ECONOMIC AND AGRONOMIC POSITIONING OF FUNGICIDES IN CONDITIONS OF WATER DEFICIT IN SOYBEAN CULTURES †

[POSICIONAMIENTO ECONÓMICO Y AGRONÓMICO DE LOS FUNGICIDAS EN CONDICIONES DE DÉFICIT HÍDRICO EN CULTIVOS DE SOJA]

Christian Szambelam Zimmermann1, Ivan Ricardo Carvalho1*, Murilo Vieira Loro2, Aline Luiza Schmidt1, and Helaine Claire Ferreira de Almeida3

1Universidade Regional do Noroeste do Rio Grande do Sul, Departamento de Estudos Agrários, Avenida do Comércio, nº3.000, Bairro Universitário, CEP 98700-000, Ijuí, RS, Brazil. E-mail: cristian_zimmer@gmail.com, carvalhoivir@gmail.com, aline_schmidt@gmail.com
2Universidade Federal de Santa Maria, Centro de Ciência Rurais, Departamento de Engenharia Rural, Avenida Roraima, Cidade Universitária, Camobi, Prédio 42, CEP 97105-900, Santa Maria, RS, Brazil. nuviloloro@gmail.com.
3Federal University of Mato Grosso do Sul, Rodovia MS-306, Km105 - Zona Rural, Chapadão do Sul - MS, 79560-000, Brazil. claire_helaine@gmail.com. *Corresponding author

SUMMARY

Background. The management of diseases in soybean crops demands a great economic cost with fungicide applications. Therefore, unnecessary or excessive applications can promote negative environmental results. Therefore, understanding the weather conditions and the development of diseases in the crop is essential to avoid the unnecessary use of fungicides, consequently reducing costs and ensuring the sustainability of the activity. Aim. The aim is to identify which yield components are quantitatively affected, and which aspects of the chemical composition of the grains are qualitatively influenced by fungicide management, in addition to relating the cost-benefit rate of sequential applications or previously stipulated management in the soybean crop, as well as their economic effects under water deficit conditions. Methodology. The trial was carried out using a randomized block design composed of 4 replications. The trial consisted of 5 kinds of fungicide management, defined by the time and number of applications, during the development cycle of the Lance cultivar, in the 2019/2020 crop season. Soybean agronomic traits and nutritional attributes were evaluated. Results. Unfavorable climate conditions contributed to the low rate of Asian rust infection during the soybean development cycle. The different kinds of fungicide management influenced soybean performance for the main agronomic variables studied. For the chemical composition of the grains, there was no statistical difference in relation to the different kinds of fungicide management. Implications. The results indicate that the management of fungicides must be carried out according to the meteorological conditions of the year of cultivation. Conclusion. Management without fungicide application maximized grain yield and promoted the highest economic return. Key words: Glycine max; Positioning of fungicides; Grain yield.

RESUMEN

Antecedentes. El manejo de enfermedades en cultivos de soja demanda un gran costo económico con aplicaciones de fungicidas. Por lo tanto, las aplicaciones innecesarias o excesivas pueden promover resultados ambientales negativos. Por lo tanto, conocer las condiciones climáticas y el desarrollo de enfermedades en el cultivo es fundamental para evitar el uso innecesario de fungicidas, reduciendo consecuentemente costos y asegurando la sustentabilidad de la actividad. Objetivo. El objetivo es identificar qué componentes del rendimiento se ven afectados cuantitativamente y qué aspectos de la composición química de los granos se ven influenciados cualitativamente por el manejo de fungicidas, además de relacionar la relación costo-beneficio de las aplicaciones secuenciales o manejo previamente estipulado en el cultivo de soja. así como sus efectos económicos en condiciones de déficit hídrico. Metodología. El ensayo se llevó a cabo utilizando un diseño de bloques al azar compuesto por 4 repeticiones. El ensayo consistió en 5 tipos de manejo de fungicidas, definidos por el tiempo y...
número de aplicaciones, durante el ciclo de desarrollo del cultivar Lança, en la campaña 2019/2020. Se evaluaron las características agronómicas y los atributos nutricionales de la soja. **Resultados.** Las condiciones climáticas desfavorables contribuyeron a la baja tasa de infección por roya asiática durante el ciclo de desarrollo de la soja. Los diferentes tipos de manejo de fungicidas influyeron en el comportamiento de la soja para las principales variables agronómicas estudiadas. Para la composición química de los granos, no hubo diferencia estadística en relación a los diferentes tipos de manejo de fungicidas. **Implicaciones.** Los resultados indican que el manejo de fungicidas debe realizarse de acuerdo a las condiciones meteorológicas del año de cultivo. **Conclusión.** El manejo sin aplicación de fungicidas maximizó el rendimiento del grano y promovió el mayor retorno económico. **Palabras clave:** *Glycine max*; Posicionamiento de fungicidas; Producción de granos.

**INTRODUCTION**

Soybean (*Glycine max* L. Merril) is one of the main worldwide commodities. In Brazil it stands out as the crop with the largest extent of sown area, having bran and oil as by-products, used in human and animal food (Conah, 2019). During its cultivation, soybean is influenced by internal and external factors, due to soil characteristics, soil and climate components, genetic traits of the cultivar, phytosanitary management and insect-pest damage, diseases and weeds that ultimately determine its productive potential (Nanuci, 2020). In addition to these, climatic factors such as temperature and rainfall are decisive for the crop. Water deficit is defined as the lack of water accessible to the plant, which results in biological, chemical and biochemical modifications of the plant and seeds (Bormann, 2009).

With the advance of the crop in the sown area and due to failures when carrying out the direct sowing system and using a monoculture, there has been an increase of many phytosanitary problems that can compromise productivity, as well as the quality of the final product. Thus, the severity of some diseases, both aerial and those caused by soil fungi has been changing over the years. Therefore, we can highlight Asian rust (*Phakopsora pachyrhizi*) and some end-of-cycle diseases such as target spot (*Corynespora cassiicola*), leaf crests of cercospora (*Cercospora kikuchii*), brown spot (*Septoria glycines*), anthracnose (*Colletotrichum truncatum*) and powdery mildew (*Erysiphe diffusa*) as the diseases responsible for high costs with fungicide application (Ito, 2013; Barros, 2008). However, the importance of each disease varies between crop seasons, between regions and between properties of the same region, since it depends on a set of factors, such as the cultivar used, the sowing season, the level of technology used and especially the climatic conditions of each crop (Barros, 2008).

