Heat stability of resistance in selected tomato breeding lines against *Meloidogyne incognita* and *M. javanica* populations under elevated soil temperatures

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Summary. In tomato, the only commercially available source of resistance to root-knot nematodes (RKN) is the *Mi-1* gene that confers resistance to *Meloidogyne incognita*, *M. javanica* and *M. arenaria*. However, its effectiveness was limited at higher soil temperatures. A study was initiated with the objective to check the durability of the potential resistance genes found in some tomato breeding lines after screening in controlled glasshouse conditions ≤ 27°C by exposing them to higher soil temperatures at 28, 32 and 36°C for 24 and 48 h periods. The aggressive Jittu and Babile *M. incognita* and Jittu and Koka *M. javanica* populations originally collected from Ethiopia were used. When seedlings reached the four-leaf stage, each tube was inoculated with 50 freshly (≤ 24 h) hatched infective second-stage juveniles (J2). Immediately after inoculation, the seedlings were exposed continuously for 24 and 48 h in a warm water bath at 28, 32 and 36°C, respectively. A control was kept separately in ambient temperature (24°C ± 2°C). The external ambient temperature and the soil temperature inside the tube while in the water bath were simultaneously recorded using a TESTO data logger. Temperature, tomato breeding lines and time had a significant effect on the number of J2 of Jittu and Babile *M. incognita* and Jittu and Koka *M. javanica* populations that penetrated the roots. The utility of the potential resistance found in the breeding lines during the controlled growth chamber resistance screening experiment was limited at higher soil temperatures, especially at 32 and 36°C. At 36°C there was no significant difference found on the mean number of penetrated J2 of Jittu and Babile *M. incognita* and Jittu and Koka *M. javanica* populations inside the roots of all the tested breeding lines compared to 'Marmande' (a susceptible control) after 48 h of heat exposure after inoculation. More J2 were found in the roots of the tested breeding lines after 48 h compared to 24 h heat exposure after inoculation for each soil temperature level tested and for both populations of *M. incognita*. It is clear from our observations that local tomato breeding lines with resistance potential can be used when soil temperatures remain below 32°C. Differences were observed between breeding lines depending on the RKN population used at higher temperatures and this knowledge can help in further optimising the development of sustainable resistance under local Ethiopian circumstances.

Key words: climate change, exposure time, management, *Mi-1*, *Meloidogyne* species.

Tomato (*Solanum lycopersicum*) is among the most valuable agricultural crops worldwide (de Carvalho *et al.*, 2015). A considerable portion of the tomato production takes place in warm and hot climates (Verdejo-Lucas *et al.*, 2013) where root-knot nematodes (RKN; *Meloidogyne* spp.) are important endoparasitic pests (Ammiraju *et al.*, 2003) and infestation causes serious crop losses (de Carvalho *et al.*, 2015). Tomato yield reduction as severe as 100% have been reported (Seid *et al.*, 2015).

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Table 1. List of tomato genotypes, their sources, growth habit, pedigree and related information used for the heat stability experiment against *Meloidogyne incognita* and *M. javanica* populations originating from Ethiopia.

Tomato genotypes	Source	Growth habit	Pedigree	Other information (*e.g.*, selected for nematode resistance/tolerance)
CLN-2366A	AVRDC	Indeterminate	[(Orion F1 × (CLN698BC1F1F2-358-4-13 × CLN1314B1F3-51-25-16-6-5)] × (UC204A × CLN1314B1F3-51-25-16-6-5B-8)	Beta[1], Tm22[2]; no known nematode resistance
CLN-2366B	AVRDC	Indeterminate	[(Orion F1 × (CLN698BC1F1F2-358-4-13 × CLN1314B1F3-51-25-16-6-5)] × (UC204A × CLN1314B1F3-51-25-16-6-5B-8)	Beta, Tm2a[3]; no known nematode resistance
CLN-2366C	AVRDC	Indeterminate	[(Orion F1 × (CLN698BC1F1F2-358-4-13 × CLN1314B1F3-51-25-16-6-5)] × (UC204A × CLN1314B1F3-51-25-16-6-5B-8)	Beta, Tm22; no known nematode resistance
CLN-2037H	AVRDC	Indeterminate	CLN657BC1F2-285-0-21-0 × CLN1776BC1F1 (Moneymaker × L3708)	Ph-3[4], Tm2a, probably Bwr-12[5] but not confirmed; no known nematode resistance
CLN-2037A	AVRDC	Indeterminate	CLN657BC1F2-285-0-21-0 × CLN1776BC1F1 (Moneymaker × L3708)	Ph-3, Tm2a, probably Bwr-12 but not confirmed; no known nematode resistance
CLN-2037B	AVRDC	(Semi) Indeterminate	CLN657BC1F2-285-0-21-0 × CLN1776BC1F1 (Moneymaker × L3708)	Ph-3, Tm2a, probably Bwr-12 but not confirmed; no known nematode resistance
CL5915-206-D4-2-2	AVRDC	Indeterminate	[[(VC48-1 × Tamu Chico III) × (ahTm-2a × VC11-1UG)] × [ahTm-2a × (VC11-1-21B × Saturn)] × [[(TR/VC9-1)-4-2-5F5 × Plum)] × 71-483N]]	Tm2a, Bwr-12
'Marmande'	MARC[1]	Determinate	Unknown	Unknown

[1]Resistant for late and early blight of tomato;
[2]Tomato mosaic virus and tobamoviruses;
[3]Resistant for tomato mosaic virus;
[4]Resistant for late blight in tomato;
[5]Resistant for bacterial wilt in tomato.

