ISSN: 2320-5407 Int. J. Adv. Res. 10(09), 848-852

Journal Homepage: -www.journalijar.com

INTERNATIONAL JOURNAL OF ADVANCED RESEARCH (IJAR)

ISSN NO. 2320-5407

Article DOI:10.21474/IJAR01/15430
DOI URL: http://dx.doi.org/10.21474/IJAR01/15430

RESEARCH ARTICLE

BIOLOGICAL EFFICACY OF THE NEEM OILFOR THE CONTROL OF APHIDAE POPULATIONS

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Manuscript Info

Manuscript History
Received: 25 July 2022
Final Accepted: 28 August 2022
Published: September 2022

Key words:
Biological Efficacy, Bioactive Substances, Insecticidal Effect, Neem Oil Extract, Azadirachtaindica A. Juss, Aphidae, Greenhouse

Abstract

In this study which took place during the 2021 year we tested the biological efficacy of neem (*Azadirachtaindica*A. Juss) seed oil on aphid populations. The researchwas performed under laboratory and greenhouse conditions using cucumber (*Cucumissativus L*) plants. The laboratory experiments showed a strong aphicide effects, the pests dying before they could leave the treated cucumber leaves. The greenhouse studies confirmed the potent insecticide action, however they also showed a moderate repellent effect. The extract inhibited aphid feeding for a period of 5-7 days but was not able to completely inhibit the consumption of plants. The neem oil in a dose of 10 L/ha has the potential to be used as an aphicide, the highest biological efficacy being reached after two treatments with an interval of 10 days between them.

Introduction:-

The role of insecticides in human society is very important. Phytophagous insects can cause losses of 10 to 90% of cultivated crops [1,2]. In addition to affecting crop growth, harmful organisms can subsequently damage stored crops [3]. However, using of synthetic pesticides has a direct negative effect on human and animal health. Also the excessive or inappropriate use of synthetic insecticides causes biodiversity loss and occurrence of pests' resistance to these substances [4,5]. On the other hand, humanity is exposed to the risk of these chemicals by consuming pesticide-contaminated agricultural products, resulting in long-term health risks.

The use of secondary metabolites synthesized by some plant species as part of their natural self-defense against pathogens and pests seems to be an excellent alternative to synthetic insecticides [6,7]. Due to good biodegradability and low toxicity to non-target organisms, the economical availability of the herbal pesticides is increasing day by day. Many experts predict a huge increase of biopesticides sales over the next decade. They have the potential to grow from 4-5% of the global pesticide market to 20% by 2025. According to its quota in the global market the oil production from *Azadirachtaindica*Juss. (Meleaceae) seeds will probably be on the first place. The biologically active compounds of neem seed oil such as azadirachtin (azadirachtin A), salannin, salannol, nimbin, nimbidiol, 1-tigloyl-3-acetyl-11. hydroxymeliacarpine (azadirachtin D) belong to the triterpenoid limonoid class. Some companies have approached the standardization of their pesticide products by adding azadirachtin A, which is considered one of the most effective insecticide compound. For example, the NeemAzal T/S product of the German company Trifolio-M, with a declared content of 10,000 ppm azadirachtin A, is the best-selling in Europe. Commercial bioinsecticides based on emulsified neem oil can vary significantly in their efficacy, as the azadirachtin content of the oil may vary from 0.01 to 0.9%, depending on the ecotype and/or seed extraction conditions. In our study which took place during the 2021 year within Institute of Genetics, Physiology and Plant from the Republic of

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Moldova we tested the biological efficacy of neem seed oil on Aphidae populations in laboratory and greenhouse conditions.

Materials And Methods:

In the simplest cases (in laboratory conditions or in the field, when the number of individuals between counts does not change), the biological efficacy of an insecticide, acaricide or rodenticide is calculated using the Abbott formula:

$$C = \frac{100(A - B)}{A}$$

Where,
C - Is the percentage of mortality of individuals;
A - The average number of pests before treatment;
B - The average number of pests after treatment;

Abbott's formula is used if pests have a hidden lifestyle and their presence can only be taken into account by the number of damaged plants or their parts (roots, tubers, buds, flowers, etc.). In this case, A is taken as the number of damaged plants (plant parts) in the control, and B is the number of damaged plants (plant parts) in the experimental version. The same applies to determining the biological efficacy of rodenticides: in this case, A is the number of burrows before treatment, and B is the number of burrows that opened after treatment. In cases where it is possible to fix the number of dead individuals, for example, individuals of the Colorado potato beetle on a plantation, aphids and ticks in a laboratory experiment, the biological efficacy is determined by comparison with the control according to the formula:

$$C = \frac{100(Ba - Ab)}{Aa}$$

Where,
C - Is the percentage of pest mortality adjusted for control;
A and a - respectively, the total number of individuals in the experimental variant and control;
B and b - respectively, the number of dead individuals in the experimental variant and control;

In order to determine the biological efficacy ofneem seed oil in laboratory conditions the cucumber (*Cucumissativus* L) seedlings were planted in a greenhouse and infected with Aphididaefamily populations transferred from a natural agrocenosis. Samples were collected from cucumber leaves in the greenhouse. The leaf blades inhabited by pests were placed in Petri dishes in 4 replicas for each variant and counting of dead individuals was performed next day after treating the samples with extracts working solutions. The plant extracts were prepared in different concentrations (Table 1). A 1% ecological Pelecolinsecticide solution was used as a standard. Mathematical processing and evaluation of reliability of the obtained scientific data were performed using the ABC Pascal platform [8,9].

