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# STUDY OF THE EFFICIENCY OF DIFFERENT CONCENTRATIONS OF *LECANICILLIUM LECANII* ISOLATES IN PROTECTING CITRUS TREES FROM INFECTION BY *MICROCEROTERMES DIVERSUS* SILV. (ISOPTERA: TERMITIDAE) IN IRAQ

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## ABSTRACT

The study was conducted to evaluate the efficacy of three isolates (Indian, Korean and local) of the fungus *Lecanicillium lecanii* in protecting citrus trees from infection by the termites insect *Microcerotermes diversus* Silv. With different concentrations 2.5, 5 and 10 g / L(2.5,5,10 x$10^8$ spore / ml)by using direct spraying on tree stems and soil surrounding a diameter of 1 m under field conditions in Iraq and during the 2019 agricultural season. The results of the study demonstrated the efficiency of fungus isolates in reducing infection, reducing the number of tunnels, length of tunnels, and the number of workers in a 20 cm tunnel gradually compared to the control treatment, which increased in it. As the Indian isolation was characterized by all its concentrations after 6 months of treatment and the local isolation in all its concentrations after 5 months of treatment with its efficiency by reducing the numbers and lengths of tunnels and the number of workers, and the Korean isolation was the least efficient in the reduction. Indian isolation surpassed all its concentrations in reducing the number of workers inside the tunnels after the treatment. As for the effect of fungus isolates on the behavior of workers and the construction of tunnels during study period which lasted for six month, the effect of all fungus isolates on the behavior of workers has been shown, where the concentration exceeded 10 g/L for the Indian isolation in reducing the length of the built tunnel, and the concentrations of 5 and 10 g/L for the Korean isolation were higher compared to the rest Isolates and concentrations, while the Indian isolate outperformed the two concentrations 2.5 and 5 g/L in reducing the number of workers inside the specified tunnel, The results of the study showed the efficiency of different isolates of the fungus in reducing termite insect infestation, and that the concentrations of 2.5 and 10 g/L are the most efficient in reducing the infection of termite insects in the field conditions of Iraq.

***Keywords* :** *Lecanicillium lecanii*, Isolate, *Microcerotermes diversus* Silv., Termite, Iraq.

## Introduction

The termites insect is one of the social insects that have economic importance in most countries of the world, and which belong to the order Isoptera and live within colony consisting of specialized social, appearance and functional casts (Al-Jassani, 2019), The number of individuals within a colony ranges from hundreds to millions, and most of its species live in colonies under the surface of the soil, which makes it difficult to reach (Abe and his group, 2000), And that the species is *Microcerotermes diversus* Silv. It is considered of high economic importance in most of the province of Iraq (Al-Jassani, 1996).

Su and Scheffrahn (2000) indicated the superiority of fungus when used it in controlling termite insects compared to the rest of the pathogens used and They give a good level of resistance in the field and the ground environment provides favorable conditions for fungi in terms of moisture and darkness (Faria and Wraight, 2007). And that the termite insect is difficult to reach its nests which located under the soil surface, and that spraying the spores of the fungus on the surface of wood and the stems of plants may be more important and effective than spraying the body of the insect (Fernandez *et al.*, 2001; Vandervalk, 2007).

Alavo (2015) indicated that *V. lecanii* is a Deuteromyces fungus used to protect plants from many pests, that *V. lecanii* can be grown on all living and dead matter (Schuler, 1991), and that *V. lecanii* is effective on termites groups *Captotermes formosanus.*

Due to the importance of *M. diversus* in Iraq and the absence of a study on the use of *L. lecanii* fungus in field resistance to termite insects, the study aimed to evaluate the efficiency of *L. lecanii* in protecting citrus trees from infection with termite insects by using direct spray on the trunks of trees infected with type *M. diversus* and treating the soil surrounding the tree.

## Materials and Methods

36 citrus trees were selected severely affected by *M. diversus* silv. In an grove in the Jadriya region in Baghdad**,** Three trees were used for each treatment, representing three replications, as the treatments included three Indian, Korean and local isolates with three concentrations of 2.5, 5 and 10 g/liter 2.5,5,10 x$10^8$ spore / ml and the control treatment (distilled water only) and the pesticide Bifinthrin (Aquastar) at a concentration 20 ml/L and the *I. Fumosoroseus* fungus at a concentration 2.5 g/L was used as a control treatment

(Nassry, 2017). The treatment were distributed randomly to the trees and were informed of cards with a record of the treatment number and the replicates . The number of tunnels, its lengths, and the number of workers in a 20 cm tunnel lengths for each tree were recorded before carrying out various treatment.

