Incidence, and Identification of Three Root-Knot Nematode species Occurring in the Medicinal Herbs

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약용식물의 뿌리혹선충 발생과 분류동정

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ABSTRACTS: Soil and root samples were collected from the rhizosphere of 11 different medicinal plants to determine the incidence, density and identification of root-knot nematode species associated with medicinal herbs. About 55% of medicinal herbs examined was found to be infested with root-knot nematodes. As a result of infection caused by three root-knot nematodes, M. hapla recorded 43.3% in medicinal herbs whereas M. incognita and M. arenaria showed 7.9% and 3.7%, respectively. Forsythia koreana, Hemerocalis fulva, Hibuscus mutabilis and Petasites japonicus were the most severely infested herbs whereas Acanthopanax sessiliflorus was least infested. Population of the second stage larvae ranges from 65 to 540/300 ml soil and older plant had higher number of nematodes than younger plants. Meloidogyne hapla, M. incognita and M. arenaria were the species associated with the medicinal herbs. The most abundant nematode observed in medicinal herbs was M. hapla and followed by M. incognita and M. arenaria. M. arenaria was observed firstly on Ficus carica, one of medicinal plant.

Key words: identification, incidence, medicinal herbs, root-knot nematodes.

Medicinal herbs have always been important and are used directly or indirectly to mankind. In old days, these herbs have been collected from mountains and forests. Nowadays, the cultivation of medicinal herbs started with the advancement of agricultural techniques. In the recent years, cultivation of medicinal herbs has been increased to meet the requirements of pharmaceutical and cosmetic industries and also to earn foreign exchange by the means of exporting these commodities. Literature indicates that plant parasitic nematodes causes heavy losses of medicinal herbs (2, 5). Root-knot nematodes, Meloidogyne spp. infect almost all cultivated plants throughout the world (9). It is reported that Meloidogyne spp. have been considered as one of the most pervasive, damaging pest and major constraint for the cultivation of medicinal herbs in India (5). Medicinal herbs are known to be attacked by all the four major and common species of the root-knot nematodes, namely Meloidogyne incognita (Kofoid & White) Chitwood, M. javanica (Treub) Chitwood, M. arenaria (Neal) Chitwood and M. hapla

Chitwood in different parts of the world (7). Some preliminiray investigations indicated that *Meloidogyne hapla* and *M. incognita* are commonly associated with medicinal herbs in Korea (2, 3). However, detailed data on the incidence and density of root-knot disease and identification of species and their pattern of distribution in different regions have not been examined. Therefore, the present study was undertaken to determine the incidence and density of root-knot diseases and identify the species of root-knot nematodes associated with medicinal herbs.

MATERIALS AND METHODS

Soil samples along with roots were collected from the rhizosphere of 11 different species of medicinal plants in April, 1998 cultivated at Uisong Medicinal Plant Experiment Station (MPES) (Table 1). The herbs examined in the present study were planted in 1996 except Forsythia koreana, Hemirocalis fulva and Petasitis japonicus which were planted in 1993 and Hibiscus mutabilis in 1994. All 240 composite soil and root samples were collected within from a depth of 20 cm and 15~25

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samples were collected from each species of medicinal herb. Each sample consisted of 4~6 soil cores. Soil samples collected from same field were thoroughly mixed together. The second stage larvae and males of root-knot nematodes were isolated from 300 ml soil sub-sample by suspending them in water in a shallow tray passing through nested sieves (100 and 400 mesh), and finally separating from the residue collected on 400 mesh sieve by centrifugal sugar-floatation technique (1). The second stage larvae were counted under stereoscopic microscope of 50X magnifications, using counting dishes. Some specimens of each stage of root-knot nematode were fixed in formalin glycerin (F:G) 4:1 solution, dehydrated by Seinhorst's rapid method (6) and mounted on

slides in anhydrous glycerin for microscopic observations.

The root samples were washed thoroughly with tap water and observed for the presence of galls and eggmasses. Number of galls and egg-masses were counted in each root system. Gall index (GI) and egg mass index (EMI) were rated on 0~5 scales (8). *Meloidogyne* species were identified on the basis of perenial pattern characteristics of mature females (4) and morphological characters of females and males (Table 2).

RESULTS AND DISCUSSION

About 55% of samples collected from different species of medicinal plants at UiSong MPES (Table 1) was

Table 1. Occurrence and density of Meloidogyne spp. in medicinal herbs

TT 1 .	Korean name	No. of samples examined	% of infestation with Meloidogyne spp.				Severity index	
Herb species			M. hapla	M. incognita	M. arenaria	Total	Root gall	Egg mass
Acanthopanax sessiliflorus	오가피	20	35 (150)	_	_	35	1-3	0-2
Allium senescens	울릉부추	25	52 (172)	_	_	52	2-3	1-3
Aracus dioicus	눈개승마	20	55 (160)	_	_	55	1-3	1-2
Aralia elata	두릅나무	25	52 (203)	_	_	52	1-3	1-2
Ficus carica	무화과	15		_	60 (287)	60	2-4	2-5
Forsythia koreana	개나리	25	68 (283)		<u> </u>	68	3-5	2-5
Hemerocalis fulva	왕원추리	20	65 (263)	_	-	65	2-5	2-4
Hibiscus mutabilis	부용	25	60 (227)	_	_	60	2-5	2-4
Liriope graminiflolia	맥문동	20	40 (130)	_	_	40	1-3	1-2
Mentha arvensis	박하	25		56 (211)	_	56	2-5	2-4
Petasites japonicus	머위	20	40 (278)	25 (245)	_	65	3-5	2-5
Total		240	43.3	7.9	3.7	55.8		

¹Numbers in parenthesis are density of second stage larvae/ 300ml soil.

