Influence of 1,2 dibromo-3-chloropropane fumigation on nematode population, mycorrhizal infection, N₂ fixation and yield of field-grown groundnut

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SUMMARY

DBCP fumigation of a typical sandy soil of Central Senegal (Dior soil) on which groundnut is commonly cultivated significantly reduced the size of *Scatellonema cavenessi* populations, favoured early infection by endomycorrhizae in two of the three groundnut cultivars studied, increased nodulation and nitrogen fixation (measured by the acetylene essay), and dramatically increased crop yields. From these results, it is concluded that *S. cavenessi* does alter the physiology of the groundnut plant, by affecting the establishment and functioning of the symbioses between the plant and both endomycorrhizae and *Rhizobium*. Thus, parasitism by this nematode reduces the take-up of phosphorus by the plant as well as its ability to fix nitrogen.

RÉSUMÉ

Influence d'un traitement nématicide avec le 1,2 dibromo-3-chloropropane sur les populations de nématodes, l'infection mycorhizienne, la fixation de N_2 et les rendements de l'arachide

Le traitement nématicide d'un sol Dior typique du Centre Sénégal cultivé en arachide au moyen du DBCP réduit significativement les populations de *Scutellonema cavenessi*; améliore, chez deux sur trois des cultivars utilisés, l'infection endomycorhizienne précoce; accroît la fixation de l'azote mesurée par la méthode de l'acétylène; accroît de façon spectaculaire les rendements de l'arachide. On déduit de ces résultats qu'en altérant la physiologie de l'arachide, *S. cavenessi* handicaperait l'établissement et le fonctionnement de la double symbiose qui s'établit entre la plante, les endomycorhizes et les *Rhizobium*, réduisant ainsi d'une part l'absorption du phosphore et d'autre part son aptitude à fixer l'azote atmosphérique.

In semi-arid conditions which prevail in West Africa, crop yields on sandy soils are limited not only by water but also nematode attacks. Not only do nematodes reduce crop yields by direct effect but also by interactions with other organisms. Nematodes may decrease endomycorrhizal infection of roots (Fox & Spasoff, 1972; Schenck & Kinloch 1974, Schenck, Kinloch & Dickson, 1975) and, in the case of legumes, reduce nodulation (Hussey & Barker, 1976; Yeates *et al.*, 1977) and, consequently, N₂ fixation (Baldwin, Barker & Nelson, 1975; Germani, 1979). Thus, it is not surprising that fumigation of nematode-infested soils by some nematicides as DBCP (1,2 dibromo-3-chloropropane) or 1,3

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dichloropropene resulted in a significant increase of endomycorrhizal infection of cotton roots (Bird, Rich & Glover, 1974).

The aim of the present work was to evaluate the influence of fumigation of a nematodeinfested soil with DBCP upon (i) endomycorrhizal infection of roots, (ii) N₂ fixation, (iii) crop yields of three different cultivars of field-grown groundnut in order to determine whether soil fumigation can kill parasitic nematodes and also improve nitrogen and phosphorus nutrition of the plant by restoring symbiosis with endomycorrhizal fungi and *Rhizobium* spp.

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Material and methods

The experimental area was located near Patar, 25 km South of the « Centre de Recherche Agronomique de Bambey » (Agronomic Research Center at Bambey), Central Senegal.

The soil was a typical sandy soil of Central Senegal (vernacular name : Dior) characterized by a very low cation exchange capacity (Charreau & Nicou, 1971) and heavily infested with *Scutellonema cavenessi* Sher, 1963. The experimental site had been previously cultivated with pearl-millet (*Pennisetum typhoides* Burns, Stapf & Hubb.) according to the traditional pearl-millet/groundnut rotation system in use in the region (Charreau, 1974).

Fertilizer (6:20:10, N: P_2O_5 : K_2O mixture) was applied on the experimental area at the rate of 150 kg per ha.

The experimental design consisted of nine randomized blocks, with two treatments : fumigated with DBCP in 1977 (14 days before planting) and non fumigated. Treated plots were fumigated, at a depth of 20 cm, 14 days before sowing at a rate or 25 1/ha of commercial nematicide, Nemagon (75 % 1,2 dibromo-3-chloropropane).

Groundnut cultivars 28-206 (growth-cycle: 120 days), 55-437 (growth cycle: 90 days), and GH 119-20 (growth cycle: 120 days), obtained from the Institut Sénégalais de Recherche Agronomique, were planted at 15 cm intervals in rows 60 cm apart, on July 23, 1977. Each plot was composed of three rows of groundnut, one of each cultivar.

During the groundnut growth cycle rainfall was low (287 mm during June-October), but well distributed. Root systems and soil were sampled twice (16th and 60th day) in order to estimate endomycorrhizal infection, N_2 (C_2H_2) fixation and nematode numbers. Only five rather than nine replicates were used for each treatment.

