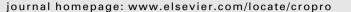
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Pathogenicity of *Scutellonema bradys* populations from different geographical areas in Benin on yam (*Dioscorea* spp.)

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ABSTRACT

Scutellonema bradys is the most important nematode species affecting yam production in Benin. Two experiments were conducted to assess the variation in pathogenicity of three separate populations of S. bradys (IsoSave, IsoToui and IsoGlaz) on seven yam cultivars of three species: Kpouna, Tabane, TDr131, Moroko (Dioscorea rotundata), Banioure, Kokoro (D. cayenensis) and Aloungan (D. alata). This was done over two consecutive yam growing seasons; 2002-2003 at the International Institute of Tropical Agriculture (IITA) Cotonou Station, Benin experimental farm and 2003-2004 in a farmer's field at Save in central Benin. S. bradys populations were obtained from yam originating from different geographical areas of Save and Toui in the northern Guinea savannah and Glazoue in the southern Guinea savannah, respectively. Four-week-old plants were each inoculated with ca. 1000 S. bradys juveniles and adults in chopped, infected pieces of yam peel. The experiments were harvested at 9 and 10 months after planting, respectively, at IITA and Save. Some differences were observed in the pathogenicity of the different nematode populations, although differences were not consistent between experiments and between yam cultivars. At harvest, in 2002–2003, the various S. bradys populations did not affect tuber weight, number of tubers produced or dry rot severity on the tubers. Highest nematode population density was observed with IsoGlaz, and Banioure was the yam cultivar most susceptible to nematode infestation. In 2003-2004, nematode inoculation reduced ($P \le 0.05$) tuber weight for yam cv's Aloungan, Moroko and TDr131. Weights of tubers from IsoToui- and IsoSave-inoculated plants were lower than those from control plants. Dry rot severity was more pronounced on cv's Moroko and TDr131 tubers than on cv's Aloungan and Banioure, while no dry rot symptoms were observed on cv Tabane tubers. IsoToui and IsoGlaz caused higher (P < 0.05) tuber dry rot severity than IsoSave. Differences in nematode population densities occurred across S. bradys populations for cv's Tabane, TDr131 and Aloungan with IsoSave resulting in the highest nematode population densities on cv's Aloungan and TDr131. IsoToui caused the highest percentage weight loss. During 5 months storage, infected tubers lost more ($P \le 0.05$) weight than uninfected control tubers. The differences in weight loss of infected tubers differed on tubers infected by the various nematode populations. Nematode multiplication during the storage period also differed among populations.

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1. Introduction

Yam (*Dioscorea* spp.) is one of the most popular and widely consumed foods in the world. It plays a staple role in the diets of many countries, notably those in Africa, South America, the Pacific Islands, and the West Indies. In 2004, 36.9 million tonnes of yam were produced globally, of which 95.8% was harvested in Africa. In

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Africa, Benin is the fourth largest producer of yam with 2.5 mt after Nigeria (27 mt), Ghana (3.8 mt) and Côte d'Ivoire (3 mt) (FAOstat, 2004). Yam is cultivated in the central and the northern parts of Benin (96.8% of total yam production area), which is characterised by a diversity of soil types (mineral, ferruginous, ferralitic, hydromorphic, vertisols), climate (beninese, sub-sudanian, tropical sudanian, atacorian) and vegetation (fallow, mangrove swamp) (Adam and Boko, 1993; Dansi et al., 2003). Also, numerous cultivars of different yam species are cultivated in Benin, where cultural practices differ among regions and according to ethnic habits (Vernier, 2001).

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Scutellonema bradys (Steiner and LeHew) Andrassy, the causal agent of dry rot, is amongst the most important constraints that challenge yam production in West Africa (Bridge, 1982). During storage, secondary organisms such as mites, fungi or bacteria soon invade the shallow lesions caused by the feeding of the nematode. This leads to the more extensive, internal decay of tubers known as "wet rot" (Adeniji, 1970). A study on the distribution, population density and incidence of plant parasitic nematodes and associated damage of marketed yam on market stalls in several West African countries during 2002 and 2003 demonstrated the presence of S. bradys on marketed yam in all countries assessed (Coyne et al., 2005a). In Benin, S. bradys was first reported on yam in 1992 (Sturhan, 1992) and has been found to be the most important nematode affecting the crop in the country (IITA, 1999). Identification of S. bradys continues to rely on morphological characters. However, although only S. bradys, within the genus Scutellonema, is reported to affect yam and cause dry rot, Scutellonema clathricaudatum Whitehead and Scutellonema cavenessi Sher are closely related, occur in West Africa, are morphologically similar and are easily confused, creating certain taxonomic confusion (Baujard and Martiny, 1995). Traditional approaches in nematode diagnosis are time consuming and expensive and results from these may not be unequivocal (Dhillon, 1998). Key descriptive characters can often vary considerably between nematode species and among races of the same species (Whitehead, 1968). Recently, molecular studies on S. bradys populations extracted from different yam cultivars collected throughout West Africa, including Benin, showed high levels of polymorphism and morphological variability (Coyne et al., 2005b). To date, however, it is not known whether variability in pathogenicity of S. bradys exists between different isolates or geographic populations, and if so, to what extent this is related to yam cultivar.

