The Effect of Palizin, Tondexir and Pymetrozine on The Green Cotton Aphid *Aphis gossypii*

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Abstract

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The aim of this study was to evaluate the effect of plant-based pesticides on the green aphid *A. gossypii* and also investigating the effect of adding citral oil to the plant pesticides. To carry out experiments on *A. gossypii* in nymph and adult stages, young soybean leaves of DPX cultivar were used (Katoul). For bioassay, first, the bulk solution was prepared and then, other concentrations (eight concentrations for each insecticide) were prepared and used. The results of this study showed that the palizin insecticide with citral oil at LC_{50} of 359.669 ml/l had the greatest effect on nymphs in-vitro, but when the same nymph stage was evaluated under greenhouse conditions, the highest efficacy was obtained with the mixture of pymetrozine and citral oil with LC_{50} of 295.242 ml/L. In the experiments performed on the adult stage under both greenhouse and laboratory conditions, the most desirable treatment was that of the mixture of herbal insecticide palizin and citral oil, and the LC_{50} obtained was 807.88 and 1249.14 ml/L under laboratory and greenhouse conditions, respectively.

Keywords: Palizin, pymetrozine, tondexir, citral oil, cotton green aphid.

Introduction

Green cotton aphid (*Aphis gossypii* Glover) is an important pest of various plants, such as cotton, soybean, cucumber, squash, tomato, etc., in different parts of the world (Abou-El-Hagag, 1998; Kapatos *et al*., 1998; Slosser *et al*., 1998). In recent years, the indiscriminate use of chemical pesticides has led to resistance of this pest to pesticides and to destruction of its natural enemies, and consequently this pest has become a major pest (Mioannidis, 1998). So far, many studies conducted on agricultural products in order to increase productivity, has led to the increased consumption of pesticides, which in turn has caused problems, such as pest resistance and toxic residues in food products. Secondary chemical compounds of some plants along with environmental compatibility, low toxicity to non-target organisms, and low stability in the environment all play an important role in natural defense and pest control and can be a good alternative to chemical insecticides in IPM programs (Daoubi *et al*., 2005). Choosing the best insecticide and using proper concentration of each insecticide for each pest can be a great help in the integrated pest management programs to reduce their re-emergence (Cloyd & Bethke, 2010). Meanwhile, some plant extracts and essential oils, called plant pesticides, have a good chance to replace chemical pesticides in order to avoid their adverse effects.

Currently, a new approach for reducing the use of chemical pesticides is the use of natural insecticides and herbal soaps. Palizin insecticide and acaricide soap is in fact a concentrated soap of coconut oil along with mint and eucalyptus extracts having insecticidal and acaricidal properties. The insecticide is physically active and causes death of insect by disrupting respiratory system and damaging exoskeleton (Amiri-Besheli & Kabiri, 2012). Another contact insecticide is garlic and pepper extracts in a solution has a physical effect on insects causing their death by disrupting their respiratory system.

So far, many studies were conducted on these plant insecticides. For example, Danaye-Tous *et al*. (2014) evaluated these two insecticides at a concentration of 2 ml/L on eggs, nymphs and pupae of pistachio psyllids and showed that tondexir pesticide has a better efficacy in reducing number of pistachio psyllid nymphs, pupae and eggs population. In addition, studies were performed on two plant pesticides, namely insecticide soap (palizin) and red pepper extract (tondexir) with two concentrations of 3 and 2.5 per thousand, and indicated that all these compounds were able to reduce the population of aphids and pomegranate mites. No significant difference was reported between the mentioned compounds and the tested concentrations.

Furthermore, Farazmand (2012) when evaluating the effect of herbal compounds of soap and coconut oil (palizin), garlic oil extract (sirinol) and red pepper extract (tondexir) at a concentration of 2000 mg/L in Iranian pomegranate orchards showed that the use of the above compounds reduced the population of *A. Punicae* (pomegranate aphid) by 73, 60 and 55%, respectively, and reduced the population of adult stages and nymph of red pomegranate mite, *Tenuipalpus Punicae* (Pritchard and Baker) by 85, 80 and 85%, respectively. Previous studies which compared mortality rate of citrus leafminer moth *Phyllocnistis Citrella* Stainton larvae showed that 96 h after treatment with the pesticides tondexir, sirinol, and palizin at a concentration of 2000 mg/l and Spinosad at a concentration of 750 mg/L, the tondexir insecticide caused the highest mortality rate (Amiri-Besheli, 2005). In another study, Atibi *et al*. (2015) studied the effect of garlic extract against citrus red mite, two-spotted spider mite, and eastern citrus mite the mortality rate reached

