

## RESEARCH NOTE

# First record of the soybean stem fly *Melanagromyza sojae* (Diptera: Agromyzidae) in the Brazilian Savannah<sup>1</sup>

Cecília Czapak<sup>2</sup>, Matheus Le Senechal Nunes<sup>2</sup>, Tiago Carvalhais<sup>2</sup>, Marcos Vinicius dos Anjos<sup>2</sup>, Rafael Ferreira Silverio<sup>2</sup>, Paula Barcelos Simões de Oliveira Lima<sup>2</sup>, Karina Cordeiro Albernaz Godinho<sup>2</sup>, Abmael Monteiro de Lima Junior<sup>3</sup>, Regina Melo Sartori Coelho<sup>3</sup>, Pablo da Costa Gontijo<sup>2</sup>

## ABSTRACT

The soybean stem fly [*Melanagromyza sojae* (Diptera: Agromyzidae)] is a pest widely distributed in the Asian continent. In Brazil, its occurrence has been reported since the 1980s, but it had not been reported in Brazilian Savannah areas yet. This is the first record of *M. sojae* in this region of great importance for the soybean world production. Specimens of the soybean stem fly were collected in a volunteer soybean area, in April 2018, in the Goiás state, Brazil, and sent for molecular identification. The similarity index of the collected specimens with *M. sojae* was higher than 99 %, thus confirming the identification of the species. The infestation level in the collection area was approximately 100 % of plants with damage caused by this insect. In Brazil, the occurrence of *M. sojae*, up to the present time, had been described as restricted to the southern states. This record raises the hypothesis that *M. sojae* is widespread in other agricultural areas of the Brazilian Savannah, and that its dissemination is favored by the maintenance of volunteer soybean.

KEYWORDS: *Glycine max*; soybean pests; integrated pest management; volunteer soybean.

The soybean stem fly [*Melanagromyza sojae* (Diptera: Agromyzidae)] is widely distributed in Asia (Wang & Gai 2001, Tapha 2012), as well as in parts of Russia (Strakhova et al. 2013), Australia (Shepard et al. 1983, Brier & Charleston 2013) and Spain (Gil-Ortiz et al. 2010), causing damage to soybean crops (Talekar 1989, Wang & Gai 2001). Its presence was also recently confirmed in Paraguay (Guedes et al. 2017). In Brazil, its occurrence has been reported since the 1980s, in the southern states of Rio Grande do Sul, Santa Catarina and Paraná (Gassen & Schneider 1985, Link et al. 2009), and

## RESUMO

Primeiro registro da mosca-da-haste da soja *Melanagromyza sojae* (Diptera: Agromyzidae) no Cerrado brasileiro

A mosca-da-haste da soja [*Melanagromyza sojae* (Diptera: Agromyzidae)] é uma praga com ampla distribuição no continente asiático. No Brasil, sua ocorrência tem sido relatada desde a década de 1980; porém, ainda não havia sido encontrada em áreas do Cerrado. Este é o primeiro relato de ocorrência de *M. sojae* nesta região de grande importância para a produção mundial de soja. Espécimes da mosca-da-haste foram coletados em área de soja voluntária, em abril de 2018, no estado de Goiás, e encaminhados para identificação molecular. O índice de similaridade dos espécimes coletados com *M. sojae* foi superior a 99 %, confirmando a identificação da espécie. O nível de infestação na área de coleta foi de aproximadamente 100 % de plantas com danos causados pelo inseto. No Brasil, a ocorrência de *M. sojae*, até o presente momento, era descrita como restrita aos estados do sul. Este registro suscita a hipótese de que *M. sojae* esteja disseminada em outras áreas agrícolas do Cerrado brasileiro, e que sua disseminação seja favorecida pela manutenção de plantas de soja voluntária.

PALAVRAS-CHAVE: *Glycine max*; pragas da soja; manejo integrado de pragas; soja voluntária.

it is considered a biosafety hazard for the country (Hirose & Moscardi 2012). The presence of *M. sojae* in Brazil was reported again in second-crop soybean, in the states of Santa Catarina and Rio Grande do Sul (Guedes et al. 2006, Arnemann et al. 2016), in 2015. However, its occurrence in the Brazilian Savannah had not been reported yet.