Selection of a representative nematode population is critical for any resistance screening procedure (Bouquet et al., 1975). Aggressive populations should be selected for screening purposes to detect genotypes possessing the highest level of nematode resistance, while a range of populations to screen against can improve the resistance selection process. The objectives of the current study were firstly to assess the variability of three Benin *S. bradys* populations in terms of pathogenicity on yam pre- and post-harvest, in relation to yam cultivar, based on seven common yam cultivars cultivated in Benin.

2. Materials and methods

2.1. Experimental sites and climatic conditions

The experiments were located at the International Institute of Tropical Agriculture (IITA) Cotonou Station, Benin experimental farm during 2002-2003 and on a farmer's field at Save in central Benin during 2003-2004. IITA-Cotonou Station is located in the coastal savannah zone at 6.25°N, 2.19°E, with an altitude of 23 m above sea level. The climate is sub-equatorial and the soil is of a hydromorphic nature, which is derived from and representative of lowland seasonally flooded areas (Adam and Boko, 1993) and characterised by N – 0.5%, P_2O_5 – 0.27 ppm, K_2O – 0.02 meq for 100 g soil, C – 0.6%, pH – 6.18, and a C/N ratio of 5.37 (Mahougbe, 1996). The minimum total monthly rainfall during the experimental period was 32 mm, recorded in June 2002 and the maximum 393 mm, recorded in August 2002. The minimum average daily temperature during the experiment was 24 °C and the maximum 32 °C. The Save site is located in the northern Guinea savannah at 8.26°N, 2.22°E, characterised by a ferruginous soil, and a sub-Sudanian climate (Adam and Boko, 1993). The minimum total monthly rainfall was 22 mm and the maximum 236 mm. The minimum average daily temperature during the experiment was 25 $^{\circ}\text{C}$ and the maximum 33 $^{\circ}\text{C}.$

2.2. Experimental design and details

The experiments comprised two factors: "nematode population" and "yam cultivar". The factor "nematode population" had four treatment levels at both experimental sites: three separate geographical populations of *S. bradys* from three localities namely Save, Toui and Glazoue in Benin and an uninfected control. The factor "yam cultivar" had four treatment levels at IITA: cv's Banioure, Kokoro, Kpouna and Tabane and five at Save: cv's Banioure, Aloungan, TDr131, Moroko and Tabane. At both sites, a randomised complete block design was used with three replicates.

Yam tubers weighing 400–500 g each (size-1 tubers) were used for planting throughout the study. The tubers were hot-watertreated at 50 °C for 30 min (Smit, 1967) to eliminate S. bradys and they were pre-sprouted to ensure plant uniformity. In the 2002-2003 experiment, yam cv's Banioure and Kokoro (D. cayenensis), Kpouna and Tabane (D. rotundata) were sourced from a farmer at Ina village in the north east of Benin and planted at IITA during April 2002. During April 2003, the experiment was repeated at Save using cv's Banioure (D. cavenensis), Aloungan (D. alata), TDr131, Moroko and Tabane (D. rotundata). Tubers of cv's Banioure and Tabane from the 2002–2003 harvests were used for planting material in 2003. Tubers of cv TDr131 were obtained from IITA-Ibadan, Nigeria, while cv's Aloungan and Moroko tubers were collected from Ina experimental station of the Benin National Institute of Agricultural Research (INRAB). For each of the two experiments, in each block, five yam tubers were planted per cultivar for each S. bradys population. Tubers were planted in mounds (ca. 50-75 cm height) prepared by gathering the surrounding top layer of soil. The plants were staked (one stake per plant) three weeks after planting. The experiments were maintained weed-free by regular hand weeding.

For both experiments, pre-plant nematode population densities in the soil were determined shortly before planting. Soil samples were taken from 30 m \times 20 m areas reserved for the experiments. Using a trowel, approximately 100 g soil was collected from each 1 m², at a depth of ca. 15–20 cm following a systematic collection procedure (Barker and Niblack, 1990). Soil from each 100 m² was combined in a plastic bag and labelled, providing a total of six bulked soil samples. Nematodes were extracted from 3 \times 50 g soil sub-samples for each bulked soil sample, over 48 h by means of a modified Baermann method (Hooper, 1990). Nematodes were identified to genus level and population densities assessed as described below.

2.3. Inoculum preparation and inoculation procedure

Three populations of *S. bradys* were used in the current study. They were obtained from heavily infected yam cv Ala tubers collected from Save ($8.06^{\circ}N$, $2.54^{\circ}E$) and Toui ($8.41^{\circ}N$, $2.41^{\circ}E$) in the northern Guinea savannah and from Glazoue ($7.94^{\circ}N$, $2.25^{\circ}E$) in the southern Guinea savannah in Benin. Inoculum population density was determined prior to inoculation by peeling tubers manually with a kitchen peeler. Several 'strips' of peel were removed from different portions of the tubers, chopped and nematodes extracted from 3×5 g sub-samples using a modified Baermann method over 48 h (Hooper, 1990). The nematode suspension was collected and nematode population density assessed from three 10 ml-aliquots using a Leica Wild M3C stereomicroscope. From the nematode population densities obtained, the weight of yam peel that contained ca. 1000 *S. bradys* juveniles and adults (combined) for each population was calculated. Consequently, 7.3, 7.9 and 8.2 g

S. bradys-infected yam peel (cv Ala) were used, respectively, for Save (IsoSave), Glazoue (IsoGlaz) and Toui (IsoToui) populations, and applied as inoculum per plant at the IITA site. The control treatment consisted of plants inoculated with 7.8 g (average of infected peel weights) of *S. bradys*-free yam peel of yam cv Ala. At Save, 9.6, 8.9 and 9.3 g *S. bradys*-infected yam peel cv Ala were applied per plant as inoculum for Save, Glazoue and Toui populations, respectively. Control treatment plants received each 9.3 g of uninfected yam peel.

