Jordan Journal of Agricultural Sciences, Volume 1, No. 1, 2005

Current Distribution of the Root-Knot Nematodes (*Meloidogyne Species and Races*) in Jordan

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ABSTRACT

Eighty-three samples of galled roots and soil were collected from infected vegetable crops and fruit trees during the period from May 2002 to August 2003. The survey covered most of the irrigated agricultural areas of Jordan namely: Southern Ghors (south of the Dead Sea), Jordan Valley, elevated uplands, and eastern desert plains. Meteorological data, host crop, cropping method, and irrigation system were recorded for the sites covered by the survey. Soil samples were analyzed for soil texture, pH, Electrical Conductivity (EC), and soil organic matter content. Nearly 84% of the collected populations were identified as *M. javanica*, 6% as *M. incognita* race 1, 4% as *M. incognita* race 2, and 6% as *M. arenaria* race 2. *Meloidogyne javanica* was distributed throughout all the investigated districts. *Meloidogyne incognita* race 1 was found in the northern part of the Jordan Valley (Dier Alla) and the eastern desert plain (Qwerah), while *M. incognita* race 2 was found in the northern part of Jordan Valley (Al-Qarin) and the elevated upland (Al-Qastall). *Meloidogyne arenaria* race 2 was found only in a limited area in the mid Jordan Valley (Dier Alla), and this is the first report on its occurrence in Jordan. Results showed that *M. javanica* had several degrees (C°) higher range than those of *M. incognita* and *M. arenaria*. However, rainfall and soil parameters indicated little direct effect on the distribution of *Meloidogyne* species in Jordan.

KEYWORDS: Ecology, Meloidogyne arenaria, incognita, Meloidogyne javanica, survey.

INTRODUCTION

Root-knot Nematodes (RKNs), *Meloidogyne* spp., are the most economically important group of plant-parasitic nematodes worldwide, attacking nearly every crop grown. These nematodes inflict great losses to various vegetable crops and fruit trees (Sasser and Freckman, 1987). It is very difficult to assess yield losses due to RKNs alone, but there is no doubt that hundreds of millions of dollars are lost annually and about 5% of the total universal crop production is destroyed by *Meloidogyne* species (Sasser *et al*., 1983; Barker *et al*., 1985; Sasser, 1987).

To date, over 70 species of *Meloidogyne* have been described, three of which are extremely polyphagous apomictic species: *M. incognita* (Kofoid and White) Chitwood, *M. javanica* (Treub.) Chitwood , and *M. arenaria* (Neal) *Chitwood* that are distributed worldwide and account for the majority of crop losses due to RKNs (Nickle, 1991).

In Jordan, two *Meloidogyne* species: *M. javanica* and *M. incognita* were identified from soil and plant samples collected from the major irrigated areas of Jordan. *Meloidogyne javanica* was found to occur in all major areas of the Jordan Rift Valley and in the elevated and eastern area, whereas *M. incognita* race 1 (Abu-Gharbieh, 1982a) and *M. incognita* race 2 (Atieh, 1986) were encountered only in the Northern part of the Jordan Valley and in the elevated areas.

No further surveys in the last two decades were made to study the occurrence and distribution of RKNs in the

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Received on 5/4/2005 and Accepted forPublication on 18/10/2005.

This Research was financially supported by the Deanship of Research, University of Jordan.

agricultural areas of Jordan. In addition to the previously reported *Meloidogyne* species and races in Jordan, possibilities exist that other species or races were not discovered by the earlier surveys or had been introduced into Jordan. The objective of this work was to study distribution of the most common *Meloidogyne* species and races in Jordan, and possible environmental factors involved.

MATERIALS AND METHODS

Collection of samples and preparation of pure nematode cultures

During the period from May 2002 to August 2003, eighty-three representative samples of plant roots and their rhizospheric soil were collected from vegetable crops and fruit trees that were found infected with RKNs. The survey covered most of the irrigated agricultural areas of Jordan that were divided into four districts varying in weather and soil factors, namely Southern Ghors (south of the Dead Sea), the Jordan Valley, elevated uplands, and eastern desert plains.

