

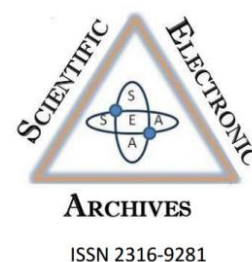
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First records of millipedes (Myriapoda, Diplopoda) associated to cultivation of Canola *Brassica* spp. (Brassicaceae) in Brazil

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Abstract: Millipedes are known by their importance in ecological systems, although some species are regarded as agricultural pests in some poly- or monocultures. Canola (Brassicaceae: *Brassica* spp.) is an important feedstock for biodiesel production and a potential ingredient in the food industry, but with several arthropod pests causing damage to its cultivation. In this perspective, this study reports the still unknown relation of millipedes with cultivation of canola. Pitfall traps were plotted in cultivation of *Brassica napus* L. var. *oleifera* in Passo Fundo, Rio Grande do Sul state, Brazil, and checked weekly during the years of 2015 and 2016. A total of 66 individuals were captured in 2015 and 41 individuals in 2016, all belonging to the families Chelodesmidae and Paradoxosomatidae (Polydesmida). The most abundant species sampled in both years was *Brasilodesmus austrocrucis* Hoffman, 1981, with 100 specimens in total. *Catharosomatini* sp. was recorded with 4 specimens, *Brasilodesmus triseriatus* (Attems, 1931) with 2 specimens, and *Oncoleptodesmus uniconus* (Attems, 1931) with only one specimen. In the present study, the sampled species were not reported causing any damage to crops, suggesting that millipedes may not be considered pests in canola.

Keywords: Chelodesmidae; Paradoxosomatidae; Passo Fundo; Polydesmida; Rapeseed oil.

Introduction

The millipede class Diplopoda comprises approximately 12,000 nominal species (Hoffman, 1980; Sierwald & Bond, 2007), distributed on all continents except Antarctica (Golovatch & Kime, 2009). Members of the class play important ecological roles as detritivorous, acting directly in the decomposition of organic matter in forests, and in the soil aeration by their movements between the soil layers (Blower, 1985; Hopkin & Read, 1992; Golovatch & Kime, 2009). Millipedes species may be restricted to specific mountains, valleys, small patches of forests, or caves (Golovatch & Kime, 2009; Enghoff, 2015; Iniesta et al., 2021). Some species have been reported as widely introduced around the world by human activities, such as gardening, cultivation of plants, and soil transport

(Iniesta et al., 2021). Additionally, species have been regarded as agricultural pests in some poly- or monocultures, feeding on seedlings of fern, soybean, coffee, lettuce, tuber, and ornamental plants (Schubart, 1942; 1945; Lordello, 1954; Domiciano & Fontes, 2001; Link & Link, 2001; Ebregt et al., 2005; Salvadori et al., 2007; David, 2015).

Cultures of *Brassica* spp. (Brassicaceae), commercially known as canola, are focused on their oil-rich seed, with important high-protein by-products to human and animal food, and biodiesel production (Gonçalves et al., 2017). Canola is considered as one of the most abundant sources of edible oil in the world (Aider & Barbana, 2011), containing around 40% of oil and 38%–43% protein (Shahidi, 1990). The canola production is broadly supplied by two

species, *Brassica napus* L. and *Brassica rapa* L. (Raymer, 2002). In Brazil, the oil production is mostly focused on the food market (Bergmann et al., 2013), with the state of Rio Grande do Sul having the largest cultivated area and the largest production with approximately 35 thousand tons of grains.

Recent studies have reported arthropod species as pests in monocultures of canola (Gu et al., 2007). In Brazil, studies have been conducted to understand the possible negative effects of these arthropods in plantations (Bianchi et al., 2019; Lucini et al., 2020; Marsaro-Júnior et al., 2019; 2021). Nonetheless, no study has been conducted to date reporting the association of millipedes with canola, so that their impact on the agricultural management remains vastly unknown.

In this perspective, this study is one of our efforts to understand the dynamics of arthropod communities in cultures of canola in Brazil. Hereby, we report for the first time millipedes species associated to the culture of *Brassica napus* in the state of Rio Grande do Sul, and their possible effects with this cultivation.