For both experiments, plants were inoculated at 4 weeks after planting. A shallow trench was dug at ca. 5 cm radius around the stem of each plant, to a depth (ca. 5–10 cm) that exposed some of the roots. The chopped tuber peel was placed around the roots and then covered with soil (Kwoseh, 2000).

2.4. Assessment of crop yield parameters

Experiments were harvested at yam maturity 9 and 10 months after planting (in December 2002 and in February 2004) at IITA and at Save, respectively. The weight and number of tubers per mound were recorded at harvest.

2.5. Assessment of nematode damage and population density

Nine tubers weighing 300–400 g each (size-2 tubers) were randomly selected per treatment and stored at ca. 25 °C (Lyonga and Ayuk-Takem, 1982; Nwankiti et al., 1988; Osagie, 1992). After 3 months storage, three of the nine size-2 tubers per treatment were randomly selected and re-weighed. Remaining tubers were subsequently used at 4 and 5 months after harvest and percentage weight loss calculated: 100 × [(weight of tuber at assessment period – 100)/weight of tuber at harvest].

At harvest, size-1 tubers were assessed for dry rot severity, while size-2 tubers were assessed for dry rot severity both at harvest and during storage. Size-1 tubers were assessed only at harvest as these were in insufficient quantity for assessment at harvest and during storage. Three tubers were assessed per treatment in a block. Dry rot was scored according to an arbitrary scale of 0–4: 0 = clean tuber; 1-4 = tuber skin showing dry rot symptoms; 1 = 1-25% low symptoms of damage; 2 = 26-50% low to moderate symptoms of damage; 3 = 51-75% moderate to severe symptoms of damage; 4 = 76-100% highly severe symptoms of damage.

Tuber nematode population densities were determined at harvest and during storage (at 3, 4 and 5 months after harvest). For each assessment, nematode population density was determined from three tubers, randomly selected per treatment as previously described. Size-2 tubers were assessed at harvest and at monthly intervals between 3 and 5 months. This was undertaken in order to compare nematode population densities on different sized tubers at harvest. Insufficient size-1 tubers prevented such comparisons during storage.

Nematode population densities were assessed from the same size-2 tubers for which weight loss assessments were made.

2.6. Data analyses

Differences in nematode population densities and percentage weight loss in tubers between treatments were compared with ANOVA, using the SAS system, Version 8 for Windows 1999, following $\log_{10}(x+1)$ transformation of nematode population densities and arcsine(x) transformation of percentage weight loss. Treatment means were separated by Fisher's protected least significant difference (LSD) test or Standard Error, $P \leq 0.05$.

3. Results

3.1. Assessment of crop yield and nematode damage at harvest

In the 2002–2003 experiment, there were no differences in yield per plant (P < 0.05) among S. bradys populations (Table 1). In 2003– 2004, vields of infected cv Banioure and cv Tabane plants were similar but the mean plant yield was greater for the control plants for cv's Aloungan, Moroko and TDr131. Plant yield differences between nematode populations were observed for cv's Aloungan, Moroko and TDr131. The mean plant yield calculated across yam cultivars indicated no yield differences between nematode treatments in 2002-2003. In 2003-2004, differences in tuber yield were observed between plants inoculated with IsoToui and that of plants inoculated with IsoGlaz and of control plants and there were yield differences between plants inoculated with IsoSave and that of control plants (Table 1). For both experiments, the number of tubers produced did not differ among S. bradys populations, except for cv Tabane in 2003-2004 (data not shown). In 2002-2003, tubers presented no symptoms of dry rot at harvest (data not shown). In 2003-2004, cv's Aloungan and Banioure tubers from plants inoculated, respectively, with IsoToui and IsoGlaz had low levels of damage. Tubers of from plants of cv's Moroko and TDr131 inoculated with IsoToui and IsoGlaz were moderately ($P \le 0.05$) affected, but IsoSave-inoculated plants had only low ($P \le 0.05$) levels of damage. No symptoms of dry rot were recorded from cv Tabane tubers. The mean value of tuber dry rot severity calculated across vam cultivars indicated higher values for IsoToui- and Iso-Glaz-inoculated plants than for IsoSave-inoculated and control plants. Dry rot severity was highest on Moroko and TDr131 tubers and lowest on Banioure and Tabane tubers (Table 2).

3.2. Assessment of nematode population density prior to planting and at harvest

No *S. bradys* were recovered from the soil during the pre-plant assessment at either the IITA or Save site.

During the 2002–2003 experiment, the highest ($P \leq 0.05$) nematode population densities at harvest on size-1 tubers were recorded for IsoGlaz on cv Tabane. The highest mean nematode population density was also recorded for IsoGlaz. For the other yam cultivars, population densities were similar across S. bradys populations. Cultivar Banioure was the most susceptible to S. bradys populations (Table 3a). In 2003-2004, the population densities for the different nematode populations differed among yam cultivars (Table 3b). For cv's TDr131 and Aloungan, higher ($P \le 0.05$) nematode population densities were recorded with IsoSave than with other nematode populations. Population densities were similar for the three nematode populations on cv's Banioure and Moroko tubers. In cv Tabane tubers, IsoToui multiplication was lower (P < 0.05) than that of IsoGlaz or IsoSave. Relatively lower nematode population densities were recovered from all cultivars tested in 2003-2004 (except for cv TDr131) than in 2002-2003. The highest mean nematode population density was recorded with IsoSave, with TDr131 appearing the most susceptible yam cultivar (Table 3). S. bradys population densities on size-1 tubers were higher than those on size-2 tubers for both experiments, except for IsoGlaz, for which the population density was higher on size-2 tubers at IITA (Table 4).