Most populations were collected from the open fields and plastic houses. Some, however, were collected from fruit tree orchards, and one sample was collected from a home garden. All fields were supplemented with drip irrigation except for one field which was under furrow irrigation.

Galled roots were washed thoroughly under tap water for 5 minutes. Small galled roots were excised and examined under a dissecting microscope for the developmental stages of the RKNs.

Large, white to light brown, egg-masses filled with eggs were selected from all crops sampled to make single egg-mass population cultures. From each galled root sample, ten egg-masses were individually picked under a dissecting microscope by forceps. The egg-masses were then transferred into tap water in an examination dish, surface-sterilized with 0.5% sodium hypochlorite (NaOCl) solution for 2 minutes, washed twice in tap water and maintained in 1 ml tap water. The egg-masses were then transferred singly, inoculated into susceptible tomato plants cv. GS 12 that were maintained under greenhouse conditions. A total of 83 *Meloidogyne* populations, representing each investigated area, were selected based on a preliminary morphological screening.

Population Identification

Identification of the 83 *Meloidogyne* populations was based on the perineal pattern morphology in addition to the North Carolina (NC) differential host test (Hartman and Sasser, 1985) and a DNA fingerprinting assay (Zijlstra *et al*., 2000) for the identification of the four major *Meloidogyne* spp.

Determination of Environmental Factors

Meteorological data were collected from all main meteorological stations in Jordan for the last 2-5 years (1999-2004). The collected meteorological data included annual air temperatures (maximum, minimum, and mean), annual absolute air temperature (maximum and minimum), number of days with minimum air temperature $\leq$ 0°C, number of days with maximum air temperature $\geq$ 35.0°C, annual average precipitation, and elevation of the station (data are not shown).

Other data including crop type, cropping method, and irrigation system were recorded for each site during the survey. The soil samples were analyzed for soil type, pH, Electrical Conductivity (EC), and soil organic matter content (Rowell, 1994).

RESULTS

Three *Meloidogyne* species were identified: *M. javanica*, *M. incognita*, and *M. arenaria* (Table 1). All *Meloidogyne* populations were composed of single species except for one mixed population composed of both *M. javanica* and *M. incognita* race 2 collected from Al-Qastall area. Different developmental stages of the RKN were observed as a disease sign associated with root galling. Various stages of *Meloidogyne* were excised from roots including females, eggmasses, and 2nd stage juveniles. Root-knot nematode populations were successfully isolated from many vegetable crops

including tomato, cucumber, eggplant, cowpea, bean, squash, peas, and garlic and from several fruit trees including banana, fig, grape, palm, apricot, and mulberry. All populations were maintained on susceptible tomato plants cultivar 'GS12'. Inoculation with the various populations resulted in the root gall formation characteristic of *Meloidogyne* species.

Table (1): Distribution of *Meloidogyne* species and races in different districts of Jordan during May 2002 to August 2003.

District	% Occurrence [1]			
	M. javanica(70) (2)	M. incognita Race 1(5)	M. incognita Race 2(3)	M. arenaria Race 2(5)
Southern Ghors	22.9	0.0	0.0	0.0
Southern Part of Jordan Valley	41.4	0.0	0.0	100
Northern Part of Jordan Valley	12.9	40.0	66.7	0.0
Elevated Uplands	21.4	0.0	33.3	0.0
Eastern Desert Plains	1.4	60.0	0.0	0.0
All districts	84.3	6.0	3.6	6.0

(1) % Occurrence = number of populations in a district/ total number of populations x100.

(2) Number of collected populations.

All samples collected from the Jordan Valley, southern Ghors, elevated uplands, and eastern desert plains were found RKN-infected. *Meloidogyne* species were found widely distributed throughout the irrigated agricultural areas and in climatically diverse and distant districts in Jordan (Table 1). Most of the RKN-infected samples were collected from the Jordan Valley, some from the elevated uplands including Jerash, Baqa'a, Amman, Ma'daba, and Al-Karak, and few from the eastern desert plains of Jordan including Sokhna and Qwerah.

