Rearing and Release of *Aphelinus mali* (Hald) (Hymenoptera: Aphelinidae), the Sole Parasitoid of Woolly Apple *Eriosoma lanigerum* (Hausmann) (Homoptera: Eriosomatidae) on Apple orchards in Ash-Shoubak

*Mazen Ateyyat, Marwan Al-Awamleh and Hassan El-Osofi*

1Faculty of Agricultural Technology, Al-Balqa’ Applied University, 19117 Al-Salt, Jordan

2National Center of Agricultural Research and Extension, Al Baqa’, Jordan

**Abstract:** *Aphelinus mali* (Hald) (Hymenoptera: Aphelinidae) is the sole parasitoid of woolly apple aphid (WAA), *Eriosoma lanigerum* (Hausmann) (Homoptera: Eriosomatidae). Five ratios of parasitoid to aphid colonies were tested to determine the suitable amount of the parasitoid that gives the highest percentage of parasitism under greenhouse conditions. Curve fitting using Table Curve program showed that releasing 6 pairs of unsexed *A. mali* adults to 100 colonies of WAA results in 98.7% of parasitism of WAA after 10 days of releasing adults of parasitoids. A valuable suppression of WAA in 2010 was recorded in two apple orchards where the parasitoid was released compared with the orchard in which no release of the parasitoid was done. However, the continuous use of pesticides by apple orchardists in the apple orchards near the experimental orchards resulted in decreasing parasitism rates in 2011 in the apple orchards where release was done due to the movement of WAA from pesticide-controlled orchards to parasitoid-controlled orchards. The government represented by ministry of agriculture is requested to apply a legislation that prevent apple orchardists use insecticides in order to give good opportunity of the parasitoid to build itself.

**Key words:** Biological control · Apple · Woolly apple aphid · Integrated Pest Management

**INTRODUCTION**

Woolly apple aphid (WAA) is an important insect pest of apple in Jordan [1]. Different predators were recorded on WAA, these are the syrphid *Syrphus confrater* [2], larvae of *Coccinella septempunctata* and *Adonia variegata* [3], *Exochomus quadripustulatus* [4] and the chrysopid *Chrysopa sceletes* [5]. *Aphelinus mali* (Hymenoptera: Aphelinidae) is the sole parasitoid that was recorded to attack WAA worldwide. It is the most important natural enemy for WAA. This parasitoid parositizes the arporeal population of WAA [6]. It is native to eastern North America and has been introduced in many regions around the world to control woolly apple aphid [7]. Studies showed that *A. mali* parasitised all stages of *E. lanigerum* reared on potted apple plants [8] and those collected from the field [9], but preferred 3rd-instar nymphs and older hosts [8]. Release impact of *A. mali* into apple orchards on the populations of the pest *E. lanigerum* was investigated in India, in 1988 [5]. The increase in parasitism by *A. mali* was assessed by surveys of the natural population before releases were made and by monitoring after releases and ranged from 49.82 to 69.08% at 3 different sites. Due to the chronic infestations of WAA, improvement of biological control has been a concern in many regions of the world for this insect pest [7]. The objective of this research was to study the impact of releasing *A. mali* on suppression of woolly apple populations in apple orchards in Ashoubak area in Jordan.
MATERIALS AND METHODS

Rearing of WAA: Fuji apple seedlings were used for rearing WAA. About 300 seedlings were used for this purpose, in which 10 seedlings were placed inside wooden cages (1 x 2 x 1 m). One hundred WAA adults were introduced inside wooden cages to establish the WAA-culture. After 2 weeks of infestation, 9 seedlings were transferred from each WAA-Culture cages to the Parasitoid-Culture cages. The remaining seedling were left to maintain the WAA-Culture.

Rearing of Aphelinus Mali: In order to determine the suitable amount of the parasitoid that gives the highest percentage of parasitism, the following ratios of parasitoid to aphid colonies were tested:

- 1 pair of unsexed *A. mali*: 10 colonies of WAA
- 1 pair of unsexed *A. mali*: 25 colonies of WAA
- 1 pair of unsexed *A. mali*: 50 colonies of WAA
- 1 pair of unsexed *A. mali*: 75 colonies of WAA
- 1 pair of unsexed *A. mali*: 100 colonies of WAA

Table curve program (Jandle Scientific) was used to find the suitable equation through which we can determine the suitable amount of the parasitoid that should be released according the population of aphid colonies in the cage. Unsexed *A. mali* adults were introduced into Parasitoid-Culture cages. In the last week of June 2010, emerged *A. mali* adults were introduced into another Parasitoid-Culture cages to maintain the Parasitoid-Culture and the remaining mummies were released into two apple orchards in Ashoubak in which about 10,000 mummies were introduced in the first orchard and about 20,000 mummies were introduced in the second orchard.

Evaluation the Impact of *A. mali* after Release: To evaluate the efficiency of releasing *A. mali*, 5 infested branches (25 cm length) were removed from 10 WAA infested trees that were chosen randomly from each of the two apple orchards in which the release of mummies was done and from an apple orchard in which no release of mummies was done. The removed branches were kept inside ice-box and transferred into the laboratory. Branches were investigated under the binocular microscope and numbers of both mummified and unparasitized aphids were recorded. Samples were taken fortnightly from August 2008 to the end of October 2011.

