



Contributions of intercropping systems for diversity and abundance of mite community on *Jatropha curcas*

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Abstract Intercropping is an important strategy of pest biological control and has the potential of increasing abundance and diversity of natural enemies. Here we assessed the effect of six management crop systems on the diversity and abundance of mite pests and predatory mites associated to physic nut crops. The following crop systems were evaluated: jack beans (*Canavalia ensiformis*), guinea grass (*Panicum maximum*), signal grass (*Brachiaria brizantha*), cowpea (*Vigna unguiculata*) in succession to corn (*Zea mays*), physic nut free of spontaneous plants between-rows, and physic nut with spontaneous plants between-rows. Total number of mites was counted and their abundance, diversity and equitability were determined. The most abundant herbivorous mite species found in all crop systems were

Polyphagotarsonemus latus and *Tetranychus bastosi*. *Amblydromalus zannoui*, *Typhlodromus aripo*, *Typhlodromus peregrinus* and *Pronematus* sp. were the most abundant predatory mites. Intercropping cowpea in succession to corn favoured the diversity and abundance of predatory mites in physic nut.

Keywords Physic nut · *Polyphagotarsonemus latus* · *Tetranychus bastosi* · Phytoseiidae · *Vigna unguiculata* · *Zea mays* · Grasses

Introduction

The physic nut (*Jatropha curcas* L. [Malpighiales: Euphorbiaceae]) is a tropical species native of equatorial America (Fairless 2007; Kumar and Sharma 2008) with potential for biodiesel and biokerosene

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production (Duraes et al. 2011). Among the problems related to cropping physic nut are the pests, mainly phytophagous mites (Sarmiento et al. 2011). Studies on the mite fauna associated to physic nut cultivation have demonstrated the existence of a diverse and abundant community, including families of phytophagous (Tarsonemidae, Tenuipalpidae and Tetranychidae), predators (Ascidae, Blattisociidae, Cunaxidae, Iolinidae, Phytoseiidae and Stigmaeidae), as well as others with diverse food habits (Acaridae, Oribatidae and Tydeidae) (Sarmiento et al. 2011; Cruz et al. 2012; Saraiva et al. 2015). The mites *Polyphagotarsonemus latus* Banks (Acari: Tarsonemidae) and *Tetranychus bastosi* Tuttle Baker and Sales (Acari: Tetranychidae) stand out as the main pests of this crop in the world (Sarmiento et al. 2011; Cruz et al. 2012).

Small farmers cultivate physic nut intercropped with beans, corn, cassava, pumpkin, peanuts, and pastures (Marques et al. 2014; Saraiva et al. 2015). The use of intercropping systems may change the abundance and diversity of the mite community in agricultural crops, which may result in an increase of predatory mites in a given management system, reducing the population of pest mites (Zhi-Yi and Zheng 1992; Saraiva et al. 2015). Intercropping systems can provide harbour, food (e.g., pollen and nectar) and suitable microclimate conditions to predators, especially in situations of prey shortage in the main crop (Rosado et al. 2015; Saraiva et al. 2015). However, a positive or negative response on the predator and pest populations depends on the management techniques between rows that can affect, for instance, the quality and quantity of pollen for predatory mites (Zannou et al. 2005). Actions such as soil fertilization, irrigation or mowing between rows of crops may cause changes in the availability of pollen and other alternative foods throughout the seasons (Tixier 2018).

Abiotic factors directly affect the mite community and their effects are extremely important for biological pest control (Skirvin and Fenlon 2003). Among the various climatic variables, temperature shows the most significant effect on mites in agroecosystems (Skirvin et al. 2002). High temperatures may act as a limiting factor for the predation of some mite species (Shipp et al. 1996), which in turn limits the efficiency of some predatory species. Some strategies such as intercropping can act by modifying microclimate conditions in crops, reducing temperatures and

contributing to the development of predatory mites (Zannou et al. 2005; Tixier 2018).

