

Asian Soybean Rust Severity Sowed in Different Seasons

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Abstract

In on-farm trials, the foliolar severity of Asian soybean rust was evaluated in 44 areas, in three regions of Mato Grosso sown in December (2019) and February (2020). Several susceptible cultivars were used in different crop systems; insect pests and weeds were controlled with different management systems by the farmers. Forty soybean leaflets from four plots replications, demarcated at random in each field were taken. In laboratory foliolar severity was appraised. For rust control in the trials conducted in February, fungicides with efficiency greater than 60% were used consisting of DMIs, QoIs and SDHIs in double or triple mixtures, always adding multisites (chlorothalonil, mancozeb, copper oxychloride). The severity was greater in the fields sown in December (4.84% than in February 0.68%). The number of fungicides spraying/ha in December was 6.4 and February 4.6. It is discussed that through the use of multisites fungicides, the mutation potential in *Phakopsora pachyrhizi* is reduced and that the spores from areas cultivated in February, probably due to unfavorable environment, do not survive during the soybean free-period. Our results indicate that the sowing period can be changed from the end of December to February, since multisites fungicides are always used.

Keywords: *Glycine max*, *Phakopsora pachyrhizi*, site specific and multisite fungicides, time of planting

1. Introduction

Mato Grosso (MT) state is the largest Brazilian soybean producer. In the 2019/20 season, an area of 9.85 million hectares was cultivated, yielding 3.55 t/ha and producing 34.96 million tons (IMEA, 2020a).

Soybean crop in MT begins to be sown on September 16th and ends on December 31st.

In the last seasons, according to data from the Anti-rust Consortium (2020), Asian soybean rust (ASR), caused by *Phakopsora pachyrhizi* H. Sydow & P. Sydow (1914), has been detected in the state since the second half of December and, in a short time, the epidemics reaches all fields.

According to the Anti-Rust Consortium (CAF, 2020) in 2018/19 season, a 3-4 fungicide sprayings per hectare have been made. This is because, in most farms, sprayings are made on an empirical basis without observing the economic damage threshold (Casa et al., 2013; Danelli et al., 2015).

The increase in doses and in number of site-specific fungicides sprayings increases the directional selection (van den Bosch et al., 2015). Therefore, the demand to decrease the number of fungicide applications to reduce the development of *P. pachyrhizi* resistance to DMIs, QoIs and SDHIs has been debated (Reis et al., 2020).

The large areas sown with soybeans in Brazil and neighboring countries, practically every month of the year, combined with the fungicide applicatin frequency, especially the site-specific, have reduced the fungus sensitivity in some regions with more favorable environmental conditions, which are the cases of late December sowing soybean in MT and other states.

In DSS, the ASR severity has been high, sometimes requiring more than seven applications of site-specific fungicides/ha/season and yet with low control efficacy. This sowing season has been demanded after 2015 season, as producers who previously sowed a little later, in February or March, to make their own or commercial

seeds, were prevented and had to do in late December, the last date allowed by the current legislation (IN 002/2015). Despite that most of seed in this region is produced from fields sown in October and November, more growers, especially small ones, need to make their own seeds at a time to get better quality.

The February sowing soybean (FSS) is less subject to the inoculum pressure from the surrounding fields, when the majority was already harvested, and associated with the lower rain frequencies, and a minimum of 6 -7 hours of continuous leaf wetting are not always satisfied reducing the severity of ASR intensity, and with lesser fungicide need compared to DSS.

The expressive number of fungicide sprays made in late DSS, allowed by IN 002/15, of 6.4, associated with the 3-4 performed in the main season, has revealed what is known as the pesticide syndrome, a vicious circle where control failures are remedied by an increased number of sprays (Doutt & Smith, 1971).

To fight ASR resistance, is necessary to reduce the number of fungicide sprayings and the combination of control methods, as in the Integrated Disease Management, to reduce the negative impact on the environment and making the production model sustainable. In addition to chemical, biological, genetic control, cultural control also important tool. Evasion or escape (Marchionato, 1949; Bergamin et al., 1995) is important to escape from environment more favorable to the pathogen and the consequent rust development.

The use of site-specific and multisite fungicides does not have the same effect on directional selection. The selection towards resistance, in *P. pachyrhizi*, has been reported only for site-specific fungicides (van den Bosch & Gilligan, 2008). In contrast, multisites do not select mutants and are, as a result, indicated as the main anti-resistance tool (Hahm, & Leroch, 2015).

The statement that FSS may increase directional selection, is not scientifically supported. On the other hand, longer than FSS, is the presence of soybean as weed in the middle of cotton crops, in an area of 1.0 million hectares which increases the period of soybean presence in the field (Reis & Guerra, 2020, unpublished data), the fungus multiplication and receiving a mean of seven-eight site-specific fungicide sprayings (IMA, 2020) is what should be the main concern and must be promptly repaired, especially in the soybean free-period.

We did not find in the available literature publications that scientifically justify avoiding soybean sowing in February, and initiatives to reduce soybean weed population in other crop species in MT.

The hypothesis formulated in this work is that soybean crop sown in December has greater ASR severity than those in February, treated with site-specific fungicides and requiring a great spraying number.

