*http://dx.doi.org/10.22161/ijeab/3.6.4* *ISSN: 2456-1878*

# Antagonistic Effect of Eight Sri Lankan Isolates of *Pseudomonas fluorescens* on, *Meloidogyne incognita* in Tomato, *Lycopersicon esculentum*

K.M.D.W.P. Nishantha[1], H.D.C.M. Jayasiri[2], H.A.L.H.K. Herath[1], M.M. Nugaliyadde[3], M.L.M.C.Dissanayake[2], Devika M. De Costa[4],

[1] Horticultural Crops Research and Development Institute, Gannoruwa, Sri Lanka
Email: wpnishantha@yahoo.com
[2] Faculty of Agricultural Sciences, Sabaragamuwa University, Belihuloya, Sri Lanka
Email: chethanimalki34@gmail.com
[3] Department of Agriculture, Peradeniya, Sri Lanka
Email : nmangalika@gmail.com
[4] Faculty of Agriculture, Universityof Peradeniya, Sri Lanka
Email : devikadecosta@gmail.com

***Abstract*—***The study was conducted to determine the efficacy of Pseudomonas fluorescens isolates collected from eight locations in the Central Province of Sri Lanka against Meloidogyne incognita in tomato. Isolates were tested under laboratory conditions to determine the efficacy on egg hatchability and mortality of second stage juveniles. A planthouse experiment was conducted using potted tomato plants to determine the potential of P. fluorescens isolates and effective application technique. All tested isolates have significantly inhibited egg hatchability and increased the juvenile mortality after 72 hours. P. fluorescens isolate from Kangkung field in Pallekelle (PK) and tomato field in Udispattuwa (UT I) recorded 95% and 95.5% inhibition of egg hatchability after 72 hours. P. fluorescens isolates collected from tomato fields in Bopane (BT II) and Udispattuwa (UT II) and from Kangkung field in Pallekelle recorded the higher mortality of second stage juveniles 93%, 87% and 83.3% respectively. The highest reduction in the root knots (96.8%, 96.3%), egg masses (98.5%, 98.2%) and lower root galling index (1 and 1) were recorded in tomato plants treated as soil drench with UT II and PK isolates respectively.The root dipping technique gave higher reduction in the number of root knots (47.4%), egg masses (44.9%) and lower root galling index (3.75) were recorded from BT II, UT II and tomato fields in Nugethenna (NT) isolates respectively. UT II and PK found to be the most effective isolates and most effective application technique determined as soil drenching ten days after transplanting under plant house conditions.*

***Keywords - Biological Control, Meloidogyne incognita, Pseudomonas fluorescens, Soil Drenching.***

## I. INTRODUCTION

Root Knot Nematodes (RKN), *Meloidogyne* spp., are widely distributed pests of several crops grown in Sri Lanka [6]. Almost 95% of food crops grown are susceptible to one or more species of RKN [2]. The yield loss due to the damage caused by *Meloidogyne* spp. is more prominent in vegetables, especially among Solanaceae and Cucurbitaceae crops than other crops [2]. The management of nematodes is challenging as they inhabit the soil and usually attack the underground parts of plants. Control of *Meloidogyne* spp. through synthetic nematicides is effective, easy to apply but are toxic to humans, animals and can cause soil and water pollution [11]. Biological control offers a good alternative to chemical control with a little hazard to the soil environment. A variety of microorganisms and natural enemies antagonistic to soil nematodes exist in the soil; these include bacteria, fungi, predatory nematodes and mites. The application of antagonistic soil microbes is considered as effective and eco-friendly for managing nematodes [7]. Bacteria are the most abundant organisms in soil and some of them, for example members of the genera *Pasteuria*, *Pseudomonas* and *Bacillus* have shown great potential for the biological control of nematodes. Aerobic endospore-forming bacteria *Pseudomonas* spp. are among the dominant populations in the rhizosphere that are able to antagonize nematodes. The antagonistic mechanisms include production of antibiotics and

*http://dx.doi.org/10.22161/ijeab/3.6.4* *ISSN: 2456-1878*

induction of systemic resistance to minimize the populations of plant parasitic nematodes [11].

