

Scutellonema bradys and *Pratylenchus* spp. associated with weeds in yam fields

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Summary – Dry root disease of yam (*Dioscorea* spp.), caused by *Scutellonema bradys*, *Pratylenchus coffeae* and *P. brachyurus*, is prominent among the plant disease problems of yam in Brazil. In order to evaluate the association of these nematode species with weeds, field sampling was performed during 2016 and 2017 in yam-growing areas with a history of the disease within the major counties in the state of Alagoas, Brazil. The frequency of occurrence of both weeds and nematodes was determined, in addition to the estimated population of nematodes in roots. Forty-three weed species were found, of which 23 were infected with at least one of the causal agents of the dry rot disease, *Pratylenchus* spp. being the most prevalent in the fields. *Ageratum conyzoides*, *Commelina benghalensis* and *Emilia coccinea* (in Zona da Mata) and *C. benghalensis* (Agreste region) were especially important due to their high frequency of occurrence and also for supporting populations of *S. bradys* and *Pratylenchus* spp. *Cyperus flavus*, *E. coccinea* and *Spermacoce verticillata* are now recorded as new weed species associated with *S. bradys*.

Keywords – *Ageratum conyzoides*, alternative hosts, Brazil, *Commelina benghalensis*, *Dioscorea* spp., dry root disease, *Emilia coccinea*, lesion nematodes, *Pratylenchus brachyurus*, *Pratylenchus coffeae*, yam nematode.

Yams (*Dioscorea* spp.) are the fourth most important root and tuber crop (Coyné & Affokpon, 2018). World production is estimated at about 73 million t, with Nigeria being the largest producer with approximately 48 million t. In South America, Brazil ranks second with approximately 250 000 t produced in an area of 26 000 ha (FAO-STAT, 2017).

Dry rot is an important field and post-harvest disease of yams, and the causal agents are the migratory endoparasitic nematodes *Scutellonema bradys* (Steiner & LeHew) Andrásy, *Pratylenchus coffeae* (Zimmermann) Filipjev & Schuurmans Stekhoven and *P. brachyurus* (Godfrey) Filipjev & Schuurmans Stekhoven. These nematode species damage yam tubers causing dark necrotic lesions in the cortex and fissures of the tuber skin (Ferraz & Brown, 2016), resulting in heavy losses of 20-30% in Brazil (Pinheiro, 2017). In the state of Alagoas, Brazil, these plant-parasitic nematodes generally occur in mixed populations, mainly *S. bradys* and *P. coffeae* (Muniz *et al.*, 2012). According to Claudius-Cole & Aworetan (2007)

and Braz *et al.* (2016), in the absence of yams, populations of these pathogens survive in the soil and other plant hosts, including weeds, which can preserve or increase the level of inoculum in the soil. Besides behaving as alternative hosts, certain weeds can protect nematodes from pesticides and in an unfavourable environment, provide nematode suppression through antagonism, contribute to changes in future nematode biotic potential, or exert indirect effects through competition with crops or by the effects of weed control strategies on nematode populations. A prominent similarity between most major weeds and plant-parasitic nematodes is that both are place-bound organisms that are passively dispersed (Thomas *et al.*, 2005). Otherwise, infected propagative material provides the main source for dissemination of nematodes (Moura, 2016; Coyné & Affokpon, 2018).

A number of reports have been published on nematodes that are commonly associated with weeds growing in the same fields as crops, such as *Radopholus similis* (Cobb) Thorne, *Helicotylenchus* spp., *Pratylenchus* spp., *Hoplo-*

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laimus seinhorsti Luc, *Meloidogyne* spp., *Rotylenchulus reniformis* Linford & Oliveira (Quénéhervé *et al.*, 2006), *P. speijeri* Luca, Troccoli, Duncan, Subbotin, Waeyenberge, Coyne, Brentu & Inserra (Brentu *et al.*, 2013) in infested banana (*Musa* spp.) fields and *R. reniformis* in cotton areas (Molin & Stetina, 2016). However, there are few studies about weeds serving as reservoirs for the causal agents of the dry rot disease of yams (Carmo *et al.*, 2014). Therefore, the objective of the present study was to survey the occurrence of *S. bradys* and *Pratylenchus* spp. in yam fields in the state of Alagoas in order to evaluate their association with weeds.

