

#### Investigation of Garlic Essential Oils for Biological Control of Agrotis Ipsilon (Hufnagel) along with Combination with Mint, Cumin, Caraway and Parsley Essential Oils

Kumar Anurag (M.Sc. Zoology) Mahendra Mahila Mahavidyalaya, Gopalganj, J.P. University, Chapra

#### Abstract:

Agrotis ipsilon (Hufnagel) also known as the black cutworm, is one of the most harmful insect pests that attack various legume crops. On the insect's third larval instar, the effects of various quantities of Garlic essential oils were assessed. Biological activity of essential oil of Garlic caused starvation and antifeedant effects in larvae fed on the treatment diet, as well as stomach and contact toxicity. Garlic oil, whose LC<sub>50</sub> ranged from 0.006 to 0.019% and was dangerous as a contact poison than a stomach poison, was the most effective treatment as plant essential oil treatment. The mixture of the oils had a potentiating impact on one another, making them more toxic. Garlic and mint made up the promising poisonous concoction. The tested oils' sublethal concentrations produced an impressively notable increase in larval and pupal duration with a delay in their development and an increase in mortality, a decrease in larval and pupal weight, and an increase in the percent of malformation for both the resulting pupae and adults. From this point of view, it is possible to suggest using the garlic plant essential oils to control the black cutworm on their hosts by spraying them as emulsions or as toxic baits alternatives to chemical pesticides. It is also possible to mix oils in the concentration range to increase their toxicity and reduce the development of insect resistance as a biological control method throughout the integrated pest control programme.

**Key Words:** Agrotis ipsilon, Essential oils, Garlic (Illium sativum), Parsley (Petroselinum crispum), Carawaya (Carum carvi), Cumin (Cuminum cyminum), Mint (Mentha piperita), Black cutworm

#### **Introduction:**

One of the most destructive insect pests, the black cutworm, *A. ipsilon*, attacks a variety of agricultural crops throughout the year, including cotton, soybeans, corn, potatoes, and tomatoes, not only in Bihar but also in many other states of our country. Due to *A. ipsilon* infestation, especially at the seedling stage, there were significant yield reductions (Ladhari et.al, 2013). The present strategy for controlling black cutworms relies heavily on pesticides, which harm the environment and/or pose a risk to human health through their residues in food,



groundwater, or accidental exposure. The need for efficient, biodegradable insecticides with high selectivity has increased due to the issues produced by pesticides and their residues (Hazaa & Alam EL-Din, 2011). New pesticides have been researched, and conventional pest control methods have been reassessed and used as alternatives. The need for efficient and biodegradable pesticides has increased due to the negative side effects of synthetic pesticides. Because of the strength of the interactions between plants and insects, plants have welldeveloped defences against herbivores and are great suppliers of new poisonous compounds for pests (Pickett et.al., 2006). Essential oils from aromatic plants are one type of natural compounds that have been presented as natural biopesticides (Isman & Grieneisen, 2014; Prakash et al., 2014). Numerous studies have been conducted on the pesticidal properties of essential oils from the Lamiaceae family (Rajendran & Sriranjini,2008; Isman et al., 2011; Ebadollahi & Jalali, 2011). The benefit of employing plant essential oils is that they are widely used for medical purposes and are accessible, indicating that they are low or non-toxic to people (Upadhyay 2013). Insects may be adversely affected by plant products in a number of ways, including toxicity, mortality, antifeedant growth inhibitory effects, suppression of reproductive behaviour and reduction of fecundity and fertility, growth inhibition, and perturbation of reproductive behaviour (reduction of fecundity and fertility) (Lambrano et al., 2014). The main objective of the current study was to assess the toxic effects of a garlic essential oils and in combination of essential oils from Mint, Cumin, Caraway and Parsley on various stages of the black cutworm A. ipsilon under controlled laboratory conditions to determine whether they could be used as a safe biological alternative to chemical pesticides as part of an integrated pest management strategy.

#### **Materials and Methods**

The insects used in this study were grown in a conventional lab culture and fed an artificial meal at a constant temperature of 23.5°C and 65.5% relative humidity. Under a 14-second light and 10-second dark exposure (EL-Kady et al.,1990). These oils were procured from laboratory supply businesses in Patna, and the hazardous and biological effects of garlic essential oils alone and in combination with mint, cumin, caraway, and parsley essential oils were assessed. By diluting with water, different quantities of each oil were created, and 0.5% of Triton X was added as an emulsifier. The created emulations were applied to different stages of the black cutworm (eggs, third larval instars, and pupae) one at a time. Eggs that were one day old were



immersed in the oil emulsion for 10 seconds before being let to hatch. The treatments' percentages of successful egg hatching were calculated after 2-4 days. The third larval instars were selected from the breeding culture and treated with the tested oils using two different techniques: Stomach effect of the oil emulsion on the 3rd larval nstar and Contact effect of the oil emulsion on the 3rd larval nstar.

