Scientific Note/Special Supplement: Pulses

Diversity and seasonal dynamics of Miridae (Hemiptera) associated with cowpea in the Piauí state, Brazil[1]

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ABSTRACT

Despite the wide range of insects associated with cowpea, the roles of certain groups, including Miridae, remain poorly understood. This study aimed to identify the diversity of Miridae species associated with cowpea and the seasonal dynamics of these species during two growing periods, in the Piauí state, Brazil. The insects were collected using sweep nets throughout the phenological cycle of the crop, covering the rainy and dry seasons. In total, 306 mirid samples, representing 13 species, were collected, being *Creontiades rubrinervis*, *Cyrtocapsus femoralis* and *Horciasinus signoreti* the most abundant ones, particularly during the reproductive stage of the crop. The highest diversity was observed during the rainy season, highlighting the effects of the season on Miridae populations in the cowpea agroecosystem.

KEYWORDS: *Vigna unguiculata* (L.) Walp., *Creontiades*, *Cyrtocapsus*, *Horciasinus*.

RESUMO

Diversidade e dinâmica sazonal de Miridae (Hemiptera) associada ao feijão-caupi no estado do Piauí, Brasil

Apesar da ampla gama de insetos associados ao feijão-caupi, os papéis de certos grupos, incluindo Miridae, ainda são pouco compreendidos. Objetivou-se identificar a diversidade de espécies de Miridae associadas ao feijão-caupi e a dinâmica sazonal dessas espécies durante dois períodos de cultivo, no estado do Piauí, Brasil. Os insetos foram coletados utilizando-se redes de varredura ao longo do ciclo fenológico da cultura, abrangendo as estações chuvosa e seca. No total, 306 amostras de mirídeos, representando 13 espécies, foram coletadas, sendo *Creontiades rubrinervis*, *Cyrtocapsus femoralis* e *Horciasinus signoreti* as mais abundantes, especialmente durante o estágio reprodutivo da cultura. A maior diversidade foi observada durante a estação chuvosa, destacando os efeitos da estação nas populações de Miridae no agroecossistema do feijão-caupi.

PALAVRAS-CHAVE: *Vigna unguiculata* (L.) Walp., *Creontiades*, *Cyrtocapsus*, *Horciasinus*.

Cowpea is commonly grown in the Northeast Region of Brazil because of numerous advantages, including nutritional benefits and physiological traits, which allow it to thrive, despite challenging environmental conditions such as high temperatures and water scarcity (Desravines et al. 2022, Raizada et al. 2023).

With the expansion of cowpea cultivation in Brazil, knowledge on previously overlooked insects, such as certain Miridae species, has grown substantially in cultivation fields.

Miridae, which is widely distributed worldwide, except in Antarctica, is the largest family within the Heteroptera suborder, with almost one-third of previously described heteropterans, including more than 1,383 genera (Wheeler 2001). In the Neotropical region, there are approximately 3,000 Miridae species (Henry & Wheeler 1988, Schuh 1995), 1,053 of which have been described in Brazil (Ferreira et al. 2006, Cassis & Schuh 2012).

Mirids range in size from 1 to 15 mm and can be morphologically identified by key characteristics, such as their elongated, oval body, with a triangular head. They present variable coloration with distinct hemeliums, with the chorus distally divided into a triangular cuneus and an apical membrane with one

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or two unequal cells divided by a short longitudinal vein, an apomorphic character for mirids (Carvalho 1952, Carvalho 1955, Schuh 1976, Wheeler 2001).

The broad distribution, polyphagia and large diversity of Miridae have prompted a search for more knowledge on this family, which was initially the focus of biological control studies in the 1920s and 1930s (Wheeler 2000a). Since then, research into predatory mirids has increased. Nevertheless, starting at the end of the 19th Century, the Miridae family has been considered an important group of phytophagous insects (Wheeler 2000a), with cotton, apple, cashew, guava, mango, sorghum and alfalfa as host crops (Wheeler 2000b). Mirids feed mainly on the cowpea meristematic tissues and flower buds, developing inside the seeds (Scott et al. 1966, Leigh 1976). Therefore, these insects can cause losses directly linked to pod production, consequently reducing seed yield (Khattat & Stewart 1975). However, studies on their diversity in cowpea are rare. Thus, this study aimed to determine the richness, abundance and diversity of mirids associated with cowpea in the Piauí state, Brazil, as well as to analyze how seasonal changes influence the species composition.

