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A SURVEY ON THE OCCURRENCE OF CITRUS NEMATODE (*TYLENCHULUS SEMIPENETRANS*) IN THE PUNJAB.

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A survey of the citrus orchards of the Punjab has revealed that most of the orchards carry heavy infestation of the citrus nematodes (*Tylenchulus semipenetrans*). Among citrus varieties, the local Gadadehi (*Citrus aurantium*) showed maximum infection of 98.4 per cent. In addition, nematodes belonging to the genera of *Pratylenchus*, *Aphelenchus*, *Tylenchorhynchus* and *Xiphinema* were also recovered from the citrus rhizosphere.

INTRODUCTION

Recent studies on the nematodes infesting the roots and rhizosphere of citrus nurseries at Lyallpur (Pakistan), by Ashraf (1969) revealed that the species of *Aphelenchus*, *Chiloplacus*, *Dorylaimus*, *Eucephalobus*, *Hoplolaimus*, *Mesodorylaimus*, *Mononchus*, *Mylonchus*, *Pratylenchus*, *Tylenchulus* and *Xiphinema* were fairly common around the roots of citrus nursery plants.

Citrus is an important fruit crop of Pakistan, only 2nd in area to mango (Khan, 1969). The climate of Pakistan is very suitable for the production of excellent quality citrus fruit, the demand for which in foreign markets is steadily increasing. It has however, been observed that on an average the economic life of a sweet orange tree does not exceed twenty years under local conditions. Except under very special circumstances the plants, usually start exhibiting symptoms of slow decline at this age. Prior to studies made by Ashraf (1969) the disease was attributed to *Colletotrichum gloeosporioides*, the cause of wither tip disease of citrus. The disease was also attributed to soil conditions, such as the presence of salts and calcium nodules, water logging etc. It is generally believed that *Colletotrichum gloeosporioides*, is a weak parasite and requires predisposition of citrus plants to adverse soil conditions before it can be attacked by the fungus. The presence of plant parasitic nematodes like *Pratylenchus* sp., *Tylenchorhynchus* sp., *Tylenchulus* sp. and *Xiphinema* sp. raised a question whether these nematodes also contributed towards the decline of citrus orchards. Preliminary studies by Ashraf (1969) proved that they cause damage to the young citrus seedlings. The citrus nematode is already reported as an

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important root pest of citrus in California by Baines *et al* (1959) and Van Gundy and Tsao (1953). The citrus nematode alone is reported to cause an annual loss ranging between 10-30 per cent of total citrus fruits produced in California.

In the present studies an attempt was made to determine overall incidence of the citrus nematode in the citrus growing areas of the Punjab province and also to find out its relation with the roots of different varieties of citrus root stocks.

Under local conditions, 'Khatti' or rough lemon (*C. limon*), is the most popular root stock used for propagating sweet orange, grape fruit and Kinnow (*C. reticulata*) varieties of citrus fruits. 'Mitha' or sweet lime (*C. aurantium*) and 'Kaghizi' lime or sour lime (*C. aurantifolia*) are propagated either from cuttings or seeds.

MATERIALS AND METHODS

Roots and soil samples of mature and young citrus plants were obtained from orchards situated in the vicinity of Lyallpur, Sargodha, Bhalwal, Qadratabad, Mandi Bah-ud-Din, Gujranwala and Sahiwal. The samples were brought to Lyallpur in cellophane bags as quickly as possible and stored at 10–15°C. An effort was made to analyze the samples as early as possible. The nematodes were isolated from soil by Cobb's sifting and gravity method. The roots were individually examined under the stereo-binocular and the number of roots infested with mature females of the citrus nematode were counted. Identification of other nematodes as well as of the larvae of the citrus nematode was made by preparing temporary mounts. Permanent slides of good specimens of each nematode were also prepared according to Thorne's method (1961). Some difficulty was encountered in dislodging the mature females of the citrus nematode from the citrus roots. In the case of freshly collected specimens of roots, the neck of the female was so firmly embeded into the root tissue, that it invariably broke when it was dislodged. The problem was solved by keeping infested roots in water at Laboratory temperature (23—35°C) for 24—36 hours. The storage of such roots probably causes disintegration of the root tissues thereby loosening their grip on the female's necks. It was also observed that the egg masses closely adheared to the posterior ends of the females in the case of freshly collected specimens. When the roots were kept soaked in water overnight the egg masses could also be easily separated from the female bodies. An attempt was made to collect samples of roots of as many citrus varieties as possible.

Most of the varieties of root stocks were obtained from the citrus root-stock experiment station, Sahiwal. The material comprised 8 varieties of *Citrus limon* L; 5 varieties of *C. reticulata* Blanco; 3 of *C. medica*; 3 of *C. aurantifolia*; 5 of *C. aurantium* L; and one each of *C. karna*; *C. sinensis* Osbeck and *C. paradisi* Macf.

RESULTS

Stereoscopic examination of root samples collected from 7 different places revealed that the infestation due to the citrus nematode ranged from 26.6 to 80.6 per cent. Maximum infestation was found on citrus roots collected from Sahiwal.

