

# Evaluation of Soybean Genotypes (*Glycine max* L. Merr.) Susceptibility to Parasitic Nematodes in Western Part of Burkina Faso

# Gilles Ibié Thio<sup>1\*</sup>, Bouma Thio<sup>1</sup>, Nofou Ouédraogo<sup>1</sup>, Frank Essem<sup>2</sup>, Inoussa Drabo<sup>1</sup>, Oumar Boro<sup>1</sup>, Fabrice Wendyam Nikiema<sup>1</sup>, Pierre Alexandre Eric Djifaby Sombié<sup>1</sup>, David Yago<sup>1</sup>, Hervé Bépio Bama<sup>1</sup>, Mahamadou Sawadogo<sup>3</sup>, Paco Sérémé<sup>1</sup>

<sup>1</sup>Institut de l'Environnement et de Recherches Agricoles (INERA), Ouagadougou, Burkina Faso <sup>2</sup>Akenten Appiah-Menka University of Skills Training and Entrepreneurial Development, Ashanti, Mampong, Ghana <sup>3</sup>Université Joseph KI-ZERBO/UFR Science de la Vie et de la Terre, Ouagadougou, Burkina Faso

Email: \*gilthiolpr@gmail.com

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# Abstract

Parasitic nematodes have been reported as one of the major constraints to soybean production worldwide. The majority of nematodes are so-called "freeliving" and feed mainly on bacteria, fungi, protozoa and other nematodes. In Burkina Faso, the presence of parasitic nematodes has been reported in crops such as rice and sorghum. The objective of this study was to identify the genera and species of nematodes associated with soybean production in Burkina Faso. Investigations were carried out on 24 genotypes of a medium-maturity group of soybean at the Farako-Ba research station. Soil samples were taken from the trial soil before its installation. At harvest, soil samples with roots were taken from each genotype in the elementary plots. The composite sample is represented by 4 lots of soil samples with roots. Extractions and counts of nematodes were performed on the different lots. In total, 7 genera of plantparasitic nematodes associated with soybean were identified. Among these genera, Pratylenchus (100% of infected genotypes), Helicotylenchus (97.28%) and Scutellonema (94.44%) were the most prevalent in terms of frequency and abundance. Some soybean genotypes were less susceptible to the genus Pratylenchus is known to be highly pathogenic in soybean. These were mainly the genotypes TGX2025-10E, TGX2023-3E and TGX2025-14E.

# **Keywords**

Parasitic Nematodes, Susceptibility, Genotypes, Soybean, Burkina Faso

# 1. Introduction

Soybean (Glycine max L. Merr.) is one of the most important oilseed legumes in the world with an estimated global grain production of over 380 million tons [1]. It is used mainly in human and animal food. Soybean is an important source of oil, protein, carbohydrates, isoflavones and minerals [2]. In Burkina Faso, soybean cultivation has become a strategic crop that contributes to building the resilience of poor households to food insecurity [3]. Despite the importance of this crop, very few improved varieties have been developed and those that are popularized are reported to have low productivity [4]. Also, several constraints such as drought, disease attacks and pests such as nematodes contribute to reducing the yields of soybean varieties [5]. The majority of nematodes are so-called "freeliving" and feed mainly on bacteria, fungi, protozoa and other nematodes [6]. It is therefore a minority of nematodes that parasitize animals and plants. Among plant parasitic nematodes, root nematodes such as Meloidogyne incognita, Meloidogyne javanica and Meloidogyne arenaria species cause enormous damage to soybean production [7]. This damage causes soybean yield losses ranging from 18% to 56% [8]. Several factors such as the number of nematodes, the virulence of the species, the resistance or tolerance of the host plant, climate, water availability, soil type, fertility, and the presence of other diseases contribute to nematode damage [9]. Furthermore, the presence of nematodes in the roots causes a blockage of rhizobial activity, which limits the impacts of nodulation and inoculations on plant development and yields [10]. Nematodes are likely to be a threat in all regions of the world where soybean is grown, and effective control methods must be sought [11]. This is the first report of parasitic nematodes from soybean in Burkina Faso, and the study was conducted from the perspective of varietal control of soybean root nematodes. Specifically, the aim is to 1) determine the composition and importance of nematode species and 2) identify sources of resistance in order to strengthen the soybean breeding program.