The study was conducted at the Embrapa's experimental field, in Teresina, Piauí state, Brazil (5º02'21.36"S; 42º47'22.44"W), during the year of 2015. The region has a tropical rainy climate (Aw'), with a predominance of summer-autumn rainfall annually ranging from 1,336.5 to 1,413.1 mm. The relative humidity ranges from 67.8 to 72.6 % and the average annual temperature is 28.2 to 28.4 ºC. Local vegetation includes sub-deciduous forests, Cerrado and Caatinga, forming diverse transitional ecosystems rich in plant species (Lima et al. 2015).

Cowpea (BRS Maratoã cultivar) was grown in a 500-m² area with 0.70-m row spacing. Planting occurred during the rainy season and later during the dry season, with irrigation. Biological control using *Bacillus thuringiensis* replaced chemical insecticides and manual weeding managed the weeds. Weekly climate data (rainfall, humidity and temperature) were collected from the Embrapa's weather station.

Mirids were collected weekly throughout the cowpea phenological cycle during the rainy (April-June 2015) and dry (September-November 2015) seasons, totaling 24 weeks of sampling. Using sweep nets, 10 sweeps were conducted at 36 points along the linear transects, with a 10-m distance between points, resulting in 864 samples. The insects were preserved in 70 % alcohol, labeled and stored in a freezer for subsequent analysis. Hemipteran specimens were screened and identified in the laboratory using a stereomicroscope.

The Miridae samples were separated using an identification key (Choate 2000). They were initially grouped into morphospecies, quantified and photographed. After the initial classification, species-level identification was performed by Paulo Sérgio Fiuza Ferreira, Miridae taxonomist from the Regional Museum of Entomology at the Universidade Federal de Viçosa, Minas Gerais state, Brazil.

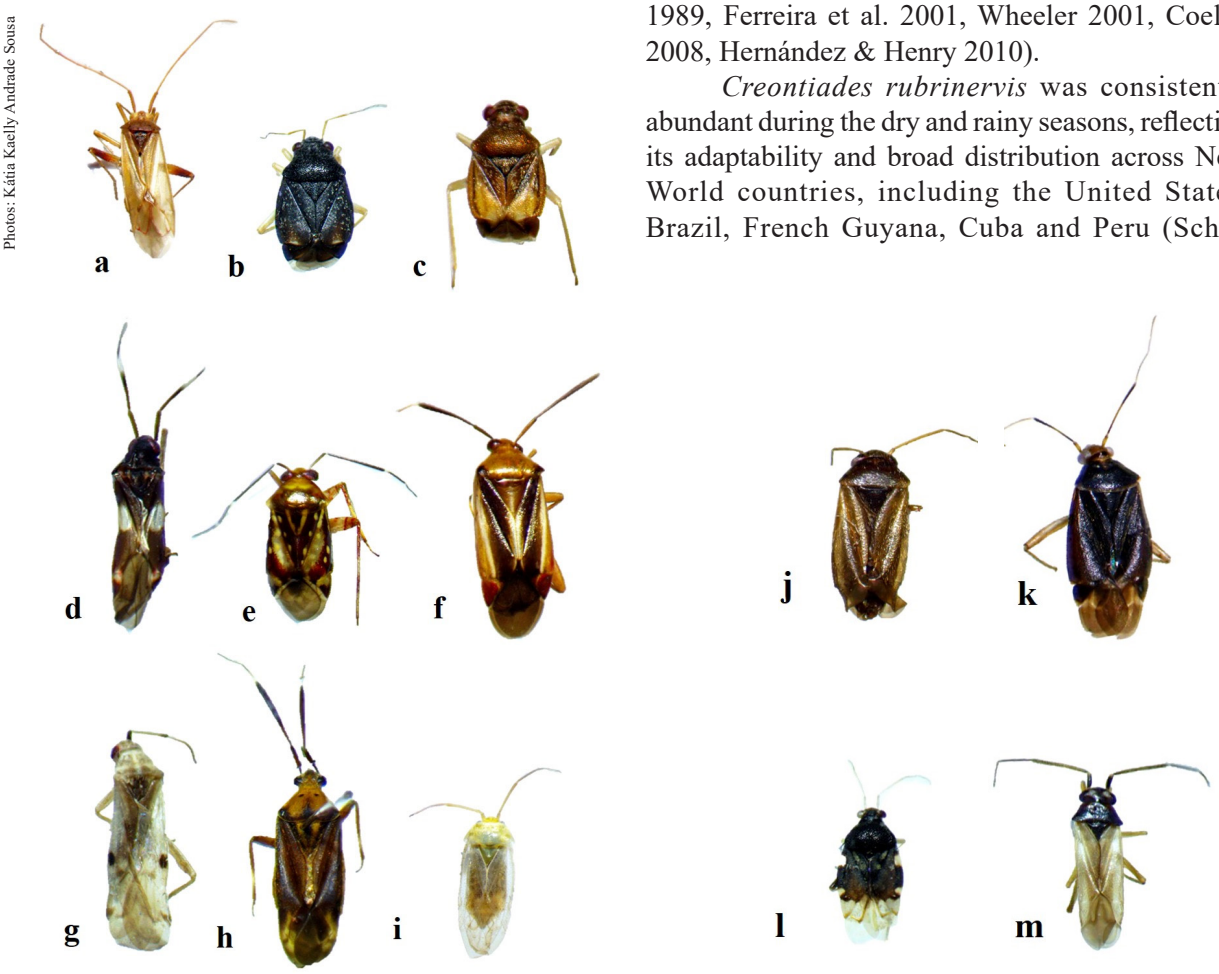
Using the obtained data, the Miridae species composition, relative frequency and seasonal occurrence were determined, as well as the population fluctuations for each season. Abundance, diversity, richness and equitability indices were determined using the Anafau software. The Pearson's correlation coefficient (r) was used to analyze the correlations between Miridae abundance and moisture, temperature and rainfall.