Management of RKN is a challenging task due to its wide host range, which hinders the practice of crop rotation (Chen *et al.*, 2006). Soil fumigants, systemic and contact nematicides, resistant rootstocks, resistant cultivars and cultural practices are commonly employed to control RKN (Devran *et al.*, 2010; Seid *et al.*, 2015). After the withdrawal of the effective and widely used soil fumigant methyl bromide from the market due to its negative effect on stratospheric ozone (Rosskopf *et al.*, 2005) and risks for non-target organisms (Ploeg, 2002; Devran *et al.*, 2010), host-plant resistance (HPR) appeared as a powerful and sustainable tool for crop protection including the management of RKN (Devran *et al.*, 2010).

In tomato, the single dominant gene *Mi-1* confers resistance but not immunity to the three most damaging species: *M. incognita*, *M. javanica* and *M. arenaria* (Milligan *et al.*, 1998) with *M. incognita* and *M. javanica* reported in Ethiopian agriculture (Mandefro & Mekete, 2002; Seid *et al.*, 2017). This gene has been the only commercially available source of resistance to RKN for the last 70 years globally (Seid *et al.*, 2015). *Mi-1* gene was found in *Solanum peruvianum* and introgressed into *S. lycopersicum* (Rodriguez *et al.*, 2013). The effectiveness of the *Mi-1* gene varies with the RKN species and population, tomato cultivar, and environmental conditions, particularly soil temperature (Devran *et al.*, 2010; Verdejo-Lucas *et al.*, 2013; Seid *et al.*, 2015). Even though there is inconsistency in the literature, increased gall formation has been reported in plants exposed to soil temperatures above 28°C (Haroon *et al.*, 1993;

Wang *et al.*, 2009; Devran *et al.*, 2010; Verdejo-Lucas *et al.*, 2013). The *Mi-1* resistance was lost after 4 days at ≥ 33°C in 1 to 3 days old seedlings exposed to heat treatment after inoculation with *M. incognita* and subsequently held at 27°C for 1 month (Dropkin, 1969). Climatic heterogeneity is a general characteristic of Ethiopia and in areas where tomato is largely produced soil temperature at times rises above 28°C (Jury & Funk, 2013). Under field conditions, plants are subjected to environmental and daily soil temperature fluctuations (Verdejo-Lucas *et al.*, 2013). Consequently, the expression of the resistance phenotype may or may not be similar to that observed under constant (generally < 28°C) temperature conditions. The incorporation of heat-stable resistance to tomato would be a valuable genetic improvement to manage *Meloidogyne* species. Thus, checking the heat stability of the potential resistance gene found in tomato breeding lines in higher soil temperatures after screening tomatoes

Fig. 1. A: plastic tubes with tomato seedlings; B: the experimental set up for the heat stability test in a warm water bath with simultanously recording of the soil temperatures using a data longer (TESTO).