**Study of the Stomach effect of the oil emulsion on the 3rd larval nstar:** To prevent cannibalism, the artificial diet is prepared by mixing a certain amount of oil with it (ml. oil/ml. food), and the prepared artificial diet is then fed to the larvae for three days in separate jars. The larvae that were still alive were moved to a normal, untreated artificial diet, where they continued to feed. After 7 days of treatment, the percentage of larval mortality was assessed. We used 30 duplicates for each concentration.

**Study of the Contact effect of the oil emulsion on the 3rd larval nstar:** Larvae were sprayed with an oil emulsion using a tiny hand sprayer, and after being allowed to feed for seven days on an untreated diet, the percentage of death was calculated. In the two methods before, the oils were swapped out for water in the control treatment. Abbott's formula (1925) was used to obtain the corrected mortality rate, and the LC<sub>50</sub> concentration was determined using the Sigma plot (2002). oil toxicity at the pupal stage. Pupae that were one day old were employed, and they were submerged in the oil emulsion for 10 seconds (much like with the treatment of eggs). Till the moths emerge, the treated pupae were left. Based on the number of adults who had emerged, the percent of pupal mortality was estimated. Pupae received a control treatment of immersion in water with Triton alone.

**Combined effect of different oils:** Third larval instars were obtained from the standard laboratory culture that was raised on artificial diet. LC<sub>50</sub> of each of the two oils (that had been previously estimated) were mixed in one diet, and the larvae were then left to feed on it for one day before being moved to a diet that had not been treated for the following seven days. At the end of the seven days, the percentage of larval mortality was calculated. At the same time, the percentage of mortality of the larvae on each oil mixed with feed separately was calculated (1965). Five replications were employed in each experiment, and Dancan (1955) was used for each test.

**Statistical analysis:-** Using Abbott's formula, the average percent mortality of the examined larvae was computed and corrected (Abbott,1925). The Finney approach was used to statistically calculate the corrected percentage of fatalities (1971). To obtain the associated



Log-concentration probit lines, the computed percentage of death was plotted against the respective concentrations on logarithmic probability paper. The 50% lethal doses for proven regression lines were established. The variance ratios were computed after statistical analysis of all the data collected for (biological studies). The 5% level was calculated using the ANOVA method using the computer programme (SPSS).

#### **Results& Discussion**

The data indicated that the mortality present was caused by Garlic oil which has the lowest LC<sub>50</sub> for all stages, it was 0.006%. on the eggs, 0.030 and 0.019% as a stomach and contact poison respectively, on the pupal stage it was 0.009%.

Plant oil	LC <sub>50</sub> for different stages(mL oil/100ml.water)				
	Egg	Larva		Dung	
		Contact	Stomach	rupa	
Garlic	0.006	0.019	0.030	0.009	

\*Table (1) Toxicity of the plant essential oils on different developmental stages of the black cut worm *A. ipsilon*.

It is clear that the eggs and pupal stages were more susceptible to oil treatment than the moveable phases, with LC<sub>50</sub> values of 0.006 and 0.009%, respectively. This was contrary to the belief that the dormant stages were more resistant to chemical insecticides. The findings demonstrated that the effectiveness of the tested oils as contact poisons was associated to the degree of oil solubility in water and lipids as well as the degree of their permeability through the integument of larvae and pupae or egg chorion. The degree to which the tested oils were acceptable to larvae consuming the oil-treated meal determined how they would react in their stomachs. The degree of their permeability through the integument and their impact on the insect haemolymph, viscera, nervous system, and neuroendocrine system may be the cause of the variable toxicity of oil in the stomach or upon contact (Fallatah, 2003). The findings were in line with those made by Odeyemi (1991), who discovered that the physical and chemical characteristics of each oil were associated to the decline in offspring of adult insects treated with those oils. He said that the oils have an impact on an insect's respiratory system by preventing breathing and causing choking. Mentha piperita, caraway, and parsley oils were found to lower mating times and fertility in female S. littoralis moths, according to Klingauf et al research's from 1982. According to Purohit et al. (1983), the control of Musca domestica, Aedes aegyptii, and Dysderus cingulatus employed Cuminum cyminum oil. According to



