

MANAGEMENT OF WOOLLY APPLE APHID, *Eriosoma lanigerum* (Hausmann) USING NEW INSECTICIDE MOLECULES AND BIOPESTICIDES

Thesis

by

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(H-2015-14-M)**

Submitted to



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The assistance and help received during the course of investigations have been fully acknowledged.

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I own entire responsibility for the errors and omissions, if any.

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Chapter-1

INTRODUCTION

Apple (*Malus x domestica* Borkhausen) is commercially the most important temperate fruit crop and is fourth among the most widely produced fruits in the world after banana, orange and grapes. China is the largest apple producing country in the world. India ranks fifth in apple production where it is mostly grown in the states of Jammu & Kashmir, Himachal Pradesh, Uttarakhand and some parts of Arunachal Pradesh and Nagaland. The area under apple cultivation in India is 3,13,000 hectares with a production of 24,97,700 Metric Tonnes. Jammu & Kashmir has remained the leading apple producing state accounting for 66.0 per cent of the total production in the country followed by Himachal Pradesh producing 7,38,700 Metric Tonnes of apple (accounting for 29.6% of total production) from an area of 1,07,700 hectares (NHB, 2015).

Apple crop is attacked by large number of insect pests which infest the crop in various stages of development affecting the crop quantitatively and qualitatively. Woolly apple aphid, *Eriosoma lanigerum* (Hausmann) is one of the major sucking pests of apple in all apple growing regions of state. Adults and immatures are reddish-purple in color with a long, white, filamentous waxy covering, hence the name woolly apple aphid. This pest remain in colonies and from a distance a colony of woolly apple aphids appears as mass of cotton on the tree.

The pest is native of Eastern North America where it originally occurred on American elm, *Ulmus americana* L., hawthorn, *Crataegus* spp., and mountain ash, *Sorbus* spp. (Patch, 1912) and is cosmopolitan in distribution except in hotter parts of the tropics. It has since adapted to apple, after cultivated apple was introduced into North America, and has spread to other apple-growing regions of the world. This pest was introduced in India during 18th century with the imported apple rootstocks from China. It was first recorded in Conoor, Tamil Nadu during 1889 and in 1909 it was reported from Kumaon Hills of Uttar Pradesh (now Uttarakhand), and in 1910 from Shimla (Lal and Singh, 1947). Unlike its native place where it exploits more than one hosts, in India only apple has been reported as its host so far. Thakur and Dogra (1980) reported *E. lanigerum* as the most important pest of apple in India.

Both aerial and edaphic plant parts are infested by this pest. It causes damage by piercing the bark (i.e. phloem tissue) and sucking the sap resulting in reduced tree vigour. Woolly apple aphid causes hypertrophy and gall formation and these galls develop splits in

reaction to feeding in as little as eight weeks. These galls are result of proliferation of nonfunctional xylem tissue, which greatly reduces water conductivity of roots i.e. up to five times than a non-infested plant according to a study and this may be the major reason why the growth of young trees infested with woolly aphid is reduced (Brown and Schmitt, 1990; Brown *et al.*, 1991). Such, root infestations can decrease the growth rate of shoot tissue, and decrease trunk diameter in newly-planted trees (Weber and Brown, 1988; Brown and Schmitt, 1990). In years of high crop-load, the number and weight of fruit per tree can be reduced, leading to economic losses (Brown *et al.*, 1995). The wounds on the trunk and stems caused by aphid colonies or other factors serve as long-term feeding and overwintering sites (Childs, 1929).

An aphelinid endoparasitoid, *Aphelinus mali* (Haldeman) introduced in Kullu valley in 1937 (Rahman and Khan, 1941) is well established in some areas and check the population of aphid, but because of re-infestation from root population which remain beyond the approach of parasitoid, it become difficult to control the pest completely. Natural populations of predatory biotic agents mainly coccinellids, syrphids and chrysopids (Rahman and Khan, 1941; Lal and Singh, 1947 and Rawat and Pawar, 1987) exercise some control on aerial parts and checks further multiplication if present in good numbers.

Use of insecticides is an important tool in the management of woolly apple aphid and large number of insecticides mainly the organophosphates and carbamates have been recommended for the same (Attri and Sharma, 1971; Hameed *et al.*, 1974; Thakur and Dogra, 1980; Thakur and Gupta, 1998; Singh and Gupta, 2006 and Gupta *et al.*, 2013).

Most of the earlier recommended insecticides against this pest are not in the Central Insecticide Board (CIB) approved usage list of insecticides. Further some new insecticide molecules with better efficacy and safety have been introduced against sucking pests. There is dire need of testing some alternate methods (like bio-pesticides) against the pest as practically no work has been done against the pest on this aspect. So in the present studies, some new insecticide molecules and bio-pesticides were evaluated against the pest with following objectives:

1. To test the efficacy of some new insecticide molecules and bio-pesticides against woolly apple aphid.
2. To observe their effect on the natural enemies.

Chapter-2

REVIEW OF LITERATURE

Management of woolly apple aphid (WAA), *Eriosoma lanigerum* (Hausmann), a major insect pest of apple, has a long history which includes use of different insecticides and biological control agents. Recently some new insecticide molecules with different modes of action have also been developed to counteract the adverse effects of use of old group insecticides. Some alternate methods like use of biopesticides is also of interest keeping in view the environmental concern.

So, in this chapter, literature pertaining to some new insecticides namely spirotetramat, flupyradifurone, flonicamid and buprofezin used in present study has been reviewed. Literature with regard to biopesticides used in present investigation have also been provided. In addition, a brief review of their effect on natural enemies has also been given. For the sake of brevity the review of literature is divided into following three subheads.

2.1 Insecticides used in recent past

Aerial application of various insecticides of different groups have been recommended earlier by a large number of workers for the management of WAA in India (Gupta *et al.*, 1969; Hameed *et al.*, 1974; Thakur and Gupta, 1998). Two applications of dimethoate (0.03%), oxydemeton methyl (0.025%) and phosphamidon (0.025%) once in March-April and again in June have been found effective against the aphid in Himachal Pradesh (Attri and Sharma, 1971).

In nurseries, root dips of fenitrothion, soil treatment with granules of dimethoate, banding of trunks with granules of the insecticides have been found useful in decreasing aphid infestations (Thakur and Dogra, 1980).

