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### REFERÊNCIA

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# Impact of anthracnose on the yield of soybean subjected to chemical control in the north region of Brazil

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#### ABSTRACT

Dias, M.D.; Pinheiro, V.F.; Café-Filho, A.C. Impact of anthracnose on the yield of soybean subjected to chemical control in the north region of Brazil. *Summa Phytopathologica*, v.42, n.1, p.18-23, 2016.

Losses due to soybean anthracnose, caused by *Colletotrichum truncatum*, have not been systematically quantified in the field, and the efficacy of chemical control of this disease is not known. This study shows an estimate of losses associated with the disease in soybean crops in the north of the country. Two trials with cv. M9144 RR were carried out in commercial fields in Tocantins State in the 2010/2011 and 2011/2012 growing seasons, in randomized blocks, with four replicates. Foliar applications were performed on plants at R1/R2 and R5.2 stages, employing CO<sub>2</sub>-pressurized equipment and application volume of 200 L ha<sup>-1</sup>. Nine fungicides and one untreated control were compared, and the disease gradients in the two seasons were obtained. The percentage of infected pods was calculated at the R6 stage. Grain yield ranged from 3,288 to 3,708 kg/ha

in the untreated plots in 2010/2011 and 2011/2012, respectively, and from 3,282 to 4,110 kg/ha in the treated plots. In the 2010/2011 season, only azoxystrobin + cyproconazole significantly reduced the disease incidence, compared to untreated control plots, not differing from the remaining treatments. In the 2011/2012 season, there were no significant differences between treated and untreated plots. Highly significant correlations (p < 0.01) were found between yield and soybean anthracnose incidence on pods in both years (r = -0.85). For each 1% increment in the disease incidence, *c*. 90 kg/ha of soybean grain were lost. The current study determined that significant losses due to anthracnose occur in commercial crops in the north of the country and highlighted the limitation of chemical control as anthracnose management method.

Keywords: Colletotrichum truncatum; Glycine max, damage

#### RESUMO

Dias, M.D.; Pinheiro, V.F.; Café-Filho, A.C. Impacto da antracnose na produtividade de soja sob controle químico na região norte do Brasil. *Summa Phytopathologica*, v.42, n.1, p.18-23, 2016.

Danos devidos à antracnose da soja, causada por *Colletotrichum truncatum* não foram quantificados sistematicamente no campo e a eficiência do controle químico desta doença não é conhecida. Este trabalho apresenta uma estimativa dos danos associados à doença em plantios de soja no norte do país. Dois experimentos com a cv. M9144 RR foram conduzidos em campos comerciais no estado do Tocantins nas safras de 2010/2011 e 2011/2012 em blocos ao acaso com quatro repetições. Aplicações foliares foram feitas em plantas nos estádios R1/R2 e R5.2 utilizando um equipamento pressurizado com CO<sub>2</sub> em volume de calda de 200 L/ha. Nove fungicidas e um controle não pulverizado foram comparados, obtendo-se gradientes de doença nas duas safras. A percentagem de vagens infectadas foi estimada no estádio R6. A produção de grãos variou entre 3.288 e 3.708 kg/ha nas parcelas não pulverizadas, respectivamente em

Palavras-chave: Colletotrichum truncatum; Glycine max, dano

Soybean [*Glycine max* (L.) Merrill] is one of the most important crops for world food security, ranking first among field crops for protein production per hectare (10). Brazil is one of the main world growers, producing *c*. 87.0 million tons on 29.3 million hectares in 2013. Other leader world producers include the United States of America, Argentina, China, and India (http://faostat.fao.org). In some places, such as Australia, soybean has been promoted as a rotation crop for its contribution to soil N content, valuable cash return, and reduction in

2010/2011 e 2011/2012 e entre 3.282 e 4.110 kg/ha nas parcelas tratadas. Na safra de 2010/2011, apenas o tratamento com azoxystrobina + ciproconazol reduziu significativamente a intensidade da doença em relação às parcelas não tratadas, não diferindo dos demais tratamentos. Na safra 2011/2012 não se detectaram diferenças significativas entre as parcelas tratadas e não tratadas. Foram encontradas correlações altamente significativas (p < 0,01) entre a produtividade e a incidência da antracnose de soja nas vagens nos dois anos (r = -0.85). Para cada incremento de 1% na incidência da doença, *c*. 90 kg/ha de grãos foram perdidos. Este trabalho determinou que danos significativos devidos à antracnose ocorrem em plantios comerciais no Norte do país e indicaram a presente limitação do controle químico como método de manejo da antracnose.

lesion nematode populations (14, 15).