A single 2 liter small sprinkler was used for each treatment, where the tree trunk was sprayed from the soil surface to the branching area approximately 1 meter from the soil surface and the tunnels were covered on the tree stems completely covering the treatment solution, The surrounding soil was sprayed around the tree with a diameter of one meter around the tree trunk. Each tree was treated with 1500 ml of treatment solution.

For the purpose of knowing the effect of the fungus on the infestation of the termite insect on citrus trees in the field, the examination was carried out after the treatment on a monthly basis, where it was recorded that there was an infection (building tunnels) or not, the presence of workers or not, the number of tunnels and its lengths, the number of workers inside a 20 cm tunnel lengths if present, and to study the effect of the fungus on behavior of tunnels building: A tunnel with a length of 20 cm was determined, and the length of the built tunnel was recorded during the study period and the number of workers present in a tunnel of 20 cm length, and the study continued for six months from 5/5/2019 to 5/11/2019.

**Statistical analysis**

The experiments were designed by adopting the Randomly complete block design RCBD and using the smallest significant difference criterion at the 0.05 level to compare the results, and the Genstat statistical program was adopted in analyzing the results.

## Results and Discussion

**1. The effect of different concentrations of *L. lecanii* isolates on the number of new tunnels on the stems of citrus trees after treatment.**

The results of the study showed that all the fungus isolates affected the reduction of the number of new tunnels building on the stems of citrus trees after the treatment, but there are differences in the effectiveness depending on the type of isolation and its concentrations, which caused a gradual decrease in the number of tunnels with the progression of the treatment time compared to the increase in the control treatment, and the local isolation has surpassed. With two concentrations of 2.5 and 10 g /L, and the Indian isolate at low concentration in reducing the tunnels number compared to all treatments.

It is evident from Table (1) that the local isolation of the fungus was the most efficient in reducing the number of tunnels. While the Korean isolation was the least effective in reducing the number of tunnels, as the number of tunnels reached 9.67, 7.33 and 7.33 tunnels / tree at concentrations of 2.5, 5 and 10 g / l, respectively. Whereas in the control treatment it was 9.67 and in the treatment of *I. fumosoroseus* 8.00 and the chemical pesticide 0.00. The statistical analysis showed that there are significant statistical differences between the general average of the number of tunnels during the readings for the months of the year after the treatment, where the Indian isolates reached 2.89, 5.56 and 5.89, the Korean isolate was 9.94, 6.39, and 7.33, and the local isolate was 3.00, 4.78 and 3.33 tunnel/tree of concentrations of 2.5, 5 and 10 g/L, respectively. And the presence of statistical differences between the cumulative average of tunnels number for the different isolates, which amounted to 4.78, 7.89 and 3.70 tunnels / tree for Indian, Korean and local isolates respectively, while the cumulative statistical analysis of the concentrations did not show any statistical differences, as the average number of tunnels reached 5.28, 5.57 and 5.52 tunnels / Tree for concentrations of 2.5, 5 and 10 g/L, respectively, Table (1).

Through the results, the superiority of the local isolation with a concentration of 2.5 and 10 g/L is evident. This superiority may be attributed to the adaptation of the local isolation of the fungus with the environmental conditions in Iraq, which results in its ability to attack individuals and cause them to die.

The superiority of the low concentration of Indian isolation is also evident, as it caused the emergence of the lowest number of tunnels compared to all concentrations and isolates, which may be attributed to the fact that this concentration is characterized by its attracting effect to the termites workers and the arrival of the fungus to the bodies of workers, its penetration and the occurrence of death.

**2. The effect of different concentrations of fungus isolates on the new tunnels lengths on the stems of citrus trees after treatment**

The results of the study showed that all of the fungus isolates had an effect on reducing the lengths of tunnels on the stems of citrus trees after the treatment, but there are differences in effectiveness depending on the type of isolation and its concentrations., The local isolation with a concentration of 2.5 and 10 gm /L and the Indian isolation at the low concentration in reducing the length of tunnels compared to the rest of the treatments.

It is evident from the results of Table (2) that the local isolation was the most effective in reducing the length of the tunnels, as the length of the tunnels after 6 months of the treatment reached 115, 292 and 91 cm/tree, followed by the Indian isolation, in which the lengths of the tunnels reached 66, 343 and 105 cm/tree, while the Korean isolation was the least effective in reducing The lengths of the tunnels were 618, 116, and 161 cm/tree in concentrations of 2.5, 5 and 10 g/L, respectively, while the control treatment was 391 cm/tree and in the trees treated with *I. fumosoroseus* 173 cm/tree, while the chemical pesticide was 0.00 cm/tree.