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69.57, 71.85 and 100% respectively. In addition, pymetrozine insecticide, a derivative of pyridine azomethine, is one of the most widely used insecticide in Iran to control aphids and whiteflies (Li *et al*., 2001; Polston & Sherwood, 2003; Jafar Beigi *et al*., 2012). Accordingly, the effect of pymetrozine was compared with two botanical insecticides, tondexir and palizin in this study.

Recently, evaluation of the negative effect of pesticides on non-target organisms has been extensively studied (Stark and Banks, 2003; Desneux *et al*., 2007). For example, research on the natural enemy of the psyllid predator *Psyllaephagus pistaciae* has shown that coconut oil has the least negative effect compared to the common insecticides Mospilan and Consultant (Kabiri & Amiri Besheli, 2012). Botanical insecticides, such as neemazal-T/S and diatomaceous earth individually or mixed against the green peach aphid *Myzus persicae* and its effect on the predator *Coccinella* spp. have been investigated. This compound had the greatest effect on peach green aphid and was reported as safe on predator insects (El-Wakeil & Saleh, 2007). Accordingly, the present study was conducted to evaluate the effect of plant pesticides including palizin and tondexir in comparison with the chemical pesticide pymetrozine on the green aphid *A. gossypii* and also investigating the effect of adding citral oil to the plant pesticides.

Materials and Methods

This study was conducted during 2018-2019. Different stages of *A. gossypii* (green cotton aphid) were collected from soybean plants grown at the Iraqi Agricultural Research Station in Gorgan. For performing experiments on green cotton aphids in nymph and adult stages, young soybean leaves of DPX cultivar (Katoul) whose seeds were used. For bioassay tests, a stock solution (5000 ml/L) was prepared and then eight dilutions for each insecticide were prepared. The following four treatments were evaluated: (a) Citral oil (emulsifying vegetable oil containing citrus peel extract) produced by Kimia Sabzevar Company at 4000, 3000, 2000, 1500, 1000, 500, 250 and 100 µl/L concentration, (b) Palizin plant pesticide (coconut oil soap, 65% SC) produced by Kimia Sabzavar Company at concentrations of 3000, 2000, 1500, 1000, 500, 250 and 100 µl/L with and without citral oil at the same concentration, (c) Tondexir plant pesticide (insecticide containing processed garlic and red pepper extracts, 80% EC) produced by Kimia Sabzavar Company at concentrations of 3000, 2000, 1500, 1000, 500, 250,and 100 µl/L with and without citral oil at the same concentration, (d) Pymetrozine insecticide (25% wettable powder) produced by Aria Shimi Company at concentrations of 3000, 2000, 1500, 1000, 500, 250 and 100 µl/L with and without citral oil at the same concentrations. All of the above treatments were applied to both nymphs and adult insects in-vitro (in a petri dish) and in greenhouse (pot) and compared with water spray as control treatment.

Investigation of the effect of pesticides on green cotton aphid under laboratory and greenhouse conditions

Aphids were collected from the infested plants at the Hashem Abad Cotton Research Station and transferred to the Entomology Laboratory for identification. An *Aphis gossipii* colony was reared on soybean plants. Wingless adult female aphids were transferred to the uninfected soybean plants and were allowed to feed for 24 h. Then adult aphids were completely removed, and one-day-old nymphs were collected, some used in different experiments, and the rest were allowed to become mature (Elbert & Cartwright, 1997). Leaves selected from healthy soybean plants grown in pots were soaked in pesticide toxic solutions for 5 seconds, and after complete drying, the leaves were cut into 6 cm diameter disc and all the surface of the petri dish was completely covered with leaves with wet cotton surrounding it to prevent aphids from drying out and escaping (Youn *et al*., 2003). 15 aphids were then removed from the infested soybean leaves using a fine brush and used for two separate treatments (nymph and adult stages). The aphids were gently placed on the leaves inside the container and the lid was then closed and 24 h later, mortality rate (%) of adult and nymph insects was measured. These experiments were performed in 3 replicates.

