

## Susceptibility of *Vitis vinifera* 'Semillon' and 'Chardonnay' to the root-knot nematode *Meloidogyne javanica*

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### Summary

A study to assess the effect of the initial population (Pi) densities (0, 200, 400, 600 and 800 second stage juveniles ( $J_2$ )  $\text{kg}^{-1}$  dry soil) of the root knot nematode, *Meloidogyne javanica*, on the growth, yield and juice characteristics of two white wine grapevine (*Vitis vinifera*) cvs. 'Semillon' and 'Chardonnay' was conducted in a vineyard located at the Centre for Irrigated Agriculture, Riverina, NSW, Australia. *M. javanica*  $J_2$  population densities in soil after harvest during 2004–2008 growing seasons increased gradually, year by year, and in most cases were higher where the initial densities were higher. Regression analysis revealed that yield, in general, was reduced significantly with the increase of the nematode population densities  $\cdot\text{kg}^{-1}$  soil for both cultivars. After six years, the nematode population had increased by ca. 9.0–22.4 fold for 'Semillon' and 6.7–18.5 fold for 'Chardonnay'. All Pi densities significantly reduced Semillon yields in all years but only the highest level (800  $J_2$   $\cdot\text{kg}^{-1}$  dry soil) affected 'Chardonnay' yields. At the end of the experiment, *M. javanica* decreased yields by 15–20 % in Semillon but only 7–13 % in 'Chardonnay'. The nematode inoculation also caused a decrease in bunch numbers in 'Semillon' but not in 'Chardonnay'. This is the first study showing that 'Chardonnay' is less susceptible to *M. javanica* than 'Semillon'.

**Key words:** Chardonnay, juice characteristics, *Meloidogyne javanica*, pruning weight, Semillon, *Vitis vinifera*, yield.

### Introduction

Production losses (5–15 %) caused by plant parasitic nematodes in grapevine (*Vitis vinifera*), have been reported in most grape-growing districts of Australia (WALKER and STIRLING 2008). According to the Australian Grape & Wine Research & Development Corporation (GWRDC) the economic impact of nematode damage has been estimated to be \$14 million *pa* across the Australian wine grape industry (SCHOLEFIELD and MORISON 2010). Several researchers have also demonstrated the impact of nematode damage by reporting that grape yields were increased after application of nematicides or fumigants, and/or planting tolerant or resistant rootstocks in nematode infected vineyards (MEAGHER 1969, SAUER 1972, 1974, CIRAMI *et al.* 1984, HARRIS 1986, WALKER 1989, EDWARDS 1989, 1991).

Among the plant parasitic nematodes detected in Australian vineyards, four root knot nematode species (*Meloidogyne javanica*, *M. hapla*, *M. incognita* and *M. arenaria*) were considered as the major problem, with being *M. javanica* predominant in many viticultural regions (SAUER 1962, MEAGHER 1969, STIRLING 1976, MCLEOD 1978) and responsible for the yield loss up to 60 % in some grapevine cultivars (SAUER 1974). However, MCKENRY *et al.* (2001) have suggested that the extent of yield loss caused by root knot nematode may vary depending on the virulence of the *Meloidogyne* species, population density in the soil, susceptibility of the grapevine cultivar and, the severity and duration of infection.

The initial population (Pi) density of *Meloidogyne* species has a significant influence on the extent of yield loss in grapevine cultivars (MCKENRY *et al.* 2001). In Australia, QUADER *et al.* (2002) demonstrated from a microplot experiment that the *M. incognita* initial population (1–25 second stage juvenile ( $J_2$ ) / 100 mL soil) affected the top growth of the susceptible cultivar 'Colombard'. On the other hand, EDWARDS (1991) suggested that for significant grapevine damage the *M. javanica* population must be more than 500  $J_2$  / 500 g soil, although experimental evidence on the extent of yield loss, associated with this population, has not been reported. As grapevines are perennial, they are long term hosts for *Meloidogyne* spp. and any initial population at planting has the potential to build up to damaging level over time that will impact on vine growth and productivity.

'Chardonnay' and 'Semillon' are two popular and premium white wine grape cultivars in Australia that occupied ca. 19 % and 4 % of total bearing areas, and produced ca. 23 % and 5 % of total wine grapes respectively during 2008–2009 growing season (GUNNING-TRANT 2010). The Riverina of south-eastern New South Wales (NSW) is one of the major white wine grape growing regions in Australia where the majority of the vineyards infected with *M. javanica* are planted with own rooted 'Chardonnay' and 'Semillon' (MCLEOD 1978). In this region, many vignerons have noticed that 'Semillon' is more susceptible to the root-knot nematodes than 'Chardonnay', with 'Semillon' showing greater yield decline from year to year. Therefore, an experiment was initiated in a Riverina vineyard to assess the relationships among the initial population (Pi) densities of *M. javanica*  $J_2$   $\cdot\text{kg}^{-1}$  dry soil at planting and growth performances, yield and juice characteristics of 'Semillon' and 'Chardonnay'.