In the 2002–2003 experiment, at harvest, the population density of IsoGlaz on the size-2 tubers was higher ($P \le 0.05$) than that of IsoSave or IsoToui (Fig. 1a). In the 2003–2004 experiment, no differences in nematode population densities were observed among the three populations (Fig. 1b). In both experiments, a few nematodes were recovered from uninoculated control tubers (Fig. 1).

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Table 1

Effect of three *Scutellonema bradys* populations on mean yield per plant (g) of seven yam cultivars planted at two sites in Benin over two growing seasons.

(a) Experiment in 2002–2003						
S. bradys population	Banioure	Kokoro		Kpouna	Tabane	Mean
IsoToui	842 ± 871.2	1303 ± 1251.5		816 ± 1020.8	996 ± 933.6	989
IsoSave	1190 ± 905.7	932 ± 970.8		888 ± 1153.4	834 ± 833.2	961
IsoGlaz	1122 ± 663.4	869 ± 754.8		890 ± 784.0	602 ± 557.0	871
Control	903 ± 648.6	688 ± 787.6		1060 ± 1620.5	959 ± 235.7	902
s.e.	ns	ns		ns	ns	ns
(b) Experiment in 2003–2004						
S. bradys population	Aloungan	Banioure	Moroko	TDr131	Tabane	Mean
IsoToui	368 ± 191.6	556 ± 190.6	467 ± 240.7	681 ± 539.2	268 ± 398.2	468 c
IsoSave	1332 ± 712.7	749 ± 301.3	281 ± 215.1	630 ± 320.6	506 ± 281.6	699 bc
IsoGlaz	1060 ± 223.0	557 ± 424.8	465 ± 241.1	991 ± 280.6	636 ± 607.6	742 ab
Control	1440 ± 771.1	779 ± 665.2	998 ± 264.2	1278 ± 461.7	346 ± 195.7	968 a
s.e.	132	ns	115	104	ns	83.69

Values represent mean yield (g) of tubers per plant with a total number of 15 plants per cultivar and per *S. bradys* population.

IsoToui, IsoSave and IsoGlaz = S. bradys populations from Toui, Save and Glazoue areas in Benin, respectively. ns = non significant at $P \le 0.05$. s.e. = standard error.

Table 2

Tuber dry rot severity caused by three populations of Scutellonema bradys at harvest on five yam cultivars during 2003–2004 growing season in Benin.

S. bradys population	2003-2004 experiment at Sa	2003–2004 experiment at Save				
	Aloungan	Banioure	Moroko	TDr131	Tabane	
IsoToui	0.5 ± 0.7	0.0 ± 0.0	2.7 ± 1.2	1.2 ± 1.1	0.0 ± 0.0	0.9
IsoSave	0.0 ± 0.0	0.0 ± 0.0	0.5 ± 1.0	0.8 ± 1.1	0.0 ± 0.0	0.3
IsoGlaz	0.0 ± 0.0	0.2 ± 0.4	$\textbf{2.4} \pm \textbf{1.7}$	2.0 ± 1.4	0.0 ± 0.0	0.9
Control	0.0 ± 0.0	0.0 ± 0.0	$\textbf{0.0} \pm \textbf{0.0}$	0.0 ± 0.0	0.0 ± 0.0	0.0
s.e	ns	ns	0.2	0.2	ns	0.2
Mean ^a	0.17 B	0.07 C	1.87 A	1.33 A	0.00 C	

IsoToui, IsoSave and IsoGlaz = S. bradys populations from Toui, Save and Glazoue areas in Benin, respectively.

Data represent mean values per plant. 15 replicates were used.

s.e. = Standard Error.

 $ns = non significant at P \le 0.05$.

^a In each column, the mean represents an average calculated from the values of tuber dry rot severity for the three *S. bradys* populations.

3.3. Assessment of nematode population density, dry rot and tuber weight loss during storage

In the 2002–2003 experiment, tubers presented no symptoms of dry rot during storage. Nevertheless, 3 months after harvest an increase in nematode population densities was observed for all three populations. From the third month after harvest to conclusion of the experiment at 5 months, nematode population densities decreased on inoculated stored tubers for all three populations, with lowest ($P \le 0.05$) population densities recorded with IsoSave (Fig. 1a).

In the 2003–2004 experiment, dry rot severity at harvest and during storage did not differ between *S. bradys* populations ($P \le 0.05$). Nematode population densities increased until the fourth month after harvest for all three populations and decreased thereafter. Four and 5 months after harvest higher ($P \le 0.05$)

Table 3

Nematode population density at harvest on tubers of seven yam cultivars inoculated with three different *Scutellonema bradys* populations during two yam-growing seasons in Benin.