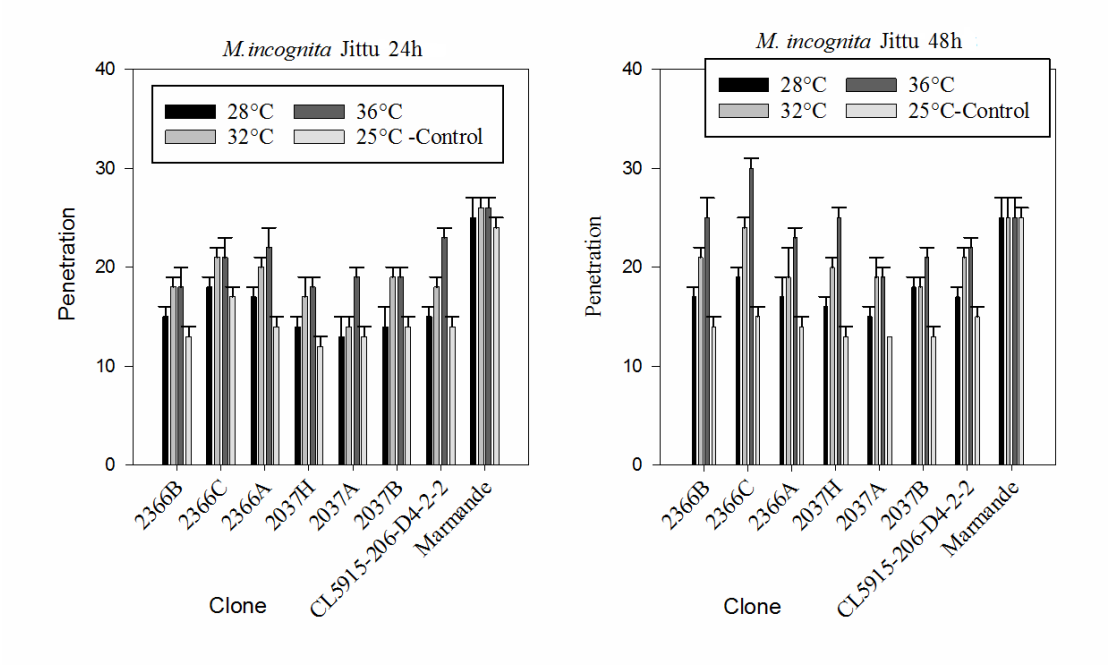

Fig. 2. Mean number of second-stage juveniles of *Meloidogyne incognita* Jittu population penetrated to the tomato roots after 24 and 48 h of inoculation at soil temperatures of 25, 28, 32 and 36°C.

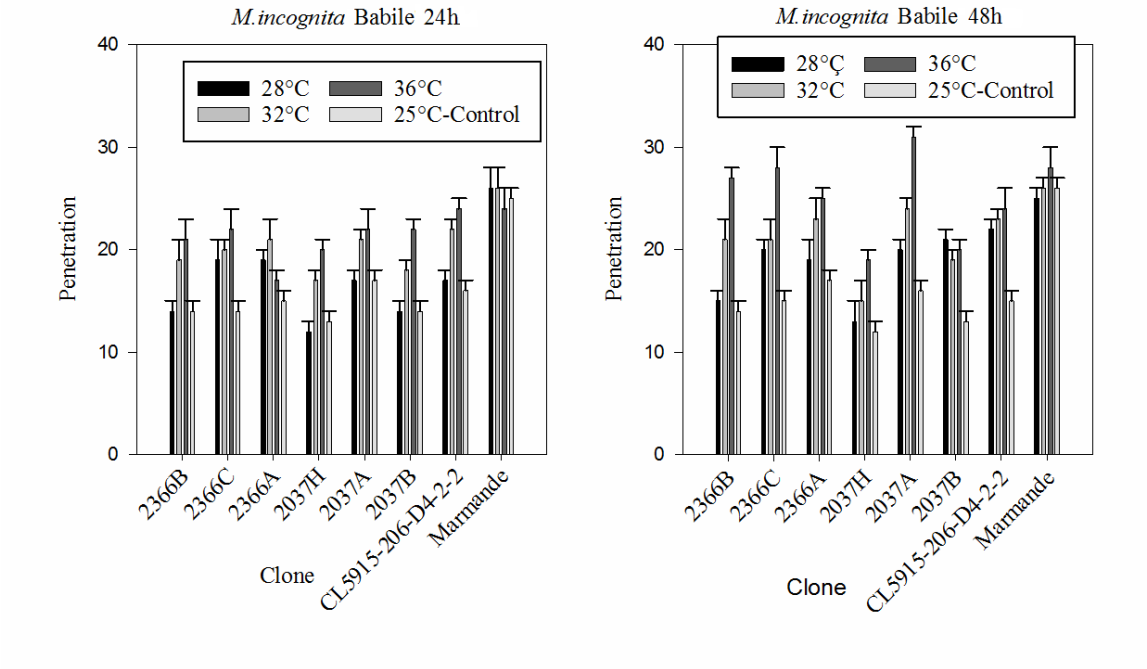

Fig. 3. Mean number of second-stage juveniles of *Meloidogyne incognita* Babile population penetrated to the tomato roots after 24 and 48 h of inoculation at soil temperatures of 25, 28, 32 and 36°C.

against RKN populations in a controlled environment is crucial. Therefore, the objective of this study was to check the durability of the potential resistance genes found in some tomato breeding lines after screening in controlled glasshouse conditions ≤ 27°C (Seid *et al.*, 2017) by exposing them to higher soil temperatures at 28, 32 and 36°C for 24 and 48 h periods.

MATERIAL AND METHODS

Tomato breeding lines. Based on the results of the growth chamber screening experiment as performed by Seid *et al.* (2017), seven tomato breeding lines (CLN-2366A, CLN-2366B, CLN-2366C, CLN-2037H, CLN-2037A, CLN-2037B and CL5915-206-D4-2-2) obtained from Asian Vegetable Research Development Centre (AVRDC) and Melkassa Agricultural Research Centre (MARC) were selected and used to check the heat stability of their potential resistance gene(s) at 28, 32 and 36°C soil temperatures. The tomato cultivar 'Marmande' was used as a susceptible check. The source of these tomato breeding lines, their growth habit, known source of resistance (if any) and pedigree are described in Table 1.