The objective of the study was to evaluate and compare the foliolar severity of ASR, as well the number of sprayings, in soybean crops sown in December 2019, currently sown for seed production, and February 2020 in MT exploring one of the main principles of plant disease, evasion or escape (Marchionato, 1949), replacing the current DSS by FSS.

2. Method

The on-farm trials were carried with soybean grown in the North, West and South regions of MT state, sown at the end of December 2019 and between February 1th and 15th, 2020 with a regional differential in environment and disease management, especially the multisites fungicides use according to the research recommendation in all DSS and FSS.

The evaluations were carried out from January to May 2020, during the entire crop cycle in both sowing times.

At first sampling, data were recorded regarding the county, farm name, producer name, soybean cultivar, sowing date, phenological crop stage, geographical coordinates of each sampling point and the sanitary crop management.

The initial leaf sampling and rust detection were made by Aprosoja trained personnel and samples sent for analysis in the laboratory of the Rio Verde Foundation for Research and Technological Development, Lucas do Rio Verde (FRV), county, Mato Grosso. After the disease was detected by the FRV laboratory, the samplings were performed by the FRV team, at 15 days interval.

Sample size. Only one green central leaflet per plant with petiole inserted in the main stem, preferably in the 3rd or 4th node was collected at every 40 steps, totaling 40 per sampling. The leaflets severity was appraised at the Plant Pathology Laboratory of FRV according to the diagrammatic scale (Godoy et al., 2006) always performed by the same personnel.

The experimental design was randomized treatments with four repetitions and the means compared by the Tukey test for the areas sown in December and February of each region.

The work was performed in DSS and FSS that received their own rust control program with commercial fungicides without any interference from the monitors regarding the fungicides used and the time of their application, including pest control, unless the recommendation (mandatory for both FSS and DSS) was always used associated to multisites in all sprayings.

The work covered 23 farms (14 DSS, 9 FSS). Eight seeded in December and at the research request seeded part of the area in the same farm, in February, for comparison under the closest possible environmental conditions, totaling 14 sown in December (D) and 17 in February (F), located in the North (5D, 9F), South (6D, 7F) and West (3D, 1F) regions of MT, and 44 sampled areas, located in the South (8D, 10F), North (9D, 10F) and West (6D, 1F) regions of MT.

For chemical rust control, in FSS and DSS, site-specific fungicides (DMI, QoI, SDHI) in double and/or triple mixtures always added with multisites (chlorothalonil, mancozeb, copper oxychloride) or multisites isolated was indicated in all areas in all applications as unique anti-resistance strategy. These fungicides showed rust control in previous field experiments greater than 60%: cyproconazole + picoxystrobin (0.3 L/ha) + mancozeb (1.5 kg/ha); epoxiconazole-pyraclostrobin-fluxapyroxade (0.8 L/ha) + copper oxychloride (0.5 L/ha); azoxystrobin-benzovindiflupyr (0.2 kg/ha) + chlorothalonil (1.5 L/ha); prothioconazole-trifloxystrobin (0.4 L/ha) + mancozeb (1.5 kg/ha); epoxiconazole-fluxapyroxade (0.3 L/ha) + copper oxychloride (0.5 L/ha); cyproconazole + mancozeb (2.0 kg/ha); and cyproconazole-picoxystrobin-benzovindiflupyr (0.6 L/ha) + mancozeb (1.5 kg/ha).

The foliolar severity data, from the last evaluation, were submitted to analysis of variance and means compared by the Tukey test at 5%.

3. Results and Discussion

It has been observed in MT since 2015 growing season when was released the State Normative Instruction 002/2015, that DSSs have greater severity than those sown in February and, therefore, receiving a great number of fungicide sprayings, as scientifically documented in a previous report (Reis et al., 2020) and in the present work.

South Region

In the experimental areas sown in December (8 trials), 9.23% leaflet severity were quantified and in February (10 trials) only 1.48% (Table 1). The highest severities occurred in fields sprayed with specific-site solo, without association with multisites (Table 1, Farm 1 and Farm 2). The highest number of applications were made in DSS (6.9) in contrast to FSS (4.3), and even so, the severity was significantly greater, indicating that both, the lack of associated multisites and the environmental conditions such high rain frequency favored the ASR.

Even with all fungicide sprayings made associated with multisites on Farm 3 (A1 and A2), Farm 5 (A1), Farm 6 (A1) and Farm 7 (A1), between seven and eight sprays, the ASR severity was greater in DSS than in FSS at same farms (Table 1). This region of the state has always been the most critical for ASR, including Primavera do Leste county, where the soybean free-period, in Brazil, was created. The frequent and intense rains that occur, especially in January make difficult to control the disease both in the surrounding fields and especially in DSS later seeding. Even though DSSs receive early rust inoculum from neighboring fields that are already being harvested. The rain in January was slightly above average, but within the normal range of the region, therefore, it is a typical season according to data from the last 24 years.

The ASR greatest severity in FSS in 2020 compared to those occurring in other regions, can be explained by the extemporaneous rains that occurred in May, 2020 (Figure 1) with volumes almost five times higher than the historical mean in the second ten days period.

