# **RESEARCH/INVESTIGACIÓN**

# FURTHER ELUCIDATION OF THE HOST RANGE OF *GLOBODERA ELLINGTONAE*

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## ABSTRACT

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*Globodera ellingtonae* was first discovered in Oregon and Idaho in 2008 and described as a new species in 2012. Knowledge of the host range of this nematode is limited, with only tomato (*Solanum lycopersicum*) and potato (*Solanum tuberosum*) reported as hosts. This study was conducted to expand the information available on the host range of *G. ellingtonae*. In greenhouse studies, a range of agricultural Solanaceous and non-solanaceous crop plants and Solanaceous weeds were inoculated with *G. ellingtonae* and nematode reproduction was determined after four months. Crops historically grown in rotation with potato at the site where *G. ellingtonae* was discovered in Oregon, alfalfa (*Medicago sativa*), wheat (*Triticum aestievum*), and oat (*Avena sativa*) were all non-hosts for the nematodes. None of the Solanaceous crop plants evaluated, *Capsicum annum* (bell and jalepeno pepper), *Nicotiana tabacum* (tobacco varietiies), *Physalis philadelphica* (tomatillo), or *Solanum melagena* (eggplant) were hosts for *G. ellingtonae*. All of the Solanaceous weed species evaluated, *Solanum nigrum, Solanum dulcamara*, and *Solanum rostratum* were hosts for *G. ellingtonae* with final population density/initial population density (*Pf/Pi*) values ranging from 1.5 to 27.0. The trap crop *Solanum sisymbriifolium* was a non-host for the nematode.

Key words: Globodera, host, potato cyst nematode, reproduction

## RESUMEN

Peetz, A.B., H. V. Baker, and I. A. Zasada. 2019. Elucidación adicional de la gama de hospedantes de *Globodera ellingtonae*. Nematropica 49:12-17.

Globodera ellingtonae se descubrió por primera vez en Oregón e Idaho en 2008, y se describió como una nueva especie en 2012. El conocimiento del rango de hospedadores de este nematodo es limitado, con solo tomate (Solanum lycopersicum) y papa (Solanum tuberosum) informados como hospedadores. Este estudio se realizó para ampliar la información disponible en el rango de hospedadores de *G. ellingtonae*. En estudios de invernadero, se inoculó una variedad de cultivos agrícolas de Solanáceas y no Solanáceas y malezas Solanáceas con *G. ellingtonae* y se determinó la reproducción del nematodo después de cuatro meses. Los cultivos que históricamente se cultivaron en rotación con papa en el sitio donde se descubrió *G. ellingtonae* en Oregón, la alfalfa (Medicago sativa), el trigo (Triticum aestievum) y la avena (Avena sativa) no fueron hospedadores de los nematodos. Ninguna de las plantas de cultivo Solanáceas evaluadas, Capsicum annum (pimiento y jalepeno), Nicotiana tabacum (variedades de tabaco), Physalis philadelphica

(tomatillo) o *Solanum melagena* (berenjena) fueron anfitriones de *G. ellingtonae*; dejar el tomate y la papa como los únicos cultivos Solanáceos demostraron ser anfitriones de *G. ellingtonae*. Todas las especies de malezas Solanceous evaluadas, *Solanum nigrum, Solanum dulcamara* y *Solanum rostratum* fueron hospedantes de *G. ellingtonae* con valores de densidad de población final / densidad de población inicial (Pf/Pi) que oscilaron entre 1.5 y 27.0. El cultivo de trampa *Solanum sisymbriifolium* no fue hospedante para el nematodo.

Palabras clave: Globodera, huésped, nematodo del quiste de la patata, reproducción

#### INTRODUCTION

A Globodera species morphologically and molecularly distinct from the guarantine potato cyst nematodes (PCN; Globodera rostochiensis and G. pallida) was first discovered in potato production fields in Oregon and Idaho in 2008 (Skantar et al., 2011). As part of this initial find, nematodes were inoculated onto potato (Solanum tuberosum) and potato was identified as a host for this nematode. In 2012, this nematode was described as *Globodera ellingtonae* (Handoo *et al.*. 2012). Data from studies including hatching assays, developmental biology, and phylogenetic analysis indicate that G. ellingtonae is more closely related to G. rostochiensis than G. pallida (Handoo et al., 2012; Phillips et al., 2015; Zasada et al., 2015). Studies on the development of G. ellingtonae in a field environment also indicate that G. ellingtonae can likely survive in the Andes Mountains of South America where its close relatives G. pallida and G. rostochiensis originated (Plantard et al., 2008; Boucher et al., 2013; Phillips et al., 2015, 2017). Beyond tomato and potato being identified as hosts for G. ellingtonae (Zasada et al., 2013; Lax et al., 2014), and tobacco as a nonhost (Lax et al., 2014), little information exists on the host range of G. ellingtonae.

