

Susceptibility of six tomato cultivars to the root-knot nematode, *Meloidogyne incognita*

Sunil K. Singh¹ and Uma R. Khurma¹

¹School of Biological, Chemical and Environmental Sciences, Faculty of Science and Technology
The University of the South Pacific, Suva, Fiji
khurma_u@usp.ac.fj

ABSTRACT

Six tomato cultivars Moneymaker, Beefsteak, Roma, Summertaste, Mini Roma and Smallfry were tested for their susceptibility to root-knot nematodes at inoculum levels of 200, 400, 600 Juveniles (J2) per pot. All were found to be susceptible to varying degrees as egg masses were present in all with Moneymaker and Roma being the most susceptible and Mini Roma, the least susceptible. The inoculum levels had a significant effect ($p<0.05$) on the number of galls and plant weights. The gall numbers and plant weights was negatively correlated, with the highest gall numbers and lowest plant weights recorded at the highest inoculum level in all cultivars except in Mini Roma in which there was little variation in gall numbers and plant weights.

Keywords: *Lycopersicon esculentum*, Resistance, inoculum level

1 INTRODUCTION

Root-knot nematodes infect a wide range of important crop plants and are particularly damaging to vegetable crops in tropical and subtropical countries (Sikora and Fernandez 2005). There are more than 90 described species in the genus *Meloidogyne* but the four most commonly occurring species are *Meloidogyne incognita*, *M. arenaria*, *M. javanica* and *M. hapla* (Sasser and Taylor 1978; Karssen 2000; Hunt *et al.* 2005). The short life cycle of 6 to 8 weeks enables root-knot nematode populations to survive well in the presence of a suitable host and their populations build up to a maximum usually as crops reach maturity (Shurtleff and Averre 2000). Host plants have varying degrees of susceptibility with some plants being highly susceptible while others are less susceptible or resistant to root-knot nematodes. The highly susceptible host plants allow the juveniles to enter the roots, reach maturity and produce many eggs while the resistant plants suppress their development and thus do not allow reproduction (Sasser and Taylor 1978; Karssen and Moens 2006). In case of severe infections and lack of appropriate control measures the yield loss is high and in some cases the plants die even before reaching maturity.

Root-knot nematodes (RKN) are one of the major pathogens of tomatoes worldwide and limit fruit production (Sikora and Fernandez 2005). Plant resistance is one of the most environmentally safe and economically viable means of controlling RKN. The RKN resistant crop varieties have comparatively better crop yield than the infected susceptible crop varieties. The resistant varieties

can be used as part of integrated pest management in combination with other methods such as use of chemical nematicides, organic soil amendments, heat treatment, soil solarization, and crop rotation with non hosts for controlling RKN. Several cultivars of tomatoes such as Montelle, Sun6082, Pik Red, Celebrity, Baja, Betterboy and Beefmaster have been developed in an attempt to produce RKN resistant cultivars (Milligan *et al.* 1998; Tisserat 2006). Tomato cultivars have varying degrees of resistance to RKN and difference in quality and quantity of fruit production. The *Mi* gene originally found in wild tomato species *Lycopersicon peruvianum* is one of the best characterized nematode resistance genes and has been genetically engineered into many commercial tomato varieties (Nono-womdim *et al.* 2002; Abad *et al.* 2003). The resistance mechanism in response to invasion by RKN involves the formation of necrotic cells at the infection site to prevent the juveniles from developing any further. However a high level of genetic variability of RKN has led to the existence of races and virulent populations which can reproduce even on plants carrying the resistance genes (Castagnone-Sereno 2006). The RKN population density also affects the yield loss and tolerance levels of the different tomato cultivars. This investigation is an attempt to identify the natural resistance of six tomato cultivars commonly available in Fiji, namely, Moneymaker, Beefsteak, Roma, Summertaste, Miniroma and Small fry, to the RKN *M. incognita* and determine the effects of varying population levels on host plant health.

