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On the field anhydrobiotic ability of *Pratylenchus thornei* and *Merlinius brevidens*

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Summary – This study on the anhydrobiotic behaviour of *Pratylenchus thornei* and *Merlinius brevidens* from dry soils has been done on a non-irrigated field under wheat cv. Gallareta, estimated here as a bad/non-host of these nematodes. *P. thornei* is shown to be more dependent than *M. brevidens* on the soil humidity for its recovery in 15 hours of migration (rehydration, reactivation and migration). *M. brevidens* emerges more easily from anhydrobiosis. The patterns of emergence are similar for both nematodes whether they are active or in state of recovery from deep anhydrobiosis. Their patterns are different when they recover from an early stage of anhydrobiosis (mild anhydrobiosis). The two nematodes have an inertia with soil depth to enter into anhydrobiosis, which seems to depend on the natural process of desiccation of the field soil. The inertia seems to be greater in *M. brevidens*.

Résumé – *Au sujet de la capacité d'anhydrobiose en champ de* Pratylenchus thornei *et de* Merlinius brevidens – La présente étude du comportement anhydrobiotique de *Pratylenchus thornei* et de *Merlinius brevidens* a été réalisée dans un champ non irrigué de blé cv. Gallareta, variété considérée comme non-hôte ou mauvais hôte pour ces nématodes. *P. thornei* paraît plus dépendant de l'humidité du sol que *M. brevidens* en ce qui concerne sa récupération après 15 h de migration (réhydratation, réactivation et migration). *M. brevidens* sort plus facilement de son état anhydrobiotique. Les schémas d'émergence sont semblables pour les deux nématodes qu'ils soient actifs ou récupérés après une anhydrobiose prononcée. Ces schémas sont par contre différents s'il s'agit d'un stade précode d'anhydrobiose (anhydrobiose modérée). Les deux nématodes font montre, en relation avec la profondeur du sol, d'une inertie à entrer en anhydrobiose qui paraît dépendre des processus naturels de dessication du sol du champ. Cette inertie semble plus prononcée chez *M. brevidens*.

Key-words : Nematodes, *Pratylenchus, Merlinius,* anhydrobiosis, quantitative recovery.

There is a correlation between species anhydrobiotic ability and their habitat conditions (Tsai & Van Gundy, 1988). Nematodes are well adapted to their environment and the natural dehydration regimes are important for nematode anhydrobiosis potential (Womersley & Ching, 1989). *Tylenchulus semipenetrans* from irrigated soil have shown an earlier stage of anhydrobiosis (Tsai, 1987).

For a better understanding of the nematode anhydrobiosis, under natural field conditions, a study has been designed and carried out on the anhydrobiotic behaviour of *Pratylenchus thornei* and *Merlinius brevidens* from dry soils. The study has been conducted along the life cycle of a wheat cultivar, bringing into consideration both soil and root habitats, as it has been suggested previously (MacGuidwin, 1989).

Material and methods

In a non-irrigated field, representative of an important area of Southern Spain, dry vertic soils with a sandy loam texture, a chosen subarea of 24 × 4 m was subdivided into 20 × 20 cm squares. Four of the latter were selected at random, as replicates, for a fortnight sampling scheme. The sampling was started at the centre of each replicate on 15 January, when the seedlings of the wheat cv. Gallareta, growing in the field, were 15-20 cm high and their root system reached in the first 5 cm a mean weight of 0.7901 grammes per 1008 g of soil, corresponding to a theoretical volume of 1125 ml (15 × 15 cm, down to 5 cm depth). It was stopped at fortnight 19, on 15 October. The same sampling volume was taken at each time and for each replicate, but down to 5, 10, 15 and 20 cm depth. The wheat was harvested between fortnights 12 and 13.

Owing to the peculiar characteristics of the soil, which cannot be handled in the laboratory when moist, the approximately 1008 g of each soil sample, after determination of its moisture content, was always diluted and suspended in water. This suspension was first sieved through a mesh to retain all the wheat roots. Then it was processed through a funnel provided at the apex with four equidistant tubes so that aliquots containing 120 g or soil were obtained at the end.