For the greenhouse experiment, plastic pots (10 cm diameter and 15 cm high) with holes to permit drainage were used (Youn *et al*., 2003). Pots were filled with soil containing river sand, garden soil, and leaf soil mix in a 1: 1:1 ratio. Growing conditions were 16:8 h light:dark photoperiod, 65±5% relative humidity and a temperature of 25±1°C. After the soybean plant reached 15 cm of height, 15 green cotton aphids were released in two treatments (nymph and adult stages) on soybean plant. For preventing escape of the aphids from plants, each pot was placed in plastic cylinders covered with wire net. The desired concentrations were applied to each pot. For spraying each treatment, a 2-liter "kingjet" sprayer with a conical nozzle was used. Mortality rate of adult insects and nymphs was measured 24 h post-treatments.

Investigation of the effect of pesticides on Bracon bee natural enemy under laboratory conditions

The effect of each pesticide (with and without citral oil) was evaluated on Bracon bee as a biological control agent (Youn *et al*., 2003). First, the test tubes were completely coated with the different seven concentrations of insecticides mentioned in the previous section. Each concentration was poured into the test tube and shaken several times, and the rest of the pesticide was emptied and the tube was air-dried. Ten Bracon bees were introduced, and the tubes were closed with a net. 24 h later, the bee mortality rate was determined. Because in the control treatment, the bee mortality rate was significantly high, a mixed solution of water and honey was used to prevent mortality in the control. This experiment continued for 96 h to evaluate the pesticides residual effect on the natural enemy.

Statistical analysis

The probit analysis and LC_{50} calculation were done using POLO-PC software and the means were compared if the effect of relevant treatments was significant. Comparison between different treatments in terms of significance was done factorially using least significance difference (LSD) test at the probability level of 0.05, with a completely randomized design using SAS software.

Results

The Effect of palizin, tondexir, and pymetrozine insecticides on aphids developmental stages under laboratory conditions

Results of mean comparison regarding LC_{50} values related to nymph stage of aphids treated by different pesticides in laboratory are summarized in Table 1.

The highest effects were related to the treatments of palizin + citral oil and pymetrozine + citral oil, and both had similar significance level. Both insecticides were also applied only and the least effect was related to the use of citral oil, but among the main treatments, tondexir insecticide with and without citral oil had less efficacy than other nymph treatments under laboratory conditions. The mean LC_{50} was determined for each of the treatments and the difference between them was significant at P=0.05 using LSD test (Table 2).

Table 1. Probit analysis of different pesticides on nymph stage of cotton aphid under laboratory conditions.

Pesticides	LC_{50} (ppm) CL*	Slope(±SE)	χ^2
Citral	1149.01 (912.12-1529.89)	1.61±0.32	0.690
Palizin	416.09 (175.46-487.29)	1.69±0.40	1.058
Pimetrozin	518.67 (292.66-844.93)	1.79±0.45	1.177
Tonexir	733.43 (458.78-1052.73	1.46±0.34	1.284
Pimetrozin+Citral	448.56 (276.89-617.61)	2.62±0.66	0.138
Palizin+Citral	356.66 (190.32-589.14)	1.48±0.31	1.811
Tondexir+citral	vbbl615.70 (376.72-990.31)	1.49±0.31	1.131

*Confidence interval

Table 2. Probit analysis of different pesticides on the nymph stage of cotton aphid under semi-field conditions.

Pesticides	LC_{50} (ppm) CL*	Slope(±SE)	χ^2
Citral	946.85 622.01-1084.01	1.816±0.419	1.318
Palizin	596.01 333.57-986.84	1.216±0.475	0.518
Pimetrozin	376.90 230.50-571.75	1.7910±0.409	1.417
Tonexir	690.20 363.85-99.31	1.269±0.311	1.285
Pimetrozin+Citral	295.42 160.97455.87	1.683±0.402	0.960
Palizin+Citral	468.83 183.96-610.97	1.451±0.332	1.742
Tondexir+citral	576.65 468.68-1037.94	1.299±0.390	1.443

*Confidence interval

The results showed that the most effective treatment in the greenhouse pot experiment was citral oil-free pymetrozine insecticide, which was able to significantly reduce the number of nymphs on the pots and Palizin with citral oil was ranked second in terms of efficacy, followed by palizin and tondexir insecticides with citral oil, and the least effective was the tondexir insecticide. Plant insecticides, such as palizin and tondexir, which have entered the market in recent years can be effective under both laboratory and greenhouse conditions. They showed good control of the cotton green aphid and when mixed with citral oil their efficacy was almost equal to that of the commonly used insecticide, pymetrozine (Tables 1 and 2).

