TARC Report

Nematological Survey on Winged Bean Fields in Okinawa with Special Reference to Varietal Difference in Susceptibility to Root-knot Nematodes

By RINKO TERUYA,* JIRO ABE,** MASAAKI NAGAMINE*** and HIROSHI NAKAMURA****

* Okinawa Prefectural Agricultural Experiment Station
(Shuri, Naha, Okinawa, 903 Japan)
, *, **** Okinawa Branch, Tropical Agriculture Research Center
(Ishigaki, Okinawa, 907-01 Japan)

Introduction

Winged bean (*Psophocarpus tetragonolobus*) is cultivated not only as an edible crop with high protein quality but also as a green manure crop because of its great nitrogen-fixing capability and vigorous vegetative growth in summer season.[1] It was reported, however, that root-knot nematodes cause damage to winged bean throughout the tropics.[2] The purpose of this survey is to know population levels of plant-parasitic nematode species and their vertical distribution in the rhizosphere of winged bean growing in fields, and also to examine varietal differences of winged bean in susceptibility to root-knot nematodes. The survey constitutes a part of the TARC's research program entitled "Winged bean introduction", and was conducted with the cooperation of the nematologist of the Okinawa Prefectural Agricultural Experiment Station.

Present address:
** Tohoku National Agricultural Experiment Station (Morioka, Iwate, 020-01 Japan)
*** Okinawa Prefectural Agricultural Experiment Station (Naha, Okinawa, 903 Japan)
**** National Agriculture Research Center (Yatabe, Ibaraki, 305 Japan)

Materials and methods

Two different fields of winged bean were used to examine (1) varietal differences in susceptibility to root-knot nematodes, and (2) vertical distribution of nematode fauna in the rhizosphere of winged bean.

(1) Examination of varietal differences

Seeds of 46 winged bean varieties including a variety (Tpt-7) of the related species, *P. palustris*, were sown into peat pots in a green house on May 12, 1982. The plants were transplanted on June 2 to the field, which had been a nursery of ornamental trees such as Chinese roses and crotons, in Ishigaki Island, 24°N lat. For each variety, 4 plants were transplanted at 50 cm distance between rows and 40 cm distance between plants on rows. Neither fertilizer nor chemical to control weeds, diseases and pests was applied. On November 5, after harvest, soil samples were taken from the soil layer, 5–15 cm in depth, located 15 cm apart from the plant stubbles, with two replication

for each variety.

(2) Examination of vertical distribution

Three varieties, i.e., "UPS-31", an early flowering variety introduced from Papua New Guinea, "Tpt-2", a medium flowering variety from Nigeria and "Colombia", a late flowering variety from Colombia, were sown with a 100 × 50 cm spacing on June 3, 1982 in an other field where sweet corn had been planted. Neither fertilizer nor chemical was applied. Soil samples were taken on November 4 from successive soil layers of 5 cm in thickness, starting from 0 cm down to 35 cm of soil depth at a site 15 cm apart from a plant of each variety by using a soil sampler, 5 cm in diameter and 5 cm long.

(3) Separation and counting of nematodes

The soil samples in plastic bags were stored in a refrigerator at 2–3°C until examination. Nematode separation from soil was made by the double layer centrifugal-floatation method.[3] When 20–25 m*l* hypo (sodium thiosulfate) solution with specific gravity of 1.2 was applied to the bottom of a 50 m*l* centrifuge tube, containing suspension of 10 g of soil in 10–20 m*l* water, two distinct layers appeared, i.e., soil suspension in the upper layer and hypo solution in the lower layer. After centrifugation for 7–10 min at 2800–3000 r.p.m., the nematode suspension between two layers was transferred to a small quantity of tap water after successive decantation of supernatant fluids to concentrate nematode suspension. Then nematodes were killed by 50–60°C hot water treatment for 3–5 min and fixed with the same amount of TAF fixative. Counting was made under a microscope of 150 magnification.

Results

1) Nematode species and their population levels in the winged bean fields

Eight genera of plant parasitic nematodes were found from 113 soil samples obtained from both the varietal difference examination field (5–15 cm deep) and the vertical distribution examination field (0–35 cm); seven genera from the former and five genera from the latter (Table 1–3). Four genera common to the both fields were *Meloidogyne* spp. (root-knot nematodes), *M. javanica* and *M. incognita*, *Helicotylenchus dihystera* (spiral nematode), *Pratylenchus zeae* (root-lesion nematode) and *Paratylenchus* spp. (pin nematodes). Three genera such as *Hoplolaimus* (lance nematodes), *Tylenchorhynchus nudus* (stylet nematode) and *Trichodorus* (stubby-root nematodes) were found only in the former field, and *Criconemoides* (ring nematodes) in deep soil beyond 30 cm of depth in the latter field.