In total, 306 Miridae samples were collected during the entire phenological cycle of the cowpea (*Vigna ungiculata*) in both seasons (Table 1). The identified species were: *Creontiades rubrinervis* Stal, 1862; *Cyrtocapsus femoralis* Reuter, 1892; *Cyrtocapsus* sp.; *Fulvius* sp. Stal; *Horcias guapeanus* Carvalho, 1976; *Horciasinus signoreti* Stal, 1859; *Macrolophus basicornis* Stal, 1860; *Notholopus filicornis* Fabricius, 1803; *Orthotylus* sp. 1; *Orthotylus* sp. 2; *Polymerus testaceipes* Stal, 1860; *Pycnoderes quadrimaculatus* Guérin-Méneville, 1857; and *Tytthus* sp. Fieber (Figure 1). Notably, this study reports the first records of associations between cowpea and *Cyrtocapsus* sp., *Fulvius* sp., *N. filicornis*, *H. guapeanus*, *Orthotylus* sp. 1, *Orthotylus* sp. 2, *P. testaceipes* and *Tytthus* sp., expanding the knowledge on the distribution and ecological interactions of these insects in the region.

Some mirid species are recognized as pests or exhibit other types of interactions with their host plants. In the case of cowpea crops, certain species commonly act as pests, such as *C. rubrinervis*, *H. signorete* and *P. quadrimaculatus*, or are associated with this crop, such as *C. femoralis*, *H. guapeabus* and *M. basicornis* (Silva et al. 1968, Alayo 1974, Ferreira & Rossi 1979, Maes & Carvalho

Table 1. Species, abundance, absolute frequency (AF%) and relative frequency (RF%) of Miridae in cowpea in Teresina, Piauí state, Brazil, in 2015.