# 2. Materials and Methods

#### 2.1. Plant Material

The planting materials used in this study comprised of twenty one (21) introduced soybean lines developed by IITA (Abuja, Nigeria) and three (3) soybean varieties (G175, G196 and G197) already used in soybean research programme in Burkina Faso used as local checks (**Table 1**). The seeds of the 21 soybean accessions were treated with an insecticide (Phostoxin) and a fungicide (Bendaco) before being transferred to Burkina Faso.

# 2.2. Experimental Design

The experimental set-up was an alpha lattice design with 3 replicates separated from each other by 2 m. Each repetition consisted of 24 entries distributed in 4 blocks, each consisting of 6 varieties. The blocks were separated from each other

Ord_ID.	Genotypes name	Genetic nature	Origin	Seed color	Maturity group
<b>V</b> 1	TGX2025-6E	Line	IITA	Yellow	Medium
V2	TGX2009-16F	Line	IITA	Yellow	Medium
<b>V</b> 3	TGX2017-5E	Line	IITA	Yellow	Medium
V4	TGX2025-9E	Line	IITA	Yellow	Medium
V5	TGX2016-3E	Line	IITA	Yellow	Medium
V6	TGX2011-6F	Line	IITA	Yellow	Medium
<b>V</b> 7	TGX1987-14F	Line	IITA	Yellow	Medium
<b>V</b> 8	TGX2017-6E	Line	IITA	Yellow	Medium
V9	TGX2025-10E	Line	IITA	Yellow	Medium
V10	TGX2015-1E	Line	IITA	Yellow	Medium
V11	TGX2025-14E	Line	IITA	Yellow	Medium
V12	TGX2008-4F	Line	IITA	Yellow	Medium
V13	TGX1989-19F	Line	IITA	Yellow	Medium
V14	TGX2022-4E	Line	IITA	Yellow	Medium
V15	TGX1993-4FN	Line	IITA	Yellow	Medium
V16	TGX2010-11F	Line	IITA	Yellow	Medium
V17	TGX2020-1E	Line	IITA	Yellow	Medium
V18	TGX2019-1E	Line	IITA	Yellow	Medium
V19	TGX2016-4E	Line	IITA	Yellow	Medium
V20	TGX2027-1E	Line	IITA	Yellow	Medium
V21	TGX2023-3E	Line	IITA	Yellow	Medium
V22	G175 (check 1)	Line	INERA	Yellow	Early
V23	G196 (check 2)	Line	INERA	Yellow	Medium
V24	G197 (check 3)	Line	INERA	Yellow	Medium

Table 1. List of the 24 soybean genotypes (TGx: Tropical *Glycine* from cross).

by 80 cm. The elementary plot was represented by 4 rows of 4 m with 50 cm of row spacing.

# 2.3. Sampling of Nematodes

Nematodes were isolated in two stages from soil samples and transferred to the Nematology Laboratory of DRREA/West. The first sample was taken during the establishment of the trial and the second one after the harvest, on October 10, 2020 (111 day after sowing). For the second sampling, the composite sample was represented by 4 soil samples with the roots of each soybean variety and the root-soil mass was put in the same labeled plastic bag and kept cold until the moment of extraction.

#### 2.4. Extraction of Nematodes from Soil Samples

The Elutriator method of [12] was used to extract nematodes from the soil during the first sampling stage. For the second sampling which was done after harvest, the modified Baerman tunnel method was used. It consisted of separating the soil roots from the sample and labeling them separately. Thus, a volume of 100 cubic centimeters (cc) of soil was taken and added to 300 mL volume of water. After 72 h of filtering on a tissue paper, the nematode suspension was collected in jars. Nematode counts were performed from a 25 mL volume suspension through a 5 Micron diameter sieve. Nematode population densities were expressed as number of nematodes/dm<sup>3</sup> of soil.

#### 2.5. Extraction of Nematodes from Root Samples

The roots were lightly shaken to remove soil clumps and rinsed under a stream of water and then lightly dried on a paper towel. The roots were cut finely with scissors, weighed and placed in a coarse mesh sieve on two layers of tissue paper and placed in a cup. A volume of 200 mL of water was poured over the roots which were left to incubate for 72 hours at room temperature. Nematode counts of the roots were performed as previously described and nematode population densities were expressed as number of nematodes/g root.