The statistical analysis showed that there are significant statistical differences between the general average of tunnels lengths during the monthly readings after the treatment, as the Indian isolate reached 159, 396 and 275 cm/tree, and the Korean isolate reached 666, 337 and 226 cm/tree, as for the local isolation 112, 305 and 124 cm/tree for concentrations of 2.5, 5 and 10 g/L, respectively. And the presence of statistical differences between the cumulative average for tunnels lengths of the different isolates, which is 277, 410 and 180 cm/tree for Indian, Korean and local isolates respectively, in addition to the presence of statistical differences between the cumulative rates of concentrations, where the local isolation was the most efficient in reducing the lengths of the tunnels, whose average lengths were 313, 346 and 208 cm / tree for concentrations of 2.5 and 5/L, respectively Table(2).

It is evident through the results that the local isolation is superior to the two concentrations of 2.5 and 10 g / L and the concentration is 2.5 g /L for the Indian isolate, The superiority of these concentrations of the two isolates may be due to the attracting effect of concentration and thus feeding and exposure to a greater extent, and thus individuals will carry the pathogen agent to its colonies, the spread of the fungus spores, and the occurrence of infection among individuals and death. And therefore, The length of the tunnels gradually decreases over time, which may mean the slow and gradual effect of the fungus on the termite insect, causing a decrease in the activity of the worker, and thus the collapse of the colony in the long run, This is consistent with what Nassry (2017) reported in a similar study when using *I. fumosoroseus* on *M. diversus* infested olive trees using direct spraying under field conditions in Iraq. And the use of pathogenic fungi is characterized by its slow effect compared to chemical methods, as well as its ability to multiply and its effect on the cast system in colonies of a termites insect (Grace and Zoberi, 1992), What increases the efficiency of the fungi is its rapid spread in the colony, so infecting one member of the colony and through contact with members of the colony transfers the fungus spore to the inside of the colony (Myles and Nutting, 1988).

As for the concentration exceeding 10 g /L, it is due to the exposure of individuals to high doses of fungus spores, and thus the arrival of sufficient quantities of the spores to the inside of the termite colonies and the occurrence of collapse in the colony.

**3. The effect of different concentrations of fungus isolates on the number of workers present in the new tunnels on the stems of citrus trees after treatment**

It was evident from the results of the study that all *L. lecanii* isolates were effective in reducing the number of workers in the 20 cm tunnel after the treatment. However, differences in effectiveness appeared in different types of isolates and concentrations. Where all the isolates caused a gradual decrease in the number of workers with the advance of the treatment time, while the control treatment showed an increase in the number of e workers, The local isolates with concentrations 2.5 and 10 g /L, and the Korean isolate with concentrations 2.5 and 5 g /L decreased the number of workers in the new tunnel with a 20 cm length.

It is evident from the results in Table (3) that all isolates were highly effective in gradually reducing the number of workers, compared to their high numbers before treatment, and Indian isolation was the most efficient in reducing the number of workers, as the number of workers after 6 months of treatment reached 0.33, 5.33 and 10. 00 workers, followed by the Korean isolation, where the number of workers reached 7.67, 2.00 and 13.33 workers, While the local isolation was the least effective in reducing the number of workers, as the number of workers was 17.33, 19.33 and 17.00 workers at concentrations of 2.5, 5 and 10 g /L, while the number of workers in the control treatment was 21.33 and 11.33 and in the treatment of *I. fumosoroseus*, while the pesticide was 0.00 workers .

It is evident from Table (3) that the local isolation of the fungus in all its concentrations was effective in reducing the number of workers inside the tunnel for a period of 5 months, reaching 9.33, 13.00 and 4.00 worker / tunnel, after it was before the treatment 33.33, 20.67 and 24.00 worker / tunnel and started Their numbers increase after the sixth month of treatment, which indicates that the spore effectiveness last for 5 months.

Statistical analysis proved the presence of significant statistical differences between the number of workers in all treatment during the monthly readings after the treatment, while there were no significant differences in the general average for the number of workers, as the number of workers for the Indian isolation reached 1.61, 8.11 and 7.39, and for the Korean isolation 8.00, 7.89 and 5.17, while the local 10.94 10.11 and 11.06 worker / tunnel for concentrations of 2.5, 5 and 10 g/L, respectively. The results also showed that there are statistical differences between the rates of the cumulative number of workers for the different isolates, which amounted to 5.70, 7.02 and 10.70 for the Indian, Korean and local isolates, respectively Table (3).