***Meloidogyne incognita* and *M. javanica* culture.** The aggressive Jittu and Babile *M. incognita* and Jittu and Koka *M. javanica* populations originally collected from Ethiopia were used (Seid *et al.*, 2017). The culture of these populations was maintained on tomato 'Marmande' in the glasshouses at Flanders Research Institute for Agriculture, Fisheries and Food (ILVO), Belgium. Inoculum was prepared from the infected tomato roots. Therefore, roots were carefully washed and cut into smaller pieces and placed on a Baermann funnel in a mistifier (Hooper *et al.*, 2005). Freshly hatched (≤ 24 h) infective second-stage juveniles (J2) were collected and used as inoculum for this study.

Heat stability experiment. A plastic tube (15 ml volume, 95 mm height, 15 mm diameter) was filled with sterilised (100°C, 16 h) soil (74% sand, 14% sandy loam, 6% clay, 5% loam, 1% organic matter content and a neutral pH). A single seed was allowed to grow per tube. Plants were watered daily and maintained at 23°C ± 2°C (11 h day), 20°C ± 1°C (13 h night), with 60 to 65% relative humidity for three consecutive weeks in a growth chamber at ILVO. When the seedlings reached four-leaf stage, they were brought to the laboratory and kept in the ambient temperature overnight. Then, each tube was inoculated with 50 freshly (≤ 24 h) hatched infective J2. Immediately after inoculation, the seedlings were exposed continuously for 24 h and 48 h in a warm water bath at 28, 32 and 36°C, respectively. A control was kept separately in ambient temperature (24°C ± 2°C). The external ambient temperature and the soil temperature inside the tube while in the water bath were simultaneously recorded using a data logger (Testo 175T2) (Fig. 1). After the exposure period, the seedlings were returned to the growth chamber. The experiment was arranged in a completely randomised design with eight replications. Eight days after inoculation, the nematodes inside the root were stained with acid fuchsine using the method described by Byrd *et al.* (1983).

Data analysis. The heat stability data were analysed using Statistica Version7 Software. Mean numbers of J2 that penetrated the tomato roots after being exposed to different soil temperature regimes: 25 (control temperature), 28, 32 and 36°C and time (24 and 48 h) was subjected to factorial ANOVA and statistically compared using Fisher's unprotected LSD at $P < 0.05$. One-way ANOVA was used to analyse the penetration of J2 in the roots of each clone separately for each temperature after inoculation followed by a post-hoc test using Tukey HSD at $P < 0.05$. J2 penetration counts were Log 10 (x+1) transformed for analysis to fulfil the criteria for normality. All graphs were drawn using Sigmaplot13 statistical software.

RESULTS

Jittu and Babile *M. incognita* populations. The effect of temperature, breeding lines and time. Temperature, tomato breeding lines and time had a significant effect on the numbers of J2 ($F_{temperature}$ = 118.06, $P < 0.01$; $F_{breeding\ lines}$ = 41.30, $P < 0.01$ and F_{time} = 23.01, $P < 0.01$) of *M. incognita* Jittu population that penetrated the tomato roots. The interaction between breeding lines × temperature, temperature × time and breeding lines × temperature × time was significant for *M. incognita* Jittu J2 penetration ($F = 2.26$, $P < 0.01$; $F = 5.96$, $P < 0.01$ and $F = 1.63$, $P < 0.01$ respectively) but the effect was less evident compared to the main effects. Similarly, temperature, breeding lines and time had a significant effect on the numbers of J2 ($F_{temperature}$ = 134.35, $P < 0.01$; $F_{breeding\ lines}$ = 47.17, $P < 0.01$ and F_{time} = 26. 44, $P < 0.01$) of *M. incognita* Babile population that penetrated the plant roots. The interaction between breeding lines × temperature, breeding lines × time, temperature × time and breeding lines × temperature × time was significant for *M. incognita* Babile population J2 penetration ($F = 4.12$, $P < 0.01$; $F = 2.05$, $P < 0.05$; $F = 8.75$, $P <$

0.01 and $F = 1.92$, $P < 0.01$, respectively) but the effect was again less compared to the main effects.

The effect of temperature levels. After 24 h heat exposure to 28°C, the number of J2 of *M. incognita* Babile population inside the roots of CLN-2366C and CLN-2366A were significantly higher compared to the control temperature (25°C). The number of Babile *M. incognita* population J2 penetrated the roots of all the tested tomato genotypes were significantly higher after 48 h exposure to 28°C compared to the control temperature except on CLN-2366B and CLN-2037H. For the Jittu *M. incognita* population, the number of J2 detected in the roots of CLN-2366B, CLN-2366A and CLN-2037H after 24 h exposure to 28°C was significantly higher compared to the control temperature. The number of J2 found inside the roots of all the tested breeding lines after 48 h exposure to 28°C were significantly higher compared to the control temperature for Jittu *M. incognita* population.