## Material and Methods

**Experimental site:** The experiment was performed in two phases: the first phase was conducted in a glasshouse and a net-house, simulating natural environment with bird exclusion, located at the National Wine and Grape Industry Centre, Wagga Wagga (35°05'S; 147°20'E) and the second phase of the experiment was conducted in a vineyard located at the Centre for Irrigated Agriculture, Riverina (34°17'S; 146°03'E), NSW. In the first phase, grapevine rootlings were prepared and inoculated with *M. javanica* J<sub>2</sub> in pots and in the second phase the inoculated rootlings were planted in a vineyard where they grew until the experiment was completed.

**Propagation of grapevine rootlings:** Initially, 50 cuttings with four buds each were obtained from one year old canes of 'Semillon' (clone DA 16162) and 'Chardonnay' (clone F1B3) during the dormant season (July-August) in 2000 and stored in a cool room at 4 °C. At the start of spring, in September, the cuttings were placed in a polystyrene box filled with moist perlite for rooting. After satisfactory root development, the rootlings were planted individually in 15 cm diameter plastic pots filled with 1.25 kg moist soil (1:1 sand and loam) pasteurised at 60 °C for one hour, equivalent to ca. 1 kg dry soil, in mid-December, 2000. The pots were placed on a 2.5 x 1.5 m metal tray in the glasshouse and drip irrigated (2 L·hour<sup>-1</sup> at three times for 5 min·day<sup>-1</sup> *i.e.* 500 mL·day<sup>-1</sup>) until June 2001. The pots were then taken outside to defoliate naturally when the ambient temperature varied between 4.0-15.1 °C at the site. After defoliation, the grapevines were pruned back to one shoot with two buds each. When bud burst was completed in spring, September to November, 30 grapevines from each cultivar with approximately similar vigour were selected for the experimental purpose. These grapevines were grouped into 5 batches of six vines per cultivar, placed on another metal tray in the net-house, irrigated three times a day (10 min each time) at 2 L·hour<sup>-1</sup> and were allowed to grow until planted in the vineyard.

**Inoculation of the grapevine rootlings with *M. javanica*:** *M. javanica* J<sub>2</sub> were extracted from infected tomato roots (*Solanum lycopersicum* cv. Grosse Lisse) following the method described by RAHMAN *et al.* (2011) and grouped into batches of 200, 400, 600 and 800 J<sub>2</sub>·mL<sup>-1</sup> of extraction. Individual grapevine rootlings of each batch of both cultivars were inoculated, in the first week of January 2000, with either 0 (control, water only), 200, 400, 600 and 800 J<sub>2</sub>. These grapevines were irrigated as above, fertilised with a slow release fertiliser Osmocote® (total N 13.8 %, P 3.5 % and K 8 %) at 3 g·pot<sup>-1</sup> twice per year and were allowed to grow until bud burst in spring, 2002. At this time, they were transferred to the drip irrigated vineyard and planted in two rows at the Centre for Irrigated Agriculture, Riverina, NSW. The rows were 2.5 m apart; one with 32 marked vine panels and the other with 28 panels. Each vine panel was 5 m long and had two vine planting positions with 3 m spacing. The annual rainfall recorded at the trial site was 235, 413, 166 and 332 mm in 2004, 2005, 2006 and 2007 respectively.

**Pre-planting nematode assessment in soil:** Two 200 g subsamples of soil from 10 vine planting spots in each row were collected in the first week of September in 2002 and processed for nematode extraction using the Whitehead tray method (WHITEHEAD and HEMMING 1965). After 5 days, suspension from each extraction tray was sieved twice using a 15 µm nylon sieve and back washed to collect the nematode in a 70 mL plastic container, and then counted under a dissecting microscope.

**Pre-planting treatment of soil with Nematicur® 400 EC:** In mid September 2002, a pit of ca. 25 cm width and ca. 20 cm depth was made at each planting location and was treated with 5 L Nematicur® 400 EC solution prepared from 0.25 mL of the original concentrated formulation (equivalent to 5.1 mL Nematicur® 400 EC/m<sup>2</sup> approximately). Two weeks after the treatment, soil from each pit was collected again to evaluate the presence of plant parasitic nematodes.

**Planting grapevine rootlings:** Three weeks after the Nematicur treatment, the vine rootlings were pulled out of the pot with minimal disturbance of soil and replanted in the Nematicur® 400 EC treated pits. Planting was done following a three replicate split plot design with cultivar (2) randomised to main plots (vine rows) and treatments as initial population (Pi) densities of *M. javanica* J<sub>2</sub> (5) randomised to subplots (panel) within main plot (vine rows). Each replicated sub-plot represents a panel consisting of two vines of the same cultivar with 2.5 (row to row) and 3.0 m (vine to vine) spacing.

**Soil sampling:** Soil samples were collected from undervine positions within two weeks after harvest in February of each year during 2004-2008 growing seasons. Two samples at a depth of ca. 25 cm and at a distance of ca. 25 cm from the vine trunk, one from either side of both vines in each panel, were collected using a narrow ended shovel, and bulked. A soil sub-sample of ca. 800 g/panel (replication) was taken to the laboratory for nematode assessment and soil moisture determination. Samples were stored for 3-5 days in a cool room at 4 °C until processing.