Material and methods

The study was carried out in the experimental area at the Centro Nacional de Pesquisa de Trigo (Embrapa Trigo), (28° 14' 02.1" S, 52° 24' 14.4" W), in Passo Fundo, state of Rio Grande do Sul, Brazil (Fig. 1). The municipality is located at 638m a.s.l., with regional climate of classified as Cfa (in Köppen classification system), and average annual temperature ranges between 16 and 18°C (Alvares et al., 2013).

The sampling site was constituted by plots of canola (*Brassica napus* L. var. *oleifera*, cv. Hyola 433) (approximately 800m²) during the months of June to September of 2015 (14 samplings), and June to October of 2016 (12 samplings). The surrounded matrix of the sampling site was

composed of other plots of crops such as wheat [*Triticum aestivum* (L.) Thell], rye (*Secale cereale* L.), and oat (*Avena sativa* L.), with natural vegetation present in the area nearby the canola cultivation (Fig. 1). The millipedes were collected with 5 pitfall traps placed randomly, without any bait or attractive substance. The traps were composed of fixing solution containing water, formaldehyde (0.5%), and colorless detergent. The traps were checked weekly during the study. The species identifications were made at the Instituto Butantan (IBSP) based on the literature and direct comparison with other specimens deposited in the zoological collection of the IBSP. All the collected specimens were identified to the lowest taxonomic level possible. All specimens were examined in 70% ethanol under a stereomicroscope Leica MZ16A and microscope Leica DM4000B. Voucher specimens are deposited at the IBSP.

Results and discussion

A total of 107 individuals were captured, 66 in 2015 and 41 in 2016, all belonging to the two polydesmidan families Chelodesmidae and Paradoxosomatidae (Table 1). For Chelodesmidae three species were identified, *Brasilodesmus austrocrucis* Hoffman, 1981 (Figs 3A, 4); *Brasilodesmus triseriatus* (Attems, 1931); *Oncoleptodesmus uniconus* (Attems, 1931); and for Paradoxosomatidae only one morphospecies was identified, *Catharosomatini* sp. (Fig. 3B). The most abundant species found in both years was *B. austrocrucis*, with 44 males and 18 females in 2015, and 19 males and 19 females in 2016, while *O. uniconus* was found only in 2016 based on a single male. The most abundant period during 2015 was in July with 28 individuals (21 males and 7 females) of *B. austrocrucis* sampled, and for 2016 was in August with 20 individuals (12 males and 8 females) of *B. austrocrucis* and two males of *B. triseriatus* sampled (Table 2; Fig. 2).

Table 1. Abundance of millipedes associated to cultivation of canola (*Brassica napus* L. var. *oleifera*) in Passo Fundo-RS, Brazil, in 2015 and 2016. Abbreviations: M, number of males; F, number of females; J, number of juveniles; T, number total; RFt, relative abundance in percentage.

Taxa	2015					2016				
	M	F	J	T	RFt (%)	M	F	J	T	RFt (%)
Polydesmida										
Chelodesmidae										
<i>Brasilodesmus austrocrucis</i>	44	18	0	62	93.9	19	19	0	38	92.7
<i>Brasilodesmus triseriatus</i>	0	0	0	0	0	2	0	0	2	4.9
<i>Oncoleptodesmus uniconus</i>	0	0	0	0	0	1	0	0	1	2.4
Paradoxosomatidae										
<i>Catharosomatini</i> sp.	0	1	3	4	6.1	0	0	0	0	0
Total =	44	19	3	66	100	22	19	0	41	100