Tomato is an attractive cash crop that provides a source of income to the rural population in the central region of Sri Lanka. Nematodes of the genus *Meloidogyne*, is known to cause more than 50% crop losses to tomato [4]. This study was conducted to determine the antagonistic properties of eight Sri Lankan isolates of *Pseudomonas fluorescens* against *Meloidogyne incognita* in Tomato (*L. esculentum*) *in vitro* and *in vivo* and to select the effective *P. fluorescens* isolate(s) for the control the *Meloidogyne* spp.

## II. METHODOLOGY

### 2.1. Preparation of water cultures of *P. fluorescens*

Soil samples collected from a Kang kung crop from Pallekelle, tomato crops from Nugethenna (one sample), Bopane (2 samples) and Udhispaththuwa (3 samples) and from a maize crop from Sooriyawewa were used for the experiments. One gram each of the above 8 samples was diluted in 100 ml of sterilized phosphate buffer solution and shake for 2 h.

A series of 10-fold dilutions was prepared by repeating 6 times under aseptic condition. The diluted soil supernatants (0.1 ml) was spread on king's medium B agar plates and incubated at 28 °C for 48 h in an incubator. Culture plates were observed under ultraviolet trans-illuminator at 366 nm for few seconds and colonies with green fluorescence were streaked on King's medium B agar plates to get pure colonies. Well-grown 48 h old uncontaminated single colonies were used to prepare water cultures of *P. fluorescens* and cell density were estimated for all the isolates.

### 2.2. Effect of *P. fluorescens* on egg hatchability of *M. incognita*

Sterilized Petri dishes were filled with the eight *P. fluorescens* suspensions at the rate of one isolate per five Petri dishes. Similarly five Petri dishes were filled distilled water (as control). Egg masses of *M. incognita* were placed on the micro sieve (75-µm aperture, 20 mm diameter) at the rate of 10 egg mass per sieve. These micro sieves were placed in the Petri dishes to touch the egg masses with *P. fluorescens* isolates or water. The experimental set-up was kept at room temperature and number of emerged juveniles was counted at 24, 48 and 72 h after inoculation. At the end of the experiment, the egg masses were treated with 1% sodium hypochlorite to dissolve the gelatin matrix around the eggs and the un-hatched eggs were counted. Percentage egg hatchability: (mean number of emerged juveniles in each treatment / Total number of juveniles and eggs in treatment) x 100 were calculated.The treatments were replicated 5 times in a Randomized Complete Block Design.

### 2.3. Effect of *P. fluorescens* on mortality of juvenile *M. incognita*

Forty, sterilized 60 mm diameter watch glasses were filled separately with 3 ml of *P. fluorescens* isolates and similarly five watch glasses were filled with 3 ml distilled water. Newly hatched second-stage juveniles of *M. incognita* were added to the bacteria suspensions at the rate of ten per watch glass. After 24, 48 and 72 h the numbers of dead juveniles were counted under a stereomicroscope. The treatments were replicated 5 times in a Randomized Complete Block Design. Percentage mortality: (Mean number of dead juveniles in the treatment / Total number of juveniles in treatment) x 100 were calculated.

### 2.4. Efficacy *of P. fluorescens* isolates for the control of *M. incognita* on tomato

A pot experiment was conducted in the plant house using the tomato variety KWR, a variety susceptible to *Meloidogyne* spp., to determine the efficacy of *P. fluorescens* isolates on the root damage using two application techniques.

#### 2.4.1. Soil drenching of *P. fluorescens* isolates to potted tomato plants

Two-week-old tomato plants potted in 15 cm dia. plastic pots at the rate of 1 plant per pot were used for the experiment. About 2 cm of top soil layer was removed near the root system and the soil were drench with 50 ml *P. fluorescens* isolates separately. In addition, two sets of plants were treated with distilled water as an untreated control. The plants were then covered with 2 cm sterilized soil layer. After 24 h each pot was inoculated with 1,000 juveniles except one set of plants treated with distilled water. The experiment was arranged in a Randomize Complete Block design with 5 replicates.

#### 2.4.2. Root dipping of tomato plants with *P. fluorescens* isolates

Roots of another set of tomato plants were dipped separately with *P. fluorescens* isolates for one minute before planting in sterilized potting media. Another set of plants were treated only with the nematodes + distilled water. The experiment was arranged in Randomize Complete Block Design with 5 replicates.