The number of J2 for Babile *M. incognita* population detected inside the roots of all the tested tomato breeding lines was considerably higher after 24 and 48 h exposure to 32°C compared to the control temperature. Remarkably higher number of J2 of Jittu *M. incognita* population were found inside the roots of all the tested breeding lines (except on CLN-2037A) after 24 h exposure to 32°C compared to the control temperature. A significantly higher number of J2 entered the roots of all the tested breeding lines for *M. incognita* Jittu population after 48 h heat exposure to 32°C compared to the control temperature (Fig. 2). A higher number of J2 invaded roots of all the tested breeding lines for both *M. incognita* populations after 24 and 48 h exposure to 36°C compared to the control temperature. Except on CLN-2366C, significantly higher number of J2 penetrated roots of all the tested breeding lines after inoculation with Babile *M. incognita* population and exposed to 32°C compared to the 28°C. For the Babile *M. incognita* population, the number of J2 in the roots of CLN-2366B, CLN-2366A, CLN-2037H and CLN-2037A were significantly higher after 48 h exposures to 32°C compared with the 28°C.

The number of J2 for *M. incognita* Babile population invaded roots of all the tested breeding lines (except on CLN-2366B) were significantly higher after 24 h exposure to 36°C compared to 28°C. After 48 h of exposure to 36°C compared to 28°C, the number of J2 for Babile *M. incognita* population that were detected in roots of CLN-2366A, CLN-2366B, CLN-2366C, CLN-2037H and CLN-2037A were significantly high (Fig. 3). The number of J2 of *M. incognita* Jittu population that was found in the roots of all the tested breeding lines was significantly higher at 36°C compared to 28°C after both (24 and 48 h) exposure time. However, after 24 h exposure to 36°C, the number of J2 detected inside the roots of CLN-2366A, CLN-2037A and CL5915-206-D4-2-2 for *M. incognita* Jittu population was significantly higher compared at 32°C. Moreover, the J2 of *M. incognita* Jittu population penetrated roots of all the tested genotypes (except CLN-2037A and CL5915-206-D4-2-2) with higher number after 48 h exposure to 36°C compared at 32°C. The breeding lines CLN-2366C, CLN-2037H and CLN-2037B supported a significantly higher number of Babile *M. incognita* J2 penetration after 24 h of exposure to 36°C compared to 32°C. The J2 of Babile *M. incognita* population found inside the roots of CLN-2366B, CLN-2366C, CLN-2037H and CLN-2037A after 48 h exposures to 36°C were considerably higher when compared with 32°C.

In general, after 24 and 48 h of heat exposure to 28, 32 and 36°C the J2 of both (Babile and Jittu) *M. incognita* populations invaded the roots of all the tested breeding lines in significantly higher numbers compared with numbers invading at the control temperature. There was no significant difference in J2 penetration of roots of all the tested breeding lines for both *M. incognita* populations after 24 and 48 h of exposure to 36°C compared numbers penetrating 'Marmande' (susceptible control). A considerably higher number of J2 was detected in the roots of all the tested tomato breeding lines for both *M. incognita* populations after 48 h heat exposure compared to the 24 h heat exposure for each soil temperature level studied.

Jittu and Koka *M. javanica* populations. The effect of temperature, breeding lines and time. Temperature, tomato breeding lines and time had a significant effect on the numbers of J2 ($F_{\text{temperature}} = 464.24$, $P < 0.01$; $F_{\text{breeding lines}} = 208.40$, $P < 0.01$ and $F_{\text{time}} = 115.60$, $P < 0.01$) of *M. javanica* Jittu population that penetrated the tested tomato genotypes roots. The interaction between breeding lines × temperature, breeding lines × time, temperature × time and breeding lines × temperature × time was significant for *M. javanica* Jittu penetration ($F = 14.49$, $P < 0.01$; $F = 7.76$, $P < 0.01$; $F = 32.88$, $P < 0.01$ and $F = 1.96$, $P < 0.01$, respectively) but the effect was again less pronounced compared to the main effects. Likewise, temperature, breeding lines and time had a significant effect on the numbers of J2 ($F_{\text{temperature}} = 218.69$, $P < 0.01$; $F_{\text{breeding lines}} = 143.67$, $P < 0.01$ and $F_{\text{time}} = 49.90$, $P < 0.01$) of *M. javanica* Koka population