Chander and Ahmed (1986), after the seeds were treated with Carum carvi oil, the percentage of eggs laid by Callosobrocus chinenses on cowpea seeds dramatically decreased. According to Mahgoub et al. (1998), parsley oil reduced the ability of C. maculates to reproduce; at a 95% concentration, the adults were stopped from laying eggs on the treated wheat seeds. According to Varma & Dubey (2001), Tribolium castanium and Sitophelus oryzaea larvae are both poisoned by mint *M. arensis*. The essential oils such as garlic (*Allium sativum*), mint (*Mintha* pipereta), and eucalyptus (Eucalyptus globulus) are renowned for their pest management qualities for controlling the grasshopper Heteracris littoralis, and our results support this finding published by Sharaby et al. (2012). According to Gurusubramanian and Krishna (1996), when Earias vitalla (Fabricius) and Dysderus koenigii (F.) eggs were exposed to the vapour of Allium sativum, the eggs' hatchability was severely reduced. They explained this by pointing to several chemical components in A. sativum (bulbs) volatiles that likely diffused into eggs and impacted key physiological and biochemical processes involved in embryonic development. These eggs' embryonic development was not quite complete, and they did not progress from crystal-clear transparency to a dark colour like the control eggs did. It was also effective as a fumigant. The main components of essential oils are lipophilic chemicals that function as poisons, feeding deterrents, and oviposition deterrents to a range of insect pests (Koul et al, 2008). Castor, mustard, groundnut, sesame, coconut, and sunflower vegetable oils were investigated by Bhargava and Meena (2002) against the cowpea pathogen Callasobruchus chinensis. Adults were much more likely to die from all oils, and the mortality rate rose with the concentration level. Pavela (2005) found that 34 essential oils were poisonous or extremely hazardous after testing them against Spodopetra littoralis larvae for insecticidal action. Similar findings were made by Tripathy & Singh (2005) in India, who noted that various vegetable oils caused Helicoverpa armigera larval death (mustard oil, sesame oil, linseed oil, castor oil, cottonseed oil and groundnut oil). Neem oil application at a dosage of 5 ML/L was effective, according to Eziah et al. (2011), in halting the development of Ephestia cautell larvae; however, mortality after 96 hours of exposure ranged from 32.5 to 55%. The observed mortality can be attributed to neem's inherent properties; mortality was dose and time dependent. According to Sharaby et al. (2012), Heteracris littoralis nymphal phases, life cycle, adults' longevity, and life span all increased statistically when compared to the control test. The female offspring of the treated first instar nymphs were prevented from laying eggs by the presence of garlic oil. Significant deformity and a high loss in egg fertility brought on by eucalyptus or

mint oil were noted. According to Sharaby et al. (2014), exposure to the vapours of *Cymbopogon citratus, Myristica fragrans* (nutmag), *Mentha citrata*, and *a-Ionone* (monoterpene) significantly decreased the lifespan of *Phthorimea operculilla* moths as well as the lifespan of newly emerged adult progeny. Other studied oils including *Pelargonium graveolens, Myristica fragrans* (Mace), and *Cinnamonium zeylanicum* had a negligible impact. Except for the oils of *C. citratus, M. fragrans* (nutmag), and *M. tragrans* (mace oil), which significantly reduced egg hatchability, none of the examined vapours had a significant impact on egg hatchability. The most effective oil vapours were sorted in ascending order as follows: *Myristica* (nutmag) *Cymbopogon Mentha* an Ionone, based on the values of damage indices. Additionally, they discovered that applying bulb powder of *Allium cepa* (50 percent cone. combined with talcum powder) on potato tubers significantly reduced both the number of deposited eggs and the adult emergence from those eggs. Oils from *Allium cepa, Pelargonium graveolens*, and *Cymbopogon citratus* significantly decreased the amount of larvae that penetrated tubers that had been treated.

Essential oil	% Antifeedant	% Starvation	LC <sub>50</sub> as stomach poison (ml/100ml.)
Garlic	- 18267	231.74	0.030

# \*Table (2): Antifeedant effect and percentage starvation of the essential oils on 3rd larval instars' of the black cu worm *A.ipsilon*.