Table 1. Asian soybean rust severity (%), spraying number and fungicide mode of action in the south region at two seeding times (December and February) in Mato Grosso state, Brazil

County	Farm/Area/Growth Stage	Sowing time		Mode of action (no.)							
		Severity(%)		Site-specific (SE)		Multisite (M) solo		M±SE		Total	
		Dec.	Feb.	Dec.	Feb.	Dec.	Feb.	Dec.	Feb.	Dec.	Feb.
Campo Verde	Farm 1-A1-R.6	27.45	-	4	-	0	-	0	-	4	-
Campo Verde	Farm 2-A1-R.6	7.09	-	2	-	0	-	4	-	6	-
Campo Verde	Farm 2-A2-R.6	8.96	-	2	-	0	-	4	-	6	-
Primavera do Leste	Farm 3-A1-R.6	3.45	-	0	-	0	-	8	-	8	-
Primavera do Leste	Farm 3-A2-R.6	7.25	-	0	-	0	-	8	-	8	-
Primavera do Leste	Farm 3-A3-R.6	-	0.3	-	0	-	0	-	4	-	4
Primavera do Leste	Farm 4-A2-R.6	-	1.06	-	0	-	0	-	4	-	4
Primavera do Leste	Farm 5-A1-R.6	4.03	-	0	-	0	-	7	-	7	-
Primavera do Leste	Farm 5-A2-R.6	-	1.21	-	0	-	0	-	4	-	4
Primavera do Leste	Farm 6-A1-R.6	7.69	-	0	-	0	-	7	-	7	-
Primavera do Leste	Farm 6-A2-R.6	-	1.18	-	0	-	0	-	4	-	4
Primavera do Leste	Farm 7-A1-R.6	7.88	-	1	-	0	-	8	-	9	-
Primavera do Leste	Farm 7-A2-R.6	-	1.67	-	0	-	0	-	4	-	4
Primavera do Leste	Farm 7-A3-R.6	-	1.74	-	0	-	3	-	3	-	6
Campo Verde	Farm 8-A1-R.6	-	3.22	-	2	-	0	-	2	-	4
Campo Verde	Farm 8-A2-R.6	-	0.2	-	2	-	0	-	2	-	4
Campo Verde	Farm 8-A3-R.6	-	0.66	-	2	-	0	-	2	-	4
Paranatinga	Farm 9-A1-R.7	-	2.73	-	0	-	0	-	5	-	5
Mean		9.23 A	1.48 B							6.9	4.3
STD		1.5	15								
C.V. (%) = 17.0											

Note. (-) not applied. Means followed by the different capital letters in the line are statistically different according to Tukey multiple range test.

3.2 North Region

In this region, there was statistical difference in severity between the two growing seasons 2.57% in December (9 trials) and 0.11% in February (10 trials) (Table 2). This high severity in the field located in the Diamantino county (Farm 15) may be explained by the high rain frequency (40% more than the historical average at the end of February and more than double at the end of March), (Figure 3), which provided better environmental conditions for the rust development. In this field, only four fungicide applications were performed and none of them with multisite, which also explains the control failure. This points out that the low fungicide efficacy especially in late sowings worsening with the lack of associated multisite. This farm is also located in a transition zone between the north and the south, with specific environmental characteristics.

Historically, the northern region of MT is less favorable to ASR, due to the lowest altitudes and warmer environment. The rust is usually detected later during the season and is less severe than in the south and west regions. The same situation occurs even in late seedings, where the rust pressure was also lower in the research conducted in the last season (Reis et al., 2020) than in other regions. In this region the number of fungicide applications in the two seeding times was similar DSS (5.4) and FSS (5.0). The greater number of sprays in this region during FSS may be the result of fiscal pressure from the State Phytosanitary Defense Service on FSS, and not for ARS own pressure.

Table 2. Asian soybean rust severity (%), spraying number and fungicide mode of action in the north region at two seeding times (December and February) in Mato Grosso state, Brazil

County	Farm/Area/Growth stage	Sowing time		Mode of action (no.)							
		Severity(%)		Site-specific (SE)		Multisite (M) solo		M±SE		Total	
		Dec.	Feb.	Dec.	Feb.	Dec.	Feb.	Dec.	Feb.	Dec.	Feb.
Claúdia	Farm 10-A1-R.6	0.3	-	4	-	0	-	0	-	4	-
Claúdia	Farm 10-A2-R.6	0.2	-	4	-	0	-	0	-	4	-
Claúdia	Farm 10-A3-R.6	0.03	-	4	-	0	-	0	-	4	-
Lucas do Rio Verde	Farm 11-A1-R.7	0.71	-	0	-	0	-	7	-	7	-
Lucas do Rio Verde	Farm 11-A2-R.7	-	0.28	-	0	-	0	-	4	-	4
Lucas do Rio Verde	Farm 12-A1-R.7	-	0.32	-	0	-	0	-	4	-	4
Marcelândia	Farm 13-A1-R5.4	0.01	-	0	-	0	-	8	-	8	-
Marcelândia	Farm 13-A2-R5.3/R6	-	0.03	-	0	-	0	-	3	-	3
Tabaporã	Farm 14-A1-R5.4	0.18	-	1	-	0	-	5	-	6	-
Tabaporã	Farm 14-A2-R5.4	0.31	-	1	-	0	-	5	-	6	-
Tabaporã	Farm 14-A3-R5.4	0.68	-	1	-	0	-	5	-	6	-
Tabaporã	Farm 14-A4-R5.4	-	0.43	-	0	-	0	-	5	-	5
Diamantino	Farm 15-A1-R5.5	20,7	-	4	-	0	-	0	-	4	-
Claúdia	Farm 16-A1-R.6	-	0	-	0	-	2	-	4	-	6
Claúdia	Farm 17-A1-R.6	-	0.01	-	0	-	0	-	4	-	4
Claúdia	Farm 17-A2-R.6	-	0.0015	-	0	-	0	-	4	-	4
Vera	Farm 18-A1-R.6	-	0.035	-	0	-	0	-	5	-	5
Vera	Farm 19-A1-R.6	-	0.17	-	0	-	4	-	2	-	6
Vera	Farm 20-A1-R.6	-	0.05	-	0	-	4	-	2	-	6
Mean		2.57 A	0.11 B							5.4	4.7
STD		0.27	0.02								
C.V. (%) = 13.9											