Sixty days after inoculation of nematodes, tomato plants were uprooted and the nematode damage was assessed by the number of egg masses per root system. The intensity of root damage was determined through the diagrammatic root knot scoring chart (John and Sam, 1980). Plant height, shoot fresh weight, shoot dry weight, root fresh weight were also measured.

### 2.4. Data analysis

Proc Catmod was performed to check for normality and homogeneity, if the results were significant, numerical data were square root-transformed prior to analysis. The data were analyzed using analysis of variance and

*http://dx.doi.org/10.22161/ijeab/3.6.4* *ISSN: 2456-1878*

treatment means were compared by Duncan's Multiple Range Test at P<0.05 level. Data were subjected to analysis using Statistical Analysis Software (SAS) package version 8.2.

## III. RESULTS

The densities of *P. fluorescens* isolates in the 8 samples ranged from 1.01 x $10^8$ to 2.00 × $10^8$ specifying that the bacterial colonies falled within a narrow range suitable for pathogenicity estimates (Table 1).

*Table.1: Cell count of different P. fluorescens isolates used for the experiments*

| Notation of the isolate | *P. fluorescens* isolates: location/ crop source | *P. fluorescens* cell count / 1 ml |
|---|---|---|
| *PK* | Pallekele - Kangkung | 1.87 × $10^8$ |
| *NT* | Nugethenna - Tomato | 1.05 × $10^8$ |
| *BT I* | Bopane - Tomato I | 2.00 × $10^8$ |
| *BTII* | Bopane - Tomato II | 1.64 × $10^8$ |
| *UT I* | Udhispaththuwa - Tomato I | 1.40 × $10^8$ |
| *UT II* | Udhispaththuwa -Tomato II | 1.01 × $10^8$ |
| *UT III* | Udhispaththuwa- TomatoIII | 1.01 × $10^8$ |
| *SM* | Sooriyawewa - Maize | 1.03 × $10^8$ |

### 3.1. Effect of *P. fluorescens* on egg hatchability of *M. incognita*

*P. fluorescens* isolates significantly reduced the egg hatchability of *M. incognita* at 24, 48, 72 h after inoculation as estimated by the chi-square < 0.05 level of probability according to Proc Catmod (Table 2). The lowest egg hatchability was observed in PK (Pallekele - Kangkung), NT (Nugethenna – Tomato) and UT 1 (Udhispaththuwa – Tomato) isolates

### 3.2. Effect of *P. fluorescens* on mortality of juvenile *M. incognita*

The mortality of *M. incognita* juveniles has shown significant effect compared to the controls from 24 to 72 h after treatment at chi-square < 0.05 level. The suppressive activity of *P. fluorescens* increases gradually with the increased exposure time. BT II (Udispattuwa isolate 2 from tomato crop) exhibited the highest mortality of juveniles after 72 h (Table 3).

*Table 2. Hatchability percentages of M. incognita egg masses treated with P. fluoresces isolate at different exposure periods*

| ***P. fluoresces* isolates** | **Mean cumulative egg hatchability % after different exposure time** | | |
|---|---|---|---|
| | 24 HAI* | 48 HAI* | 72 HAI* |
| PK | 1.07 $^{c}$ | 3.19 $^{cd}$ | 4.55 $^{e}$ |
| NT | 2.64 $^{bc}$ | 3.40 $^{cd}$ | 5.47 $^{e}$ |
| BT I | 5.10 $^{ab}$ | 8.75 $^{bc}$ | 12.09 $^{cd}$ |
| BT II | 2.37 $^{bc}$ | 7.50 $^{bcd}$ | 15.94 $^{bc}$ |
| UT I | 1.56 $^{bc}$ | 4.51 $^{cd}$ | 4.99 $^{e}$ |
| UT II | 1.11 $^{c}$ | 5.64 $^{cd}$ | 8.99 $^{d}$ |
| UT III | 9.70 $^{a}$ | 15.24 $^{ab}$ | 20.87 $^{ab}$ |
| SM | 2.86 $^{b}$ | 6.95 $^{cd}$ | 16.09 $^{bcd}$ |
| DW(Control) | 11.13 $^{a}$ | 29.71 $^{a}$ | 32.12 $^{a}$ |