Species	Rainy season			Dry season		
	Abundance	AF%	RF%	Abundance	AF%	RF%
Creontiades rubrinervis	115	38.3	19.0	6	100	100
Horciasinus signoreti	87	29.0	15.5	0	0	0
Cyrtocapsus femoralis	34	11.3	13.8	0	0	0
Orthotylus sp. 1	18	6.0	13.8	0	0	0
Horcias guapeanus	16	5.3	13.8	0	0	0
Macrolophus basicornis	11	3.7	5.2	0	0	0
Notholopus filicornis	8	2.7	6.9	0	0	0
Pycnoderes quadrimaculatus	4	1.3	3.4	0	0	0
Polymerus testaceipes	3	1.0	1.7	0	0	0
Cyrtocapsus sp.	1	0.3	1.7	0	0	0
Fulvius sp.	1	0.3	1.7	0	0	0
Orthotylus sp. 2	1	0.3	1.7	0	0	0
Tytthus sp.	1	0.3	1.7	0	0	0
Total	300	100	100	6	100	100



1989, Ferreira et al. 2001, Wheeler 2001, Coelho 2008, Hernández & Henry 2010).

Creontiades rubrinervis was consistently abundant during the dry and rainy seasons, reflecting its adaptability and broad distribution across New World countries, including the United States, Brazil, French Guyana, Cuba and Peru (Schuh

Figure 1. Miridae species collected from cowpea in Teresina, Piauí state, Brazil, in 2015. a) *Creontiades rubrinervis*; b) *Cyrtocapsus femoralis*; c) *Cyrtocapsus* sp.; d) *Fulvius* sp.; e) *Horcias guapeanus*; f) *Horciasinus signoreti*; g) *Macrolophus basicornis*; h) *Notholopus filicornis*; i) *Orthotylus* sp. 1; j) *Orthotylus* sp. 2; k) *Polymerus testaceipes*; l) *Pycnoderes quadrimaculatus*; m) *Tytthus* sp.

1995, Hernández & Henry 2010). In cotton crops, particularly in Arizona and northern Mexico, mirids are considered the most important pests, because of their capacity to persist and reproduce throughout the season, leading to substantial economic losses and control costs (Wheeler 2001). In addition to cotton, *C. rubrinervis* is found in crops such as watermelon, cucumber, chickpea, soy, green bean, common bean, cowpea, sesame and sorghum (Silva et al. 1968, Alayo 1974, Ferreira & Rossi 1979, Maes & Carvalho 1989, Ferreira et al. 2001, Wheeler 2001, Hernández & Henry 2010). Although this species is abundant in cowpea fields, its role as a pest of this crop is not well-understood. This gap likely stems from the challenges in distinguishing its damage from that caused by other well-known phytophagous insects, leading to an underestimation of its impact on cowpea.

Horciasinus signoreti was the second most abundant and frequent species, representing the first recorded occurrence of this species in the Piauí state. In cowpea, *H. signoreti* can attack plants during the seed development and maturation phases, peaking during the rainy season (Silva & Magalhães 1980). This finding validates the results of the present study, as it was observed that the population peak occurred during the developmental phase of the seeds. This species is widely distributed in South America, including Colombia, Brazil, Peru, Paraguay, Bolivia, Venezuela and Argentina (Carvalho et al. 2000). In Brazil, this species has been found in 17 states: Minas Gerais, Rio de Janeiro, Santa Catarina, Mato Grosso, Goiás, Paraná, Pernambuco, Pará, Amazonas, Amapá, Bahia, São Paulo, Ceará, Rondônia, Paraíba, Espírito Santo and Rio Grande do Sul (Coelho 2008). In addition to cowpea, *H. signoreti* is associated with common bean (*Phaseolus vulgaris*), beetroot (*Beta vulgaris* L.), soy [*Glycine hispida* (Moench) Maxim.], Mexican cotton (*Gossypium hirsutum* L.) and Congo grass (*Brachiaria ruziziensis* Germ. & Evrard) (Ferreira et al. 2001, Coelho 2008).

The third most abundant mirid was *C. femoralis*, which was present primarily during the pod-filling stage. Currently, little is known about *C. femoralis*. There are only a few records of this species in Brazil, French Guyana and Venezuela (Carvalho 1957, Schuh 1995). In Brazil, *C. femoralis* has been found only in Minas Gerais, Piauí, Espírito Santo and Rio Grande do Sul (Coelho 2008). In addition to cowpea, *C. femoralis* is associated with *Ipomoea batatas* (L.) Lam., *Brachiaria brizantha* (A. Rich.) Stapf. and *Panicum maximum* cv. Mombaça Jacq (Nogueira et al. 2019).