Note. (-) not applied. Means followed by the different capital letters in the line are statistically different according to Tukey multiple range test.

3.3 West Region

Under this situation, a statistical difference was detected in the rust leaflet severity between the two growing seasons 2.74% in December (6 trials) and 0.45% in February (1 trial) (Table 3).

In his region, also known for the high historical rust pressure in the last soybean growing seasons. On average, it was the region with the highest number of DSS sprays, with up to nine applications. For this reason, due to the aggressiveness of ASR in these December fields, when the grower decided to reduce the spraying number, Farm 21, the rust went out of control in one of its area.

It is likely that the high number of sprayings in only one farm sowed in February, Farm 23-A3, was due to the application of multisite solo, with a low protection period but which, despite the greater frequency of rain, in April and May (Figures 2 and 3). ASR was significantly less in these fields than in those of late December, despite receiving seven to nine sprayings, including multisites on Farms 22 and 23, showing the importance of environmental conditions just by changing the sowing date.

Table 3. Asian soybean rust severity (%), spraying number and fungicide mode of action in the west region at two seeding times (December and February) in Mato Grosso state, Brazil

County	Farm/Area/Growth Stage	Sowing time		Mode of action (no.)							
		Severity(%)		Site-specific (SE)		Multisite (M) solo		M±SE		Total	
		Dec.	Feb.	Dec.	Feb.	Dec.	Feb.	Dec.	Feb.	Dec.	Feb.
Campo Novo	Farm 21-A1-R5.3	10.8	-	1	-	2	-	3	-	6	-
Campo Novo	Farm 21-A2-R5.4	1.92	-	1	-	2	-	3	-	6	-
Campos de Júlio	Farm 22-A1-R.6	0.9	-	5	-	0	-	4	-	9	-
Campos de Júlio	Farm 22-A2-R5.5	1.5	-	5	-	0	-	4	-	9	-
Campos de Júlio	Farm 23-A1-R5.5	0.83	-	2	-	1	-	4	-	7	-
Campos de Júlio	Farm 23-A2-R5.5	0.43	-	2	-	1	-	4	-	7	-
Campos de Júlio	Farm 23-A3-R5.3	-	0.45	-	0	-	7	-	0	-	7
Mean		2.74 A	0.45 B							7.3	7
STD		1.04	0.22								
C.V. (%) = 38.3											

Note. (-) not applied. Means followed by the different capital letters in the line are statistically different according to Tukey multiple range test.

In the three regions, regarding the overall mean, there was a significant difference between the sowing times, DSS (4.84%) and FSS (0.68%) foliolar severity (C.V. 6.4% and standard deviation for DSS 0.33 and for FSS 0.08).

Considering the number of sprayings performed in the two sowing dates, the experimental areas in DSS received an average of 6.4 applications against 4.6 in FSS. Although the number of applications made in February seems high, when compared to those performed in the previous work (Reis et al., 2020), in the present season the growers followed the research recommendation, and made applications of multisite, especially mancozeb, (associated to site-specifics). Some growers sprayed only multisites and other biological products, and these products have been applied more than necessary.

Also due to the pressure of the State Sanitary Defense Agency inspection, many farmers made unnecessary applications, sometimes preventively in the FSS and even too late in the season, resulting in higher number of sprayings. Producer that used only multisite in FSS, achieved effective disease control. If we do not consider the use of multisites solo, but only the site-specific or used in mixture with multisites, in DSS, 6.2 applications were performed on average, against 3.6 in FSS, therefore 42% reduction in spraying number. All producers of FSS sprayed multisites associated, or not, to site-specific, while 50% of the farmers of DSS used at least once site-specific solo, but used multisites on average in 88.9% applications, despite the recommendation that multisites should be used in all of them. For crop, including December, there was no obligation to use multisites in tank mixture with site-specific, but was a research recommendation. For FSS we suggest that this practice should be mandatory in the future.

This strategy was used considering that *P. pachyrhizi* does not undergo mutations to multisites applied solo, or in tank mix, or in co-formulations, in all sprayings. Mutations at the target site conferring resistance only occurs against site-specific-fungicides (Hahm & Leroch, 2015). In contrast, multisite inhibitor fungicides, are not prone to target site resistance (Hahm & Leroch, 2015) for they act in different points of the fungus metabolism, and for this reason, they play a fundamental role in anti-resistance strategy (McGrath, 2004).