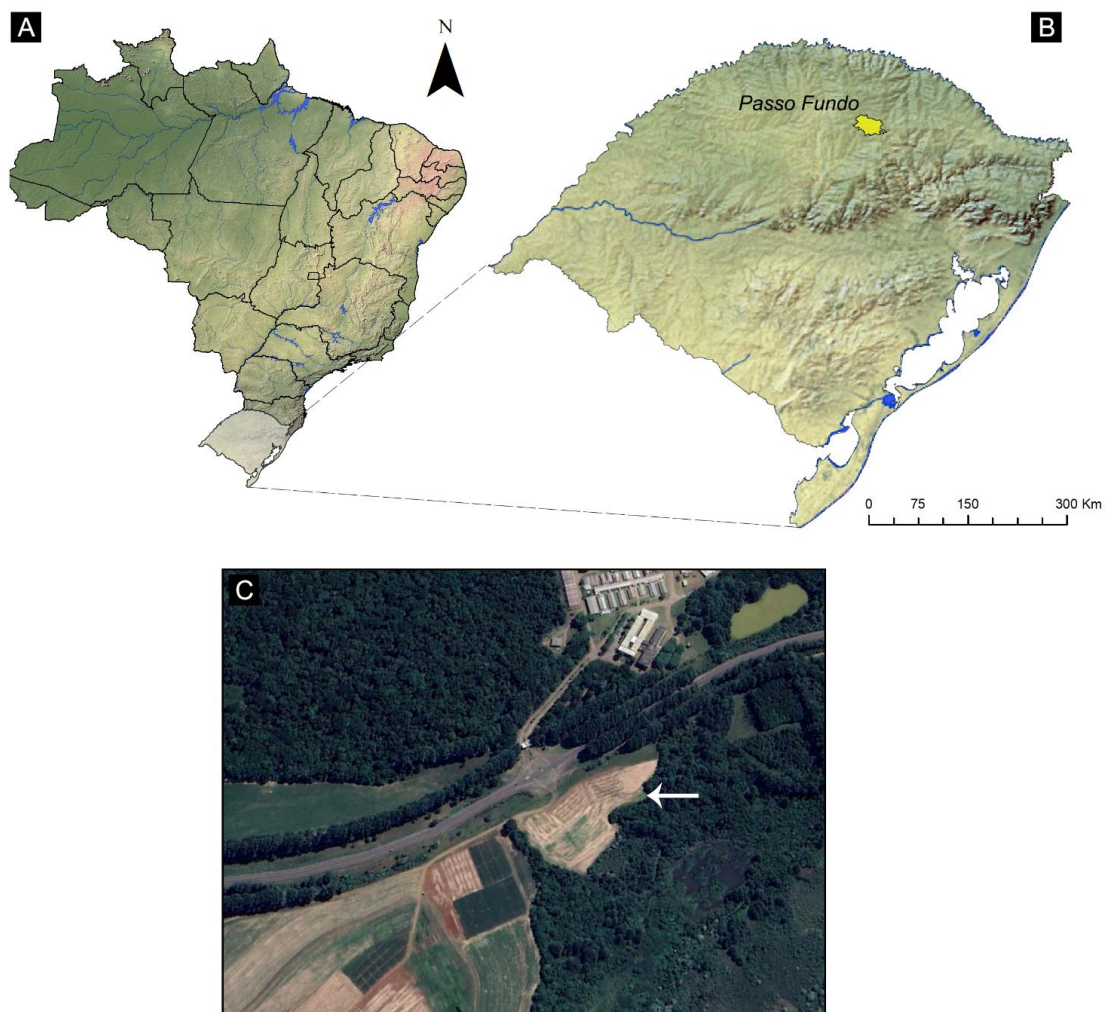


Figure 1. Map of the sampling site. A) Map of Brazil, with Rio Grande do Sul state highlighted; B) Rio Grande do Sul state, with the municipality of Passo Fundo in yellow; C) Experimental area at the Embrapa Trigo. The white arrow indicates the exact location of the cultivation of canola (*Brassica napus* L. var. *oleifera*).