**Nematode extraction:** Two 200 g sub-samples of soil from each replication were processed for nematode extraction using the method described above. Nematode populations from two duplicate samples from each replication were averaged and numbers were calculated to population density kg<sup>-1</sup> dry soil by using the soil moisture percentage (oven dried at 105 °C for two days) recorded from each sample in each season.

**Yield, bunch numbers and 50 berry weights:** On the day of harvest, 50 berries from the two grapevines of both cultivars in each replicated plot (panel) were collected randomly to determine the juice characteristics. Then, all grape bunches in each replicated plot (panel) were hand picked, counted and weighed. The 50 berries weight was included in the total berry weight of the respective replication of each treatment.

**Measuring pruning weight and trunk circumference:** Shoots of each grapevine were pruned back to two buds by the end of July, winter, during

each of the growing seasons, and weighed. At the end of the experiment in 2008, trunk diameter of each grapevine along the irrigation line (ca. 15 cm above the soil level) was measured on the same day of pruning, using a digital slide calliper (Mitutoyo Corp., Model No. CD-6" C, Japan), and converted to circumference.

**Juice characteristics:** Juice of 50 berries, from each replication of the different treatments, was extracted separately using a commercial juicer (Panasonic, model no. MJ 66 PRA, Japan). °Brix from juice was recorded immediately using a refractometer (PR 32 Palette, serial no. 0165152, ATAGO, Japan). Juice was refrigerated for 2-4 weeks and then titratable acidity (TA) and pH were measured using the method Tim Talk8 Titration manger program (Radiometer Copenhagen, Radiometer Analytical S.A., Lyon, France).

**Data analysis:** Linear mixed models fitted using ASREML (GILMOUR *et al.* 2005) with treatment (Pi density), cultivar, year and their interactions as fixed effects and blocking structures of replicate, main plot and sub plot as random effects were used to model traits. Nematode populations were transformed into  $\text{Log}_e[M. javanica J_2 + 1]$  to meet normality assumptions.

Bivariate linear mixed models were fitted to pairs of traits. These pairs included  $\text{Log}_e[M. javanica J_2 + 1]$  and one of yield, number of bunches/vine, weight of 50 berries, pruning weight and juice characteristics. In each model, trait, trait x year, trait x cultivar and trait x year x cultivar were fitted as fixed effects while trait x treatment, trait x cultivar x treatment, trait x year x treatment and trait x cultivar x year x treatment were fitted as random effects. Blocking structures from the designed experiment including replicate, main plot and sub plot for each trait were fitted as random effects. To model the relationship between the two traits at the treatment mean level, correlation structures were fitted at various levels of the model, namely, the overall mean level (trait x treatment), the cultivar level (trait x cultivar x treatment), the year level (trait x year x treatment) and the interaction level (trait x cultivar x year x treatment). The resulting regressions fitted to treatment means are presented in this article.

## Results

**Pre-planting populations of *M. javanica* in soil:** *M. javanica* was absent from the soil but a few (< 20/kg dry soil) root lesion nematodes (*Pratylenchus* spp.) were present (data not shown). However, no *Pratylenchus* spp. was recoded after soil treatment with Nematicur® 400 EC.

***M. javanica* J<sub>2</sub> Population changes in soil after planting inoculated grapevines:** *M. javanica* J<sub>2</sub> populations in soil after harvest in each February, from 2004 to 2008, varied significantly considering the effect of Pi densities ( $P < 0.001$ ), cultivar ( $P = 0.003$ ), year ( $P < 0.001$ ) and their two or three way interactions ( $P < 0.001$ ) (Tab. 1). In Semillon, populations generally increased over the six year period with minor decreases in years 2005 and 2008 (Fig 1 a) whereas in Chardonnay it increased steadily until 2006 and then decreased in years 2007 and 2008 (Fig. 1b). At the end of the experiment in 2008, the populations increased by ca. 9.0-22.4 and 6.7-18.5 fold in Semillon and Chardonnay respectively (Tab. 2, Fig. 1 a, b).

**Grape yield:** Grape yields were significantly affected by *M. javanica* J<sub>2</sub> Pi densities, year (Y), cultivar (C) x Pi and C x Y either at  $P = 0.013$  or  $P < 0.001$  (Tab. 1). It was observed that uninoculated control Semillon vines had higher yields than the inoculated vines in every year (Tab. 2). For 'Chardonnay', grape yields were significantly lower than the control at Pi = 800, when field populations kg<sup>-1</sup> dry soil varied between 664 (2004) and 14,764 (2008), and at Pi = 600 in 2005 and 2007 (Tab. 2). The inoculated 'Semillon' and 'Chardonnay' vines, throughout the trial period, produced 4.7-6.4 kg and 1.4-2.5 kg less grapes, respectively, when compared with their uninoculated control vines. This is equivalent to ca. 6.3-8.5 and 1.9-3.3 t·ha<sup>-1</sup> yield loss in 'Semillon' and 'Chardonnay' respectively with 2.5 (row-row) x 3 (vine-vine) m vine spacing. At the end of the experiment in 2008, inoculated grapevines produced ca. 15-20 % and 7-13 % less yield, respectively, in 'Semillon' and 'Chardonnay' when compared with the uninoculated control. These yield losses were associated with