Two species from the *Orthotylus* genus, which is the most diverse genus in the Miridae family (Pagola-Carte & Ribes 2007), were also identified, namely *Orthotylus* sp.1 and *Orthotylus* sp.2. In Brazil, thirteen species of *Orthotylus* have already been registered in the states of Minas Gerais, Bahia, Santa Catarina (Ferreira & Rossi 1979, Ferreira et al. 2001, Coelho 2008), and in Piauí for the first time in the present study. The *Orthotylus* genus includes species from the Orthotylini tribe, which presents variable dietary habits, both phytophagous and predatory. Plant-sucking species generally cause foliar symptoms, such as chlorosis, lesions and necrosis, and are capable of feeding on inflorescences (Wheeler 2001).

Very little is known about the *H. guapeanus* mirid, which is found in Brazil, particularly in the Minas Gerais state, where it feeds on Mexican cotton (*Gossypium hirsutum*) and cowpea plants (Ferreira et al. 2001, Nogueira et al. 2019). Additionally, research on *N. filicornis* is limited, because it is recorded in Peru, French Guiana and Brazil (Schuh 1995, Chérot & Carpintero 2016). This study provides the first record of *N. filicornis* in cowpea crops in Piauí.

Polymerus testaceipes was first recorded in a cowpea crop in Piauí in the present study. This species is widely distributed throughout the New World, from the state of Florida (USA) to Argentina (Carvalho 1959, Almeida & Ferreira 1984, Schuh 1995, Coelho 2008). In Brazil, it has been recorded in 16 states: Amazonas, Acre, Rondônia, Pará, Goiás, Bahia, Pernambuco, Alagoas, Espírito Santo, Paraná, Minas Gerais, Rio de Janeiro, Santa Catarina, São Paulo, Mato Grosso and Rio Grande do Sul (Coelho 2008). *P. testaceipes* has been recorded on several host plants, including *Bidens pilosa* L., *Borreria verticillata* (L.) G. Meyer, *Parthenium hysterophorus* L., cotton (*Gossypium* sp.), tomato (*Lycopersicon esculentum* Mill.), *Amaranthus dubius* Marts. ex Thell., *A. gracilis* Desf. ex Poir, *A. spinosus* L., *Eryngium agavifolium* Griseb., *Daucus carota* L., *Dahlia* sp., *Helianthus* sp., *Cleome* sp. and *N. tabacum* L. (Coelho 2008).

Pycnoderes quadrimaculatus is a pest species widely distributed in mirids in the Caribbean,

Mexico, Central America, South America and the USA (Johnston 1940, Carvalho 1955, Wheeler 2001). In Brazil, it has already been recorded in the states of Minas Gerais, Mato Grosso, Piauí, Espírito Santo and Rio Grande do Sul (Ferreira et al. 2006, Coelho 2008, Nogueira et al. 2019). In addition to cowpea, *P. quadrimaculatus* is associated with cucumber (*Cucumis sativus* L.), pumpkin (*Cucurbita maxima* Duchesne, *C. pepo* L.), common bean (*P. vulgaris*), arum lily [*Zantedeschia aethiopica* (L.) Spreng], *Brachiaria brizantha* (A. Rich.) Stapf., sweet potato (*Ipomoea batatas* L.), melon (*Cucumis melo* var. *cantalupensis* Naudin) and *Sinapis alba* L. (Johnston 1940, Bruner et al. 1945, Zimmerman 1948, Wheeler 2000a, Ferreira et al. 2001, Ruiz 2014, Nogueira et al. 2019).

In addition to phytophagous mirids, predator species such as *M. basicornis* and *Tytthus* sp. were identified, which also have a zoophytophagous feeding habit, meaning that, in the absence of prey, these insects can also feed on plants. Possible mycophagous species, such as *Fulvius* sp., were also identified. The presence of these predatory, mycophagous and zoophytophagous species is crucial for maintaining the balance of pest insect populations, as they play an important role in the natural biological control of crops.