It is evident through the results of Table (3) the superiority of Indian isolation in all its concentrations, and this may be attributed to the effect of the fungus on the behaviors of the workers and its repellent effect on the workers, thus reducing infection during different periods of time. In this regard, Su (1982) mentioned the occurrence of termite dispersal behaviors in environments containing biological control agents, pesticides, and slow-acting substances. The termites does not die directly at the treatment site, but rather its death requires time and is considered to expel the individuals of the termites in the event of their presence and to add its to the soil (Su, 1982).

**4. The effect of different concentrations of fungus isolates on the behavior of worker workers in building tunnels (20 cm specified)**

**A-Impact on the length of the tunnel built by the workers**

The results of the study showed that all *L. lecanii* isolates showed effectiveness in reducing the length of the specified tunnel and affected the behavior of workers in terms of its building on citrus trees after treatment, but there is a variation in their effect according to the different types of isolation and concentration. As all isolates caused a gradual decrease in the length of the specified tunnel with the progression of the time period, compared to the fact that the length of the old tunnel was not affected by the control treatment. The local isolation with high concentration and the Indian isolation at the low concentration, while the Korean isolation with 5 gm / liter concentration decreased the specified tunnel length compared to the rest of the treatments.

It is evident from the results of Table (4) that the Indian isolation of the fungus was the most efficient in reducing the length of the specified tunnel, as it reached after 6 months of treatment 6.67, 13.33 and 0.00 cm, followed by the Korean isolation, where it was 13.33, 6.67 and 6.67 cm, while the local isolation was the least effective. In reducing the specified 20 cm tunnel length, which was 13.33, 13.33 and 11.33 cm, at concentrations of 2.5, 5 and 10 g /L, respectively, while the control treatment was 20.00 cm, and in *I. fumosoroseus* treatment it was 8.00 and the pesticide was 0.00 cm.

The statistical analysis proved that there were no statistical differences between the general average of tunnels lengths except for the 5 gm / L concentration for the Indian isolation and the 2.5 concentration for the Korean isolate, which only differed significantly between them, and that all the concentrations differed significantly from the control treatment. It is also evident that the concentration exceeds 10 g / liter for the Indian isolate, as the reason for the absence of the tunnel after six months of treatment compared to all other concentrations and isolates.

**B. The effect on the number of workers**

It was evident from the results of the study that all of the fungus isolates had an effect on reducing the number of workers in the old tunnel on the stems of citrus trees after the treatment, but there are differences in effectiveness depending on the type of isolation and their concentrations, which caused a gradual decrease in the number of workers present in the old tunnel with the progression of the treatment time compared to its increase in a control treatment, The Indian isolate with its concentrations of 2.5 and 5 gm/L and the Korean isolate with the low concentration in reducing the number of workers compared to all treatments.

It is evident from Table (5) that the Indian isolation of the fungus was the most efficient in reducing the number of workers, as it reached after 6 months after the treatment 0.67, 4.00 and 0.00 workers, followed by the effect of the Korean isolation, as the number of workers in it reached 3.00, 6.00 and 2.67 female workers Whereas, local isolation was the least effective in reducing the number of workers in the specified tunnel, reaching 28.67, 12.67 and 9.67 workers for concentrations of 2.5, 5 and 10 g /L, respectively, while the control treatment was 17.3 workers, and the treatment of *I. fumosoroseus* was 6.3 workers and in the chemical pesticide 0.00 employed.

It is evident from Table (5) that local isolation was very effective in gradually reducing the number of workers, especially the concentrations of 5 and 10 g / L after 5 months of treatment, which amounted to 18.33, 8.67 and 1.67 workers / tunnels, after they were 33.33, 20.67 and 24.00 workers / tunnel before the treatment. For the concentrations of 2.5 and 5,10 g / L, respectively, its numbers began to increase after 6 months of treatment in all concentrations, indicating the deterioration of the strain and the death of its spores.

The statistical analysis showed that there are significant statistical differences between the general rate of the workers number during the monthly readings after the treatment, as it reached 0.56, 3.33 and 8.83 for the Indian isolation, 9.17, 7.94 and 5.17 for the Korean isolation, and for the local isolation 18.94, 8.72 and 5.72 for the concentrations of 2.5, 5 and 10 g/L. respectively, And the existence of statistical differences between the average of cumulative number of workers for the different isolates, which was 4.24, 7.43 and 11.13 workers for Indian, Korean and local isolates, respectively. While the cumulative statistical analysis of the concentrations did not show any statistical differences between the average number of workers, where the number of workers reached 9.56, 6.67 and 6.57 workers at concentrations of 2.5, 5 and 10 g /L, respectively, Table (5). The results showed that the Indian isolate was superior to the concentrations 2.5 and 5 g /L, while the high concentration caused no infection after 6 months after the treatment and the local strain was superior to the two concentrations 5 and 10 g / L after five months of treatment.