* Mean values within a column followed by the same letter(s) are not significantly different at p<0.05 based on the Duncan's multiple range test.Key to cultures PK = Pallekele/ Kangkung; NT = Nugethenna /Tomato; BT1 and II = Bopane/ Tomato: UT I, II, III= Udhispaththuwa/ Tomato: SM = Sooriyawewa/ Maize: DW = Distilled water

*Table 3. Mortality (%) of M. incognita juveniles when exposed different isolates of P. fluorescens for 24, 48 and 72h periods*

| ***P. fluoresces* isolates** | **Juveniles (J2) mortality percentages after different exposure periods** | | |
|---|---|---|---|
| | **24 h*** | **48 h*** | **72 h*** |
| PK | 53.33 $^{ab}$ | 70.00 $^{a}$ | 83.33 $^{abc}$ |
| NT | 50.00 $^{ab}$ | 60.00 $^{b}$ | 70.00 $^{abcd}$ |
| BT I | 30.00 $^{b}$ | 56.67 $^{ab}$ | 63.33 $^{cd}$ |
| BT II | 36.67 $^{ab}$ | 60.00 $^{ab}$ | 93.33 $^{a}$ |
| UT I | 46.67 $^{ab}$ | 53.33 $^{ab}$ | 60.00 $^{d}$ |
| UT II | 40.00 $^{ab}$ | 50.00 $^{a}$ | 86.67 $^{ab}$ |
| UT III | 56.67 $^{a}$ | 60.00 $^{ab}$ | 66.67 $^{bcd}$ |
| SM | 46.67 $^{ab}$ | 60.00 $^{ab}$ | 66.67 $^{bcd}$ |
| DW | 0 | 3.33 $^{c}$ | 13.33 $^{e}$ |

* Mean values within a column followed by the same letter(s) are not significantly different at p<0.05 based on the Duncan's multiple range test.

Key to cultures PK = Pallekele/ Kangkung; NT = Nugethenna /Tomato; BT1 and II = Bopane/ Tomato: UT I, II, III= Udhispaththuwa/ Tomato: SM = Sooriyawewa/ Maize: DW = Distilled water

### 3.3. *In vivo* experiments to determine the efficacy *of P. fluorescens* isolates for the control of *M. incognita* on tomato

#### 3.3.1. Efficacy *of P. fluorescens* isolates on root knot development:

*http://dx.doi.org/10.22161/ijeab/3.6.4* *ISSN: 2456-1878*

We observed that the tested *P. fluoresces* isolates and application technique significantly influenced on the root knots per root system and the number of egg masses per root; chi-square < 0.05 probability level according to Proc Catmod and significant difference between application methods at P< 0.05 level of probability according to Duncan's multiple range test except the NT and positive control (Fig 1 and 2). *P. fluorescens* isolates from PK (Pallekele/ Kangkung crop); UT II (Udhispaththuwa/ Tomato crop 2) and BT II (Bopane /tomato crop 2) recorded a significantly low number of root knots when the isolated were soil drenched. It was observed that root knot count per root significantly influenced by the application technique; and soil drenching as the most effective application technique for all tested isolates.

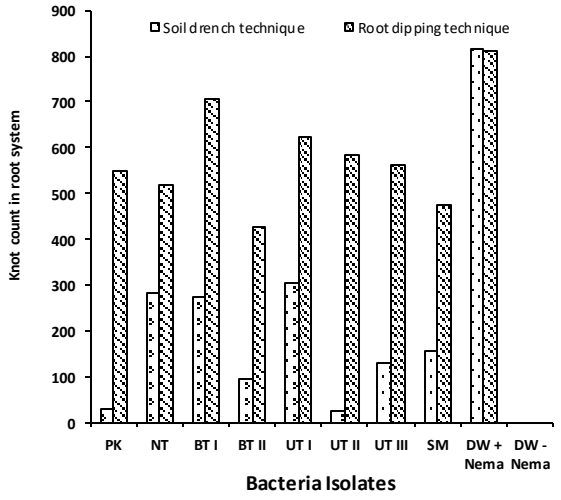


*Fig 1:Root knot counts in tomato treated with different P. fluorescens isolates under different application techniques*