Table 2. Characteristics used for the identification of Meloidogyne species

Sexual stage	Characters	Root-knot nematodes						
	Characters	M. hapla	M. incognita	M. arenaria				
Females	Perenial pattern	Dorsal arch flattened. Lateral lines distinct, striae smooth to wavy. Tail terminal area marked by punctations.	Dorsal arch high, composed of smooth to wavy striae. Some striae forked near the lateral lines.	Dorsal arch flattened to rounded. The striae in the arch slightly indented at the lateral lines and form a shoulder on the arch.				
	Stylet	Small compred to other two species. The conus of stylet slightly curved dorsally. Knobs rounded and distinctly set off	Stylet cone distinctly curved dorsally. Stylet knobs broadly elongate, set off from the shaft and anteriorly indented.	Very robust; shaft increase in width posteriorly and gradually merges with the knobs. The knobs wide and rounded pos-				
Males	Lip region	The lip cap high and much narrower than the lip region. Lip region non-annulated set off from the body.	The lip cap large and rounded, centrally concave. Lip region marked with 2-3 incomplete annulations Lip cap low and sloped pos-	Lip region continuous with body, 2-3 incomplete annulations are present on the lip region.				
	Stylet	Much thinner and shorter than of <i>M. incognita</i> and <i>M. arenaria</i> . Knobs rounded and set off from the shaft.	Stylet tip blunt. Shaft narrows near the knobs. Knobs set off from the shaft, anteriorly indented and broadly elongate to rounded.	Stylet cone is pointed and lumen opening marked on the ventral side by slight prutuberance. Knobs large, anteriorly indented and gradually merge with the shaft.				

 $^{^{2}}$ Root-knot index scale; 0 = no gall; $1 = 1 \sim 2$ galls; $2 = 3 \sim 10$ galls; $3 = 11 \sim 30$ galls; $4 = 31 \sim 100$ galls and 5 = >100 galls per root system.

found to be infested with root-knot nematodes. The highest incidence of root-knot nematode recorded 68% and was observed on Forsythia koreana. The incidence on Hemerocalis fulva, Petasites japonicus, Hibiscus mutabilis were recorded as: 65%, 65% and 60%, respectively. The lowest recorded 35% and was found in Acanthopanax sessiliflorus. Similarly, the severity of the disease also appeared to be high in Forsythia koreana, Hemerocalis fulva, Petasites japonicus and Hibiscus mutabilis as gall index (GI) and egg-mass index (EMI) were in range of 3~5 and 2~5, respectively. In the other plants, GI and EMI were observed in the range of 1~3 and 0~3, respectively (Table 1).

Three species of root-knot nematode such as Meloidogyne hapla, M. incognita and M. arenaria were identified to be associated with different species of medicinal plants at Ui Song MPES (Table 1). These species were identified on the basis of their perenial pattern characteristics and morphology of lip region and stylet (Table 2). As a result of infection caused by three root-knot nematodes, M. hapla recorded 43.3% in medicinal herbs, whereas M. incognita and M. arenaria showed 7.9% and 3.7%, respectively. The population densities of the second stage larvae of Meloigogyne spp. in the rhizosphere of different medicinal plants were obtained in the range of 65~540/300 ml soil. The older paints harboured more nematodes than younger plants. The population differences may be attributed to the period of association of nematodes with plants. Severely infested plants showed the typical symptoms such as yellowing and shedding of leaves, stunted plant growth, reduced root system and large number of galls on roots. Studies for the effect of M. incognita on Mentha spp. showed that increase in initial population of nematodes resulted in decrease of plant growth, chlorophyll contents, vield and reduction in I-menthol (7).

The two species, *M. hapla* and *M. incognita* are mainly responsible for diseases of the medicinal herbs caused by root-knot nematodes in Korea. Since it is the first record that *M. arenaria* is found on a medicinal herbs, occurrence of *M. arenaria* on *Ficus carica* is quite significant. *M. javanica* was not encounted in this area, which is probably due to unsuitable climatic conditions of the area.

The present study showed that most of cultivated medicinal herbs at Ui Song MPES were infested with several species of root-knot nematodes. Medicinal herbs and other susceptible crops in this area may suffered yield losses. Our knowledge on problems caused by

nematodes such as their association, biology, pathogenicity and management is very little.

Therfore it is resonable to solve urgent problems such as the incidence, prevalence, severity and actual damage caused by root-knot nematodes in medicinal herbs in Korea.

요 약

약용식물에 기생하는 뿌리혹선충의 종류와 발생량을 알아보기 위하여 11종의 약초에 대하여 조사한 결과 뿌리혹선충에 기생된 약용식물의 비율은 55%였으며 그중 M. hapla는 43.3%, M. incognita는 7.9%, 그리고 M. arenaria는 3.7%의 비율로 조사한 한식물에 기생하고 있었다. 약초별로 보면 의성개나리, 울릉부추, 무화과와 박하는 감염정도가 심하였고, 오가피는 기생정도가 낮았다. 2령유충의 검출수는 토양 300 ml당 65~540마리였다. 조사된 약초에서 발견된 뿌리혹선충의 좋은 M. hapla가 가장많았으며, 그다음이 M. incognita였으며 M. arenaria는 약용으로 이용하는 식물 중 무화과에서 처음으로 기록되었다.

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