(a) Experiment in 2002–2003						
S. bradys population	Banioure	Kokoro	I	Kpouna	Tabane	Mean
IsoToui	787 ± 83.5 a	560 ±		641 ± 62.3 a	652 ± 59.6 b	660 b
IsoSave	$681\pm68.3~\mathrm{a}$	$622 \pm$		605 ± 48.7 a	$574 \pm 44.9 \text{ b}$	620 b
IsoGlaz	804 ± 84.0 a	$621 \pm$	57.2 a	659 ± 60.0 a	771 ± 82.9 a	714 a
Control	$0\pm0.0~b$	5 ± 1	3.0 b	8 ± 6.9 b	$11 \pm 9.1 \text{ c}$	6 c
Mean ^a	757 A	601 B		635 B	666 B	
b) Experiment in 2003–2004						
S. bradys population	Aloungan	Banioure	Moroko	TDr131	Tabane	Mean
IsoToui	93 ± 39.6 b	196 ± 31.3 a	204 ± 56.8 a	$491 \pm 232.2 \text{ b}$	$121\pm68.4~b$	221 b
IsoSave	183 ± 79.3 a	174 ± 52.6 a	$181 \pm 39.3 \text{ a}$	655 ± 433.3 a	193 ± 72.2 a	277 a
IsoGlaz	$119\pm38.5~\mathrm{b}$	131 ± 18.4 a	171 ± 38.7 a	$509\pm109.1~\mathrm{b}$	227 ± 115.0 a	230 b
Control	$0\pm0.0\ c$	0 ± 0.0 b	5.58 ± 6.5 b	$4.64\pm2.4~\mathrm{c}$	$3.50 \pm 1.5 c$	3 c
Mean ^a	132 B	167 B	185 B	552 A	181 B	

IsoToui, IsoSave and IsoGlaz = *S. bradys* populations from Toui, Save and Glazoue areas. Values represent nematode population densities on tubers weighing 400–500 g each. For a given yam cultivar, columns with the same letter are not significantly different according to Fisher's protected LSD test, $P \le 0.05$. Statistical analyses was undertaken on $\log_{10}(x+1)$ transformed data.

^a In each column, the mean represents an average value calculated from the three *S. bradys* population densities.

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Table 4

Population density of three Scutellonema bradys populations on size-1 and size-2 yam tubers in two experiments in Benin over two growing seasons^a.

S. bradys population	2002–2003 experiment at	IITA ^b	2003–2004 experiment a	t Save ^b
	Size-1 tubers	Size-2 tubers	Size-1 tubers	Size-2 tubers
Control	2 ± 1.1 a	2 ± 1.3 a	0 ± 0.7 a	1 ± 1.4 a
IsoGlaz	$713\pm243.4~b$	947 ± 234.1 a	231 ± 124.6 a	$141\pm63.8~b$
IsoSave	620 ± 143.2 a	$411\pm132.4~b$	$277 \pm 129.5 \text{ a}$	$157\pm86.1~b$
IsoToui	$660\pm144.9~a$	$552\pm234.3~b$	$281\pm230.1~\text{a}$	$175 \pm 121.6 \ b$

IsoToui, IsoSave and IsoGlaz = S. *bradys* populations from Toui, Save and Glazoue areas in Benin.

For each *S. bradys* population, nematode densities followed by the same letter in a line for a given experiment are not significantly different according to Fisher's protected LSD test, $P \le 0.05$; analyses was undertaken on $\log_{10}(x+1)$ transformed data.

^a Size-1 tubers are tubers weighing 400–500 g each at harvest. Those tubers were not stored. Size-2 tubers are tubers weighing 300–400 g each at harvest and they were stored. Data represent nematode population densities in 5 g yam tuber peel.

Values represent mean nematode densities for four yam cultivars at IITA and five at Save.

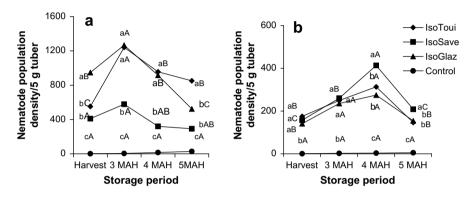


Fig. 1. Population densities on tubers during a 5-month storage period at 25 °C of three *Scutellonema bradys* populations used to inoculate yam cultivars planted at IITA, Benin Station in 2002 (Fig. 1a) and at Save in 2003 (Fig. 1b) in Benin. MAH = Months After Harvest. Size of tubers at harvest: 300–400 g. IsoToui, IsoSave and IsoGlaz = *S. bradys* populations from Toui, Save and Glazoue areas. For each *S. bradys* population, means with the same lower case letter are not significantly different according to Fisher's protected LSD test, $P \le 0.05$. Statistical analyses was undertaken on $\log_{10}(x+1)$ transformed data.

population densities of nematodes were recovered for IsoSave, compared with IsoToui- or IsoGlaz-inoculated tubers (Fig. 1b).

During 2002–2003, 4 and 5 months after harvest, no differences in size-2 tuber weight loss were recorded between S. bradys population treatments. Weight loss of tubers was not affected (P < 0.05) by nematodes when compared to tuber weight loss from uninoculated control treatments. Three months after harvest, the greatest weight loss was recorded for IsoSave (Fig. 2a). In 2003-2004, assessment at the third and the fifth months after harvest showed that tuber weight loss did not differ (P < 0.05) between nematode population treatments. But 4 months after harvest, tuber weight loss was greatest for IsoToui, followed by IsoGlaz and Iso-Save (Fig. 2b). In 2002–2003, the mean tuber weight loss calculated across the storage period showed differences in weight loss of tubers between IsoSave-inoculated plants and IsoGlaz-inoculated and control plants and higher weight losses were observed for tubers from IsoToui- and IsoSave-inoculated plants, compared with tubers from control plants (Table 5). In 2003-2004, differences in weight loss of tubers were observed between inoculated and control plants with the highest weight loss observed for tubers from IsoToui-inoculated plants (Table 5).