The large area under soybean cultivation in Brazil, including the incorporation of new growing areas in the northern and midwestern regions, mostly under monoculture and non-tillage system, affects the prevalence and the intensity of diseases. Among these, most prominent is anthracnose, whose causal agent in soybean is generally delimited by the taxon *Colletotrichum truncatum* (Schwein.) Andrus & W.D. Moore, although at least four other species with falcate conidia

have been associated to the disease (*C. coccodes, C. destructivum, C. gloeosporioides* and *C. graminicola* (9). In addition, Yang et al. (16, 17) have recently described two other species with falcate conidia causing soybean anthracnose in the USA, *C. chlorohyti* and *C. incanum*. Regardless of the complex nature of the causal agents, anthracnose leads to estimated yield losses of 16-26% in the United States, 30-50% in Thailand, and up to 100% in Brazil and in India (9). In Australia it was first reported mainly as a seedling disease and was considered a potentially serious pathogen (12), while Chen and others (3) considered it the most destructive disease of soybean in Taiwan. In Brazil the disease was first reported in 1961 in the state of Rio Grande do Sul. Since the 1980s it has been reported as prevalent in the Savannah-like Brazilian "Cerrado" region, causing severe damage to the soybean crop (2).

Anthracnose takes its main source of inoculum from infected seeds and crop residue, so it is one of the first diseases to establish in the crop, with lesions appearing on the cotyledons. Symptoms are later observed on stems and leaves. However, the most significant damage occurs during the reproductive phase, including twisted and aborted pods. They become twisted and fall off the plants, with direct impact on the yield (9, 13). Weather conditions favorable to epidemics include temperatures above 25 °C and leaf wetting that lasts longer than 24 h (1, 13). These conditions are frequent in the mid-western and northern regions of Brazil, which are occupied by the Cerrado biome, where the growing period coincides with the wet season.

Almost no information on the chemical control of anthracnose is available and whether or not there is a grain yield return for the investment on chemical control of the disease is not known. Nevertheless, frequent reports on the increase in the importance of anthracnose in the northern and mid-western regions indicate that the active ingredients that compose the chemical control program for fungal diseases in soybean are not effective against anthracnose (5). Specifically, estimates of damage to crop yield in the state of Tocantins, located in the northern region, have not yet been undertaken, although a high prevalence of anthracnose has been reported (5, 6).

Field studies, conducted in commercial areas in Distrito Federal (mid-western region), have recorded a mean incidence of 24% of typical anthracnose lesions in cotyledons (6). Among the measures recommended for reducing the incidence of anthracnose are crop rotation, wide-row spacing, appropriate plant density, seed treatment and suitable soil fertilization, especially regarding potassium (1). Chemical control was noted with some benzimidazoles alone or in a mixture with triazoles (1). However, unpublished accounts have pointed to a significant loss in yield due to anthracnose, especially in the northern and mid-western regions of Brazil. Furthermore, frequent reports from growers suggest the limited effectiveness of chemicals in controlling this disease.

Given the scarcity of quantitative information on damage associated with anthracnose, this study aimed to estimate the magnitude of yield losses under commercial crop conditions. To achieve this goal, the effectiveness of the main fungicides that are currently used in managing the soybean disease complex was estimated for the control of anthracnose.

#### MATERIALS AND METHODS

Two trials were carried out in commercial soybean fields in Alvorada County, state of Tocantins, in the periods from November to March in two consecutive growing seasons, 2010/2011 and 2011/2012.