Materials and methods

FIELD SURVEY AND IDENTIFICATION OF WEED SPECIES

The survey was performed between 2016 and 2017 in two climatically different regions, involving 11 counties within the state of Alagoas, which are representative of the major yam-growing areas. These counties included Paulo Jacinto, Chã Preta, Viçosa, Quebrangulo, Mar Vermelho, Branquinha, Flexeiras and Murici (called Zona da Mata, with 28 areas); Taquarana, Arapiraca and Limoeiro de Anadia (Agreste region, with 13 areas), with a total of 41 areas.

Weeds were sampled during yam cultivation, at 5-9 months after planting, in areas with a history of dry rot disease incidence. Five plants of each weed species were randomly selected from each area, based on highest frequency, and removed from the soil using a hand trowel. Roots were separated from soil by gently tapping off, and the whole plants were sealed in plastic bags, labelled, placed in cooler boxes, and taken to the Phytopathology Laboratory at the Federal University of Alagoas, Brazil, within 8 h after sampling. At the laboratory, the aerial portions were removed for botanical classification using scissors, and the root systems, combined for each weed species, were then stored in a refrigerator at a temperature between 5-7°C for a maximum period of 24 h before nematode extraction.

Identification of weed species was performed according to the classification system of the Angiosperm Phylogeny Group (APG III, 2009), based on morphological characteristics, with the aid of specific literature (Kissman & Groth, 1997, 1999, 2000; Lorenzi, 2006, 2014).

EXTRACTION, QUANTIFICATION AND FREQUENCY OF NEMATODES ON WEEDS

Roots were washed under tap water, cut into 1-3 cm pieces and composite samples of 5 g roots were processed according Coolen & D'Herde (1972). After extraction, nematodes were killed and fixed in hot 4% formaldehyde solution. The population densities of nematode suspensions were assessed from 1 ml aliquots with the aid of Peters counting slides under an inverted light microscope at 100× magnification. The identification of nematodes was based on morphological characters of mature females according to Mai & Mullin (1996) and Castillo & Vovlas (2007).

The frequency of occurrence of weeds and nematodes was calculated according to Ntidi *et al.* (2012): $FOW = \left(\frac{NLW}{NL}\right) \times 100$, where FOW is the frequency of occurrence of the weed, NLW is the number of localities where the weed species occurred and NL is the number of localities sampled. $FON = \left(\frac{NN}{NL}\right) \times 100$ where FON is the frequency of occurrence of nematodes and NN is the number of times the nematode species occurred in roots of each weed species.

Results

FIELD SURVEY AND IDENTIFICATION OF WEED SPECIES

Forty-three different weed plants belonging to 19 families were found, with the Asteraceae family being predominant (11 species), followed by Cyperaceae with six species (Table 1). The most frequent weed species in Zona da Mata were *Commelina benghalensis* (82%), followed by *Gnaphalium* sp. (79%), *Ageratum conyzoides* (71%), *Hyptis pectinata* (64%) and *Emilia coccinea* (61.0%). In the Agreste region the prevalent weed species were *C. benghalensis* and *A. conyzoides* with 100% and 31% occurrence, respectively.

IDENTIFICATION, FREQUENCY, AND POPULATION DENSITIES OF NEMATODES ON WEEDS

Pratylenchus spp. (*P. coffeae* and *Pratylenchus* sp.) were identified in the areas surveyed, with *P. coffeae* being the predominant species accounting for up to 96% of the overall *Pratylenchus* populations.