The established host range for PCN is narrow and restricted to plant species belonging to the family Solanaceae (Sullivan *et al.*, 2007). *Globodera pallida* and *G. rostochiensis* have highly specialized survival strategies and hostspecific hatching cues are usually required for the nematodes to hatch (Perry and Gaur, 1996). Additionally, these nematodes establish a complex relationship with their host to enable successful reproduction (von Mende *et al.*, 1998). Recently, the host range of each PCN was expanded to include Solanaceous weeds commonly found in or near agricultural fields. New reported hosts for *G*. pallida included Solanum physalifolium while Solanum dulcamara, Solanum nigrum, and Solanum villosum were reported as hosts for *G.* rostochiensis (Boydston *et al.*, 2010; Rott *et al.*, 2011; Mimee *et al.*, 2014).

Knowledge of the host range of plant-parasitic nematodes is critical information when making informed management decisions such as which crops to rotate to as well as which cover crops to use. From a policy perspective, weeds and cover crops alike can act as alternate hosts of plantparasitic nematodes, keeping populations alive amidst eradication efforts and thus rendering these efforts unsuccessful (Forge *et al.*, 2000; Boydston *et al.*, 2004; Boydston *et al.*, 2010). As such, our objective was to expand the known host range of *G. ellingtonae* with particular focus on potential Solanaceous host plant species and other crop species grown in rotation with potato.

# **MATERIALS AND METHODS**

Cysts of *G. ellingtonae* were reared on 'Russet Burbank' potato (*Solanum tuberosum*) in a field at Powell Butte, OR, USA. Soil was collected from the site and air-dried prior to extraction of cysts for inoculum. Cysts were extracted from soil using a USDA cyst extractor (Ayoub, 1980). Extracted cysts were handpicked, enumerated, and then crushed with a rubber stopper on a 250- over a 25- $\mu$ m sieve, with eggs being retained on the 25- $\mu$ m sieve. Eggs were washed from the sieve into a 50ml tube and then enumerated using an inverted microscope.

Several Solanum weed species were tested for host status for *G. ellingtonae* including *S. dulcamara* (accession PI 643457), *S. nigrum* (accessions PI 304600 and PI 381290), *S. rostratum* (accession PI 420997), and *S. sisymbriifolium* (accession unknown). Seeds were obtained from the United States Department of

Agriculture Germplasm Resources Information Network (USDA GRIN; https://www.ars-grin.gov/ in Beltsville, MD) and Chuck Brown (USDA-ARS Prosser, WA). Seeds were notched using a micro scalpel, placed on an 85-mm Grade 1 filter paper (Whatman; Buckinghamshire, UK), moistened with deionized water, and subsequently incubated in sealed 100 x 15 mm petri plates (VWR; Radnor, PA) in complete darkness at 18°C for 3-5 days. Germinated seeds were then placed in 6-pack containers containing soilless media to continue to root. Crop plants were also evaluated including oats (Avena sativa), bell and jalapeño pepper (Capsicum annum), alfalfa (Medicago sativa), 'K326' tobacco (Nicotiana tabacum), 'TM900 Mexican Strain' tomatillo (Physalis 'Wheeler' (Triticum philadelphica), wheat aestivum), 'Rutgers' tomato (Solanum lycopersicum), and 'Black Beauty' eggplant (Solanum melongena). These were all direct seeded into 6-pack containers containing soilless media. An unknown variety of tobacco was also included in the experiment. Tobacco plants were received as seedlings from Karen Keller (USDA-ARS, Corvallis, OR). 'Désirée' potato (S. tuberosum) was included as a positive control in each experiment and seedlings were produced by placing tubers in a 25- x 25-cm bin containing soilless media to germinate. When weed and crop seedlings were approximately 5 to 7 cm tall, they were transplanted into 10-cm round clay pots containing approximately 500 g of a 1:1 steampasteurized sand and Willamette loam mix. Globodera ellingtonae was inoculated onto plant roots either by pipetting ~2,500 eggs directly onto roots in 2 to 3 ml water or by placing 10 cysts containing eggs onto the root system prior to covering roots with soil to achieve an initial density (Pi) of approximately 5 eggs/g soil.

