FIELD DAMAGE IN POTATO BY LESION NEMATODE *PRATYLENCHUS PENETRANS*, ITS ASSOCIATION WITH TUBER SYMPTOMS AND ITS SURVIVAL IN STORAGE

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Summary. Soil samples from an area of depressed growth in a potato (S*olanum tuberosum*) cv. Saturna field in Grue, eastern Norway, yielded large numbers of root lesion nematodes, *Pratylenchus penetrans*. The yield of potatoes was reduced by 50% in the affected area of the field. Transect-sampling showed plant growth to be negatively correlated with densities of *P. penetrans* and suggested a damage threshold of potato to the nematode of 100 specimens per 250 g of soil. Common scab (*Streptomyces scabies*) occurred frequently in the affected area. *Pratylenchus penetrans* was present in roots, underground stems, stolons and tubers. In tubers, nematodes were detected inside cross-lesions typical of common scab, and occurred also in the outermost 0.5 mm of tissue associated with such lesions. On potato cv. Saturna grown in a greenhouse, *P. penetrans* alone induced tuber lesions similar to those of common scab. Also, the combined inoculation of the bacterium and the nematode seemed to enhance symptom expression. *Pratylenchus penetrans* survives storage of potatoes, and new infections may develop from infected tubers used as seed. Hence, potato tubers do appear to be an important means for the spread of *P. penetrans* to new areas.

Keywords: Damage threshold, Norway, *Solanum tuberosum*, *Streptomyces scabies*, yield loss.

In the summer of 1996, an area of depressed growth in a field of potato (*Solanum tuberosum* L.) cv. Saturna [resistant to pathotype Ro1 of *Globodera rostochiensis* (Woll.) Skarbilovich], suggestive of cyst nematode damage, was detected in Grue, eastern Norway (Fig. 1). However, analyses of soil samples did not detect potato cyst nematodes, but did demonstrate the occurrence of a large number of lesion nematodes *Pratylenchus penetrans* (Cobb) Filipjev *et* Schuurmans Stekhoven. Tubers from the area of depressed growth had severe symptoms similar to those caused by the common scab bacterium, *Streptomyces scabies* (Thaxter) Lambert *et* Loira.

The objectives of this study were to investigate: *i*) a possible relationship between numbers of *P. penetrans* and damage to potato; *ii*) the symptom expression of nematodes and common scab; and *iii*) the survival of the nematode in stored potato tubers.

MATERIALS AND METHODS

Relationships between damage and nematode density. The previous crop in the affected field had been barley for the last 3 years. In August 1996, when potatoes were in growth stage IV (tuber bulking), nine samples were taken on a transect from the centre of the growth depression to the first apparently healthy row of potatoes, cv. Saturna (Fig. 1), using an auger with a half cylindrical blade (20 cm × 3.0 cm). Plants and tubers from the field were also collected for further studies. Two months later the infected field was harvested. The tuber yield and size were graded using a commercial scale. The frequencies of scab symptoms on tubers were also assessed, using a standardized grading scale (Bjor and Roer, 1980). These observations were made on tubers collected from the affected area and from unaffected parts of the field.

Nematodes were extracted from soil using the Seinhorst elutriator (Seinhorst, 1988), killed by heat, fixed in formalin-acetic acid 4:1, mounted on temporary slides and identified under a LEITZ DMR interference contrast microscope with Leica Qwin "Image Processing and Analysis System". Identification of *Pratylenchus* nematodes was made by morphometric studies of adult females and males using the key of Loof (1978).

To observe nematodes in tuber tissue, potato tubers were boiled in 0.05% acid fuchsin in lacto-glycerol for 7 minutes (Hooper, 1986). After cooling, 1 cm diameter circular areas of the peel were cut out with a cork borer, the outer part of the skin was examined, and approximately 2-3 mm thin sections were mounted on slides and examined under the microscope. The occurrence of nematodes was documented by photography. Underground stems and stolons were examined for lesions using a binocular microscope, and segments with lesions were separated into two portions. One portion was boiled in 0.05% acid fuchsin in lacto-glycerol for 7 minutes (Hooper, 1986) and examined, while the other portion was chopped into pieces of 1 cm length and nematodes extracted with the Bærmann funnel (Hooper, 1986). From tubers with scab symptoms, nematode were extracted using the method described by Koen and Hogewind (1967), fixed and identified as described before.

Development of symptoms. The development of symptoms on potato was studied in a greenhouse. A standard population of the scab bacterium *S. scabies*, obtained from the national programme for potato breeding, was reared on potato dextrose agar at 22-23 °C for 3 weeks. The cultures were then homogenized in water to produce the inoculum suspension, which was inoculated according to standard procedures of Bjor and Roer (1980).