The mean LC_{50} was determined for each of the treatments and differences was evaluated for each of the insecticides using the LSD test at P=0.05 (Table 3). Results obtained showed that the addition of citral oil to all treatments reduced the concentration of pesticides to control the adult stage of cotton green aphid and thus, doubled the effectiveness of the plant insecticides tondexir and palizin when mixed with oil.

Table 3. Probit analysis of different pesticides on adult stage of cotton aphid under laboratory conditions.

Pesticides	LC_{50}(ppm) CL*	Slope(±SE)	χ^2
Citral	61.15 12.19-65.1	1.707±0.30	1.052
Palizin	62.938 501.03-930.92	1.509±0.31	1.681
Pimetrozin	1363.17 809.99-1732.96	1.265±0.38	0.159
Tonexir	1851.99 1156.15-2347.05	1.319±0.32	0.401
Pimetrozin+Citral	1143.33 691.69-1473.33	1.819±0.17	0.313
Palizin+Citral	807.88 624.82-1063.54	1.370±0.30	1.484
Tondexir+citral	1291.87 887.04-1402.02	1.799±0.12	0.611

*Confidence interval

The least effect was obtained by the tondexir insecticide, citral oil alone, and palizin insecticide, respectively, which were not significantly different from each other (Table 4). Interestingly, the adult cotton green aphid under greenhouse conditions showed resistance to the concentrations used in this experiment.

Considering all the data presented above it can be concluded that the addition of citral oil to all pesticide treatments had increased their efficacy which led to severe reduction in the aphids population.

Statistical analysis (ANOVA and LSD) of the effect of different pesticides at different concentrations on mortality rate of cotton green aphid nymphs under semi-field conditions showed significant differences between the

different treatments. Likewise, the effect of different pesticides at different concentrations on cotton green aphid adults under laboratory conditions showed significant differences between different treatments. Similarly, the effect of different pesticides at different concentrations on adults of cotton green aphid adults under semi-field conditions showed significant differences between different treatments.

Table 4. The Probit analysis of different pesticides on adult stage of cotton green aphid under semi-field conditions.

Pesticides	LC_{50} (ppm) CL*	Slope(±SE)	χ^2
Citral	1948.14 1452.36-2053.59	2.311±0.53	1.021
Palizin	1789.13 1027.64-2289.33	1.170±0.31	0.864
Pimetrozin	1557.99 954.12-2238.90	1.513±0.29	1.452
Tonexir	2157.98 1342.91-2831.33	1.101±0.38	1.620
Pimetrozin+Citral	1389.59 967.34-1900.69	2.086±0.47	0.425
Palizin+Citral	1249.14 736.88-1331.03	1.505±0.37	1.038
Tondexir+citral	1260.41 747.75-1659.11	1.585±0.44	0.019