The root-knot nematodes were found from all soil samples from both fields, except Tpt-9 plot in the former field, although a great diversity in population levels was observed. The spiral nematodes were also so frequent that they were detected in all soil samples from the latter field and in 24 out of 46 winged bean varieties, although their population levels in the surface soil layers of top 15 cm were as low as less than five per 10 g of soil with an exception of two varieties. The nematode populations of other genera were also very low.

2) Vertical distribution of nematodes in the rhizosphere of winged bean

The root-knot nematode larvae were found throughout the soil layer of 0–35 cm in depth. However, most of them were distributed in the upper half of the soil layer. The distribution pattern of the nematodes seems to be influenced by different plant growth and root distribution of different varieties of winged bean, i.e., in the rhizosphere of the earlier flowering variety more number of the nematode was distributed in deeper soil. The amount of root-knot nematode larvae in the soil below 10 cm of depth was 63, 50, and 38%, for UPS-31 (early variety), Tpt-2 (medium early variety) and Colombia (late variety), respectively (Table 2 and Fig. 1).

The spiral nematode populations tended to distribute in comparatively deep soil beyond 15 cm. Although population levels of other genera were very low, root-lesion nematode occurred in comparatively upper soil, and ring

Table 1. Population levels of plant parasitic nematodes other than root-knot nematodes in the varietal difference examination field

Nematode species	Winged bean variety	No. of nematodes in 10 g of soil
Helicotylenchus dihystera		
	UPS-31	11
	BBG	6
	Tpt-31	4
	UPS-122	4
	LBNC-1	3
	M 13-1	3
	MHS-14	3
	Ishigaki-2	3
	Tpt-17	2
	Tpt-26	2
	UPS-132	2
	902-1	2
	6035-A	2
	Florida	2
	Tpt-1	1
	Tpt-6	1
	Tpt-8	1
	Tpt-14	1
	Tpt-16	1
	Tpt-32	1
	Tpt-33	1
	Tpt-48	1
	IN-1	1
	UGM-1	1
Pratylenchus zeae		
	UPS-122	4
	Tpt-30	2
	BBG	2
	Ishigaki-1	2
	Ishigaki-3	2
	Tpt-1	1
	Tpt-6	1
	Tpt-16	1
	Tpt-17	1
	Tpt-31	1
	UPS-31	1
	902-1	1
	M 13-1	1
Paratylenchus spp.		
	UPS-31	1
	UPS-132	1
	902-1	1
	1126-A	1
	Florida	1
Hoplolaimus spp.		
	Tpt-14	1
	UPS-31	1
	902-1	1
Tylenchorhynchus nudus		
	BBG	2
	MHS-14	1
Trichodorus spp.		
	M 13-1	1

nematodes in deep soil.

3) *Susceptibility of the winged beans to the root-knot nematodes*

Population density of root-knot nematode larvae in the rhizosphere of winged bean is regarded as reflecting nematode reproduction as well as host plant susceptibility to the nematodes in the field. Varietal differences in the susceptibility can also be evaluated by successive examinations, throughout a growing season, of the degree of gall formation by each of *Meloidogyne* species. The larval population density showed a wide variation, suggesting varietal differences in the susceptibility to the root-knot nematodes; i.e., 16 varieties with a high population density (100 larvae/10 g of soil), 10 varieties with a moderate population density (10 larvae), and 20 varieties with a low population density (1 larva) as shown in Table 3. Varieties introduced from Indonesia showed a comparatively low larval population density, and varieties from Papua New Guinea and Africa showed a wide variation in the population.

Discussion

One of the characteristics of the nematode fauna in the rhizosphere of winged bean fields, as compared with that of other crops in this region, appears that not too many genera of parasitic nematodes are found; i.e., out of 21 genera found by one of the present authors in Okinawa region,[6] 17 were detected in sugar cane fields, 12 pineapple fields and 8 in winged bean fields. In particular, *Paratrophorus* or *Rotylenchus*, the most prevailing genera, were not detected in this survey. Further investigation will be needed to assure

Table 2. Vertical distribution of plant parasitic nematodes (number of nematodes per 10 g of soil) in the rhizosphere of three varieties of winged bean

Soil depth	*Meloidogyne* spp.	*Helicotylenchus dihystera*	*Pratylenchus zeae*	*Criconemoides* spp.	*Paratylenchus* spp.
		Variety : UPS-31			
0- 5 cm	144	1			
5-10	537	3	1		
10-15	735	2			
15-20	387	9			1
20-25	21	20			
25-30	3	6			
30-35	2	11			
		Variety : Tpt-2			
0- 5	21	3	1		
5-10	106	2	1		
10-15	56	1			
15-20	40	12	1		
20-25	30	10			
25-30	5	16			
30-35	3	6		5	
		Variety : Colombia			
0- 5	97	3			
5-10	104	2			
10-15	65	2			
15-20	6	3			
20-25	3	7			
25-30	3	4			
30-35	2	4		1	

whether this fact is specific to winged bean or resulted from the preceding crops.