The directional selection is proportional to the number of site-specific applications (European and Mediterranean Plant Protection Organization, 1998). It must be emphasized that mutations occur only for site-specific (DMI, QoI, SDHI) and not for the multisites (Hahm & Leroch, 2015). And, the larger the spore population produced, the greater the chance of selecting mutant individuals.

Therefore, in the present work, care has been taken to apply site-specific + multisite mixtures in all areas and applications as an anti-resistance strategy, to avoid directional selection.

Most areas sown on the same farm under similar environmental conditions, but due to the variation in rainfall resulted in ASR severities of 6.5 to 13.7% high in DSS (Farm 3-A1 and Farm 3-A2 compared with Farm 3-A3, Tab 1) and with half the number of sprays performed in FSS. In addition, it is likely that the spore population reduction resulting from February fields, compared to late December, even with the use of site-specific applications with multisites, suggests that coupled with a greater rain frequency in the January (Figure 1) the highest rust severity

was detected in the DSS. It is likely that for the DSS, the spore population from the fields in the entire region that begin to be harvested in January, also influences rust severity in DSS.

Moreover, there were cases where the difference in severity between DSS and FSS was smaller, in the same farm, indicating that the comparison of sowing times and ASR severity should be done in spatially isolated areas to minimize the interference among experimental plots, old to the youngest. This situation should not occur if DSS is not near to FSS. This occurred in all regions where there were trials done in both seeding times on the same farm, (Tables 1, 2 and 3), but still with lower severities in FSS, indicating that the escape, changing the DSS to FSS, is a practice to be implemented.

It should be considered that in FSS and DSS to rationalize the use of fungicides the Economic Damage Threshold (EDT) principle (Casa et al., 2013) was used to time the first spraying. In this way, the first applications were not “preventive”, nor “in closing the rows”, reducing more the number of unnecessary applications and contributing to greater economic and environmental sustainability of soybean in Mato Grosso. Moreover, van den Bosch et al. (2015) stated that in conclusion, ‘current evidence does not support early or protective treatment being a resistance management tactic’.

It is necessary to rationalize the use of fungicides not only the exceeding, curative and inefficient applications made in the DSS as demonstrated in the previous work (Reis et al., 2020), and in the previous year 7.4. In the last season (2019/2020) the difference was greater between DSS and FSS ASR control, since not all farmers used multisites (Reis et al., 2020). Despite the use of multisites in most DSS sprayings, the control showed low efficiency. It is likely that this was due to low multisite use and that the higher rainfall frequencies leached the non penetrant fungicides.

The intensity of the directional selection occurs: (a) by the use of site-specific fungicides without multisite (DMI, QoI, SDHI) the only ones that select mutations; (b) the time period of site-specific use solo or in double or triple mixtures in the control of *P. pachyrhizi*, for 18 years in Brazil and MT state; (c) as a function of the inoculum density of *P. pachyrhizi* related to the grown soybean area and of soybean weed plants in the middle of cotton crops; (d) to a greater degree by the number of sprayings in the crop, and for site-specific fungicides. In the normal soybean season 10 million ha and 3-4 sprayings per ha of site-specific fungicides are performed.

The rate of multisites use in Brazilian soybean crop was 30% of the spraying number in the last growing season. In addition, the risk of up to four sprayings of the same site-specific fungicides (DMI, QoI, SDHI) in the experimental areas coordinated the Anti-rust National Consortium has to be considered, including products with especial temporary registration in the Ministry of Agriculture are used. Despite the importance of this work, its risk should be explained considering the sowing time of the experimental areas, the large number experimental sites over the country, especially in MT, which can aggravate the ASR management due to the existence soybean weed plants amid cotton fields near the experimental areas. It is likely that spores can migrate to soybean weeds during the soybean free-period. Perhaps in other states this situation is not as serious as in MT, the main producing state in Brazil sowing almost one million hectares in the second season, in areas where there was soybean grown in the first season.

It is likely that the risk of new mutation emergence may be reduced by the use of multisite in all applications and in all grown areas sown in February. Reports confirm the relationship between the number of sprays and the development of resistance (Ishii & Hollomon, 2015). Therefore, there is no risk for multisites as they do not select *P. pachyrhizi* mutants, especially in FSS, where the rain frequency is lower, resulting a longer protection period.

In addition, the lowest risk in the development of resistance, due to the use of associated multisites, and probably due to the lower spore density produced in the February fields, our results showed a significant lower reduction in FSS than in the DSS, indicating that the replacement of the DSS by FSS is feasible.

The replacement of DSS by FSS, with harvest done in May, or until 06/15, when the soybean free-period starts for FSS, continues to avoid the ‘green bridge and protecting the soybean free-period, when soybean green plants should not exist on the field. This should be extended to all types of soybean cultivations in open areas, including the work of advancing generations and genetic breeding currently allowed in MT, and prohibiting commercial soybean planting for seed production during the free-period in other states whose environmental conditions may influence the Asian rust spread to the fields of the next season.

It should be considered that the FSS fields harvested before June do not enter the soybean free-period, and therefore, do not work as ‘green bridges’.

Regarding the practical importance of the mutations reported in *P. pachyrhizi*, the statement by Ishii and Hollomon (2015) is cited the knowledge of mutations cannot be used by growers.

