Infestation by the cyst forming nematode *Globodera pallida* of potato tubers in Southern Italy

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Abstract

A case of heavy infestation on tubers of three potato (Solanum tuberosum L) cultivars (Nicola, Spunta and Sieglinde) by the cyst nematode Globodera pallida Stone, 1973 was observed in Southern Italy on early producing potato cultivars in spring. Tubers were covered by white females and cysts but no other external symptoms were detectible on their surfaces. Detailed observations were directed to study the response of phellem and secondary cortex tissues induced by the expansion of syncytia during nematode feeding activity. The micro details of histological changes observed on serially sectioned infected tissues are described and illustrated. The nematode's feeding activity was confined to the outer part of the tuber while the host-parasite relationships were similar to those induced by the same nematode in the feeder roots.

Introduction

Potato (Solanum tuberosum L.) is one of the world's most important food crops and suffers an annual 10-12% loss due to plant parasitic nematode infection (Sasser and Frekman, 1986). Many plant parasitic nematode species are widely distributed throughout the world potato producing areas. The main genera of importance are Ditylenchus, Globodera, Meloidogyne, Nacobbus, Pratylenchus, Paratrichodorus and Trichodorus. Among them, potato cyst nematodes (PCN), Globodera rostochiensis (Woll.) and G. pallida Stone, are the most economically important throughout temperate regions of the word (Brodie et al., 1993). Both PCN species occur separately or together in most of the major potato producing areas of Italy, limiting the acreage available for this crop (Ambrogioni, 1977; Greco et al., 1982).

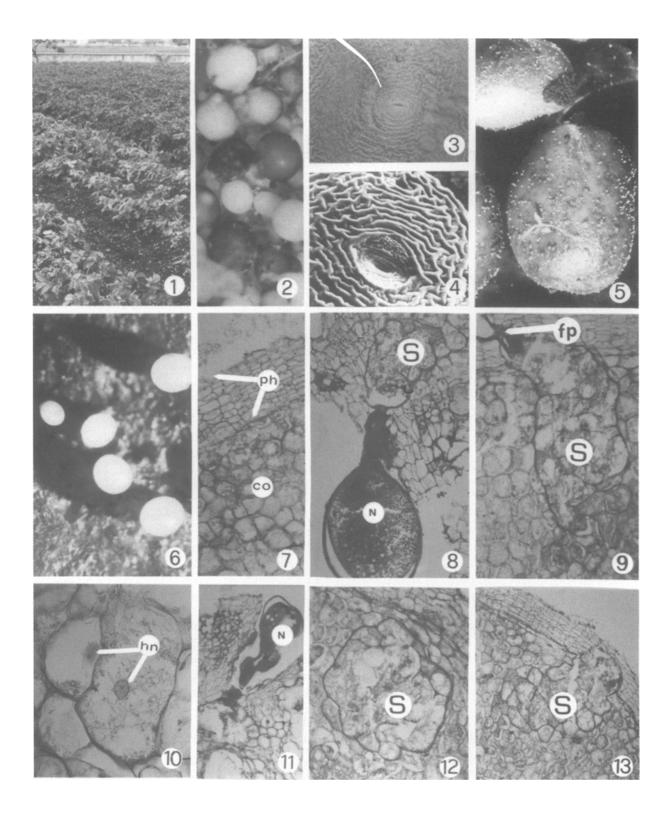
Not only roots but also tubers may be infected by PCN species and many records can be found in the literature. Sasser (1971), Inagaki (1974) and Jensen et al. (1979) provided micrographs showing tubers infected by *G. rostochiensis*, while Siggeirsson and Quigley (1983) noticed heavy losses of potato yield,

in Iceland, when tubers grew in warm soil heavily infested by this nematode species. Numerous other observations of PCN infecting potato tubers have been made by Prof. L. Miller (personal communication) in Bolivia, Mexico, Perù, New Zealand etc., but in all cases no taxonomic studies were made to elucidate which PCN species was involved in the tuber parasitism

In May 1994 and 1995, heavily infested potato tubers (Nicola, Spunta and Sienglende) were noticed near Bari, Southern Italy, grown in fields heavily infested with *G. pallida*. The tubers were covered by white females an a few cysts (Figure 5). Because of the uncommon and heavy infestation, detailed histological investigations were conducted on the anatomical alterations induced by the nematode on potato tuber phellem and secondary cortex tissues.