Table 1

*P*-values for initial population (Pi) densities of *Meloidogyne javanica* (*M.j.*) kg<sup>-1</sup> dry soil, grapevine cultivars, year and their interactions on root-knot nematode populations kg<sup>-1</sup> dry soil in vineyard and associated grapevine yield and yield parameters, during 2004-2008 growing seasons, Riverina, NSW, Australia

Source of variation	<i>P</i> - values					
	<i>M. j.</i> kg <sup>-1</sup> dry soil	Yield (kg)	No. of bunches	50 Berry weight (g)	Pruning weight (g)	Trunk circum. (cm), 2008
Pi densities	<0.001	<0.001	<0.001	0.792	0.266	0.396
Cultivar (C)	0.003	0.063	<0.001	0.005	0.189	0.413
Year (Y)	<0.001	<0.001	<0.001	<0.001	<0.001	-
Pi x C	<0.001	0.013	0.01	0.986	0.778	0.282
C x Y	<0.001	<0.001	0.012	<0.001	0.778	-
Pi x Y	<0.001	0.235	0.634	0.881	0.913	-
Pi x C x Y	<0.001	0.569	0.96	0.356	0.746	-

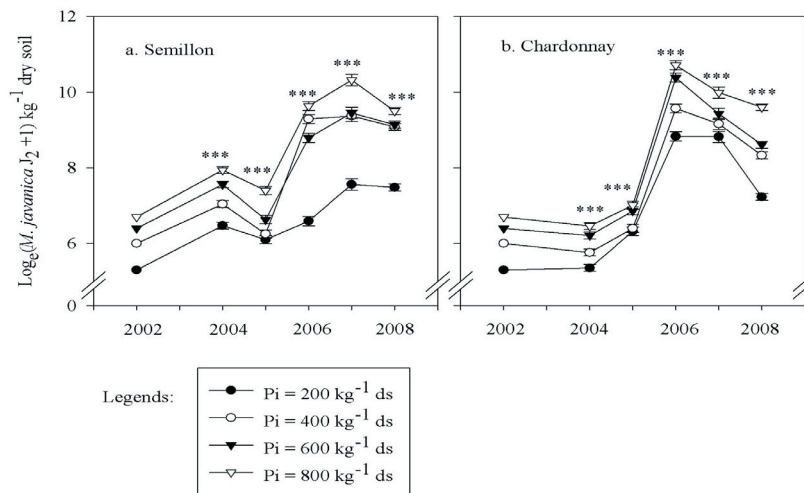


Fig. 1: Initial population density (Pi) of *Meloidogyne javanica*  $J_2$   $kg^{-1}$  dry soil changes during 2002-2008 growing seasons in vineyard, Riverina, NSW, Australia. Each data point is the average of 3 replications; Bar =  $\pm$  SE; \*\*\* indicates significant difference at  $P < 0.001$ ; ds = dry soil; Data points, in 2002, represent the Pi densities used for inoculating the grapevine rootlings; no data was collected in 2003.

Table 2

Fresh grape yield (kg / grapevine) of 'Semillon' and 'Chardonnay' under different initial population (Pi) densities of *Meloidogyne javanica*  $J_2$  (*M. j.*)  $kg^{-1}$  dry soil, 2002, along with the corresponding population densities  $kg^{-1}$  dry soil at harvest<sup>a</sup> during 2004-2008 growing seasons, Riverina, NSW, Australia

Pi density	Semillon									
	2004		2005		2006		2007		2008	
	<i>M.j.</i> <sup>a</sup>	Yield*	<i>M.j.</i>	Yield	<i>M.j.</i>	Yield	<i>M.j.</i>	Yield	<i>M.j.</i>	Yield
0	0	13.8 b	72	16.9 b	0	31.1 b	0	23.1 b	0	31.5 b
200	664	9.0 a	445	12.1 a	734	26.3 a	1997	18.3 a	1807	26.8 a
400	1096	8.1 a	544	11.2 a	10937	25.3 a	12087	17.4 a	8954	25.8 a
600	1997	7.8 a	734	10.9 a	6633	25.1 a	13359	17.1 a	8954	25.6 a
800	2696	7.4 a	1635	10.5 a	14764	24.7 a	9731	16.7 a	13359	25.2 a
Chardonnay										
0	0	10.3 b	71	14.8 b	0	17.8 b	0	17.5 b	0	18.8 b
200	199	8.7 ab	544	13.2 ab	6633	16.2 ab	6633	15.9 ab	1338	17.2 ab
400	229	8.9 ab	601	13.4 ab	14764	16.4 ab	9896	16.0 ab	4023	17.4 ab
600	492	8.3 ab	991	12.8 a	2859	15.7 ab	12087	15.4 a	5431	16.8 ab
800	664	7.9 a	1096	12.4 a	4355	15.4 a	2025	15.0 a	14764	16.4 a
LSD (5 %)		1.95		1.94		2.45		1.84		2.18

\* Yield comparisons have been made within a cultivar. Predicted values (average of 3 replications) in a column with the same letter(s) do not differ significantly at 5%.