### 3.3.2. Effect of *P. fluorescens* on egg laying of *M. incognita*:

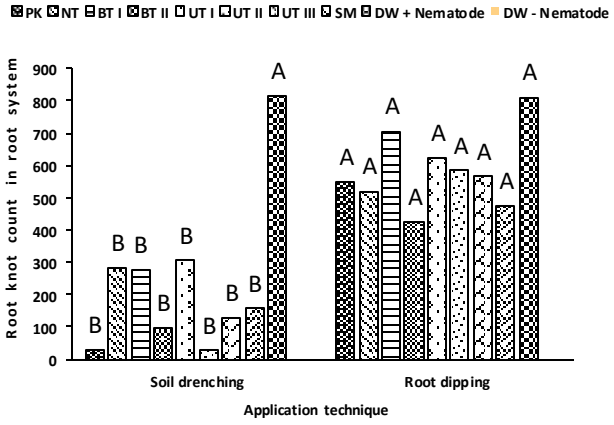


*Fig.2: Rootknot counts per tomato root system treated with eight P. fluorescesusing two different application techniques (root drenching and root dipping) (n= 5)*

Significant differences among the treatments were observed as shown by chi-square <0.05 probability level and application methods at P< 0.05 level of probability according to Proc Catmod and Duncan's multiple range test. The *P. fluorescens* isolates UT II (Udhispaththuwa Tomato crop 2) and PK (Pallekele Kangkung crop) showed the lowest number of *M. incognita* egg masses of when the treatments were soil drenched.

Soil drenching technique recorded the low number of *M. incognita* egg masses in all *P. fluorescens* isolates when compared to the root dipping technique. PK, NT, BT I, BT II, UT I, UT II, UT III, SM isolates recorded low number of egg masses in soil drench technique than root dipping technique.

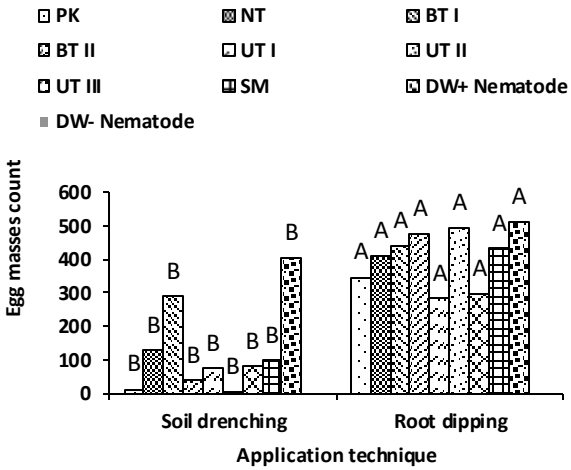


*Fig.3: Egg masses count per tomato root system treated with eight bacterial isolates using two different application techniques (root drenching and root dipping) (n= 5)*

### 3.3.3. Effect of *P. fluorescens* on root knot score on tomato:

The mean value of root knot scores in *P. fluorescens* isolates. Moreover, there is significant difference between control and all other isolates in soil drench techniques at chi-square < 0.05 probability level of according to Proc Catmod. There is no significant difference between two application techniques at P< 0.05 level of probability according to Duncan's multiple range test. According to table 5, Bacteria isolates of PK, UT II and shows the significant low root knot score (score =1) compared to control treatment in soil drenching technique. In root dipping technique, NT isolate showed the low mean number of root knot score (score =3.75).

*http://dx.doi.org/10.22161/ijeab/3.6.4* *ISSN: 2456-1878*

*Table 4. Root knot scores of tomato plants treated with different bacterial isolates as soil drenching and root dipping .*

| *P. fluoresces* isolates | Mean value of Root knot score ± SD | |
|---|---|---|
| | Soil drench technique* | Root dipping technique* |
| *PK* | 1 ± 0 d | 4.25 ± 0.95 abc |
| *NT* | 3 ± 1.15 bc | 3.75 ± 0.5 c |
| *BT I* | 5 ± 0.95 a | 4.75 ± 0.95 abc |
| *BT II* | 1.25 ± 0.5 d | 5 ± 0 ab |
| *UT I* | 4 ± 1.15 ab | 4.75 ± 0.5 abc |
| *UT II* | 1 ± 0 d | 4.75 ± 0.5 abc |
| *UT III* | 2.25 ± 0.95 c | 5.25 ± 0.57 a |
| *SM* | 2.75 ± 1.5 bc | 5 ± 1.15 ab |
| DW + *Nema* | 5.25 ± 0.5 a | 5 ± 0.81ab |
| DW only | 0 | 0 |