Molinari (1986) while working on the management of WAA, reported that aerial spray application of mineral oil in winters followed by that of chlorpyrifos or oxydemeton-methyl during growing period particularly after flowering (when predators are less abundant) is quite effective. Reasonably good control of the aphid using pirimicarb without adverse effects to the natural enemy (*A. mali*) of the pest has been reported by Staubli and Chapuis (1989), however, it was also pointed out by the workers that the chemical lacked persistence and was also ineffective in killing hidden colonies of WAA.

A single spray of OP insecticide, vamidothion, was reported to give good control of woolly apple aphid in Africa but tolerance of the pest (WAA) to vamidothion has been observed in South Africa (Pringle *et al.*, 1994).

A new addition to the chemical arsenal against the woolly aphid is the systemic aphicide imidacloprid (Confidor) which is having good systemic root activity and chemical also provides excellent control of woolly aphid on trees (Loveless, 1999). Five different insecticides namely imidacloprid (0.04%), acetamiprid (0.01%), thiamethoxam (0.05%), acephate (0.12%) and profenofos (0.10%) were evaluated against the subterranean population of the woolly apple aphid, *E. lanigerum* by drenching treatment of apple seedlings raised in earthen pots. All the test insecticides were found at par with phorate (1 g a.i./plant) and significantly superior over 0.08 per cent chlorpyrifos and were effective for 30 days against the root form of the aphid except chlorpyrifos which remained effective only for 10 days (Singh and Gupta, 2006).

Ateyyat (2012) tested four insecticides (imidacloprid, pirimicarb, cartap hydrochloride and petroleum oil) for their suitability in an integrated pest management programme of woolly apple aphid in Jordan. To determine the degree of selectivity and suitability of the insecticides in the programme, three indices were developed. The study revealed that imidacloprid was most effective in suppressing the aphid population. The index used to measure the negative impact of insecticides on the parasitoid, *A. mali* indicated that pirimicarb was the safest chemical among all the four test chemicals while, imidacloprid proved most toxic to the mummified aphids. Both the above indices were integrated in a selectivity index to determine their IPM compatibility. The selectivity index indicated that pirimicarb is the most suitable insecticide for controlling WAA as its application resulted in significant reduction of WAA with very low negative impact on the parasitoid.

2.2 New insecticides and some biopesticides

Broad spectrum nature, higher persistence, adverse effect on the useful fauna (natural enemies, pollinators) and environment (residues in food, water etc.) and development of resistance in many insect pests against organophosphates and carbamates, demanded for the development of new pesticide molecules, overcoming all above ill effects. As such, new chemicals were synthesized by various manufacturers all over the world but so far, practically little work appears to have been done on management of woolly apple aphid using these

insecticides. Hence, the literature pertaining to the new group insecticides used in the present studies against some other sucking pests has also been reviewed.

2.2.1 Spirotetramat

Spirotetramat is an innovative new insecticide used to control sucking insect pests. It belongs to a new chemical class, tetramic acids, and interferes with lipid synthesis. It is highly systemic with two way distribution mechanism via the phloem and xylem in plants, to eliminate sucking insect pests (Brück *et al.*, 2009).

Nauen *et al.* (2008) reported that under laboratory conditions, when apple seedlings infested with woolly apple aphid were treated with spirotetramat (20-100 µg a.i. per leaf) resulted in 70-90 per cent reduction in woolly apple aphid population after 28 days of application.

Studies conducted to evaluate bioefficacy, persistence and median lethal concentration of spirotetramat as foliar application against mealy bug, *Phenacoccus solani* (Ferris) in Tamil Nadu Agricultural University on cotton revealed that the insecticide was highly effective in checking the population of mealy bug when applied @ 75 g a.i./ha (Kumar *et al.*, 2008a). Another study by Kumar *et al.* (2008b) against the aphid, *Aphis gossypi* at similar dosage revealed that spirotetramat was highly effective in checking the population of the pest and was on par with the standard check, imidacloprid @ 25 g a.i./ha. The study also revealed that all treatments of the insecticide resulted in increased seed cotton yield under field conditions.

Field studies were conducted to evaluate spirotetramat as foliar application for its bioefficacy against cotton whitefly, *Bemisia tabaci* in Tamil Nadu and it was revealed that spirotetramat (@ 75 g a.i./ha) reduced whitefly population up to 89.7 per cent over control and the chemical persisted up to 25 days (Kumar *et al.*, 2009).

Bioefficacy studies were carried out by Udikeri *et al.* (2009) at Agricultural Research Station, Dharwad, Karnataka, against sucking pests of cotton using spirotetramat 150 OD and a combiprodut of thiamethoxam 141 SC + lambda cyhalothrin 106 SC. The studies showed that the population of thrips, leafhoppers and aphids was brought below ETL with three sprays during 2006 and two sprays in 2007. Significantly highest seed cotton yield of 20.32 q/ha (2006) and 29.22 q/ha (2007) was obtained with the combiprodut (@ 300ml/ha) and

spirotetramat (@500ml/ha), respectively and were on par with acetamiprid 20 SP, a standard check used in the studies.

The effectiveness of spirotetramat (Movento 100 SC) at two doses viz., 1.8 l/ha and 2.25 l/ha for the control of pear psylla (*Cacopsylla pyri*) on pear trees was investigated in five field experiments in Poland, during 2009- 2011. Two treatments were applied in June; the first treatment in the early stage of hatching, whilst the second application was carried out about two weeks later. In all the experiments, higher dose of insecticide effectively reduced the pest population by 75.3-91.4 per cent after one week and by 83.7-97.6 per cent after two weeks of single application, which was 58.4-91.3 per cent and 56-92.6 per cent at lower dose of the insecticide. In comparison with standard insecticides viz. diflubenzuron and novaluron the efficacy of spirotetramat was higher (Krystyna *et al.*, 2012).

Sunda *et al.* (2015) conducted an experiment to investigate the field efficacy of some newer insecticides against insect pests of brinjal and it was revealed that combination of spirotetramat + imidacloprid (@ 75 + 75 g a.i./ha) was found most effective for the control of jassids and white flies as it accounted for more than 76.11 per cent control of these pests whereas, spirotetramat + imidacloprid (@ 60 + 60 g a.i./ha) combination was found statistically at par with imidacloprid (@ 75 g a.i./ha).

Three field trials using spirotetramat, imidacloprid, thiamethoxam and flonicamid to control *Myzus persicae* were laid out in two nectarine farms containing three varieties each at Italy during spring. The results revealed that spirotetramat was the most persistent insecticide whereas flonicamid the most efficacious of all the four test chemicals used (Pasqualini and Scannavini, 2015).

