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# Express Pest Risk Analysis:

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**(*Meloidogyne ethiopica* and *Meloidogyne luci)***

# Prepared by:

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The PRA was reviewed and comments provided by the Expert working group for PRA (dr.Gregor Urek, dr.Irena Mavrič Pleško (Agricultural Institute of Slovenia), dr. Sebastjan Radišek (Slovenian Institute of Hop Research and Brewing) and Administration for Food Safety, Veterinary Sector and Plant health (Anita Benko Beloglavec, dr. Alenka Zupančič).

| **Summary1** of the Express Pest Risk Analysis for “***Meloidogyne ethiopica* and *Meloidogyne luci***” | | | |
| --- | --- | --- | --- |
| Root knot nematodes *M. ethiopica* and *M. luci* are serious pests of several agricultural crops belonging to monocotyledons, dicotyledons, including herbaceous and woody plants. All previously reported populations of *M. ethiopica* in Europe and Turkey were reclassified as *M. luci.* In Brazil and Chile, *M. ethiopica* is considered to be a damaging species on kiwi and grapevine, as infestations lead to a reduction of plant growth, fruit size and quality. It is widely distributed in vineyards in Chile. In Brazil, *M. ethiopica* has also been reported to cause multiple galls on potato root systems, as well as protuberances on tubers. Similarly, *M. luci* has been reported on several vegetable crops and has also been shown as a potential threat to the potato production in Portugal.  *M. ethiopica* and *M. luci* can be introduced by several pathways but the highest risk for entry and spread in the PRA area is considered the pathway of host plants for planting with or without soil attached. In addition, plant parts like tubers may present also high risk of spreading in case of pest establishment outdoors. Human assisted spread of the pests is the most important one. The probability of establishment in the protected area is high. *M. luci* has already been detected in the open field production in Europe (Mediterranean climate), in corn and kiwi production in Greece, and potato production in Portugal. There is a high risk of *M. luci* establishment outdoors in the PRA area, as this species was proved to overwinter outdoors at the open field in continental and Mediterranean climate. Once the RKNs have been introduced, it is in general difficult to control or eradicate them, especially outdoors at the open field. On the other hand the ability of *M. ethiopica* to overwinter at the open field in continental and Mediterranean climate has not been tested yet. The endangered area in the PRA area for *M. luci* is therefore: protected production area, vegetable production area, potato production area, corn and other cereals production area, vineyards, kiwi and peach production area. | | | |
| **PRA area:** whole territory of Slovenia | | | |
| **Describe the endangered area:** protected production area, vegetable production area, potato  production area, corn and other cereals production area, vineyards, kiwi and peach production area | | | |
| **Main conclusions**  Root knot nematodes *M. ethiopica* and *M. luci* can be introduced by several pathways:   * High risk for entry in the PRA area: host plants for planting with roots, with or without soil attached (planting material of vegetables, ornamental plants, fruit trees and grapevine), non-host plants for planting with soil attached and in addition, plant parts like tubers may present also high risk of spreading in case of pest establishment outdoors. * Moderate risk: soil and growing medium * Low risk: non-hosts plant parts contaminated with soil (such as roots, tubers, bulbs), for consumption or processing, soil attached to equipment and machinery, travellers   Human assisted spread of the pests is the most important.  *M. ethiopica* and *M. luci* have very wide host range including majority of vegetable crops which are cultivated in the protected area. The probability of establishment in the protected area is high. *M. luci* has already been detected outdoors in Europe (Mediterranean area), in corn and kiwi production in Greece, and potato production in Portugal. There is a high risk of *M. luci* establishment outdoors in the PRA area, as this species was proved to overwinter outdoors in European temperate regions and countries of Mediterranean area. Once the RKNs have been introduced, it is in general difficult to control or eradicate them, especially outdoors. On the other hand the ability of *M. ethiopica* overwintering in the open field in temperate and Mediterranean areas has not been tested yet.  The potential impact of both pest species in the PRA area is high especially in the light of climate change scenarios. The models of climate changes predict an increase of average temperatures and more frequent periods of drought, floods and heat waves. Higher temperatures would allow development of more generations of RKNs in a growing season and consequently higher yield losses. In addition, the findings of virulent populations of *M. luci* in Turkey that can reproduce on tomato plants bearing *Mi*-1 resistance gene adds to the concern about the damage potential of this pest.  Phytosanitary measures to prevent the spread of these pests within the PRA area in case of an outbreak are necessary. The measures should include prohibition of movement of plants for planting originating from infested area, ban of movement of soil form infested area, cleaning and disinfection of facilities, tools and machinery, destruction of nematode infested plant waste, survey, sampling scheme and laboratory testing (within infested area and the surroundings). | | | |
| **Phytosanitary risk for the *endangered area* (***Individual ratings for likelihood of entry and establishment, and for magnitude of spread and impact are provided in the document***)** | High  | Moderate □ | Low □ |
| **Level of uncertainty of assessment**  (see Q 17 for the justification of the rating. Individual ratings of uncertainty of entry, establishment, spread and impact are provided in the document) | High□ | Moderate  | Low □ |
| ***Other recommendations:***   * Inform EPPO or EU * Surveys are recommended to confirm the pests status * Additional research is needed to support phytosanitary decisions (needed research on the damage potential of both species to major crops (early, late potato cultivars, corn etc.); the ability of *M. ethiopica* to overwinter at the open field in continental and Mediterranean climate;   assessment of the efficiency of biological and low risk control/management options) | | | |

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# Stage 1. Initiation

**Reason for performing the PRA:**

*M. luci* is a root knot nematode (RKN) which has recently been discovered in several European countries. Pest specimens, previously found in Europe and Turkey have been identified as *M. ethiopica* but recent studies justified reclassification to *M. luci* (Janssen et al. 2016; Gerič Stare et al. 2017a). RKN *M. luci* was detected for the second time in Slovenia, near Ljubljana in 2015 (Gerič Stare et al. 2017b), while the first found was in 2003. Sister species *M. ethiopica* and *M. luci* were found to be closely related and no differential host has been discovered so far. The pest risk assessment therefore comprises both species, *M. ethiopica* and *M. luci*.