Table 1. Susceptibility of tomato cultivars to varying RKN inoculum levels

Cultivar	Inoculum level	Av. Gall No.	Av. Egg Mass	Av. Plant Weight	Av. % Wt Reduction compared to control
Moneymaker	control	0	0	7.46	0
	200 J2	16	10	6.05	18.9
	400 J2	21	12	5.99	19.71
	600 J2	26	12	4.81	35.52
Roma	control	0	0	8.67	0
	200 J2	13	6	7.4	14.65
	400 J2	19	11	6.66	23.18
	600 J2	26	17	5.92	31.72
Summertaste	control	0	0	3.41	0
	200 J2	9	3	2.73	19.94
	400 J2	13	3	2.3	32.55
	600 J2	22	8	2.18	36.07
Small fry	control	0	0	8.65	0
	200 J2	11	5	7.79	9.94
	400 J2	15	9	6.99	19.19
	600 J2	21	12	5.9	31.79
Beefsteak	control	0	0	3.09	0
	200 J2	12	7	2.85	7.77
	400 J2	15	8	2.31	25.24
	600 J2	20	11	2.01	34.95
Mini Roma	control	0	0	2.56	0
	200 J2	5	1	2.33	8.98
	400 J2	6	1	2.37	7.42
	600 J2	6	2	2.34	8.59

2 METHODS

The root-knot nematode *Meloidogyne incognita* (Kofoid and White) was used for the experiments. The RKN culture was initiated from single egg masses and propagated on tomato cultivar, Moneymaker in the glass house at The University of the South Pacific. Egg masses from the infected host plant were hand picked and placed in distilled water kept in a BOD incubator at $28 \pm 1^\circ\text{C}$ to obtain the second stage juveniles (J2). The juvenile suspension was standardised to final concentration of 100 juveniles per millilitre of distilled water.

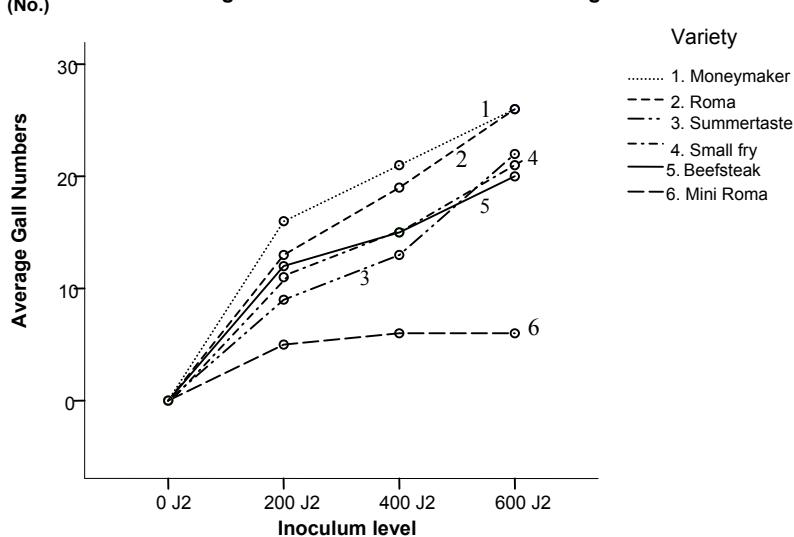
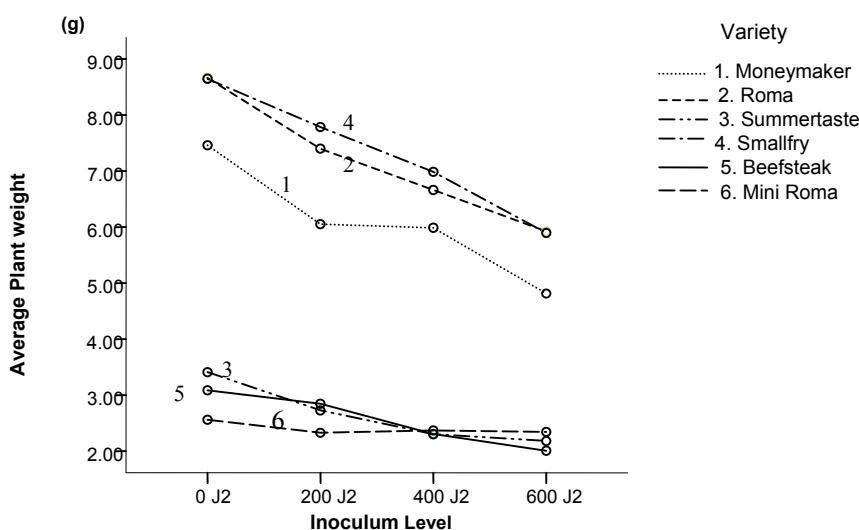
Experiments were conducted in 2 litre pots each containing 900 grams of autoclaved soil. A three week old tomato seedling was planted into each of the pots for each of the six cultivars: Moneymaker, Beefsteak, Roma, Summertaste, Mini Roma and Smallfry. The J2 inoculum was added 1 day after transplanting at the rate of 200, 400 and 600 J2 per pot using 1 ml micropipette into two wells near the roots. Four replicates were kept for each of the inoculum levels including a control without any inoculation (0 J2). The pots were arranged in a random block design and kept in the glass house and watered once every second day. The experiments ran for 8 weeks starting from the second week of December 2006. All the varieties are recommended to be grown in the summer period by the supplier.