Regarding the greenhouse experiments (Fig.2) there were used Merengue F1 hybrid cucumber seeds, sown in the end of April while the aphids inhabited the plants naturally. The cucumbers were treated with four variants (each variant in three repetitions): experimental variants - neem oil in two doses (0.8 Litre per Hectare (L/ha) and 1.0 L/ha), standard (Pelecol EO – 10 L/ha) and control. The parcels were placed according to the randomized block method. During the vegetation period, 2 treatments were performed. The treatment was performed according to the biological characteristics, the numerical density and the spread degree of the pests. The biological efficacy was determined from data obtained from pre-treatment period and post-treatment on day 3, day 5 and day 7. The calculation was done according to the formula of Henderson & Tilton [10].

Results And Discussions:-

The maximum biological efficacy in laboratory conditions was observed when the leaves were treated with 0.5% *Azadirachtaindica* A. Juss extract (Table 1, V2 (0,5%N)). The pests died before they could leave the treated cucumber leaves (Fig.1).

Table 1:- Biological efficacy of neem oil extract in the control of melon aphid(*Aphis gossypii*Glov.) on *Cucumissativus* L (laboratory conditions).

Variant	The number of livingaphids,%	The number of dead aphids,%	Biological efficacy, %
Control	96,6	22,8	-
Standard	70	52,5	**52,2**
V1 (0,1%N)	90	71	**56,1**
V2 (0,5%N)	86,9	80,7	**70,1**