**PRA area:** whole territory of Slovenia

# Stage 2. Pest risk assessment

1. **Taxonomy:**

*Meloidogyne ethiopica* Whitehead, 1968 Domain: Eukaryota

Kingdom: Metazoa Phylum: Nematoda Family: Meloidogynidae Genus: *Meloidogyne*

Species: *Meloidogyne ethiopica*

And

*Meloidogyne luci* Carneiro et al., 2014 Domain: Eukaryota

Kingdom: Metazoa Phylum: Nematoda Family: Meloidogynidae Genus: *Meloidogyne* Species: *Meloidogyne luci*

Common name (of the genus): root knot nematodes (RKN)

# Pests overview

*M. ethiopica* was described by Whitehead in 1968 from the Mlalo region of Tanzania. It has been reported from Africa - Kenya, Ethiopia, Mozambique, Zimbabwe, Tanzania, South Africa (Whitehead 1968, 1969), South America - Chile, Brazil, Peru (Carneiro et al. 2003, 2004), Europe - Slovenia (Širca et al. 2004), Greece (Conceição et al. 2012), and Italy (Maleita et al. 2012) and Asia - Turkey (Aydinli et al. 2013). *M. ethiopica* can parasitize several crops including monocotyledons, dicotyledons, and herbaceous and woody plants (Carneiro et al. 2003, 2004; Lima et al. 2009; O’Bannon 1975; Strajnar et al. 2009, 2010; Whitehead 1968). It is widely distributed in vineyards in Chile (Aballay et al. 2009). In Brazil, *M. ethiopica* affects the root growth, yield and quality of grapevines and kiwi, causing serious crop damage and economic losses (Carneiro et al. 2003). In Chile, *M. ethiopica* was found to be the major root knot nematode parasite of grapevines (*Vitis vinifera*) and other crops (Carneiro et al., 2007). Furthermore, Medina et al. (2014) reported *M. ethiopica* on potato plants from Parana state, Brazil, showing root systems with multiple galls and tubers with protuberances.

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*M. ethiopica* is very difficult to distinguish from the sibling species *M. inornata* and the recently described *M. luci* (Carneiro et al. 2014) due to morphological and morphometrical similarity and intraspecific variation in the group. Besides, the perineal patterns of *M. ethiopica* are highly variable and similar to *M. luci* (Carneiro et al. 2014) and were characterized as varying from the *M. arenaria* type to the “*acrita*” type of *M. incognita* (Golden 1992). Analysing phenotypes of malate dehydrogenase (MDH) and esterase (EST) isozymes has proved to be a useful approach in RKN species identification (Esbenshade and Triantaphyllou 1985; 1990). Isozyme phenotyping is currently the most useful procedure for routine differentiation of *M. ethiopica* from other RKN species as the esterase E3 is species-specific (Carneiro et al. 2004). However, E3 is not the only esterase phenotype of *M. ethiopica*. Mandefro and Dagne (2000) observed phenotype E2 in a *M. ethiopica* population from Africa. In *M. ethiopica* populations reported from Slovenia, Italy, Greece and Turkey, esterase phenotype E3 has also been reported. However, a closer examination showed that all these populations have a three banded esterase pattern where the upper band is slightly lower than the one in *M. ethiopica* populations from Brazil, Chile and Peru. The pattern has recently been recognised as L3 and specific for a newly described species *M. luci* (Carneiro et al. 2014)*.* The molecular approach also confirmed a very close relationship between *M. ethiopica* and *M. luci* and also *M. inornata*. Sequence of COII/lRNA, a region of mtDNA proved to be very useful for analysing the phylogenetic relationship of the very closely related RKN species and populations. Low levels of heteroplasmy in this region allowed the formation of a monophyletic clade for *M. luci* with a clear separation (Geric Stare et al., 2017a). Close relationship between these three species has also been shown on coding mtDNA regions by Janssen et al*.* (2016). Therefore all previously reported populations of *M. ethiopica* in Europe and Turkey were reclassified as *M. luci* (Geric Stare et al., 2017a).

Both *M. ethiopica* and *M. luci* cause severe damage to many host plants by affecting the development of the root systems which are distorted by multiple small and large galls. As a consequence, the water supply to the shoots is impaired. Affected plants can also show above ground symptoms such as stunting and wilting. Based on crop damage and economic losses that *M. ethiopica* has been causing on grapevines and kiwi (Carneiro et al. 2003, 2007) *M. ethiopica* has been included on the EPPO Alert list of harmful organisms in 2011. *M. ethiopica* is also reported to affect potato production (Medina et al. 2014) and *M. luci* has been shown as a potential threat to the potato production in Europe (Maleita et al*.* 2016). Further, *M. luci* has been reported on several vegetables, flowers and fruits species (Carneiro et al. 2014). In addition, the potential of *M. ethiopica* to survive outdoors under a continental climate even in areas where soil temperatures fall below zero during winter, as well as under a sub- Mediterranean climate (Strajnar et al. 2011) now applies to *M. luci* as the identity of Slovene *M. ethiopica* population has been reclassified into *M. luci* (Janssen et al. 2016; Geric Stare et al., 2017a). This indicates that *M. luci* could establish and spread in the PRA area.

In addition, the findings of virulent populations of *M. luci* in Turkey that can reproduce on tomato plants bearing *Mi*-1 resistance gene (Aydinli & Mennan, 2016) adds to the concern about the damage potential of this pest. The *Mi*-1 resistance gene has been introgressed into many commercial tomato cultivars to control the root knot nematode species *M. incognita, M. javanica* and *M. arenaria*. It has also been shown that *Mi*-1 prevents the reproduction of *M. luci* (Strajnar & Širca, 2011; Conceição et al., 2012). *Mi*-1 tomato cultivars are widely used to control root knot nematode species including *M. luci.* However, the finding of *M. luci* virulent populations may present serious threat to vegetable production as there are very limited efficient management options available.

As both *M. ethiopica* and *M. luci* can damage a large number of economically important crops, the EPPO Secretariat decided to include both species in the EPPO Alert List.