**Table 1 :** The number of tunnels in citrus trees treated with isolates and different concentrations of *L. Lecanii*

| Average | Date of Sample (month)after treatment | | | | | | | Contrate g/L | Fungus Isolate |
|---|---|---|---|---|---|---|---|---|---|
| | 201911//5 | 2019/10/5 | 2019/9/5 | 2019/8/5 | 2019/7/5 | 2019/6/5 | Pre treatment | | |
| 2.89 | 1.33 | 1.67 | 1.67 | 4.67 | 3.67 | 4.33 | 4.00 | 2.5 | H |
| 5.56 | 5.67 | 5.67 | 6.67 | 5.00 | 5.00 | 5.33 | 3.67 | 5 | |
| 5.89 | 4.67 | 9.33 | 6.67 | 6.00 | 3.67 | 5.00 | 3.00 | 10 | |
| | | | | | | | | | |
| 9.94 | 9.67 | 15.00 | 9.00 | 10.33 | 7.67 | 8.00 | 5.00 | 2.5 | K |
| 6.39 | 7.33 | 8.33 | 3.67 | 6.33 | 5.67 | 7.00 | 3.33 | 5 | |
| 7.33 | 7.33 | 8.33 | 5.67 | 8.33 | 6.00 | 8.33 | 5.00 | 10 | |
| | | | | | | | | | |
| 3.00 | 3.67 | 2.67 | 3.00 | 3.00 | 3.00 | 2.67 | 3.33 | 2.5 | M |
| 4.78 | 5.33 | 3.67 | 5.67 | 5.33 | 3.67 | 5.00 | 2.33 | 5 | |
| 3.33 | 2.33 | 4.33 | 3.33 | 3.33 | 2.67 | 4.00 | 3.00 | 10 | |
| 2.40 | 5.88 (N.S) | | | | | | | LSD | |
| | 5.26 | 6.56 | 5.04 | 5.81 | 4.56 | 5.52 | 3.63 | Average of Date | |
| | 1.96 (N.S) | | | | | | | LSD | |
| | 8.00 | 4.67 | 5.33 | 4.00 | 3.67 | 5.33 | 2.67 | Isaria(Average of control | |
| | 9.67 | 10.00 | 8.00 | 8.33 | 8.00 | 7.33 | 4.33 | Distilled water(Average of control | |
| | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 6.67 | pesticide | |
| | 7.38 (N.S) | 9.15 (N.S) | 4.48 | 6.45 | 3.90 | 4.85 | 2.81 | LSD | |
| Average of Isolate | The sixth | The fifth | The fourth | The third | The second | The first | Fungus Isolate | | |
| 4.78 | 3.89 | 5.56 | 5.00 | 5.22 | 4.11 | 4.89 | H | | |
| 7.89 | 8.11 | 10.56 | 6.11 | 8.33 | 6.44 | 7.78 | K | | |
| 3.70 | 3.78 | 3.56 | 4.00 | 3.89 | 3.11 | 3.89 | M | | |
| 1.39 | 3.39 (N.S) | | | | | | LSD | | |
| Average of Concentration | The sixth | The fifth | The fourth | The third | The second | The first | Constration (g/L) | | |
| 5.28 | 4.89 | 6.44 | 4.56 | 6.00 | 4.78 | 5.00 | 2.5 | | |
| 5.57 | 6.11 | 5.89 | 5.33 | 5.56 | 4.78 | 5.78 | 5 | | |
| 5.52 | 4.78 | 7.33 | 5.22 | 5.89 | 4.11 | 5.78 | 10 | | |
| 1.39 (N.S) | 3.39 (N.S) | | | | | | LSD | | |
| M=Local Isolate K=Korean Isolate H=Indian Isolate | | | | | | | | | |