4. Discussion

Although the populations used in this study appeared to multiply at different rates, this did not affect the number of tubers produced or dry rot severity over the duration of the study. The effect of the nematodes on dry rot severity at harvest has also been observed by Ayala and Acosta (1971) and Bridge (1973) who also concluded that S. bradys did not cause reduction in yield of yam. However, they reported that S. bradys caused a marked reduction in the marketable value of the tubers during storage. The dry rot symptoms observed on tubers from Save, although no symptoms of tuber dry rot were observed at IITA, indicate that yam tuber dry rot is possibly a consequence of climate in combination with *S. bradys*. which is dependent on yam cultivar. The results obtained, in terms of tuber weight and nematode population density, provides further information that variability between S. bradys populations likely exists and which may be related to molecular and morphological differences (Coyne et al., 2005b). The fact that population densities of the different populations varied between experiments may be related to the conditions at each experimental site. Environmental factors such as rainfall, temperature, humidity, and soil type (Mai, 1985; Curran et al., 1986; Sasser, 1990) may be more favourable for multiplication of the nematodes at IITA than at Save. Luc and Hoestra (1960) indicated that in West Africa, the red soil type was more favourable to S. bradys than fine or coarse sand soil conditions. Initiation of growth of size-1 tubers may have begun before that of size-2 tubers (Tourei and Ahoussou, 1982), which may explain the earlier recorded infection of size-1 tubers and the higher nematode population densities recovered at harvest on size-1 tubers, compared with those on size-2 tubers. The decrease in nematode population densities with duration of storage may be related to the high nematode population densities on the tubers during those periods, leading to competition for nutrients on small tubers (300-400 g each). However, this does not fully explain the situation, as nematode population densities of much higher levels have been observed on marketed yam in Nigeria (Bridge, 1973). The

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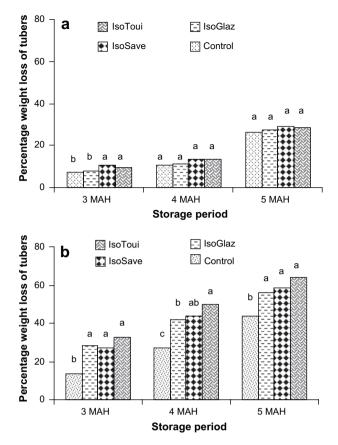


Fig. 2. Percentage weight loss of yam tubers inoculated with three populations of *Scutellonema bradys* at planting and stored for 5 months after harvest at 25 °C in Benin. Data for experiment in 2002–2003 are represented in Fig. 2a and those for 2003–2004 in Fig. 2b. Each column represents the mean across yam cultivars per *S. bradys* population. For a given assessment period, columns with the same letter are not significantly different at *P* \leq 0.05 according to Fisher's protected LSD test. IsoToui, IsoSave and IsoGlaz = *S. bradys* populations from Toui, Save and Glazoue areas. Statistical analyses was undertaken on $\log_{10}(x+1)$ transformed data. Size of tubers at harvest: 300–400 g = size-2.

decrease in nematode population densities during storage may also be related to the physiological changes in the yam tubers, which can affect multiplication of nematodes. Adesiyan et al. (1975) found that the increase in monosaccharides in *S. bradys*-infected yam tubers explained one mode by which the nematode predisposes yam tubers to infection by secondary pathogens (fungi and bacteria). The activities of these invaders subsequently speed up

Table 5

Mean percentage weight loss of yam tubers inoculated with three populations of *Scutellonema bradys* at planting and stored for five months after harvest at 25 $^\circ C$ in Benin.

S. bradys population	2002-2003 experiment at IITA	2003-2004 experiment at Save
IsoToui	$17.13 \pm 9.05 \text{ ab}$	48.86 ± 22.73 a
IsoSave	$17.88\pm9.03~a$	$42.90 \pm 21.12 \ b$
IsoGlaz	$15.48 \pm 10.27 \text{ bc}$	$40.03\pm19.49~b$
Control	14.69 ± 9.45 c	28.29 ± 17.91 c

IsoToui, IsoSave and IsoGlaz = *S. bradys* populations from Toui, Save and Glazoue areas in Benin.

Values for the experiments at IITA and Save represent, respectively, means for four and five yam cultivars. Each value represents mean tuber weight loss for three, four and five months of storage.

For each *S. bradys* population, tuber weight loss followed by the same letter within a column are not significantly different according to Fisher's protected LSD test, $P \le 0.05$.

other physiological processes, such as respiration and water loss, which can affect tuber storage life and *S. bradys* multiplication adversely (Cadet and Quénéhervé, 1994). This contradicts other observations, however, where *S. bradys* populations increased 9- to 14-fold on *D. rotundata* tubers over a 5–6 month storage period Bridge (1973), and five- to eight-fold on *D. alata* and *D. cayenensis* over a similar period Adesiyan (1977). The tubers stored by Bridge (1973) and Adesiyan (1977) were possibly larger than those assessed in the current study. The nematodes recovered from the control tubers were most probably a result of a small number of nematodes surviving the hot water treatment of the planting material and which multiplied during the vegetative period of yam plants and storage of tubers.