Of these eight genera, *Meloidogyne* appeared most important to cause severe damage to winged bean due to its high reproduction capacity on susceptible varieties as shown by a high population density of more than 500 larvae per 10 g of soil. Spiral nematodes occur with a high frequency, but population density is too low to cause practical injury to winged bean.

It was observed that larvae of *Meloidogyne incognita* occurring in fields of tomato and bitter gourd, both of which are susceptible, continued to increase in number from May to November when air temperature fluctuated between 20°C and 32°C.[5] This observation agreed with the report that optimum hatch of *M. incognita* and *M. javanica* occurred between 25°C and 30°C and between 20°C and 30°C, respectively.[2] The beginning of November, when the soil sampling was made in this survey, may be suitable to estimate the distribution and population levels of root-knot nematode larvae in soil, because growth of winged bean was proved to synchronize with the root-knot nematode reproduction.

A high degree of variability in the resistance to the root-knot nematodes was observed among winged bean varieties tested. This suggests the possibility of selecting resistant varieties which will be suitable for a cover crop to be grown in rotation with susceptible crops in an attempt to decrease nematode population levels as well as to enhance soil fertility.

Summary

Vertical distribution of parasitic nematodes in the rhizosphere (0–35 cm in soil depth) of winged bean and differences in susceptibility

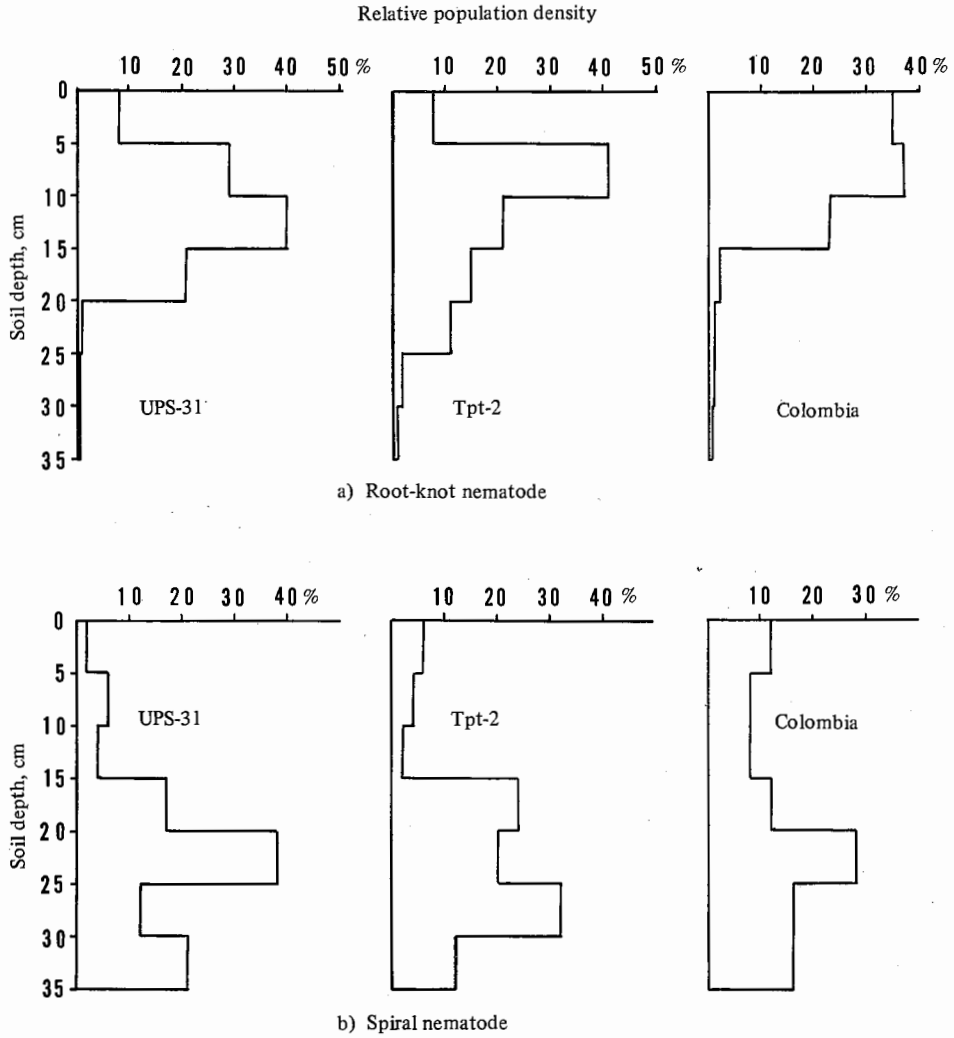

Fig. 1. Vertical distribution of root-knot nematodes and spiral nematodes in the winged bean field

to root-knot nematodes among 46 winged bean varieties were surveyed in subtropical Ishigaki Island, Okinawa.