Pratylenchus penetrans was reared in a greenhouse on lettuce (*Lactuca sativa* L.). Micro-tubers of potato cv. Saturna, produced from meristems, were planted in 48 two-litre pots containing sterile sand. There were four treatments each with twelve pots: *i*) Control, potato only; *ii*) 2000 *P. penetrans* per pot; *iii*) 100 ml bacterium suspension per pot; *iv*) 2000 *P. penetrans* and 100 ml bacterium suspension per pot.

The plants were grown at day/night temperatures of 15 and 10 °C, and a light period of 18 hours. The extension, severity and index of scab symptoms on tubers (Bjor and Roer, 1980) were evaluated in all plots at 8, 11 and 14 weeks post-inoculation. Treatment means based on four randomly selected pots were compared with Student's t-test. A statistically significant difference was recognized at $P \leq 0.05$, and a tendency was recorded in the probability range of $0.10 \geq P \geq 0.05$.

Survival during storage. To study the survival of nematodes, infected tubers from the field were stored at 4 °C, for 20 weeks, then transferred to five pots with sterile sand, and grown for 3 months in a greenhouse. After harvest, soil, roots, stolons, tubers and skin lesions were examined for the presence of lesion nematodes as described before.

RESULTS

Damage and nematode densities. The area of depressed growth in the potato crop covered approximately 300 m^2 of the field (Fig. 1). Growth of potato was greatly reduced in the centre of the patch, with non-vigorous and stunted plants, and increased gradually towards the margins. Plant growth was negatively correlated with densities of *P. penetrans* in the soil. The nematode density in the centre of the affected area was 900 specimens/250 g of soil, compared with 40 specimens/250 g of soil at the margins, where plant growth seemed unaffected. In August, the first signs of plant damage coincided with a nematode population density of 110 individuals/250 g of soil.

At harvest in October, the tuber yield in the depressed area was of 15,000 kg/ha compared with 30,000 kg/ha in areas with normal growth. Scab symptoms were more severe in the affected area compared to the area with normal growth.

Fig. 1. Damage by lesion nematodes, *Pratylenchus penetrans*, in a cv. Saturna potato field in Grue, Norway.

Symptoms and nematode association. Roots and stolons from the field showed brownish root lesions and necrosis in cortical cells. Large numbers of *P. penetrans* were extracted in all stages of development from roots, underground stems, stolons and tubers. Root systems were partially or almost totally destroyed by nematodes and probably secondary infections by micro-organisms. Tubers from the affected area had a high incidence of common scab, *S. scabies*, symptoms (Fig. 2) as the tuber skins were marked by cross-shaped lesions typical of the pathogen (Fig. 3). Examination of such lesions demonstrated the presence of *P. penetrans* in the lesion tissue and in the tissue 0.5 mm under the tuber skin close to these cross-lesions.

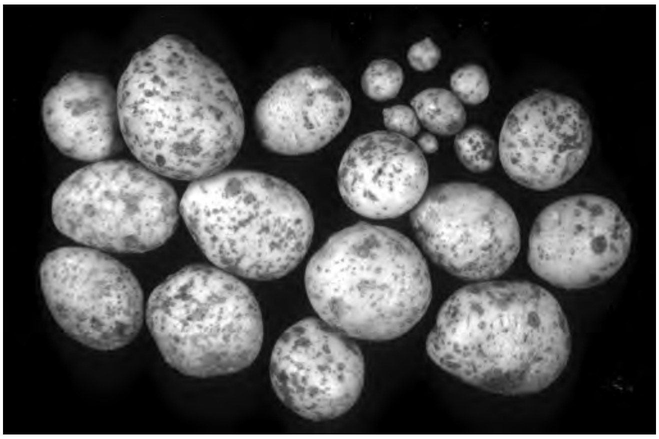

Fig. 2. Symptoms of common scab *Streptomyces scabies* on potato (cv. Saturna) from a field infested by the lesion nematodes *P. penetrans* in Grue, Norway.

Symptom expression. In the greenhouse study, the expression of scab symptoms (Table I) in the form of such cross-shaped lesions was found in all treatments, including a low frequency also in the control treatment. *Pratylenchus penetrans* alone induced the formation of cross lesions on tubers (Fig. 4) similar to the lesions caused by *S. scabies*.