1,807-13,359 and 1,338-14,764 *M. javanica*  $J_2$   $kg^{-1}$  dry soil at harvest, respectively, in 'Semillon' and 'Chardonnay' vines (Tab. 2). Bivariate analysis for the relationship among overall density of  $\text{Log}_e[M. javanica J_2 + 1]$   $kg^{-1}$  dry soil and yield at the mean level of Pi densities (averaged across year and cultivar) indicated that yields were reduced significantly with the increase of the nematode population densities (Fig. 2). When all five Pi densities (0, 200, 400, 600, 800) were included in the regression analysis, 'yield = 20.083-0.536\* $\text{Log}_e[M. javanica J_2 + 1]$ ', the correlation was -0.998 (Fig 2 a). When only four inoculated treatments (200, 400, 600, 800) were considered, the correlation across the treatment means (Pi densities) was -0.993 which also gave a significant regression equation of 'yield = 24.171- 1.056\* $\text{Log}_e[M. javanica J_2 + 1]$ ' (Fig. 2 b). Bi-

variate analysis for the relationship between  $\text{Log}_e[M. javanica J_2 + 1]$   $kg^{-1}$  dry soil and yield of a cultivar (averaged across 5 years for Pi=200, 400, 600 and 800) also showed that yields for both cultivars declined significantly with increasing  $\text{Log}_e[M. javanica J_2 + 1]$   $kg^{-1}$  dry soil (Fig. 3). The uninoculated control treatment for 'Semillon' (Pi = 0) fitted well with this linear trend (Fig. 3 b) but it was a poor fit to the trend line for 'Chardonnay' indicating the relationship for 'Chardonnay' is not linear (Fig 3 a).

Bunches per grapevine: The number of bunches differed significantly ( $P < 0.05$  or  $P < 0.001$ ) among the Pi densities of nematodes (Pi), cultivar (C), year (Y) and their two way interactions except Pi x Y. The three way interaction (Pi x C x Y) was not significant (Tab. 1). Uninoculated control vines produced significantly more

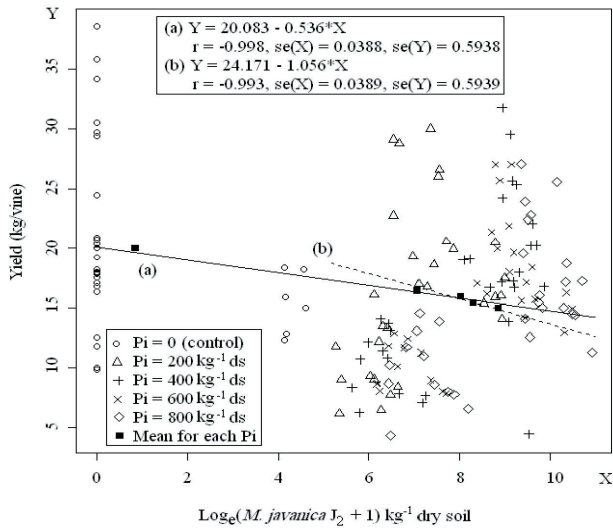


Fig. 2: Relationship among overall population densities of *Meloidogyne javanica*  $J_2$ : $kg^{-1}$  dry soil (averaged across 5 years and two cultivars) and grape yield, showing the difference in the regression when yield of uninoculated control treatment was included (a) or excluded (b). Pi = Initial population density; ds = dry soil.

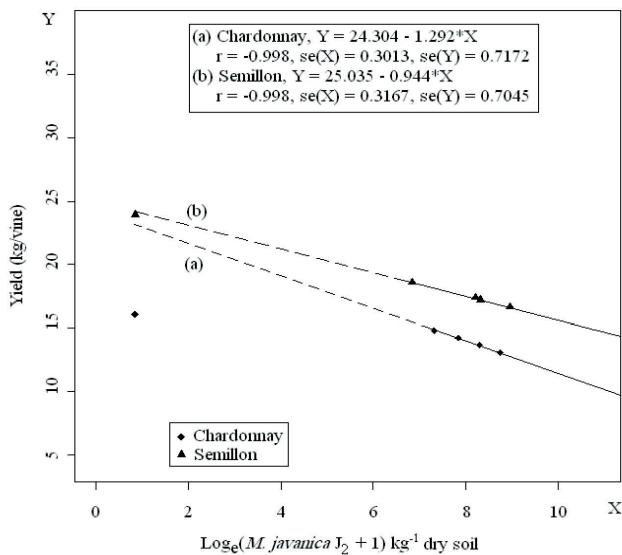


Fig. 3: Relationship among population densities of *Meloidogyne javanica*  $J_2$ : $kg^{-1}$  dry soil and yield (average over 5 years) of grapevine cultivars (a) 'Chardonnay' and (b) 'Semillon'. ds = dry soil.

bunches than the inoculated vines throughout the trial period in 'Semillon' but not in 'Chardonnay'. In most cases, the number of bunches increased, in both cultivars, with the progression of the vine age (Tab. 3).

In 'Semillon', the average number of bunches per vine across five years decreased significantly ( $r = -0.966$ ) with the increase in nematode densities (Number of bunches =  $148.275 - 0.754 * \text{Log}_e[M. javanica J_2 + 1]$  (Fig. 4 b) but did not decrease significantly in 'Chardonnay' (Fig 4 a). A poor fit of the mean of the uninoculated control treatment to trend line was observed in 'Semillon' (Fig. 4 b).