The data on the fluctuation of the yearly rainfall (mm), in 10 days periods, were taken from weather stations located in two regions of Mato Grosso, South North and Center. Poxoreú (OMM: 83358), South; Latitude: 15°50', Longitude: 54°23', Altitude 450.00 m; Matupá (OMM: 83214), North; Latitude: 10°15', Longitude: 54°55', Altitude 285.00 m) and Diamantino (OMM: 83309), Center; Latitude: 14°24', Longitude: 56°27', Altitude 286.00 m) where data were collected (Figures 1, 2, and 3).

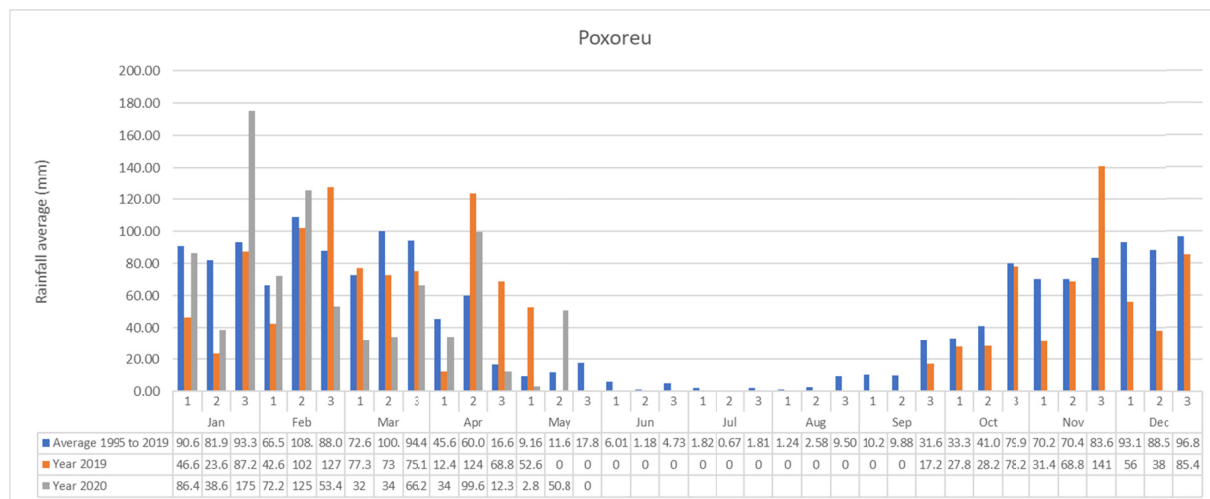


Figure 1. Rainfall average (mm) per ‘ten-day’ period from January to May 2020, compared to 2019 year and to the average from 1995 to 2019 in Poxoreu, MT. INMET. The blue bars represent the decennial historical average, from 1995 to 2018 and the oranges the rainfall occurred in 2019

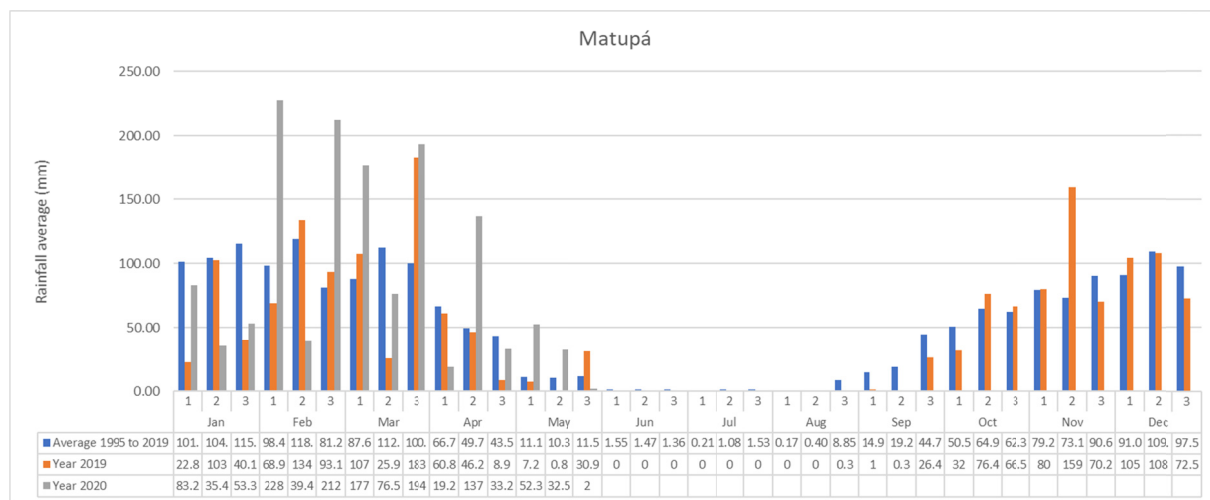


Figure 2. Rainfall average (mm) per ‘ten-day’ period from January to May 2020, compared to 2019 year and to the average from 1995 to 2019 in Matupá, MT. INMET. The blue bars represent the decennial historical average, from 1995 to 2018 and the oranges the rainfall occurred in 2019