Pots containing plants were placed in 99 x 51 x 16.5 cm plastic bins with 25 pots per bin (Sterilite; Townsend, MA). Holes were drilled in the bottom of the bins and screens were secured to the 10 drilled holes. Experiments were arranged in a randomized complete block design with 4 to 6 replications per treatment for the Solanaceous weeds and Solanaceous crop species experiments, respectively; each experiment was conducted twice. Plants were watered with 9-45-15 (N-P-K) fertilizer (Scotts, Marysville, OH) immediately after transplanting. Plants were grown in a greenhouse under long-day conditions, 16-hr

photoperiod, with 23/18°C day/night temperatures and were fertilized twice each week with 20-20-20 (N-P-K) fertilizer (Scotts). The plants were grown in the greenhouse for four months or until the plants naturally senesced. At harvest, the aboveground portion of the plant was removed and discarded. The soil from the pots, along with any tubers or roots were spread on trays to dry. Tubers and roots were removed and discarded from dry soil. The total amount of dry soil was weighed and 200-g subsamples were collected from each sample. Cysts were extracted, collected, counted, crushed, and the egg density determined as described above.

For each plant, final egg density (*Pf*) was divided by Pi to obtain Pf/Pi. The relative susceptibility (RS) of the plants was calculated by dividing the *Pf* of the test plant by the average *Pf* for 'Désirée' potato (EPPO, 2006), a measure that has been used in previous studies evaluating Solanaceous weeds as host for *G. rostochiensis* (Rott *et al.*, 2011; Mimee *et al.*, 2014). Each host was given a RS score with 1 indicating the highest level of resistance.

# **RESULTS AND DISCUSSION**

This is the first report of the host status of several Solanaceous plants for G. ellingtonae. Prior to this report, the host status of only three plant species had been considered, with tomato and potato as hosts (Zasada et al., 2013; Lax et al., 2014) and tobacco as a non-host (Lax et al., 2014) for G. ellingtonae. Similar to G. rostochiensis and G. pallida, G. ellingtonae has a very narrow agricultural crop host range; only tomato and potato are hosts of the plants evaluated (Table 1). Similar to our previous findings (Zasada et al., 2013), potato, in general, is a better host for G. ellingtonae than tomato. In this study the average *Pf/Pi* was 4.3 times greater on potato than tomato, similar to the 5.3 times difference in reproduction between potato and tomato in a previous study (Zasada et al., 2013). However, root diffusates of potato and tomato elicited similar hatch of G. ellingtonae in in vitro assays (Zasada et al., 2013). To date, the following potato varieties have been demonstrated to be hosts for G. ellingtonae: Désirée, Russet Burbank, Yukon Gold, Modoc, Norland, Umatilla, and Colorada (Zasada et al., 2013; Lax et al., 2014). The only other

Crop species	Cultivar	Common name	Pf/Pi <sup>x</sup>	RS score <sup>y</sup>				
Avena sativa	NA <sup>z</sup>	Oat	0°	9				
Capsicum annuum	NA	Green bell	0	9				
C. annuum	NA	Jalepeño	0	9				
Medicago sativa	NA	Alfalfa	0	9				
Nicotiana tabacum	NA	Tobacco	0	9				
N. tabacum	K326	Tobacco	0	9				
Physalis philadelphica	TM900 Mexican Strain	Tomatillo	0	9				
Solanum lycopersicum	Rutgers	Tomato	5.4 (±1.1)	1				
S. melongena	Black Beauty	Eggplant	0	9				
S. tuberosum	Désirée	Potato	33.1 (±2.7)	1				
Triticum aestivum	Wheeler	Wheat	0	9				

Table 1. Host status of Solanaceous crop species and crop species grown in rotation with potato (Solanum tuberosum) in Oregon for Globodera ellingtonae.

 $^{x}Pf/Pi$  is the multiplication rate expressed as a ratio between final egg population (*Pf*) divided by the initial egg population (*Pi*). Values are the mean (+ standard error) of 12 observations from repeated experiments.

 $^{y}RS$  = Relative susceptibility, which is calculated as *Pf* of test plant/*Pf* of 'Désirée' potato (EPPO, 2006).

<sup>z</sup>NA=Not Available

Solanaceous crop reported to be a host for a *Globodera* species is eggplant (Ambrogioni *et al.*, 2000). There was no reproduction of *G. ellingtonae* on bell pepper, eggplant, and on the two cultivars of tobacco evaluated (Table 1). Lax *et al.* (2014) reported no reproduction on 'K326' tobacco as was observed in this study. The inclusion of the non-solanaceous crop plants, alfalfa, oat, and wheat (Table 1) was to demonstrate that only potato was a host in the crop rotation scheme employed at the farm where *G. ellingtonae* was found in Oregon.