In this study, the first occurrence of *M. sojae* in the Brazilian Savannah was verified by the Laboratory of Integrated Pest Management team of the Escola de Agronomia, Universidade Federal de Goiás (MIP-UFG), at the end of April 2018, in Silvânia

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2. Universidade Federal de Goiás, Escola de Agronomia, Goiânia, GO, Brasil. E-mails: [ceciczepak@yahoo.com.br](mailto:ceciczepak@yahoo.com.br), [matheuslsn@hotmail.com](mailto:matheuslsn@hotmail.com), [carvalhais.tiago@gmail.com](mailto:carvalhais.tiago@gmail.com), [viniciusdosanjos2009@hotmail.com](mailto:viniciusdosanjos2009@hotmail.com), [rafaelsilverio18@hotmail.com](mailto:rafaelsilverio18@hotmail.com), [paulabsol.agro@gmail.com](mailto:paulabsol.agro@gmail.com), [kcalbernaz@gmail.com](mailto:kcalbernaz@gmail.com), [pablocgontijo@gmail.com](mailto:pablocgontijo@gmail.com).

3. Ministério da Agricultura, Pecuária e Abastecimento, Goiânia, GO, Brasil. E-mails: [abmael.lima@agricultura.gov.br](mailto:abmael.lima@agricultura.gov.br), [regina.sartori@agricultura.gov.br](mailto:regina.sartori@agricultura.gov.br).

(16°26'6,33"S; 48°50'4,22"W), Goiás state, Brazil, in an area of 223 ha with volunteer soybean (*tiguera*, in Portuguese), in the reproductive phase. To estimate the infestation level in the area, ten samples, from ten plants each, were randomly collected. Approximately 100 % of the plants in the area showed damage from *M. sojae* (Figure 1). Longitudinal cuts of the infested plants allowed to verify the presence of larvae and pupae. The rotting tissues of the soybean stem had a soft consistency and reddish-brown color.

In May 2018, volunteer soybean plants infested by *M. sojae* were collected in the reported area, in the

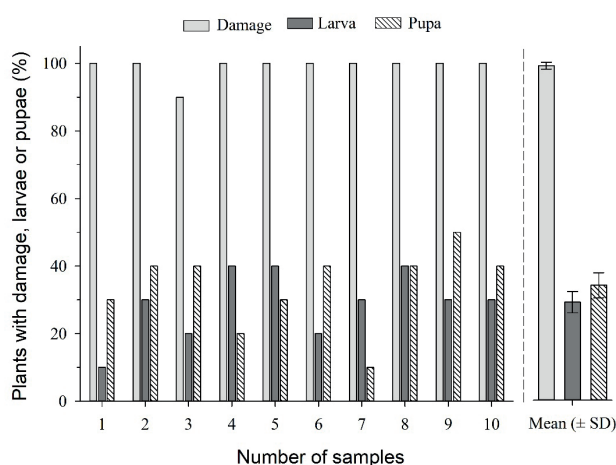


Figure 1. Infestation of the soybean stem fly *Melanagromyza sojae*, in area of volunteer soybean, in Silvânia, Goiás state, Brazil (April 2018).

presence of technicians from the Agência Goiana de Agrodefesa (Agrodefesa) (Collection term n° 22323, series A). Samples collected during the inspections were sent to the laboratory (MIP-UFG), in Goiânia, where the material was screened and *M. sojae* larvae were transferred to eppendorfs (1.0 mL) containing alcohol (99 %). Part of the collected material (six larvae and pupae) was identified and deposited in a collection maintained by the same laboratory, and another part was kept in a freezer (-18 °C). Later, the material was sent to the Laboratório Nacional Agropecuário de Goiás (Lanagro-GO/MAPA, Goiânia, Goiás), where the molecular characterization of the *M. sojae* larvae samples was carried out by amplifying the sequence of the cytochrome C oxidase gene. The DNA extraction from the larvae (three samples) was performed with the DNeasy Blood and Tissue Kit (Qiagen) and amplifications were performed with primer pairs: LCO 1490 (5'-GGT-CAACAAATCATAAAGATATTGG-3') and HCO 2198 (5'-TAAACTTCAGGGTGACCAAAAAA-TCA-3'), at 0.2 uM (Folmer et al. 1994). The PCR for the primer pairs was performed on the Veriti 96-well thermal cycler (Life Technologies of Brazil), programmed for 1 cycle of 95 °C for 15 min; 35 cycles of 94 °C for 30 seconds; 51 °C for 1 min; and 72 °C for 1 min, with a final extension of 72 °C for 10 min. The PCR product was purified by enzymatic reaction with the ExoSAP kit (USB Corporation). Sequencing was performed with the ABI Prism 3500