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**3. Is the pest a vector? Yes** ☐ **No** 

**4. Is a vector needed for pest entry or spread? Yes** ☐ **No** 

# Regulatory status of the pest

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*Meloidogyne ethiopica*: EPPO Alert List pest since 2011, no other status

*Meloidogyne luci*: EPPO Alert List pest since 2017, no other status

# Distribution

*Meloidogyne ethiopica* distribution

| ***Continent*** | ***Distribution*** *(list countries, or provide a general indication , e.g. present in West Africa)* | ***Provide comments on the pest status in the different countries where it occurs*** *(e.g. widespread, native, introduced….)* | ***Reference*** |
| --- | --- | --- | --- |
| *Africa* | Ethiopia, Kenya  ,Mozambique, South Africa, Tanzania, Zimbabwe | Native, Present, no details | Whitehead, 1969;  Mandefro & Dagne, 2000 Carneiro et al., 2004; |
| *America* | Brazil, Chile, Peru | Brazil (present in 5 provinces); Chile (widespread: area of 1000 km distance, causing economic damage) | Carneiro et al., 2003; Gomes et  al., 2005;  Carneiro et al., 2007;  Somavilla et al., 2011;  Murga- Gutierrez et al., 2012; |
| *Asia* | Absent | Reclassified in *M. luci* | Gerič Stare et al. 2017a; |
| *Europe* | Absent | Reclassified in *M. luci* | Gerič Stare et al. 2017a; |
| *Oceania* | Absent | No records |  |

*Meloidogyne luci* distribution

| ***Continent*** | ***Distribution*** *(list countries, or provide a general indication , e.g. present in West Africa)* | ***Provide comments on the pest status in the different countries where it occurs*** *(e.g. widespread, native, introduced….)* | ***Reference*** |
| --- | --- | --- | --- |
| *Africa* | Absent | No records |  |
| *America* | Brazil, Chile | Brazil (Present, few occurrences) Chile (Present, one occurrence) | Carneiro et al., 2014; Belle et al., 2016a; |
| *Asia* | Iran, Turkey | Iran (Present, no details)  Turkey (widespread in Samsun province) | Aydinli et al., 2013; Gerič Stare et al.  2017a; |
| *Europe* | Greece, Italy, Portugal, Slovenia | Greece (Present, few occurrences) Italy (Present, one occurrence) Portugal (Present, one occurrence) | Sirca et al., 2004; Strajnar  et al., 2009; Conceição et al., 2012;  Maleita et al. 2012; Maleita |

| ***Continent*** | ***Distribution*** *(list countries, or provide a general indication , e.g. present in West Africa)* | ***Provide comments on the pest status in the different countries where it occurs*** *(e.g. widespread, native, introduced….)* | ***Reference*** |
| --- | --- | --- | --- |
|  |  | Slovenia (Present, two occurrences; first occurrence eradicated; second occurrence under eradication) | et al. 2016; Gerič Stare et al. 2017b; |
| *Oceania* | Absent | No records |  |

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# Host plants /habitats\* and their distribution in the PRA area

There is no dependable differential host plant between *M. ethiopica* and *M. luci* known so far. It seems that these two species are so closely related that there is no biological difference between them in terms of parasitizing different plant species. Host species lists for *M. ethiopica* and *M. luci* were prepared based on recorded infestation in the field and tests of host status for each species in literature.

In assessing whether the host is a major or minor crop two factors has been taken into account, the importance of crop in agricultural production and host suitability of plant to the root knot nematode

*M. ethiopica* or *M. luci* (e.g. soybean is considered to be a major crop, since the agricultural area with soybean is increasing each year and the crop plants are a very good host for *M. luci* and *M. ethiopica*).

Host species list for *Meloidogyne ethiopica*

| **Host Scientific name (common name)**  ***/ habitats\**** | **Presence in PRA area (Yes/No)** | **Comments (e.g. total area, major/minor crop in the PRA area, *major/minor habitats*\*)** | ***Reference*** |
| --- | --- | --- | --- |
| *Acacia mearnsii*  (Black wattle) | No | - | Whitehead 1968,1969 |
| *Actinidia deliciosa*  (Kiwi) | Yes | Minor crop for PRA area /  Major crop for some PRA area regions | Carneiro et al. 2003, 2004 |
| *Agave sisalana*  (Sisal) | No | - | O’Bannon 1975. |
| *Ageratum conyzoides*  (Billygoat-weed) | No | - | O’Bannon 1975. |
| *Arachis hypogaea*  (Peanut) | No | - | Lima et al. 2009 |
| *Asparagus officinalis*  (Asparagus) | Yes | Minor crop | Murga-Gutierrez et al. 2012 |
| *Avena sativa*  (White oat) | Yes | Minor crop | Lima et al. 2009 |
| *Avena strigose*  (Black oat) | No | - | Lima et al. 2009 |
| *Brassica napus*  (Rapeseed) | Yes | Minor crop | Lima et al. 2009 |
| *Brassica oleracea*  (Cabbage) | Yes | Major crop | O'Bannon 1975;  Whitehead, 1968, 1969 Carneiro et al., 2014 |
| *Cajanus cajan*  (Dwarf pigeon pea) | Yes | Minor crop | Lima et al. 2009 |
| *Capsicum annum*  (Pepper) | Yes | Major crop | Carneiro *et al*., 2004 |
| *Capsicum frutescens*  (Chili Pepper) | Yes | Minor crop | Whitehead 1968, 1969 |
| *Canavalia ensiformis*  (Jack-bean) | No | - | Lima et al. 2009 |
| *Carthamus tinctorius*  (Safflower) | Yes | Minor crop | Lima et al. 2009 |