Eight nematode genera were found, and root-knot nematodes (*Meloidogyne incognita* and *M. javanica*) were most predominant. Spiral nematodes also occurred frequently, but their population levels were very low.

Vertical distribution of root-knot nematode larvae indicated a tendency that the upper soil layer contained more number of the larvae than the lower layer, although the distribution was slightly influenced by root distribution of different varieties of winged bean. It was apparent that the spiral nematode occurred in comparatively deep soil below 15 cm of depth.

A high degree of variability in the population density of root-knot nematode larvae in the rhizosphere was observed among winged bean varieties tested, i.e., 16 varieties showed a high population density, 10 varieties a moderate density, and 20 varieties a low

Table 3. Number of larvae of *Meloidogyne incognita* and *M. javanica* in the rhizosphere of the winged bean varieties tested

Variety	Origin	No. of nematodes per 10 g of soil
Variety with high population level		
UPS-132	P. N. G.	847
Tpt-11	Nigeria	754
902-1	Indonesia	446
UPS-31	P. N. G.	340
Tpt-18	Nigeria	320
Florida	U. S. A.	303
Tpt-32	Nigeria	301
Tpt-7	Nigeria	287
Ishigaki-3	Japan	276
Tpt-26	Nigeria	272
Tpt-6	Nigeria	259
Tpt-33	Nigeria	196
Tpt-3	Nigeria	191
Tpt-15	Nigeria	177
MHS-14	Thailand	114
Tpt-22	Nigeria	108
Variety with intermediate population level		
Colombia	Colombia	84
Tpt-2	Nigeria	82
Tpt-17	Nigeria	72
Tpt-8	Nigeria	62
Tpt-12	Nigeria	56
UPS-122	P. N. G.	30
Tpt-4	Nigeria	24
Tpt-29	Nigeria	24
1126-A	Indonesia	17
Ohama	Japan	14
Variety with low population level		
LBNC-1	India	9
Tpt-14	Nigeria	7
UPS-45	P. N. G.	7
Ishigaki-2	Japan	7
IN-1	Indonesia	7
M 13-1	Malaysia	7
Tpt-30	Nigeria	6
Tpt-48	Nigeria	5
6035-A	—	5
Tpt-16	Nigeria	4
Tpt-10	Nigeria	3
Tpt-31	Nigeria	3
BBG	Indonesia	2
1126-B	Indonesia	2
Tpt-1	Nigeria	1
Tpt-19	Nigeria	1
Ishigaki-1	Japan	1
Pyimana	Burma	1
UGM-1	Indonesia	1
Tpt-9	Nigeria	0

density less than 10 larvae per 10 g of soil.

Acknowledgement

We wish to thank Dr. M. Ichinohe, Hokkaido National Agricultural Experiment Station, for his valuable comments on the manuscript. Thanks are also due to Dr. T. N. Khan, Department of Agriculture, Western Australia, and to Dr. N. Q. Hg, IITA, Nigeria, for their courtesy to supply winged bean seeds.

References

1) Masefield, G. B.: *Psophocarpus tetragonolobus*—A crop with a future? *Field Crops Abs.*, **26**, 157–160 (1973).
2) Price, T. V. & Linge, D. S.: Studies on the root-knot nematodes of winged bean (*Psophocarpus tetragonolobus*) in Papua New Guinea. *Trop. Agric.* (Trinidad), **56**, 345–352 (1979).
3) Takagi, Y.: Double layers centrifugal-floatation method for the separation of nematodes. *Jpn. J. Appl. Ent. Zool.*, **14**, 108–110 (1970) [In Japanese with English summary].
4) Teruya, R.: Host range of a plant-parasitic nematode species, *Meloidogyne incognita*, in Okinawa. *Okinawa Nogyo*, **5**, 52–57 (1966) [In Japanese with English summary].
5) Teruya, R.: Studies on the control of the plant parasitic nematodes. II. On the seasonal fluctuation of the population density of *Meloidogyne incognita* larvae and plant injury in the vegetable field. *Bull. Ryukyu Agr. Exp. Sta.*, **5**, 23–28 (1969) [In Japanese with English summary].
6) Teruya, R.: Plant-parasitic nematodes in Okinawa. *Shokubutsu Boeki*, **25**, 458–460 (1971) [In Japanese].

(Received for publication, May 23, 1983)