The plants were carefully dug out at the end of the eighth week and gently washed to remove soil from the roots. The plant roots were examined under a stereoscopic

microscope (Olympus Model SZ 50) and the number of galls and egg masses present were recorded. The fresh plant weights (shoot plus roots) were also recorded for each of the plant. Average values are recorded in Table 1. The results were subjected to Univariate ANOVA and trends plotted using SPSS software Version 13.0 (Figures 1, 2 and 3).

3 RESULTS

All the six tomato cultivars were susceptible to root-knot nematodes to varying degrees. Egg masses were recorded in all six cultivars thus indicating that none of the tested cultivars was resistant to RKN. A direct relationship was observed between the gall numbers and the inoculation level for all cultivars except for Mini Roma which had relatively little variation in gall number and plant weight with increasing inoculum levels (Figure 1). The plant weights in all cultivars except Mini Roma decreased as compared to the controls with increasing inoculum levels (Figure 2). The greatest percentage reduction in plant weight compared to the control was observed for the cultivar Summertaste while Mini Roma had the least variation in the percentage plant weight loss (Figure 3). Mini Roma was found to be the least susceptible cultivar while Moneymaker and Roma were found to be the most susceptible cultivar having the highest gall numbers. The inoculum level was found to have significant effect ($p < 0.05$) on the number of galls and plant weight.

Figure 1: Effect of inoculum levels on gall numbers**Figure 2. Effect of inoculum level on plant weight**

4 DISCUSSION

The susceptibility of a plant to RKN depends on the ability of RKN juveniles to penetrate the roots of the plant and cause the formation of giant cells which appears as knots (galls) on the roots (Chen and Dickson 2004). The juveniles feed and moult twice before developing into the adult stage (Siddiqi 2000). The adult female RKN stays inside the giant cells and continues to feed and produces egg mass in a gelatinous matrix protruding out of the root gall. The egg masses give rise to infective juveniles (J2) which may infect other uninfected roots of the same plant or migrate and infect the nearby plants. In case of a plant resistant to RKN, the juveniles are either unable to penetrate the roots, or die after penetration or are unable to complete their development, or females are unable to