**Table 2 :** The lengths of tunnels built on the stems of citrus trees treated with isolates and concentrations of *L. lecanii*

| Average | Sample (Month)after treatment Date of | | | | | | Pre Treatment | Concetrate g/L | Fungus Isolate |
|---|---|---|---|---|---|---|---|---|---|
| | 2019/11/5 | 2019/10/5 | 2019/9/5 | 2019/8/5 | 2019/7/5 | 2019/6/5 | | | |
| 159 | 66 | 106 | 136 | 173 | 242 | 234 | 230 | 2.5 | H |
| 396 | 343 | 393 | 483 | 387 | 378 | 393 | 433 | 5 | |
| 275 | 105 | 275 | 300 | 307 | 322 | 341 | 243 | 10 | |
| | | | | | | | | | |
| 666 | 618 | 746 | 692 | 676 | 591 | 673 | 730 | 2.5 | K |
| 337 | 116 | 362 | 343 | 367 | 399 | 435 | 294 | 5 | |
| 226 | 161 | 189 | 161 | 239 | 274 | 331 | 350 | 10 | |
| | | | | | | | | | |
| 112 | 115 | 103 | 125 | 100 | 133 | 98 | 132 | 2.5 | M |
| 305 | 292 | 299 | 345 | 293 | 308 | 292 | 212 | 5 | |
| 124 | 91 | 129 | 124 | 134 | 134 | 134 | 162 | 10 | |
| 118 | 288 | | | | | | | LSD | |
| | 212 | 289 | 301 | 297 | 309 | 326 | 309 | average of Date | |
| | 96 | | | | | | | LSD | |
| | 173 | 191 | 204 | 221 | 285 | 250 | 279 | )Isaria( Average of Control | |
| | 391 | 524 | 489 | 532 | 577 | 539 | 511 | Average of Control (Distilled water) | |
| | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 527 | Pesticides | |
| | 255 | 289 | 295 | 291 | 290 | 266 | 313 | LSD | |
| Average of Isolate | The Sixth | The fifth | The fourth | The third | The second | The first | Fungus Isolate | | |
| 277 | 171 | 258 | 307 | 289 | 314 | 323 | H | | |
| 410 | 299 | 432 | 399 | 427 | 421 | 480 | K | | |
| 180 | 166 | 177 | 198 | 176 | 192 | 175 | M | | |
| 68 | 166 | | | | | | LSD | | |
| Average of Concetrate | The Sixth | The fifth | The fourth | The third | The second | The first | Concetrate(g/L) | | |
| 313 | 266 | 318 | 318 | 316 | 322 | 335 | 2.5 | | |
| 346 | 251 | 351 | 390 | 349 | 362 | 373 | 5 | | |
| 208 | 119 | 198 | 195 | 227 | 243 | 269 | 10 | | |
| 68 | 166 | | | | | | LSD | | |
| H=Indian Isolate | | | K=Korean Isolate | | | M=Local Isolate | | | |

**Table 3 :** Number of worker workers present in the new 20 cm tunnel on tree trunks treated with isolates and concentrations of the fungus *L. Lecanii*

| Average | Sample Date(Month)after treatment | | | | | | Pre treatment | Concetrate g/L | Fungus Isolate |
|---|---|---|---|---|---|---|---|---|---|
| | 2019/11/5 | 2019/10/5 | 2019/9/5 | 2019/8/5 | 2019/7/5 | 2019/6/5 | | | |
| 1.61 | 0.33 | 1.33 | 3.00 | 1.33 | 1.00 | 2.67 | 4.67 | 2.5 | H |
| 8.11 | 5.33 | 10.67 | 14.67 | 3.00 | 7.00 | 8.00 | 9.67 | 5 | |
| 7.39 | 10.00 | 12.67 | 5.00 | 2.33 | 5.67 | 8.67 | 18.00 | 10 | |
| | | | | | | | | | |
| 8.00 | 7.67 | 12.33 | 8.33 | 4.00 | 9.33 | 6.33 | 19.00 | 2.5 | K |
| 7.89 | 2.00 | 4.33 | 5.00 | 17.33 | 10.33 | 8.33 | 20.67 | 5 | |
| 5.17 | 13.33 | 4.67 | 2.67 | 2.33 | 2.00 | 6.00 | 13.67 | 10 | |
| | | | | | | | | | |
| 10.94 | 17.33 | 9.33 | 11.33 | 5.00 | 14.67 | 8.00 | 33.33 | 2.5 | M |
| 10.11 | 19.33 | 13.00 | 15.00 | 2.67 | 3.00 | 7.67 | 20.67 | 5 | |
| 11.06 | 17.00 | 4.00 | 5.67 | 4.33 | 25.67 | 9.67 | 24.00 | 10 | |
| 2.92 (N.S) | 12.39 (N.S) | | | | | | | LSD | |
| | 10.26 | 8.04 | 7.85 | 4.70 | 8.74 | 7.26 | 15.69 | average of Date | |
| | 4.13 (N.S) | | | | | | | LSD | |
| | 11.33 | 8.00 | 5.00 | 3.00 | 2.00 | 2.67 | 10.00 | )Isaria( Average of Control | |
| | 21.33 | 18.00 | 7.33 | 13.33 | 7.67 | 11.67 | 5.00 | Average of Control (Distilled water) | |
| | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 9.67 | Pesticides | |
| | 13.37 | 14.11 (N.S) | 14.0 (N.S) | 9.21 | 11.61 | 5.79 | 22.19 (N.S) | LSD | |
| Average of Isolate | The sixth | The fifth | The fourth | The third | The second | The first | Fungus Isolate | | |
| 5.70 | 5.22 | 8.22 | 7.56 | 2.22 | 4.56 | 6.44 | H | | |
| 7.02 | 7.67 | 7.11 | 5.33 | 7.89 | 7.22 | 6.89 | K | | |
| 10.70 | 17.89 | 8.78 | 10.67 | 4.00 | 14.44 | 8.44 | M | | |
| 2.92 | 7.15 (N.S) | | | | | | LSD | | |
| Average of Concentrate | The sixth | The fifth | The fourth | The third | The second | The first | Concetrate (g/L) | | |
| 6.85 | 8.44 | 7.67 | 7.56 | 3.44 | 8.33 | 5.67 | 2.5 | | |
| 8.70 | 8.89 | 9.33 | 11.56 | 7.67 | 6.78 | 8.00 | 5 | | |
| 7.87 | 13.44 | 7.11 | 4.44 | 3.00 | 11.11 | 8.11 | 10 | | |
| 2.92 (N.S) | 7.15 (N.S) | | | | | | LSD | | |
| H=Indian Isolate | | | K=Korean Isolate | | | M=Local Isolate | | | |