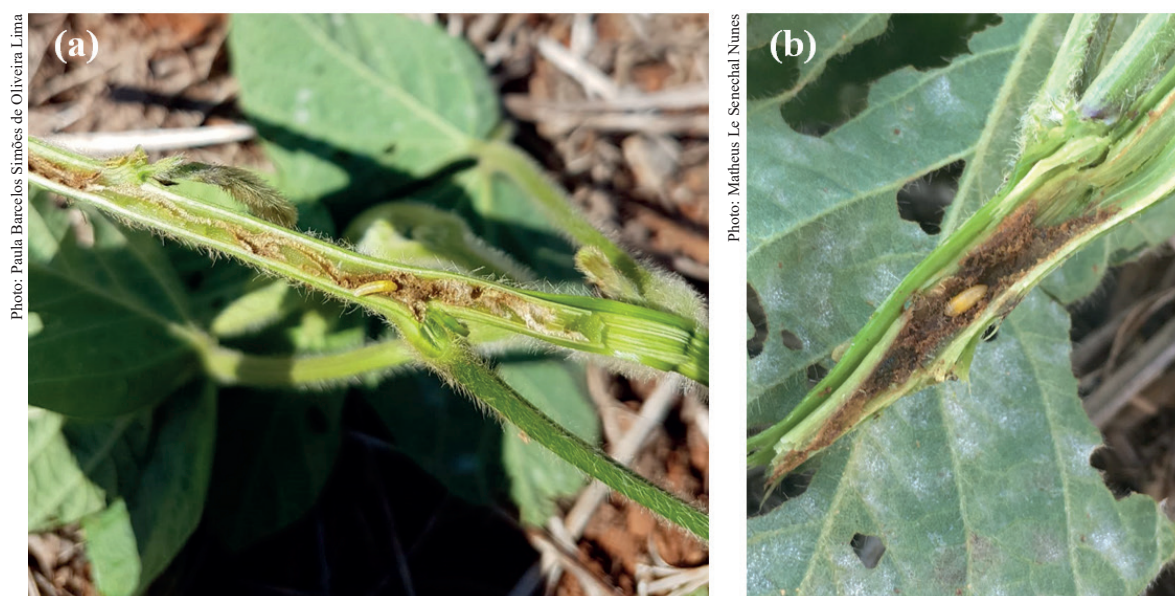


Figure 2. Larva (a) and pupa (b) of the *Melanagromyza sojae* stem fly in volunteer soybean, in area of the Brazilian Savannah (April 2018).

DNA Analyzer (Life Technologies of Brazil). Each sequence generated in the ABI file by the genetic analyzer was saved in a useful comparison unit (FASTA) to search in an authenticated standard database. The acceptance criterion adopted was to obtain a bidirectional sequence of the expected length (at least 500 bp in length), with less than 2 % ambiguous bases in contig sequence, or a single reading with at least 98 % of high quality bases. Post-sequencing analyzes were performed using the Sequencing Analysis Software 6 (Applied Biosystems). The treated sequences were compared with the validated sequences deposited in Bold Systems (Ratnasingham & Hebert 2007). The similarity index found with the *M. sojae* species was higher than 99 %, what confirms the identification of the species.

The soybean stem fly (*M. sojae*) prefers to infest plants from the Fabaceae family, such as soybean (Hirose & Moscardi 2012). The small black adult flies (2-3 mm) are rarely observed in the field, but, in large infestations, they may be seen in inflorescences and foliage. Their endophytic oviposition is performed on the abaxial face of the new leaves, with the eggs inserted into the plant tissues (Brier & Charleston 2013). After 2 to 3 days, the larvae that initiate the infestation begin to hatch, burrowing the nearest vein and then advancing through the petiole to the stem, forming galleries in parts or their entire extension. The larval period is 7-11 days. After this period, the pupae are formed within the galleries in the plant stems, lasting about 10 days. The complete life cycle varies from 16 to 26 days (Hirose & Moscardi 2012).

Stem fly infestation can reduce the stem diameter, plant height and distance from internodes, and a thickening at the base of the stem may be observed, which results in dwarfism or nutrient deficiency. In the reproductive phase, the number of pods is reduced. In addition, later planted soybeans infested by *M. sojae* may reach up to 21 % reduced grain yield (Gassen & Schneider 1985). In India, the high-intensity infestation of *M. sojae* has caused more than 80 % of losses in the soybean production (Plantwise 2015). To control the soybean stem fly infestation, crop rotation, adequate soil preparation and avoidance of late planting are the most effective control practices (Hirose & Moscardi 2012). Research on the management of this insect with insecticides is also under development (Curioletti et al. 2018).

After the identification of *M. sojae* was confirmed and its occurrence reported in the Brazilian Savannah to the proper inspecting agencies at Agrodefesa, they intensified the inspection in voluntary soybean areas and verified other plants infested by *M. sojae* within 20 km from the original site. *M. sojae* may be widespread in other agricultural areas of the Brazilian Savannah, and voluntary soybean plants originating from the remaining grains after harvest have facilitate the maintenance and dispersion of this pest. Inspection in areas with voluntary soybean is necessary, because the destruction of these plants may reduce the incidence of *M. sojae*, or even prevent the pest from spreading to other regions of the state or country. These measures are necessary to avoid *M. sojae* becoming established in high population levels and causing greater damages to the next soybean crops.

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