Intercropping can provide both food for predators and better microclimate conditions. Plant species used in intercropping, however, should be carefully chosen, as the abundance and diversity of predatory mites may differ according to leaf anatomy and flowering season of associated crops (Zannou et al. 2005). In deciduous plants, such as physic nut, the choice of the species used in intercropping is even more important. Choosing the right species may result in providing alternative food, shelter and favourable microclimate conditions for predatory mites in a period in which high temperature, extremely low RH and complete absence of leaves coincide.

Suitable knowledge of the intercropping systems that favour the abundance and diversity of predatory mites is fundamental for the adoption of pest control techniques such as conservation biological control. Such information contributes to the incorporation of sustainable and efficient practices in order to reduce the dependence of pesticides and their negative impacts on the environment and human health (Onstad and Crain 2019). The aim of the present study was to evaluate the effect of intercropping systems on the diversity and the abundance of predatory and phytophagous mites in physic nut crops in tropical areas.

Material and methods

Experimental system

The experiment was carried out between August 2012 and July 2013 at the Experimental Campus of the Federal University of Tocantins, Gurupi—TO, Brazil (11° 43' 45" S, 49° 04' 07" W and 278 m altitude). The region presents an equatorial climate with dry winters (Rubel and Kottek 2010). The average annual temperature is 26 °C, with two well-defined seasons in the year, a rainy from November to April, and a dry season from May to October, with an annual average rainfall of 1184.20 mm. Under these conditions, physic nut plants show a natural defoliation between July and September.

Data were collected in a physic nut plantation of 18 months, with spacing of 5 m between planting rows and 2 m between plants, without irrigation. The experiment was a randomized complete block design

with six treatments and four replicates for each of the systems totalizing 24 plots. Each plot (560 m²) had four planting rows with 14 physic nut plants per row. The two central rows of the plot were considered the useful area, discarding one plant at each end, making a total of 24 physic nut plants (240 m²). The following crop management systems were evaluated: (1) physic nut with jack beans (*Canavalia ensiformis* (L.) DC [Fabales: Fabaceae]), (2) physic nut with guinea grass (*Panicum maximum* Jacq. [Poales: Poaceae]), (3) physic nut with signal grass (*Brachiaria brizantha* (Sm.) Griseb. [Poales: Poaceae]), (4) physic nut with cowpea (*Vigna unguiculata* L., Walp. [Fabales: Fabaceae]) in succession to corn (*Zea mays* L. [Poales: Poaceae]), (5) physic nut free of spontaneous plants between rows (by weeding), and (6) physic nut with spontaneous plants between rows. These systems were chosen because they include the main cultivation systems in central Brazil where physic nut is cultivated. Farmers who integrate physic nut production and cattle breeding for meat production commonly use the two grasses: small farmers crop cowpea and corn for food production in integration to physic nut cultivation. Local farmers use jack beans as green manure. Mowing and keeping the rows always clean of spontaneous plants are cultural practices adopted by farmers who cultivate physic nut as a monoculture. Some farmers also adopt the leaving of spontaneous plants growing between rows.

No pesticide was applied on the physic nut crop during the experiments, neither on other associated crops or spontaneous plants. In the plots with pastures (guinea grass and signal grass), two mowing were performed during the rainy season. Jack bean was sown at the beginning of the rainy season (November) and incorporated as green manure at the end of the rainy season (April). Corn was sown in November and maintained for 75 days (up to February), when it was harvested. Cowpea was sown after the corn harvest and it was harvest in April. In plots with maintenance of spontaneous plants, their growth was allowed between rows during all cultivation period, except in the physic nut rows where spontaneous plants were removed (0.5 m each side) by constant weeding.

In the local conditions of the experiments, jack bean began flowering at 65 days after planting, reaching flowering peak at 90 days after planting. The grasses did not flower during the experiments due to constant mowing. Cowpea flowered at 32 days after planting

with peak of flowering at 45 days after planting. Corn began to release pollen from the panicles at 35 days after planting and continued releasing it until 92 days.