Asian rust, caused by fungus *Phakopsora pachyrhizi* is one of the most severe diseases that affects the soybean crop currently, due to its expansion capacity that occurs through the spread by the wind and due to rapid infection in the plant, which, depending on the degree of severity, can make the harvest unfeasible (Yorinori, 2004). Thus, in addition to some practices such as the use of resistant cultivars, the choice of the best sowing season, adjustment of plant density, crop rotation, treatment and use of seeds of quality and balanced fertilizations, the use of chemical fungicides has become essential, increasing the cost of production and requiring greater knowledge regarding crop monitoring, in addition to the use of more efficient products and dosages and greater information regarding the application technology (Godoy et al., 2009; Barros, 2008). When considering control methods, chemicals with the use of fungicides have been the most efficient method, since in addition to facilitating the application, they allow for results in the short term. However, in order for the chemical control to be efficient, field monitoring becomes a fundamental practice, with the identification of initial symptoms and preventive applications (Nanuci, 2020).

So, because many farmers have been experiencing a degree of resistance to a large number of applications of fungicides on their farms due to the high cost of the plant protection product and which will result in significant gains in the cost of production, there is a need for a study to answer some relevant questions to many farmers, such as what the principles that guide the decision of whether or not to apply a fungicide are, what time is best to perform the first application, the number of applications necessary to obtain the maximum profit, which application that is not carried out will cause the least damage (in the beginning or in the end).

Several recent studies have focused on showing results that may contribute to a more environmentally and economically sustainable management, especially in crops such as white oat (Dornelles et al., 2020; Dornelles et al., 2021) and soybean (Knebel et al., 2021), revealing that there is a great dynamic of relationships to position the management of fungicides. Thus, all these questions seek to evaluate to what extent the largest number of applications is profitable for the farmer and this way establish a relationship between the management that delivers the best action, that is, the best disease control associated with the lowest cost and maximum grain yield, especially in the case of a year with water deficit.

Therefore, the present work aims to demonstrate the best positioning of fungicides in the soybean culture, seeking to determine the maximum efficiency of action, weighted by the moment to start the phytosanitary management. It seeks to identify which yield components are affected quantitatively,
and which aspects of the chemical composition of the grains are influenced qualitatively by the management of fungicides, in addition to relating the cost-benefit rate of sequential applications or previously stipulated management in the soybean culture, as well as their economic reflexes in conditions of water deficit.

MATERIALS AND METHODS

The trial was carried out during the 2019/2020 crop season, in a property geographically located in the municipality of Ijuí, in the state of Rio Grande do Sul, Brazil, at 28°19’29”S 53°50’30”O, and an altitude of 320 meters. The soil is classified as typical dYROFERR red latosol (Embrapa, 2020). The climate, according to the Köppen climate classification, is of the humid subtropical type (Cfa).

The soybean genotype used was cultivar Brasmx Lança IPRO (58i60RSF IPRO), with maturation group 5.8.

The experimental design of randomized blocks with five treatments and four replications was used, totaling 20 experimental units. Each experimental unit consisted of six seeding lines spaced in 0.45 meters and ten meters long. The treatments consisted of five different methods of application of fungicides, defined by the moment of entry (phenological stage) and the number of applications, as shown in Table 1. For the fungicide applications, the combination of five different treatments using products of a systemic action and protective multisites were sought. Regardless of the number of applications performed, the same management order was followed, seeking to provide each treatment with the same condition, isolating only the effects of the time of entry and number of applications. During the performance of the trial the applications were carried out between 6:30 pm and 7 pm, that is, in periods with mild temperature conditions, seeking to minimize phytotoxicity problems, due to the predominance of high temperatures and low rainfall. The sprays were carried out at a height of 0.5 meters in relation to the canopy of the crop with the aid of a manual spray costal of the jetto brand, with a capacity of 20 liters, using a fan-type application tip.

The soybean sowing was carried out on November 16, 2019, with the aid of a Stara Seeder containing 6 sowing lines spaced in 0.45 meters, previously adjusted to distribute 14.7 seeds per linear meter. The fertilizer management was carried out with 300 kg ha⁻¹ of the Topmix 02-23-23 formulated fertilizer (Yara Evolution).

To collect the data, the two central lines of each experimental unit were used, discounting 1.0 meter between the blocks, avoiding edging effects. The evaluations were divided into two moments, that is, during the monitoring of the crop in the field where they were evaluated: initial and final plant population (IP, unit.; FP unit.); this procedure was developed with the aid of a tape measure, counting five meters and two meters from the centerline of each plot; chlorophyll content in the leaves (CHLORINE, %): for this procedure, the equipment used was cloroFiLOG, which allows an instant analysis of the chlorophyll content of the leaves; obtained through the measurement of four plants of the two centerlines of each experimental unit; this procedure was performed one day before and one day after each phytosanitary management, seeking to identify possible changes caused by the use of fungicides; number of flowers (NF, unit): obtained through the direct counting of four plants in the two centerlines of each experimental unit, when the culture was in the full flowering stage (R2 stage); plant tissue analysis: for this evaluation ten plants of each experimental unit were sampled and sent to the Bromatology Laboratory of the UNIJUÍ, where the percentage of protein in the tissue was determined (PPT, %); neutral detergent fiber in the tissue (NDFT, %); acid detergent fiber in the tissue (ADFT, %); lipids in the tissue (LIPT, %); mineral material in the tissue (MMT, %); lignin in the tissue (LIGT, %); carbohydrates in the tissue (CHOT, %); calcium in the tissue (CaT %); phosphorus in the tissue (PT %); potassium in the tissue (KT, %); magnesium in the tissue (MgT, %); sulfur in the tissue (ST, %).