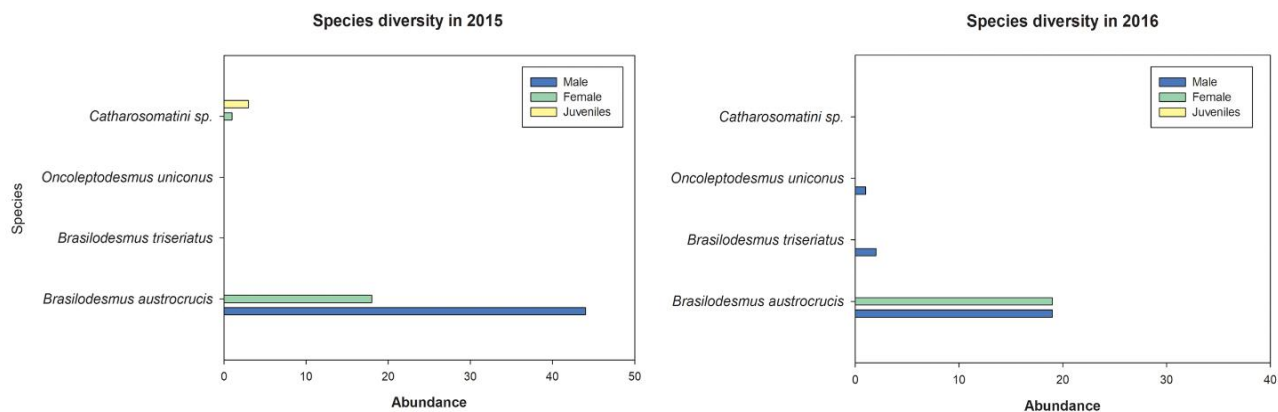


Figure 2. Abundance of millipedes associated to cultivation of canola (*Brassica napus* L. var. *oleifera*) in Passo Fundo-RS, Brazil, throughout the period of 2015 and 2016.

Table 2. Abundance of millipedes associated to cultivation of canola (*Brassica napus* L. var. *oleifera*) in Passo Fundo-RS, Brazil, in 2015 and 2016. Abbreviations: M, males; F, females; J, juveniles; T, total. The months are represented in Roman numerals.

	Data of sampling	Rate												
		<i>Brasilodesmus austrocrucis</i> *			<i>Brasilodesmus triseriatus</i> *			<i>Oncoleptodesmus uniconus</i>			Catharosomatini sp.			
		M	F	J	M	F	J	M	F	J	M	F	J	T =
2015	01.vi	3	0	0	0	0	0	0	0	0	0	0	1	4
	16.vi	4	2	0	0	0	0	0	0	0	0	0	0	6
	22.vi	0	1	0	0	0	0	0	0	0	0	0	0	1
	29.vi	5	3	0	0	0	0	0	0	0	0	0	0	8
	06.vii	13	4	0	0	0	0	0	0	0	0	0	0	17
	13.vii	6	2	0	0	0	0	0	0	0	0	0	1	9
	22.vii	2	1	0	0	0	0	0	0	0	0	0	0	3
	03.viii	3	2	0	0	0	0	0	0	0	0	0	0	5
	10.viii	2	0	0	0	0	0	0	0	0	0	0	0	2
	24.viii	2	1	0	0	0	0	0	0	0	0	1	0	4
	31.viii	2	0	0	0	0	0	0	0	0	0	0	1	3
	08.ix	0	1	0	0	0	0	0	0	0	0	0	0	1
	14.ix	1	1	0	0	0	0	0	0	0	0	0	0	2
	21.ix	1	0	0	0	0	0	0	0	0	0	0	0	1
	2016	27.vi	1	5	0	0	0	0	0	0	0	0	0	0
05.vii		0	1	0	0	0	0	0	0	0	0	0	0	1
11.vii		3	0	0	0	0	0	1	0	0	0	0	0	4
18.vii		3	2	0	0	0	0	0	0	0	0	0	0	5
08.viii		7	6	0	1	0	0	0	0	0	0	0	0	14
15.viii		1	1	0	0	0	0	0	0	0	0	0	0	2
22.viii		3	1	0	0	0	0	0	0	0	0	0	0	4
30.viii		1	0	0	0	0	0	0	0	0	0	0	0	1
19.ix		0	1	0	0	0	0	0	0	0	0	0	0	1
26.ix		0	1	0	0	0	0	0	0	0	0	0	0	1
03.x		0	0	0	1	0	0	0	0	0	0	0	0	1
10.x	0	1	0	0	0	0	0	0	0	0	0	0	1	
	T =	63	37	0	2	0	0	1	0	0	0	1	3	107

*First record in Rio Grande do Sul state.

