

## **Evaluation of soybean (*Glycinemax L. Merrill*) varieties for resistance to root-knot nematode (*Meloidogyne spp.*) under field conditions**

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### **ABSTRACT**

Field experiment was conducted to evaluate the resistance of fourteen varieties of soybean to *Meloidogyne spp.* infection at Umudike. Result obtained indicated that there were no significant differences on plant height, mean number of leaves per plant, on the varieties used. Only mean number of nodules (3.3) per plant, mean flowering dates (44 DAP) exhibited significant differences. Varieties TGX1789-7F, TGX1681-3F, TGX1440-1E, TGX1805-33F, NCRISOY-6 and NCRISOY-1 were moderately susceptible. Varieties TGX1485-1D, TGX1805-31F, TGX1843-6E and Cameroon late were observed to be resistant to the nematode infection. The resistant varieties recorded for nematode population (2433) at planting, nematode population (2566) at mid planting periods only. There was no significant difference on the grain yield. Grain weight for each treatments and variety showed TGX1019-2EN as having the lowest yield (0.56kg). Variety TGX1807-19F maintained a higher yield (1.42kg) and resistance to nematode attack, while varieties TGX1805-33F and NCRISOY-1 had high nematode populations and galls (3.3, 3.7) respectively. However, these varieties yielded the highest grain by weight than other varieties ie 0.94kg and 1.06kg respectively.

**Keywords:** Soybean, varieties, root-knot nematode, resistance, susceptibility.

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### **INTRODUCTION**

Soybean (*Glycinemax L. Merrill*) is one of the important crops selected for active production, research and utilization in Africa. It is a good source of protein for human and livestock throughout the world. *Meloidogyne spp.* (root-knot nematode) constitute a major problem wherever is grown in the world, more especially in Nigeria. Although nematicides are effective for control in terms of time of action, they are not always cost effective (Luzziet *al.*, 1995) and they are not environmentally friendly. Most rotation crops are also susceptible to *Meloidogyne spp.* Soybean farmers need to depend largely on resistant crops for sustainable yield and profitable cropping. Presently in pest and disease management, recent research efforts are focused on crop resistance, an ideal solution to environmental hazards caused by continuous and routine use of chemical pesticides (Afolami, 2000).

In Nigeria, nationally co-ordinated trials on soybean at IITA, and other commodity research institutes have released many elite varieties of soybean selected for good agronomic characters, early maturity, medium maturity and resistance to bacterial and fungal diseases. Some others are still being assessed as lines. The resistance of these improved varieties and lines to root-knot nematodes is gradually receiving attention (Iheukwumereet *al.*, 1995; Afolami and Atungwu, 1999). Most researchers screening or evaluating lines for resistance to *Meloidogyne spp.* rated the test crops based either on Taylor and Sasser's (1978) 0-5 gall index-scale or the standardized method of Sasseret *al.*, (1984). The use of gall-index alone as a measure of plant damage could at best be a preliminary indicator of plant resistance, but most studies on host resistance determination stop at this preliminary phase probably because it is fast or due to the large number of varieties and lines to be screened within a short period of

time (Afolami, 2000). Using grain yield as additional measure of plant damage, the work of Atungwu (1998) and Afolami (2000) showed some discrepancies over the status of varieties rated using the gall index alone. In this case therefore there is need to integrate gall index with the ultimate effect of *Meloidogyne* spp. on crop yield for a definite host resistance determination, based on the argument of previous researchers (Cook, 1974; Canto-Saenz, 1985). True resistance must show superior yield advantage on the varieties so labelled compared to the susceptible ones in the presence of the root-knot nematode (Canto-Saenz, 1983; Afolami, 2000). The objective of this study was to evaluate varieties for resistance to root-knot nematode *Meloidogyne* spp.

## MATERIALS AND METHODS

The experiment was laid out in a Randomized Complete Block Design (RCBD) made up of 12m by 48m plots in a naturally infested field. Before the main crop was planted ie soybean, two test crops were planted on the field to indicate the level of nematode in the soil ie Tomatoes (Roma variety) and Indian spinach. When the soils were sampled and analysed, an average of 250 juveniles of *Meloidogyne* spp. were recorded. Based on this, during planting of the main crop (soybean) about 250 juveniles per 250g of soil was already indicated on the soil (field). Two to three seeds of the fourteen varieties were sown per hole at the depth of 2.5cm to 4cm and 75cm by 5cm spacing. Treatments were replicated threetimes. Each plot was made up of four rows of 4m by 0.75m each.