Total weight of fifty grape berries: The total weight of 50 berries did not differ significantly among the *M. javanica*  $J_2$  Pi densities but differed signifi-

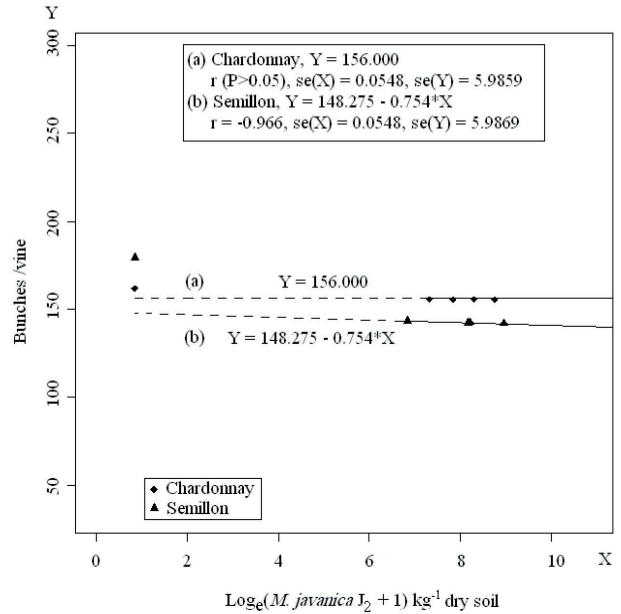


Fig. 4: Relationship among population densities of *Meloidogyne javanica*  $J_2$ : $kg^{-1}$  dry soil and number of bunches/grapevine (average over 5 years) in (a) 'Chardonnay' and (b) 'Semillon'. ds = dry soil.

cantly among the cultivar (C), year (Y) or cultivar x year interaction at  $P < 0.01$  or  $P < 0.001$  (Tab. 1). The three way interaction effect of Pi x cultivar x year was not significant. The average 50 berry weight ranged between 43-78 g and 42-64 g in 'Semillon' and 'Chardonnay', respectively, at various growing seasons (data not shown).

Juice characteristics of grape berries: The Pi densities of *M. javanica* had a significant effect on TA but not on pH and °Brix (Tab. 4). Although, there was significant year effect for all these juice characteristics, varietal effect was only significant for pH and °Brix. At the interaction level, only C x Y had significant effects on °Brix and TA (Tab. 4).

Since the Pi x C x Y interaction was not significant for TA, we examined the effects of C x Y and Pi densities separately. Averaged across Pi densities, 'Semillon' had significantly lower TA than 'Chardonnay', in 2004 and 2008, while 'Chardonnay' had lower TA than 'Semillon' in 2005 (Tab. 5). The effect of Pi densities (averaged across cultivars and years) on TA was significant but not consistent. TA for Pi = 200 (4.59) and Pi = 600 (4.57) were significantly lower than TA for Pi = 0 and Pi = 800 (both TA 4.73, LSD 5 % = 0.11). TA for Pi = 400 (4.68) was not significantly different to either of these TA values (data not shown).

Although *M. javanica*  $J_2$  Pi densities did not have any adverse effects on pH and °Brix, there were differences between cultivars across years. In all years, the pH and °Brix were significantly higher in 'Chardonnay' than 'Semillon'. The averages across the Pi densities for pH and °Brix varied, respectively, between 3.70-3.98 and 18.61-24.14 for 'Chardonnay' and between 3.58-3.86 and 16.39-21.57, respectively, for 'Semillon' (Tab. 5).

Regression analysis on the relationship between  $\text{Log}_e[M. javanica J_2 + 1]$   $kg^{-1}$  dry soil and juice characteristics produced no significant trends either at overall

Table 3

Effect of initial population (Pi) densities of *Meloidogyne javanica* J<sub>2</sub> kg<sup>-1</sup> dry soil, 2002, on the average number of bunches of 'Semillon' and 'Chardonnay', during 2004-2008 growing seasons, in vineyard

Pi density	Number of bunches/ grapevine*									
	Semillon					Chardonnay				
	2004	2005	2006	2007	2008	2004	2005	2006	2007	2008
0	88 b	170 b	196 b	223 b	236 b	80	148	142	213	232
200	48 a	130 a	156 a	183 a	197 a	74	142	136	206	225
400	44 a	126 a	152 a	179 a	192 a	73	141	135	205	224
600	50 a	132 a	158 a	185 a	198 a	73	141	135	206	225
800	46 a	127 a	153 a	180 a	194 a	72	140	134	204	223
LSD (5 %)	17	17	21	19	19	ns	ns	ns	ns	ns

\* Predicted values (average of 3 replications) in a column with the same letter(s) do not differ significantly at 5 %; ns = not significant.