The harvest of the experimental units was carried out on March 27, 2020, manually, collecting ten representative plants of each plot. The following was evaluated: plant height (PH, cm); height of the insertion of the first legume (HIFL, cm); number of branches (NB, unit.); number of legumes on the main stem (NLMS, unit.); number of legumes on the branches (NLB, unit.); number of legumes with one grain (NL1, unit.); number of legumes with two grains (NL2, unit.); number of legumes with three grains (NL3, unit.); number of legumes with four grains (NL4, unit.); grain mass per plant (GMP, g); number of grains per plant (NGP, unit.); mass of a thousand grains (MTG, g) and grain yield (GY, kg). From the grain production obtained from each treatment the percentage of green grains (GG, %) was evaluated, through the counting of four samples of 100 grains, and the grain analysis, carried out through NIRS (Nearinfrared Reflectance) from two samples for each treatment, in which the following was determined: the percentage of protein in the grain (PTNG, %); oil in the grain (OLG, %); fiber in the grain (FBG, %); mineral material in the grain (MMG, %); palmitic acid in the grain (APALMG, %); stearic acid in the grain (AESTG, %); oleic acid in the grain (AOLG, %); linoleic acid in the grain (ALINOG, %); linolenic acid in the grain (ALENICG, %).

The data were submitted to the assumptions of the statistical model, normality, homogeneity, independence and additivity of the model. Subsequently, the analysis of variance at 5% probability was performed by the F test, for the effects of fungicide management. The variables that
showed significance were dismembered to the main effects through the grouping of means by Scott Knott. In order to identify the tendency of association among the traits, the linear correlation was determined at 5% probability by the t test. The mean Euclidean distance was estimated and the matrix was submitted to the UPGMA grouping. Eigenvalues and eigenvectors were extracted for the effects of variables and fungicide management, and based on this, the Biplot analysis of the principal components was carried out. All statistical analyses were performed by the Rstudio software, using packages ggplot2, metan, Agricolae and Exp. Des. Pt.

**RESULTS AND DISCUSSION**

According to the meteorological data (Figure 1), the occurrence of water deficit can be observed in the crop season, since during the experimental period there was a total accumulated rainfall of 264,0 mm, of which approximately 187 mm was in the reproductive stage under poorly distributed rains, especially during grain filling; this rainfall volume is considered to be less than the minimum amount required by the crop to obtain satisfactory yield. Soybean has a total water demand from 450 to 850 mm, and during the development of the crop the period of greatest water demand occurs during flowering and the filling of grains, demanding between 7 and 8 mm day\(^{-1}\). (Farias et al., 2007; Carvalho et al., 2013). The effects of water deficit on plants are linked to changes in their morphological and physiological traits as a way to adjust themselves to the conditions of the external environment. Among these modifications the ones that stand out are the reduction of the foliar water potential, the stomatal closure, the decrease of the photosynthetic rate, the reduction of the aerial part, acceleration of the senescence and abscission of leaves, resulting in less developed plants with lower grain yield (Ferrari et al., 2015).

### Table 1. Description of treatments with trade name of fungicides, active ingredients (i.a.), application dosages and time of application.

<table>
<thead>
<tr>
<th>Management</th>
<th>Products/Dose</th>
<th>Application</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>I</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1st application: 0,4 L ha(^{-1}) Fox (Protioconazol + Trifloxistrobina) + 1,5 Kg ha(^{-1}) Unizeb Gold (Mancozebe) + 0,4 L ha(^{-1}) Aureo (mineral oil)</td>
<td>33 DAS</td>
<td></td>
</tr>
<tr>
<td>2nd application: 500 ml ha(^{-1}) Fox XPRO (Protioconazol + Trifloxistrobina + Bixafem) + 1,5 Kg ha(^{-1}) Unizeb Gold (Mancozebe) + 0,4 L ha(^{-1}) Aureo (mineral oil)</td>
<td>48 DAS</td>
<td></td>
</tr>
<tr>
<td>3rd application: 0,35 L ha(^{-1}) Sphere Max (Trifloxistrobina + Ciproconazol)) + 1,5 Kg ha(^{-1}) Unizeb Gold (Mancozebe) + 0,4 L ha(^{-1}) Aureo (mineral oil)</td>
<td>63 DAS</td>
<td></td>
</tr>
<tr>
<td>4th application: 0,6 L ha(^{-1}) Fusão (Metominostrobina + Tebucanon) + 1,5 Kg ha(^{-1}) Unizeb Gold (Mancozebe) + 0,4 L ha(^{-1}) Aureo (mineral oil)</td>
<td>78 DAS</td>
<td></td>
</tr>
<tr>
<td>5th application: 0,35 L ha(^{-1}) Sphere Max (Trifloxistrobina + Ciproconazol)) + 400 ml ha(^{-1}) Aureo (mineral oil)</td>
<td>95 DAS</td>
<td></td>
</tr>
<tr>
<td><strong>II</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1st application: 0,4 L ha(^{-1}) Fox (Protioconazol + Trifloxistrobina) + 1,5 Kg ha(^{-1}) Unizeb Gold (Mancozebe) + 0,4 L ha(^{-1}) Aureo (mineral oil)</td>
<td>47 DAS</td>
<td></td>
</tr>
<tr>
<td>2nd application: 500 ml ha(^{-1}) Fox XPRO (Protioconazol + Trifloxistrobina + Bixafem) + 1,5 Kg ha(^{-1}) Unizeb Gold (Mancozebe) + 0,4 L ha(^{-1}) Aureo (mineral oil)</td>
<td>62 DAS</td>
<td></td>
</tr>
<tr>
<td>3rd application: 0,35 L ha(^{-1}) Sphere Max (Trifloxistrobina + Ciproconazol)) + 1,5 Kg ha(^{-1}) Unizeb Gold (Mancozebe) + 400 ml ha(^{-1}) Aureo (mineral oil)</td>
<td>77 DAS</td>
<td></td>
</tr>
<tr>
<td>4th application: 0,6 L ha(^{-1}) Fusão (Metominostrobina + Tebucanon) + 1,5 Kg ha(^{-1}) Unizeb Gold (Mancozebe) + 0,4 L ha(^{-1}) Aureo (mineral oil)</td>
<td>95 DAS</td>
<td></td>
</tr>
<tr>
<td><strong>III</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1st application: 0,4 L ha(^{-1}) Fox (Protioconazol + Trifloxistrobina) + 1,5 Kg ha(^{-1}) Unizeb Gold (Mancozebe) + 0,4 L ha(^{-1}) Aureo (mineral oil)</td>
<td>52 DAS</td>
<td></td>
</tr>
<tr>
<td>2nd application: 500 ml ha(^{-1}) Fox XPRO (Protioconazol + Trifloxistrobina + Bixafem) + 1,5 Kg ha(^{-1}) Unizeb Gold (Mancozebe) + 0,4 L ha(^{-1}) Aureo (mineral oil)</td>
<td>68 DAS</td>
<td></td>
</tr>
<tr>
<td>3rd application: 0,35 L ha(^{-1}) Sphere Max (Trifloxistrobina + Ciproconazol)) + 1,5 Kg ha(^{-1}) Unizeb Gold (Mancozebe) + 400 ml ha(^{-1}) Aureo (mineral oil)</td>
<td>83 DAS</td>
<td></td>
</tr>
<tr>
<td><strong>IV</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1st application: 0,4 L ha(^{-1}) Fox (Protioconazol + Trifloxistrobina) + 1,5 Kg ha(^{-1}) Unizeb Gold (Mancozebe) + 0,4 L ha(^{-1}) Aureo (mineral oil)</td>
<td>62 DAS</td>
<td></td>
</tr>
<tr>
<td>2nd application: 500 ml ha(^{-1}) Fox XPRO (Protioconazol + Trifloxistrobina + Bixafem) + 1,5 Kg ha(^{-1}) Unizeb Gold (Mancozebe) + 0,4 L ha(^{-1}) Aureo (mineral oil)</td>
<td>77 DAS</td>
<td></td>
</tr>
</tbody>
</table>