*Mean values with in a column followed by simple letters are not significantly different at p<0.05 based on the Duncan's multiple range test according to treatments,

Key to cultures PK = Pallekele/ Kang Kung; NT = Nugethenna /Tomato; BT1 and II = Bopane/ Tomato: UT I, II, III= Udhispaththuwa/ Tomato: SM = Sooriyawewa/ Maize, DW = Distilled water

### 3.3.4. Effect of P. fluorescens on fresh root weight of tomato:

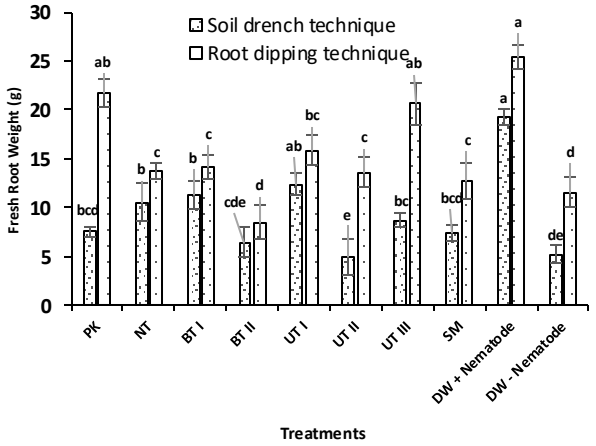


*Fig.4: Root fresh weight in different P. fluorescesisolations applied with two different soil application techniques*

Fresh root weight was significantly lower in bacterial isolates treated treatments than nematode present treatment at P< 0.05 level of probability. Highest fresh root weight was recorded in nematode present treatment in both soil application techniques. The lowest fresh root weight was recorded in UT II (5 g) bacterial isolate treatment in soil drenching technique and BT II (9 g) bacteria isolate treatment in root dipping technique respectively (Figure 5).Tomato plants treated with the isolates as soil drench technique recorded the lowest root fresh weight than root dipping technique.

### 3.3.5. Effect of *P. fluorescens* on plant growth parameters:

Tomato plants treated with the different isolates as soil drenching and root dipping, shows variations in shoot length, shoot fresh weight and shoot dry weight. There were no significant difference among treatments in dry shoot weight at P< 0.05 level of probability according to Duncan's multiple range test (Table 5 and 6).

Root knot nematodes are soil pathogen. They directly attack to the root system of plants and main symptoms were occurred in the below ground parts of the plant. Secondary symptoms will have occurred in the above ground plant parts. Alsothe plants were maintained under plant house conditions and because of that, the effect of bacterial treatments on above ground plant parameters were not clearly expressed.

*Table 5. Growth parameters of tomato plants when soil drench with P. fluoresces isolates*

| *P. fluoresces isolates* | Shoot Length (cm)* | Shoot Fresh Weight(g)* | Shoot Dry Weight(g)* |
|---|---|---|---|
| *PK* | 91.45 c | 68.48 d | 13.23 ab |
| *NT* | 105.8 ab | 93.52 bc | 17.01 ab |
| *BT I* | 109.52 ab | 105.70 ab | 17.50 ab |
| *BT II* | 104.52 b | 91.08 bc | 16.89 ab |
| *UT I* | 108 ab | 110.28 a | 18.38 a |
| *UT II* | 89.65 c | 84.20 c | 16.57 ab |
| *UT III* | 113.07 a | 95.62 abc | 17.03 ab |
| *SM* | 87.5 c | 104.74 ab | 16.70 ab |
| DW + Nematode | 88.17 c | 91.85 bc | 14.77 ab |
| DW - Nematode | 105.62 ab | 92.47 bc | 15.50 ab |

*Mean values within a column followed by the same letter(s) are not significantly different at p<0.05 based on the Duncan's multiple range test.

