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An undescribed *Steinernema* sp. (Nematoda: Steinernematidae) from Germany and the Scandinavian Subarctic

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Summary. Infective-stage juveniles of an undescribed *Steinernema* species were isolated by direct extraction from soil samples in Germany. The samples originated from five non-arable sites in the northern region of the country. The morphology of the juveniles resembled that of *S. intermedium* and *S. affine*. *Steinernema* juveniles isolated from a subarctic heath in northern Sweden appear to belong to the same undescribed species. The potential hosts appear to prefer natural, non-arable, moist habitats.
Key words: Arctic, entomopathogenic nematode, Germany, soil, *Steinernema*, Sweden.

Studies on the natural occurrence of entomopathogenic nematodes in Germany revealed the presence of a total of eleven *Steinernema* and two *Heterorhabditis* species, among them several still undescribed *Steinernema* species (Sturhan, 1996). One of the undescribed species has been reported as "*Steinernema* sp. E" (Sturhan, 1995, 1997). Among nematodes isolated from soil samples collected from a subarctic heath in northern Sweden infective-stage juveniles of a *Steinernema* species were found, which were morphologically similar to the infective juveniles of the undescribed "spec. E" from Germany. The morphological characters of the infective-stage juveniles of this species are described and details of the sampling sites in Germany and Sweden are given.

MATERIALS AND METHODS

Nematodes were extracted from bulk soil samples collected in Germany by sieving-decanting or centrifugation-flotation methods (Sturhan, 1995) and from the Subarctic soil samples (one core each, divided into subsamples of 0-3 cm and 3-6 cm soil depths) using a modified Baermann method (Ruess *et al.*, 1998). The nematodes were fixed in hot TAF or hot 4% formalin and transferred to pure glycerine by a slow evaporation method for morphological studies. Voucher specimens on permanent microscopical slides are deposited in the German Nematode Collection (DNST), Biologische Bundesanstalt, Münster, Germany.

RESULTS

Morphology of the infective juveniles. Morphometric characters of the infective-stage juveniles of three populations from Germany and Sweden are given in Table 1. Body of heat-relaxed specimens slightly curved (Fig. 1C), gradually tapering towards anterior and posterior ends, to about 95 %, 35 % and 65 % of maximum body width at base of oesophagus, lip region and at anus. Cuticle annulated; annules 1.3-1.6 μm wide at mid-body. Lateral fields with weak marginal, well-demarcated submarginal and three less distinct central lines (Fig. 1D, E). Deirids in centre of the lateral fields immediately posterior to nerve ring; phasmids at ventral side of the lateral fields, at 30-33 % of the tail length posterior to the anus.

Lip region rounded, often slightly flattened anteriorly, continuous with body contour or only weakly offset by a slight depression, 9-11 μm wide at base (Fig. 1A). Oesophagus with slightly widened anterior part, isthmus surrounded by nerve ring and weakly valvated terminal bulb; cardia distinct. Excretory pore slightly anterior to middle of oesophagus, with wide duct; excretory glands at ventral side near oesophago-intestinal junction, extending to about one body width beyond oesophageal bulb. Hemizonid at isthmus level, about 4 μm long. Bacterial vesicle posterior to cardia, mostly between one and two corresponding body widths long, usually filled with thread-like bacteria. Lumen of intestine col-

Table 1. Morphometric characters of infective-stage juveniles of *Steinernema* sp. E.

Character	Population		
	Langeoog	Selenter See	Swedish Subarctic
n	10	10	10
Length (μm)	743 (550-850)	780 (630-910)	731 (630-830)
Head to excretory pore in % of oesophagus length	45.5 (40-50)	45.1 (39-48)	46.6 (43-51)
Tail length (μm)	81 (71-89)	90 (80-99)	78 (70-82.5)
Hyaline tail length (μm)	45 (38-51.5)	51 (47-56)	45.5 (36.5-50.5)
Hyaline tail length in % of tail length	55 (52-60)	57 (54-60)	58 (52-62)
a	27.5 (23-29.5)	31 (29-34)	26 (24.5-27.5)
b	5.7 (5.2-6.0)	5.7 (4.9-6.4)	5.5 (4.6-6.2)
c	9.3 (8.5-10.3)	8.6 (7.9-9.2)	9.4 (8.4-10.0)
c'	4.8 (4.5-5.4)	6.0 (5.0-6.7)	5.0 (4.5-5.6)
h'*	6.3 (5.4-7.0)	7.8 (6.5-9.6)	5.8 (5.2-6.7)

*Hyaline tail length divided by width at proximal end of hyaline tail portion.

lapsed, rectum straight, about 1.5 anal body widths long.