## 2.6. Nematodes Count

Nematode frequencies were calculated from the formula

$$F = \frac{e}{n} \times 100$$

with e the number of samples containing the genus of interest, and n the total number of samples. Nematode abundance was determined by the formula

$$A = \sum xi/e$$

with *xi* the number of individuals of the genus under consideration per liter of soil or per gram of dry root and *e* the number of samples in which the genus under consideration was present.

## 2.7. Data Analysis

The data analysis was performed using Excel for the generation of the graphs. After the transformation of the raw data into  $Log_{10}(X + 1)$  where X is the observed data, an analysis of variance (ANOVA) was performed using GenStat Release 12.1 on all variables. All treatment means were compared using the Duncan multiple comparisons test at a 5% level of significance. The principal component analysis and Cluster analysis were performed with R software.

#### **3. Results**

# 3.1. Main Parasitic Nematode Genera Associated with Soybean

Nematological analysis of soil and root samples of soybean varieties revealed the presence of seven (7) genera of plant-parasitic nematodes which are the genera *Meloidogyne, Pratylenchus, Helicotylenchus, Scutellonema, Xiphinema, Tylen-chorhynchus* and *Paratrichodorus.* The inventory reveals that the genera *Pratylenchus, Helicotylenchus* and *Scutellonema* are the most important in terms of frequency and abundance. The specie *Pratylenchus brachyurus* represented the highest density with 7805 nematodes/dm<sup>3</sup> followed by *Helicotylenchus dihystera* with 362 nematodes/dm<sup>3</sup> and *Scutellonema cavenessi* with 251 nematodes/dm<sup>3</sup>. The lowest density was found in the specie *Paratrichodorus* are polyphagous and favor the transmission of viral diseases. **Table 2** presents the different genera encountered and their frequencies.

#### 3.2. Response of Soybean Genotypes to Parasitic Nematodes

The results of the analyses of variance (ANOVA) indicated no significant differences among soybean genotypes for the population densities of soil and root nematodes belonging to the genera *Pratylenchus*, *Helicotylenchus* and *Scutellonema*. Table 3 gives the mean values of population densities of the main soil and root parasitic nematodes according to soybean genotypes.

#### 3.3. Interaction between Soil Nematodes and Soybean Genotypes

For the *Pratylenchus brachyurus* communities present in the soil, the mean values of densities ranged from 2233 to 17,000 N/dm<sup>3</sup> of soil with a mean of 7805 N/dm<sup>3</sup> of soil. Genotype TGX1989-19F and TGX2015-1E were the least infested with 2233 and 2250 N/dm<sup>3</sup> of soil respectively. Genotype TGX2027-1E was the most attacked with 17,000 N/dm<sup>3</sup> of soil.

	Frequency (%)		Abundance		
Genera	Soil	Root	Soil nematodes (N/dm³ of soil)	Root-knot nematodes (N/g of root)	
Pratylenchus	100	100	7804.86	61.68	
Helicotylenchus	65.28	65.28	361.81	3.41	
Scutellonema	50	50	250.69	2.73	
Tylenchorhynchus	29.16	-	21.53	-	
Xiphinema	34.72	-	29.86	-	
Meloidogyne	9.72	-	7.64	-	
Paratrichodorus	1.38	-	1.39	-	

Table 2. The main genera of parasitic nematodes, their frequency and abundance.