Within the four ridges, two middle ridges (rows) were harvested for net yield determination. The two border rows covering the two middle rows that was used as net yield were used to determine the level of galls, nodulations, plant heights and 50% flowering, number of leaves per plant, nematode population in the soil at planting, mid planting and at harvesting.

Soil samples were collected at planting, mid planting and at harvest. This was done by random collection of 2kg soil samples from each plot at zigzag form using hand trowels. From each soil sample 200mls of soil was used for the nematode extraction.

For weed suppression, a mixture of grammaxon and galex was applied to the field immediately after planting at the rate of 5 litre/ha each. Two hand weeding operations were carried out before maturity and harvesting of the crop. The extraction of nematode from the soil for the trial was carried out at planting, mid planting and at harvest by random collection of up to 2kg soil sample at zigzag form from each plot having each treatment.

The extraction of nematode was done in the soil samples by use of pie-pan modification of the Baermann funnel method (Hooper, 1969). Each soil sample was mixed thoroughly and 200cm<sup>3</sup> of each of the soil samples was placed into a plastic sieve plate, which had its bottom covered with serviettes table tissues. The set-up was gently added through the tray. Enough water to soak the soil in the plastic plate was gently added through the tray. The set-up was then left undisturbed for 72 hours (3 days) before decanting the suspension into a beaker. The suspension was later decanted to 100 mls before it was used for counting of the nematode population in the soil with the aid of a microscope. A sample (1 ml) of each suspension was drawn with injection syringe after it has been properly mixed with magnetic stirrer into a petri dish and placed under a light microscope. A tally counter was used for the counting of the nematode whatever that was counted out was multiplied by 100ml of water used. This gave the population of the nematode in each treatment.

Data obtained from the trial was subjected to statistical analysis using Analysis of Variance (ANOVA) Procedure and Duncan Multiple Range Test (DRMT) was used for mean separation.

## RESULTS

Results of the effect of *Meloidogyne* spp. on mean plant height, mean number of leaves per plant, mean flowering dates (50%) is shown in Table i. The plant height was not affected by the nematode infection. There was no significant difference in the mean height of all the varieties tested. However, heights of varieties NCRISOY-1 and TGX 1807-19F were higher when compared with other varieties tested. The lowest height was recorded in variety TGX 1789-7F and this was not significantly different from others.

The data for the mean number of leaves per plant indicated no significant difference in the number of leaves per plant. Highest number of leaves was recorded for variety NCRISOY-1. Similarly, there were no significant differences indicated on the varieties on days of flowering (50% of flowering) for the trial. Some varieties TGX 1807-19F, Cameroon late and TGX 1440-1E, showed a significantly longer days to flower, while the lowest flowering date was recorded in varieties TGX 1485-1D, TGX 168-3F, TGX 1789-7F.

**Table i: Effect of *Meloidogyne* spp. on mean plant height (cm), mean number\* of leaves per plant, mean flowering dates (Days after sowing until 50% of plants flowered)**

Treatment	Mean Plant height (cm)	Mean Number of leaves per plant	Mean Flowering (Days after sowing until 50% of plants flowered)
TGX 1485-1D	42.67 <sup>a</sup>	12.7 <sup>a</sup>	34.0 <sup>c**</sup>
TGX 1799-8F	40.23 <sup>a</sup>	14.3 <sup>a</sup>	36.7 <sup>c**</sup>
TGX 1019-EN	48.40 <sup>a</sup>	11.0 <sup>a</sup>	40.7 <sup>ab**</sup>
TGX 1681-3F	44.27 <sup>a</sup>	15.0 <sup>a</sup>	34.7 <sup>c</sup>
TGX 1789-7F	36.43 <sup>a</sup>	14.7 <sup>a</sup>	33.0 <sup>c</sup>
NCRISOY-1	57.17 <sup>a</sup>	20.7 <sup>a</sup>	40.7 <sup>ab</sup>
TGX 1440-1E	43.20 <sup>a</sup>	15.3 <sup>a</sup>	43.3 <sup>a</sup>
TGX 1805-31F	41.37 <sup>a</sup>	16.3 <sup>a</sup>	41.3 <sup>ab</sup>
TGX 1807-19F	52.23 <sup>a</sup>	17.0 <sup>a</sup>	44.0 <sup>a</sup>
TGX 1805-33F	48.83 <sup>a</sup>	13.0 <sup>a</sup>	42.7 <sup>ab</sup>
NCRISOY-6	48.03 <sup>a</sup>	13.0 <sup>a</sup>	42.3 <sup>a</sup>
TGX 1843-6F	45.50 <sup>a</sup>	15.7 <sup>a</sup>	42.0 <sup>ab</sup>
TGX 1835-1E	47.73 <sup>a</sup>	15.0 <sup>a</sup>	42.3 <sup>a</sup>
Cameroon late	39.00 <sup>a</sup>	11.7 <sup>a</sup>	43.7 <sup>a</sup>