All of the Solanaceous weed species included in the study were hosts for G. ellingtonae, except for S. sisvmbriifolium (Table 2). Sullivan et al. (2007) reported S. nigrum as a non-host for G. rostochiensis with an RS score of 9. However, later research indicated that S. nigrum has a range of susceptibility against G. rostochiensis. Interestingly, this range seems to be dependent on the origin of the Globodera population, as well as where the plant was collected. This varying host status was significant for S. villosum and G. rostochiensis (Rott et al., 2011; Mimee et al., 2013). Additional examples of this complex relationship were also observed between S. dulcamara and G. rostochiensis (Mimee et al., 2013). Several Solanaceous weed species were tested but not included in our results due to difficulty in obtaining enough viable seed to repeat experiments. Among those, S. ptycanthum (accession 64750068), S. sarrachoides (accession 954750073), S. triflorum, and S. villosum (accessions 804750186 and 884750018) were

assessed during a single experiment (data not shown). All of these species would be considered hosts for *G. ellingtonae* with *S. sarrachoides*, *S. trifolrum*, and *S. villosum* being excellent hosts (RS = 1) and *S. ptycanthum* a good host (RS = 5). For comparison with other PCN host ranges, *S. ptycanthum* and *S. triflorum* were reported to be a poor hosts for *G. rostochiensis* (Rott *et al.*, 2011). Of particular interest is the likely host status of *S. sarrachoides*. It has been reported as a non-host for *G. rostochiensis* but a suitable host for *G. pallida* (Boydston *et al.*, 2010; Rott *et al.*, 2011). Additionally, we have observed replication of *G. ellingtonae* on *S. sarrachoides* in the field (Zasada, unpublished data).

Solanum sisymbriifolium has been identified as a trap crop for PCN (Scholte, 2000). While S. sisymbriifolium does induce egg hatch of G. pallida and G. rostochiensis, it does not allow the nematodes to complete their life cycle; S. sisymbriifolum is also not a host for G. ellingtonae (Table 2). From a management perspective, there may be temporal limitations of crop growth period (207-227 days to reach 90-95% hatch) of S. sisymbriifolium, which may be an impediment to utilizing it as a trap crop in the field (Timmermans et al., 2006). However, a recent study evaluated the impact of S. sisymbriifolium on the ability of G. pallida to parasitize potato after exposure to a S. sisymbriifolium trap crop (Dandurand and Knudson, 2016). In this study, the progeny were as low as 1/1,000 of the initial population, indicating S. sisymbriifolium root diffusate may impact G.

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to reports for <i>Grobbacra partia</i> ( <i>Gp</i> ) and <i>Grobbacra rostocnichsts</i> ( <i>Gr</i> ) from the mendance.								
Solanum species	Accession	Pf/Pi <sup>v</sup>	$Ge \operatorname{RS}^{w}$	$Gp \operatorname{RS}^{x}$	$Gr \operatorname{RS}^{\operatorname{y}}$			
S. dulcamara	PI 643457	3.6 (±6.2)	2	7	4-9			
S. nigrum	PI 304600	27.0 (±7.1)	1	7	3-9			
S. nigrum	PI 381290	14.3 (±11.3)	1		8-9			
S. rostratum	PI 420997	$1.4 (\pm 0.9)$	5		9			
S. sisymbriifolium	NA <sup>z</sup>		9		9			
S. sisymbriifolium II	NA		9					
S. tuberosum 'Désirée'		9.7 (±3.1)	1					

Table 2. Host status of Solanaceous weed species for *Globodera ellingtonae* (Ge) and comparison of host status to reports for *Globodera pallida* (Gp) and *Globodera rostochiensis* (Gr) from the literature.

<sup>v</sup>*Pf*/*Pi* is the multiplication rate expressed as a ration between final egg population (*Pf*) divided by the initial egg population (*Pi*). Values are the mean ( $\pm$  standard error) of 8 to 12 observations from repeated experiments. <sup>w</sup>RS = Relative susceptibility which is calculated as *Pf* of test plant/*Pf* of 'Désirée' potato.

<sup>x</sup>Boydston et al., 2010

<sup>y</sup>Rott et al., 2011 and Mimee et al., 2014

 $^{z}NA = not available.$ 

*pallida* reproduction following its removal and subsequent crop rotation to a known host. *Solanum sisymbriifolium* may contain glycoalkaloids with unknown nematicidal activity, and the fruit is toxic to some mollusks (Bagalwa *et al.*, 2010; Dias *et al.*, 2012). The potential biocidal activities of this species against *Globodera* species warrants further investigation.

Knowledge of the potential for Solanaceous weed species to support nematode reproduction is critical when trying to manage or exclude a nematode from an area. For example, if G. ellingtonae is introduced to a different region where an alternate host is growing, its population density may potentially increase even in the absence of potato production. Depending upon the density of the host Solanaceous weeds in a field, the potential increase in population density of G. ellingtonae could be more rapid and greater than if potato was planted on a rotational basis because a host would be present every year on which G. ellingtonae could reproduce. Future regarding control of recommendations G. ellingtonae include cultural controls such as fallow and a strict weed control program, as well as further research into the full potential of S. sisymbriifolium as a trap crop in a field setting.

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