For both regions, Zona da Mata and Agreste, *C. benghalensis*, *Gnaphalium* sp., *A. conyzoides*, *H. pectinata* and *E. coccinea* were associated with at least one nematode

Table 1. Family, scientific and common names of weed species collected in the major yam-growing areas in the state of Alagoas, Brazil during 2016 and 2017.

Family	Scientific name	Common name
Amaranthaceae	<i>Amaranthus deflexus</i>	Largefruit amaranth
Amaranthaceae	<i>Amaranthus retroflexus</i>	Redroot amaranth
Amaranthaceae	<i>Amaranthus spinosus</i>	Spiny pigweed
Asteraceae	<i>Acanthospermum hispidum</i>	Starbur
Asteraceae	<i>Ageratum conyzoides</i>	Billygoat weed
Asteraceae	<i>Centratherum punctatum</i>	Brazilian button flower
Asteraceae	<i>Conyza</i> sp.	Hairy fleabane
Asteraceae	<i>Eclipta alba</i>	False daisy
Asteraceae	<i>Emilia coccinea</i>	Scarlet tasselflower
Asteraceae	<i>Emilia fosbergii</i>	Florida tasselflower
Asteraceae	<i>Gnaphalium</i> sp.	Cudweed
Asteraceae	<i>Praxelis pauciflora</i>	Common white snakeroot
Asteraceae	<i>Spilanthes acmella</i>	Toothache
Asteraceae	<i>Tridax procumbens</i>	Tridax dayse
Boraginaceae	<i>Heliotropium indicum</i>	Indian heliotrope
Caryophyllaceae	<i>Drymaria cordata</i>	Tropical chickweed
Commelinaceae	<i>Commelina benghalensis</i>	Wandering Jew
Cyperaceae	<i>Cyperus distans</i>	Piedmont flatsedge
Cyperaceae	<i>Cyperus esculentus</i>	Yellow nutsedge
Cyperaceae	<i>Cyperus flavus</i>	Denton's flatsedge
Cyperaceae	<i>Cyperus iria</i>	Rice flatsedge
Cyperaceae	<i>Cyperus laxus</i>	Sedge
Cyperaceae	<i>Cyperus surinamensis</i>	Tropical flatsedge
Euphorbiaceae	<i>Euphorbia hyssopifolia</i>	Hyssop leaf sandmat
Fabaceae	<i>Indigofera</i> sp.	Indigo
Fabaceae	<i>Mimosa pudica</i>	Common sensitive plant
Lamiaceae	<i>Hyptis pectinata</i>	Comb hyptis
Malvaceae	<i>Corchorus olerarius</i>	Jute
Malvaceae	<i>Sida</i> sp.	Fanpetals
Malvaceae	<i>Waltheria</i> sp.	Sleepy morning
Molluginaceae	<i>Mollugo verticillata</i>	Green carpetweed
Onagraceae	<i>Ludwigia leptocarpa</i>	Anglestem primrose-willow
Phyllanthaceae	<i>Phyllanthus</i> sp.	Seed-under-leaf
Piperaceae	<i>Peperomia transparens</i>	Peperomia
Plantaginaceae	<i>Scoparia dulcis</i>	Sweet broom weed
Poaceae	<i>Brachiaria</i> sp.	Signal grass
Poaceae	<i>Digitaria</i> sp.	Crabgrass
Poaceae	<i>Eleusine indica</i>	Goosegrass
Portulacaceae	<i>Portulaca oleracea</i>	Little hogweed
Rubiaceae	<i>Richardia brasiliensis</i>	White eye
Rubiaceae	<i>Richardia grandiflora</i>	Largeflower Mexican clover
Rubiaceae	<i>Spermacoce verticillata</i>	Shrubby false buttonwood
Solanaceae	<i>Solanum americanum</i>	American black nightshade

species (Tables 2, 3). Seven of the 42 weed species found in Zona da Mata region were parasitised by both *S. bradys* and *Pratylenchus* spp., while 15 were hosts only for *Pratylenchus* spp. and 20 weed species showed no nematodes

infecting their roots (Table 2). By contrast, in the Agreste region, 24 weed species were recorded, and among these species only *C. benghalensis* and *Richardia brasiliensis* were hosts for both *S. bradys* and *Pratylenchus* spp;

Table 2. Frequency of occurrence of weed species, frequency and population densities of *Scutellonema bradys* and *Pratylenchus* spp. (g root)⁻¹ in Zona da Mata Region.

