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**INTRODUCTION**

Wheat-aphid *Schizaphis graminum* (Rondani) which caused directly-losses of yields by sap sucking from invested plants and indirectly-losses as a transporter of fungal and viral diseases, (Abro, et al., 2004). In Upper Egypt, the wheat damage caused by aphid-insects was recorded mostly infestations reached up to "23%" (El-Heneidy and Adly, 2012). The bird-cherry or oat-aphid *Rhopalosiphum padi* L.; green-bug *S. graminum*; *Rhopalosiphum maidis* Fitch. and *Sitobion avenae* F. one of the most important types of insect-aphids in Arab Republic of Egypt (El-Fatih, 2000). Aphids are commonly controlled by pesticides. Though, pesticide-resistance has researched to finding a new method to insect-aphids control. In addition to, randomly using for the several synthetic-insecticides to mammalians protects from dangerous health-hazards. The integrated pest management concluded entomopathogenic-fungi as a biological control agent, can be used in controlling against aphids attack, which in turn works often led to the natural-mortality of population density. Such as, the entomopathogenic fungi (EPF), *Metarhizium anisopliae*, Metsch., which is used as a pathogen for many pests and is used within the Integrated-Pest-Management as biological-control in all parts of the world. (Sandhu, et al., 2012), therefore the wheat-aphid *M. anisopliae*, considered one of a favorable method for biological-control against insect-pests (Zibaei et al., 2011). The current investigation aim to studding and suggestion some alternatives methods within the Integrated-Pest-Management "IPM" to controlling of wheat-aphid, *S. graminum* by determining the toxicity of four isolates of entomopathogenic fungi, *Metarhizium anisopliae* (Metsch.) against *S. graminum* adult females under laboratory conditions to evaluate their effects and efficiency as natural insecticides.

**MATERIALS AND METHODS**

**Rearing of Wheat Aphid:**

Aphid colonies were maintained according to (El-Gendy, 2009). The Laboratory strain of wheat aphid, *Schizaphis graminum* (Rondani) was obtained from a colony cultured at Biological control Department, Plant Prot. Res. Inst., Agric. Res. Centre, Dokki-Giza-Egypt. These strains were reared for several generations under laboratory conditions on wheat plants in plastic pots. Aphid colonies were prevented from external contamination by placing infested plants in cages covered with a muslin cloth.

**Bioassay:**

Four conc. of spores suspension 1×10^5, 1×10^6, 1×10^7 and 1×10^8 conidia/ml were prepared in (0.1%) TritonX-100 (added as surfactant) for each of entomopathogenic fungi isolates, *Metarhizium anisopliae* (M 1, M 2, M 6 and M 9) were isolated in "Bioinsecticides Production Unit" Plant Prot. Res. Inst., Agric. Rese. Center - Giza-Egypt in previous research (Altahawi et al., 2020). Spray method was used to test the virulence of fungi isolates. "Abbott's formula", Abbott 1925, used in correct of mortalities%. Ldp line software "Bakr, 2000" used to calculating the values of "LC50, LC90 and slope" according to "Finney, 1971".

**RESULTS AND DISCUSSION**

**Virulence of entomopathogenic isolates against aphid, *S. graminum***:

Four isolates of the entomopathogenic fungi, *Metarhizium anisopliae* were chosen according to the highest mortality of the previous experiment (M 01, M 02,
Mortality % of S. graminum to entomopathogenic fungi M. anisopliae isolates. Percentage mortality values after exposing to concentrations-series (1×10⁸, 1×10⁹, 1×10⁷ and 1×10⁶ spores/ml) illustrated in (Table 1 and Fig. 1) after 10-days from treatments. Mortality % showed gradual-increasing over with increasing of spores-conc. and exposure-time. Low-conc. of (1×10⁸ spores/ml) resulted (48, 53, 32 and 38%) for (M1, M2, M6 and M9) respectively. After 10-days from treatments, but high-conc. of (1×10⁹ spores/ml) resulted (88, 91, 76 and 79%) for (M1, M2, M6 and M9) respectively. When mortality was assessed after the same consecutive days, respectively. Fig (2), illustrated that, sporulation of M. anisopliae on cadavers of adult stage of S. graminum which prove the pathogenicity of the entomopathogenic fungi against aphid.

Table 1. Mortality % of S. graminum treated with series concentrations of (M 01, M 02, M 06 and M 09) isolates after ten days of treatment.

<table>
<thead>
<tr>
<th>Line Name</th>
<th>Concentrations</th>
<th>M 01</th>
<th>M 02</th>
<th>M 06</th>
<th>M 09</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1×10⁸</td>
<td>48</td>
<td>53</td>
<td>32</td>
<td>38</td>
</tr>
<tr>
<td></td>
<td>1×10⁹</td>
<td>60</td>
<td>69</td>
<td>42</td>
<td>45</td>
</tr>
<tr>
<td></td>
<td>1×10⁷</td>
<td>74</td>
<td>82</td>
<td>60</td>
<td>67</td>
</tr>
<tr>
<td></td>
<td>1×10⁶</td>
<td>88</td>
<td>91</td>
<td>76</td>
<td>79</td>
</tr>
</tbody>
</table>

Results presented in Table (2), and illustrated in Fig. (3) revealed that, M02 isolate give the most effect against S. graminum, the LC₅₀ value of M2 was 1×10⁷ spores/ml "slope 0.278", while it give the greater LC₅₀ value for M01, M06 and M09 which were (2.7×10⁷, 2.3×10⁹, and 7.8×10⁹) spores/ml respectively. Mortality means were 7.45, 10.3, 7.35 and 8.47 for M1, M2, M6 and M9 respectively. Results obtained indicated that, the M2 isolate give highly-mortality in short time, the LT₅₀ value was 3.94 days, while the isolates others M1, M6 and M9, the LT₅₀ values were 4.32, 5.1 and 5.04 days, respectively as shown in Table (3) & Fig(4).

