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HISTOPATHOGENESIS OF SUSCEPTIBLE AND RESISTANT PEACH ROOTSTOCKS INFECTED WITH THE ROOT-KNOT NEMATODE*, Meloidogyne incognita.*

Farahat, A.A.*;H.I.El-Naggar*;H.Hendyand Ahlam El-Ghonaimy****
*** Fac. of Agric., Cairo University**
**** Desert Research Center, Cairo**

ABSTRACT

The histological changes resulted from the infection with *Meloidogyne incognita* in root tissues of the susceptible (Meetghamr) and the resistant (Florida) peach rootstocks were studied. In the susceptible rootstock, clusters of numerous typical multinucleated giant cells entirely filled with dense cytoplasm and highly thickened cell walls and relatively larger than those in the resistant rootstock were observed. However, in such resistant rootstock (Florida), although some penetrated stages succeeded to feed normally forming typical giant cells , vacuolated giant cells of moderate thickened cell walls , necrotic areas, hyperplastic cells and, cavities were more frequently observed. Withal, numerous penetrated stages failed to develop to reach maturity.

Keywords: Histopathology, peach, *Meloidogyne incognita*.

ITRODUCTION

Since the middle of the last century , peach rootstcks, Nemaguard, Nemared, Okinawa and Florida are known all over the world to be resistant to the root –knot nematodes *M. incognita*, *M. javanica*, *M. arenaria* (Sharpe, 1958, Sharpe *et al.*, 1969 & Brooks and Olmo, 1961). Histopathogenesis of such nematodes in different plant hosts was comprehensively studied. The changes resulted from nematode infection in root tissues are mainly the formation of enlarged multinucleated cells with dense cytoplasm and thick cell walls or the so called giant cells which considered the feeding sites of the root-knot nematodes. The number, size and shape, number and size of nuclei, cytoplasm densing, cell wall thickening; and the site at which they are formed varied according to many factors, *i.e.* plant cultivar, host suitability, nematode species and/or races. Hypertrophied, hyperplastic cells of parenchyma, necrotic areas of collapsed and compressed cells around nematodes, syncytia formation, the disruption and disorganization of vascular cylender, all were noticed in infected root tissues of different plants (Osman, 1968; Huang, 1969; Siddiqui, 1971; Hendy, 1979; Kheir and Farahat, 1981; Sosa-Moss *et al.*, 1983; Sudha and Prabhoo, 1983; Hasan and Jain, 1985; Acosta *et al*., 1986; Orion *et al.*, 1987; Yahia *et al*., 1994; Bilqees and Jabeen, 1994; Pedrosa *et al.*, 1996; Mostafa, 2001 and Saeed, 2005).

Variations in histological responses of susceptible and resistant hosts were also documented by Sosa-Moss *et al*., (1983) whom found that females of *M*. *arenaria* reached maturity and produced eggs in root tissues of resistant tobacco, but considerable necrosis was induced. *M. hapla* and *M. javanica* developed on resistant and susceptible cultivars but there were necrotic lesions at some infection sites in the resistant cultivars. Fewer giant cells, smaller in size and with fewer nuclei were formed in resistant lines (Singh *et al*., 1984). They also observed dead cells (hypersensitive reaction) around

infecting root-knot nematode larvae occurred in roots of resistant lines. Typical multinucleated cells limited to the vascular tissue in roots of resistant maize and several mature females and egg-masses of *M. javanica* were observed by Asmus *et al* (2000).

The present study was carried out to find the histopathological changes resulted from the infection with *M. incognita* in root tissues of resistant and susceptible peach rootstocks.

MATERIALS AND METHODS

One year old seedlings of susceptible (Meetghamr) and resistant (Florida) rootstocks of uniform size were transplanted singly in 30 cm-diameter clay pots filled with sterilized sandy loam soil. After adaptation, seedlings were inoculated with 5000 J2 of the root-knot nematode, *Meloidogyne incognita* by pouring the inoculum in holes around the root system and immediately covered with wetted sandy soil. After three months, plants were taken off and infected portions (small galls) of each rootstock were selected and fixed in formaline acetic acid alcohol (FAA). They were then dehydrated in graded series of a mixture of ethanol and butanol alcohols, infiltrated and embedded in paraffin wax. The processed materials were sectioned by a rotary microtome then stained with safranine and fast green according to the procedures of Sass (1964). Microscopic examinations were made with binocular microscope, using an oil immersion lens as needed.

RESULTS

The sectioned galls were microscopically examined and showed that, in both the susceptible and resistant rootstocks, the root-knot nematode , *M. incognita* was able to form typical multinucleated giant cells limited in both the cortical and vascular tissues (Fig 1 a-d).

In the susceptible rootstock (Meetghamr), numerous functional multinucleated giant cells of normal appearance and different size were formed in both cortex and stellar regions. The giant cells are always in clusters and dark stained cytoplasm entirely filled the cells which are clearly thickened walls (Fig 2 a-d). The number of the giant cells in the close vicinity to the female varied between 5 to more than 15 (Fig. 2 d). The number of nuclei per giant cell was variable and synsctia were not surrounded with necrosis in the majority of cases.