**Table 4 :** Tunnel lengths 20 cm specified in trees treated with isolates and different concentrations of *L. lecanii* fungus

| Average | Date of sample(Month)after treatment | | | | | | | Concetrate g/L | Fungus Isolate |
|---|---|---|---|---|---|---|---|---|---|
| | 2019/11/5 | 2019/10/5 | 2019/9/5 | 2019/8/5 | 2019/7/5 | 2019/6/5 | Pre treatment | | |
| 8.89 | 6.67 | 6.67 | 6.67 | 6.67 | 20.00 | 6.67 | 20.00 | C1 | H |
| 15.56 | 13.33 | 13.33 | 13.33 | 13.33 | 20.00 | 20.00 | 20.00 | C2 | |
| 14.72 | 0.00 | 8.33 | 20.00 | 20.00 | 20.00 | 20.00 | 20.00 | C3 | |
| | | | | | | | | | |
| 17.00 | 13.33 | 18.67 | 18.33 | 16.67 | 15.00 | 20.00 | 20.00 | C1 | K |
| 10.33 | 6.67 | 6.67 | 6.67 | 15.33 | 13.33 | 13.33 | 20.00 | C2 | |
| 12.22 | 6.67 | 6.67 | 6.67 | 13.33 | 20.00 | 20.00 | 20.00 | C3 | |
| | | | | | | | | | |
| 13.56 | 13.33 | 13.33 | 13.33 | 13.33 | 13.33 | 14.67 | 20.00 | C1 | M |
| 13.33 | 13.33 | 13.33 | 13.33 | 13.33 | 13.33 | 13.33 | 20.00 | C2 | |
| 12.89 | 11.33 | 11.67 | 11.67 | 13.33 | 11.67 | 17.67 | 20.00 | C3 | |
| 6.33 | 15.51 (N.S) | | | | | | | LSD | |
| | 9.41 | 10.96 | 12.22 | 13.92 | 16.30 | 16.19 | 20.00 | Average of Date | |
| | 5.17 | | | | | | | LSD | |
| | 8.0 | 6.7 | 6.7 | 6.7 | 8.0 | 9.0 | 20.00 | )Isaria(Average of control | |
| | 20.0 | 20.0 | 20.0 | 13.3 | 13.0 | 20.0 | 20.00 | Average of Control(Distill water) | |
| | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 20.00 | Pesticides | |
| | 16.50 (N.S) | 16.54 (N.S) | 15.76 (N.S) | 16.61 (N.S) | 14.0 (N.S) | 11.96 | - | LSD | |
| Average of Isolate | The sixth | The fifth | The fourth | The third | The second | Thr first | Fungus Isolate | | |
| 13.06 | 6.67 | 9.44 | 13.33 | 13.33 | 20.00 | 15.56 | H | | |
| 13.19 | 8.89 | 10.67 | 10.56 | 15.11 | 16.11 | 17.78 | K | | |
| 13.26 | 12.67 | 12.78 | 12.78 | 13.33 | 12.78 | 15.22 | M | | |
| 3.65 (N.S) | 8.96 (N.S) | | | | | | LSD | | |
| Average of conctrat | The sixth | The fifth | The fourth | The third | The second | Thr first | Concetrate(g/L) | | |
| 13.15 | 11.11 | 12.89 | 12.78 | 12.22 | 16.11 | 13.78 | C1 | | |
| 13.07 | 11.11 | 11.11 | 11.11 | 14.00 | 15.56 | 15.56 | C2 | | |
| 13.28 | 6.00 | 8.89 | 12.78 | 15.56 | 17.22 | 19.22 | C3 | | |
| 3.65 (N.S) | 8.96 (N.S) | | | | | | LSD | | |
| H=Indian Isolate | | | K=Korean Isolate | | | M=Local Isolate | | | |