| **Host Scientific name (common name)**  ***/ habitats\**** | **Presence in PRA area (Yes/No)** | **Comments (e.g. total area, major/minor crop in the PRA area, *major/minor habitats*\*)** | ***Reference*** |
| --- | --- | --- | --- |
| *Citrullus lantanus*  (Watermelon) | Yes | Minor crop | Carneiro *et al*., 2003; Carneiro *et al*., 2004 |
| *Clitoria ternatea*  (Butterfly pea) | Yes | Ornamental Minor crop | Lima et al. 2009 |
| *Crotalaria anaguroydes*  (Rattlepod) | No | - | Lima et al. 2009 |
| *Crotalaria apioclice*  (Rattlepod) | No | - | Lima et al. 2009 |
| *Crotalaria grantiana*  (Rattlepod) | No | - | Lima et al. 2009 |
| *Crotalaria juncea*  (Rattlepod) | No | - | Lima et al. 2009 |
| *Crotalaria lanceolata*  (Rattlepod) | No | - | Lima et al. 2009 |
| *Crotalaria okoelvka*  (Rattlepod) | No | - | Lima et al. 2009 |
| *Crotalaria spectabilis*  (Rattlepod) | No | - | Lima et al. 2009 |
| *Cucumis melo*  (Melon) | Yes | Major crop | Bellé et al. 2017b |
| *Cucumis sativus*  (Cucumber) | Yes | Major crop | Carneiro et al., 2014 |
| Cucurbita sp.  (Pumpkin) | Yes | Major crop | Whitehead 1969; |
| *Datura stramonium*  (Jimson weed) | Yes | weed | O’Bannon 1975. |
| *Dahlstedtia pentaphylla*  (Timbó) | No | - | Lima et al. 2009 |
| *Dolichos lablab*  (Hyacinth bean) | Yes | Ornamental Minor crop | Lima et al., 2009 |
| *Eleusine coracana*  (Finger millet) | Yes | Minor crop | Lima et al. 2009 |
| *Ensete ventricosum*  (Ethiopian banana) | Yes | Ornamental, minor crop | Mandefro & Dagne 2000 |
| *Eruca sativa*  (Aragula) | Yes | Minor crop | Carneiro et al., 2014 |
| *Euchlaena mexicana*  (synonim [*Zea*](http://www.theplantlist.org/tpl1.1/record/kew-450400)[*mexicana*)](http://www.theplantlist.org/tpl1.1/record/kew-450400) | No | - | Lima et al. 2009 |
| *Fagopyrum esculentum*  (Buckwheat) | Yes | Major crop | Lima et al. 2009 |
| *Glycine max*  (Soybean) | Yes | Major crop | O’Bannon 1975. Castro et al., 2003, Carneiro et  al., 2003 |
| *Glycine wightti*  (Cooper) | No | - | Lima et al. 2009 |
| *Gossypium hirsutum*  (Cotton) | No | - | O’Bannon, 1975 |
| *Helianthus annuus*  (Sunflower) | Yes | Minor crop | Lima et al. 2009 |
| *Jatropha curcas*  (Barbados nut) | No | - | Lima et al. 2009 |
| *Lactuca sativa*  (Lettuce) | Yes | Major crop | O'Bannon 1975. |
| *Lolium multiflorum*  (Italian ryegrass) | Yes | Minor crop | Lima et al. 2009 |

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| **Host Scientific name (common name)**  ***/ habitats\**** | **Presence in PRA area (Yes/No)** | **Comments (e.g. total area, major/minor crop in the PRA area, *major/minor habitats*\*)** | ***Reference*** |
| --- | --- | --- | --- |
| *Lupinus albus*  (White lupin) | Yes | Minor crop | Lima et al. 2009 |
| *Lupinus angustifolius*  (Blue lupin) | Yes | Minor crop | Lima et al. 2009 |
| *Lycopersicon*  *esculentum* (Tomato) | Yes | Major crop | Carneiro et al., 2003 Mandefro i Dagne 2000 |
| *Medicago sativa*  (Alfalfa) | Yes | Major crop | Lima et al. 2009 |
| *Mucuna aterrima*  (Black velvet bean) | No | - | Lima et al. 2009 |
| *Mucuna cinerea*  (Grey velvet bean) | No | - | Lima et al. 2009 |
| *Mucuna deeringiana*  (Dwarf velvet bean) | No | - | Lima et al. 2009 |
| *Mucuna puriens*  (Green velvet bean) |  | - | Lima et al. 2009 |
| *Nicotiana tabacum*  (Tobacco) | No | - | Whitehead 1968, 1969 |
| *Ornithopus compressus*  (Yellow serradella) | No | - | Lima et al. 2009 |
| *Oxalis corniculata*  (Creeping woodsorrel) | Yes | weed | [Belle](http://www.plantwise.org/KnowledgeBank/Datasheet.aspx?dsid=33239) et al. 2016b |
| *Oryza sativa*  (Rice) | No | - | Lima et al. 2009 |
| *Pennisetum glaucum*  (Pearl millet) | Yes | Minor crop | Lima et al. 2009 |
| *Pisum sativum* subsp*.*  *arvense*) (Forage pea) | Yes | Major crop | Lima et al. 2009 |
| *Pisum sativum*  (Pea) | Yes | Minor crop | Lima et al. 2009 |
| *Phaseolus vulgaris*  (Bean) | Yes | Major crop | Mandefro & Dagne 2000 Bellé et al. 2017a |
| *Polymnia sonchifolia*  (Yacon) | No | - | Carneiro & Almeida, 2005 |
| *Psidium guajava*  (Guava) | No | - | Dias et al. 2010 |
| *Prunus persica*  (Peach) | Yes | Major crop | Carneiro et al., 2003 Somavilla et al. 2009 |
| *Raphanus sativus*  (Radish) | Yes | Minor crop | Lima et al. 2009 |
| *Saccharum* spp.  (Sugarcane) | No | - | Bellé et al. 2017c |
| *Setaria italica*  (Foxtail millet) | Yes | Minor crop | Lima et al. 2009 |
| *Secale cereale*  (Rye) | Yes | Major crop | Lima et al. 2009 |
| *Sida rhombifolia*  (Arrowleaf sida) | No | - | Gomez et al. 2005; Lima-Medina et al. 2013 |
| *Solanum melongena*  (Eggplant) | Yes | Major crop | Carneiro et al. 2014 |
| *Solanum nigrum*  (Nightshade) | Yes | weed | O’Bannon 1975. |
| *Solanum tuberosum*  (Potato) | Yes | Major crop | Whitehead 1968, 1969;  Lima-Medina et al., 2011 |
| *Spinacia oleracea*  (Spinach) | Yes | Minor crop | Carneiro et al. 2014 |