Key to cultures PK = Pallekele/ Kangkung; NT = Nugethenna /Tomato; BT1 and II = Bopane/ Tomato: UT I, II, III= Udhispaththuwa/ Tomato: SM = Sooriyawewa/ Maize

*http://dx.doi.org/10.22161/ijeab/3.6.4* *ISSN: 2456-1878*

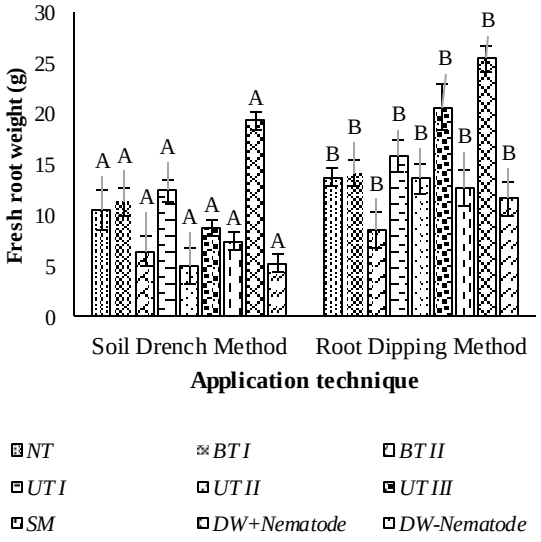


*Fig.5: Root fresh weight in two different application techniques of eight P. fluoresces isolates*

*Table 6. Growth parameters of tomato plants when root dipped with P. fluorescesisolatesbefore planting*

| ***P. fluoresces* isolates** | **Shoot Length (cm)*** | **Shoot Fresh Wt(g)*** | **Shoot Dry Wt(g)*** |
|---|---|---|---|
| PK | 95.1 b | 114.12 a | 19.57ab |
| NT | 100.1b | 100.18 bc | 16.92 ab |
| BT1 | 99.8 b | 100.02 bc | 17.52 ab |
| BT2 | 110.2 a | 86.15 de | 15.37 b |
| UT1 | 96.6 b | 90.38 cde | 16.19 ab |
| UT2 | 96.6 b | 85.01 e | 16.96 ab |
| UT3 | 100.8 b | 121.51 a | 20.27 b |
| SM | 101.4 b | 90.87 cde | 16.83 ab |
| DW + Nematode | 84.05 c | 95.59 bcd | 18.55 ab |
| DW –Nem | 0 | 0 | 0 |

*Means within a column followed by the same letter(s) are not significantly different at p<0.05 based on the Duncan's multiple range test.

Key to cultures PK = Pallekele/ Kangkung; NT = Nugethenna /Tomato; BT1 and II = Bopane/ Tomato: UT I, II, III= Udhispaththuwa/ Tomato: SM = Sooriyawewa/ Maize

## IV. DISCUSSION

Some reports says that, *P. fluorescens* has nematicidal activity to root knot nematode, *M. javanica*, juveniles and nematicidal activity against potato cyst nematode, *Globodera rostochiensis* eggs as the same might contribute as mechanism of *M. incognita* mortality and egg hatchability [9].

*P. fluorescens* can produce large number of toxic secondary metabolites such as; phenazine, indole, compounds, phenyl-pyrroles and pterines [1]. These metabolites might also be toxic to *M. incognita* juveniles [1]. The production of the metabolite 2, 4-diacetylphloroglucinol (2, 4-DAPG) by *P. fluorescens* strain CHA0 induced mortality in juvenile of root-knot nematodes [13].

Some *P. fluorescens* strains are known to contain 1-aminocyclopropane-1carboxylic acid which inhibits ethylene production in roots and henceminimizes colonization of root knot nematodes and root knot development [9]. Also reported that application of *P. fluorescens* bacteria led to reduce the number of egg masses of nematodes [4].

Some articles described DAPG produced form *P. fluorescens* strains, reduced the mobility and survival the second stage juveniles, the infective stages of some plant-parasitic nematodes [10]. The production of this antibiotic in the rhizosphere of plants suppress nematode penetration of roots [12]. In addition, it is known that DAPG affect root morphology and such changes in root architecture that may alter the number of available infection sites and, therefore, lead to a complex response with regards to nematode suppression [10].

The percentage of gall formation and root gall index found to decrease when *P. fluorescens* were introduced prior to *M. incognita* infestationon tomato plants [1]. Similar results were observed that the highest reduction in the numbers of second-stage juveniles in soil, host root galls and egg mass indices when *P. fluorescens* was drenched before planting [5]. However, it was reported that, strains CHA805 and CHA89 had no significant impact on nematode population densities in soil and root-knots in tomato and soybean crops [10].