*Confidence interval

Discussion

Due to the fact that *Aphis gossypii* is one of the most important polyphagous pests of most vegetables grown in house gardens and greenhouses in Iran, and consumed as fresh products. In addition, the natural enemies of this pest should be protected by using low-risk insecticides to control these pests including botanical insecticides. Based on the results obtained in this study, all stages of the green cotton aphid, except for the nymph stage under greenhouse conditions, the most desirable treatment was a mixture of palizin insecticide with citral oil. Previous studies showed monoterpene compounds, present in the pesticides used in this study, are suitable alternatives to chemical insecticides and their insecticidal properties have the least risk to human's health and the environment (Sokovich *et al*., 2009). Furthermore, different effects of these compounds on insects have been studied and confirmed by various researchers (Li *et al*., 2001; Stampolous *et al*., 2007; Santon *et al*., 1997). Tondexir plant insecticide is actually a hot pepper extract derived from compounds of hot pepper species. Previous work showed that this compound plays an important role as insect-repellent with insecticidal properties (Gudeva, 2012). It has been shown that A. gossypii population on cucumber was reduced by 75.9 and 90.6%, respectively (Bani Ameri, 2008). Heidari *et al*., (2016) tested the effect of citral oil (5ml/l) and tondexir insecticide (2 and 3 ml/l) along with 1 ml/l of soap on the Australian bug *(Icerya purchasi* Maskell) and showed that the highest mortality rate was obtained by the tondexir treatment, and 1.5 ml/l of palizin insecticide had the least effect on this pest, which is not in agreement consistent with the results obtained in this study. This difference may be due to the type of pest and its feeding habit. In a previous study, evaluating the effect of three botanical insecticides including tondexir, palizin, and sirinol on citrus mealybug in the laboratory showed that tondexir and palizin pesticides at a concentration of 3000 ppm produced a higher mortality rate than the sirinol pesticide with a concentration of 3500 ppm (Ahmadi *et al*., 2012), which is in agreement with the results of this study. In a similar study conducted by Amiri Beheshti (2009) on the effect of plant pesticides and mineral oil on larvae of the citrus leafminer moth of *Phyllocnistis Citrella* Stainton, it was found that the plant pesticides palizin and sirinol caused a higher mortality rate compared to BT and mineral oil. However, the results of this study showed that tondexir insecticide could not control nymphs and adults of cotton green aphids as much as butterfly larvae, which could be due to the difference in biology of the two pests. Another insecticide evaluated in this study was pymetrozine insecticide. This compound has a chemical structure and a special effect and is used more against aphids and whiteflies (Talebi-Jahromi, 2007). Pymetrozine prevents sucking insects from entering the plants vascular system, resulting in cessation of feeding and death of insects (Ausborn *et al*., 2005), but as an insecticide, however, it has side effects on the environment and natural enemies. Previous experiments confirmed that pymetrozine had a good effect on the weevil aphid, but its comparison with palizin as a new plant insecticide showed that in most cases, effectiveness of palizin insecticide was significantly higher.

It can be concluded from this study that although the chemical insecticide pymetrozine still has a good ability to control the cotton green aphid, but the plant insecticide palizin can be used as a safer insecticide with better performance for the control of this pest, and it is recommended to replace conventional pesticides, as this insecticide has lower LC_{50} than other treatments. It is also important to note that in all treatments, when plant and chemical insecticides were used in combination with citral oil, their performance was significantly improved.

الملخص

مقدام، م، ب. أميري–بيشيلي و م. شريف. 2022. تأثير المبيدات النباتية Palizin، Tondexir وPymetrozine على منّ القطن الأخضر *Aphis gossypii*. مجلة وقاية النبات العربية، 40(4): 340–345. https://doi.org/10.22268/AJPP-40.4.340345

هدفت هذه الدراسة لتقييم فعالية بعض المبيدات الحشرية النباتية (مستخلصات نباتية) إزاء حشرة المنّ الأخضر *A. gossypii*، ومدى زيادة فعاليتها بتدعيمها بزيت الليمون، حُدِّدَت المقاييس (القراءات) الحيوية لفعالية المبيدات على الحوريات والحشرات الكاملة المُكاثرة على أوراق فتيّة لنبات فول الصويا (الصنف DPX Katoul) سواءً مختبرياً أو في الدفيئة، استخدم في هذه الدراسة المحلول الأساسي لكلّ مبيد نباتي وسلسلة من تخفيفاته (ثمانية تخفيفات). أظهرت النتائج المختبرية وجود تأثير فعَّال للمبيد palizin المدعَّم بزيت الليمون على حوريات حشرة المنّ الأخضر، إذ بلغ التركيز المميت النصفي (LC_{50}) 359.669 مل/ل، في حين كانت النتائج مغايرة عندما نفذت التجربة على حُوريات الحشرة ذات العمر الحُورِي نفسه في الدفيئة الزجاجية، إذ تفوق مزيج المبيد pymetrozine مع زيت الليمون، إذ بلغت قيمة التركيز المميت النصفي 295.242 مل/لتر. أمّا في ظروف الدفيئة فقد أثبت مزيج المبيد palizin وزيت الليمون أعلى فعالية على الحشرات الكاملة سواءً في ظروف المختبر أو الدفيئة الزجاجية، حيث بلغت قيمة التركيز المميت النصفي 807.88 و 1249.14 مل/لتر تحت ظروف المختبر والدفيئة الزجاجية، على التوالي.

كلمات مفتاحية: Palizin، pymetrozine، tondexir، زيت الليمون، منّ القطن الأخضر.

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