DAS: days after sowing; NA: no application; Spray volume of 100 L ha\(^{-1}\)
Soybean adapts better in regions where temperatures fluctuate between 20 and 30°C, and the ideal temperature for its development is around 30°C (Farias et al., 2007). At temperatures below 10°C and above 40°C, adverse effects occur to metabolism reducing growth, and temperature extremes, when associated with water deficit, are even more aggravating to plant development (Ferrari et al., 2015). Therefore, it is noticed that during the vegetative stages of the culture the temperature fluctuated from 11°C to 38°C, not reaching the critical limits. During the stages of flowering and filling of grains it is noted that the maximum temperature was close to the thermal preference of the crop, not exceeding 35°C, and the minimum temperature obtained a decline, reaching the peak of 8°C at the end of the filling of grains (100 DAS), reaching lower limits than the base temperature of the crop.

Asian rust, as well as other fungal diseases are influenced by the presence of water in the liquid phase and high temperatures. Water from foliar wetting is considered the most important variable for the development of diseases, since most pathogens require the presence of liquid water on plants for spore germination and tissue infection. Air temperature is also a determining factor for the infection process, and temperatures favorable to the development of soybean are also suitable for the development of rust. Therefore, it is estimated that the minimum period of foliar wetting for infection to occur is 6 hours, associated with temperatures between 20°C and 25°C (Lelis et al., 2009). This information, together with the meteorological data of the 2019/2020 crop season, provides an explanation for the low incidence of diseases such as Asian rust on the soybean crop.

During the applications, temperature and humidity are determining factors to ensure greater efficacy of fungicides and avoid antagonistic effects on plants. According to the meteorological data, it is noticed that during the applications (AP), most of the fungicide management was hindered due to the presence of higher temperatures during the management, with maximum temperatures above 30°C and because the plants are in stress conditions due to low soil moisture. In conditions of water deficit, the plant tends to express different morphological and biochemical responses, as a form of resistance to the dehydration process, which can confer different results regarding the diffusion of the fungicide in leaves, and the pathogenicity of fungi (Stefanello, 2014). These changes due to water stress, together with high temperatures can increase problems, such as phytotoxicity due to the difficulty of the plant in metabolizing the active ingredient. According to Azevedo and Freire (2006), the ideal conditions for the application are temperatures above 15°C and below 32°C, with minimum relative humidity around 50-55%.

The evaluation of the chlorophyll content of the plants is an indication of the sanity of the leaf tissue, revealing stress situations by pests, diseases, nutrition and climatic conditions such as water deficit and high temperatures, or it is also the response of the plant to the application of a plant protection product. Thus, higher chlorophyll levels are associated with reduced yellowing and senescence of plants, prolonging the period of the photosynthetic activity, and it may contribute to greater mass accumulation at the end of the cycle (Alves and Juliatti, 2018). Therefore, by analyzing the chlorophyll content of plants one day after the application of the fungicide, it was possible to identify that for most of the treatments there was an increase in the chlorophyll content of the plants. However, it is not possible to state that fungicides caused increases in the chlorophyll content of the plants one day after application, as similar effects were observed for management absence of application (MI), and this variation may result from the evaluation of different plants during the performance of the trial. Similar effects were observed by Alves and Juliatti (2018) when they evaluated different fungicides for the management of Asian rust in soybean, in which the application of mancozeb alone or in combination with other fungicides have provided increases in the concentration of chlorophyll-b; this difference is not related to the severity of the disease, but it is due to the existence of a mechanism for the protection of the photosynthetic apparatus. For Marques (2017), the use of mancozeb increases the concentrations of sulfur (S), manganese (Mn) and zinc (Zn) in the tissues, enabling increases in the green coloration of the culture.

In addition, there is a small increase in the final chlorophyll content of the plants submitted to management absence of fungicide (MI). A similar result was reported by Mochko (2017), when comparing treatments with the absence and the presence of chemical control for Asian rust in soybean, in which it was found that plants submitted to fungicide application presented a lower chlorophyll content if compared to plants without disease control. During the month of January (50 days after sowing) with the beginning of the reproductive stage, a reduction in the chlorophyll concentration of plants was observed, possibly being associated with chlorophyll degradation, due to the low water regime and high temperatures with maximums above 35°C. According to Streir et al. (2005), the beginning of chlorophyll degradation in senescent tissues is initiated through external factors such as water stress, changes in temperature, luminosity, increased levels of ethylene or due to the combination of these factors. From 60 days after sowing (DAS) with reductions in temperature maximums and resumption of rainfall, there was a gain in chlorophyll content exceeding the limits previously reached.
Figure 1. Description of meteorological variables during the conduction of the experiments and evaluation of chlorophyll in soybean plants. A: Meteorological data (November 16, 2019 to March 27, 2020) of maximum temperature (Tmax, °C), average temperature (Tmed, °C), minimum temperature (Tmin, °C). B: maximum relative humidity (Urmax, %), minimum relative humidity (Urmin, %) and precipitation (Prec, mm). C: Chlorophyll content evaluated one day before (Before) and one day after (Later) of each fungicide management during the soybean crop cycle. Source: IRDeR weather station 2019/2020.