A trial conducted in Virginia, USA, to control grapes mealy bug, *Pseudococcus maritimus* by using two chemicals viz., dinotefuran (Scorpion, 0.292 L/ha) and spirotetramat (Movento, 0.439 L/ha) revealed that, both the chemicals significantly reduced mealybug counts but spirotetramat showed high persistence and reduced mealy bug population (Jones and Mizuho, 2016).

2.2.2 Flupyradifurone

Flupyradifurone is the first representative of the novel butenolide class of insecticides active against various sucking pests and showing an excellent safety profile against non-

target insects and has been used to control a variety of sucking insect pests many parts of the world (Nauen *et al.*, 2015).

Bioefficacy of eight insecticides viz., spinosad, fipronil, imidacloprid, acetamiprid, thiamethoxam, flupyradifurone, dichlorvos and nimbecidine was evaluated against mulberry thrips, *Pseudodendrothrips mori* (Niwa) in Karnataka. Among different treatments, fipronil, flupyradifurone, acetamiprid, imidacloprid and thiamethoxam were found effective in reducing the population of the pest, to 0.97, 0.99, 1.03, 1.05 to 1.08 individuals per top three leaves, respectively, at 5 days after spray as compared with 15.31 individuals per three leaves recorded in control (Patil *et al.*, 2013).

In a trial conducted against whitefly, *B. tabaci* on tomato crop using three chemicals viz., cyazypyr, flupyradifurone and pyrifluquinazon in Florida it was revealed that flupyradifurone and cyazypyr reduced both the whiteflies population and incidence of tomato yellow leaf curl virus significantly (Smith and Nagle, 2014).

2.2.3 Flonicamid

Flonicamid, a novel class insecticide from nicotinoiden group possessing unique chemical structure. This compound is very active against a wide range of aphid species and some other species of sucking insects. It rapidly inhibits the feeding behavior of aphids and provides long-lasting control. The chemical shows no cross-resistance to conventional insecticides, exhibits excellent systemic and translaminar activity and has no negative impact on beneficial insects and mites. Furthermore, it has a favorable toxicological, environmental and ecotoxicological profile. These characteristics make flonicamid well suited for resistant management strategies and integrated pest-management programs (Morita *et al.*, 2014).

Four insecticides including a new molecule flonicamid @ 50, 75 and 150g a.i. /ha, imidacloprid, thiamethoxam (both @ 25g a.i. /ha) and clothianidin (@ 12.5g a.i. /ha) were field screened against brown plant hopper, *Nilaparvata lugens* (Stal.) infesting rice both during kharif and summer seasons at the Central Research farm of Odisha University of Agriculture and Technology, Bhubaneswar. Significantly low BPH population (1.40-1.30/hill) was observed in flonicamid @ 150 g a.i. /ha treatment with a reduction of 88.73 and 90.30 per cent over untreated control during kharif and summer seasons, respectively (Misra, 2009).

Ten insecticides were evaluated against major sucking pests infesting the Bt cotton at KVK farm, JAU, Targhadia (Rajkot) during Kharif, 2012-13. Among test products, five were

bio-pesticides viz., neem oil (1.0 %), NSKE (neem seed kernel extract) (5.0 %), azadirachtin (0.0009 %), *Verticillium lecanii* (@ 2.5 kg/ha) and *Beauveria bassiana*(@ 2.5 kg/ha) and five were synthetic chemical pesticides viz., acetamiprid (0.004 %), thiamethoxam (0.01 %), imidacloprid (0.0089 %), dinotefuran (0.008 %) and flonicamid (0.02 %). The results showed that the chemical pesticides caused higher mortality, while bio-pesticides caused moderate to lower mortality of sucking pests. Among the insecticide treatments, flonicamid (0.02 %) was found more effective against all major sucking pests, acetamiprid (0.004 %) against aphid and whitefly, dinotefuran (0.008 %) and imidacloprid (0.0089 %) against jassid and, thiamethoxam (0.01 %) was found effective against thrips. In relation to toxicity of insecticides on predators (Coccinellids and *Chrysoperla*) of sucking pests, all the bio-pesticides were found safer to predators, while, chemical pesticides were found moderate to highly toxic (Ghelani *et al.*, 2014).

Kalasariya (2016) evaluated bioefficacy of insecticide spray schedules against aphid, *Lipaphis erysimi* (Kaltenbach) in mustard in a field experiment conducted at Junagadh Agricultural University, Junagadh during Rabi 2012-13 and 2013-14. The results revealed that flonicamid (0.02 %) at seedling stage, flubendamide (0.014 %) at pre-flowering stage, azadirachtin (0.15 %) at 50 per cent flowering stage and acephate + fenvalerate (0.028 %) at 50 per cent pod formation stage were significantly the most effective treatments which recorded lowest population of the aphid on the crop.

2.2.4 Buprofezin

Buprofezin is an insecticide which is used for the control of insect pests such as, mealy bugs, leaf hopper and whitefly on vegetable crops. It acts as a growth regulator, inhibiting chitin synthesis in insects (Uchida *et al.*, 1985).

Buprofezin as foliar spray at 1125 and 1500 ml/ha was tested for its bio-efficacy against grape mealy bug, *Maconellicoccus hirsutus* (Green) in comparison to carbosulfan and chlorpyrifos during rabi 2002 and 2003 seasons. The results showed that buprofezin reduced the nymphal and adult population and bunch infestation of *M. hirsutus* and increased the fruit yield in comparison to untreated check and recommended insecticides. The study also revealed that the efficacy of both the doses was statistically on par, the lower dose (1125 ml/ha) being sufficient to effectively mitigate the mealy bug colonies (Muthukrishnan *et al.*, 2005).

A field trial was conducted by Hegde and Nidagundi (2009) at the Agricultural Research Station, Siruguppa for two consecutive kharif seasons of 2004 and 2005 to study the effect of buprofezin 25 SC at two different doses (1 ml/L and 0.75 ml/L), against plant hoppers (brown plant hopper and white backed plant hopper) and their mirid predator, *Cyrtorhinus lividipennis* (Reuter) on rice. The results clearly indicated that buprofezin at higher dosage (1 ml/L) recorded the lowest plant hopper population at 10 days after spray which was significantly superior to the lower dose (0.75 ml/L) and at par with standard checks thiamethoxam 25 WG (@ 0.2 g/L) and imidacloprid 17.8 SL (@ 0.3 ml/L). Buprofezin at both dosages tested, recorded significantly higher predatory mirid bug population over other treatments while, the higher dose (1 ml /L) recorded highest yield and was on par with the lower dose (0.75 ml/L).