Weeds (number of samples)	FOW (%)	<i>Scutellonema bradys</i>		<i>Pratylenchus</i> spp.	
		FON (%)	PD	FON (%)	PD
<i>Acanthospermum hispidum</i> (2)	7.14	0	0	0	0
<i>Ageratum conyzoides</i> (20)	71.43	17.85	4 (0-6)*	64.29	8 (0-38)
<i>Amaranthus deflexus</i> (3)	10.71	0	0	0	0
<i>Amaranthus retroflexus</i> (1)	3.57	0	0	3.57	2
<i>Amaranthus spinosus</i> (1)	3.57	0	0	0	0
<i>Brachiaria</i> sp. (3)	10.71	0	0	10.71	14 (4-22)
<i>Centratherum punctatum</i> (1)	3.57	0	0	0	0
<i>Commelina benghalensis</i> (23)	82.14	39.29	3 (0-6)	78.57	9 (0-32)
<i>Conyza</i> sp. (6)	21.43	0	0	0	0
<i>Corchorus olitorius</i> (1)	3.57	0	0	0	0
<i>Cyperus distans</i> (2)	7.14	0	0	0	0
<i>Cyperus esculentus</i> (8)	28.57	0	0	28.57	6 (2-18)
<i>Cyperus flavus</i> (2)	7.14	3.57	4 (0-4)	7.14	2
<i>Cyperus iria</i> (1)	3.57	0	0	0	0
<i>Cyperus laxus</i> (7)	25.00	0	0	25.00	15 (2-42)
<i>Cyperus surinamensis</i> (1)	3.57	0	0	0	0
<i>Digitaria</i> sp. (2)	7.14	0	0	7.14	12 (2-22)
<i>Drymaria cordata</i> (1)	3.57	0	0	3.57	2
<i>Eclipta alba</i> (1)	3.57	0	0	0	0
<i>Eleusine indica</i> (8)	28.57	0	0	0	0
<i>Emilia coccinea</i> (17)	60.71	7.14	3 (0-4)	60.71	7 (2-30)
<i>Emilia fosbergii</i> (1)	3.57	0	0	0	0
<i>Euphorbia hyssopifolia</i> (2)	7.14	0	0	0	0
<i>Gnaphalium</i> sp. (22)	78.57	0	0	67.88	4 (0-22)
<i>Heliotropium indicum</i> (1)	3.57	0	0	0	0
<i>Hyptis pectinata</i> (18)	64.29	0	0	57.14	6 (0-18)
<i>Indigofera</i> sp. (1)	3.57	0	0	3.57	12
<i>Ludwigia leptocarpa</i> (3)	10.71	0	0	0	0
<i>Mimosa pudica</i> (9)	32.14	0	0	32.14	14 (2-30)
<i>Mollugo verticillata</i> (7)	25.00	0	0	25.00	4 (2-12)
<i>Peperomia transparens</i> (2)	7.14	0	0	0	0
<i>Phyllanthus</i> sp. (8)	28.57	7.14	8 (0-12)	25.00	9 (0-36)
<i>Portulaca oleracea</i> (1)	3.57	3.57	2	3.57	4
<i>Praxelis pauciflora</i> (5)	17.86	0	0	0	0
<i>Richardia grandiflora</i> (2)	7.14	0	0	7.14	4 (2-6)
<i>Scoparia dulcis</i> (2)	7.14	0	0	7.14	16 (14-18)
<i>Sida</i> sp. (5)	17.86	0	0	17.88	5 (2-16)
<i>Solanum americanum</i> (2)	7.14	0	0	0	0
<i>Spermacoce verticillata</i> (13)	46.43	3.57	2 (0-2)	42.88	5 (0-14)
<i>Spilanthes acmella</i> (13)	46.43	0	0	42.88	3 (0-8)
<i>Tridax procumbens</i> (1)	3.57	0	0	0	0
<i>Waltheria</i> sp. (1)	3.57	0	0	0	0

FOW = Frequency of occurrence of weed species. FON = Frequency of occurrence of nematodes on weed roots. PD = nematode population densities. *Mean (range).