The analysis of variance showed that for the variation factor of fungicide management, there was a significant difference at 5.00% of probability (p-value<0.05) for all variables. The mean grouping test by Scott Knott (Figure 2), performed for variables plant height (PH), height of the insertion of the first legume (HIFL), number of branches, number of legumes on the main stem (NLMS), grain mass per plant (GMP) and grain yield (GY), revealed that when there was a significant difference the best results were obtained for management I. In general, when analyzing variable plant height (PH) it is
noted that management MI, MII, MIV and MV stood out, obtaining the largest gain in height, being the smallest size obtained for the plants that were submitted to management MI. For trait height of the insertion of the first legume (HIFL), it is noted that the MII, MIII, MIV and MV treatments obtained their zone of production started at a higher level from the soil when compared to the plants that received the MI management. The number of branches (NB) was boosted when the first application was started later with a smaller number of kinds of management, highlighting MI, MIV and MV management, and the early or anticipated management with the use of a greater number of total applications resulted in lower lateral growth. Variable number of legumes on the main stem (NLMS) was maximized with management MI and MII, with the lowest tendencies obtained for management MIII, MIV and MV, which obtained lower results.

When evaluating the initial and final plant population ratio (IFR), it is noted that for most of the management there was a reduction in the final population (FP) if compared to the initial population (IP), which is directly associated with plant death during the development of the culture, and management MI, MIII and MV presented statistically equal results with the largest final population, while management MIV differed by the smallest number of final plants. For grain mass per plant (GMP) and grain yield, the same tendencies were obtained, with the best result obtained for management MI (absence of application) and the lowest averages with statistically equal results were obtained for management MIII, MIV and MII, and MV. Among the worst averages, management MIII stands out, with the grain mass per plant of 8.25 g and grain yield of 2017 kg ha\(^{-1}\). This result can be explained due to the phytotoxicity problems caused as a result of the late application of fungicides with higher concentration of triazoles as observed in management MIII, which, in its last application performed during the soybean reproductive stage (R4-R5), received the management with the fusion fungicide (Methomimosrobin + Tebuconazole). According to Barroso (2008), some cultivars are more sensitive to some fungicides, and stress conditions for the plant, such as the lack of soil moisture and air associated with high temperatures, increase the chances of phytotoxicity problems.

When contrasting the study ends, which are found in management MI and MII, distinct effects are perceived. Therefore, for this study, the absence of fungicide application resulted in higher plant height (PH), reduction of the insertion of the first legume (HIFL), a greater number of branches, maintaining good legume production on the main stem (NLMS), resulting in higher averages in the grain mass per plant (GMP) and grain yield (GY) of 10.58 g and 2586 kg ha\(^{-1}\). For management MII, which comprised five applications with the first one in the V4 stage, an increase in the total height of plants (PH) was observed; therefore, the insertion of the first legume was at a higher level and with fewer branches, and the productivity was formed with greater representativeness on the main stem due to a greater predominance of legumes, resulting in grain mass per plant of 9.36 g and grain yield of 2287 kg ha\(^{-1}\); this is a lower result, when compared to the management with no application. According to Navarro Júnior and Costa (2002), the most important components in determining soybean crop yield are the number of legumes per unit of area, number of grains per legume and average grain mass. Thus, it is noticed that the increase in the number of fungicide applications in years with water deficit does not result in benefits, also obtained in crops with high rainfall, due to the lower incidence of diseases, because the optimal environmental conditions for the development of the pathogen were not established. Lichstbn & Godoy (2006), in a study aimed at evaluating the effect of fungicide application on the morphology and content of epicuticular wax, in two coffee cultivars, observed that the application of fungicide decreased the wax content and changed its morphology, causing ruptures and disappearance of crystalloids, making the plant more susceptible to diseases, pests and water stress. In addition, the low water volume causes the plants to change some of their structures as a way to reduce loss and optimize the use of water, such as the early closure of the stomata, increased oxidative stress (H\(_2\)O\(_2\)), which acts as an antifungal agent, and leaves with more cutinized walls of the epidermis and with greater layers of waxes. These changes hinder the process of infection of the fungus, delaying the appearance of the first rust pustule, reducing the severity in the plants (Stefanello, 2014.)

Therefore, when observing the tissue analysis, it is noted that management MI stood out for variable percentage of protein in the tissue (PPIT), which would probably be linked to the higher nitrogen content in the tissue. In addition, it can be noted that this management presented a lower percentage of total minerals in the tissues when compared to the MII and MIV management, with most of the minerals formed by calcium, phosphorus, magnesium and sulfur. According to Crusciol et al. (2001), nitrogen and magnesium are part of the chlorophyll molecule, which is formed by four nitrogen atoms linked to one magnesium atmo, and the chlorophyll synthesis is fundamental for the interception of solar radiation during the photosynthetic process. These results corroborate with the results found, in which the chlorophyll content was superior for the MI management; in addition, they help to explain the responses obtained for this treatment as to the maximization of grain yield.

For the MII and MIV management, different responses are observed in relation to the MI
management, due to the lower concentrations of proteins in the leaves, which may be related to a lower nitrogen content. In addition, increases in the percentage of lipids in the tissue (LIPT), reduction in the content of lignin (LIGT), increase in the percentage of carbohydrates in the tissue (CHOT) and increase in mineral material (MMT) such as phosphorus (PT) and potassium (KT) are observed. Potassium is the most abundant cation in the cytoplasm, which acts on the maintenance of the osmotic potential of the cells and tissues, as an enzymatic activator, and also on nitrogen absorption and protein synthesis (Cruz et al., 2013). A lower concentration of protein in the tissue (PPIT) was observed when the MV management was performed; along with this response, an increase in the content of tissue lipids (LIPT) and lignin (LIGT) was noted, which may be related to the increase of waxes and lignification of the tissues, as a defense response of the plant to the stress caused by low rainfall, high temperatures or even due to the effect of fungicide management in plants that already came from a stress. Some results showed a certain similarity between the MV and MIII treatments, which presented low concentrations of mineral material in the tissue (MMT) when compared to the other treatments, with high calcium and magnesium levels and low phosphorus, potassium and sulfur levels in the tissue. It is noticed that for these treatments there were increments for variables neutral detergent fiber in the tissue (NDFT), acid detergent fiber in the tissue (ADFT) and lignin content in the tissue (LIGT). Therefore, these results are in agreement with the ones by Cruz et al. (2013), in which the levels of ND and DF are directly related to the concentration of calcium in the plant, since this nutrient is essential for the structural integrity of the cell wall.

