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Short note

A new culturing method for the rice white tip nematode, *Aphelenchoides besseyi* Christie, 1942, on carrot discs

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The rice white tip nematode, *Aphelenchoides besseyi*, first described by Christie (1942), belongs to the Class Aphelenchida and family Aphelenchoididae. It was discovered by Kakuta in 1915 (Van Nieuwenhuyzen, 1977; Mc Gawley *et al.*, 1984). According to Franklin and Siddiqi (1972), the species is a synonym of the pest *Aphelenchoides oryzae* Yokoo, 1948.

Aphelenchoides besseyi has been reported from USA, Africa, Bangladesh, El Salvador, Indonesia, India, Italy, Korea, Cuba, Hungary, Pakistan, Taiwan and Russia (Franklin and Siddiqi, 1972). In Turkey it was first reported in 1995 in Ipsala (Edirne) and Gonen (Balikesir) (Ozturk and Enneli, 1997). Typical symptoms of this nematode are found only on the aboveground parts of the rice plant and include 3 to 5 cm length of the tip of the leaves turning white, the panicle leaf often becoming twisted and curled, and a shorter panicle that is often atrophied at the tips (Figure 1a, b, c).

Genetic resistance offers one of the best control methods for *A. besseyi*; however, laboratory screening for resistance requires mass rearing of the nematode. Multiplication of *A. besseyi* was achieved using carrot discs in the laboratory, which has been reported by Moody *et al.* (1973) to work successfully with the root lesion nematodes *Pratylenchus vulnus*. Using this method, an average of 10,000 *A.besseyi* individuals can be obtained from a carrot culture in approximately 4 months.

The protocol used to produce these cultures is a modification of the published method by Moody *et al.* (1973) and requires newly harvested fresh, healthy carrots. These were peeled in a laminar flow cabinet using a sterile peeler which had been flamed with a Bunsen burner. The peeled carrots were then surface sterilized by placing them in an ethyl alcohol solution (95%) for 5 min. The alcohol solution was then drained and the carrots are flamed with a Bunsen burner. The carrots were then cut into pieces 1 cm thick, and 2-3 pieces were placed in 9 cm diameter Petri dishes of. *Aphelenchoides besseyi* individuals were collected from rice panicles using a modified Baermann funnel method. The nematodes were surface sterilized using streptomycin sulphate (0.1%) for 10 min, then one female was transferred onto a single carrot disc (average diameter 3-4 cm) in a Petri dish, under sterile conditions in a laminar flow chamber using a dissecting microscope at ×40 magnification. The nematodes were added within 2 h after preparing the carrots. The Petri dishes were sealed tightly with paraffin tape to prevent contamination and placed in an incubator at 22±1 °C. Harvesting cultures was done as required by slicing the carrot into thin pieces (approx. 1 mm) and placing them in a Petri dishes containing sterile distilled water. The nematodes migrated from the carrot into into the water over a period of 3-4 h. The nematodes were concentrated by passing the water solution through a 20 µm sieve. The experiment was replicated more than 30 times, and in most cases, after a period of four months, up to 10,000 *A. besseyi* per Petri dish could be obtained. Obtaining nematodes in a short