A field experiment in Gujarat was carried out to evaluate the bioefficacy of buprofezin against mealy bug in cotton. Three doses viz. 250 g a.i. /ha, 312.5 g a.i. /ha and 625 g a.i. /ha of the chemical were tried. The experiment revealed that efficacy of insecticide was dose dependent and more than 95 per cent reduction in mealy bug population over control was recorded after three days of spray at all the three doses. (Patel *et al.*, 2010). The insecticide was more effective at two higher doses compared to its lower dose. Efficacy of chemical against early and later instar nymphs of the mealy bug has also evaluated and was also found dose dependent. It was more toxic to early instars than later instar nymphs. It was most effective against early as well as later instars nymphs at the higher dose (625 g a.i. /ha). At lower doses (250 g a.i. /ha and 312.5 g a.i. /ha), its efficacy was comparable to chlorpyrifos (400 g a.i. /ha) and carbaryl (1000 g a.i. /ha).

Field studies were undertaken to study the comparative bioefficacy of buprofezin at two doses viz., 75 g a.i./ha and 150 g a.i./ha against sucking pests of cotton and its safety to natural enemies during kharif, 2008 and 2009 in farmers field in Odisha. The study revealed that when applied @ 150 g a.i. /ha, the insecticide was highly effective in checking the sucking pests namely *B. tabaci*, *Amrasca biguttula biguttula*, *Aphis gossypii* and *Scirtothrips dorsalis*, in both the years and it had no adverse effect on the population of natural enemies (Sontakke *et al.*, 2013).

Das and Islam (2014) conducted a study using four insecticides viz., Fipronil, imidacloprid, buprofezin and thiamethoxam + emamectin benzoate to evaluate their toxicity against sucking pests (*Amrasca devastans* and *B. tabaci*) infesting brinjal during winters. The

study revealed that fipronil, imidacloprid and buprofezin proved effective while the combiproduct, thiamethoxam + emamectin benzoate proved least effective in controlling these two species.

An experiment was conducted by Mandal *et al.* (2015) to evaluate the efficacy of few new groups of insecticides against two important sucking pests of cotton viz., cotton jassid (*A. biguttula biguttula*) and whitefly (*B. tabaci*) during the pre-kharif season of 2011 and 2012. The insecticides viz., imidacloprid (18, 27 & 36 g a.i./ha), spiromesifen (100, 150 & 200 g a.i./ha) and buprofezin (150, 200 & 300 g a.i./ha) were applied twice at 15 days interval. From the experiment it was concluded that among the treatments, the most effective insecticide in controlling the jassid population was buprofezin (300 g a.i./ha) followed by imidacloprid (36 g a.i./ha) and least effective was spiromesifen (100 g a.i./ha). The order of bioefficacy on the basis of per cent reduction of cotton jassid over control plot was: buprofezin > imidacloprid > spiromesifen. But in case of whitefly, the most effective insecticide was spiromesifen (150 g a.i./ha) followed by buprofezin (300 g a.i./ha) and least effective was buprofezin (150 g a.i./ha). The order of bioefficacy on the basis of per cent reduction of cotton whitefly over control was: spiromesifen > buprofezin > imidacloprid.

Two field experiments were conducted during kharif of 2011 and 2012 at Agricultural Research Station Raichur by Nadagouda *et al.* (2015) to test the bioefficacy of buprofezin against sucking pests of *Bt* cotton and its safety to natural enemies at different doses and it was found that the higher dose (300g a.i./ha) recorded significantly low leafhopper population of 0.32 per leaf at seven days after spraying. This was followed by its next lower dosage (250g a.i./ha). The molecule also brought about significant reduction of whiteflies and mealybugs. It did not exhibit any phytotoxicity on *Bt* cotton and was safe to the natural enemies viz. spiders and coccinellids as well. It was also observed that buprofezin (300 g a.i./ha) recorded the highest seed cotton yield and this was at par with its next lower dosage (250 g a.i./ha), both being significantly superior to imidacloprid and acephate.

2.2.5 Biopesticides

Against woolly apple aphid

A number of different insect pathogenic fungi have been recorded infecting woolly apple aphid (Damavandian and Pringle, 2007). The majority of entomopathogenic fungi (EPF) are found in the order Entomophthorales, of which approximately 200 species are regarded as pathogens of insects and mites (Benny *et al.*, 2014). Since *Beauveria bassiana*

and *Metarhizium anisopliae* already exist in commercial formulations, the efficacy of large-scale field applications need to be investigated.

Among botanicals, insecticides with semi systemic activity, such as azadirachtin or neem oil-based products, have been tested, but their effectiveness has usually been found to be insufficient (Caldwell *et al.*, 2013). Other plant protection products that are increasingly being used for woolly aphid control, are paraffin oil-based products, applied in combination with sulphur-based products (Kelderer *et al.*, 2016).

Against other sucking pests

Field evaluation of fish oil insecticidal rosin soap against soft green scale, *Coccus viridis* (Green) on citrus by Singh and Rao (1979) in Karnataka, India, showed that this pest on mandarin could be effectively controlled using fish oil insecticidal rosin soap (4%). A 3-year study by Rishi (1982) on the main fruit pests of Jammu & Kashmir, India, with particular reference to *Quadraspidiotus perniciosus* (Comst.), was carried out in an apple orchard containing pear, plum, peach and walnut trees also to evaluate the possibilities of controlling the scale using selective insecticides at the most favourable times from the point of view of biology and environmental conditions. The application of conventional oil-emulsion sprays in conjunction with Bordeaux mixture, fish-oil soap and 40 per cent nicotine sulfate at vulnerable stages of the life-cycle resulted in a 40-60 per cent decline in its overall incidence, and also showed increased effectiveness against other key pests.

Similarly, Karupuchamy *et al.* (1998) studied seasonal incidence and management of aphid, *Aphis punicae* on pomegranate at Tamil Nadu in India and found that fish oil rosin soap at 2.5 per cent was highly effective in controlling the aphid population. Beattie *et al.* (2002) reported that oil content of fish emulsion fertilizer suppressed oviposition of citrus leaf miner on lemon (*Citrus limon* L.) trees.

The bio-agent, *Beauveria bassiana* was found to reduce white fly population in cotton by 39.7 to 72.6 per cent. However, it was ineffective against jassids, as it could bring about only 10-14 per cent reduction in population. Neem oil was more effective against jassids (20.4-42.5% reduction) than that of neem seed kernel extract (NSKE). However, both the neem products tested (neem oil & NSKE) were found to reduce more than 50 per cent of whitefly population on the crop (Jat and Jeyakumar, 2006).