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| **Host Scientific name (common name)**  ***/ habitats\**** | **Presence in PRA area (Yes/No)** | **Comments (e.g. total area, major/minor crop in the PRA area, *major/minor habitats*\*)** | ***Reference*** |
| --- | --- | --- | --- |
| *Tephrosia candida*  (White hoarypea) | No | - | Lima et al. 2009 |
| *Triticum aestivum*  (Triticale) | Yes | Minor crop | Lima et al. 2009 |
| *Vicia faba*  (Broad bean) | Yes | Minor crop | Whitehead 1969 |
| *Vigna umbellata*  [(Ricebean)](http://www.feedipedia.org/content/feeds?species=13546) | Yes | Minor crop | Lima et al. 2009 |
| *Vigna unguiculata*  (Cowpea) | Yes | Minor crop | Whitehead, 1968 Lima et al. 2009 |
| *Vigna radiata*  (Mungbean) | Yes | Minor crop | Lima et al. 2009 |
| *Vicia sativa*  (Common vetch) | Yes | Minor crop | Lima et al. 2009 |
| *Vicia villosa*  (Hairy vetch) | Yes | Minor crop | Lima et al. 2009 |
| *Vitis*  (Grapevine) | Yes | Major crop | Carneiro et al. 2003, 2004 |
| *Zea mays*  (Corn) | Yes | Major crop | Lima et al. 2009 |

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Host species list for *Meloidogyne luci*

| **Host Scientific name (common name)**  ***/ habitats\**** | **Presence in PRA area (Yes/No)** | **Comments (e.g. total area, major/minor crop in the PRA area, *major/minor habitats*\*)** | ***Reference*** |
| --- | --- | --- | --- |
| *Abelmoschus*  *esculentus* (Okra) | No | - | Carneiro et al., 2014 |
| *Actinidia deliciosa*  (Kiwi) | Yes | Minor crop for PRA area /  Major crop for some PRA area regions | Carneiro et al., 2014 |
| *Allium cepa* var. cepa  (Onion) | Yes | Minor crop | Strajnar et al., 2009 |
| *Antirrhinum majus*  (Snapdragon) | Yes | Ornamental Minor crop | Carneiro et al., 2014 |
| *Apium graveolens*  (Celery) | Yes | Minor crop | Strajnar et al., 2009 |
| *Brassica oleracea*  var. capitate (Cabbage) | Yes | Major crop | Strajnar et al., 2009 |
| *Brassica oleracea*  var. botrytis (Cauliflower) | Yes | Minor crop | Strajnar et al., 2009 |
| *Brassica oleracea*  var. gongylodes (Kohlrabi) | Yes | Minor crop | Strajnar et al., 2009 |
| *Brassica oleracea*  var. italic (Broccoli) | Yes | Minor crop | Strajnar et al., 2009 Carneiro et al., 2014 |
| *Brassica oleracea*  var. subauda (Kale) | Yes | Minor crop | Strajnar et al., 2009 |
| *Beta vulgaris* var.  cicla (Chard) | Yes | Minor crop | Strajnar et al., 2011 |
| *Beta vulgaris* var.  conditiva (Beet) | Yes | Minor crop | Strajnar et al., 2009 |
| *Capsicum annuum*  (Pepper) | Yes | Major crop | Carneiro et al., 2014 |
| *Cichorium endivia*  (Endive) | Yes | Major crop | Strajnar et al., 2009 |

| **Host Scientific name (common name)**  ***/ habitats\**** | **Presence in PRA area (Yes/No)** | **Comments (e.g. total area, major/minor crop in the PRA area, *major/minor habitats*\*)** | ***Reference*** |
| --- | --- | --- | --- |
| *Cichorium intybus*  var. foliosum (Chicory) | Yes | Major crop | Strajnar et al., 2009 |
| *Cucumis melo*  (Melon) | Yes | Major crop | Strajnar et al., 2009 |
| *Citrullus lantanus*  (Watermelon) | Yes | Minor crop | Geric Stare et al., 2017a |
| *Cucumis sativus*  (Cucumber) | Yes | Major crop | Strajnar et al., 2009 Carneiro et al., 2014 |
| *Daucus carota*  (Carrot) | Yes | Minor crop | Strajnar et al., 2009 |
| *Foeniculum vulgare*  var. azoricum (Florence fennel) | Yes | Minor crop | Strajnar et al., 2009 |
| *Glycine max* L.  (Soybean) | Yes | Major crop | Bellé et al., 2016a |
| *Helianthus annuus* L.  (Sunflower) | Yes | Minor crop | Strajnar et al., 2009 |
| *Hordeum vulgare* L.  (Barley) | Yes | Major crop | Strajnar et al., 2009 |
| *Hylotelephinum spectabile* L. (Sedum) | Yes | Ornamental minor crop | Carneiro et al., 2014 |
| *Lactuca sativa*  (Lettuce) | Yes | Major crop | Strajnar et al., 2009 Carneiro et al., 2014 |
| *Lavandula spica* L  (Lavender) | Yes | Ornamental minor crop | Carneiro et al., 2014 |
| *Lycopersicum*  *esculentum* (Tomato) | Yes | Major crop | Strajnar et al., 2009 |
| Nicotiana tabacum L.  (tobacco) | No | - | Carneiro et al., 2014 |
| *Phaseolus vulgaris*  (Common Bean) | Yes | Major crop | Strajnar et al., 2009 Carneiro et al., 2014 |
| *Pisum sativum* (Pea) | Yes | Major crop | Strajnar et al., 2009 |
| *Polymnia sonchifolia*  Poepp. Endl (Yakon) | No | - | Carneiro et al., 2014 |
| *Raphanus sativus* var.  *radicula* (Radish) | Yes | Minor crop | Strajnar et al., 2009 |
| *Rosa* sp. (Rose) | Yes | Ornamental minor crop | Carneiro et al., 2014 |
| *Rumex patientia* L  (Curled dock) | Yes | Weed | Strajnar et al., 2009 |
| *Solanum melongena*  (Eggplant) | Yes | Major crop | Strajnar et al., 2009 |
| *Solanum tuberosum*  (Potato) | Yes | Major crop | Maleita et al., 2016 |
| *Spinacia oleracea*  (Spinach) | Yes | Minor crop | Strajnar et al., 2009 |
| *Vitis vinifera*  (Grapevine) | Yes | Major crop | Carneiro et al., 2014 |
| Zea mays (Corn) | Yes | Major crop | Strajnar et al., 2011 |
| *Zea mays* var.  *saccharata* (Sweet corn) | Yes | Minor crop | Strajnar et al., 2009 |