RESULTS AND DISCUSSION

Five ratios of the parasitoid *Aphelinus mali* to woolly apple aphid were used to find the best release ratio that results in the highest percent of parasitism under greenhouse conditions (Table 1). Results showed that releasing one pair of the parasitoid to 10 colonies of WAA resulted in 100 % parasitism rate and this rate of release was significantly not different with the release rate of one pair of the parasitoid to 25 colonies of WAA that resulted in a parasitism percent of 96.3 (Table 1).

Table 1: Percent of parasitism of woolly apple aphid by *Aphelinus mali* using different release ratios

<table>
<thead>
<tr>
<th>Unsexed pair of <em>A. mali</em>:WAA colonies (% of parasitoid to WAA)</th>
<th>Percent of Parasitism ± SE</th>
</tr>
</thead>
<tbody>
<tr>
<td>1:10(0.1)</td>
<td>100a±0.00</td>
</tr>
<tr>
<td>1:25(0.04)</td>
<td>96.3a±1.86</td>
</tr>
<tr>
<td>1:50(0.02)</td>
<td>37.7b±8.97</td>
</tr>
<tr>
<td>1:75(0.013)</td>
<td>18c±2.08</td>
</tr>
<tr>
<td>1:100(0.01)</td>
<td>14c±2.08</td>
</tr>
</tbody>
</table>

Due to the great variations recorded in the fecundity and longevity of *A. mali* recorded by Lundie[10], who showed no relationship with the longevity of adult female of the parasitoid with egg laying capacity. For this, we used TableCurve program (Jandle Scientific) to find the best equation that describes the ratio of parasitoid release with the available number of woolly apple aphid colonies in the rearing cages based on releasing unsexed pairs of the adult parasitoids. The resulted equation is:

\[ \text{Ln(percent of parasitism of WAA)} = a + b/(\text{Percent of } A. mali \text{ to WAA})^{1.5} \]

| Parm | Value  | Std Error | t-value | 95% Confidence Limits | P>|t| |
|------|--------|-----------|---------|-----------------------|-----|
| a    | 4.747245505 | 0.058569890 | 81.05266257 | 4.620712951 | 4.873778059 | 0.00000 |
| b    | -0.00227963 | 0.000321888 | -7.08204563 | -0.00297502 | -0.00158423 | 0.00001 |
Curve fitting (Fig. 1) using the above equation indicates that we have to release 6 pairs of unsexed *A. mali* adults to 100 colonies of WAA to achieve 98.7% of parasitism of WAA after 10 days of releasing adults of parasitoids.

At the start of the twenty-first century, the opportunities and need for effective biological control are greater than ever: pest resistance continues to be a problem, pesticides are being withdrawn on environmental grounds without suitable replacements [11]. In Jordan, control of woolly apple aphid depends mainly on insecticides and particularly on imidacloprid that is used as soil drench application. However, apple cultivation in Ash-Shoubak area depends on ground water irrigation. So, using imidacloprid as drench results in polluting the ground water. For this, biological control of WAA by using its sole parasitoid provides an alternative safe solution to imidacloprid. Successful inoculative biological control of woolly apple aphid in apple orchards throughout Europe by the release of *A. mali* was recorded [12]. In the present study, *A. mali* was reared on two years old WAA-infested seedlings and the produced mummies were released in June 2009 into two apple orchards in Ash-Shoubak area in which about 20,000 mummies were released in the first orchard and 10,000 mummies were released in the second apple orchard. The release was in the form of inoculative biological control as this type of biological control has been most successful with perennial crops (fruit plantations and forests), where the long-term nature of the ecosystem enables the interactions between pest and natural enemy to become fully established over a period of time. Before releasing *A. mali* in June 2009, no valuable variations in WAA populations in the two apple orchards where release of *A. mali* was done and in the orchard in which no release was done (Fig. 2). However, valuable decrease in WAA populations were recorded in 2010 in the orchards where release was done compared with populations in the orchard where no release was done (Fig. 2). In 2011, no valuable variations were recorded in populations of WAA in the three apple orchards (Fig. 2). Even though no clear differences in the number of mummified aphids were recorded in the three orchards in 2010, but a slight increase in mummified aphids were recorded in both orchards in which *A. mali* was released in 2011 (Fig. 3). Significant increase in parasitism rate was recorded in 2010 after release of parasitoid mummies (Fig. 4). The highest rate of parasitism was recorded in November 2010 in the orchard in which 20,000 mummies were released in June 2010 (Fig. 4). Also, both orchards where release was done showed an increase in parasitism rate compared with the orchard where no release was done and generally the parasitism rate was lower than that recorded in 2010 as the highest recorded rate of parasitism was 23%. Even though a valuable increase in the number of mummified aphids were recorded in 2011 in the orchards where release was
Finally, it seems that introduction of *A. mali* in Ash-Shoubak does not result in a satisfied suppression of WAA populations and this because Ash-Shoubak is a small area that has an extensive plantations of Apple and most orchardists still use pesticides in controlling the pest that resulted in its movement to orchards where no pesticides used. So, for a successful biological control of WAA in Ash-Shoubak by using its sole parasitoid

Fig. 4: Percentage of parasitism of WAA by *A. mali* per five 25-cm length twigs of *Golden delicious* apple. Orchard I: no release of *A. mali*, Orchard II: Release of 10,000 mummified aphids, Orchard III: Release of 20,000 mummified aphids. Release was done in the last week of June 2010.

Fig. 5: Monthly average temperature (°C) and average monthly relative humidity (%) in Ash-Shoubak area from August 2009 to October 2011.

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