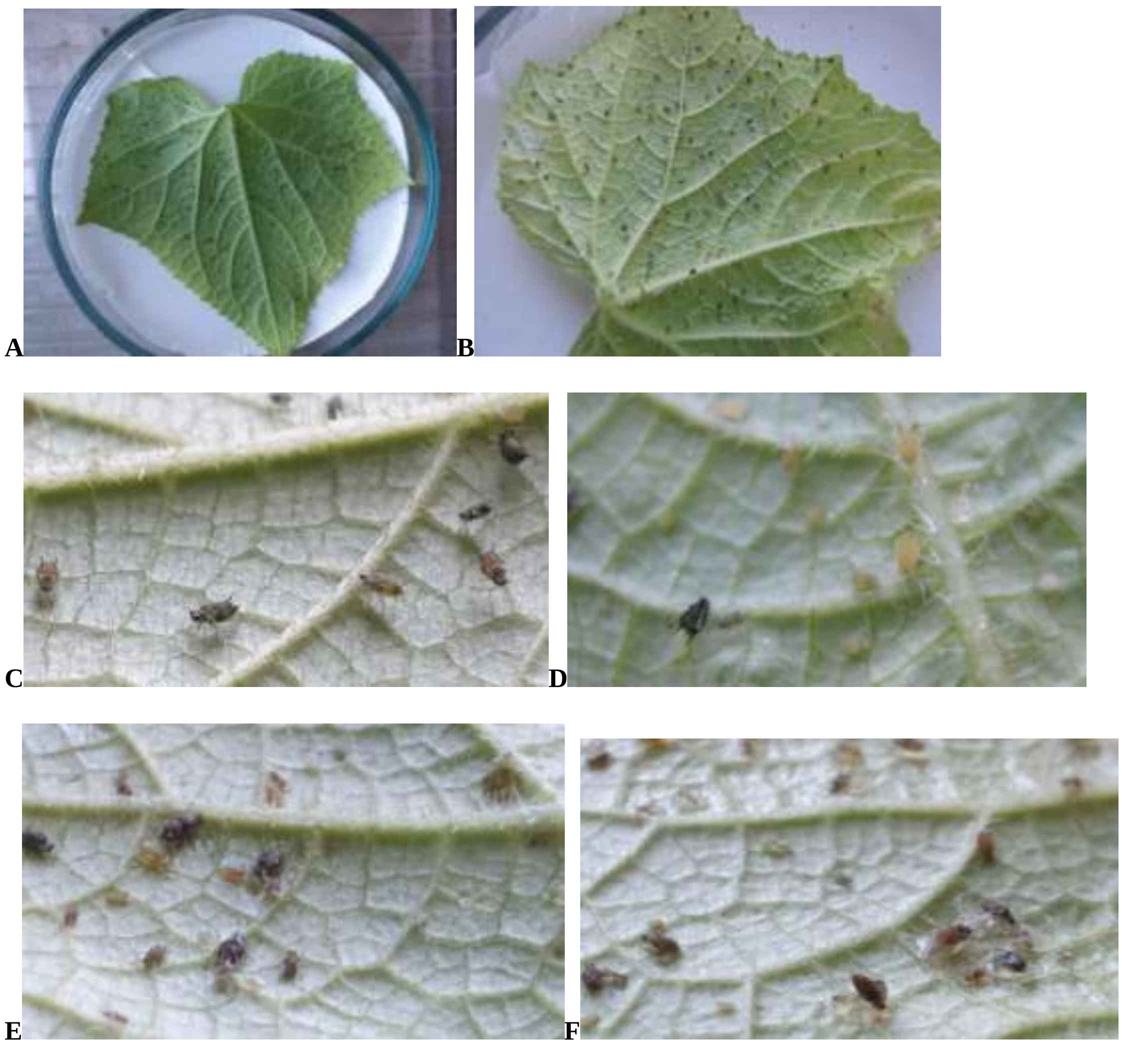

Fig 1:- *Aphis gossypii*Glov. on the leaves of *Cucumissativus* L
(A, B - general view of the leaf affected by the pest;
C, D - living aphids before treatment;
E, F – dead aphids after treatment with neem extract)

Regarding the greenhouse experiments the biological efficacy of the neem oil after first treatment (Table 2) was registered in the high dose (10.0 L/ha) with 81.84% in the 3rd day after treatment, while the 8.0 L/ha dose showed a result of 75.41 %. The standard (Pelecol - 10.0 L/ha) efficacy was 89.47%. On the 5th day of evidence it was observed that the biological efficacy of studied products increased compared with the standard, having a value of 83.78% for 10.0 L/ha dose and 76.99% for 8.0 L/ ha. On the 7th day after treatment it was observed a slightly lower efficacy compared to the 3rd day.

Table 2:- Biological efficacy of the neem oil extract in the controlof melon aphid (*Aphis gossypii*Glov.) on Merengue F1 hybrid cucumber(first treatment in greenhouse conditions, date: 24.06.2021).

Variant	Repetition	The average numerical density of aphids on a leaf				Biological efficacy, %		
		Before treatment	In the days of evidence					
			3 days	5days	7days	3 days	5days	7days
Neem oil - 8,0 L/ha	avg.	39.00	17.75	21.50	29.75	**75.41**	**76.99**	**74.12**
Neem oil - 10,0 L/ha	avg.	45.00	15.00	17.50	23.75	**81.84**	**83.78**	**82.21**
Standard (Pelecol-10,0 L/ha)	avg.	46.00	8.84	13.17	16.50	**89.47**	**88.06**	**87.86**
Control	avg.	46.00	77.00	100.00	124.00	-	-	-
DEM_{05}		**3.54**	**3.15**	**4.6**	**5.06**	-	-	-

The second and last treatment was performed 10 days after the first treatment (Table 3). After this last treatment on the 5th day of evidence, the 10.0 L/ha neem experimental variant recorded highest biological efficiency - 90.05%. This was slightly higher than Pelecol result (89.09%). On the 7th day after treatment, the biological efficacy was slightly reduced but still present, as Azadirachtin - the active substance in the neem oil, inhibits the feeding of aphids and acts as a repellent.

Table 3:- Biological efficacy of the neem oil extract in the controlof melon aphid (*Aphis gossypii*Glov.) on Merengue F1 hybrid cucumber(second treatment in greenhouse conditions, date: 05.07.2021).

Variant	Repetition	The average numerical density of aphids on a leaf				Biological efficacy, %		
		Before treatment	In the days of evidence					
			3 days	5days	7days	3 days	5days	7days
Neem oil - 8,0 L/ha	avg.	30.00	7.25	7.70	9.50	**81.36**	**82.08**	**80.83**
Neem oil - 10,0 L/ha	avg.	24.00	3.67	3.50	4.25	**88.04**	**90.05**	**89.71**
Standard (Pelecol-10,0 L/ha)	avg.	17.00	2.50	2.75	3.75	**88.41**	**89.09**	**86.40**
Control	avg.	127.00	163.00	190.00	211.00	-	-	-
DEM_{05}		**4.47**	**2.76**	**6.7**	**3.34**	-	-	-

A  **B** 

Fig.2:- Greenhouse cucumber plants (A - cucumber seedlings; B – the row with plants treated with neem extract (left) and plants from the control row (right)).

The laboratory experiments on the neem extract showed strong aphicide effects, the pests dying before they could leave the treated cucumber leaves. The greenhouse studies confirmed the potent insecticide action, however they also showed a moderate repellent effect. The extract inhibited aphid feeding for a period of 5-7 days but was not able to completely inhibit the consumption of plants. The neem oil in a dose of 10 L/ha has the potential to be used as an aphicide, the highest biological efficacy being reached after two treatments with an interval of 10 days between them.

The research was performed within the institutional project no. 20.80009.5107.19: "Strengthening the capacities for forecasting and combating harmful organisms and phytosanitary risk analysis in integrated plant protection".

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