The differences in weight loss between *S. bradys*-infected and control tubers (especially in the 2003–2004 experiment) confirm observations by Wood et al. (1980), who indicated that diseased tubers harvested from the field in Nigeria lost up to 29% more weight than healthy tubers. Weight loss occurs during storage due to moisture loss through epidermal layers (Ayala and Acosta, 1971; Bridge, 1973) and as a result of natural metabolic processes, such as respiration. These metabolic processes are more accentuated in tubers infected with nematodes (Cadet and Quénéhervé, 1994) and nematode feeding sites are zones of high metabolic activity (Jones, 1981).

Variation in pathogenicity and the existence of host races and pathotypes or biotypes of plant-parasitic nematodes has previously been recognised for a number of species, such as root-knot nematodes (Meloidogyne spp.), cyst nematodes (Heterodera spp. and Globodera spp.) and stem nematodes (Ditylenchus spp.) (Starr et al., 2002; Luc et al., 2005) and particularly for Radopholus similis (Dochez, 2004). Variants may differ in their reproductive potential on host species or cultivars, which consequently has important implications for the development of pest management strategies. The identification of biotypes or highly virulent populations is particularly important in breeding for resistance against these pests (Bakker et al., 1993). The results of the current study provide preliminary indication that populations of S. bradys with different levels of virulence may exist. The results were not consistent between experiments, but the second year's experiment indicated greater detrimental effect with IsoToui and IsoGlaz on weight of tubers, percentage weight loss and dry rot of tubers, and nematode population density than with IsoSave. Further studies would therefore be necessary with higher initial nematode population densities, and possibly more susceptible yam cultivars to more clearly define differences between the different S. bradys populations.

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References

Adam, K.S., Boko, M., 1993. Le Benin. Agence de Coopération Culturelle et Technique. Les Editions du Flamboyant/EDICEF, Cotonou, Benin.

- Adeniji, M.O., 1970. Fungi associated with storage decay of yam in Nigeria. Phytopathology 60, 590–592.
- Adesiyan, S.O., 1977. Penetration and multiplication of *Scutellonema bradys* in yams (*Dioscorea* spp.). Nematol. Mediterr 5, 313–317.

Adesiyan, S.O., Odihirin, R.A., Adeniji, M.O., 1975. Changes in carbohydrate constituents induced in the yam tuber (*Dioscorea rotundata* Poir) by a plant parasitic nematode *Scutellonema bradys*. Int. Biodeterior. Bull. 11 (4), 124–126.

Ayala, A., Acosta, N., 1971. Observations on yam (*Dioscorea alata*) nematodes. Nematropica 1, 39–40.

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- Bakker, J., Folfertsma, R.T., Roupe van der Voort, J.N.A.M., Boer de, J.N.A.M., Gommers, F.J., 1993. Changing concepts and molecular approaches in the management of virulence genes in potato cyst nematodes. Ann. Rev. Phytopathol 31, 169–190.
- Barker, K.R., Niblack, T.L., 1990. Soil sampling methods and procedures for field diagnosis. In: Zuckerman, B.M., Mai, W.F., Krusberg, L.R. (Eds.), Plant Nematology Laboratory Manual, revised ed. The University of Massachusetts Agricultural Experiment Station, Amherst, Massachusetts, pp. 10–19.
- Baujard, P., Martiny, B., 1995. Ecology and pathogenicity of the Hoplolaimidae (Nemata) from the Sahelian zone of West Africa. 1. Scutellonema clathricaudatum Whitehead, 1959. Fund. Appl. Nematol 18, 347–353.
- Bouquet, D.J., Williams, C., Birchfield, W., 1975. Resistance in soybeans to five Louisiana populations of the root knot nematode. Plant Dis. Rep. 59, 197–200.
- Bridge, J., 1982. Nematodes of yams. In: Miege, J., Lyonga, S.N. (Eds.), Yams Ignames. Oxford Science Publications, pp. 253–264.
- Bridge, J., 1973. Nematodes as pest of yams in Nigeria. Medelelingen Fakulteit Landbouwwetenschapp Gent. 38, 841–852.
 Cadet, P., Quénéhervé, P., 1994. Fluctuations naturelles de Scutellonema bradys
- Cadet, P., Quénéhervé, P., 1994. Fluctuations naturelles de Scutellonema bradys (Nematoda: Hoplolaimidae) au cours de la croissance et du stockage de l'igname (Dioscorea alata) à la Martinique. Nematologica 40, 587–600.
- Coyne, D.L., Tchabi, A., Baimey, H., Labuschagne, N., Rotifa, A., 2005a. Distribution and prevalence of nematodes (*Scutellonema bradys* and *Meloidogyne* spp.) on marketed yam (*Dioscorea* spp.) in West Africa. Field Crops Res. 96, 142–150.
- Coyne, D.L., Williamson, V.M., Al Banna, L., Tchabi, A., Baimey, H., Labuschagne, N. and Rotifa, I. 2005b. Analysis of the pathogenic, morphological and genetic variability of *Scutellonema bradys* on yam in West Africa. In: 17th Nematological Society of South Africa Symposium at Hans Merensky, South Africa, 22–26 May 2005. Book of Abstracts p. 10.
- Curran, J., McClure, M.A., Webster, J.M., 1986. Genotypic differentiation of *Meloi-dogyne* populations by detection of Restriction Fragment Length difference in total DNA. J. Nematol. 18, 83–86.
- Dansi, A., Vernier, P., Marchand, J.-L., 2003. Les variétés d'igname cultivées: Savoir Faire paysan au Bénin. FAO, Groupe de Travail Interdépartemental sur la biodiversité dans l'Alimentation et l'Agriculture., 37 pp
- Dhillon, S., 1998. Use of New techniques to develop nematode diagnostic tools. 1–5 September 1997. In: Sharma, S.B., Johansen, C., Midha, S.K. (Eds.), Nematode Pests in Rice-wheat-legume Cropping Systems. Proceedings of a Regional Training Course. CCS Haryana Agricultural University, Hisar, Haryana, India, pp. 30–35.
- Dochez, C., 2004. Breeding for resistance to *Radopholus similis* in East African highland bananas (*Musa* spp.). PhD Thesis. Katholieke Universiteit Leuven, Belgium.
- FAOstat data, 2004. Food and Agriculture Organization of the United Nations, Rome. Hooper, D.J., 1990. Extraction and processing of plant and soil nematodes. In:
- Luc, M., Sikora, R.A., Bridge, J. (Eds.), Plant Parasitic Nematodes in Subtropical and Tropical Agriculture. CAB International, Wallingford, UK, pp. 45–68.
 Int. Inst. Trop. Agric (IITA). 1999. Annual Report. Ibadan, Nigeria. IITA.
- Jones, M.G.K., 1981. Host cell responses to endoparasitic attack: structure and function of giant cells and syncytia. Ann. Appl. Biol. 97, 353–372.