\* Values are mean of 3 replicates

\*\* Means in the same column followed by the same letter are not significantly different using Duncan's Multiple Range Test.

The result obtained from the effect of *Meloidogyne* spp. on mean number of nodules per plant and mean gall index scores per plant, is shown in Table ii. The nodules scores were significantly low. However the highest nodule scores were recorded in varieties TGX 1805-31F, while the lowest nodule scores were obtained from variety TGX 1835-1E. There was no nodule recorded in variety TGX 1835-1E.

The highest gall index were obtained in varieties TGX 1805-33F, TGX 1799-8F and NCRISOY-1 whereas, the lowest gall index was recorded in varieties, TGX 1485-1D, TGX 1019-2EN, TGX 1789-7F, TGX 1805-31F, NCRISOY-6, TGX 1843-6E, TGX 1835-1E and Cameroon late. The rest of the varieties recorded gall index scores that rated them moderately resistant. The mean number of root-knot nematode juveniles recovered from 200g soil samples and mean grain yield is shown in Table ii. Mean number of root-knot nematode juveniles recovered from 200g of soil is shown in Table ii. The result indicated significant difference between the varieties tested on the mean nematode population at planting and mid planting only. There was no significant difference on the mean nematode population at harvest.

There was no significant difference between the mean nematode population recorded in varieties NCRISOY-1 and TGX 1805-33F, but they differed significantly from the other varieties in the nematode population at planting. There was no significant differences observed in the mean nematode population of TGX 1019-2EN, TGX 1789-7F, TGX 1789-7F; TGX 1805-31F, TGX 1843-6E, NCRISOY-6 and Cameroon late at planting but they differed significantly from the remaining varieties in the nematode population recovered at planting. Treatment TGX 1485-1D, TGX 1799-3F, TGX 1681-3F and TGX 1843-6E, did not differed significantly in their mean nematode population at planting but had the lowest nematode population at planting when compared with other varieties.

In the mid planting there was no significant difference in nematode population in varieties NCRISOY-1 and TGX 1805-33F. But these two varieties differed significantly from the other varieties in the mean nematode population in the mid planting. However, TGX 1805-33F and NCRISOY-1 recorded the highest mean nematode population in the mid planting. There was no significant differences observed in the mean nematode population of varieties TGX 1485-1D, TGX 1799-8F, TGX 1789-7F, TGX 1807-19F and TGX 1835-1E at mid planting but they differed significantly from the remaining varieties in the nematode population recovered at mid planting. Varieties TGX 1019-2EN, TGX 1681-3F, TGX 1805-31F, did not differed significantly from the remaining varieties in the nematode population recorded at mid planting. The lowest nematode population is recorded in TGX 1843-6E and NCRISOY-6.

Generally, there was reduction in the mean nematode population at harvest. The highest mean number of nematode population was obtained at harvest in varieties NCRISOY-1, and this was however, not significant when compared with other varieties. However, varieties NCRISOY-1, TGX 1805-33F maintained the highest mean number of nematode population. The mean grain yield is shown on Table ii. Although there was no significant difference in the yield of the varieties, the highest grain was recorded by variety TGX 1807-19F. However, variety TGX 1805-33F had the highest grain by weight than other varieties, but this though high yielding, but non-resistant variety because of high numbers of nematode population and galls. Variety TGX 1019-2EN gave the lowest grain yield.