The crop was harvested on October 20th 1977 (cv. 55-437) and November 10th 1977 (cv. 28-206 and GH 119-20). Crop yield estimations were based on the plant harvest of each of the nine replicates.

Interpretation of data was performed using Mann-Whitney test, (Snedecor & Cochran, 1967) for acetylene reducing activity per plant (ARAP) and mycorrhizal infection percentage; classical parametric « t » test for other data.

ESTIMATION OF ENDOMYCORRHIZAL INFECTION

Five root systems per plot were examined. Roots were stored in formalin-acetic acidethanol (¹) and then treated and stained according to the method of Phillips and Hayman (1970). A microscopic examination was made under 250 magnification on the total length of each 1 cm root segment. For each root system, 12-15 segments were observed. The extent of mycorrhizal infection was expressed as percentage of microscope fields with endomycorrhizal infected roots.

Estimation of N_2 (C_2H_2) fixation

The basic procedure for the measurement of N₂ fixation was that of Hardy et al., (1968), with some modifications. Immediately after harvest, root systems were placed in 570 ml serum bottles with a rubber stopper. Through the stopper, 50 ml acetylene and 0.4 ml propane (tracer gas) were injected. Bottles were incubated for 30 mn at ambiant temperature (c. 28°). The incubation atmosphere was sampled using a 10 ml Vacutainer tube. The ethylene assay was performed using a hydrogen flame ionization chromatograph (Varian aerograph 1200). The stainless steel column 150×0.3 cm was packed with Spherosyl X OB 075 80-100 mesh + 10 % Na₃PO₄. The injector, column and detector temperatures were respectively 105, 40 and 180°, N₂ flow 30 ml per minute, H₂ flow 30 ml per minute, compressed air flow- 300 ml per minute. ARAP was expressed as nanomoles C₂H₂ reduced per plant per hour.

CROP YIELDS

Aerial parts and pods were harvested separately and their weights determined after drying at 70-80°. The nitrogen content of plant tissues was determined by Kjeldahl analysis. Phospho-

⁽¹⁾ Formalin (13 ml), acetic acid (5 ml), 50 % ethanol (200 ml).

rus analysis was performed using the molybdophosphoric blue color method after wet oxidation of plant tissues by means of the HNO_3 - $HCIO_4$ acid mixture.

ENUMERATION OF NEMATODES

Nematodes were extracted from fresh soil samples by elutriation (Seinhorst, 1962) and from roots by mist chambers (Seinhorst, 1950). Results were expressed as numbers of *Scutellonema cavenessi* per dm³ of soil or 100 g of roots (fresh weight).

Results and discussion

EFFECT OF NEMATICIDE UPON NEMATODE POPULATIONS

DBCP fumigation significantly reduced Scutellonema cavenessi populations (Tab. 2). The effect of the treatment was dramatic when plots were fumigated in the same year as the crop was grown.

Table 1

Effect of DBCP fumigation on mycorrhizal infection of field-grown groundnut expressed as percentage of infected roots.

	Groundnut cultivars							
<i>,</i>	cv. 55-437	cv. 28-206	cv. GH 119-20					
First sampling								
(16th day) No fumigation Fumigation	28.4 a (a) 33.0 ab (a)	22.8 a (a) 59.6 b (b)	53.8 a (b) 76.5 a (b)					
Second samplir	ng		•					
(60th day) No fumigation Fumigation	54.0 bc (a) 56.2 c (a)	63.6 b (a) 40.7 ab (a)	57.0 a (a) 57.4 a (a)					

Numbers in columns not having same letter and numbers in rows not having same letter between brackets differ P = .05 by Mann-Whitney test (Snedecor & Cochran, 1967).

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EFFECT OF NEMATICIDE UPON ENDOMYCOR-RHIZAL INFECTION

Mycorrhizal infection, as measured at the first sampling (16th day), was significantly higher in fumigated plots for two cultivars (28-206 and GH 119-20).

Data in Table 1 suggest differences between the three cultivars since endomycorrhizal infection was lower in cv. 55-437 than other cultivars.

Since groundnut (cv. 28-206 and GH 119-20) roots were heavily infected in the fumigated plots as early as the 16th day, endomycorrhizal infection of some cultivars appeared to be an early process in the edaphic and climatic conditions of Senegal, provided that there is no limiting factor, such as nematode injury.

No significant difference between treated and untreated plots was found at late sampling (60th day). Thus, DBCP treatment stimulated an early endomycorrhizal infection.

These results support the conclusion of Bird, Rich and Glover (1974) that nematodes might eventually limit mycorrhizal infection, but the related mechanism is still unknown.