One aliquot per replicate was processed by differential sedimentation in water in an elutriator, sieving of the supernatant through four 53 µm sieves and then active migration of the nematodes, through a filter, to clean tap water. Migration through the nematode filter (rehydration plus migration) was left to proceed for along 135 hours, which were divided into six times (T1-T6; T1 corresponds with the first 15 h and T2-T6 with

Table 1. *Soil moisture (% of soil weight) at nineteen different sampling times (fortnights) and four depths (Values are the mean of four replicates).*

Fortnight	Depths			
	0-5 cm	5-10 cm	10-15 cm	15-20 cm
F1	15.63	13.54	14.83	14.14
F2	4.23	5.64	8.55	7.95
F3	4.79	5.48	6.05	6.34
F4	17.09	17.80	15.18	13.94
F5	10.27	10.74	11.22	11.42
F6	5.30	6.71	9.03	8.97
F7	9.05	12.81	12.86	12.97
F8	4.69	7.42	6.70	7.24
F9	3.49	5.42	6.59	6.44
F10	5.15	7.28	6.70	6.94
F11	14.27	13.19	13.02	11.07
F12	15.14	13.94	14.67	13.94
F13	5.01	13.64	12.00	13.54
F14	3.08	6.75	10.67	8.70
F15	2.90	5.70	6.90	5.90
F16	2.50	4.70	6.00	9.80
F17	2.23	3.16	3.62	5.06
F18	3.88	2.84	4.23	5.59
F19	14.25	13.81	14.62	15.85
Means	7.52	8.98	9.66	9.78

Table 2. *Absolute and relative densities (Dens) of* Pratylenchus thornei *in soil (S) and roots (R) of wheat cv. Gallareta. corresponding to 120 g of soil, at four different sampling times (fortnights F1, F6, F12, F19) and four depths. (Values are the mean of four replicates).*

		F1 (15 janv.)		F6 (30 mars)		F12 (30 jun)		F19 (15 oct.)	
Depths		Dens	%	Dens	%	Dens	%	Dens	%
(0-5)	S	467	96.1	21	40.4	20	87.0	46	97.9
	R	19	3.9	31	59.6	3	13.0	1	2.1
	T	486		52		23		47	
(5-10)	S	1213	90.1	96	41.9	177	88.9	205	99.0
	R	134	9.9	133	58.1	22	11.1	2	1.0
	T	1347		229		199		207	
(10-15)	S	1825	94.3	303	67.0	144	87.8	184	99.5
	R	111	5.7	149	33.0	20	12.2	1	0.5
	T	1936		452		164		185	
(15-20)	S	1279	95.6	244	72.2	140	89.2	128	100
	R	59	4.4	94	27.8	17	10.8	0	0
	T	1338		338		157		128	
Means	S	1196	93.7	166	62.2	120	88.2	141	99.3
	R	81	6.3	101	37.8	16	11.8	1	0.7
	T	1277		267		136		142	

Table 3. *Correlations between nematode population density and wheat cv. Gallareta root weight/soil humidity, at four depths, over the nineteen fortnights sampling scheme. (From the means of four replicates) (*, **, *** significance of the " r " value at the 5 %, 1 %, 0.1 % probability levels) (T1, T2, T3 respectively 15, 39 and 63 hours of migration).*

	Depths			
	(0-5)	(5-10)	(10-15)	(15-20)
	ROOT WEIGHT			
P. thornei				
Soil	−0.539*	−0.546*	−0.438	+0.267
Roots	+0.067	+0.593**	+0.528*	+0.583**
% (in roots)	+0.634**	+0.858***	+0.652**	+0.415
M. brevidens				
Soil	−0.156	−0.484*	−0.497*	−0.466*
	SOIL HUMIDITY			
P. thornei				
T1	+0.637**	+0.727***	+0.557*	+0.309
T2	−0.688**	−0.666**	−0.735***	−0.512 *
T3	−0.563*	−0.560*	−0.538*	−0.234
M. brevidens				
T1	+0.236	+0.426	+0.126	+0.202
T2	−0.692**	−0.795***	−0.714***	−0.512*
T3	−0.579**	−0.687**	−0.641**	−0.282

additional periods of 24 h). After each time the water embedded in the nematode filters was changed and these were replaced in a fresh volume of tap water to stimulate the nematode activity (Tobar *et al.*, 1995).

The roots from the 1008 g of soil replicates were weighed, cut into small pieces and processed in a mistifyer for 14 days to recover the endoparasitic *Pratylenchus thornei.*

Absolute and relative values, transformed to $\log_{10}$ and angles, respectively, were studied by analysis of the variance. The resulting estimations were compared by means of the corresponding least significant difference.

Results and discussion

SOIL HUMIDITY, ROOT DEVELOPMENT AND HOST ABILITY OF WHEAT CV. GALLARETA

Soil humidity went up and down significantly throughout the whole sampling scheme (Table 1), from fortnights 1 to 19 (F1-F19). Over all, the mean humidity of this dry soil decreased from the 20 cm depth up to the field surface.