In Florida (the resistant rootstock) root tissues, the juveniles which were able to penetrate the root, were able in many instances to form the typical giant cells and develop to mature females however, others failed to form their feeding sites and accordingly degenerated (Fig.3 a-d). Many other females although they were able to form few small giant cells in the cortical layer, they were not able to develop to mature females and they were of abnormal appearance (Fig, 3 e,f).

F1

1

F2

F3

3

3

Even those females that succeeded to form typical giant cells, many of these cells were non-functional of vacuolated cytoplasm and small numbers of dark stained nuclei (Fig. 3 g,h). Necrotic areas were of common occurrence beside the giant cells (Fig 3 g) or surrounding the females (Fig. 3 d,I). Cavities were also clearly observed in the infected tissues of resistant rootstock and hyperplastic cells as well (Fig. 1b, 3g,h).

In general, giant cells in the resistant rootstock were somewhat smaller than those in the susceptible one with fewer numbers of nuclei and the thickening of cell walls was not as much as that in the susceptible rootstock. As well, such giant cells may be scattered (Fig. 3j) or in clusters (Fig. 3h) in Florida root tissue, however, they were always in clusters in the susceptible rootstock, Meetghamr.

DISCUSSION

Results indicated that *Meloidogyne incognita* induced typical giant cells in the susceptible and resistant peach rootstocks. Likewise, Sosa-Moss *et al*., (1983) stated that different root-knot nematode species reached maturity in tissues of resistant tobacco cultivar, even though noticed necrotic lesions were notcied. Also, Asmus *et al.* (2000) observed typical multinucleated cells limited to the vascular tissues of resistant maize with even several mature females and egg-masses of *M. javanica..*

The obtained results emphasized that both rootstocks are hosts of the root-knot nematode, *M. incognita*. Yet, the distinct variations between both of them in their reaction to nematode infection illustrated the differences in their suitability to nematode reproduction. The failure of some penetrated stages in producing giant cells or even producing abnormal ones resulted in their failure to develop to mature females in tissues of Florida. Malo (1967) said that the resistance of Nemaguard and Okinawa peach rootstocks is based on walling-off the giant cells followed by their breakdown. The disappearance of giant cells, which are the main source of food to the nematode growth and reproduction, eventually leads to their death.

Some other histological characteristics of the resistant peach rootstock are the occurrence of vacuolated non-functional giant cells, necrosis and cavities as well as the moderate thickening of cell walls. All are signs of host resistance and indicators of the success of such rootstock (Florida) in hindering reproduction of the root-knot nematode, *M. incognita* (El-Ghonaimy *et al*, 2005).Likewise, Sano and Nakasono (1986) observed that *M. incognita* giant cells in the non-host plants *Macroptilium atropureum* and *Crotolaria spectabilis* were similar to those in tomato roots but they noticed vacuolated cytoplasm and moderate thickening of cell walls in *Crotolaria*.

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التغيرات التشريحية في جذور أصناف الخوخ المقاومة والقابلة للإصابة بنيماتودا تعقد الجذور

أحمد فرحات* ، حسن النجار* ، حسن هندي **، أحلام الغنيمي**

*** كلية الزراعة – جامعة القاهرة**

**** مركز بحوث الصحراء**

تم في هذا البحث دراسة التغيرات التشريحية التي تنتج عن الإصابة بنيماتودا تعقد الجذور وذلك في صنفي الخوخ الحساس للإصابة (ميت غمر) والصنف المقاوم (فلوريدا) بغرض التعرف علي طبيعة المقاومة لهذه الآفة في أشجار الخوخ وأوضحت الدراسة ما يلي:

استطاعت معظم اليرقات التي اخترقت جذور الصنف الحساس التغذية بصورة طبيعية مكونة الخلايا العملاقة التي كانت دائماً أكبر حجماً وأكثر عددا وجدرها أكثر تغلظاً وتحتوي على سيتوبلازم أكثر كثافة يملأ كل الخلية وبه عدد أكبر من الأنوية من تلك التي لوحظت في الصنف المقاوم . أما في الصنف المقاوم فقد استطاعت بعض اليرقات فقط أن تتغذى بصورة طبيعية مكونة خلايا عملاقة أصغر حجما وأقل عددا وجدرها أقل تغلظاً وبها سيتوبلازم أقل كثافة وربما كانت متناثرة وليست في تجمعات كما في الصنف الحساس. أيضا لوحظ في أنسجة الصنف المقاوم وجود الخلايا الميتة المحيطة بالخلايا العملاقة ووجود الفراغات والإناث الغير قادرة على إتمام نموها الى إناث بالغة. أوضحت الدراسة علي العموم وجود اختلافات تشريحية واضحة بين كل من الصنف الحساس والصنف المقاوم لنيماتودا تعقد الجذور.