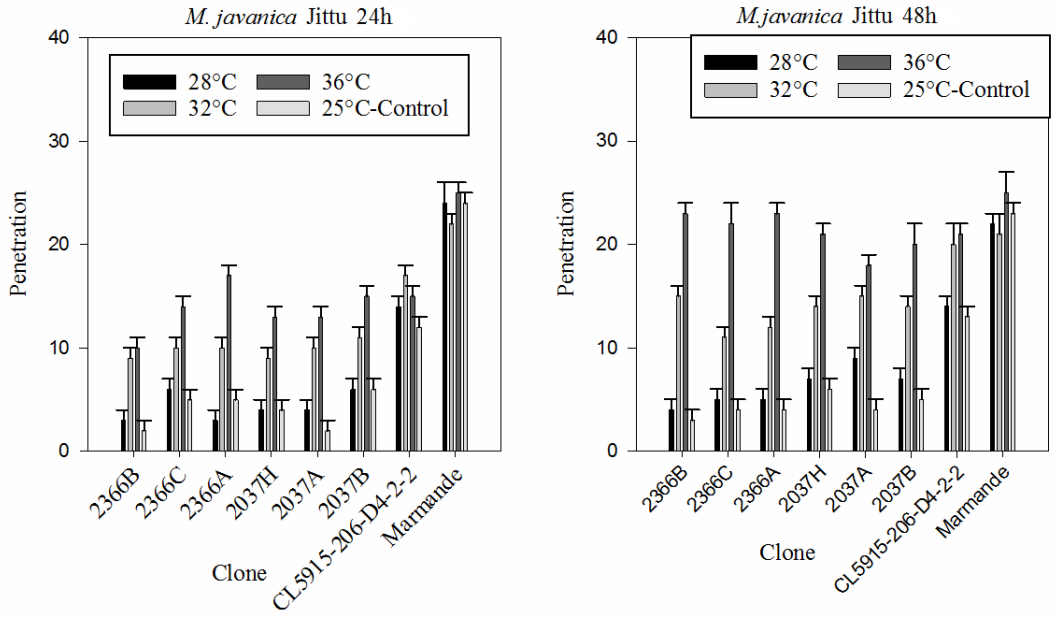

Fig. 4. Mean number of second-stage juveniles of *Meloidogyne javanica* Jittu population penetrated to the tomato roots after 24 and 48 h of inoculation at soil temperatures of 25, 28, 32 and 36°C.

that penetrated the plant roots. The interaction between breeding lines × temperature, temperature × time and breeding lines × temperature × time was significant for *M. javanica* Koka population penetration ($F = 7.834$, $P < 0.01$; $F = 5.55$, $P < 0.01$ and $F = 2.84$, $P < 0.01$, respectively) but the effect was less pronounced compared to the main effects.

The effect of temperature levels. There was no significant difference found in the number of penetrated J2 for *M. javanica* Jittu population inside the roots of all the tested breeding lines (except on CLN-2037A) after 24 and 48 h exposure to 28°C compared numbers penetrating at the control temperature (Fig. 4). After 24 h exposures to 28°C compared to the control temperature, the number of J2 of *M. javanica* Koka population that invaded the roots of all the tested breeding lines was not significantly different. Conversely, after 48 h exposure to 28°C compared to the control temperature, the number of J2 from *M. javanica* Koka population found inside the roots of CLN-2366B, CLN-2366C, CLN-2037H and CL5915-206-D4-2-2 were significantly different (Fig. 5). After 24 and 48 h exposure of both *M. javanica* populations to 32°C, the number of J2 detected in the roots of all tested breeding lines (except on the CLN-2037H after 48 h exposure with *M. javanica* Koka population) was significantly higher compared to the control temperature. The number of J2 detected inside the roots of all the tested breeding lines was significantly higher for both *M. javanica* populations after both exposure times to 36°C compared to the control temperature.

After both exposure times to 32 and 36°C, the number of J2 that entered the roots of all the tested breeding lines for both *M. javanica* populations was significantly different compared to the exposure at 28°C. The number of J2 detected inside the roots of all the tested breeding lines (except on the CLN-2366B and CL5915-206-D4-2-2) was significantly higher for *M. javanica* Jittu population after 24 h exposure to 36°C compared to 32°C. Furthermore, the number of J2 found in the roots of all the tested breeding lines (except on CL5915-206-D4-2-2) was significantly higher for *M. javanica* Jittu population after 48 h exposure to 36°C compared to the exposure at 32°C. The roots of CLN-2366A, CLN-2366B and CLN-2366C were found penetrated with a higher number of J2 from both *M. javanica* populations after both exposure times at 36°C compared to 32°C. Generally, a significantly higher mean number of J2 from both *M. javanica* populations penetrated the roots of all the tested breeding lines at 36°C after 48 h exposure compared to the 24 h exposure time.

DISCUSSION

All the tested tomato breeding lines in this study showed resistance gene inactivation for Jittu and Babile populations of *M. incognita* after exposure to