A field experiment was conducted by Anitha and Nandihalli (2008) to find out the efficacy of certain botanicals and mycopathogens against okra leafhopper, *A. biguttula*

biguttula and aphid, *Aphis gossypii* at Agricultural Research Station, Dharwad, Karnataka. Results revealed that, among botanicals and mycopathogens, neem oil (2%) was found effective in controlling the leafhopper population (1.41 and 4.11 leafhoppers/ 3 leaves after 15 days of first and second spray, respectively) while NSKE (5%) was found effective against aphid (2.10 and 5.22 aphids/ 3 leaves at 15 DAS of first and second spray, respectively). Among biorationals, NSKE proved to be the most economical treatment as realized by highest net returns.

Ahirwar *et al.* (2010) conducted a field trial to evaluate field efficacy of natural and indigenous products on sucking pests (jassids, mirid bug and whitefly) of sesame. Results revealed that incidence of nymph and adult population of the pests decreased significantly by both the natural and indigenous product treatments viz. neem oil, neem seed kernel extract, neem leaf extract, garlic bud + red pepper extract, cow urine, and cow butter milk as compared to the control. Of all the tested products, neem seed kernel extract gave maximum control of pest population followed by neem oil and neem leaf extract. Cow urine and cow butter milk were recorded to be least effective.

Chand *et al.* (2010) evaluated efficacy and economics of two indigenous plant products viz. seed extracts of *Strychnos nuxvomica* (Loganeaceae) and *Pachyrrhizus erosus* (Papilionaceae), using petroleum ether as solvent and an entomopathogenic fungus, *Beauveria bassiana*, against the aphids, viz. *L. erysimi* and *M. persicae* on broccoli. *S. nuxvomica*, when used @ 4.0 ml/L. of water, resulted in 86.82 per cent reduction of aphids. *B. bassiana* and *P. erosus* @ 3.0 g/L. and 4.0 ml/L. of water, respectively, caused 84.18 per cent and 83.52 per cent, reduction in aphid population and yield of 111.55 and 104.25 q/ha, respectively.

A field experiment was conducted at Agricultural College and Research Institute, Maduraito to evaluate the field efficacy of botanicals and fish oil rosin soap for the management of major pests of coleus (*Coleus forskohlii* Briq.). Ten rounds of application of six botanicals and one animal origin insecticide at fortnightly interval were made and the incidence of thrips (*Scirtothrips dorsalis* Hood), scale insects (*Orthezia insignis* Browne) and defoliator (*Orphanostigma abruptalis* Wlk.) on coleus were recorded. Azadirachtin (0.15%) @ 1.5 ml/L was effective in reducing the incidence of thrips and defoliator, recording a mean leaf damage of 6.1 and 6.6 per cent for thrips and defoliator. Fish oil rosin soap (FORS) (@ 25g/L) was effective against scale insects of coleus and recorded the lowest mean leaf

damage of 4.8 per cent. Azadirachtin recorded the highest yield of wet tubers/ha (Thangavel *et al.*, 2011).

A field study was carried out to find out the effect of different botanical pesticides and a chemical pesticide on the sucking pests of cotton, *Gossypium hirsutum*. Of the different botanicals used, neem seed kernel extract (5%) was found to be the most effective followed by *Pongamia glabra* seed kernel extract (5%), neem oil (3%) and *Pongamia glabra* oil (3%) in the decreasing order against the sucking pests (leafhopper and aphids) of cotton. Maximum population reduction was noticed on the 3rd day after treatment (Vinodhini and Malaikozhundan, 2011).

An experiment was conducted at farmers field in Tamil Nadu for the management of mealy bug of custard apple using four biopesticides viz., *Verticilium lecanii* (1.15%), *V. lecanii* (3%), *Beauveria bassiana* (1.15%), neem seed kernel extract (5%) and four insecticides namely buprofezin (0.02%), imidacloprid (0.005%), diafenthiuron (0.02%) and dichlorvos (0.05%). The results revealed that buprofezin (0.02%) was most effective as compared with other test products viz. *V. lecanii* (1.15%) @ 3 g/L, *V. lecanii* (3%) @ 3 g/L, *B. bassiana* (1.15%) @ 5 g/L, neem seed kernel extract at the rate of (5%) @ 50 g/L, imidacloprid (0.005%), diafenthiuron (0.02%) and dichlorvos (0.05%) and control (water spray) (Kulkarni and Patil, 2013).

In a study conducted in Bangladesh by Akmal *et al.* (2013), *B. bassiana* was used to evaluate its pathogenicity against adults of four aphid species namely *Schizaphis graminum*, *Rhopalosiphum padi*, *Brevicoryne brassicae* and *L. erysimi*, and their natural enemy *Coccinella septempunctata*. *B. bassiana* was found effective at all concentrations i.e. 1×10^6 , 1×10^7 and 1×10^8 spores/ml on all aphid species, but the uppermost concentration (1×10^8 spores/ml) provided maximum control within short period of time. *B. bassiana* showed little or no detrimental effects to *C. septempunctata*. By testing the field efficacy of *B. bassiana* against aphid species it is concluded that, this insect pathogenic fungus can be used as potential biocontrol agent for the management of aphids.

2.3 Effect of test insecticides and biopesticides on different natural enemies

Altogether, 73 species of natural enemies of woolly apple aphid have been recorded from worldwide, most common being *A. Mali* (Asante, 1997). The predators known to feed

on WAA colonies include syrphids, chrysopids, coccinellids, predatory hemipterans and earwigs (Asante, 1997; Gontijo *et al.*, 2012).

In India, for the control of woolly aphid, *A. mali* was introduced from UK at Saharanpur (Uttar Pradesh). The parasitoid failed to establish and provide effective control of woolly aphid in the Kumaon hills because of the intense activity of coccinellid beetle, *Coccinella septempunctata* which fed indiscriminately on parasitized as well as non-parasitized woolly aphids. *C. septempunctata*, however provided satisfactory control of the pest. The parasitoid could control woolly aphid in Kullu valley in Himachal Pradesh and has spread to Kashmir valley. It was transferred to Coonoor (Tamil Nadu) from Kullu where it got established. At present, it has established in all apple growing areas of the country, but is more effective in valleys rather than on mountain slopes. The aphilenid has in fact emerged as a key population regulatory agent of *E. lanigerum* in the areas of its successful establishment (Singh, 2004).