Tail slender conoid, often with weak irregular narrowings, straight or tip slightly curved ventrad or dorsad (Fig. 1B). Hyaline posterior part always longer than half tail length; close to tail tip often with irregular granular inclusions. Tail terminus finely rounded.

Differential diagnosis. In general appearance, shape of the lip region and in particular structure of the lateral field *Steinernema* spec. E most closely resembles *S. affine* (Bovien, 1937) and *S. intermedium* (Poinar, 1985). The body of heat-killed specimens of both species is generally more C-shaped than in *Steinernema* sp. E and the tail terminus is always sharply pointed. In *S. affine* the extreme tail tip is refractile, the tail and the hyaline tail portion are shorter (less than half tail length). In *S. intermedium* the tail has generally a somewhat irregular outline with a dorsal depression at about half tail length and the hyaline portion covers around half tail length. Occasionally the tail shape of *Steinernema* sp. E is similar to that of *S. intermedium.*

Distribution and ecology. Till present *Steinernema* sp. E has been found at five sites in Germany, all located in the northern part of the country: Isle of Langeoog, moist depression with grasses and moss behind the coastal dunes, humous loamy sand, pH 5.9, collected 14 May 1983; Heeslingen near Zeven, river bank vegetation with *Alnus glutinosa* (L.), *Humulus lupulus* L., *Urtica dioica* L., *Glechoma hederacea* L. and Gramineae, loamy sand, pH 5.9, collected 30 July 1990; Selenter See, Schleswig-Holstein, vegetation at lake shore with grasses and various dicots, sandy soil, pH 7.6, soil sample taken at 3 July, 1985, and at the same site at 24 May, 1992; Berlin-Dahlem, unused part of the experimental field of the Biologische Bundesanstalt, with grasses and various herbs, sandy soil, collected 1 February 1994; Kleinmachnow near Potsdam, bog with *Alnus glutinosa* and *Acer pseudo-platanus* L., grasses and *Urtica dioica,* very moist humous soil, collected 18 October 1993. Resampling at the last two sites in November 1998 revealed no additional specimens of "spec. E".

From the bulk soil sample taken in May 1992 at the Selenter See besides "spec. E" the species *S. affine, S. feltiae* (Filipjev, 1934) and *S. kraussei* (Steiner, 1923) were isolated. *Steinernema affine* was also present in the soil sample from Berlin-Dahlem. With the exception of the sample from Langeoog the number of infective-stage juveniles extracted from the soil samples was only low.

In Sweden soil samples were taken in the Subarctic near Abisko, northern Swedish Lapland (68° 20'N, 20° 51'E) in August 1996. The site is a subalpine heath close to the tree limit at 450 m a.s.l. on a slope facing WNW. The climate of this region is subarctic, with a growing season from mid June till early September. In 1996 the mean annual precipitation was 278 mm and the mean annual air temperature -0.5 °C. The bedrock of the site consists mainly of base-rich mica schists and the humus layer is well-developed with at least 10 cm depth; soil pH was 7.1, soil organic matter content 82%. The vegetation is a species-rich dwarf shrub heath community with *Cassiope tetragona* (L.) as the dominant plant. At the site the following treatments were established every growing season since 1989: Temperature enhancement (soil +1.5 °C, air +7.7 °C), application of fertilizer (10, 2.6 and 9 g m^{-2} N, P and K, respectively), temperature enhancement + fertilizer, and untreated control. For more details on climate, vegetation, soil and treatments see Havström *et al.* (1993) and Jonasson *et al.* (1993).