Macrolophus basicornis is distributed in Guatemala, Cuba, Venezuela and Brazil (Bruner et al. 1945, Carvalho et al. 2000, Coelho 2008, Nogueira et al. 2019). In Minas Gerais, it was found in *Nicotiana tabacum* L. (Nogueira et al. 2019), as well as in Santa Catarina, Goiás, Rio de Janeiro, Rio Grande do Sul (Coelho 2008) and Piauí in cowpea plants (Nogueira et al. 2019). Currently, this species is being studied as a potential biological control agent for *Tuta absoluta* Meyrick, 1917 (Lepidoptera: Gelechiidae) in tomato crops, as nymphs and adults of *M. basicornis* effectively feed on the eggs and larvae of *T. absoluta* in the tomato plant (Bueno et al. 2013, Silva et al. 2016, Van Lenteren et al. 2017), in addition to preying on aphids and larvae of lepidopterans (Hernández & Henry 2010).

Tytthus sp. was collected for the first time in Piauí in the present study. It is known for egg-preying insects, especially those of the Delphacidae family (Hemiptera: Auchenorrhyncha), which is important for the biological control of rice and sugarcane crops, where Delphacidae are highly relevant pests (Wheeler 2001). The *Tytthus* genus is distributed throughout the entire northern hemisphere, but some species are also found in the tropics, in China, South America, Australia and the Indo-Pacific region, with a total of 24 species described (Wheeler 2001).

Fulvius sp. is another predatory and possibly a mycetophagous species (Gorczyca 2000, Ferreira & Henry 2002) that was found for the first time in cowpea crops in Piauí as part of this study. This species is among the most numerous and widely distributed genres within the Cylapinae subfamily. It is composed of more than 80 species (Carvalho & Costa 1994, Schuh 1995, Yasunaga 2000, Ferreira & Henry 2002, Sadowska-Woda 2005, Gorczyca 2006) that occur mainly in the Afrotropical, Neotropical and Oriental regions; however, very few studies have researched the biology of this genus.

The population fluctuations of the Miridae species in both seasons revealed variations in richness and diversity indices, when correlated with abiotic factors. The highest abundance and species richness were detected in the rainy season (April to June), when 13 Miridae species were recorded (Table 1; Figure 2). During the dry season (September to November), only *C. rubrinervis* was collected from cowpea crops (Table 1; Figure 2). During the rainy season, *C. rubrinervis*, *H. signoreti* and *C. femoralis* presented the highest relative frequencies (Table 1), with Miridae diversity calculated using the Shannon-Wiener index at H = 1.8489 (confidence interval: 1.831415-1.866320; p = 0.05). The richness index (Margalef) and uniformity or equitability were calculated as ALFA = 2.1810 and E = 0.7710, respectively.

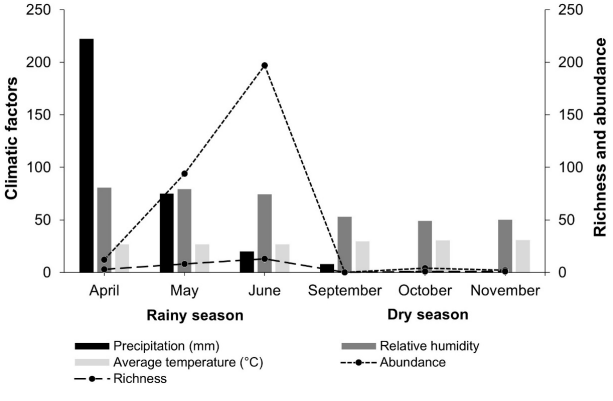


Figure 2. Richness and abundance of Miridae in cowpea during the rainy and dry seasons in Teresina, Piauí state, Brazil, in 2015, using an analysis of the relationship with climatic factors.

The abundance of mirids was negatively correlated with temperature ($r = -0.53$; $p < 0.05$) and rainfall ($r = -0.09$; $p > 0.5$), and positively correlated with humidity ($r = 0.47$; $p < 0.05$). The rainfall, relative humidity and abundance reached their highest levels between April and June. In April, rainfall was 222.4 mm, relative humidity 80.7 % and abundance 2.4 individuals. In May, rainfall decreased to 75 mm, relative humidity remained at 79.3 % and abundance increased to 23.5 individuals. In June, rainfall was further reduced to 20 mm, relative humidity remained at 79.3 % and abundance stabilized at 49.2 individuals. The increase in Miridae abundance can be attributed to the high relative humidity during these months. This is consistent with studies showing that high humidity favors the survival and proliferation of insects, as observed in *Eurystylus oldi* Poppius (Hemiptera: Miridae) in Mali, where its abundance increased with higher humidity (Ratnadass & Butler 2003). Other studies also indicate that elevated humidity (60-80 % RH) improves the fecundity and survival of immature species such as *Apolygus lucorum* (Meyer-Dür) (Heteroptera: Miridae) (Lu & Wu 2011).