Table 3. Frequency of occurrence of weed species, frequency and population densities of *Scutellonema bradys* and *Pratylenchus* spp. (g root)⁻¹, in Agreste Region, Brazil.

Weeds (number of samples)	FOW (%)	<i>Scutellonema bradys</i>		<i>Pratylenchus</i> spp.	
		FON (%)	PD	FON (%)	PD
<i>Acanthospermum hispidum</i> (2)	15.38	0	0	0	0
<i>Ageratum conyzoides</i> (4)	30.77	0	0	15.38	4 (0-4)
<i>Amaranthus retroflexus</i> (2)	15.38	0	0	7.69	6 (0-6)
<i>Amaranthus spinosus</i> (2)	15.38	0	0	0	0
<i>Centratherum punctatum</i> (1)	7.69	0	0	0	0
<i>Commelina benghalensis</i> (13)	100.00	53.85	2 (0-4)*	76.92	4 (0-8)
<i>Conyza</i> sp.(1)	7.69	0	0	0	0
<i>Cyperus esculentus</i> (1)	7.69	0	0	0	0
<i>Digitaria</i> sp. (1)	7.69	0	0	7.69	2
<i>Drymaria cordata</i> (1)	7.69	0	0	7.69	2
<i>Eclipta alba</i> (1)	15.38	0	0	0	0
<i>Eleusine indica</i> (1)	15.38	0	0	0	0
<i>Emilia coccinea</i> (1)	7.69	0	0	7.69	4
<i>Gnaphalium</i> sp. (2)	15.38	0	0	7.69	6 (0-4)
<i>Heliotropium indicum</i> (1)	7.69	0	0	0	0
<i>Mimosa pudica</i> (1)	7.69	0	0	7.69	2
<i>Phyllanthus</i> sp. (2)	7.69	0	0	7.69	4 (0-4)
<i>Portulaca oleracea</i> (2)	15.38	0	0	15.38	2 (2-4)
<i>Praxelis pauciflora</i> (1)	7.69	0	0	0	0
<i>Richardia brasiliensis</i> (2)	7.69	7.69	2 (0-2)	7.69	2 (0-4)
<i>Richardia grandiflora</i> (2)	15.38	0	0	7.69	4 (0-2)
<i>Solanum americanum</i> (1)	7.69	0	0	0	0
<i>Spilanthes acmella</i> (2)	15.38	0	0	15.38	2
<i>Tridax procumbens</i> (1)	7.69	0	0	0	0

FOW = Frequency of occurrence of weed species. FON = Frequency of occurrence of nematodes on weed roots. PD = nematode population densities. *Mean (range).

11 species were parasitised by only *Pratylenchus* spp., whereas 11 species were not infected by nematodes (Table 3).

In the Zona da Mata region, the highest frequency of occurrence for *S. bradys* was observed in roots of *C. benghalensis* (39%) and *A. conyzoides* (18%), whereas the highest population density of this nematode was observed in roots of *Phyllanthus* sp. with 8 indiv. (g root)⁻¹. However, this weed species showed a low FOW (7%). Seven weed species showed FOW ranging from 79 to 43%, concerning their association with *Pratylenchus* spp., particularly *C. benghalensis*, *Gnaphalium* sp., *A. conyzoides* and *E. coccinea*. The highest population densities for *Pratylenchus* spp. were observed in roots of *Brachiaria* sp., *Cyperus laxus*, *Mimosa pudica* and *Scoparia dulcis* (14-16 indiv. (g root)⁻¹) (Table 2). In the Agreste region, the highest frequency of occurrence for *S. bradys* and *Pratylenchus* spp. was also observed in *C. benghalensis*

with 54 and 77%, and mean population density ranging from 2 to 4 indiv. (g root)⁻¹, respectively (Table 3).

