR severity of 83%, the maximum yield 3,876 kg/ha and the unsprayed treatment yield of 1,355 kg/ha, therefore the reduction was 1,355 kg/ha or 34.95%, lower than those mentioned in the literature. Each severity point reduced 17.0 kg in a 3,876 kg/ha actual gross yield.

In the first spraying cost was considered soybean kneading by sprayer wheels in commercial farms (32 m long boom, 35 cm width tires), fuel (75 HP engine), labor, fungicide price (dose/ha), yield potential kg/ha), soybean prince (one US\$ = R\$ 5.20; US\$ 26.47/60 kg, Dec., 2020—Cotrijal, Não Me Toque county, Rio Grande do Sul state, Brazil), totaling US\$ 46.00/ha, or 104 kg/ha soybean. In the calculation the costs were transformed into soybean grains weight (kg/ha) (Table 2).

Treatments	Fungicide cost		Total cost ^v	Control	Damage ^x	Yield (I	Yield (Kg/ha)	
	Dose (Kg/ha)	US \$/ha	Soy (Kg/ha)	(Kg/ha)	(%)	(Kg/ha)	Gross ^y	Net ^z
Unsprayed	0	0	0	0	0	1355	<u>(2521)</u> f	1355
Dife + cypr	0.3	9.03	20.43	411	9 g	765	3111 d	2700
Fenp	0.3	8.37	18.91	397	18 f	606	3271 d	2874
Chlo + tebu	2.5	20.19	45.63	643	49 c	306	3570 c	2927
Pico + cypr	0.3	9.81	22.17	427	36 e	423	3453 c	3027
Azox + cypr	0.3	8.13	18.38	392	9 g	924	2953 e	2561
Trifl +cypr	0.2	9.51	21.51	421	22 f	731	3145 d	2725
Trif + tebu	0.5	7.79	17.60	385	33 e	602	3275 d	2890
Meto + tebu	0.725	11.85	26.77	469	35 e	642	3235 d	2766
Pico + tebu	0.5	15.38	34.77	543	40 d	357	3520 c	2977
Carb + tebu + kres	1.25	18.04	40.67	598	22 f	658	3218 d	2621
Trifl + prot	0.4	20.98	47.42	659	41 d	301	3575 c	2916
Azox + benz	0.2	16.23	36.68	560	39 d	457	3419 c	2859
Pico + benPico + benz	0.6	22.50	50.85	691	57 b	101	3775 a	3085
Pyr + flux	0.35	22.42	50.68	689	46 c	359	3517 c	2829
Pyra + epox + flux	0.8	23.13	52.28	704	45 c	200	3677 b	2973
Azox + cypr + mcz	2.0	20.54	46.42	650	50 c	174	3702 b	3053
Trif+ prot +bixa	0.5	28.85	65.19	823	34 e	222	3655 b	2832
Pico + tebu + mcz	2.5	29.71	67.15	763	78 a	0	3876 a	3036
C.V. (%)					10.6		2.8	

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Table 2. Asian soybean rus	t spraying cost,	and treatments effect on	control, damage,	and grain yield

Note. ^vTotal cost: kneading by sprayer wheels, fuel, labor, fungicide price (four spraying/ha), yield potential kg/ha, soybean price (US\$ 26.47/60 kg); ^xDamage: the highest gross yield 3,876 kg/ha minus the yield in each treatment; ^yGross yield: actual yield; ^zNet yield: Gross yield minus the spraying cost (kg/ha). Means follow by the same letter in the columns do not differ by Scott-Knott's test.

Our work shows that the profit is relate to the control efficacy and cost of application (Table 2).

In the last growing seasons experiments have shown a reduction in the efficacy of Asian soybean rust control by commercial fungicides (Reis et al., 2017). This statement can be confirmed by the mixtures performance with control less than 50% (Figure 3). Most of the commercial chemicals used by growers do not contain in their formulation multisite such as chlorothalonil, mancozeb or copper oxychloride (Figure 2), and there is a need for tank mixing which is little used. This has delayed the use of multisite across the soybean grown area and in all sprayings. Therefore, there is a need to shorten the time for all fungicides sold for soybean rust control to contain multisite in their formulation, as happened with those used to potato, tomato and grapevine downy mildew control (Duvauchelle & Ruccia, 2015).

The hypothesis was not accepted only for two fungicides that showed control higher than 50%.

It may be concluded that, season after season, the sensitivity of *P. pachyrhizi* to site-specific fungicides is decreasing. One should keep in mind that the maximum yield is obtained with control > 80%.

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