Table ii: Effect of *Meloidogyne* spp. on mean nodule scores per plant, mean gall index, mean number of nematode population recovered in 200g soil sample at planting, mid planting and at harvest and mean grains yield in (kg)

Treatment	Mean Nodule score(s)***	Mean Gall index score(s)	Nematode population at planting	Nematode population at mid planting (53 DAP)	Mean population at harvest (62 DAP)	Grain yield (kg)
TGX 1485-1D	2.3 <sup>c</sup>	1.0 <sup>c**</sup>	966.7 <sup>c**</sup>	1066.7 <sup>ab**</sup>	533.3 <sup>a</sup>	1.02 <sup>a</sup>
TGX 1799-8F	1.8 <sup>g**</sup>	3.7 <sup>c**</sup>	1133.3 <sup>c</sup>	1100.0 <sup>bc</sup>	600.0 <sup>b</sup>	0.73 <sup>a</sup>
TGX 1019-EN	1.8 <sup>g</sup>	1.0 <sup>c</sup>	1233.3 <sup>bc**</sup>	1000.0 <sup>c</sup>	500.0 <sup>a</sup>	0.56 <sup>a</sup>
TGX 1681-3F	2.8 <sup>c**</sup>	2.7 <sup>b**</sup>	1066.7 <sup>c</sup>	1000.0 <sup>c</sup>	466.7 <sup>a</sup>	0.75 <sup>a</sup>
TGX 1789-7F	2.0 <sup>f**</sup>	1.3 <sup>c</sup>	1100.0 <sup>bc</sup>	1033.3 <sup>bc**</sup>	700.0 <sup>a</sup>	0.75 <sup>a</sup>
NCRISOY-1	2.7 <sup>d**</sup>	3.7 <sup>a</sup>	2433.3 <sup>a**</sup>	2333.3 <sup>a</sup>	833.3 <sup>a</sup>	1.06 <sup>a</sup>
TGX 1440-1E	2.8 <sup>c</sup>	2.3 <sup>b</sup>	1600.0 <sup>b</sup>	1300.0 <sup>bb</sup>	500.0 <sup>a</sup>	0.57 <sup>a</sup>
TGX1805-31F	3.3 <sup>a</sup>	1.0 <sup>c</sup>	1333.3 <sup>bc</sup>	1000.0 <sup>c</sup>	566.7 <sup>a</sup>	0.67 <sup>a</sup>
TGX1807-19F	2.7 <sup>c</sup>	2.0 <sup>b</sup>	1400.0 <sup>bc</sup>	1100.0 <sup>b</sup>	733.3 <sup>a</sup>	1.42 <sup>a</sup>
TGX1805-33F	3.0 <sup>b</sup>	3.3 <sup>a</sup>	2400.0 <sup>a</sup>	2566.7 <sup>a</sup>	533.3 <sup>a</sup>	0.94 <sup>a</sup>
NCRISOY-6	2.0 <sup>f</sup>	1.0 <sup>c</sup>	1166.7 <sup>bc</sup>	900.0 <sup>cd</sup>	566.7 <sup>a</sup>	0.86 <sup>a</sup>
TGX 1843-6F	1.5 <sup>h</sup>	1.0 <sup>c</sup>	1033.3 <sup>c</sup>	866.7 <sup>cd</sup>	466.7 <sup>a</sup>	1.07 <sup>a</sup>
TGX 1835-1E	1.0 <sup>i</sup>	1.0 <sup>c</sup>	1100.0 <sup>bc</sup>	1066.7 <sup>bc</sup>	366.7 <sup>a</sup>	1.25 <sup>a</sup>
Cameroon late	2.5 <sup>a</sup>	1.0 <sup>c</sup>	1166.7 <sup>bc</sup>	1266.7 <sup>b</sup>	533.3 <sup>a</sup>	1.10 <sup>a</sup>

\* Values are mean of 3 replicates

\*\* Means in the same column followed by the same letter are not significantly different using Duncan's Multiple Range Test.

## \*\*\* Scale used for scoring nodules (1-5)

1	=	No nodule
2	=	Few nodules mostly small
3	=	More nodules some large
4	=	Many nodules several large
5	=	Abundant nodules many large

Source: Nationally co-ordinated trial logbook (2000)

## \*\*\* Scale use for gall index (0-5)

Number of gall or egg masses	Gall or egg mass index	Resistance rating
0	0	Immune (I)
1-2	1	Resistance (R)
3-10	2	Moderately Resistant (MR)
11-30	3	Moderately Susceptible (MS)
31-100	4	Susceptible (S)
100+	5	Highly Susceptible (HS)

Taylor and Sasser (1978) method

## DISCUSSION

The result of this investigation has shown that *Meloidogyne* spp. affected the parameters of considered on the trials (soybean).