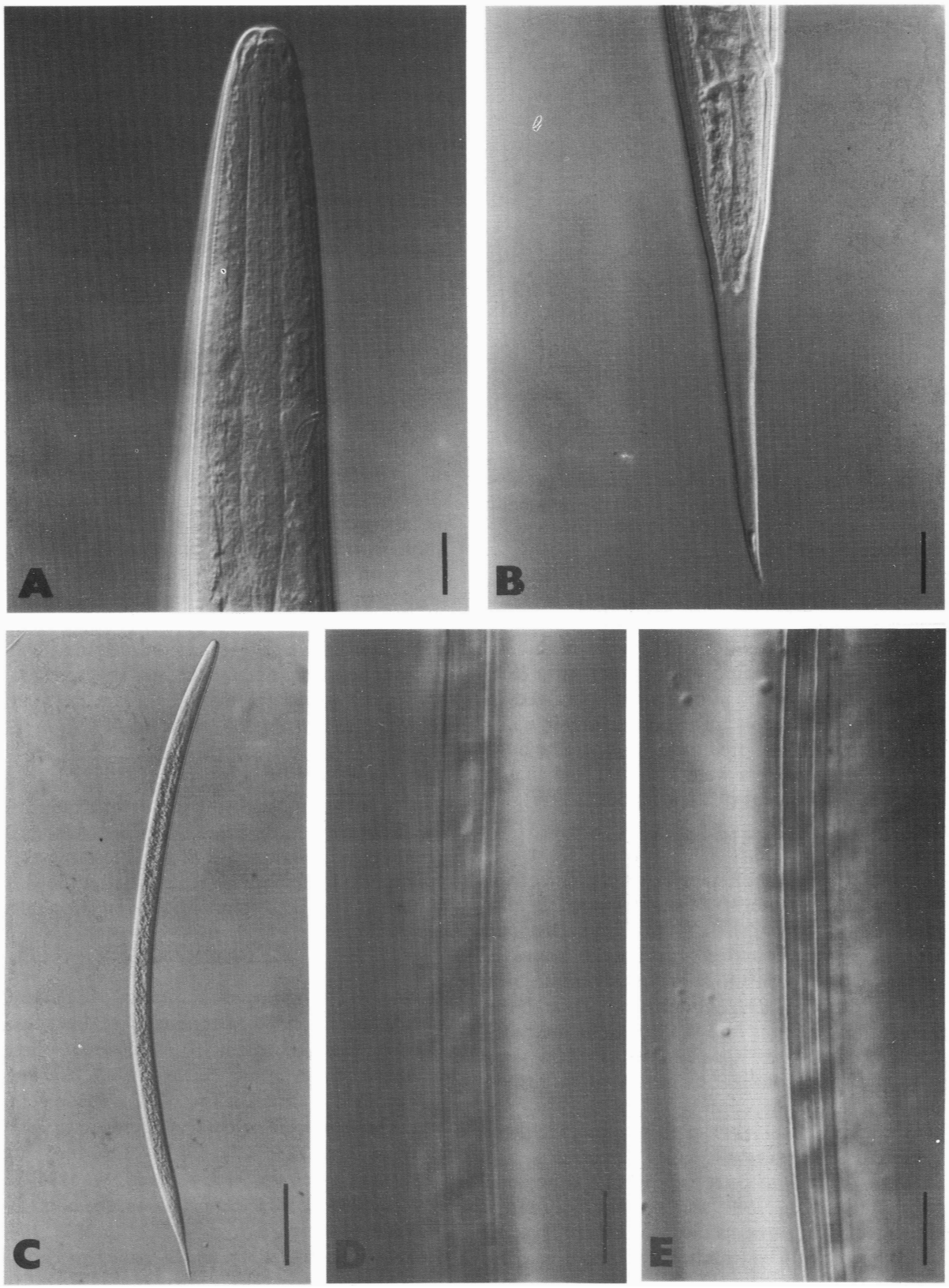

Fig. 1. Infective juveniles of *Steinernema* sp. E. A: Anterior end; B: Tail; C: Habitus; D, E: Lateral field at mid-body. Scale bars: A,B,D,E - 10 µm, C - 100 µm.

