

# Morpho-anatomical Changes in *Gladiolus* due to Infection by *Aphelenchoides besseyi* from West Bengal, India

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## ABSTRACT

A critical study on the morpho-anatomical changes in the leaves of *Gladiolus* sp. consequent to infection by *Aphelenchoides besseyi* Christie, a nematode, was carried out. *Gladiolus* sp. cv. 'Red Majesty' was found naturally infected by *A. besseyi* during January, 2009 at the University Central Research Farm, West Bengal, India. Elongated, vein-bounded, light brown to deep brown streaks developed on the leaf and progressed both ways but never reached the leaf tips. Streaks become necrotic and black, subsequently dried up gradually. A huge number of nematodes was isolated from the infected leaves of *Gladiolus*; they were identified as *A. besseyi*. Morpho-anatomical changes of the foliar nematode-infected leaves comprised of disintegrations of epidermal tissues followed by formation of vacuoles of diverse shapes and sizes. Stomata as well as disintegrated epidermal tissues might be a suitable route for *A. besseyi* to gain access into the internal leaf tissues of *Gladiolus*. The present finding of foliar nematode infection through stomata and disintegrated epidermal tissues may be helpful in disease management of *Gladiolus*.

**Keywords:** foliar nematode, histopathology, symptomatology

## INTRODUCTION

There are records of infestation of *Aphelenchoides besseyi* Christie in ornamental plants all over the world. Approximately 44 plant species have so far been identified as hosts of *A. besseyi*, of which 19 species e.g. *Cattleya*, *Chrysanthemum morifolium*, *C. maximum*, *Cymbidium*, *Dahlia variabilis*, *Dendrobium nobile*, *Ficus elastica*, *Hibiscus brachenridgii*, *Hydrangea macrophylla*, *Impatiens balsamina*, *Oncidium*, *Phalaenopsis*, *Polianthes tuberosa*, *Saintpaulia ionantha*, *Tagetes* sp., *Torenia fournieri*, *Tithonia diversifolia*, *Vanda* sp. and *Zinnia elegans* belong to ornamental groups (Fortuner and Williams 1975; Uchida and Sipes 1998). In India, rice (Dastur 1936), tuberose (Chakraborti and Ghosh 1993), *Setaria italica* (Lal and Mathur 1988) and *Panicum melacium* (Gokte *et al.* 1990) were recorded as hosts of foliar nematode, *A. besseyi*. Excluding tuberose there is no record of any ornamental acting as host of *A. besseyi* in India. Surprisingly, *Gladiolus* sp. was found as an alternative host of *A. besseyi* showing all the symptomatology like foliar nematode disease of tuberose. West Bengal is an important flower growing state of India where 2120 million spikes plus 52 thousand tonnes of loose flowers were produced from 21.07 thousand ha of land during 2008-09 (Anon 2010). *Gladiolus* occupies 11% of the total flower growing area of the state. Therefore, *Gladiolus* assumes an important position among floricultural crops of the state. Any setback in *Gladiolus* cultivation due to such disease infestation may cause huge loss to the growers. Usually farmers are not well aware of this foliar nematode disease and their symptoms and control. Disease caused by them if remains undiagnosed for several years, losses can be increased. Therefore, the present experiment on the infestation of *A. besseyi* in *Gladiolus* was undertaken with a view to understanding the symptom and disease development in the plant due to nematode attack, mode of entry and nature of feeding of the nematode in the plant and the morpho-anatomical changes in the leaf tissues associated with nematode attack.

## MATERIALS AND METHODS

*Gladiolus* sp. cv. 'Red Majesty' was found infected in nature by the foliar nematode at the Central Research Farm of BCKV, Nadia, West Bengal. Infected plants were brought to the laboratory; infected leaves were cut into pieces by scissors, placed in water and incubate for 24 hours. Nematodes migrate out of the leaves when the lesions were soaked in water. Nematodes were identified as *A. besseyi* Christie by the first author following taxonomic characters mentioned by Franklin and Siddiqi (1972). The disease symptom caused by *A. besseyi* was studied in detail under field conditions. Transverse sections of the leaves of *Gladiolus* sp. were passed through different grades of alcohol (Jiangsu Huaxi International Trade Co. Ltd.) starting from 30 to 100% and stained with 50% saffranin (E. Merck India Ltd.) and 95% light green (Loba Chemie Pvt. Ltd.) following a standard anatomical staining procedure. Finally, the sections were kept in clove oil (E. Merck) and xylene (E. Merck) mixture (1: 1, v/v) for 1 min for cleaning and mounted on glass slides in a drop of DPX (E. Merck) medium. The slides were placed on a hot-plate overnight or until the DPX solution fully dried up.