Table 4

*P*-values for initial population (Pi) densities of *Meloidogyne javanica* J<sub>2</sub> kg<sup>-1</sup> dry soil, grapevine cultivars, year and their interactions on juice characteristics

Source of variation	<i>P</i> -value for juice characteristics		
	pH	°Brix	TA gL <sup>-1</sup>
Pi densities	0.226	0.717	0.017
Cultivar (C)	<0.001	0.031	0.120
Year (Y)	<0.001	<0.001	<0.001
Pi x C	0.274	0.796	0.423
C x Y	0.148	<0.001	<0.001
Pi x Y	0.367	0.473	0.407
Pi x C x Y	0.859	0.985	0.794

treatment (Pi densities) or cultivar mean level (data not shown).

## Discussion

Population dynamics of *M. javanica* J<sub>2</sub> in soil: Soil analysis, immediately after grape harvest in each season, indicated that the *M. javanica* J<sub>2</sub> populations kg<sup>-1</sup> dry soil increased from the Pi densities, suggesting that both 'Semillon' and 'Chardonnay' are hosts for *M. javanica*, in accordance with the findings of WALKER *et al.* (1994) and HARDIE and CIRAMI (1988).

The changes seen in *M. javanica* J<sub>2</sub> populations over the trial period of this study may have been caused by the variation in weather conditions, and changes in abundance of natural enemies and competitors in soil from year to year as suggested by JONES and KEMPTON (1978). The grapevine row in this trial was maintained weed free by applying herbicide which may have caused depletion of organic matter and the decrease of the beneficial nematodes in the soil, thus allowing the populations of *M. javanica* to increase. RAHMAN *et al.* (2009) reported that herbicide treated grapevine rows had less beneficial nematodes (bacteria feeders, predators, omnivores and fungal feeders) and consequently more plant parasitic nematodes, than grapevine rows with a permanent sward.

With few exceptions, nematode counts immediately after harvest indicated that soil planted with 'Semillon' had higher *M. javanica* populations than soil planted with 'Chardonnay'. It is likely that this variation is due to differences in susceptibility and physiological traits of the cultivars. Although both cultivars seemed to be good hosts to *M. javanica* in this trial, the differences in physiological plant responses on post-infection nematode development and reproduction are unknown. MCKENRY *et al.* (2001) presented a similar view based on the variation in nematode reproduction and vine response among 18 *Vitis* cultivars that were inoculated with *Meloidogyne* spp. They have suggested that differences in plant physiological responses or defence mechanisms may be the reason for such variation. We have not examined the root populations, which could give a better idea on the varietal response on post nematode development.

Water stress in the plant system may be another physiological trait which might influence nematode invasion, development and reproduction. It has been reported that the transpiration rate is higher in 'Semillon' than in 'Chardonnay', making 'Semillon' more prone to water stress than 'Chardonnay' when grown in similar conditions (ROGIERS *et al.* 2009). It is likely that water stress in 'Semillon' may have weakened the plant's physiological system leading to greater invasion and multiplication of the nematodes in the roots with eventual release into soil. It will be interesting to investigate this aspect in future.

Yield and yield parameters of grapevine cultivars: We found that uninoculated control vines (Pi = 0) produced higher grape yields than the inoculated vines of 'Semillon' and 'Chardonnay' suggesting that *M. javanica* infections have an effect on yield loss in these cultivars. Higher grape yields in vines with no or low infection of nematodes compared with vines with high infections has also been observed by EDWARDS (1991) and SAUER (1972) in own rooted Sultana cultivar. Our results also suggest that the rate of yield loss, caused by *M. javanica* infection, varied between the two cultivars. The final yields of inoculated vines were 15-20 % lower than the uninoculated vines for Semillon, but only 7-13 % lower for 'Chardonnay'. Thus, the two cultivars responded differently to dam-

Table 5

Juice characteristics of 'Semillon' and 'Chardonnay' cultivars

Juice characteristic	Cultivar	Year				
		2004	2005	2006	2007	2008
pH	Semillon	3.86 a	3.58 a	3.85 a	3.72 a	3.58 a
	Chardonnay	3.98 b	3.70 b	3.97 b	3.85 b	3.70 b
	LSD	0.03	0.03	0.03	0.03	0.03
Brix°	Semillon	21.57 a	19.85 a	17.20 a	16.39 a	17.78 a
	Chardonnay	24.14 b	20.72 b	21.59 b	18.61 b	21.48 b
	LSD	1.21	0.81	1.30	0.97	0.96
TA	Semillon	3.64 a	5.30 b	4.27 a	4.72 a	5.13 a
	Chardonnay	4.08 b	4.95 a	4.35 a	4.75 a	5.41 b
	LSD	0.18	0.18	0.18	0.18	0.18

Each value is the average of 15 observations; values with same letter in a column do not differ significantly at 5 %.