According to Table 2, the tested garlic oils acted as feeding deterrents or antifeedants against A. ipsilon larvae, causing starvation (231.17%) and the highest antifeeding activity (-18.267%). Numerous plant extracts and botanical oils have been shown to inhibit insect feeding. For instance, Rajasekaran and Kumaraswami (1985) discovered that when sesame oil was added to an aqueous extract of neem seed kernel, the antifeedant activity against S. littoralis larvae was increased; Khadr and others 1986) demonstrated that the larvae of S. littoralis were significantly inhibited from feeding by the leaf extract of Melia azadrach; According to Sabbour and Abd El-Aziz (2002), eucalyptus oil was able to deter A. ipsilon and S. littoralis from feeding during their third larval instar. Shershby and co. 2004) demonstrated that cinquef oil improved its ability to repel A. ipsilon larvae; According to Erturk (2006), different plant extracts from Aesculus hippocastanum, Viscum album, Sambucus nigra, Buxus sempervirens, Diospyros kaki, Artemisia absinthum, Alnus glutiosa Goertn, Origanum vulgare, Hypericum androsaemum, and Ocimum basilicum stopped Thaumetopoae solitaria larvae The biological effects of essential oils isolated from Cymbopogon nardus, C. flexuosus, and C. martini on two

Lepidoptera larvae were recently examined by Labrano et al. (2014). All of the examined oils demonstrated antifeedant acticity and dermal contact lethality against Acharia fusca and Euprosterna elaeasa (Lepidoptera: Emacodidae) in varying amounts. All oils had strong toxicity and antifeeding effects on insects.

Essential oil	% Mortality	Co-Toxicity factor	Type of interaction
Garlic	32		
Garlic +Mint	86	65.38	Potentiating
Garlic + Cumin	72	38.46	Potentiating
Garlic + Carawaya	80	63.26	Potentiating
Garlic + Parsley	76	68.89	Potentiating

Table (3): Effect of essential oil combination on their type of toxicity against the 3rd larval instar of the black cut worm A.ipsilon

The combination of the tested oils had an impressive impact on raising the percentages of larval mortality Table (3). When compared to the mortality produced by each oil independently (23% for garlic and 20% for mint), all combinations of the oils had a potentiating impact. However, the combination that increased mortality percentage the most was (Garlic+Mint), coursing death percentage reaching (86%) in that case. It is evident from Table 3's combinations that all combinations resulted in higher death rates than they would have had they compared each oil alone. Combining the oils improved their effectiveness and toxicity against the larvae. According to Jember & Hassanali (2002), the importance of oil combinations or their terpen components in boosting their toxicity for controlling stored grain pests is due to the varying contents of each oil and the variable side effects such constituents have on insects. This lends credence to the theory that the contents of the oils determine how poisonous they are. Vegetable oils from cotton, linseed, safflower, pundi, honge, and sesame were tested as synergists of synthetic pyrethroids (fenvalerate, deltamethrin, and cypermethrin) against *Plutella xylostella* in India by Vastrad et al. in 2002. The larval mortality with Honge and Sesame oils in combination with synthetic pyrethroids rose with concentration, and they also reported the maximum larval mortality with sesame oils. According to Sharaby et al. (2014), talcum powder mixed with pelargonium or allium provided effective protection against Phtorimaea operculella infection for the potato tubers during a lengthy storage period (30-40 days). The synergistic effects of complex mixtures, such as essential oils, are believed to be crucial in plant defence against herbivore predators, according to Labrano et al. (2014). Complex essential oils may be more effective than single pure molecules since plants often present their defences as a collection of compounds. For all of these considerations, we can conclude that



plant essential oils, particularly those of garlic and mint, could be regarded as a natural option for the treatment of *A. ipsilon*.

#### Conclusion

Because of their benefits for the environment and the safety of mammals, the use of plant essential oils has been regarded as a significant option for pest management. The present study's findings suggest that garlic oils have a harmful effect on all stages of *A. ipsilon*, are toxic to the larvae's stomach and skin, and have antifeedant and starving properties in all of the investigated oils. When the oils were combined, they had a potentiating impact on one another and became more toxic. (Garlic + Mint) was the potentially hazardous combo. In this light, it is possible to suggest using the plant essential oils to control the black cutworm on their hosts by spraying them as emulsions or as toxic baits alternatives to chemical pesticides. It is also possible to mix oils to increase their toxicity and reduce the development of insect resistance as a biological control method throughout the integrated pest management programme. The acquired results pointed to a fascinating prospect to create bioinsecticides based on plant essential oils for the management of this harmful lepidopteran pest and other pests that can have an impact on output. However, more research and work on the field evaluation are required.

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