Microscopical observations showed that only very few vesicles and arbuscules were found in roots of groundnut as reported by Ross and Harper (1973).

Effect of DBCP upon $N_{\,2} \ \left(C_{\,2} H_{\,2} \right)$ fixation

Table 1, shows that no significant effect on N_2 fixation could be detected on either sampling date.

However, time course curves of acetylene reducing activity per plant (ARAP) (Germani, unpubl.) indicate that ARAP of cv. 55-437 and GH 119-20 fumigated plots was markedly higher than that occurring in non-fumigated plots when plants were 40-50 days old, but that such a difference was not detected up when plants were either 16 or 60 days old.

Thus, DBCP fumigation appeared to improve ARAP of cultivars 55-437 and GH 119-20 but not that of cv. 28-206. It is interesting to note that whereas ARAP of cv. 28-206 was not improved by fumigation, its endomycorrhizal infection was greatly enhanced by this treatment, at least during the period ranging from day 40 to 50 when ARAP of groundnut is

TABLE 2

Effect of soil fumigation with DBCP on nematode population, N_2 fixation, endomycorrhizal root infection and yields of field-grown groundnut (cv. 55-437; 28-206; GH 119-20).

	cv. 55-437		cv. 28-206		cv. GH 119-20	
· ·	DBCP	Control	DBCP	Control	DBCP	Control
Early sampling (16th day)						
Number of nematodes (¹) Number of nematodes (²) A R A P (³) % mycorrhizal roots	0 (●●●) 0 (●●●) 9800 (N.S.) 33.0 (N.S.)	$848 \\ 688 \\ 6228 \\ 28.4$	0 (●●●) 0 (●●●) 12.007 (N.S.) 59.6 (●)	$\begin{array}{c} 828 \\ 1450 \\ 4362 \\ 22.8 \end{array}$	0 (●●●) 0 (●●●) 12.082 (N.S.) 76.05 (●)	$968 \\ 1760 \\ 4908 \\ 53.8$
Late sampling (60th day)						
Number of nematodes (¹) Number of nematodes (²) A R A P(³) % mycorrhizal roots	16 (●●●) 0 (●●●) 7176 (N.S.) 56.2 (N.S.)	$\begin{array}{r} 248\\ 33362\\ 3941\\ 54.0 \end{array}$	80 (●●●) 4 (●●●) 5498 (N.S.) 40.7 (N.S.)	3892 15688 9706 63.6	4 (000) 0 (000) 6665 (N.S.) 57.0 (N.S.)	$2704 \\ 26058 \\ 5981 \\ 57.4$
Crop yield (kg per ha)						
Pods (d.w.) Aerial parts (d.w.) Seed N Seed P	$\begin{array}{c} 1197 (\bullet \bullet \bullet) \\ 3279 (\bullet \bullet \bullet) \\ 30.20 (\bullet \bullet \bullet) \\ 3.02 (\bullet \bullet \bullet) \end{array}$	$\begin{array}{c} 497 \\ 866 \\ 13.30 \\ 1.29 \end{array}$	$\begin{array}{c} 1893 (\bullet \bullet \bullet) \\ 3071 (\bullet \bullet \bullet) \\ 44.50 (\bullet \bullet \bullet) \\ 3.56 (\bullet \bullet \bullet) \end{array}$	$793 \\ 1485 \\ 18.90 \\ 1.79$	$\begin{array}{c} 1707 (\bullet \bullet \bullet) \\ 3975 (\bullet \bullet \bullet) \\ 45.90 (\bullet \bullet \bullet) \\ 4.59 (\bullet \bullet \bullet) \end{array}$	746 1829 19.70 1.86

(1) numbers per dm³ soil

(2) numbers per 100 g (d.w.) roots

(3) ARAP : Acetylene reducing activity expressed as micromoles $C_{a}H_{2}$ reduced per plant per h.

: Significantly different (P = .001)

: Significantly different (P = .05) N.S. : Not significantly different $(P = \ge .10)$

known to be the highest (Germani, unpubl.; Ducerf, pers. comm.).

EFFECT OF NEMATICIDE UPON CROP YIELDS

DBCP increased crop yields expressed as dry weight basis (Tab. 2) confirming previous experimental results (Germani, 1979).

In conclusion, Scutellonema cavenessi, presumably impedes the establishment and functioning of the double plant symbiosis (Mosse, Powell & Hayman, 1976; Daft & El-Giahmi, 1976) (i) with endomycorrhizae (ii) with *Rhizobium*, thus indirectly reducing plant phosphorus uptake, and possibly its water uptake (Safir, Boyer & Gerdemann, 1971), and also, its N_2 fixing ability. Such consequences are presumably most harmful to the plant especially in phosphorus and nitrogen deficient soils which commonly occur in semiarid West Africa.

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