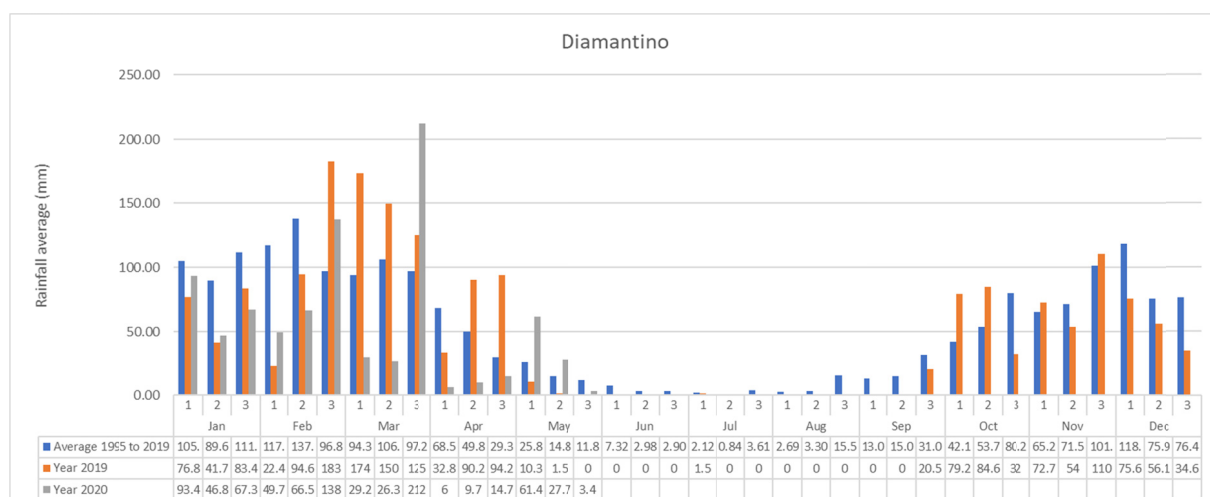


Figure 3. Rainfall average (mm) per ten-day period from January to May 2020, compared to 2019 year and to the average from 1995 to 2019 in Diamantino, MT. INMET. The blue bars represent the decennial historical average, from 1995 to 2018 and the oranges the rainfall occurred in 2019

The rain fall in DSS was 925.9 to 1573.7 mm in FSS from 582.3 to 1203.1 mm (Table 4). The wetting requirement period of the soybean leaves for *P. pachyrhizi* infection is more than 8 h with a temperature in this period between 18 to 27 °C (Melching et al., 1989; Reis et al., 2004). In our work the wet period was not measured, however it may be related to rain frequency, but infection does not relate to rain intensity or volume (Del Ponte et al., 2006).

Table 4. Monthly average and total rainfall (mm) from December (2019) to April 2020 and from February to May of 2020 during the soybean cycle in three regions of Mato Grosso state, INMET

Ten-day period	Rainfall (mm) (Year/month)									
	2019			2020					Total (Dec./Apr.)	Total (Feb./May)
	December	January	February	March	April	May				
<i>Poxoréu</i>										
1	56.0	86.4	72.2	32.0	34.0	2.8	280.6	141.0		
2	38.0	38.6	125.0	34.0	99.6	50.8	335.2	309.0		
3	85.4	175.0	53.4	66.2	12.3	0.0	392.3	131.9		
Total	179.4	300.0	250.6	132.2	145.9	53.6	1008.1	582.3		
<i>Matupá</i>										
1	105.0	83.2	228.0	177.0	19.2	52.3	612.4	476.5		
2	108.0	35.4	39.4	76.5	137.0	32.5	396.3	285.4		
3	72.5	53.3	212.0	194.0	33.2	2.0	565.0	441.2		
Total	285.5	171.9	479.4	447.5	189.4	86.8	1,573.7	1,203.1		
<i>Diamantino</i>										
1	75.6	93.4	49.7	29.2	6.0	61.4	253.9	146.3		
2	56.1	46.8	66.5	26.3	9.7	27.7	205.4	130.2		
3	34.6	67.3	138.0	212.0	14.7	3.4	466.6	368.1		
Total	166.3	207.5	254.2	267.5	30.4	92.5	925.9	644.6		

In the DSS and FSS the total rainfall was more than the soybean water requirement (450 mm) (Table 1). In contrast, the rainfall mean in the period from 1995 to 2019, during the soybean free-period (June to August), the mean rainfall was just 29.1 mm.

The same environmental condition, lower rain frequency from April in MT (Figures 1, 2 and 3) which contributes to the better efficiency of fungicides and lesser rust severity in FSS, is the same condition that can lead to reduction in productivity as there may be periods of drought at the grain filling phase, which results in smaller seeds and consequent lower yield. However, this does not affect the seed quality, which, despite having a 1,000 grains weight below the average of the cultivar, still their physiology is not impaired.

In this season, for example, although there were periods with rain above the historical average, it is observed that in all regions (Figures 1, 2 and 3) there was less rainfall in April and May than the historical series and this led to the significant reduction in productivity in some fields carried out in February. This did not happen in some irrigated areas where, although the irrigation system was activated, leaf wetting was not enough to increase the ASR progress.

Fields sown at the end of December always received higher volumes and frequency of rain in the subsequent months when most soybean fields are being harvested, which have already received 3-4 fungicide sprayings. Under high rain frequencies ASR control is even more difficult. The spores removed and disseminated from these field can be deposited on soybean weed plants amid cotton crops.