Soybean genotypes	Nematode community from soil (N/dm³ of soil)			Nematodes community from root (N/g of root)	
	Prat	Hel	Scu	Prat	Scu
TGX2025-6E	12367a	200a	433a	21.18b	5.44a
TGX2009-16F	3967a	367a	217a	50.16b	2.80a
TGX2017-5E	8900a	283a	450a	40.11	0a
TGX2025-9E	11917a	283a	333a	91.90ab	1.85a
TGX2016-3E	5183a	150a	150a	53.15b	0a
TGX2011-6F	7583a	367a	217a	202.81a	4.76a
TGX1987-14F	14450a	267a	283a	110.62ab	3.05a
TGX2017-6E	7283a	183a	233a	60.51b	3.84a
TGX2025-10E	3400a	317a	233a	16.56b	3.51a
TGX2015-1E	2250a	500a	233a	53.93b	3.93a
TGX2025-14E	7333a	250a	133a	39.04b	2.05a
TGX2008-4F	8250a	267a	100a	24.40b	0.79a
TGX1989-19F	2233a	467a	383a	49.51b	0a
TGX2022-4E	4550a	400a	150a	56.49b	2.12a
TGX1993-4FN	7117a	483a	350a	40.69b	3.78a
TGX2010-11F	9750a	450a	300a	104.96ab	4.34a
TGX2020-1E	8233a	217a	233a	63.45ab	4.17a
TGX2019-1E	10500a	383a	100a	39.76b	1.85a
TGX2016-4E	15333a	667a	433a	149.36ab	7.58a
TGX2027-1E	17000a	650a	300a	21.78b	2.60a
TGX2023-3E	5600a	233a	150a	33.42b	1.57a
G175 (check 1)	3033a	167a	150a	57.50b	1.75a
G196 (check 2)	6000a	333a	200a	55.44b	2.20a
G197 (check 3)	5083a	800a	250a	43.70b	1.55a
Mean	7805	362	251	61.68	2.73
CV (%)	94.7	100.3	91.6	108.1	169.8
P value	0.565 <sup>NS</sup>	0.876 <sup>NS</sup>	0.889 <sup>NS</sup>	$0.245^{NS}$	0.976 <sup>NS</sup>

Table 3. Mean values of population densities of the main parasitic nematodes genera.

Prat: *Pratylenchus*, Hel: *Helicotylenchus*, Scu: *Scutellonema*; NS: No Significant. Means followed by the same letter in a column are not significantly different at  $P \le 0.05$ .

For the *Helicotylenchus dihystera* communities present in the soil, the average values of population densities varied between 150 and 800 N/dm<sup>3</sup> of soil. Genotypes G175, TGX2016-3E, TGX2017-6E and TGX2025-6E showed the lowest le-

vels of infestation while the highest densities were obtained with the check variety G197 at 800 nematodes/dm<sup>3</sup> of soil.

For the *Scutellonema cavenessi* communities present in the soil, the average values of population densities were significantly low compared to the first two genera. Population densities ranged from 100 to 450 N/dm<sup>3</sup> of soil. Genotypes TGX2008-4F and TGX2019-1E had the lowest levels of infestation.

# 3.4. Interaction between Root-Knot Nematodes and Soybean Genotypes

At the root level, population densities were very low compared to those present in the soil. The genera *Pratylenchus brachyurus* and *Helicotylenchus dihystera* were the most representative with an average of 61.68 and 2.73 N/g of roots, respectively.

For the *Pratylenchus brachyurus* community, genotype TGX2025-10E showed the lowest level of infestation with 16.56 N/g of root. It was followed by geno-types TGX2025-6E and TGX2027-1E with 21.18 and 21.78 N/g of root, respectively. Genotype TGX2011-6F recorded the highest density value of 202.81 N/g of root. For the nematode community of the specie *Scutellonema cavenessi*, the average values of population densities were very low and ranged from 0 to 7.58 N/g of root with an average of 2.73 N/g of root. Genotypes TGX2017-5E, TGX2016-3E and TGX1989-19F with each 0 N/g of root were free of infestation while variety TGX2016-4E showed the highest infestation with 7.58 N/g of root.

#### 3.5. PCA Analysis

Figure 1 shows the results of the principal component analysis (PCA) of different agronomic and parasitic nematode distribution parameters as well as the



**Figure 1.** Variables and genotypes distribution in the PCA Biplot design. **Legendre: Flo50**: days to 50% flowering; **Mat50**: days to 50% maturity; **NbNod**: number of nodules per plant; **NbPodPlt**: number of pods per plant; **PH**: plant height; **SW100**: hundred seeds weight.

distribution of individuals in the biplot. Analysis distinguished two axes that explain 42.9% of the total genetic variability within the soybean genotypes and parasitic nematode densities. All variables are somewhat well represented in the biplot whereas SW100, soils nematodes Pratylenchus brachyurus (SNPra), Scutellonema cavenessi (SNScu) and Helicotylenchus dihystera (SNHel), Mat50 and GY weakly represented in the biplot (Figure 1), they do not strongly contribute to the formation of either axe 1 or axe 2. However, we can distinguish correlated variables and groups of variables. Soils nematodes SNHel, SNPra, SNScu and root nematode RNScu communities constitutes a group of correlated variables and were positively correlated to axis 1 (26.7% of the total variability). Among the parasitic nematodes, Scutellonema cavenessi from root (RNScu) contributed around 12% of the total variability according to the contribution diagram in Figure 1 and individual TGX2016-4E is highlighted to be well performing for that feature. Scutellonema cavenessi (SNScu) and Pratylenchus brachyurus (SNPra) from soil contributed each between 9% to 12% to the variability. Helicotylenchus dihystera from soil (SNHel) contributed very little to the variability, i.e., 6% of the total variability. Flo50 and NbNod are also well represented, correlated and contributed each up to 8% of the total variability. Individuals are widely dispersed in the diagram and show how much variability is within the plant materials studied.