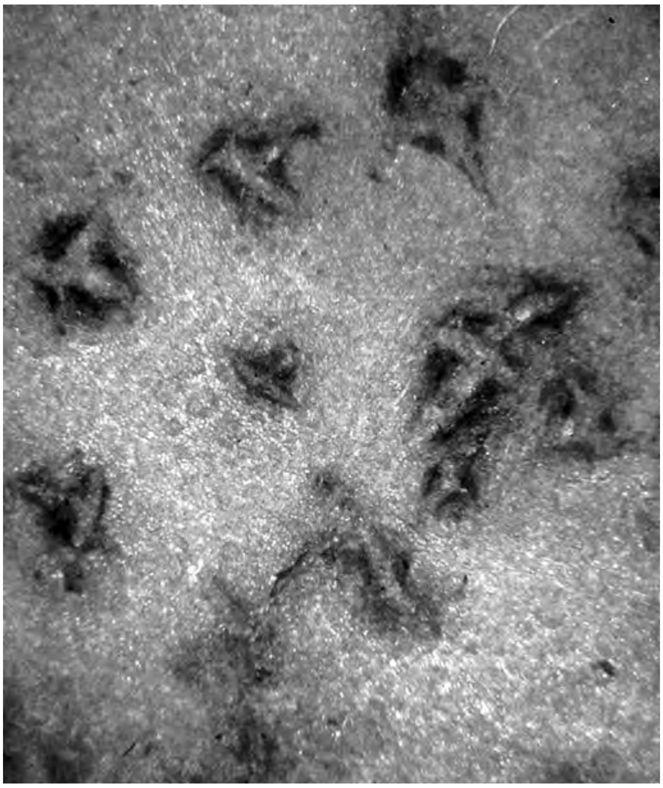

Fig. 3. Detail of a tuber from the affected field showing cross-lesions typical of common scab S. *scabies*.

Fig. 4. Cross-lesions on tubers from plants inoculated with 2000 specimens of *P. penetrans* per pot in a greenhouse.

Eight weeks after inoculation, only plants inoculated with *P. penetrans* alone had significantly (P = 0.0418) more "extensive symptoms of scab" than control plants. However, no significant difference could be detected compared with the treatment with *S. scabies* alone and the treatment with *S. scabies* and nematodes combined. The "severity of scab symptoms" was significantly (P = 0.0162) more for plants inoculated with *P. penetrans* alone than for all other treatments, but no significant differences of "scab index" were observed.

After 11 weeks, the nematode-bacterium treatment had a significantly (P = 0.0517) higher score (3.5) for "extension of scab symptoms" than controls (0.5). For "severity of scab symptoms" the nematode-bacterium combination tended (P = 0.0807) to have a higher score (2.3) than controls (0.5). Also, a score of 2.3 was recorded in the treatment with *P. penetrans* alone, but this was not significantly different from the other treatments. For "scab index" there was a tendency (P = 0.0969) for the nematode-bacterium treatment to have a higher score (32.4) compared to the control treatment (1.9).

At 14 weeks, the only significant difference recorded was in the "extension of scab symptoms", which was 2.8 in the nematode-bacterium treatment and 0 in the control.

Table I. Scab symptom expression in potato (cv. Saturna) inoculated with lesion nematodes *Pratylenchus penetrans* and the common scab bacterium *Streptomyces scabies* alone and combined. Mean numbers (n = 4) followed by the same letter are not statistically different at (P = 0.05). Symptom indices according to Bjor and Roer (1980).

Treatment	Extension of symptoms* Week post-inoculation			Severity of symptoms** Week post-inoculation			Scab index*** Week post-inoculation		
	8	11	14	8	11	14	8	11	14
Control	0.8a	0.5a	0a	0.5a	0.5a	0a	4.9a	1.9a	0a
P. penetrans	3.3b	3.3ab	2.1ab	2.5b	2.3a	1.3a	23.1a	28.5a	12.8a
S. scabies	2.3ab	2.3ab	1.6ab	1.3a	1.5a	1.3a	10.6a	20.5a	11.8a
P. penetrans + *S. scabies*	2.8ab	3.5bc[t]	2.8b	1.0a	2.3ab[t]	1.3a	10.5a	32.4ab[t]	14.3a

[t]= tendency to difference when 0.10 ≥ P ≥ 0.05.
*Extension of symptoms = % of tuber surface covered.

Score Relative surface covered	0	1	2	3	4	5	6	7	8	9
% of surface covered	0	<5	5-10	10-25	25-50	50-75	75-90	90-95	95-99	100

**Severity of symptoms score 1= superficial scab; 2= medium deep scab; 3= deep scab.
The product of the two assessments was used to calculated a scab index having a maximum value of 100.
***Scab index = Severity of symptoms x Extension of symptoms x 100/27.

Nematode survival. When symptomatic tubers, which first had been stored at 4 °C for 20 weeks, were transferred to pots with sterile sand, and the potato plants allowed to develop, new cultures of *P. penetrans* were established. Examination at harvest of soil, roots, stolons, tubers and cross-lesions confirmed the presence of *P. penetrans*.

DISCUSSION

Although the lesion nematodes *P. penetrans*, *P. crenatus* Loof and *P. fallax* Seinhorst are common in cultivated fields in Norway, the relationship between lesion nematodes and the potato crop has not been studied in detail. In the literature, more than ten species of lesion nematodes have been reported from potato (Thorne, 1961; Koen, 1965; Loof, 1978; Brodie *et al.*, 1993).