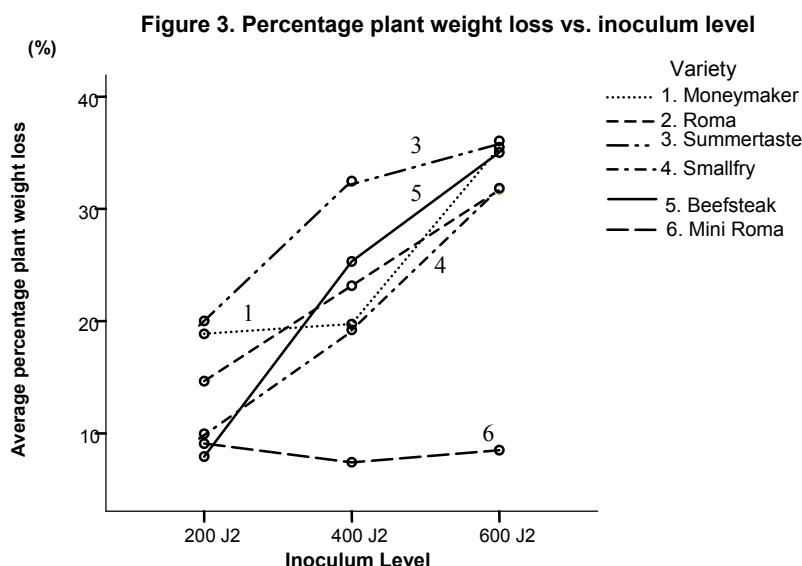
reproduce. The *Mi* gene confers resistance by localized tissue necrosis around the region where the juveniles penetrate, thus juveniles are unable to establish feeding sites resulting in their death or migration out of the roots (Milligan *et al.* 1998; Lo'pez-Pérez *et al.* 2006).

The presence of root galls along with egg mass on all six varieties of tomato plants indicates that none of the varieties is resistant to root-knot nematodes. However significant differences in the number of galls present on each of the six varieties indicate different levels of susceptibility. The level of susceptibility is controlled by the presence of resistance genes such as *Mi* gene and genetic background of the tomato cultivar (Castagnone-Sereno 2006; Jacquet *et al.* 2005). The homozygous or heterozygous state of the *Mi* locus has been found to affect

the degree of resistance to RKN, with the cultivars having the heterozygous form of the *Mi* gene being more susceptible than the homozygous cultivars (Jacquet *et al.* 2005). The variation in the susceptibility to RKN in the six tomato cultivars screened is likely to be due to the genetic differences between the cultivars and thus explains the variation in gall numbers and egg masses. Moneymaker and Roma were found to be the most susceptible as greatest number of juveniles penetrated and completed their development to maturity as shown by the high gall numbers and egg masses present. Mini Roma was the least susceptible variety as only a limited number of juveniles were able to penetrate, develop to maturity and lay egg masses.

Increasing inoculum levels led to an increase in the number of galls with greater reduction in plant weight for all varieties except for Mini Roma. This shows that when the inoculum levels are high, greater number of juveniles are able to infect the plant roots which results in reduced nutrient and water uptake by the roots and consequently poor plant growth (Karssen and Moens 2006). In Mini Roma, even higher inoculum (600 J2) could not establish a larger population, indicating the presence of some genetic resistance and consequently insignificant decrease in plant weight with increasing inoculum levels. Further trials with higher inoculum levels or planting in heavily infested soil would give a more conclusive result about the susceptibility of each cultivar and the effect of RKN on yield quantity and quality.

The variability in pathogenicity is also dependent on the genetic variability of RKN population and species composition but since the populations used for inoculation were raised from a single egg mass under the same conditions and host plants it is likely to have had minimal effect in this investigation. There is sophisticated interaction between the host plant and root-knot nematodes and a number of studies have found resistance breaking pathotypes of RKN that are able to parasitize even RKN resistant plants (Jacquet *et al.* 2005; Abad *et al.* 2003; Baicheva *et al.* 2002) which is a major limiting factor in using plant resistance as a means for controlling root-knot nematodes. However, identification and use of RKN resistant and tolerant varieties can still be a viable means of minimising loss caused by RKN. Another factor which needs to be taken into consideration in any further investigation is the quality and quantity of fruit production of the resistant, less susceptible and highly susceptible varieties because at times the resistant varieties do not produce fruit with the desirable taste and quality (Lo'pez-Pérez *et al.* 2006). Grafting of desired varieties on the roots of the less susceptible and resistant varieties is an alternative but requires technical knowledge and has additional costs associated with getting the grafted plants to the farmers. The susceptibility of the different tomato varieties has important implications on the yield and economic returns thus information on susceptibility to RKN can be useful to farmers while selecting the variety for planting on RKN infested fields.