#### 3.6. Cluster Analysis in Soybean Genotypes

The cluster analysis resulting from the hierarchical ascending classification (HAC) grouped the genotypes of soybean into three clusters (**Figure 2**). Cluster 1 consisted of 11 genotypes including genotypes TGX2011-6F, TGX1993-4FN and TGX2025-9E. The genotypes in this group are characterized by their low soil densities to species of the specie *Helicotylenchus dihystera*. Cluster 2 consisted of 3 genotypes TGX2016-4E, TGX1987-14F, and TGX2027-1E that exhibited the highest root population densities to the genus *Pratylenchus*. Genotypes of this group also exhibited the highest soil population densities to the species *Pratylenchus*.



Figure 2. Dendrogram of cluster analysis of soybean genotypes susceptibility to parasitic nematodes.

*brachyurus, Helicotylenchus dihystera* and *Scutellonema*. Cluster 3 contains 10 genotypes including the three checks varieties (G175, G196 and G197) that showed the lowest root and soil population densities to the specie *Pratylenchus brachyurus*. These genotypes can be used as sources of resistance in varietal control against species of the three major nematode genera identified including the genus *Pratylenchus*.

### 4. Discussion

Many genera and species of nematodes have been associated with damage in several plant species worldwide [13] [14]. This damage causes yield losses in most plants ranging from 10% - 15% of production [15]. Among the nematode genera, the genera Heterodera, Meloidogyne, and Pratylenchus are the most widespread in the world [16] [17]. More than 100 nematode species, comprising 50 genera, have been reported in association with soybeans [14]. In the present study, the species Pratylenchus brachyurus, Helicotylenchus dihystera and Scutellonema cavenessi were the most important in terms of frequency and abundance. In contrast, species of the genus Heterodera were not observed at the Farako-Bâ site. These three genera belong to the obligate phytophagous group and pose a real threat to soybean cultivation in Burkina Faso [18]. Similar studies have reported the presence of these three genera in soybean crops in several regions of Africa [12]. Yield losses in soybean due to nematode parasitism are quite considerable and can reach up to 30% - 100% of production [19]. Among these parasitic nematodes associated in soybean, the specie Pratylenchus brachyurus is known to be one of the most damaging [13] [20] [21]. Similar studies on parasitic nematodes of rice in Burkina Faso have isolated species of the genera Meloidogyne, Heterodera, Scutellonema, and Pratylenchus in strict rainfed and lowland ecologies [22]. [23] reported the impact of soil parasitic nematodes Pratylenchus brachyurus on cowpea production despite the use of different levels of mineral fertilization to control its colonization in Burkina Faso. The presence of parasitic nematodes in soils and soybean roots poses a serious threat to soybean production in Burkina Faso and could compromise cropping systems in rotation with cereals such as sorghum [24].

# **5.** Conclusions

Soybeans, a recently introduced crop is gaining importance due to the high national demand for food and feed. The popularization of new soybean varieties is essential but this introduction requires the evaluation of new materials.

Our study which aimed at investigating the susceptibility of 24 soybean genotypes to nematodes identified seven (7) genera of nematode parasites on the crop in Burkina Faso. Among these genera, *Pratylenchus*, *Helicotylenchus* and *Scutellonema* can be considered the most important ones at the Farako-Bâ site. Considering the root populations of the genus *Pratylenchus* known to be highly pathogenic in soybean, group 2 genotypes were identified as the least susceptible to parasitic nematodes and can be used as sources of resistance in varietal control of these nematodes. Further studies will identify the different species of the different genera and better guide varietal control of these parasitic nematodes in soybean. This will allow a better assessment of the behavior of soybean genotypes against plant-parasitic nematodes.

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# **Conflicts of Interest**

The authors declare no conflicts of interest regarding the publication of this paper.

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