**Table 5 :** Number of worker workers present in the 20 cm tunnel specified on the trunks of citrus trees treated with different isolates and concentrations of *L. lecanii* fungus

| Average | Date of sample(Month)after treatment | | | | | | | Concentrate g/L)( | Fungus Isolate |
|---|---|---|---|---|---|---|---|---|---|
| | 2019/11/5 | 2019/10/5 | 201/9/5 | 2019/8/5 | 2019/7/5 | 2019/6/5 | Pre treatment | | |
| 0.56 | 0.67 | 0.00 | 0.00 | 0.00 | 1.67 | 1.00 | 4.67 | C1 | H |
| 3.33 | 4.00 | 11.00 | 1.00 | 0.33 | 1.33 | 2.33 | 9.67 | C2 | |
| 8.83 | 0.00 | 8.67 | 3.00 | 7.33 | 20.33 | 13.67 | 18.00 | C3 | |
| | | | | | | | | | |
| 9.17 | 3.00 | 7.33 | 8.67 | 21.00 | 7.67 | 7.33 | 19.00 | C1 | K |
| 7.94 | 6.00 | 4.33 | 12.33 | 9.00 | 7.00 | 9.00 | 20.67 | C2 | |
| 5.17 | 2.67 | 3.33 | 3.00 | 5.00 | 5.67 | 11.33 | 13.67 | C3 | |
| | | | | | | | | | |
| 18.94 | 28.67 | 18.33 | 30.67 | 21.33 | 4.33 | 10.33 | 33.33 | C1 | M |
| 8.72 | 12.67 | 8.67 | 7.33 | 12.00 | 5.00 | 6.67 | 20.67 | C2 | |
| 5.72 | 9.67 | 1.67 | 6.00 | 5.00 | 4.00 | 8.00 | 24.00 | C3 | |
| 7.78 | 19.01 (N.S) | | | | | | | LSD | |
| | 7.48 | 7.04 | 8.00 | 9.00 | 6.33 | 7.74 | 18.19 | Average of Date | |
| | 6.36 (N.S) | | | | | | | LSD | |
| | 6.3 | 0.7 | 0.7 | 0.0 | 2.7 | 0.0 | 10.00 | )Isaria(Average of control | |
| | 17.3 | 23.3 | 10.7 | 5.7 | 6.0 | 8.0 | 5.00 | Average of Control(Distill water) | |
| | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 9.67 | Pesticides | |
| | 22.3 (N.S) | 15.0 (N.S) | 27.1 (N.S) | 19.7 (N.S) | 11.90 (N.S) | 10.31 (N.S) | 22.19 (N.S) | LSD | |
| Average of Isolate | The sixth | The fifth | Thr fourth | The third | The second | The first | Fungus Isolate | | |
| 4.24 | 1.56 | 6.56 | 1.33 | 2.56 | 7.78 | 5.67 | H | | |
| 7.43 | 3.89 | 5.00 | 8.00 | 11.67 | 6.78 | 9.22 | K | | |
| 11.13 | 17.00 | 9.56 | 14.67 | 12.78 | 4.44 | 8.33 | M | | |
| 4.49 | 11.00 (N.S) | | | | | | LSD | | |
| Average of Concetrat | The sixth | The fifth | Thr fourth | The third | The second | The first | Concetrat(g/L) | | |
| 9.56 | 10.78 | 8.56 | 13.11 | 14.11 | 4.56 | 6.22 | C1 | | |
| 6.67 | 7.56 | 8.00 | 6.89 | 7.11 | 4.44 | 6.00 | C2 | | |
| 6.57 | 4.11 | 4.56 | 4.00 | 5.78 | 10.00 | 11.00 | C3 | | |
| 4.49 (N.S) | 11.00 (N.S) | | | | | | LSD | | |
| H=Indian Isolate | | | K=Korean Isolate | | | M=Local Isolate | | | |

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