Mite sampling

Sampling for evaluation of mite fauna on the physic nut plants started with the appearance of their first leaves in October 2012 and continued until July 2013, and it was done fortnightly. We randomly sampled five physic nut plants from the useful area in each plot every two weeks. In order to get homogeneous sampling of the whole plant in the useful area, each plant was stratified into three parts: apical, middle, and basal (Saraiva et al. 2015). From each sampled plant, a branch was chosen in each stratum. The third and fourth leaves that were totally expanded from the branch were collected, as these are considered the best leaves for sampling mites in the crop (Evaristo et al. 2013). After collection, each sample was stored in plastic bags and kept refrigerated at 10 °C until the time of processing. Washing and sieving with a 0.037 mm mesh was used to recover the mites (Saraiva et al. 2015). They were collected and transferred to tubes containing alcohol at 70% (v/v), counted and mounted on slides with Hoyer medium (Cruz et al. 2012; Saraiva et al. 2015).

Mite specimen identification was done using the dichotomous keys by Chant and McMurtry (1994), Lofego (1998) and Moraes et al. (2004) for Phytoseiidae; Bolland et al. (1998) for Tetranychidae; Welbourn et al. (2003) for Tenuipalpidae; Gerson (1992) for Cheyletidae; and Halliday et al. (1998) for Ascidae and Blattisocidae. Slides were deposited in the collection of the Entomology Laboratory of the Federal University of Tocantins, Gurupi, TO, Brazil.

Data analysis

The total numbers of phytophagous and predatory mites that were counted fortnightly from August 2012 to July 2013 were used as data. Diversity, abundance and equitability of mites in the six crop management systems were determined. The survey of the identified mites is presented including the number of mites per species and the number of mites in each of the systems. Shannon–Wiener index (Shannon 1948) was used to determined diversity and Pielou index (Pielou 1966) for equitability. For both, we use a dispersion measure

calculating the confidence interval (CI) of the arithmetic mean with a 5% probability.

The abundance was determined by the number of individuals of each species for each plant in the plots. After normality (Kolmogorov–Smirnov) and homoscedasticity (Levene) tests, the abundance data were subjected to one-way analysis of variance (ANOVA). If a significant effect was detected, the means were compared by Tukey's HSD post-hoc test. Statistical analyses were performed using the software R v3.6.3 (R Development Core Team 2020).

Results

In total, 11,573 mites of 35 species grouped in nine families were recorded on physic nut plants in the entire experiment. The predatory mites belong to seven families (Ascidae, Blattisociidae, Cheyletidae, Cunaxidae, Iolinidae, Stigmaeidae and Phytoseiidae), and the phytophagous to two families (Tarsonemidae and Tetranychidae). The family with more species was Phytoseiidae, with 17. The families with the highest number of collected individuals were Tarsonemidae, Tetranychidae and Phytoseiidae with 9274, 1821 and 415 mites, respectively (Table 1).

Predatory mites represent 4.13% of all collected mites with a total of 478 individuals. These mites were grouped into seven families and divided into 27 species. *Amblydromalus zannoui* Sourassoua, Sarmiento & Moraes (Acari: Phytoseiidae) and *Typhlodromalus aripo* De Leon (Acari: Phytoseiidae) were the ones with the highest number of collected individuals (101 and 91 individuals, respectively). Three species of the Phytoseiidae (*A. zannoui*, *T. aripo* and *Typhlodromus peregrinus* Nesbitt (Acari: Phytoseiidae)), and one of the Iolinidae family (*Pronematus* sp.) were present in all evaluated systems.

Herbivorous mites represented 95.87% of all collected mites with a total of 11,095 individuals. They were distributed in eight species and grouped into two families. The species *P. latus* and *T. bastosi* were the ones with the highest numbers among the collected individuals (9270 and 1801 individuals, respectively) and the only phytophagous mites present in all management crop systems.

The highest diversity of mite species, determined by the Shannon–Wiener index (H), was found in the intercropping of physic nut with cowpea in succession

to corn (H = 0.8988), followed by the physic nut maintained free of spontaneous plants (H = 0.8833), and by physic nut with signal grass (H = 0.7484) (Table 2). Physic nut intercropped with guinea grass had the lowest diversity value (H = 0.2505). The highest values observed for the Pielou equitability index (J) were for the intercropping systems of physic nut with cowpea in succession to corn (J = 0.2828) and physic nut free of spontaneous plants between rows (J = 0.2818). The lowest J value was observed for the intercropping of physic nut with jack beans (J = 0.0981) (Table 2).