Sowing took place in the second half of November, using cv. M9144RR, following the technical recommendations for the crop. Spacing was 0.5 m between rows, with 13 plants per linear meter. Fungicide applications were carried out at phenological stage R1 of Fehr and Caviness (8) and at stage R5 (20 days after the first application), using a CO<sub>2</sub> pressurizer, with a double-fan tip and an application volume of 200 L ha-1. Experimental design was in randomized blocks with four replicates, and experimental units consisted of areas of 18 m<sup>2</sup> (six rows of six meters long). Nine fungicides, selected from among those most used by soybean farmers for rust (Phakopsora pachyrhizi) control, were evaluated for their efficiency in anthracnose control. Some formulations were applied with the addition of adjuvants in the form of vegetable oil (Aureo<sup>®</sup>, Bayer CropScience) or mineral oil (Nimbus<sup>®</sup>, Syngenta e Assist<sup>®</sup>, Basf S.A.). The treatments, the respective levels and adjuvants (when used) were: trifloxystrobin + tebuconazole (Nativo<sup>®</sup> + Aureo<sup>®</sup>) 0.5 L/ha; trifloxystrobin + tebuconazole + carbendazim (Nativo<sup>®</sup> +  $Derosal^{\otimes} + Aureo^{\otimes}$ ) 0.5 + 1.0 L/ha; azoxystrobin + cyproconazole (PrioriXtra<sup>®</sup> + Nimbus<sup>®</sup>) 0.3 L/ha; picoxystrobin + cyproconazole (Aproach Prima<sup>®</sup> + Aureo<sup>®</sup>) 0.3 L/ ha; pyraclostrobin + epoxiconazole (Opera<sup>®</sup> + Assist<sup>®</sup>) 0.5 L/ha; trifloxystrobin + cyproconazole (Sphere Max<sup>®</sup> + Aureo<sup>®</sup>) 0.3 L/ha; chlorothalonil (Daconil<sup>®</sup>) 1.5 kg/ha; trifloxystrobin + prothioconazole (Fox<sup>®</sup> + Aureo<sup>®</sup>) 0.3 L/ha; picoxystrobin + tebuconazole + carbendazim (Horos® + Bendazol®) 0.5 + 0.8 L/ha. The treatments picoxystrobin + cyproconazole (Aproach Prima<sup>®</sup>) and picoxystrobin + tebuconazole + carbendazim (Horos<sup>®</sup> + Bendazol®) were evaluated in the second assay only (2011/2012 season).

The incidence of infected pods at phenological stage R6 was estimated based on the percentage of the number of infected pods over the total number of pods per plant, in 10 plants randomly distributed in the experimental unit. The yield was determined in 5 m<sup>2</sup>, equivalent to the two central rows of each plot, eliminating 0.5 m of the border. Correlation analysis, statistical analysis and separation of means (Tukey, at 5% probability) were carried out with the program SISVAR (7).

#### **RESULTS AND DISCUSSION**

Incidence of the disease and damage to yield, growing season 2010/2011: Significantly different incidences of soybean anthracnose developed following applications of fungicides in the 2010/2011 growing season; in the untreated plots, 16.3% of the pods presented anthracnose symptoms. Among the plots treated with fungicides, the percentage of symptomatic pods varied from 9.5% to 15.5 % (Fig. 1A). Only the azoxystrobin + cyproconazole treatment, with 9.5% incidence, differed significantly from the control (p < 0.05) but was not distinguished from the other treatments. Yield varied between 3,282 kg/ ha and 3,966 kg/ha. The highest yield was recorded for the azoxystrobin + cyproconazole treatment, which differed significantly (p < 0.05) from the untreated control (Fig. 2A). Even with 42% of reduction in anthracnose incidence in the best treatment, compared to non-sprayed controls, a 9.5% disease incidence is considered excessively high.

A strong negative correlation was detected between yield and disease incidence, with r = -0.8485 (P < 0.01, 6 d.f., Fig. 3A). The unsprayed control presented a mean reduction in yield of 678 kg/ha when compared with the azoxystrobin + cyproconazole treatment. No significant incidence of other diseases was noted, including in the unsprayed treatment.

*Incidence of the disease and damage to yield, growing season* 2011/2012: In the 2011/2012 season the percentage of symptomatic pods in the untreated plots (15.6%) was similar to that in the previous



**Figure 1.** Incidence of soybean pod anthracnose symptoms in field plots treated with synthetic fungicides in Northern Brazil. (A): 2010/2011 harvest; (B): 2011/2012 harvest. Columns with the same letters are not significantly different (Tukey, 0.05). Incidence was not significantly different in the 2011/2012 season.

growing season. However, significant differences were not detected in the disease incidence between treatments (10.7% to 13.3%) and the untreated control (Fig. 1B). Yields varied between 3,708 kg/ha(control) and 4,110 kg/ha, but the treatments did not differ statistically (Fig. 2B). The incidence of anthracnose was high once again, even in the treatment with the lowest incidence of diseased pods (10.7%), which did not differ from the non-sprayed control treatment.

An important negative correlation between the incidence of anthracnose in the pods and the yield was observed once more (r =

-0.8488, P < 0.01, 8 d.f., Fig. 3B). There were no noteworthy incidences of other diseases during the second assay.