Materials and methods

Potato plants, tubers, soil and root samples were collected 30 days before harvesting to confirm nematode identity, determine population densities and



describe epigeal symptoms on infected potato plants. Although all cultivars (Sieglinde, Nicola and Spunta) were found to be infected by Globodera pallida only cv. Nicola tubers were selected for histopathological observations. Portions ($5 \times 8 \times 5$ mm) of the outer part of infected tubers, or sufficiently small newly formed tubers to obtain whole-piece serial sections, were fixed in FAC (formaldehyde-acetic acid-chromic acid) solution, dehydrated in a tertiary butyl alcohol series (50-100%) and embedded in 58 °C m.p. histowax. Embedded material was sectioned transversely and longitudinally with a rotary microtome and the sections mounted on glass slides for staining with safranin and fast green according to Johansen's (1940) method. Syncytia induced by G. pallida on potato tubers were compared with those induced by the nematode on the adventitious potato roots.

Observations

Nematode identification. Detailed morphometric observations based on larval body length, stylet length, shape of stylet knobs, female colour and perineal pattern configuration (Figures 3 and 4) agreed well with those given by Stone (1973), for *G. pallida*.

Field symptoms. In the field under observation the heavy infestation of G. pallida caused stunted growth, poor root development and early potato plant death (Figures 1 and 2). Nematode population levels (measured as root infection rates and soil infestation 30 days before harvest were 230–280 specimens (including all life stages)/g of root tissue and 70–80 eggs and juveniles/cm of soil. At harvest time (May) almost all tubers (Figures 5 and 6) were completely covered by numerous white females but only a few cysts (ratio 20:1); no other external symptoms were detected on them. Only 25% of the immature females present on tuber surfaces became dark brown cysts when infected potato tubers were stored at 22–24 °C and 70% R.H. in the dark.

Histopathology. Histological examination of sectioned healthy (Figure 7) and nematode-infected potato tuber portions (Figures 8 and 13) showed that the host parasite relationships are similar to those observed in adventitious feeder roots. Nematode feeding sites in tubers consisted in the expansion of large syncytia formations 14–20 cell layers deep. Juvenile stages, white females and cysts always fed at the outer extremity of the syncytial formation, with their bodies protruding from the tuber surface, although nematode penetration (Figure 11) occurred in some cases when the nematode fed on very deep syncytia in the secondary cortex.

Syncytia induced by *G. pallida* on potato tubers probably originate from phellem cells (disposed in 8–10 cell layers), when infective juveniles insert their stylets into the initial syncytium cell. Syncytium formation and expansion, in fact, occurred on the periphery of sectioned tubers, reaching a length of up to 200–780 μ m and involving phellem and secondary cortex tissues (Figures 8, 9, 12, 13). Wall dissolution of the cells involved occurs in well developed active syncytia (Figures 9, 12, 13), while thickened cell walls, granulated cytoplasm and hypertrophied nuclei characterize the syncytia cells (Figure 10).

Remarks

Tuber infection by G. pallida is uncommon and probably it does not interfere with tuber development especially in early potato cropping. Yield losses are attributed to root infections but tuber infections, may be a signal of heavy soil nematode-population levels that require serious sanitation measures in order to prevent spread of nematode infections. Bio-ecological investigations and confirmation of pathotype identity are required to provide the information necessary for proper management of the nematode.

Figures 1–13. (1) Aboveground symptoms of infected potato cv. Nicola plants, caused by a large G. pallida population density. Note the early senescence of plants; (2) Mature females and cysts of G. pallida extracted from the soil 30 days before harvesting of potato tubers; (Figures 3 and 4) Perineal patterns of G. pallida as seen by light and scanning electron microscopy; (5) Potato tubers cv. Sieglinde covered with G. pallida females; (6) Enlarged portion of (Figure 5); (7) Transverse section of the outer part of a healthy tuber showing the eight-cell layer of phellem or cork (ph) and a portion of secondary cortex (co); (Figures 8–13) Multinucleate syncytia (S) formed by incorporation of a number of cells adjacent to the feeding cell at the feeding point (fp), following cell wall dissolution, which begins with and involves phellem and/or secondary cortex cells (Figures 8, 9, 12, 13). Note that the nematode (N) feeds from outside the tuber surface (Figure 8) or partially penetrated into several cell layers (Figure 11). Hypertrophied nuclei (hn) of the syncytial cells in the granulated cytoplasm are arrowed in Figure 10.

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