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# Pathways for entry

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| **Possible pathways**  *(in order of importance)* | **Short description explaining why it is considered as a pathway** | Pathway prohibited in the PRA area?  Yes/No | Pest already intercepted on the pathway?  Yes/No |
| --- | --- | --- | --- |
| Host plants for planting (except seeds, bulbs and tubers) with or without soil attached | Infested plant roots (early or later stage) carry viable pest; in case of planting infected material; | No for all hosts/countries  Yes- for some hosts from some non-EU or non- European countries.  See points 9., 11., 13., 15.,  18. of Annex III CD 2000/29/EC  and specific requirements for all soil attached to plants originating in some non-EU countries and non-European countries.  See point 34. of Annex IV CD 2000/29/EC | No |
| Plants for planting of non- hosts with soil | Infested soil attached (pest in egg stage and J2) | No for all hosts/countries  Yes- for some hosts from some non-EU or non- European countries.  See points 1,2,3, 9.1, 16, 17,19. of Annex III CD 2000/29/EC.  Specific requirements for all soil attached to plants originating in some non- EU countries and non- European countries. See point 34. of Annex IV CD 2000/29/EC | No |
| Bulbs, tubers, corms, rhizomes as plants for planting | Infested tubers (e.g. potato) carry viable pest; in case of planting infected material; | No for all hosts/countries  Yes- for some hosts from non-EU or non-European countries.  See points 10.of Annex III CD 2000/29/EC | No |
| Host plant parts contaminated with soil (such as roots, tubers, bulbs, corms, rhizomes), for consumption or processing. | Infested consumption tubers, roots (e.g. infested consumption potato, celery, carrots etc.); | No for all hosts/countries  Yes- for some hosts from non-EU or non-European countries.  See points 12.of Annex III CD 2000/29/EC. | No |

| **Possible pathways**  *(in order of importance)* | **Short description explaining why it is considered as a pathway** | Pathway prohibited in the PRA area?  Yes/No | Pest already intercepted on the pathway?  Yes/No |
| --- | --- | --- | --- |
| Non-host plant parts contaminated with soil (such as roots, tubers, bulbs, corms, rhizomes), for consumption or processing. | Infested soil attached (pest in egg stage and J2) | No |  |
| Soil/ growing medium | There is a possibility to introduce the pest in egg stage and J2. | No for all hosts/countries  Yes- from some non-EU countries and non- European countries.  See point 14. of Annex III CD 2000/29/EC | No |
| Plant waste | There is possibility to introduce the pest in egg stage; | No | No |

*Rating of the likelihood of entry Low ☐ Moderate  High ☐*

*Rating of uncertainty Low ☐ Moderate  High ☐*

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Pathways for entry of *M. luci* from EU countries, where *M. luci* is present, are not regulated (Italy, Greece, Portugal). It is not known how RKN *M. luci* was introduced in Slovenia. However, *M. luci* was detected in Slovenia twice. The location of infestation from 2015 is situated more than 100 km away from location found in 2003 and no direct connections between these farms could be found. This indicates that infestation at the new location might be a result of a new entry into the county. Therefore rating of the likelihood of entry and uncertainty was assessed as moderate.

# Likelihood of establishment outdoors in the PRA area

*M. luci* has already been detected in the open field production in Europe (Mediterranean climate) in corn and kiwi production in Greece, and potato production in Portugal. In addition, the potential of *M. luci* to survive outdoors under a continental climate even in areas where soil temperatures fall below zero during winter, as well as under a sub-Mediterranean climate (Strajnar et al. 2011) indicates that *M. luci* could establish in most parts of the PRA area

*M. luci*

*Rating of the likelihood of establishment outdoors Low ☐ Moderate ☐ High* 

*Rating of uncertainty Low*  *Moderate ☐ High ☐*

*M. ethiopica*

*Rating of the likelihood of establishment outdoors Low ☐ Moderate*  *High* ☐

*Rating of uncertainty Low ☐ Moderate*  *High* ☐

# Likelihood of establishment in protected conditions in the PRA area

Both pest species have very wide host range including majority of vegetable crops which are cultivated in the protected areas. The probability of establishment in the protected area is high.

*M. ethiopica* & *M. luci*

*Rating of the likelihood of establishment in protected conditions*

*Low ☐ Moderate ☐ High* 

*Rating of uncertainty*

*Low*  *Moderate ☐ High ☐*

# Spread in the PRA area

* + Natural spread

Natural spreading of RKNs is very slow in the soil and can reach up to few meters from the infestation in a growing season.

* + Human assisted spread

Human assisted spread is much more important. The spreading may occur by agricultural machinery (tractor wheals, tillage and cultivator equipment, any equipment that have direct soil contact). Further, nematodes can be spread by irrigation, movement of crop plants and crop products, or by infested soil and growing media.

Plants for planting (vegetables, ornamental plants, fruit trees and grapevine) with or without soil attached are considered the main pathway for spread of *M. ethiopica* and *M. luci* within the PRA area. In addition, plant parts like tubers may present also high risk of spreading in case of pest establishment outdoors, especially in potato tuber seed production where latent infestations may also occur (tubers with no visible symptoms of infestation).

*M. ethiopica* & *M. luci*

*Rating of the magnitude of spread Low ☐ Moderate*  *High ☐*

*Rating of uncertainty Low ☐ Moderate*  *High ☐*

# Impact in the current area of distribution

*M. ethiopica* is widely distributed in vineyards in Chile (Aballay et al. 2009) and was found to be the major root knot nematode parasite of grapevines (*Vitis vinifera*) and other crops in Chile (Carneiro et al., 2007). In Brazil, *M. ethiopica* affects the root growth, yield and quality of grapevines and kiwi, and cause serious economic losses (Carneiro et al. 2003). Furthermore, Medina et al. (2014) reported *M. ethiopica* on potato plants from Parana state, Brazil, showing root systems with multiple galls and tubers with protuberances. *M. luci* has been reported on several vegetables but there are no reports of economic damage caused by *M. luci* except from Slovenia.