Among all species of mites identified, only the phytophagous *P. latus* and *T. bastosi* and the predatory mites *Pronematus* sp., *T. peregrinus*, *A. zannoui* and *T. aripo* were present in all the systems evaluated. Therefore, their abundance was compared between the evaluated crop systems (Table 3). These species add up to a total of 11,336 individuals, corresponding to 97.95% of the total mites collected in the surveys (Table 1).

P. latus showed the highest abundance in all systems. There was no significant difference among the adopted crop management systems in its abundance ($F_{5,18} = 0.85$; $P = 0.534$). (Table 3). For *T. bastosi*, we observed lower population of mites per plant on physic nut with spontaneous plants (0.09 ± 0.02), guinea grass (0.33 ± 0.20) and signal grass (0.83 ± 0.37), indicating a negative effect of these systems on this mite population ($F_{5,18} = 95.43$; $P < 0.001$). A higher abundance of *T. bastosi* was found on physic nut in the management crop system free of spontaneous plants between rows (14.43 ± 1.33) (Table 3).

The abundance of the predatory mites *Pronematus* sp. ($F_{5,18} = 1.59$; $P = 0.213$) and *T. aripo* ($F_{5,18} = 0.64$; $P = 0.670$) on physic nut plants was not influenced by the crop management system (Table 3). For the other two predatory species, *T. peregrinus* ($F_{5,18} = 37.89$; $P < 0.001$) and *A. zannoui* ($F_{5,18} = 11.63$; $P < 0.001$), higher abundance was found when physic nut was in the system with cowpea in succession to corn (0.23 ± 0.02 and 0.59 ± 0.03 , respectively). Among the other crop systems, *T. peregrinus* and *A. zannoui* abundance did not show statistically significant differences (Table 3).

Table 1 Total numbers of mites collected on physic nut plants in different management systems

Family	Species	Planting systems						Σ of species
		JB	SP	GG	FSP	CC	SG	
Ascidae	<i>Asca germani</i>	–	–	4	–	–	4	8
	<i>Asca</i> sp.	–	–	5	–	–	–	5
	<i>Lasioseius</i> sp.	–	–	1	–	–	2	3
Blattisociidae	<i>Aceodromus convolvuli</i>	–	–	–	–	2	1	3
	<i>Aceodromus</i> sp.	–	–	–	–	–	1	1
Cheyletidae	<i>Cheletogenes ornatus</i>	–	–	–	–	1	–	1
Cunaxidae	Not identified	–	1	–	–	–	–	1
Iolinidae	<i>Pronematus</i> sp.	6	3	4	6	4	14	37
Stigmaeidae	<i>Agistemus brasiliensis</i>	–	2	–	–	–	1	3
	<i>Zetzellia</i> sp.	–	1	–	–	–	–	1
Phytoseiidae	<i>Amblydromalus zannoui</i>	14	9	13	14	47	4	101
	<i>Amblyseius neochiapensis</i>	3	–	1	7	13	4	28
	<i>Amblyseius tamatavensis</i>	–	–	1	4	12	–	17
	<i>Amblyseius</i> sp.	–	1	–	2	2	1	6
	<i>Euseius citrifolius</i>	–	–	2	2	1	4	9
	<i>Euseius concordis</i>	–	4	1	6	22	1	34
	<i>Galendromus</i> sp.	–	–	–	–	1	–	1
	<i>Iphiseiodes zuluagai</i>	–	–	–	2	2	5	9
	<i>Neoseiulus</i> sp.	–	–	1	1	–	1	3
	<i>Paraphytoseius multidentatus</i>	–	–	–	–	1	–	1
	<i>Phytoseius guianensis</i>	–	1	–	1	–	–	2
	<i>Proprioseiopsis cannaensis</i>	2	1	–	4	3	10	20
	<i>Proprioseiopsis</i> sp.	–	–	–	–	–	1	1
	<i>Typhlodromalus aripo</i>	12	13	9	21	23	13	91
	<i>Typhlodromalus peregrinus</i>	6	4	2	5	18	1	36
	<i>Typhlodromalus</i> sp.	–	–	–	–	1	–	1
<i>Typhlodromus transvaalensis</i>	3	2	1	–	46	3	55	
Σ predatory mites in each system		46	41	35	75	196	63	456 ^a
Tarsonemidae	<i>Polyphagotarsonemus latus</i>	1748	1430	1400	1743	1821	1128	9270
	<i>Brevipalpus</i> sp.	1	–	–	–	–	–	1
	<i>Tenuipalpus</i> sp.	–	–	1	–	–	–	1
	Not identified	1	–	–	1	–	–	2
Tetranychidae	<i>Mononychellus planki</i>	–	–	–	1	1	–	2
	<i>Oligonychus</i> sp.	–	–	–	1	–	–	1
	<i>Tetranychus bastosi</i>	26	66	7	1154	346	202	1801
	<i>Tetranychus</i> sp.	–	–	–	15	2	–	17
Σ phytophagous mites in each system		1776	1496	1408	2915	2170	1330	11095 ^b