In a laboratory study, toxicity of pirimicarb, imidacloprid, dimethoate, lambda-cyhalothrin, flonicamid and spinosad to two-spot ladybird, *Adalia bipunctata*, was evaluated via residual contact and ingestion through feeding on contaminated green peach aphids (*M. persicae*). The study revealed that flonicamid and spinosad had no lethal effects on larvae and female adults. Pirimicarb was harmless to the predator by ingestion exposure but showed some residual toxicity at high concentrations to both larval and adult stages. Beside these, all the insecticides used were highly toxic to the *A. bipunctata*. Findings suggest that pest management programs in agricultural crops using dimethoate, lambda-cyhalothrin and, to a lesser degree, imidacloprid, are detrimental to *A. bipunctata*, whereas pirimicarb, flonicamid and spinosad are more compatible with the predator (Jalali *et al.*, 2009).

In a study carried out by Rogers *et al.* (2011) it was found that carbaryl caused 85 per cent mortality of *A. mali* after 21 days of treatment. The toxicity of five insecticides (lambda-cyhalothrin, dimethoate, flonicamid, mineral oil, spirotetramat) and a biopesticide *B. bassiana* to *Aphelinus certus*, a parasitoid of soybean aphid, was assessed under laboratory conditions. In a screening bioassay, the descending order of toxicity after 48 hours of application was, dimethoate >lambda-cyhalothrin>flonicamid >mineral oil >*B. bassiana*>spirotetramat. In general, lambda-cyhalothrin and dimethoate both were found harmful to *A. certus*. The other insecticides proved harmless to *A. certus* and are potential candidates for inclusion in soybean aphid management programs (Frewin *et al.*, 2012).

In an experiment lethal and sub lethal side effects of spirotetramat on adults and larvae of *Cryptolaemus montrouzieri* (Mulsant) were evaluated under laboratory conditions by topical application and by ingestion of treated individuals of *Planococcus citri* (Risso). The lethal and sub lethal effects of spirotetramat were compared to those of chlorpyrifos and pyriproxyfen. In the experiment spirotetramat proved harmless when directly applied on larvae and adults of *C. montrouzieri*, since it did not affect survival, longevity, fecundity, egg hatching, and offspring survival. Contrary to this, chlorpyrifos was classified as moderately toxic for adults due to its effects on fecundity, egg hatching and offspring survival while pyriproxyfen was classified as harmful for larvae due to the acute effect on pupal mortality. When larvae and adults of *C. montrouzieri* were fed with treated prey, spirotetramat again proved harmless in comparison to other two which were found to be harmful (Planes *et al.*, 2012).

Neetan and Aggarwal (2012) compared the toxicity of insect growth regulators and neonicotinoid-based insecticides recommended for the management of cotton mealy bug, *Phenacoccus solenopsis* (Tinsley) against different developmental stages of predatory coccinellid, *Cheilomenes sexmaculata* (Fabricius). Among various chemicals, buprofezin recorded significantly higher (76.66%) hatching of eggs of *C. sexmaculata*, followed by endosulfan (73.33%), imidacloprid (70.00%) and chlorpyrifos (63.33%). The emergence of adults from the treated pupae was highest in buprofezin (83.33%) followed by endosulfan (76.67%) and profenophos (53.33%). Buprofezin was found to be the safest insecticide, followed by endosulfan to all the stages of *C. sexmaculata*.

In a study undertaken by Gupta *et al.* (2013), it was revealed that parasitized aphids collected from chlorpyrifos (0.04 %) treated apple trees resulted only in 12.5 per cent emergence of the parasitoid, *A. mali* whereas it was 59.7 per cent in case parasitized aphids collected from thiamethoxam (0.025 %) treated apple trees.

Studies were carried on the adults of *Aphytis melinus* DeBach (Hymenoptera: Aphelinidae), a parasitoid of California red scale, *A. aurantii* (Maskell) (Hemiptera: Diaspididae), to evaluate the lethal effects of freshly applied and laboratory aged residues of eighteen selected pesticides recommended in IPM for citrus. According to their toxicity on the parasitoid, the pesticides could be divided into four groups on the basis of harmful effect and persistence. Etofenprox and chlorpyrifos were harmful and persistent; abamectin was moderately harmful and slightly persistent; dimethoate, chlorpyrifos-methyl, and

spirodiclofen were slightly harmful and moderately persistent; while the remaining 12 pesticides (azadirachtin, etoxazole, fenbutaestan, hexythiazox, mancozeb, petroleum oil, pirimicarb, propargite, pymetrozine, pyriproxyfen, spirotetramat, and tebufenpyrad) were considered harmless to the parasitoid (Vanaclocha *et al.*, 2013).

In another study conducted by Pandi *et al.* (2013) to evaluate the toxicity of some insecticides to grubs of *C. sexmaculata* it was found that among the insecticides tested, acetamiprid showed high toxicity to grubs, followed by thiamethoxam, imidacloprid, buprofezin and neembaan. Imidacloprid showed 14.8 times more toxicity in comparison to buprofezin, whereas neembaan produced less than 50 per cent mortality irrespective of the concentration tested. However, field recommended dosage of acetamiprid (0.006%) and thiamethoxam (0.005%) caused almost 50 per cent mortality of the predator. Whereas, buprofezin at recommended dose (0.013 %) was considered to be safer, because it produced 10, 16, and 22 per cent mortality at 24, 48, and 72 hours after treatment, respectively, under laboratory conditions.

The selective toxicity of three doses viz. 1.25, 2.5, 3.75 ml/l of spirotetramat 150 OD against the life stages of *Chrysoperla zastrowi sillemi* was studied under in vitro conditions. The highest dose of test insecticide caused 28.88 per cent mortality and hence can be considered 'harmless' to *C. z. sillemi* as per the categorization of IOBC (International Organization on Biological Control). There was no adverse effect of spirotetramat 150 OD on the egg hatching percentage, rate of pupation, adult emergence and fecundity of the predator and hence can be considered as a safer molecule to be integrated with *chrysoperla* in insect pest management programs (Amla *et al.*, 2015).

The toxicity of 11 modern insecticides applied at their maximum field recommended rate was tested to two life stages (adults and mummies) of *Eretmocerus mundus* (Mercet), an important natural enemy of *B. tabaci* in California, USA. Laboratory and persistence tests were conducted and effects assessed not only in terms of mortality but on the reproductive performance of the natural enemy as well. Flubendiamide, methoxyfenozide, spiromesifen and flonicamid were harmless, while spinosad and sulfoxaflor were harmful, and rest (spirotetramat, metaflumizone, deltamethrin, abamectin and emamectin) were slightly harmful (Maria *et al.*, 2015).