Meloidogyne species and races were found distributed in all investigated areas with predominance of *M. javanica (84%). Meloidogyne javanica* was widely distributed in all districts including southern Ghors, Jordan Valley, the elevated uplands and eastern desert plains (Table1). Among the seventy *M. javanica* populations detected, nearly 65% were found in the subtropical areas of the southern Ghors and southern the Jordan Valley. However, *M. incognita* was completely absent in these areas. This species was found in the relatively cooler climates in the northern Jordan Valley, elevated uplands and eastern desert plains. *Meloidogyne incognita* race 1 was found in the central part of the Jordan Valley (Dier Alla location) and eastern desert plains (Qwerah location), while *M. incognita* race 2 was found in the northern part of the Jordan Valley (Al-Qarin location) and elevated uplands (Al-Qastall location). *Meloidogyne arenaria* race 2 was recovered only from one location (Dier Alla area) in the central part of the Jordan Valley (Table 1).

All *Meloidogyne* populations were collected from regions with an average annual air temperature in the range of 15 to 25˚C (data not shown), with an absolute annual minimum air temperature in the range of approximately –5 to 7˚C and absolute annual maximum air temperature in the range of approximately 37.0 to 48.5˚C. The number of days with minimum air temperature ($\leq$ 0˚C) ranged between 0 to 12 days while the number of days with maximum air temperature ($\geq$ 35˚C) ranged between 4 to 60 days in the investigated areas. Also, total precipitation in these areas ranged between 75 to 450 mm annually and the number of rainy days ranged between 20 to 80 days. The average annual relative humidity ranged between 50 to 69%.

Soil textures in the investigated areas ranged from loamy sand to clay soil. All soil samples harboring *M. arenaria* and *M. incognita* were sandy clay loam; except for one case with clay soil. Soil pH values of the collected samples ranged from 7.04 to 8.3. Soil EC values

ranged from 0.27 to 5.43 mmhos/cm. Soil organic matter contents ranged from 0.67 to 5.39%

DISCUSSION

The present study revealed that three of the four most common species of *Meloidogyne* (*M. javanica*, *M. incognita*, and *M. arenaria*) occurred in Jordan; with the former (*M. javanica*) being the most common. Two physiological races (race 1 and 2) of *M. incognita* and one physiological race (race 2) of *M. arenaria* were identified. The two races of *M. incognita* were previously reported from Jordan in separate studies; (race 1 by Abu-Gharbieh (1982a) and race 2 by Ateih (1986)). Both *M. javanica* and *M. incognita* were previously reported from Jordan (Abu-Gharbieh and Hammou, 1970) while *M. arenaria* reported in this study is a new record from Jordan. The identification is precise and reliable because it was based on a combination of different diagnostic methods including DNA fingerprinting. The sequence characterized amplified regions-polymerase chain reaction (SCAR-PCR) based assay, was used in this study. It was reliable and consistent in differentiating the four common *Meloidogyne* species worldwide (Zijlstra *et al.*, 2000).

One mixed population composed of both *M. javanica* and *M. incognita* race 2 was collected from Al-Qastall area. Abu-Gharbieh (1982b) also reported similar mixed population of the two species but from Al-Zarqa area.

Although *M. javanica* was found in almost all areas investigated, *M. incognita* and *M. arenaria* were found in the relatively cooler areas of the Jordan Valley (northern part) and elevated uplands. This is in general agreement with earlier findings (Abu-Gharbieh, 1982) that *M. javanica* has 4-5°C higher optimum temperature range than *M. incognita* and also with findings in the USA where it was found that the northern limits of continued existence of *M. incognita* and *M. arenaria*, is a few hundred miles farther north of the limits for *M. javanica*. (Taylor and Sasser, 1978). *Meloidogyne incognita* was also found in one area (Qwerah location) of the eastern desert plain despite the relatively hot weather conditions. The microclimate provided by evergreen orchard plantation and the regular irrigation at Qwerah location may be a possible explanation for the survival of *M. incognita* populations under such hot summer conditions.