JB Physic nut with jack beans, SP physic nut with spontaneous plants, GG physic nut with guinea grass, FSP physic nut free of spontaneous plants between rows, CC physic nut with cowpea in succession to corn, SG physic nut with signal grass

^aTotal sum of predatory mites

^bTotal sum of phytophagous mites

Table 2 Number of species, Shannon–Wiener diversity index (H), and Pielou equitability index (E) of adult mites collected on physic nut plants in different crop management systems

Crop management system	Number of species	Diversity (H) [95% CI]	Equitability (E) [95% CI]
Physic nut with jack beans	11	0.2590 [0.2578, 0.2601]	0.0981 [0.0976, 0.0985]
Physic nut with spontaneous plants	14	0.3721 [0.3707, 0.3736]	0.1342 [0.1337, 0.1347]
Physic nut with guinea grass	16	0.2505 [0.2490, 0.2520]	0.0884 [0.0879, 0.0889]
Physic nut free of spontaneous plants between rows	19	0.8833 [0.8827, 0.8840]	0.2818 [0.2816, 0.2820]
Physic nut with cowpea in succession to corn	21	0.8988 [0.8976, 0.8999]	0.2828 [0.2824, 0.2831]
Physic nut with signal grass	15	0.7484 [0.7466, 0.7503]	0.2421 [0.2415, 0.2427]

Estimated values and 95% confidence intervals (CI) are indicated for Shannon–Wiener diversity index (H), and Pielou equitability index (E)

Discussion

Associated plants can act as alternative food for herbivorous mites in times of scarcity in the main crop and also provide shelter and alternative food for predatory mites (Childers and Ueckermann 2015; Tixier 2018). Appropriate intercropping strategies can modify the abundance and diversity of predatory mites by altering biotic and abiotic factors between crop rows. Such modifications, if implemented correctly, can lead to improvements in biological pest control (Tixier 2018).

Intercropping physic nut with cowpea in succession to corn enhances the occurrence of the predatory mites *T. aripo* and *A. zannoui*. The first species is a phytoseiid mite that has potential use for biological control of pest mites from Tarsonemidae and Tetranychidae families (Cruz et al. 2012). *Amblydromalus zannoui* is a new species of phytoseiids, with its first specimens collected in physic nuts from the same region where the present work was carried out (Sourassou et al. 2017). Before its identification at the species level, *A. zannoui* had already been reported as important natural agent for control of *P. latus* and *T. bastosi* (Sarmiento et al. 2011; Cruz et al. 2012) in physic nuts, which led to the identification of the species.