In order to show possible modifications in grain quality due to different kinds of fungicide management, the mean grouping test by Scott Knott was performed for the variables that demonstrated a significant effect in the analysis of variance. The chemical composition of soybean grains is influenced by genetic factors and by the environmental conditions during cultivation. This can be explained through the carbon balance of photosynthesis and nitrogen, since under normal conditions these two elements are interconnected, in which the carbon demand by the grain depends on the availability of nitrogen. Therefore, the imbalance of these two elements may result in different responses to the chemical composition of the grains (Pipolo et al., 2015).

Thus, it is noticed that the differences contained for variables percentage of protein in the grain (PTNG), oleic acid in the grain (AOLG) and linolenic acid in the grain (ALENICG) were not expressive, and there was the formation of a single grouping for the evaluated treatments, demonstrating that the fungicide management did not influence the chemical composition of the grains. However, for variable percentage of green grains (GG), the formation of three different groupings was identified, and management MV presented the largest number of green grains with an average of 50%; next, with statistically equal effects, management MI and MIII with respectively 43% and 42%, and MII and MIV were the ones that presented the lowest percentage with 39% and 37% of green grains. The percentage of green grains varies depending on the type, intensity and when stress occurs, which is associated with the premature death or forced maturation of the plant. The occurrence of green grains is usually associated with the attack of diseases, insects or by the water deficit associated with high temperature conditions during the final phases of grain filling and maturation (Neto et al., 2005). Under normal conditions, the chlorophyll content decreases during the maturation process due to the activity of the chlorophyllase enzyme, which degrades the chlorophyll, forming the yellowish color of the grains or seeds. Therefore, when the grain maturation process is forced by the premature death of the plant, the chlorophyllase enzyme may cease its activity before all the chlorophyll is degraded, resulting in the formation of greenish-looking grains (Rangel et al., 2011).

Therefore, the presence of green grains is linked to the occurrence of water deficit associated with high temperatures between the filling and the maturation of the crop, which caused the premature death of the plants, anticipating the harvest by approximately 15 days. The difference in the percentage of green grains (GG) between the management may be linked to the effects of fungicides on the plants that were subjected to stress conditions. According to Pinto et al. (2011), when evaluating the influence of the application of fungicide on the occurrence of green seeds in soybean, it was noticed that the application of fungicide had no effect on this variable, and it may be related to the conditions of mild temperatures and high relative air humidity during the crop development.

The Pearson linear correlation estimate (Figure 3) revealed the magnitude and sense of the associations between morphological traits, which act as direct and indirect components in yield and bromatological traits linked to the chemical composition of grains and plant tissue. Estimates of linear relationships between soybean traits are of great importance for the indirect selection of traits with low heritability or difficult to measure. Studies carried out by Ferrari et al. (2018), Nardino et al. (2016) and Carvalho et al. (2021) have shown some relationships, however, few results are presented with managements without fungicide.

In this context, for the 13 variables, 78 associations were revealed, of which 11 are significant. Therefore, the correlation revealed a strong positive significant association ($r=0.96$) between variable...
Figure 2. Scott-Knott mean clustering test for variables plant height (PH), height of the insertion of the first legume (HIFL), number of branches (NB), number of legumes on the main stem (NLMS), grain mass per plant (GMP), grain yield (GY), initial population (IP), final population (FP), ratio of initial and final population (IFR), protein in the tissue (PPIT), acid detergent fiber in the tissue (ADFT), neutral detergent fiber in the tissue (NDFT), lipids in the tissue (LIPT), mineral material in the tissue (MMT), lignin in the tissue (LIGT), carbohydrates in the tissue (CHOT), calcium in the tissue (CaT), phosphorus in the tissue (PT), potassium in the tissue (KT), magnesium in the tissue (MgT), sulfur in the tissue (ST), percentage of protein in the grain (PTNG), oleic acid in the grain (AOLG), linolenic acid in the grain (ALENICG), green grains (GG) of the soybean culture subjected to different kinds of management fungicide. MI: management I; MII: management II; MIII: management III; MIV: management IV; MV: management V.
number of legumes composed of two grains (NL2) and the mass of one thousand grains (MTG), in which the increase of these legumes may have directly contributed to the individual mass gain during the grain filling process. According to Weber (2017), high yields would be achieved with two grains per legume, 1950 legumes per m² and 190 g mass of one thousand grains. According to Navarro Júnior and Costa (2002), the average grain mass is genetically determined, but it is influenced by the environment. In addition, a strong positive correlation (r=0.99) was identified between grain yield (GY) and the number of grains per plant (NGP). A study carried out by Souza et al. (2015) agrees with the results found, highlighting that different climate conditions between two crop seasons influenced the number of grains per plant, which directly affects soybean grain yield.

Negative associations were observed between variables neutral detergent fiber in the tissue (NDFT) and percentage of phosphorus in the tissue (PT) (r=-0.87). The percentage of carbohydrates in the tissue (CHOT) showed a strong positive association with the percentage of potassium in the tissue (KT) (r=0.94) and a negative one with the percentage of magnesium in the tissue (MgT) (r=-0.95). The percentage of phosphorus in the tissue showed a significant positive tendency with the percentage of potassium in the tissue (KT) (r=0.96) and with the percentage of oil in the grain (r=0.88). According to Souza (2012), when evaluating different sources and doses of phosphorus, it was observed that the treatments did not influence the soybean oil content, possibly because this nutrient was not directly associated with the reserve lipid biosynthesis. Trait percentage of oil in the grain showed a strong positive significant association with the percentage of palmitic acid in the grain (APALMG) (r=0.88), and a negative one with the percentage of oil acid in the grain (AOLG), (r=-0.99), that is, the increase in the oil content in the grains can contribute to increases in the concentration of palmitic acid and a decrease in the oleic acid. Just like the percentage of