Table 2. LC₅₀ and mortality means between (M01, M02, M06 and M09) isolates on S. graminum.

<table>
<thead>
<tr>
<th>Lines-name</th>
<th>LC₅₀</th>
<th>Mortality %</th>
<th>The index slope</th>
<th>LC₅₀</th>
<th>Mean±SE</th>
</tr>
</thead>
<tbody>
<tr>
<td>M 01</td>
<td>5.7×10⁷, 1.68×10⁸</td>
<td>35.51 (0.278)</td>
<td>1.13×10¹²</td>
<td>7.45±0.85bc</td>
<td></td>
</tr>
<tr>
<td>M 02</td>
<td>1×10⁷, 2.3×10⁹</td>
<td>100 (0.253)</td>
<td>1.13×10¹²</td>
<td>10.32±2.35a</td>
<td></td>
</tr>
<tr>
<td>M 06</td>
<td>3.8×10⁹, 6.9×10⁹</td>
<td>4.23 (0.29)</td>
<td>6.05×10¹²</td>
<td>7.35±0.86b</td>
<td></td>
</tr>
<tr>
<td>M 09</td>
<td>7.8×10⁹, 1.3×10¹⁰</td>
<td>12.46 (0.319)</td>
<td>8.8×10¹¹</td>
<td>8.47±0.99bc</td>
<td></td>
</tr>
</tbody>
</table>

The index compared with (M2)
One way ANOVA statistical analysis indicated that significant levels of effect (F_{5,156}=2.96; P \leq 0.05) between isolates M01, M02, M06 and M09 on adult aphid.

The obtained results in this study showed that all tested isolates infected aphid treated with conc. (1×10^6, 1×10^7, 1×10^8 and 1×10^9 spores/ml) respectively. Pathogenicity of fungus increased with the increase of concentration and time. The results were compatible with Vu et al.(2007), where, the all isolates pathogenicity was reconfirmed and accumulated-mortality rates ranged among “33.3 and 100%” within (7 days), in addition, Aranya et al. (2010), who found highest concentration of 10^6 spores/ml, at the LT_{50}= 5.54 days.

Also, results were compatible with, (Lee et al. 2015, and Aker and Abaci 2016) who proved that the use of M. anisopliae can provide protection against green peach aphid Mycus persicae and hazelnut aphids Myzocallis coryli, respectively and it can be effective as biocontrol agent.

The present results were consistent with laboratory bioassay studies carried out by Entesar et al. (2020), evaluated four native of entomopathogenic fungi isolates B. bassiana and M. anisopliae on wheat-aphid S. grahamum R. at three concentrations (1×10^6, 1×10^7 and 1×10^8 conidia).Isolate (B1) was more effective against adults, The LC_{50} values of B1 were 3.11×10^6 spores/ml while M1, M2, and B2 revealed greater LC_{50} values, 6.09×10^6, 2.32×10^7 and 1.15×10^8 spores/ml, respectively.

Results were in same trend with Sahar et al. (2016), when evaluated four-conc. 1×10^6, 1×10^7, 1×10^8 and 1×10^9 spores/ml, for each entomopathogenic fungi, B. bassiana, M. anisopliae, P. lilacinus and L. antillanum against the adults of cowpea aphid, A. craccivora. Also the results were in harmony with Inas (2020) where M. anisopliae, have high toxicity and mortality rates against Cabbage Aphids, Brevicoryne brassicae occurred after 3rd days from treatments. The maximum percent of mortality (100%) occurred after the 10th day from treatment with the 3rd concentration (1×10^9 spores/ m). Furthermore Tang et al. (2019) suggested that, the entomopathogenic fungi M. anisopliae is a good as a bio-control agent against S. furcifera and N. lugens with (LT_{30}) of approximately four-days were observed with high conidial-conc. of (1×10^8 conidia/ml) of M. anisopliae, with alternative strategy for pest control. On the same line, these results were compatible with (Ogarkov and Ogarkova, 1997, and Ismail, et al., 2016) they found that, the most common fungi used for insect control belong to the genera Beauveria, Metarhizium, Paecilomyces, Verticillium, Aschersonia, and Conidiobolus.

REFERENCES


Inas, M. Y. Mostafa(2020). Entomopathogenic Fungi against Cabbage Aphids,


القدرة الإمارض لعزلات جديدة من الفطر الممرض للحشرات "$Metarhizium anisopliae$" تحت الظروف المعملية "*Schizaphis graminum*" مع إعداد بحوث وقائية للنباتات ، مركز البحوث الزراعية ، القلي ، الجزة ، كلية العلوم الزراعية البيئية ، جامعة العريش ، شمال سيناء تم اختيار أربع عزلات محلية من فطر ممرض للحشرات "$Metarhizium anisopliae$" من مجموعة من مربعات "Schizaphis graminum" الممرض للحشرات وهي (M 01، M 02، M 06 و M 09) وقد تم استخدام أربعة تركيزات من عزلات الفطر "$Metarhizium anisopliae$" من العزلات معاً بشكل معزول من جميع الحشرات ومثقلة معملياً ضد من الفطر "Schizaphis graminum" الجرياني لكل من العزلات الفطرية الأربعة 1×10⁶ و 1×10⁷ و 1×10⁸ و 1×10⁹ جراثمة/م لوح الجرياني الكاملة عمر يوم وقد أظهرت النتائج أن العزلة LC₉₀ كانت الأكثر فاعلية كما أظهرت خطوط السمية M 02