The root weights increased until F6 (30 March); they went down then from 1.2 g (0.8436-1.5591) at 0-5 cm, to 0.1838 (0.1393-0.2451) at 15-20 cm, per 120 g of soil. They decreased from then on until F19 (15 October).

Though the highest relative densities of *P. thornei* are located in roots at F6 (30 March), when the tillers of

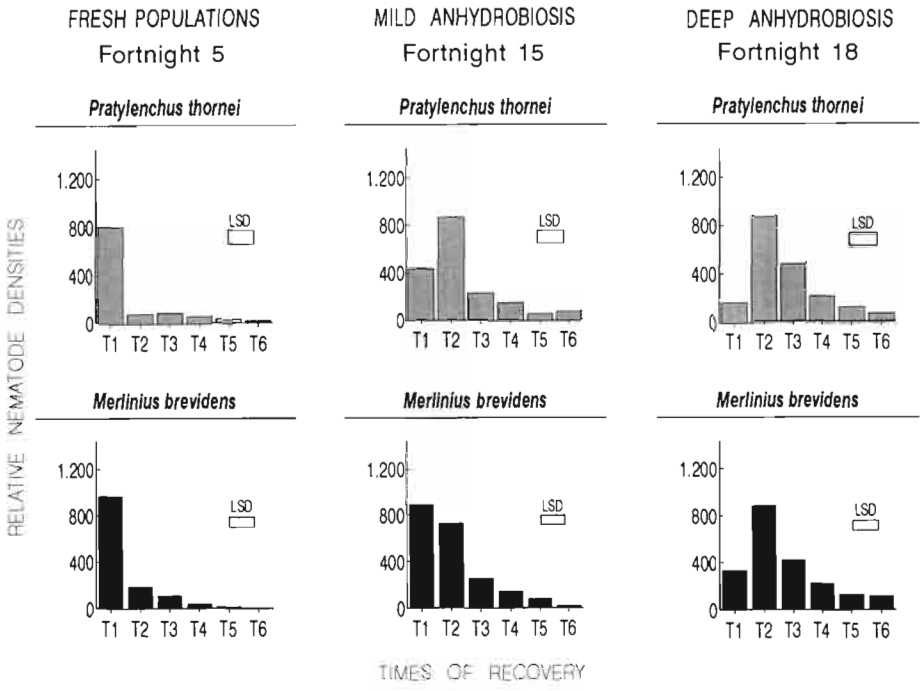

Fig. 1. *Effect of sampling time on the rate of recovery from anhydrobiosis in* Pratylenchus thornei *and* Merlinius brevidens. *T1 = 15 h; T2 = T1 + 24 h; T3 = T2 + 24 h; 14 = 13 + 24 h; T5 = T4 + 24 h; T6 = T5 + 24 h.*

wheat cv. Gallareta are spouting, similarly to a previous finding (Esmenjaud *et al.,* 1990), the absolute nematode densities fall abruptly under the cultivation of the wheat (Table 2). The correlation coefficients for absolute and relative numbers of *P. thornei* in roots with the root weight are positive and significant (Table 3) and the « r » values for the densities in soil are negative. *Merlinius brevidens* densities give a negative correlation against root weights. Consequently, it is shown that both nematodes either do not reproduce or do not reproduce actively on this wheat variety.

Soil humidity in relation to the recovery of *P. thornei* and *M. brevidens* from dry soil

It has been established that a quantitative recovery of 95 % or more of the *P. thornei* and *M. brevidens* populations from dry soil is reached at time T6 (135 h) of recovery (Tobar *et al.,* 1995). Nevertheless, the most outstanding differences are shown by the times T1 (15 h), T2 (39 h), T3 (63 h).

The " r " values for densities of *P. thornei* in soil against soil humidity are significant/positive for T1 and significant/negative for T2 and T3 (Table 3). *M. brevidens* gives the same results for T2 and T3, but its " r " values for T1, though positive, are not significant. The greater density were obtained from T1, the smaller ones to get from T2 and T3.

P. thornei seems to be more dependent on soil humidity for its recovery in T1 than *M. brevidens,* which seems to emerge more easily from anhydrobiosis.