Discussion

Management methods recommended for dry rot disease of yams are based on the use of healthy propagative material planted on areas free of the nematodes (Moura, 2016). Hence, efforts to reduce wild plant hosts that support nematodes should be considered in order to deprive the pathogens from alternative food sources during the yam cropping season. The presence of alternative hosts reduces the efficacy of management techniques designed to lower plant-parasitic nematode populations and thereby enhances crop injury that is proportional to the size of the nematode population (Thomas *et al.*, 2005).

There are few studies on the host range of weed plants for *S. bradys*. Under glasshouse conditions, Ade-

siyan (1976) considered *Corchorus olitorius* and *Tridax procumbens* as poor host and non-host of this nematode species, respectively. However, in a different study, *T. procumbens* was considered as non-host/poor host for this nematode species, depending on the type of inoculum (naturally infested *S. bradys* soil or sterilised soil infested with the nematode) (Kayode & Claudius-Cole, 2017). By contrast, Claudius-Cole & Aworetan (2007) verified that *C. benghalensis* supports the reproduction of this nematode. The findings of the present study are in line with reports by these authors.

The current study also showed an association between *S. bradys* and *A. conyzoides*, *Portulaca oleracea* and *R. brasiliensis*, different to the findings previously reported by Carmo *et al.* (2014), which classified these weed species as non-hosts for this nematode species. These differences can be attributed to the genetic variability of the weed and nematode populations and also to environmental factors. For the first time, *S. bradys* was observed in association with *C. flavus*, *E. coccinea* and *Spermacoce verticillata*.

The higher prevalence of *P. coffeae* can be justified by its wide host range of over 250 plant species, covering almost all plant families (Burke *et al.*, 2005). In terms of host status of weeds to *Pratylenchus* spp., Quénehervé *et al.* (1995, 2006) found that *Amaranthus spinosus*, *Emilia fosbergii*, *Eleusine indica* and *Solanum americanum* were not parasitised by the nematode, in accordance with the present study. Furthermore, *M. pudica* and *P. oleraceae*, referred to as hosts for *P. coffeae* (Bendixen, 1988), were also infected by this nematode species in the present study. The lowest nematode population densities observed in the Agreste region when compared to Zona da Mata, could be attributed to differences in environmental factors and also to a lower number of samples collected.

Many weed species were not associated with the causal agents of dry rot disease of yam. However, this result does not necessarily mean that such association cannot occur; for example, *S. bradys* was associated with *A. conyzoides*, *E. coccinea*, *Phyllanthus* sp. and *P. oleracea* in Zona da Mata, but the nematode was not detected in the Agreste region. In addition, association between *C. esculentus* and *Pratylenchus* spp. was not observed in this region. Thus, glasshouse tests are necessary in order to confirm the host status for these weeds.

Considering that some weed plant species were found to host causative agents of the dry rot of yam, management practices that suppress these plant species will contribute to reduce nematode populations for the next cropping

season. In addition, weed plants that were non-hosts for plant-parasitic nematodes could be tested for the management of these pathogens, through the test of their extracts, as observed by Ferreira *et al.* (2013) who used the weed *T. procumbens* against *Meloidogyne incognita*.

In conclusion, *C. benghalensis*, *A. conyzoides* and *E. coccinea* were the most frequently found weed species in yam-growing areas in Alagoas, being associated with *S. bradys* and *Pratylenchus* spp. *Cyperus flavus*, *E. coccinea* and *S. verticillata* are now recorded as new weed species being associated with *S. bradys* in field conditions.

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