On the other hand, FSS occurs when most soybean fields have already been harvested. In the 2019/20 season reaching 68.7% of the crops harvested on February 8th, 2020 in the West, 64.21% in the North and more than 52% in the Southeast (IMEA, 2020b), which implies reduced inoculum pressure. If we consider the end of February, the time when seeding can still be done without going into the soybean free-period, 95.6%, 96.2% and 85.8% of the soybean crop had already been harvested respectively in the western, north and southeast regions of MT, reducing the inoculum pressure for new sowings that could still be done respecting the soybean free-period (IMEA, 2020b).

In addition to the rain lower frequency in the months of April onwards (Figures 1, 2 and 3), the rust intensity can be reduced due to the less favorable environmental conditions for its development and more efficient control. Yields on irrigated fields reached 3.0-3.4 tons/ha. In DSS, maximum yields were also obtained in the northern region, with up to 3.1 tons/ha.

After harvesting the December and February fields, the soybean free-period in MT starts on June 16th. During this time, the environment is adverse to the soybean plant (Figures 1, 2 and 3) and for *P. pachyrhizi* development.

The fungus spores are dry and are removed from uredia and transported by wind and deposited by sedimentation or impact by the wind on any surface. If they are deposited in soybean leaves and under more than 8h of continuous leaf wetness, low light intensity, they germinate, penetrate the leaves, initiating the infectious and sporulation process (Melching et al., 1989; Reis et al., 2004; Blum et al., 2015). On the other hand, these required conditions are not always met in the months following February sown, especially after April. Despite in the last two years (2019/2020) (Figures 1, 2 and 3) the rainfall in this month was higher than the historical average, even so, due the low rain frequency, the ASR was less severe and control more efficient.

3.4 Survival of Soybean Plants During the Period of Water Deficit

The total rainfall from June to August (90-day period) in the period from 1995 to 2019 in Poxoréu, MT was 29.5 mm, in Matupá 16.6 mm, and in Diamantino 41.3 mm (Figure 1, 2, 3), with an average of 29.1 mm.

The optimum soil temperature for soybean germination is 28 °C and the optimum soil moisture between 10 to 15% in the tested soil type (Tyagi & Tripathi, 1983). According to FAO (1986) the water requirements for maximum production of soybean ranged from 450-700 mm well distributed over the growing season, similar Farias et al. (2007) report considering also 450 mm as the minimum water requirement for soybeans to complete its cycle. Comparing those hydric requirements to the available water in Mato Grosso of 17-41 (mean 29.1) mm, we raise the hypothesis that should be no infected volunteer live soybean plants at the end of the soybean free-period in the farms (September 15th). The rust inoculum is unable to survive, except in irrigated areas, as it was before the soybean free-period has been implemented in MT.

However, soybean weed plants are found especially inside cotton crops, which, due to the inefficiency of the herbicides used in their control, germinate, and the new seedlings receive inoculum from the surrounding plants and their cycles entering the soybean free-period. This is certainly the main problems to be solved and not the excessive concern with the February sown soybean.

Spores produced in plants sown in February and harvested until May face an inhospitable environment to survive from June to September 15th. In addition to the water deficit, mainly the spore lethality by solar irradiation (Isard et al., 2006; Nicolini et al., 2010). During 90 days, for the reasons explained, no secondary cycles should occur under these adverse environmental conditions.

3.5 Environment Unfavorable to Infection

Finally, even in the presence of volunteer soybean plants in August, the requirements for infection > 8 h continuous leaf wetness (Melching et al., 1989) should not be fulfilled and deserve investigation. Surveys of soybean voluntary plants (weeds) in cotton fields (unpublished data) show that those born in January show the

greatest rust severity and decreasing in intensity in those born later. However, when such weed plants are born in sequence, they become infected by the inoculum coming from the older ones.

It can be inferred that the inoculum from within the state would be available mainly due to the presence of volunteer soybean plants during the soybean free-period.

The other hypothesis is that the inoculum annually arriving in MT state, may come from outside the state, specifically from authorized sown fields during the soybean free-period, and also from authorized experimental areas in this period of the year, within the MT.

Therefore, the advantages of soybean cultivation in February were the lowest rust intensity, likely due to the lowest rainfall frequency (Reis et al., 2020), great spore exposure to lethal solar radiation (Isard et al., 2006; Nicolini et al., 2010), plants height under a short photoperiod, resulting in better penetration of the fungicide mixture and, consequently, greater efficiency of the fungicides so that less spraying was required (Reis et al., 2020).

4. Conclusions

The continuous directional selection is taking place: (a) in the normal soybean season (September to December) in 10 million hectares through the use of site-specific fungicides (DMI, IQe, ISDH); (b) high number of spraying 5.0/area/season; and (c) the soybean as a weed present in cotton crops, 1.0 million hectares, infected by *P. pachyrhizi*, is certainly the one that may be contributing to the reduction of the fungus sensitivity to a greater degree than FSS fields. This serious situation must be considered by the State Plant Protection Service and be a research priority.

The results of our research indicate that the sowing period can be changed from the end of December to February, but always with the use of multisites fungicides. This change, in accordance with the basic principle of disease control, escape, implies a significant reduction in risks, less environmental damage and a reduction in economic costs and is still in accordance with the principles of IN 002/2015, especially with the preservation of the soybean free-period and avoiding the pressure of selection of Asian Rust, due the curative and large numbers of sprays, which occur in the end of the year sowing.

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