Patterns of emergence from anhydrobiosis of *P. thornei* and *M. brevidens*

To see the patterns of emergence from anhydrobiosis given by *P. thornei* and *M. brevidens* from wet and dry soil, three fortnights are chosen from the whole sampling scheme : F5 (15 March), with mean soil humidity 10.9 % (10.27-11.42); F15 (15 August), 5.35 % (2.90-6.90); and F18 (30 September), 4.14 % (2.84-5.59). The last two had the soil similarly dried, though the second for a longer time.

Figure 1 shows : the typical similar patterns, given by fresh *P. thornei* and *M. brevidens* populations (fortnight F5); the different patterns for the two nematodes, when recovered from a mild anhydrobiosis (fortnight F15), which show a faster recovery for *M. brevidens,* given previously as a distinction between endo and ectoparasitic nematode way of life (Talavera *et al.,* 1992); and the similar/almost equal patterns (fortnight F18), shown again by the two nematodes, when recovered from deep anhydrobiosis.

Similarly to what has been seen by de Guiran (1979) in anhydrobiotic quiescence of *Meloidogyne incognita,* the longer *P. thornei* and *M. brevidens* stay under an-

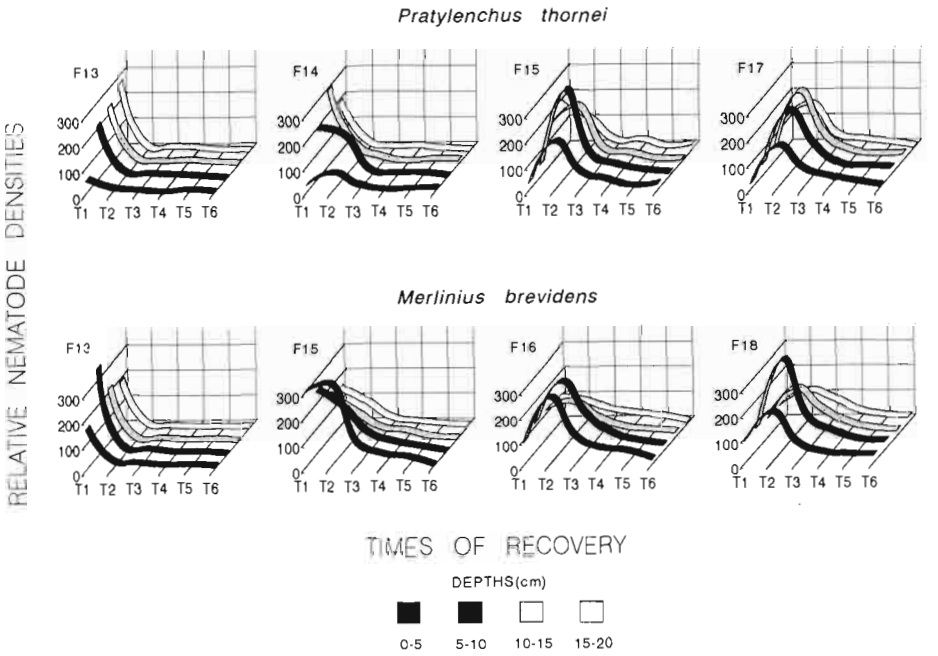

Fig. 2. *Effect of depth of sampling on the rate of recovery from anhydrobiosis in* Pratylenchus thornei *and* Merlinius brevidens *on four critical fortnights. T1 = 15 h; T2 = T1 + 24 h; T3 = T2 + 24 h; T4 = T3 + 24 h; T5 = T4 + 24 h; T6 = T5 + 24 h.*

hydrobiosis the longer time needed for their rehydration and reactivation.

INERTIA, WITH SOIL DEPTH, IN THE RETURN FROM ANHYDROBIOSIS OF *P. THORNEI* AND *M. BREVIDENS*

The induction of anhydrobiosis is determined by the physical forces exerted by the water film surrounding the nematodes (Demeure *et al.*, 1979). Consequently, the nematode anhydrobiosis is progressing downwards as the soil is losing water from the upper to the lower layers over the fortnights.

The rehydration/reactivation of *P. thornei* and *M. brevidens* interchanges its maximum rate of recovery from T1 (15 h) to T2 (39 h) as their state of anhydrobiosis takes longer, which is inversely related to the depth (Fig. 2).

P. thornei has a greater ability to enter into anhydrobiosis. Its state of anhydrobiosis in relation to depth at F13, F14, F15 and F17 is the most similar to that of *M. brevidens* at F13, F15, F16 and F18. Again *M. brevidens* shows the need for a longer time under anhydrobiosis to have an emergence similar to that of *P. thornei.*

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