Figure 3. Pearson linear correlation estimates, significant at * 5.0%, ** 1.0% and *** 0.05% probability by the t test. number of legumes with two grains (NL2); grain yield (GY); number of grains per plant (NGP); mass of a thousand grains (MTG); oil in the grain (OLG); acid palmitic in the grain (APALMG); oleic acid in the grain (AOLG); neutral detergent fiber in the tissue (NDFT); carbohydrates in the tissue (CHOT); phosphorus in the tissue (PT); potassium in the tissue (KT); magnesium in the tissue (MgT); chlorophyll (CLORO).
oil in the grain, the percentage of palmitic acid showed a negative correlation with the percentage of oleic acid in the grain (AOLG), \( r = -0.91 \). According to Pipolo et al. (2015), high temperatures during soybean grain filling contribute to the increase in oleic acid concentration, while lower temperatures increase the concentration of linoleic and linolenic acids. For industrial processing, the production of oils with greater oxidative stability is sought, that is, with higher oleic acid contents, and lower linoleic and linolenic acid contents, improving their usefulness at higher temperatures, for pharmaceutical purposes, cosmetics and industrial products such as biodiesel (Bellinasso et al., 2021; Figueiredo, 2017).

The chlorophyll content of the leaves (CLORO) correlated positively with variables percentage of phosphorus in the tissue (PT) \( (r = 0.90) \) and with the oil content of the grains (OLG), \( (r = 0.88) \). Possibly, higher leaf chlorophyll levels may be related to plants with greater capacity to intercept solar radiation, as well as higher nitrogen input in the leaves, resulting in a greater photosynthetic activity, biomass conversion and grain production. According to Pipolo et al. (2015), the balance of the carbon supply of photosynthesis and nitrogen affects the chemical composition of the grain, which may explain the variations in protein and oil concentrations due to environmental factors. Stefanello (2014), when evaluating the behavior of fungicides in soybean plants, observed that the chlorophyll content of the leaves correlated positively with the mass of one hundred grains and the grain yield of the crop, reporting that grain growth is dependent on the export of leaf photoassimilates.

We performed the analysis of the average Euclidean Distance in order to show the dissimilarity among five different fungicide treatments, which were determined by the time and the number of applications. However, when analyzing the dendrogram (Figure 4A) it can be identified that there was the formation of two large distinct groups, one formed by management M1 (absence of application) and the other by management MIV, MII, MV and MIII, the latter being the most dissimilar management within the second group, differing for variables plant height (PH), initial plant population (IP) and the ratio between the initial and final plant population (IFR). For this second group, MII and MIV are distinguished with greater similarity to each other, with statistically equal results for variables: height of the insertion of the first legume (HIFL), plant height (PH), grain mass per plant (GMP), grain yield (GY), protein in the grain (PTNG), oleic acid in the grain (AOLG), linolenic acid (aleticg), ratio between the initial and final population (IFR), percentage of green grains (GG), acid detergent fiber (ADFT), neutral detergent fiber (NDFT), lipids in the tissue (lipt), mineral material in the tissue (MMT), lignin in the tissue (LIGT), carbohydrates in the tissue (CHOT), calcium in the tissue (CaT), phosphorus in the tissue (PT), potassium in the tissue (KT) and magnesium in the tissue (MgT). Therefore, it was observed that the management with the absence of fungicide application was the only one that did not show similarity with the other kinds of management, distinguishing for variables height of the insertion of the first legume (HIFL), grain mass per plant (GMP) and grain yield (GY).

We performed the analysis of the principal components (BI PLOT) in order to select the management of fungicides according to the variable to which it correlates (figure 4B); four distinct responses regarding the tendencies for each treatment in relation to the variables analyzed are noted, and this analysis allows an explainability of the data set of 71.1%. Thus, the use of the M1 management during the development of the soybean culture showed a greater tendency for variables number of grains per plant (NGP), grain mass per plant (GMP), acid detergent fiber in the tissue (ADFT), number of legumes on the main stem (NLMS) and number of legumes with two grains (NL2). Therefore, it is noted that this treatment acted positively on the mass of grains per plant (GMP), which may be related to the maximization of variables number of legumes on the main stem (NLMS), number of branches (NB) and increase of the production zone by reducing the height of the insertion of the first legume (HIFL).

The use of the MII management allowed greater affinity for variable mineral material in the tissue (MMT), which may be related to the maximization of phosphorus and potassium levels in the tissue. Greater tendencies were observed for the percentage of linoleic acid in the grains (ALINOG), which is negatively correlated to the increase in oleic acid concentrations. The management also demonstrated affinity for ratios between the initial and final population (IFR) and percentage of protein in the grain (PTNG). Proteins occupy about 40 % of the grain composition, and this is influenced by genetic and environmental traits during grain filling (Brunini et al., 2016). The MIII management demonstrated greater affinity with variables percentage of mineral material in the grain (MMG), percentage of stearic acid in the grain (AESTG) and number of branches (NB). The soybean grain is rich in minerals that are divided into macro and micronutrients, and the increase of these are of paramount importance in human and animal food (Bellinasso et al., 2021). For management MIV, the variables that stood out were: percentage of carbohydrates in the tissue (CHOT), which is positively influenced in the treatment, and neutral detergent fiber in the tissue (NDFT). In management MV, a greater tendency was identified for variables percentage of lipids in the tissue (Lipt), percentage of phosphorus in the tissue (PT), which can be negatively influenced, and percentage of lignin in the tissue (LIGT).
Figure 4. A: Dendrogram with the dissimilarity among different kinds of fungicide management for 13 variables measured in the soybean culture, using the mean Euclidean distance and the UPGMA grouping method. B: Principal components (BIPLOT). Height of insertion of the first legume (HIFL); plant height (PH); number of branches (NB); number of legumes on the main stem (NLMS); number of legumes on the branches (NLB); number of legumes with one grain (NL1); number of legumes with two grains (NL2); number of legumes with three grains (NL3); mass of grains per plant (GMP); grain yield (GY); number of grains per plant (NGP); mass of a thousand grains (MMG); protein in the grain (PTNG); oil in the grain (OLG); fiber in the grain (FBG); mineral material in the grain (MGG); acid palmitic in the grain (APALMG); stearic acid in the grain (AESTG); oleic acid in the grain (AOLG); linoleic acid in the grain (ALINOG); linolenic acid (ALENI); number of flowers (NF); initial population (IP); final population (FP); initial and final population rate (IFR); green grains (GG); protein in the tissue (PPIT); neutral detergent fiber in the tissue (NDFT); acid detergent fiber in the tissue (ADFT); lipids in the tissue (LIPT); mineral material in the tissue (MMT); lignin in the tissue (LIGT); carbohydrates in the tissue (CHOT); calcium in the tissue (CaT); phosphorus in the tissue (PT); potassium in the tissue (KT); magnesium in the tissue (MgT); sulfur in the tissue (ST).