A two year study was conducted to evaluate toxicity of foliar application of spirotetramat in comparison to some common insecticides against spiders and coccinellid

predators on okra *Abelmoschus esculentus* (L.) in Coimbatore district during 2013 to 2014. The results revealed that spirotetramat (@ 60 and 75 g a.i./ha) recorded significantly less toxicity on natural enemies especially to spiders and coccinellids which was at par with higher dose (@ 90 g a.i./ha) of spirotetramat. All the treatments of insecticide were found to be moderately safe to natural enemies when compared to thiamethoxam and dicofol (Gajalakshmi *et al.*, 2015).

Chapter-3

MATERIALS AND METHODS

The present investigations were carried out in the Fruit Entomology Laboratory of the Department of Entomology, College of Horticulture, Dr Y S Parmar University of Horticulture and Forestry, Nauni (Solan) and in the farmer's field at Matiana, District Shimla, Himachal Pradesh.

Field study

For the management of aerial form of woolly apple aphid, an apple orchard (20 years old) having aphid infestation was selected (Plate-1). Two concentrations of each of test insecticides/biopesticides namely, spirotetramat, flupyradifurone, flonicamid, buprofezin, *B. bassiana*, fish oil, neem oil and azadirachtin were evaluated which were compared with the standard treatments i.e. chlorpyrifos (0.04 %) and thiamethoxam (0.025 %), and control, wherein only water spray was given. For making proper emulsion of neem oil, 0.01 per cent sticker (Indron-AE) was added in the spray solution. The spray application was done in autumn (October, 2016) with the high volume sprayer till the point of run off, and after each treatment, the pump was cleaned thoroughly with water to prevent any contamination. The details of the treatment are provided in Table-1.

The trial was laid out in randomized block design having three replications with a tree as replicate. Four branches were selected and tagged for taking observations on colony count and colony size.

Colony count:

Pre-treatment aphid colony count on 4 marked branches/tree was taken before spray application in all the trees taken for experimentation. Thereafter, the colony count was recorded on the same branches 3, 7, 14, 21 and 28 days of spray application. The reduction in colony count over pre-treatment count was calculated using following formula:

$$\text{Per cent reduction in colony count} = \frac{\text{Pre treatment count} - \text{Post treatment count}}{\text{Pre treatment count}} \times 100$$

Table 1. Details of different treatments used for management of woolly apple aphid

Test product	TREATMENT	CONCENTRATION (%)
Spirotetramat	T ₁	0.007
	T ₂	0.015
Flupyradifurone	T ₃	0.017
	T ₄	0.03
Flonicamid	T ₅	0.025
	T ₆	0.05
Buprofezin	T ₇	0.025
	T ₈	0.05
<i>Beauveria bassiana</i>	T ₉	1.0 (1x10 ¹⁰ cfus/ lt.)
	T ₁₀	2.0 (2x10 ¹⁰ cfus/ lt.)
Fish oil	T ₁₁	0.5
	T ₁₂	1.0
Neem oil	T ₁₃	1.0
	T ₁₄	2.0.
Azadirachtin	T ₁₅	0.01
	T ₁₆	0.02
Thiamethoxam	T ₁₇	0.025
Chlorpyrifos	T ₁₈	0.04
Control	T ₁₉	water spray

Colony size:

The colony size was recorded in 4 colonies/ selected branch (16 colonies/tree) with the help of a Vernier caliper before spray and 3, 7, 14, 21, and 28 days after spray. The efficacy of different treatments was judged by per cent reduction in colony size over the pre-treatment count.

$$\text{Per cent reduction in colony size} = \frac{\text{Pre treatment size} - \text{Post treatment size}}{\text{Pre treatment size}} \times 100$$



a) Infested tree



b) Close-up of woolly apple aphid colonies

Plate 1. Woolly apple aphid infestation

Table 2. Information regarding test insecticides and biopesticides used in present study

Technical name	Trade name	Manufacturer
Spirotetramat	Movento 150 OD	Bayer Crop Sciences
Flupyradifurone	Sivanto 200 SL	Bayer Crop Sciences
Flonicamid	Ulala 50 WG	United Phosphorus Limited
Buprofezin	Lapa 25 SC	Syngenta India Limited
Thiamethoxam	Actara 25 WG	Syngenta India Limited
Chlorpyrifos	Dursban 20 EC	Dow Agro Sciences
<i>B. bassiana</i>	Daman (1x10 ⁹ cfus/g)	International Panacea Limited
Fish oil	Petro Fish oil	Petro Star
Neem oil	Neem tel	Vaidya Bal Mukund and Sons
Azadirachtin	Neem Baan (10000 ppm)	Pest Control India

Effect of different treatments on Natural Enemies

***Aphelinus mali*:**

The effect of test insecticides/biopesticides was observed on Aphelinid endoparasitoid, *A. mali*. The parasitized aphids (lacking woolly strands and showing mummification) (Plate-2) were collected from the experimental trees 3, 7 and 14 days after spray and were brought to the laboratory where the parasitized aphids without exit holes were separated by observing under Stereoscopic Zoom Microscope using camel hair brush. Twenty parasitized aphids from each treatment collected after 3, 7 and 14 days of foliar application were kept in clean glass vials (15 ml Corning) for further observations in the laboratory (Plate-3). The data on emergence of parasitoid adults were recorded upto three months and thereafter the per cent emergence of parasitoid adults was calculated.

Coccinellids:

Since in the field the coccinellid population was quite low, so the toxicity of the test insecticides and biopesticides was evaluated against two common species namely *Coccinella septempunctata* and *Hippodamia variegata* which were collected from the field and were fed till three days with mustard aphids. The toxicity was evaluated using residue film method (Plate-4) where test insecticide and biopesticides at the field concentrations were sprayed on the Petri plates (4.5x4.5 cm diameter) with the help of an atomizer, on the bottom and sides and also on the lower sides of the upper Petri plate (Lid) and were allowed to dry, so that a film of the test product was made on the Petri plates. Twenty adults were released thereafter and the mortality was recorded after 24, 48 and 72 hours of release.

Statistical analysis

The data recorded under field conditions and laboratory conditions were subjected to statistical analysis to find out the significance of the results obtained. The data obtained under field and laboratory conditions were analyzed using Randomized Block Design and Completely Randomized Design respectively (Gomez and Gomez, 1983) using OPSTAT package.