REFERENCES

- Abad, P., Favery, B., Rosso, M. and Castagnone-Sereno, P. 2003. Root-knot nematode parasitism and host response: Molecular basis of a sophisticated interaction. *Molecular Plant Pathology*. **4**:217-224.
- Baicheva, O., Salkova, D. and Palazova, G. 2002. Root-knot nematodes (*Meloidogyne Goeldi*, 1987) – species composition, pathogenicity, some problems for investigation. *Experimental Pathology and Parasitology*. **5**:21-24.
- Castagnone-Sereno, P. 2006. Genetic variability and adaptive evolution in parthenogenetic root-knot nematodes. *Heredity*. **96**:282-289.
- Chen, Z.X., Chen, S.Y. and Dickson, D.W. 2004. Nematology advances and perspectives Vol. 2, *Nematode Management and Utilization*. Tsinghua University press China, 636pp.
- Hunt, D.J., Luc, M. and Manzanilla-Lopez, R.H. 2005. Identification, morphology and Biology of Plant Parasitic nematodes. In: Luc, M., Sikora, R.A. and Bridge, J. (Eds). *Plant parasitic nematodes in subtropical and tropical agriculture*. 2nd edition, CABI publishing, pp. 11-52.
- Jacquet, M., Bongiovanni, M., Martinez, M., Verschave, P., Wajnberg, E. and Catagnone-Sereno, P. 2005. Variation in resistance to the root-knot nematode *Meloidogyne incognita* in tomato genotypes bearing the *Mi* gene. *Plant pathology*. **54**:93-99.
- Karssen, G. 2000. *The Plant-parasitic Nematode Genus Meloidogyne Goeldi, 1892 (Tylenchida) in Europe*. Brill Academic Publishers, Leiden, The Netherlands, 160 pp.
- Karssen, G. and Moens, M. 2006. Root-knot nematodes. In: Perry, R.N. and Moens, M. (Eds). *Plant Nematology*. CABI publishing, pp 59-90.
- Lo'pez-Perez, J-A. Strange, M.L., Kaloshian, I., and Ploeg, A.T. 2005. Differential response of *Mi* gene resistant tomato rootstocks to root-knot nematodes (*Meloidogyne incognita*). *Crop protection*. **25**:382-388.
- Milligan, S.B., Bodeau, J., Yaghoobi, J., Kaloshian, I., Zabel, P. and Williamson, V.M. 1998. The root knot nematode resistance gene *Mi* from Tomato is a member of the Leucine Zipper, Nucleotide binding Leucine-rich repeat family of plant genes. *The Plant Cell*. **10**:1307-1319.
- Nono-Womdim, R., Swai, I.S., Mrossou, L.K., Chadha, M.L. and Opena, R.T. 2002. Identification of root-knot nematode species occurring on tomatoes in Tanzania and resistant lines for their control. *Plant Disease*. **86**:127-130.
- Sasser, J.N. and Taylor, A.L. 1978. *Biology, identification and control of root-knot nematodes (Meloidogyne species)*. Raleigh, NC: North Carolina State University Graphics, 111 pp.
- Shurtleff, M.C. and Averre, C.W.III. 2000. *Diagnosing plant disease caused by plant parasitic nematodes*. The American phytopatological society, 187 pp.
- Siddiqi, M. R. 2000. *Tylenchida Parasites of plants and insects*. 2nd Edition, CABI Publishing, 833pp.
- Sikora, R.A. and Fernandez, E. (2005). Nematode parasites of vegetables. In: Luc, M., Sikora, R.A. and Bridge, J. (Eds). *Plant parasitic nematodes in subtropical and tropical agriculture*. 2nd edition, CABI publishing, pp. 319-392.
- Tisserat, N. 2006. Root knot nematode of tomato. *Fact sheets tomato*. Extension plant pathology –Kansas State University, Manhattan.
<http://www.plantpath.ksu.edu/pages/extention>.