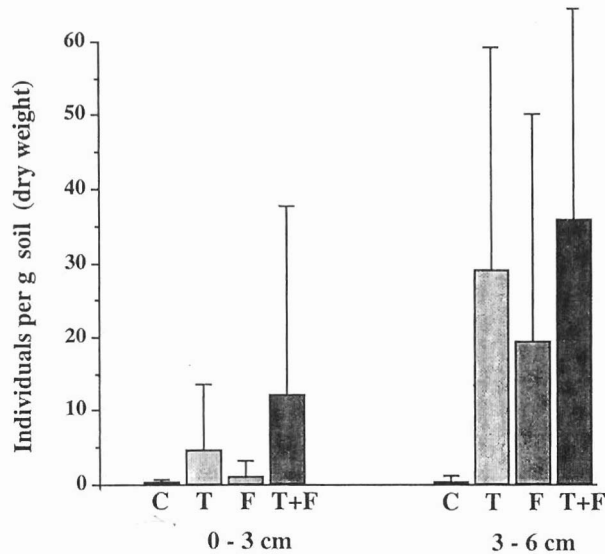

Fig. 2. Numbers of *Steinernema* sp. E infective juveniles per gram of soil (dry weight) of two soil layers in a Swedish subarctic heath (C - control, T - elevated temperature, F - fertilizer applied, n = 6).

The population density of *Steinernema* was remarkably high at the subarctic sampling site. The average numbers of juveniles were higher in the 3-6 cm soil layer than in the upper 0-3 cm soil layer (Fig. 2). A strong increase of numbers was observed in plots with elevated temperature, particularly in combination with fertilizer-treatment. Generally densities in the treated plots were 4 to 100 fold higher compared to the control (0.3 specimens/g DW).

DISCUSSION

The morphological characters of the infective-stage juveniles characterise *Steinernema* sp. E as a member of the *S. affine* group. Of this group *S. affine, S. intermedium* and an undescribed species from UK have been reported for Europe previously (Hominick *et al.*, 1995, Sturhan, 1997, and others). Though the juveniles from Germany and the Subarctic are morphologically similar, the species identity of the geographically distant populations has to be confirmed by further studies.

In Germany *Steinernema* sp. E is among the most rarely found *Steinernema* species, amounting for about 0.5% of all records of entomopathogenic nematodes (Sturhan, 1997 and unpublished data). The presence of the same species in northern Sweden indicates that it might be widely distributed in (northern) Europe. It might be possible that it has been "overlooked" in surveys conducted in other European countries using the baiting technique with *Galleria* larvae.

The record of *Steinernema* for northern Sweden is of particular interest, since there are few records of entomopathogenic nematodes from north of the polar circle *i.e.* Vänninen *et al.* (1989) isolated *Steinernema* (probably *S. feltiae*) from soil from one site in northern Finland, and Haukeland Salinas (1996) found *Steinernema* sp. in two samples from Tromsø and the Kirkenes region in northern Norway. Other entomopathogenic nematodes reported for Scandinavia are *S. carpocapsae* (Weiser), *S. intermedium* (Poinar) and *Heterorhabditis* sp. (Burman *et al.*, 1986; Haukeland, 1993). In a survey of entomopathogenic nematodes in Western Canada *Steinernema* was found in British Columbia but not further north in the Yukon Territory (Mracek & Webster, 1993). Tests of Antarctic soils for insect parasitic nematodes showed no evidence of steinernematids or heterorhabditids (Griffin *et al.*, 1990).

Till now nothing is known about potential hosts of *Steinernema* sp. E, but the sampling sites in Germany and Sweden indicate that it might be hosts preferring non-arable and moist soils. The higher numbers of *Steinernema* juveniles at the subarctic sites with fertilization or/and elevated temperature suggest that the increased vegetation cover at these plots may have attracted the host. The composition of the soil fauna at high latitudes is dominated by microarthropods, nematodes and a significant range of Diptera larvae (Heal, 1997). The latter possibly could act as host for *Steinernema*.

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Sturhan D., Ruess L. Неописанный вид *Steinernema* (Nematoda: Steinernematidae) из Германии и Скандинавской Субарктики.

Резюме. Инвазионные личинки неописанного вида *Steinernema* были выделены методом прямой экстракции из почвенных образцов Германии. Образцы были собраны в пяти точках северной части страны из мест, не подвергавшихся сельскохозяйственной обработке. По своей морфологии выделенные личинки сходны с *S. intermedium* и *S. affine*. Личинки *Steinernema* sp., выделенные из почвы субарктических пустошей на севере Швеции, также, по-видимому, принадлежат к этому неописанному виду. Потенциальные хозяева этих нематод предпочитают, вероятно, сырые некультивируемые места обитания.