Examination of roots of different root-stock varieties of citrus revealed that certain varieties were free from the citrus nematode, for example 'Galgal' (*Citrus medica*); 'Mithee' (*C. limon*); 'Baramasi' (*C. reticulata*); 'Chakotra' (*C. grandis*) and grape fruit (*C. paradisi*). Among the remaining varieties of citrus root stocks, 'Jatti Khatti', 'Jullundari Khatti', 'Narangi Shahdarah', 'Gadadehi' & 'Kaghzi' lime showed more than 90 per cent of roots infested with the citrus nematode. It may be mentioned here that a root piece of 'Gadadehi', one centimeter long, carried 12 mature females on it and a similar piece of 'Jatti Khatti' root carried a maximum number of 8 mature females. Both are very susceptible to the attack of the citrus nematode. Results of infestation percentage are given in Table I below :—

TABLE I. *Per cent infestation of roots of different varieties of citrus root-stocks with citrus nematode*

	Root stock variety	Range of Infestation (per cent)
1.	Macroptera, Lemon fine, Citron of commerce, 'Jullundari' and 'Jatti Khatti', 'Narangi Shahdarah', 'Gadadehi' and 'Kaghzi' lime.	83.71—98.41
2.	'Jambeeri' Ceylone, Mandarine, Sweet lime and Lemon Ceylone	70.7 —74.44
3.	Trifoliate orange, Cloepetra, Sweet orange, 'Tarujabba' Sour orange and Attoni.	30.54—54.10
4.	Eureka lemon, 'Jambeeri' Lyallpur, 'Karna' sour orange, 'Mokri', 'Mithotra', European lemon and Seville kimb	2.47—19.19
5.	Pomello, Pummello, 'Baramasi', 'Mithee and 'Galgal'.	0

The results show that most of the popular citrus root stocks such as 'Jatti khatti' and 'Jullundari khatti' are very susceptible to the attack of citrus nematode. The only root stock which could replace these root stocks is 'Jamberi' Lyallpur which showed only 6.09 per cent infestation. All other root stocks are either of only academic interest or too susceptible to the attack of citrus nematode.

In the USA, almost entire citrus industry is based on sour orange root stocks which under the local conditions has excelled all other varieties of root-stocks including the rough lemon. But the sweet oranges budded on sour orange have also fallen a prey to the notorious quick-decline or Tristeza disease in the USA. In Pakistan, the use of sour orange, (*C. aurantium*) is extremely limited on account of comparatively drier climate, and to some extent due to lack of appreciation for fine quality oranges. Since the virus causing quick-decline becomes ineffective when the sweet orange is budded onto rough lemon stock, the disease has not been reported from Pakistan so far. Sour orange as a root stock used in the experimental garden at Sahiwal has shown only 2.96 per cent attack of the citrus nematode. This could easily replace rough lemon in Sahiwal area but for the fear of introduction of quick decline disease into Pakistan, as the insect vectors of the Tristeza virus are already there.

Regarding the distribution of the citrus nematode in the Punjab, the results are given in Table 2.

TABLE 2. *Prevelence (Per cent roots infested) of the citrus nematode at various places of the Punjab province*

1.	Lyallpur	64.4 Per cent
2.	Sahiwal	80.6 ,,
3.	Sargodha	28.3 ,,
4.	Bhalwal	27.3 ,,
5.	Mandi Bah-ud-Din	25.6 ,,
6.	Qudrat-abad	41.0 ,,
7.	Gujranwala	40.8 ,,

The results show that the maximum infestation due to the citrus nematode occurs round about Sahiwal and the minimum in Sargodha region. This fact generally reflects in the conditions of citrus orchards at both the places.

DISCUSSION

The survey undertaken to determine the prevelence of the citrus nematode in the citrus growing areas of the Punjab province has revealed that almost one hundred per cent citrus orchards were infested with the nematode. The number of nematodes per root length was more in Sahiwal area than in Sargodha area. This corroborates with the general growth conditions of the citrus trees in these areas, Sargodha being the highest producer of quality fruits in the whole of the Punjab.

The most common root-stock used for the propagation of both sweet orange and kinnow mandarines is rough lemon (*C. limon*) which is one of the most susceptible root-stocks. This is probably the cause of wide spread distribution of the citrus nematode in the Punjab. Slow decline or the wither-tip disease of citrus is of common occurrence in the Punjab, particularly in Lyallpur, Sahiwal and down South. This situation can also be attributed to the use of rough lemon as the most popular root-stock used for the propagation of citrus fruits. Among various varieties of rough lemon. 'Jambeeri' Lyallpur appears to be somewhat resistant to the attack of the citrus nematode. This could probably replace the ordinary 'Jatti Khatti' or 'Jullundari Khatti' (*C. limon*) varieties as a promising resistant root-stock. This needs to be further investigated.

The presence of other nematodes such as *Aphelenchus, Pratylenchus, Hoplolaimus, Tylenchorhynchus & Xiphinema* in the rhizosphere of citrus plants also points out towards the possibility of their being harmful to the roots of citrus plants. Christie (1953) has already proved that the ecto-parasitic nematodes were as harmful to the plants as endoparasitic ones. The above mentioned nematodes are some of the major plant parasitic nematodes. They may also be contributing towards the incidence of the slow decline of citrus in the Punjab. Feldmesser & Hannon (1965) reported that *Pratylenchus* sp. was associated with the decline of citrus in USA. They have reported 2.5% reduction in citrus growth and yield. Malo (1969) has reported *Hoplolaimus* sp. and *Xiphinema* sp. in the rhizosphere of citrus trees in central Florida. Further studies on the role played by these nematodes in the causation of the slow decline of citrus in the Punjab is imperative. Khan (1969) has already expressed the views that stringent nursery laws need to be promulgated to produce high quality disease free citrus nursery plants. Unless the citrus nematode is controlled in the citrus nurseries, the disease will continue to play havoc in the orchards.

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