Through calculations, the cost-benefit of the use of different fungicide positions in the soybean crop was calculated, in order to allow a broader viewing of the representativeness of each kind of management in the production cost of the crop, and through this to identify the management that delivers the greatest economic return to the farmer, especially in years with unfavorable climatic conditions to phytosanitary management, with the predominance of water deficit, as observed during the performance of this trial. For the purposes of calculations, the amount of R$ 90.00 was considered for the soybean bag; in addition, all inputs used, such as: seeds, fertilizers, insecticides, acaricides, mineral oil and fungicides were accounted for; the value of the products and of the soybean which were predominant during the experimental period of the 2019/2020 crop season were considered. Therefore, when analyzing the data, it is noticed that fungicide management entails significant increases in the cost of production for the farmer, having a representativeness of 32%, 29%, 25% and 19% in the total cost of the crop when five, four, three and two fungicide applications are carried out, respectively. These results become even more significant in crops in which low rainfall indexes are predominant, in which the productive capacity of the crop is limited, and any expensive expenditure entails significant effects on the profitability of the farmer.

Thus, there is no recipe for optimal phytosanitary management, but one that delivers to the farmer the most efficient control associated with greater profitability, because each crop has a distinct behavior in the predominance of diseases that are strongly associated with climatic conditions. For pathologies such as soybean Asian rust to develop, the disease triangle is necessary, that is, an interaction among the host, pathogen and the environment; if one of these does not interact with the other two the disease does not develop (Lelis et al., 2009). Gardiano et al. (2020), in a work with 13 different fungicide treatments, found that the application of one to seven days after the detection of the first spores presented a lower percentage of infected leaf area if compared to the treatments that received the application later. All of these reinforce
the importance of monitoring from the beginning of the crop cycle, to determine the interval between applications and when to perform the first application, as this is decisive for the success of the control, since after its establishment and having favorable conditions, rust can cause significant losses in productivity (Godoy et al., 2009).

In addition to losses related to the occurrence of diseases, losses caused by the incorrect positioning of fungicides in years of water deficit and high temperatures can also be a problem faced by many farmers, even though they are often unnoticed. Therefore, there is a limitation of studies related to crops with atypical climatic conditions regarding the functionality of phytosanitary management, as well as their effects on plants subjected to water stress. In addition to negatively influencing productivity, the management of fungicide poorly positioned in years with stress conditions end up causing unnecessary expenses to the farmer. However, for crops that have rainfall rates below the demand requirement of the culture, coupled with high temperatures and low relative humidity of the air, it is necessary to carry out studies with the use of lighter, less harmful molecules to the plants, focusing on the use of strobilurins associated with protection products such as mancozeb, since unfavorable climate conditions and the occurrence of fungal diseases allow the reduction of the use of trizolos that can be more harmful to the plant under stress. According to Marques (2017), the use of protective fungicide mancozeb has produced an effect on the physiological parameters of plants, and reduced the phytotoxicity caused by other fungicides such as in the mixture with trifloxystrobin + prothioconazole. Studies with estrubilurine pyraclostrobin showed an increase in the photosynthetic rate of soybeans; this increase in the liquid photosynthesis is linked to changes in the CO$_2$ compensation point, which favors its absorption, as opposed to its release by respiration. In addition, strobilurins act as moderators of biotic and abiotic stresses by participating in the metabolism of nitric acid and stress hormones such as the abscisic acid, decreasing the energy loss caused by the respiratory process, especially in atypical conditions, maximizing productivity. The fungicides based on strobilurine have the characteristic of promoting the increase in the chlorophyll content due to the increase in nitrogen assimilation and reduction of ethylene production, resulting in the “green effect”, contributing to less stress on plants, generating greater productivity (Mochko, 2017).

**CONCLUSION**

Unfavorable climate conditions contributed to the low rate of Asian rust infection in soybean. Fungicide management influenced soybean performance for the main agronomic variables studied, while no effect was observed in qualitative variables.

Grain yield was higher for the management with no fungicide application and provided a better cost-benefit ratio, and this result was influenced by variables plant height, height of the insertion of the first legume, number of branches and number of vegetables on the main stem.

Fungicide management must be carried out according to the climatic conditions favorable to the incidence of diseases, in order to obtain a greater cost benefit of soybean cultivation.

**Acknowledgments.** Does not apply.

**Funding.** The authors did not receive funding

**Conflict of interest.** The authors declare that they have no conflict of interest in carrying out the research work from which they derived the data used.

**Compliance with ethical standards.** Does not apply.

**Data availability.** Data are available from <Ivan Ricardo Carvalho, carvalho.irc@gmail.com> upon reasonable request with the corresponding author.

**Author contribution statement (CRediT)** - C. S. Zimmermann - Conceptualization, formal analysis and writing original draft., I. R. Carvalho - Conceptualization, supervision and formal analysis., M. V. Loro - Conceptualization, supervision and formal analysis., A. L. Schmidt - Data curation, writing-review & editing., H. C. F. Almeida - Conceptualization, formal analysis and writing original draft.

**REFERENCES**


Revista de Agricultura, 96(1), pp.294-313. https://doi.org/10.37856/bja.v96i1.4260


Souza, N.M. 2012. Teor de óleo na cultura da soja em função de fontes e doses de fósforo. 64 f. Dissertação (Mestrado em Agronomia) – Universidade Estadual de Ponta Grossa, Ponta Grossa.


Weber, P.S. 2017. Componentes de rendimento e grupos de maturidade relativa que influenciam o potencial de produtividade em soja. 2017. 34p. Trabalho de Conclusão de curso (Graduação em Agronomia). Universidade Federal de Santa Maria, Santa Maria, RS.