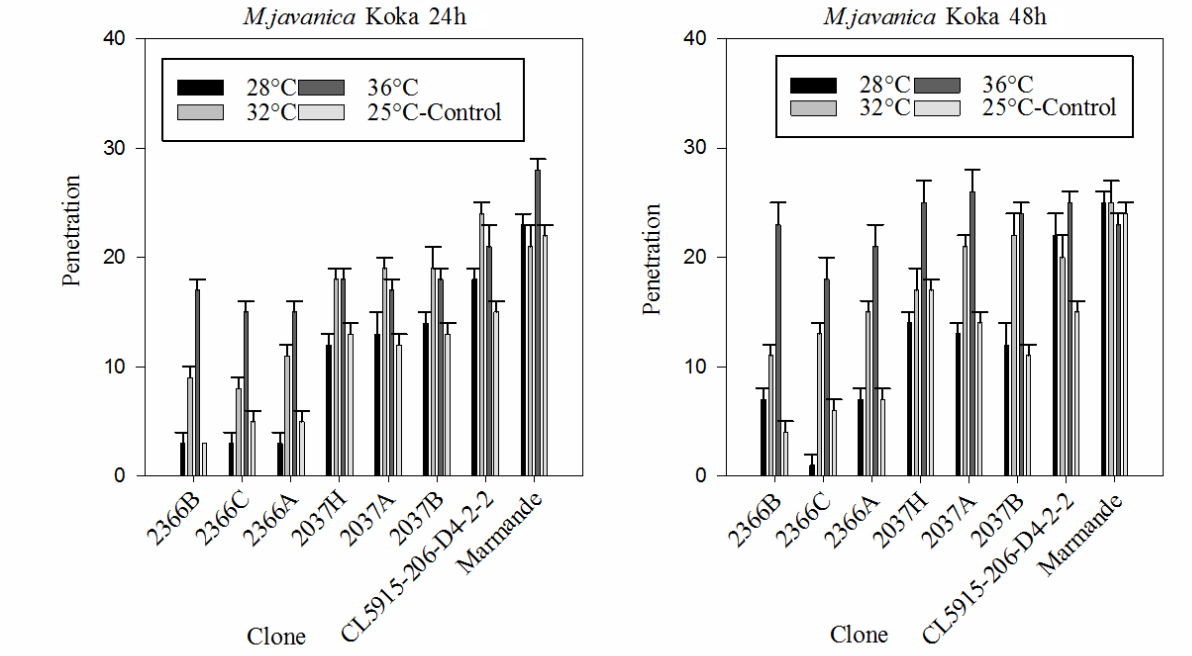

Fig. 5. Mean number of second-stage juveniles of *Meloidogyne javanica* Koka population penetrated to the tomato roots after 24 and 48 h of inoculation at soil temperatures of 25, 28, 32 and 36°C.

28, 32 and 36°C during 24 and 48 h. For Jittu and Koka populations of *M. javanica* this effect was only seen after 24 and 48 h at 32 and 36°C. This finding is in agreement with several reports of *Mi-1* gene inactivation characterised by higher egg masses and root gall production above 28°C soil temperature both in glasshouse and field conditions (Dropkin, 1969; Ammati *et al.*, 1986; Tzortzakakis & Gowen, 1996; Noling, 2000; Wang *et al.*, 2009; Devran *et al.*, 2010). The *Mi-1* mediated resistance was found inactivated by *M. incognita* in the 'Motelle' and 'VFNT' at 32°C (Ammiraju *et al.*, 2003). The utility of the potential resistance found in these breeding lines during the controlled growth chamber resistance screening experiment is limited at higher soil temperatures especially at 32 and 36°C. Hence, we suggest that soil temperatures at 32 and 36°C will reduce the effectiveness of the potential resistance genes contained in the tested tomato breeding lines. The effect of temperature was stronger on *M. javanica* compared to *M. incognita*, indicating that *M. javanica* could be more aggressive on the potential resistance gene(s) contained in the tested tomato breeding lines as temperature increases. The genetic background of these tomato breeding lines to determine the presence of nematode resistance gene(s) should be assessed through marker analysis (Cortada *et al.*, 2012). In this study, the potential resistance gene(s) present in the tested breeding lines seemed less strong for *M. incognita* populations. However, in Seid *et al.* (2017), these tomato breeding lines there were a statistically significant difference in the number of egg mass produced per root system compared to the standard control ('Marmande') (Seid *et al.*, 2017), indicating the type of resistance in these lines seemed post-infection resistance, which is manifested after nematodes penetrated roots due to failed development or less production of eggs per egg mass (Anwar & McKenry, 2000).

Tomato breeding lines such as CLN-2366A, CLN-2366B and CLN-2366C can be used as a potential resistance gene source after genetic improvement especially at soil temperatures below 32°C and in areas where *M. javanica* is predominant. In our study, J2 of all *M. incognita* and *M. javanica* populations penetrated the breeding lines tested after 24 and 48 h heat exposure irrespective of the soil temperature levels. Higher numbers of J2 were found after 48 h of exposure compared to 24 h exposures. This exposure time could be comparable to field-like heat exposures in tropical agriculture (including Ethiopia) during the day and the night. Soil temperatures fluctuates monthly, daily and even, at times, hourly mainly by variations in air temperature and solar radiations