There was no significant difference recorded on the mean heights of the trial (naturally infected by nematode) and this agreed with the reports by Malek and Jenkins (1964) who observed that growth of plants are not retarded to any appreciable extent until the number of nematodes reaches a very high level. This could be the possible reason the height of soybean were not affected much by nematode infection. There was a significant difference in the mean nodule scores. However, the nodule scores of some varieties did not differ significantly from each other. This observation agreed with the findings of Taha and Raski (1969) who reported that *Meloidogyne* spp. are not known to severely inhibit nitrogen fixation, regardless of their tendency to invade Rhizobium nodules. Furthermore, Robinson (1961) revealed that nodules attacked by nematodes during early stages of development become galls but when attacked at a later stage, remain nodules. Varieties TGX 1807-19F, TGX 1805-33F recorded higher nodule score. The higher the inoculum level, the higher the number of galls and root-knot nematodes. However, the highest mean gall indices observed were on varieties TGX 1805-33F, TGX 1799-8F and NCRISOY-1. Using Taylor and Sasser (1978) gall rating index, the above mentioned varieties are classified as moderately susceptible (MS). The low galling indices observed on most varieties could be due to local inoculum level or that most of the varieties were resistant to *Meloidogyne* spp. Gall formation hinders transportation of water and minerals and nutrients from roots to the aerial parts (Dropkin and Nelson, 1960).

Significant differences were recorded on mean nematode population at planting and mid planting. There was no significant difference on the mean nematode population at harvest. However, varieties NCRISOY-1, TGX 1805-33F maintained the highest mean number of nematode population in the trial.

Yield data obtained showed that significant variety, TGX 1805-19F had a higher grain yield by weight. However, variety TGX 1805-33F had the highest grain by weight than other varieties but this variety though high yielding, but non-resistant variety because of high numbers of nematode population and gall index attached to it. The treatment TGX 1805-19F that had relatively low nematode populations and gall formations could be considered as the highest yielding variety. TGX 1019-2EN, yielded the lowest grain by weight and was susceptible to nematodes attack. The experiment confirms the failure rate of TGX 1019-2EN in the previous work of Afolami and Atungwu (2000).

To achieve control of root-knot nematode, a resistant cultivar must prevent a large proportion of reproduction, usually 90% or more as compared to susceptible cultivars of the same species (Taylor and Sasser, 1978). In this regard, the varieties used in this study could be classified into immune or resistant, moderate and susceptible varieties to root-knot nematode.

### CONCLUSION

The study has shown that root-knot nematode, *Meloidogyne* spp. has some pathological influence on growth and development of plant. The results obtained suggest that fourteen different cultivars of soybean reacted differently to the same population of nematode. These differences were significant (at 5% level) in some cases. Six soybean cultivars (TGX 1835-1E, TGX 1843-6E, Cameroon late, TGX 1805-31F, TGX 1485-1D and TGX 1807-19F), showed some resistance to the nematode attack, while TGX 1681-3F, TGX 1789-7F, TGX 1440-1E, were moderately resistant and the rest, TGX 1799-8F, TGX 1019-2EN, NCRISOY-1, NCRISOY-6 and TGX 1805-33F were moderately susceptible to nematode attack (*Meloidogyne* spp.) apart from root galling, the moderately susceptible cultivars showed reduction in their vegetative growth as well as discoloration or chlorosis of leaves. Their vigour was also impaired in varying amounts due to the nematode attack. Based on the results obtained it is important that soybean farmers need to depend largely on crop resistance for sustainable yield and profitable cropping. Although nematicides are difficult to get even when you get them the quantities are too small if they are to be used in large area of land. The findings of this study are not conclusive further research works are needed in the varieties (TGX 1835-1E, TGX 1843-6E, Cameroon late, TGX 1805-31F, TGX 1485-1D and TGX 1807-19F) that showed resistance to *Meloidogyne* spp. to be tried on other ecological zones that are endemic with *Meloidogyne* spp. attack.

This will help to prove their further resistance to nematode attack. After this the varieties could be released for use by the farmers in the area infested with *Meloidogyne* spp. The high yielding non-resistant varieties (TGX 1805-33F, NCRISOY-1) should be subjected to selective bio-pesticide treatment. The susceptible varieties TGX 1799-8F, NCRISOY-1, NCRISOY-6, TGX 1019-2EN and TGX 1805-33F recorded in this study, especially TGX 1019-2EN which has been confirmed in other areas should not be included in areas with *Meloidogyne* infestation.

The varieties should also be subjected to different levels of nematode population (*Meloidogyne* spp.) to ascertain their reactions to them. For accurate reporting of data in nematology, galling should not be used alone as a measure of susceptibility.

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