The field symptoms noted by us in Grue have also been observed in other potato fields in eastern Norway (Skien, Solør), and the symptoms correspond well with previous reports (Brodie *et al.*, 1993). In Norway, commercial fumigants, organophosphates or carbamate nematicides have not been used since the early 1970s. One of the most efficient methods of controlling lesion nematodes is bare fallow, but this is difficult to achieve in a country with a small acreage devoted to agriculture. These factors might have allowed lesion nematodes to reach damaging levels in recent years.

Published information shows that *P. penetrans* can reduce yields of potato by 30 to 70% (Thorne, 1961; Bernard and Laughlin, 1976; Olthof, 1989; Lazarovits *et al.*, 1991; Philis, 1995). Also, the economic threshold of potato to *P. penetrans* is reported to be of 100-250 specimens per 250 g soil (Oostenbrink, 1966; Brodie *et al.*, 1993). Despite the fact that we do not have exact data on potato yield, or information on the initial population densities of the nematodes, our observations nevertheless suggest a damage threshold of potato cv. Saturna to *P. penetrans* of about 100 individuals/250 g soil. The yield reduction of 50% observed by us is of the same magnitude as reported in the literature.

The lesion nematode species *P. brachyurus* (Godfred) Filipjev *et* Schuurmans Stekhoven, *P. scribneri* Steiner in Sherbakoff *et* Stanley, *P. neglectus* (Rensch) Filipjev *et* Schuurmans Stekhoven, and *P. penetrans* have previously been reported from potato tubers (Koen, 1965; Brodie *et al.*, 1993). Here we report on the close association of *P. penetrans* with cross-lesions on the tuber skin. This extends the range of lesion nematode symptoms beyond those of superficial small lesions and pustules reported earlier (Thorne, 1961; Koen, 1965; Brodie *et al.*, 1993).

The similarity of cross-lesions caused by *P. penetrans* to symptoms caused by the common scab bacterium, *S. scabies*, is obvious. As demonstrated in the greenhouse experiment, *P. penetrans* alone may significantly increase the extension and severity of "scab-like" symptoms on tubers, and similar trends were observed when *P. penetrans* and *S. scabies* were inoculated together. The occurrence of a low frequency of symptoms also in the control treatment may relate to the fact that skin-cracks in potato may be caused by internal pressures during tuber growth (Stevenson *et al.*, 2001). Skin cracks have also been reported to occur in the control treatments of previous tests with common scab (Bjor and Roer, 1980). Our observations suggest that both organisms are capable of increasing the frequency of cross-lesions on tubers.

The high incidence of scab symptoms observed in the field was unexpected since the potato cv. Saturna has a relatively high tolerance against common scab. In resistance tests, potato cv. Saturna has a grade of 6 on a scale of 0 to 9, where 9 is the highest degree of resistance against common scab (Førsund, 1992). Similar scab symptoms, in connection with lesion nematode infections, have been observed on potato tubers of cv. Oleva, which also is relatively tolerant and classified as grade 4 (Møllerhagen and Nybråten, 2007). It cannot be excluded that nematode-induced symptoms *per se* or interactions between both organisms may have contributed to the surprisingly high scab frequency recorded in the field.

The fact that the symptoms induced by the lesion nematode *P. penetrans* may be mistaken for symptoms of common scab, *S. scabies*, has important implications for scab control. This relates in particular to recommended maintenance of high soil moisture at and during weeks 4-9 after tuber set (Stevenson *et al.*, 2001). If symptoms are related to nematode infection rather than to the scab bacterium, this recommendation would allow for a rapid build-up of lesion nematodes, which would in turn result in more symptoms and a decrease in marketability of the tubers.

It was reported by Koen (1965) that the "potato peel nematode" (*P. brachyurus*) could spread with infested tubers. We demonstrated that this is also true for *P. penetrans*, and that this species can survive normal Norwegian storage procedures for potatoes. Survival of *P. penetrans* in stored potato tubers for 19 weeks at +7 °C has been reported from Ontario, Canada, (Olthof and Wolynetz, 1991), while storage for 20 or more weeks at +5 °C caused complete mortality of *P. brachyurus* (Koen and Hogewind, 1967). Hence, species seem to differ in their ability to survive in storage.

In conclusion, the lesion nematode *P. penetrans* may cause considerable yield loss in the potato crop. The bacterium *S. scabies*, together with *P. penetrans*, may have an additive effect in the expression of symptoms of common scab. Tuber symptoms induced by the nematode may easily be confused with symptoms of *S. scabies*. Mistaking nematode symptoms for scab, and as routine control keeping soil moisture high, may aggravate nematode damage. The occurrence of *P. penetrans* in tubers and its survival in storage needs to be considered in the production of seed potato tubers.

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