Even with the intercropping management strategies resulting in a high diversity of predatory mites, we observed the presence of phytophagous mites such as *P. latus* and *T. bastosi* in the systems. This is of great importance for conservation biological control purposes, which aims to maintain the predator and prey population in equilibrium but at low economic threshold (Straub et al. 2008). In general, predatory

mites were less abundant than herbivores, but this is justified due to need of a large amount of prey to maintain a certain population of predators (McMurtry and Croft 1997). Intercropping physic nut with cowpea in succession to corn tended to have higher abundance values for *T. peregrinus* and *A. zannoui* mites. Possibly, the provision of pollen by corn and its nutritional input acted as a complementary food to prey in the system. The contribution of pollen associated with the dry climate produces a food reserve that can complement the diet of predators during the transition from the rainy to the dry season, thus guaranteeing the presence of a higher population throughout the sample period. Marques et al. (2014) stated that corn pollen may be an alternative and complementary food source for *Iphiseiodes zuluagai* Denmark & Muma (Acari: Phytoseiidae) and *Euseius concordis* Chant (Acari: Phytoseiidae), both predators of *P. latus* and *T. bastosi* and from the same family as *T. aripo* and *A. zannoui*.

This study presented a general view of how the intercalary management systems influence the diversity, abundance, and equitability of mites focusing on the sampling carried out in the main crop. New studies addressing concomitant sampling on crops and associated plants may open doors for understanding the factors responsible for changes in mite diversity and abundance in agroecosystems. Hypotheses such as the existence of migration of phytophagous mites from crop to intercropped plants (and in the opposite direction) or whether the presence of predatory mites in critical times of the crop is influenced by the supply of alternative food at less favorable times of the year (e.g., when physic nut shows natural defoliation) should be clarified. This information can guide the

Table 3 Abundance of adult mites (average number of mites per plant in the plot \pm SE) collected on physic nut plants in different crop management systems

Family	Species	Crop management system					
		Physic nut with jack beans	Physic nut with spontaneous plants	Physic nut with guinea grass	Physic nut free of spontaneous plants between rows	Physic nut with cowpea in succession to corn	Physic nut with signal grass
Tarsonemidae	<i>Polyphagotarsonemus latus</i>	14.1 \pm 2.12 ^a	17.5 \pm 2.75 ^a	21.85 \pm 2.96 ^a	21.79 \pm 3.28 ^a	22.76 \pm 3.88 ^a	17.88 \pm 5.84 ^a
Tetranychidae	<i>Tetranychus bastosi</i>	2.53 \pm 0.253 ^{bc}	0.09 \pm 0.01 ^c	0.33 \pm 0.06 ^c	14.43 \pm 1.34 ^a	4.33 \pm 0.15 ^b	0.83 \pm 0.05 ^c
Iolinidae	<i>Pronematus</i> sp.	0.18 \pm 0.08 ^a	0.05 \pm 0.02 ^a	0.08 \pm 0.03 ^a	0.08 \pm 0.01 ^a	0.05 \pm 0.02 ^a	0.04 \pm 0.02 ^a
Phytoseiidae	<i>Typhlodromalus peregrinus</i>	0.01 \pm 0.00 ^b	0.03 \pm 0.00 ^b	0.08 \pm 0.01 ^b	0.06 \pm 0.01 ^b	0.23 \pm 0.03 ^a	0.05 \pm 0.01 ^b
	<i>Amblydromalus zanoui</i>	0.05 \pm 0.02 ^b	0.16 \pm 0.06 ^b	0.18 \pm 0.04 ^b	0.18 \pm 0.10 ^b	0.59 \pm 0.03 ^a	0.11 \pm 0.04 ^b
	<i>Typhlodromalus aripo</i>	0.16 \pm 0.06 ^a	0.11 \pm 0.01 ^a	0.15 \pm 0.07 ^a	0.26 \pm 0.18 ^a	0.29 \pm 0.04 ^a	0.16 \pm 0.01 ^a

Means followed by different letters within line are significantly different by Tukey's test ($p > 0.05$)

adoption of practices that stimulate predatory mite migration from associated plants to physic nut, especially on those crop management with high biodiversity and high predator population, as observed with the temporal association of corn and cowpea.

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Compliance with ethical standards

Research involving human and/or animal participants

This article does not contain any studies with human participants performed by any of the authors. All applicable international, national, and/or institutional guidelines for the care and use of animals were followed.

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