Rainfall alone does not appear to influence dominance of the three detected *Meloidogyne* species in their respective areas, partly due to regular irrigation. In the southern Ghors, the Jordan Valley, Sokhna, and Qwerah, the annual precipitation is less than 250 mm. However, possibilities are that *M. javanica* has the edge over *M. incognita* and *M. arenaria* in survival through dual adverse conditions of very high summer temperatures, long dry summer, and very low rainfall.

Soil pH may have little direct effect on *Meloidogyne* populations activities where in most cases it ranged between 7- 8.5 in various areas. The pH was in the range favorable for plant growth thus for the nematode. Also, soil EC ranged from approximately 1-5 mmhos /cm in most cases.

In conclusion, *M. javanica* is the most widely distributed species while *M. incognita* and *M. arenaria* were restricted to the relatively cooler agricultural areas in Jordan. *M. javanica* has around 4-5°C higher optimum temperature range than *M. incognita* and *M. arenaria*. Rainfall alone does not appear to influence dominance of the three *Meloidogyne* species in the respective areas. However, *M. javanica* can survive dual adverse conditions of very high summer temperatures and very low rainfall more than *M. incognita* and *M. arenaria.* Under the Jordanian conditions, soil type, pH, EC, and organic matter contents may have had little direct effect on the distribution of *Meloidogyne* species and races in the various areas investigated in Jordan.

Since *M. arenaria* is confined to one small area in the central part of the Jordan Valley, an eradication or confinement campaign must be considered. Quarantine services are recommended to prevent the introduction of new *Meloidogyne* species or races into Jordan.

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توزيع أنواع و سلالات نيماتودا تعقد الجذور (*Meloidogyne* species and races) في الأردن

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ملخـص

تم إجراء حصر لنيماتودا تعقد الجذور (*Meloidogyne* spp.) في الأردن، خلال الفترة من أيار 2002 إلى آب 2003م. تضمن الحصر جمع ثلاثة و ثمانين مجتمعاً للنيماتودا من محاصيل خضر وأشجار فاكهة متعددة في مقاطعات ومناطق مروية متعددة ومتنوعة مناخياً في الأردن شملت: وادي الأردن والأغوار الجنوبية والمناطق المرتفعة والمناطق الصحراوية الشرقية. تم تسجيل العوامل البيئية التالية: المعلومات المناخية، نوع المحصول، طريقة الزراعة، ونوع نظام الري لكل موقع شمله الحصر. وكذلك تم اجراء تحليل لعينات التربة المأخوذة من منطقة الجذور المصابة بنيماتودا تعقد الجذور لتحديد نوع التربة، درجة حموضتها، درجة توصـيلها الكهربائي، ومحتـواها من المادة العـضوية. تم تـعريف سبعين مجتمعاً لنيماتودا تعقد الجذور تتـبع النوع *M. javanica* الذي تبين أنه واسع الانتشار، وخمسة تتبع النوع *M. incognita* (السـلالة 1) ، وثلاثة تتبع النوع *M. incognita* (السلالة 2)، و خمسة تتبع النوع *M. arenaria* (السلالة 2). تم تسجيل النوع *M. arenaria* لأول مرة في الأردن في هذه الدراسة. ووجد النـوع *M. incognita* (السلالة1) في الجزء الشمالي من وادي الأردن وفي المنطقة الصحراوية الشرقية (موقع القويرة)، ووجـد النوع *M. incognita* (السلالة2) في الجزء الشمالي من وادي الأردن وفي المناطق المرتفعة (موقع القسطل)، بينما وجد النوع *M. arenaria* (السلالة2) في الجزء الأوسط من وادي الأردن (موقع دير علا) فقط. كذلك أظهرت النتائج أن للنوع *M. javanica* 4-5 درجات أعلى في مدى الحرارة المثلى للنوعين *M. incognita* و *M. arenaria*. من ناحية أخرى، يبدو أن لكل من كمية الأمطار ونوع التربة ودرجة حموضتها ودرجة توصيلها الكهربائي ونسبة المادة العضوية فيها، أثراً ضئيلاً مباشراً على توزيع هذه الأنواع من *Meloidogyne* في الأردن.

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