age caused by *M. javanica*. Similar cultivar differences in susceptibility to root knot nematode have also been reported for cotton (DAVIS and MAY, 2005). Estimation of soil-borne nematode Pi density can assist growers to adopt appropriate control strategies to minimise yield losses caused by pest nematodes. Results from this study indicate that for Semillon, the initial *M. javanica* densities of 200-800 J<sub>2</sub>·kg<sup>-1</sup> dry soil and 800 J<sub>2</sub>·kg<sup>-1</sup> dry soil for 'Chardonnay' caused significant yield loss. In 2008, six years after planting, the *M. javanica* densities had increased to 1,807-13,359 and 1,338-14,764 kg<sup>-1</sup> dry soil in 'Semillon' and 'Chardonnay' respectively. Our results showed that *M. javanica* J<sub>2</sub> population densities (200 and 800 J<sub>2</sub>·kg<sup>-1</sup> dry soil at planting) in six years increased and caused ca. 15-20 % yield loss in 'Semillon'. In 'Chardonnay', only the Pi = 800 J<sub>2</sub>·kg<sup>-1</sup> dry soil reduced the yield. The difference detected in yield loss between the cultivars revealed that 'Semillon' and 'Chardonnay' respond differently to *M. javanica*. These results indicate that it is very likely that the *M. javanica* population densities between 200 and 800 J<sub>2</sub>·kg<sup>-1</sup> dry soil will increase over 5-6 years period and may cause ca. 7-20 % yield loss depending on the susceptibility of the cultivar. According to MCKENRY (1992), the damaging level for grapevine growth and yield in vineyards in USA, caused by root-knot nematodes should be higher than 500 J<sub>2</sub>·kg<sup>-1</sup> soil.

Yield loss of a particular crop is often expressed by the linear relationship between Log<sub>e</sub>[*M. javanica* J<sub>2</sub> + 1] and yield. Regression analysis of our data showed that yields decreased with the increase of *M. javanica* populations in soil in both cultivars. However the rate of decrease varied between 'Semillon' and 'Chardonnay'. The trend line for four inoculated treatments (Pi = 200, 400, 600, 800) on yield reduction fitted well with the Pi = 0 values in 'Semillon' but not in 'Chardonnay' suggesting a possible curvilinear trend for 'Chardonnay'. Therefore, the 'Semillon' productivity was more affected with the increase of *M. javanica* densities in soil than the Chardonnay which revealed to be more tolerant until population densities kg<sup>-1</sup> soil increase to 600 or more. Along with the degree of susceptibility, variation in plant vigour and root density may be the other possible reason for differences to nematode damage of the tested cultivars. ROGIERS *et al.* (2009) also

noted that 'Chardonnay' was more vigorous than 'Semillon' and had greater root density and longer roots than 'Semillon'. These physiological attributes found in 'Chardonnay' may compensate for the damage caused by nematodes and grapevine cultivars with these characteristics have been considered as tolerant to some pest nematode species (MCKENRY *et al.* 2001).

'Semillon' vines inoculated with *M. javanica* had significantly lower numbers of bunches compared to uninoculated control vines, and bunch numbers were correlated with Log<sub>e</sub>[*M. javanica* J<sub>2</sub> + 1] kg<sub>1</sub> dry soil, suggesting that *M. javanica* had an effect on bunch initiation in 'Semillon' but not in 'Chardonnay'. Significantly lower bunch numbers in own rooted 'Shiraz' (susceptible to nematode) vines than the vines grafted on nematode resistant rootstock were also observed when planted in a nematode infested vineyard (CIRAMI *et al.* 1984).

*M. javanica* had no effect on berry development in any of the cultivars in this study, which is consistent with the results of HARRIS (1986).

**Pruning weight and trunk circumference:** Our results did not show any significant relationship between soil *M. javanica* population density and pruning weight or trunk circumferences suggesting that more time may be needed to demonstrate the effect of *M. javanica* root damage on grapevine top growth. WALKER (1997), in a pot experiment, also did not find any significant reduction in shoot dry weight in Colombard (susceptible to *Meloidogyne spp.*) when inoculated with 5,000 eggs of *M. javanica*. HARRIS (1986) did not observe any significant differences in cane weight of Sultana vines infected with *M. javanica* (8-483 J<sub>2</sub> / 500 g soil) prior to grape harvest and SAUER (1967) found no significant differences in trunk circumferences in ungrafted grapevines seven years after planting in a *M. javanica* infected vineyard.

**Juice characteristics:** This study demonstrated that Pi densities of *M. javanica* had no adverse effects on juice pH and °Brix. No consistent effect of Pi densities on TA, within a cultivar for each year, could be determined, despite the significance of the overall effect of Pi densities. This is similar to the results obtained by CIRAMI *et al.* (1984) and RAHMAN *et al.* (2001, 2011) who

reported no major impact of parasitic nematode infection on juice characteristics. The reason for differences of TA due to Pi densities between the cultivars, within a year, is not known but differences in amount of other elements in the juice, such as K in high concentration, may decrease the concentration of free acids (KODUR 2011).

The increased °Brix in 'Chardonnay' compared to 'Semillon' was probably caused by the lower yields and thus increased ripening rates for 'Chardonnay'. Although not studied, the higher °Brix level in 'Chardonnay' may also be related with the leaf growth rate (JACKSON 1986).

### Conclusion

This is the first study showing that 'Semillon' is more susceptible to the root-knot nematode *M. javanica* than 'Chardonnay', and that *M. javanica* J<sub>2</sub> densities between 200 and 800 kg<sup>-1</sup> dry soil, which may occur in Australian vineyards at planting, will increase enough over 5-6 years period, causing 15-20 % yield loss in 'Semillon'. Only the highest level (800 J<sub>2</sub>-kg<sup>-1</sup> dry soil) at planting affected 'Chardonnay' yields by 7-13 %.

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