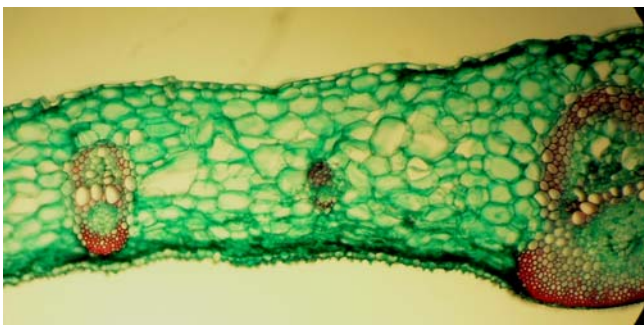
## RESULTS AND DISCUSSION

### Symptomatology

Leaf symptom of infected *Gladiolus* begin with elongated, vein-restricted, light brown to deep brown streak that become necrotic and black subsequently (Figs. 1A, 1B). Symptoms of foliar nematode infection typically appear as vein-delimited streaks due to their inability to cross the major leaf veins once inside the plant (Chizhov *et al.* 2006). Symptoms appeared at the vegetative stage of the plant mostly on the basal and mid portion of the leaf and progressed both ways but never reaching the apex. Infected leaves later on die and abscise. Floral stalk of the infected plant become shortened with less number of florets as compared to healthier ones (Fig. 1C). Corms of the infected plants are undersized and shriveled (Fig. 1D). There was



**Fig. 1** Symptoms of foliar nematode infection in *Gladiolus*. (A) Elongated, vein-restricted, brown to black necrotic streak; (B) close-up view of the leaf streak; (C) floral stalk of infected plant bears less number of florets; (D) corms of the infected plant are small and shriveled.



**Fig. 2** Transverse section of healthy *Gladiolus* leaf.

hardly any record of infestation of *A. besseyi* on *Gladiolus* though *A. subtenius* on *Gladiolus* has been reported from Iran to cause diseases of corms and pseudostem (Abbas *et al.* 2006). *A. besseyi* feed ectoparasitically on vegetative tissues (Singh and Mathur 2004; Duncan and Moens 2006) but endoparasitic feeding on foliage part was also recorded on some ornamental plants (Volvas *et al.* 2005). During endoparasitic feeding in leaves, foliar nematodes cause brown to black or chlorotic, vein-delimited lesions that may become necrotic (Dunn 1997; Uchida and Sipes 1998).

### Observation on morpho-anatomical changes

Disintegrations of epidermal tissues as well as development of number of vacuoles of different shapes and sizes are formed as a result of nematode infection (Figs. 2, 3). Nematode feeding and movement within the plant causes damage by disintegrating tissues. This damage becomes visible in the form of tissue discolouration, formation of rots and inhibition of plant growth. Detailed morpho-anatomical changes in this crop due to infection by *A. besseyi* are lacking. Easy access to the internal tissues through stomata as well as disintegrated epidermal tissues might be a suitable route for *A. besseyi* in *Gladiolus*, an alternate ornamental host crop other than tuberose in West Bengal. Foliar nematodes, *Aphelenchoides* spp. enter and exit through wounds or natural openings such as stomatal pores on leaves. The method of entry varies among host plants (Wallace 1959; Strumpel 1967).

### CONCLUSION

*Gladiolus* sp. has been identified as an alternative host of *A. besseyi* Christie in addition to rice, *P. tuberosa* and two weeds like *S. italica* and *P. melacium* of Poaceae in India. This appears to be the first record of its kind from West



**Fig. 3** Transverse section of *Gladiolus* leaf showing morpho-anatomical changes due to infection by *Aphelenchoides besseyi* 'Christie'.

Bengal. *Gladiolus* growers like tuberose growers have to be equally careful in future about the foliar nematode management.

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