In contrast, the rainfall, relative humidity and abundance were lower during the dry season, from September to November, whereas the average temperatures were higher. In September, rainfall was 7.8 mm, the average relative humidity was 53 % and abundance was zero. In October, rainfall decreased to 1.4 mm, relative humidity was 48.9 % and abundance increased slightly to one individual. In November, rainfall was 0 mm, relative humidity increased to 50.3 % and abundance remained at one individual. During the dry period, the high average temperature of 30.2 ºC likely surpassed the tolerance threshold of mirids, reducing their activity and reproduction rates. However, the presence of Miridae during the dry season, although in lower abundance, indicates that some species can survive under adverse climatic conditions. This resilience may pose a challenge for integrated pest management, especially considering that some of the collected species are already reported as pests in other crops of economic importance. For example, *H. signoreti* has been recorded as predominant in cowpea crops in the Amazonas, focusing on the reproductive phenological stage (Alves et al. 2023). The adaptation of these species to different climatic conditions highlights the need for management strategies that consider population dynamics throughout the crop cycle and seasonal variations.

Compared with the dry season, the rainy season, characterized by higher humidity and lower temperatures, favored an increase in the mirid population, whereas extreme conditions such as high temperatures, low humidity and lack of rain hindered the appearance of insects (Figure 2).

The Mirid species were more prevalent during the reproductive phase, particularly during the flowering stage of the cowpea lifecycle (Figure 3). Only *C. rubrinervi* was collected during the vegetative phase with trifoliate leaves (V4) in the cowpea phenological cycle. After the reproductive phase with pre-flowering (V5), the most frequent species were *C. rubrinervis*, *H. signoreti* and *C. femoralis* (Figure 3). Peak populations of *C. rubrinervis* and *H. signoreti* occurred during the reproductive phase with pod-filling (R8), whereas those of *C. femoralis* occurred during the physiological maturation phase of the plant (R9) (Figure 3).

Creontiades rubrinervis and *Horciasinus signoreti* were the most abundant species in the cowpea crops.

For the first time, an association between cowpea and *Cyrtocapsus sp.*, *Fulvius sp.*, *Notholopus filicornis*, *H. guapeanus*, *Orthotylus sp. 1*, *Orthotylus sp. 2*, *P. testaceipes* and *Tytthus sp.* was recorded.

The rainy season favored both the diversity and abundance of Miridae in the Piauí state, Brazil.

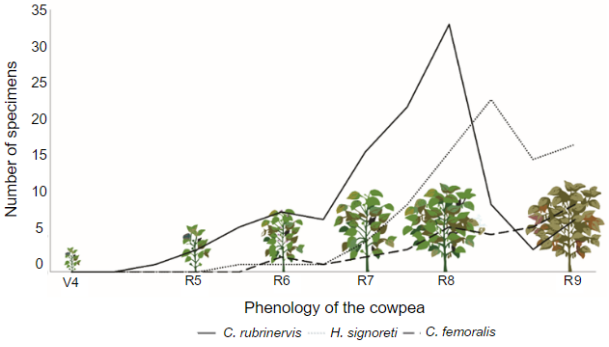


Figure 3. Fluctuations in the cowpea phenology for the most abundant species (*Creontiades rubrinervis*, *Horciasinus signoreti* and *Cyrtocapsus femoralis*) during the rainy season, in Teresina, Piauí state, Brazil, in 2015. V4: vegetative phase with trifoliate plants; R5: reproductive phase with pre-flowering; R6: reproductive phase with flowering; R7: reproductive phase with pod formation; R8: reproductive phase with pod filling; R9: physiological maturation.

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