The results from the two years of assays conducted under commercial field conditions show that anthracnose can be an important limiting factor for soybean production in the state of Tocantins and probably in areas that have similar weather conditions to those prevalent in the northern and mid-western regions of Brazil, where reports of significant yield reductions are frequent. The absence of significant incidence of other diseases provides a strong indication



**Figure 2.** Soybean grain yield in field plots treated with synthetic fungicides in Northern Brazil. (A): 2010/2011 harvest; (B): 2011/2012 harvest. Columns with the same letters are not significantly different (Tukey, 0.05). Grain yield was not significantly different in the 2011/2012 season (P > 0.05)

that the observed losses are attributable to anthracnose and reinforces the importance of this disease in the Cerrado biome of the northmidwestern region of the country. The negative correlation between yield and incidence of anthracnose in the pods showed that for every 1% increment in the disease incidence in pods, in the range of 9 to 17 % incidence, about 90-91 kg of soybean will be lost per hectare (Fig. 3A,B). Indeed, given the high disease incidence in the treatments with the lowest disease levels in both trials, yield loss due to anthracnose may have been underestimated, since there was no treatment with total control. Therefore, the estimates of negative impact on grain yield reported here may be regarded as conservative.

The results show that chemical control of anthracnose under



Figure 3. Correlation between incidence of soybean pod anthracnose symptoms and grain yield in field plots treated with fungicides in Northern Brazil. (A): 2010/2011 crop season; (B): 2011/2012 crop season

the evaluated conditions are not satisfactory, reaching maximum efficiencies of only 41.7% in the 2010/2011 season and of 31.4% in the 2011/2012 season. This is not unusual in the chemical control of soybean diseases. Recent studies showed that applications of triazoles did not reduce soybean brown spot (Septoria glycines) or favorably impact yield in Ohio. Other fungicides (pyraclostrobin and azoxystrobin) significantly reduced the final levels of brown spot, but yield increase due to chemical control was detected in only half of six assays (4). Nevertheless, the range of fungicides studied here provided the establishment of gradients for the disease and for the yield, which were adequate to estimate damage to grain production. Even at relatively modest (first year), or non-significant (second year), disease control results, the use of fungicides were correlated to higher yields in relation to the unsprayed control. The data presented in this study were obtained under natural conditions and may be useful for soybean growers elsewhere, such as in other parts of the world with ecological environments similar to the one prevailing in the hot and humid Cerrado planting season.

The high natural incidence, remaining consistent for two consecutive seasons in commercial fields and the losses associated with the disease show the need for appropriate anthracnose management in the north-midwestern region of Brazil, where the planting season is hot and wet, ideal conditions for anthracnose epidemics to develop. Therefore, control should not be based on the application of fungicides but on the integration of other agricultural practices, such as crop rotation, balanced fertilization, appropriate plant population size and use of good-quality seeds, in order to obtain an added or synergetic effect in the management of anthracnose. Although anthracnose is presently very important in the regions investigated in the current study, other regions with similar microclimates may be at risk.

The limited efficiency of the tested products may be attributed to various factors. One of the motives may be related to the timing of fungicide application in relation to the plant's infection period. Another cause may be associated with the low sensitivity of the pathogen to the tested active ingredients, which is presently unknown. According to Klingelfuss and Yorinori (11), *Colletotrichum* is present in the

aerial organs of the plant well before the appearance of anthracnose symptoms. Therefore, when applications of fungicides are carried out after symptoms appear, there is no reducing effect on the disease. These authors did not observe any effect of the application of the fungicide difenoconazole on soybean anthracnose. A third reason may be related to the variability of the *Colletotrichum* populations associated with soybean anthracnose, which may involve more than one species. In fact, the identity of the *Colletotrichum* species occurring in soybean in Brazil has not been studied. A better knowledge of the *Colletotrichum* species associated with the soybean crop is an important piece of information not only for the chemical control, but also for the development of soybean resistance breeding programs. Soybean anthracnose is a complex system, with many unresolved questions about the identity of the causal agent, the fungicide management and the need to develop a specific integrated control program.

This study presents a first estimate of the quantification of soybean yield losses due to soybean anthracnose, in the Cerrado of the northmidwestern region of Brazil, where the area sown with the crop is spreading fast. In two consecutive seasons, the fungicides applied and evaluated at R1 growth stage and again at 20 days after the first application were insufficient for a complete control of anthracnose. Significant negative correlations were found between yield and anthracnose incidence in both years (r = -0.85), and for each 1% increment in diseased pods, *c*. 90 kg/ha (3.32 bushels/ha) of soybean grain was lost. Our results suggest that anthracnose may also evolve into a worldwide threat to the soybean industry, especially in areas with a weather type similar to the one prevailing in the Cerrado biome, where the growing period coincides with the wet season.

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