- Kwoseh, C.K., 2000. Identification of resistance to major nematode pest of yams (*Dioscorea* spp.) in West Africa. PhD thesis. The University of Reading, UK.
- Luc, L., Hoestra, H., 1960. Cited by the Commonwealth Institute of Helminthology. Description of Plant-Parasitic Nematodes. Set 1, No. 10.
- Luc, M., Sikora, R., Bridge, J., 2005. Plant Parasitic Nematodes in Subtropical and Tropical Agriculture, Revised second ed. CAB International, UK.
- Lyonga, S.N., Ayuk-Takem, J.A., 1982. Investigations on selection and production of the edible yams (*Dioscorea* spp.) in the western highlands of the United Republic of Cameroon. In: Miege, J., Lyonga, S.N. (Eds.), Yams – Ignames. Oxford Science Publications, pp. 60–87.
- Mahougbe, C.A., 1996. Influence de la date d'application des extraits de neem, Azadirachta indica (A. Juss) sur les thrips de fleurs Megalurothrips sjostedti (Trybom), et leurs ennemis naturels dans la culture du niébé, Vigna unguiculata (L.) Walp. Mémoire de fin de cycle pour l'obtention du DEAT. Lycée Agricole Medji de Sékou, Allada, Bénin.
- Mai, W.F., 1985. Plant-parasitic nematodes: their threat to agriculture. In: Sasser, J.N., Carter, C.C. (Eds.), An Advanced Treatise on Meloidogyne. Biology and Control, vol. 1. Department of Plant Pathology/United States Agency for International Development, USA, pp. 10–17.
- Nwankiti, A.O., Atu, U.G., Alozie, S.O., Igbokwe, M., Okpokiri, A., Nnodu, E., 1988. Effect of modifying two traditional yam barn structures on storability of four cultivars of yam (*Dioscorea* spp.). Trop. Agric. 65, 125–128.
- Osagie, A.U., 1992. The Yam Tuber in Storage. Postharvest Research Unit. University of Benin, Benin City, Nigeria.
- Sasser, J.N., 1990. Preface. In: Luc, M., Sikora, R.A., Bridge, J. (Eds.), Plant Parasitic Nematodes in Subtropical and Tropical Agriculture. CAB International, Wallingford, UK, pp. ix-x.
- Smit, J.J., 1967. Nematodes. In: Coursey, D.G. (Ed.), Yams. Longmans, London. UK, pp. 115–120.
- Starr, J.L., Cook, R., Bridge, J., 2002. Plant Resistance to Parasitic Nematodes. CAB International, UK.
- Sturhan, D., 1992. Rapport de mission sur l'identification et la lutte contre les nématodes au Projet Bénino-Allemand de la Protection des Végétaux au Bénin. SPV, Porto-Novo, Bénin [Abstr.].
- Toure, B., Ahoussou, N., 1982. Etude du comportement en collection des ignames (*Dioscorea* spp.) dans deux régions écologiques différentes de la Côte d'Ivoire. In: Miege, J., Lyonga, S.N. (Eds.), Yams-Ignames. Oxford Science Publications, pp. 23–30.
- Vernier, P., 2001. Enquêtes sur les systèmes de culture à base d'igname dans le département du Borgou et du Zou, Bénin. Campagne 1996–1997. Rapport de travail. Unité de Coordination des Recherches sur les Ignames. CIRAD – IITA, Montpellier, France.
- Whitehead, A.B., 1968. Taxonomy of *Meloidogyne* (Nematoda: Leteroderidae) with description of four new species. Trans. Zool. Soc. Lond 31, 263–401.
- Wood, T.G., Smith, R.W., Johnson, R.A., Komolafe, P.O., 1980. Termite damage and crop loss studies in Nigeria – Pre-harvest losses to yams due to termites and other soil pests. Trop. Pest Manag. 26, 355–370.