In Slovenia, *M. luci* was found at two locations. In the greenhouse in Primorska from 2003 the pest was eradicated due to omission of the agricultural production. Second location was detected in 2015 in a greenhouse in the village near Ljubljana. Infested tomato plants showed symptoms of decline, leaf chlorosis and large galls were present on the roots. Nematode infestation was relatively high as more than 80 % of plants were severely damaged. Phytosanitary measures were imposed in the infested greenhouse including the use of dazomet, a soil fumigant. All infested plant material from the affected greenhouse was destroyed. However, the eradication program implemented in the past two years has not been successful. Therefore only resistant tomato cultivars carrying *Mi* gene and poor- to non-host crops in combination with registered chemical nematicide were allowed to be grown in the infested greenhouse. The success of this management program for nematode control will be assessed in the future. The infested area measures approximately 1100 m2.

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*M. ethiopica* & *M. luci*

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*Rating of the magnitude of impact in the current area of distribution*

*Low ☐ Moderate ☐ High* 

*Rating of uncertainty*

*Low ☐ Moderate*  *High ☐*

Rating of the magnitude of impact in the current area of distribution was assessed as high because of high impact of *M. luci* at current area of distribution in PRA area.

Moderate rating of uncertainty was assessed due to lack of available data on economic damage caused by both species at current area of distribution.

# Potential impact in the PRA area

*M. ethiopica* and *M. luci* could survive under protected cultivation (glasshouses) across the PRA area. Climatic conditions for establishment of *M. luci* in the PRA area outdoors are suitable since this species was proved to overwinter in continental and Mediterranean climate. Besides it was already detected outdoors in Europe (Mediterranean climate), in corn and kiwi production in Greece, and potato production in Portugal. Once the RKNs is introduced into the PRA area, it is in general difficult to control or eradicate them, especially outdoors at the open field. The ability of *M. ethiopica* to overwinter at the open field in continental and Mediterranean climate has not been tested yet.

The models of climate changes predict an increase of average temperatures and more frequent periods of drought, floods and heat waves. Higher temperatures will allow development of more generations of RKNs in a growing season and consequently higher yield losses. The potential impact of both pest species in the PRA area is high.

Will impacts be largely the same as in the current area of distribution? No

The potential impact of pests on outdoor crops in the PRA area is difficult to estimate. Epidemiological studies such as the assessment of the damage potential of both pest species for major crops such as potatoes, corn, soybean, grapevine, fruit trees etc. in the PRA area climate are not available. We can expect losses in the protected cultivation (with soil as natural growing medium) and the same impact as in the current area of distribution.

# If No

Rating of the magnitude of impact in the area of potential establishment

Low ☐ Moderate ☐ High 

Rating of uncertainty

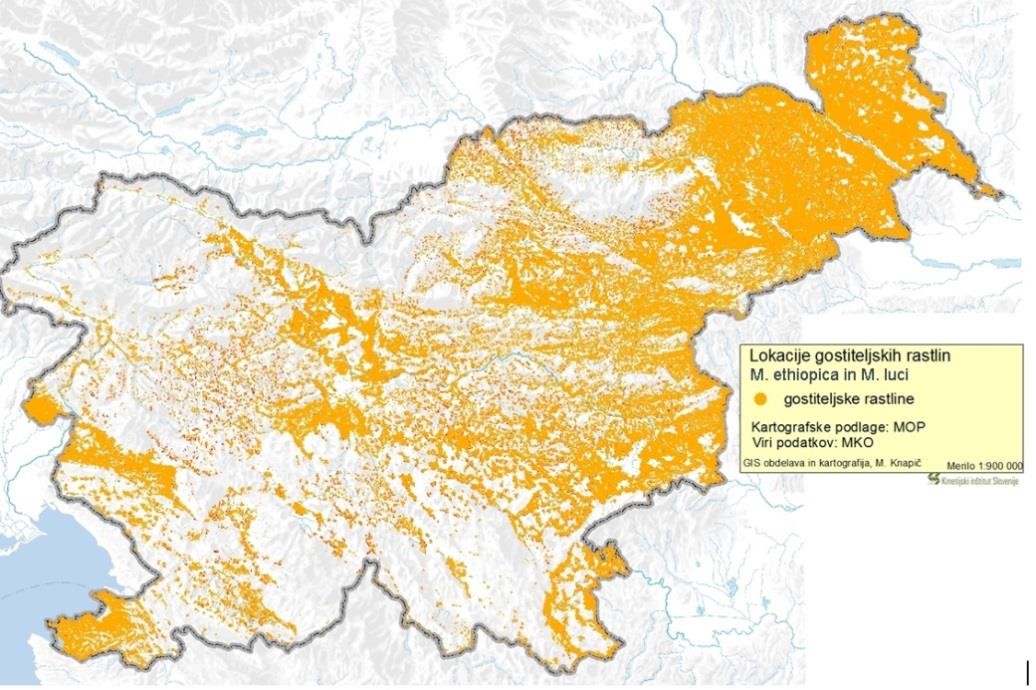
Low ☐ Moderate  High ☐

1. **Identification of the endangered area**

Endangered area in PRA area is:

* Protected area production
* Vegetable production area
* Potato production area
* Corn and other cereals production area
* Vineyards
* Kiwi and peach production area

Figure: Distribution of the most important major crops in Slovenia (covers almost all national agricultural production land).



# Overall assessment of risk

Root knot nematodes *M. ethiopica* and *M. luci* can be introduced by several pathways but the highest risk for entry and spread in the PRA area is considered the pathway of host plants for planting (except seeds, bulbs and tubers) with or without soil attached, like the planting material of vegetables, ornamental plants, fruit trees and grapevine. Furthermore, plant parts like tubers may present also high risk of spread in case of pest establishment outdoors, especially in potato tuber seed production where latent infestations may also occur (tubers with no visible symptoms of infestation). Human assisted spread of the pests is the most important.