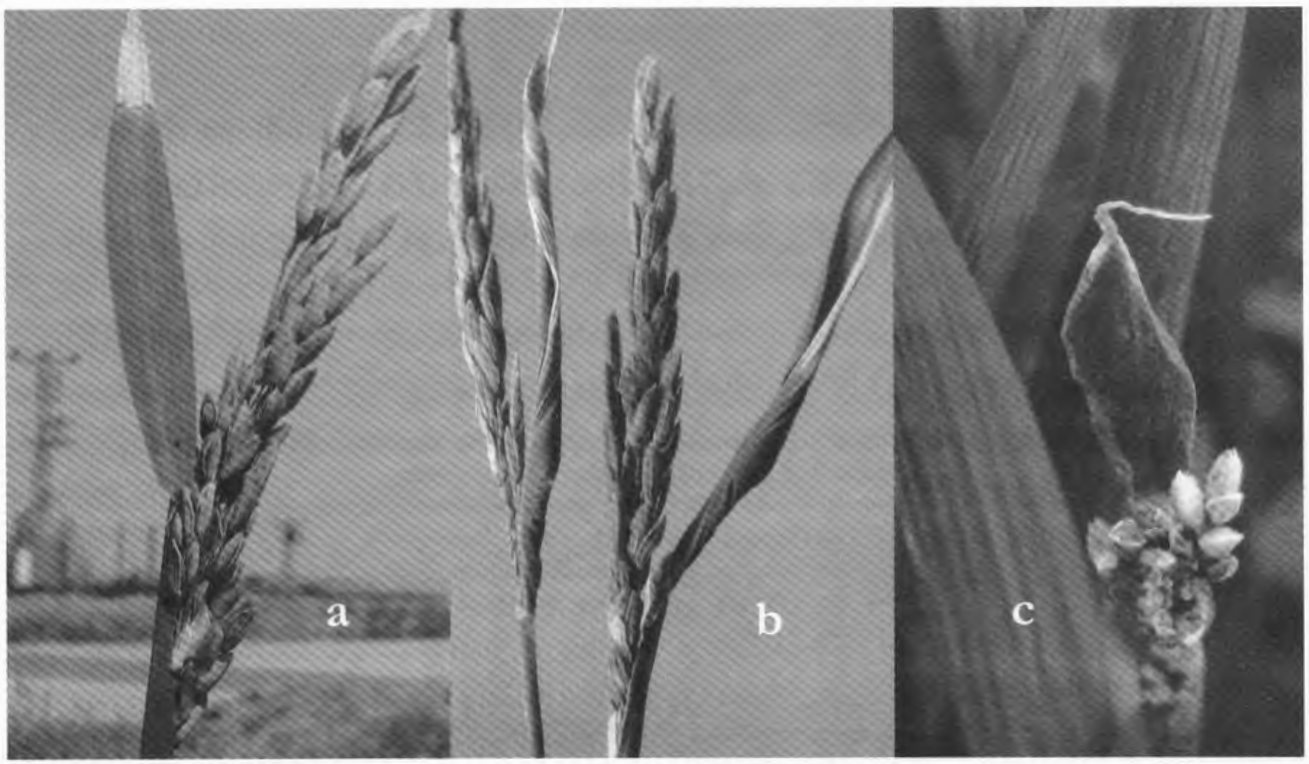

Fig. 1. *Aphelenchoides besseyi* damage on leaves of rice, a: flag leaf tip is white for a distance of 3 - 5 cm; b: the panicle leaves are severely affected, often twisted and curled; c: the panicle is shorter and often atrophied at the tips.

time and in desired amounts by this method will facilitate research on plant reactions against nematodes.

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REFERENCES

FRANKLIN, M.T. AND SIDDIQI, M.R. 1972. *Aphelenchoides besseyi. C.I.H.Description of Plant Parasitic Nematodes.* Set 1, No. 4.

MC GAWLEY, E.C., RUSH, M.C. AND HOLLIS, J.P. 1984. Occrrence of *Aphelenchoides besseyi* in Louisiana rice seed and its interaction with *Sclerotium oryzae* in selected cultivars, *Journal of Nematology*, 16 (1): 65-68.

MOODY, E.H., LOWNSBERY, B.F., AHMED, J.H., 1973. Culture of root lesion nematode *Pratylenchus vulnus* on carrot discs. *Journal of Nematology*. 5: 226-255

OZTURK, G. AND ENELI, S. 1997. Determination of *Aphelenchoides besseyi* Christie (Aphelenchida: Aphelenchoidiae) (the white tip nematode) harmful on rice for the first time in Turkey. *Turkish Journal of Entomology* 21:129-132.

VAN NIEUWENHUYZEN, A.A. 1977. Nematode diseases of rice, Plant Protection for the Rice Crop, West Africa Rice Development Association, 240-245.