(Cheon *et al.*, 2014). In the face of global warming and climate change, this is even more evident. A single mid-day heat exposure of 35°C during 3 h was sufficient to break the *Mi-1* gene resistance in 'Amelia' by a *M. incognita* population (de Carvalho *et al.*, 2015). Similarly, in our study all the tested tomato breeding lines resistance was lost after 24 or 48 h of exposure to 36°C and there was no significant difference found between the tested breeding lines and the susceptible control 'Marmande'. The *Mi-1* resistance was hypothesised either to acclimatise or recover from exposure to high temperatures (Zacheo *et al.*, 1995; de Carvalho *et al.*, 2015). There is still some inconsistency in the literature and several authors have reported as *Mi-1* conferred resistance was found effective at temperatures ≥ 34°C (Abdul-Baki *et al.*, 1996; Verdejo-Lucas *et al.*, 2013). The *Mi-9* obtained from the *S. peruvianum* complex and a homologue of *Mi-1* gene was found unaffected by temperature (Bleve-Zacheo *et al.*, 2007) but it is not yet commercially available. As confirmed with the tested tomato breeding lines in this study and reported in different crop species such as bean (Mullin *et al.*, 1991), pepper (Thies & Fery, 2000), sweet potato (Jatala & Russell, 1972), alfalfa (Griffin, 1969) and cotton (Carter, 1982) temperature sensitivity is a characteristic of several RKN resistance genes.

Checking the heat stability of resistance genes in time at higher temperatures and assessing whether the resistance genes could be reversed or not is crucial for further development of sustainable cultivars. Adjustment of planting date to avoid planting at the hottest season when monthly soil temperature rises above 28°C is also very important especially in areas with enough irrigation potential in the vicinity. Systematically designed crop rotation schemes in which plants are included that can reduce the soil temperature (*i.e.*, using crops with higher canopy coverage) and the nematode population (a non-host crop) should be developed and used. A mechanism to increase soil aeration and thereby cool down the soil temperature might also be reliable especially until the plant successfully establishes in the field after transplanting. It is also recommended to soften the soil between the plants without affecting the tomato roots so that soil will be aerated and soil temperature cools down. Most of the tomato growing areas in Ethiopia are characterised by sandy soil (less thermal conductivity and diffusivity) and it can be feasible to look for alternatives to cool down the soil temperatures. We also recommend checking the soil temperature prior to transplanting tomato seedlings using a simple soil thermometer and avoid too dry and too wet extreme field conditions. It is clear from our observations that local tomato breeding lines with resistance potential can be used when soil temperatures remain below 32°C. Differences were observed between breeding lines depending on the RKN population at higher temperatures and this knowledge can help in further optimising the development of sustainable resistance under local Ethiopian circumstances.

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A. Seid, C. Fininsa, T.M. Mekete, W.M.L. Wesemael and W. Decraemer. Тепловая стабильность устойчивости некоторых селекционных линий томатов к популяциям *Meloidogyne incognita* и *M. javanica* при повышенных температурах почвы.

Резюме. Единственным коммерчески доступным источником устойчивости томатов к галлообразующим нематодам является ген *Mi-1*, который и определяет способность подавлять развитие *Meloidogyne incognita*, *M. javanica* и *M. arenaria*. Эффективность этого гена существенно снижается при высоких температурах почвы. Проведено изучение устойчивости этих генов, выявленных при выращивании селекционных линий томатов в теплицах при температуре ≤ 27°C, при повышенных температурах среды: культивированию растений при 28°C, 32°C и 36°C на протяжении 24 и 48 часов. В экспериментах были использованы высокоинвазивные эфиопские популяции *M. incognita* 'Jittu' и 'Babile' и популяции *M. javanica* 'Jittu' и 'Koka'. Когда проростки томатов в пробирках достигали стадии четырех листьев, в каждую из пробирок добавляли по 50 только что вылупившихся (в пределах 24 часов) личинок мелойдогин второй стадии. Сразу после внесения личинок проростки помещали на 24 и 48 ч в водяную баню при 28°C, 32°C и 36°C. Контрольные проростки томата содержали при 24°C ± 2°C. Показания температуры среды и почвы в пробирках постоянно записывали с помощью регистратора данных TESTO. Показано, что температура, селекционная линия томатов и время экспозиции оказывали существенное воздействие на число проникающих в корни личинок мелойдогин. Эффективность потенциальной устойчивости в селекционных линиях томатов существенно снижалась при повышенных температурах почвы, в особенности при 32°C и 36°C. При 36°C не было статистических отличий в числе проникших личинок *M. incognita* 'Jittu' и 'Babile' и *M. javanica* 'Jittu' и 'Koka' в сравнении с сортом 'Marmande' (восприимчивый к мелойдогинам сорт томатов) при экспозиции в 48 ч. Показано, что число личинок, проникших в корни после экспозиции в 48 ч, было выше, чем при экспозиции в 24 ч для каждой из изученных температур и для двух популяций *M. incognita*. Полученные результаты показывают, что потенциал природной генетической устойчивости к мелойдогинозу у местных селекционных линий может быть использован, если температура почвы не превышает 32°C. Различия в устойчивости изученных селекционных линий томатов при высоких температурах почвы к разным популяциям мелойдогин могут оказаться полезными при оптимизации методов выращивания томатов в условиях Эфиопии.