*M. ethiopica* and *M. luci* have very wide host range including majority of vegetable crops which are cultivated in the protected areas. The probability of establishment in the protected area is high. *M. luci* has already been detected in the open field production in Europe (Mediterranean climate), in corn and kiwi production in Greece, and potato production in Portugal. There is a high risk of *M. luci* establishment outdoors in the PRA area, as this species was proved to overwinter in open field in continental and Mediterranean climate. Once the RKNs have been introduced, it is in general difficult to control or eradicate them, especially outdoors at the open field. The ability of *M. ethiopica* to overwinter at the open field in continental and Mediterranean climate has not been tested yet. The endangered area in the PRA area is therefore: protected production area, vegetable production area, potato production area, corn and other cereals production area, vineyards, kiwi and peach production area.

Phytosanitary measures to prevent the spread of these pests into non-infested area in case of an outbreak are necessary. The measures should include prohibition of movement of plants for planting production outside infested area, movement of soil outside infested area, disinfection of equipment and footwear, destruction of nematode infested plant waste, and intensive monitoring of infested area and the surroundings.

The potential impact of both pest species in the PRA area is high especially in the light of climate change scenarios. The models of climate changes predict an increase of average temperatures and more frequent periods of drought, floods and heat waves. Higher temperatures would allow development of more generations of RKNs in a growing season and consequently higher yield losses.

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# Stage 3. Pest risk management

1. **Phytosanitary measures**
   1. Measures on individual pathways

Measures for host plants for planting, roots, bulbs, tubers, corms, rhizomes are listed in the table below as the high risk pathway, in order to prevent entry of *M. ethiopica and M. luci* into host production systems.

For soil (and growing media), the risk of entry relates mostly to soil from where RKNs are present, but many EPPO countries prohibit import of soil from third countries. Internal movement of soil within the EU is not regulated, but the likelihood of entry from Italy, Greece and Portugal is considered as low (due to the types of environments in which the nematode is currently present). Measures were not studied, but are likely to be similar to those for other nematodes.

| **Possible pathways *(in order of importance))*** | **Measures identified** |
| --- | --- |
| Host plants for planting with roots (including rooted cuttings, bulbs, tubers, corms, rhizomes; except plants in tissue culture), with or without  soil or growing media | Pest free area Or  Pest-free production site or pest-free place of production (with all production sites pest-free)  Or  Growing under complete physical isolation (EPPO Standard PM5/8) (with requirements appropriate for RKN) |
| Host plant parts (bulbs, tubers, corms, rhizomes) with soil or growing media | Pest free area Or  Pest-free production site (or pest-free place of production with all production sites pest-free)  Or  Growing under complete physical isolation (EPPO Standard PM5/8) (with requirements appropriate for *RKN*) |

# Eradication and containment

* Prevention of spread

At the current *M. luci* infested area several phytosanitary measures were implemented to prevent the spread of the pest into non-infested areas. The measures include prohibition of movement of plants for planting and soil from infested area, disinfection of equipment and footwear, destruction of nematode infested plant waste, and intensive survey of the infested area and surroundings.

* Eradication/control

Once RKN have been introduced, it is in general difficult to control or eradicate them mainly because of their polyphagous nature and limited management options. At the place of production RKNs can be controlled with a proper crop rotation of poor and non-host crops, resistant varieties and the usage of nematocides. Efficient crop rotation is very difficult to set up in vegetable production as there are many vegetable crops that are good hosts for both species. Several commercial tomato and pepper varieties exhibit resistance and tolerance against RKN. However, the resistance breaking populations of *M. luci* have already been reported from Turkey. No other host crop exhibit resistance.

There are two registered nematocides in Slovenia, a soil fumigant dazomet for protected areas and open field, and fluopyram for protected areas only. Dazomet was used in *M. luci* eradication program at 2015 outbreak but did not reach successful effect as the RKN population rendered again in next growing season, maybe due to unsuitable application. The efficiency of fluopyram at the same location is

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currently under investigation. A product which employs a biological mode of action with *Bacillus firmus* bacteria strain that prevents nematodes from causing damage is currently in the registration process (Terefe et al., 2009)

# Uncertainty

The main sources of uncertainty within the risk assessment:

* the likelihood of pest entry,
* the magnitude of spread in the PRA area,
* the magnitude of impact in the area of potential establishment.

It is presumed that *M. luci* was introduced in Slovenia twice, in separate entries into the county. The pathways of entry in both cases have not been identified. At the same time origin of the pest is difficult to suspect because of very limited known pest distribution in European and at the world scale.

The uncertainty for the magnitude of spread and potential impact in the PRA was also assessed as “moderate” mainly because of uncertain entry pathways and the lack of epidemiological data for the PRA area climate conditions. Both pest species might colonise entire PRA area. It is difficult to estimate the influence of pests on outdoor crops, but we can observe losses in the protected cultivation (with soil as natural growing medium). Epidemiological studies such as the assessment of the damage potential of both pest species for major crops such as potatoes, corn, soybean, grapevine, fruit trees etc. in the PRA area climate would enable informative phytosanitary decisions. Several tomato and pepper varieties exhibit resistance and tolerance against RKN however the resistance breaking populations of

*M. luci* have already been reported from Turkey.

The main source of uncertainty within the risk of management is lack of efficient pests control measures. Because of limiting chemical pesticide usage trend, more environmentally friendly approaches will be use in the future. Performing pest management studies of genetic resistance sources and applications of soil amendments formulation like essential oils or biomasses from plants with biocidal properties, as well as microbial bio-nematicides based on nematode-parasitic fungi or bacteria are therefore of the utmost importance.

# Remarks

The biggest concern for *M. luci* is the spread at the open field agricultural production area (see 9. likelihood of establishment outdoors in the PRA area). The ability of *M. ethiopica* to overwinter in the open field in continental and Mediterranean climate has not been tested yet. Because of the polyphagous nature of both pests and limited management options the pests (in case of a wide spread) would be very difficult to control. In addition, the climate changes are in favour of such tropical pests which complete more reproduction cycles at higher summer temperatures and thus cause greater damage.

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**Appendix 1. Symptoms of *M. luci* infestation.**

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*M. luci infestation of tomatoes in Slovenia in 2015. (Foto: KIS) M. luci infested tomato root system. (Foto: KIS)*