## V. CONCLUSION

The *Pseudomonas fluorescens* isolated from tomato rhizosphere from Nugethenna (NT) and Udhispaththuwa (UT I) effectively minimize egg hatchability of *Meloidogyne incognita*. *P fluorescens* isolated from tomato rhizosphere from Bopane (BT II) and Udhispaththuwa (UT II) effectively controlled juveniles of *M. incognita* in tomato. The tomato plants treated with *P. fluorescens* isolated from Kang Kung from Pallekele (PK) and UT II had lower number of root knots and egg masses. These experiments indicated that the tomato rhizosphere from Udhispaththuwa and Kang-Kung rhizosphere from Pallekele contained effective *P. fluorescens* isolates that can be used for the management of *M. incognita*. The effective application technique determined as soil drenching ten days after transplanting under plant house conditions.

## REFERENCES

[1] Ashoub, A.H. and Amara, M.T. (2010). Biocontrol activity of some bacterial genera against root-knot

nematod, *Meloidogyne incognita*. *Journal of American Science*. 6(10): 321-328.

[2] Ekanayak, H.M.R.K., Jayalath, J.P.I.S. and Nishantha, K.M.D.W.P. (2000). Effect of Meloidogyne incognita on plant growth, absorption and translocation of nutrients and water in tomato plants. *Annals of the Sri Lanka Department of Agriculture*. 2: 93-100.

[3] Ekanayake, H.M.R. and Toida, Y. (1997). Nematode parasites on agricultural crops and their distribution in Sri Lanka. *JIRCAS Journal for Scientific Papers*.

[4] Ketabchi, S., Charehgani, H. and Majzoob, S. (2016). Impact of rhizosphere antagonistic bacteria and urea fertilizer on roccot knot nematode (Meloidogyne incognita) under greenhouse condition. *JAPS, Journal of Animal and Plant Sciences*. 26(6): 1780-1786.

[5] Khan, M.R. and Haque, Z., 2011. Soil application of Pseudomonas fluorescens and *Trichodermaharzianum* reduces root-knot nematode, *Meloidogyne incognita*, on tobacco. *Phytopathologia Mediterranea*. 50(2): 257-266.

[6] Lamberti, F., Ekanayake, H.M.R.K., Sasanelli, N. and Zacheo, F. (1993). Control of root-knot nematodes in Sri Lanka. *Nematologia Mediterranea*. 21(2): 227-234.

[7] Park, J., Seo, Y. and Kim, Y.H. (2014). Biological control of Meloidogyne hapla using an antagonistic bacterium. *The plant pathology journal*. 30(3): 288.

[8] Prernachandra, W.T.S.D., Lasanthi A.H.P., Deepananda, K.H.M.A. and Jayasinghe R.C. (2007). Infestations of root-knot nematodes, *Meloidogyne* species, associated with selected vegetable crop in Matara district in Sri Lanka. In: *proceedings forth academic session*,159-163.

[9] Seenivasan, N., David, P.M.M., Vivekanandan, P. and Samiyappan, R. (2012). Biological control of rice root-knot nematode, Meloidogyne graminicola through mixture of Pseudomonas fluorescens strains. *Biocontrol science and technology*. 22(6): 611-632.

[10] Siddiqui, I.A., Haas, D. and Heeb, S. (2005). Extracellular protease of Pseudomonas fluorescens CHA0, a biocontrol factor with activity against the root-knot nematode Meloidogyne incognita. *Applied and Environmental Microbiology*. 71(9): 5646-5649.

[11] Tian, B., Yang, J. and Zhang, K.Q. (2007). Bacteria used in the biological control of plant-parasitic nematodes: populations, mechanisms of action, and future prospects. *FEMS microbiology ecology*. 61(2): 197-213.

[12] Timper, P., Kone, D., Yin, J., Ji, P. and Gardener, B.B.M. (2009). Evaluation of an antibiotic-producing strain of Pseudomonas fluorescens for suppression of plant-parasitic nematodes. *Journal of nematology*. 41(3): 234.

[13] Wei, L., Shao, Y., Wan, J., Feng, H., Zhu, H., Huang, H. and Zhou, Y., 2014. Isolation and characterization of a rhizobacterial antagonist of root-knot nematodes. *PloS one*, *9*(1):85988.