The species *Brasilodesmus austrocrucis* and *B. triseriatus* are reported for the first time in the Brazilian state of Rio Grande do Sul, since the single known records are from their type localities in the state of Santa Catarina (Attems, 1931; Hoffman, 1981). The tribe Catharosomatini Brolemann, 1929 encompasses 12 genera and about 100 species, distributed in South and Central America, with several records from the state of Rio Grande do Sul. One of the most remarkable features of this group is the poorly developed to missing paraterga (Nguyen & Sierwald, 2013), in comparison to chelodesmid species. For the *O. uniconus*, the record in Passo Fundo extends its distribution in 163 km from the type locality in Santa Cruz do Sul, also in the state

of Rio Grande do Sul (Schubart, 1958). Overall, the record of these species is quite unexpected, since they have not been reported since their original descriptions. In fact, most records of millipedes species in Brazil are only from their descriptions, mainly from the beginning of the 20th century by European naturalists (Hoffman, 1980; Bouzan et al., 2018), which demonstrates our poor knowledge of the millipedes distribution in the country.

The association of these species with canola is the first record of millipedes in that culture in Brazil. To date, the association of millipedes has been reported only in cultures of fern, ornamental plants, and edible plants of families as Apiaceae, Asteraceae, Fabaceae, Rubiaceae, and

Solanaceae, for instance (Schubart, 1942; 1945; Lordello, 1954; Domiciano & Fontes, 2001; Link & Link, 2001; Salvadori et al., 2007). For most of these records, the millipedes species found are regarded as agricultural pests, including the introduced paradoxosomatid species *Oxidus gracilis* (C.L. Koch, 1847) and *Orthomorpha coarctata* (Saussure, 1860) (Iniesta et al., 2020; 2021). Both species were not reported in our study. To date, no one chelodesmid species has been considered as pest in mono- polycultures in Brazil, even if sporadically

found near tubercles or rootlets (see Schubart, 1942).

Although the sampled millipedes species in pitfall traps was found associated with canola, it is not suggestive that these species were causing any damage to crops. Additionally, the records obtained for *B. triseriatus*, *O. uniconus*, and Catharosomatini sp. can be regarded as occasionally in canola, considering their low abundance (less than 8% in both years) compared with the most abundant *B. austrocrucis*.

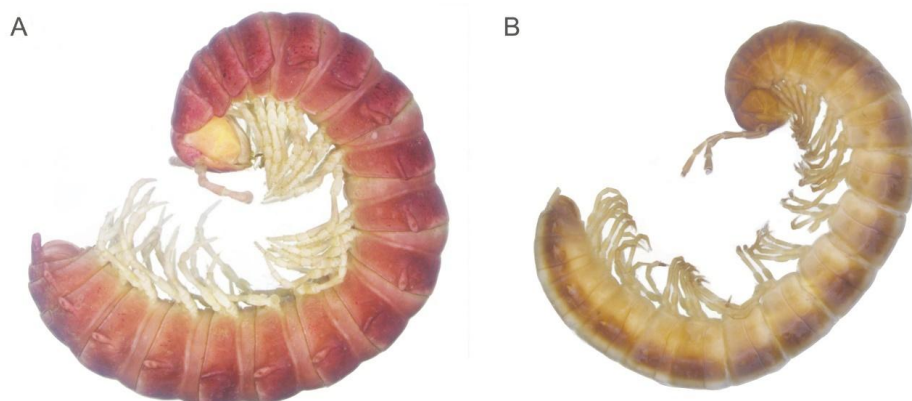


Figure 3. Millipedes associated to cultivation of canola (*Brassica napus* L. var. *oleifera*) in Passo Fundo-RS, Brazil, in lateral view. A) *Brasilodesmus austrocrucis*; B) Catharosomatini sp. Images not to scale. Photos: Luiz F.M. Iniesta.



Figure 4. Living specimen of *Brasilodesmus austrocrucis* associated to cultivation of canola (*Brassica napus* L. var. *oleifera*) in Passo Fundo-RS, Brazil. Photo: Alberto L. Marsaro Júnior.

Conclusion

Based on our results, the millipede species associated to the cultivation of canola were *Brasilodesmus austrocrucis*, *Brasilodesmus triseriatus*, *Oncoleptodesmus uniconus*, and Catharosomatini sp., with the former the most abundant species in 2015 and 2016. It was not observed any damage to the culture regarding the presence of millipedes, suggesting that the sampled species should not be regarded as pests on this culture. Nonetheless, further surveys are still needed to understand the relation of millipedes with

this culture, mainly the ecological factors that can be related to this agricultural system.

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