Plate 2. Parasitized mummified aphids among woolly apple aphid colonies



Plate 3. Collected parasitized aphids kept in the lab for adult emergence

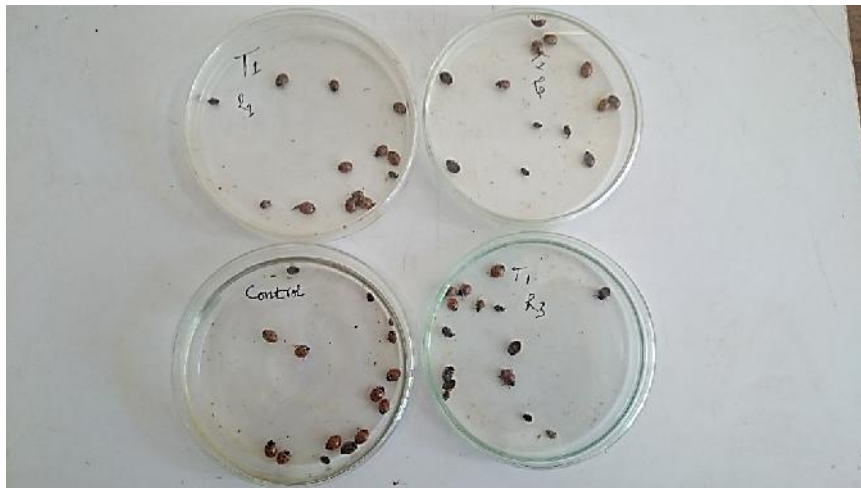


Plate 4. Estimation of toxicity to coccinellid using residual film method

Chapter-4

RESULTS AND DISCUSSION

Four insecticides viz. spirotetramat (Movento, 150 OD), flupyradifurone (Sivanto, 200 SL), flonicamid (Ulala, 50 WG), buprofezin (Lapa, 25 SC) and four biopesticides viz., neem oil, azadirachtin (Neem Ban, 10000 ppm), fish oil and *Beauveria bassiana* (Daman, 1×10^9 cfus/g) were sprayed at two selected concentrations each. Their effect was measured by taking into consideration two parameters i.e. colony size and colony count of the aerial population of the pest. The results so obtained are presented in the Tables 3 and 4. In addition, effect of the test products on natural enemies is also presented and discussed in this chapter.

4.1 Observations on colony count

When the data (Table 3) were recorded three days after treatment, spirotetramat at its higher concentration (0.015 %) proved most effective and resulted in 38.33 per cent reduction in colony count of woolly apple aphid which was at par with flonicamid (0.05 %) with 35.37 per cent reduction in colony count. Buprofezin was the least effective treatment and resulted in 14.99 and 16.30 per cent reduction in colony count at its lower (0.025 %) and higher (0.05 %) concentrations, respectively, being at par.

Amongst biopesticides, 17.50, 19.03, 19.86 and 16.76 per cent reduction in colony count was observed at higher concentrations of *B. bassiana*, fish oil, neem oil and azadirachtin respectively, which were at par with each other.

Spirotetramat (0.015 %) and flonicamid (0.05 %) with 38.33 and 35.37 per cent reduction were at par with thiamethoxam (0.025 %) where 38.80 per cent reduction in colony count was recorded after three days of treatment. Rest all the treatments were inferior to thiamethoxam (38.80 per cent reduction) as well as chlorpyrifos (0.04 %) where 51.80 per cent reduction in colony count was noticed just after three days of treatment. Among earlier recommended insecticides, chlorpyrifos (0.04 %), proved superior over thiamethoxam (0.025 %) after 3 days of treatment.

After seven days of treatment, again, spirotetramat (0.015 %) and flonicamid (0.05 %) were the most effective treatments among test insecticides as well as biopesticides as it resulted in 62.16 and 60.06 per cent reduction in colony count, respectively, being at par.

Table 3. Bioefficacy of insecticides and biopesticides against woolly apple aphid, *E. lanigerum* infesting apple (based on colony count)

Treatment	Concentration (%)	Percent reduction in colony count over pre-count (DAT)					Mean
		3	7	14	21	28	
Spirotetramat	0.007	21.60 (27.64)	33.26 (35.19)	43.63 (41.31)	46.73 (43.10)	51.26 (45.70)	39.30 (38.59)
	0.015	38.33 (38.23)	62.16 (52.03)	71.80 (57.93)	81.63 (64.66)	88.46 (70.18)	68.48 (56.61)
Flupyradifurone	0.017	15.34 (23.01)	20.20 (26.63)	25.46 (30.26)	32.50 (34.73)	38.35 (38.23)	26.38 (30.57)
	0.03	20.53 (26.91)	28.30 (32.16)	35.07 (36.28)	48.60 (44.17)	56.20 (48.55)	37.75 (37.62)
Flonicamid	0.025	20.66 (27.01)	29.10 (32.59)	35.20 (36.37)	45.06 (42.15)	53.00 (46.70)	36.60 (36.96)
	0.05	35.37 (36.47)	60.06 (50.84)	67.40 (55.16)	70.00 (56.82)	73.73 (59.17)	61.31 (51.69)
Buprofezin	0.025	14.99 (22.73)	20.16 (26.52)	29.50 (32.84)	36.66 (37.24)	38.30 (38.18)	27.92 (31.50)
	0.05	16.30 (23.77)	25.50 (30.31)	36.80 (37.32)	44.20 (41.63)	58.50 (49.88)	36.26 (36.58)
<i>B. bassiana</i>	1x10 ¹⁰ cfus/lt.	10.26 (18.62)	20.20 (26.62)	25.63 (30.37)	25.63 (30.37)	22.30 (28.12)	20.81 (26.82)
	2x10 ¹⁰ cfus/lt.	17.50 (24.61)	22.80 (28.52)	28.60 (32.29)	36.40 (37.07)	40.50 (39.49)	29.17 (32.40)
Fish oil	0.5	13.95 (21.83)	18.80 (25.65)	24.18 (29.35)	26.50 (30.93)	26.50 (30.93)	21.99 (27.74)
	1.0	19.03 (25.80)	25.90 (30.53)	30.50 (33.47)	40.60 (39.55)	40.60 (39.55)	31.32 (33.78)
Neem oil	1.0	14.16 (22.06)	18.30 (25.27)	25.83 (30.50)	28.30 (32.07)	28.53 (32.25)	23.03 (28.43)
	2.0	19.86 (26.41)	25.20 (30.06)	30.23 (33.30)	35.50 (36.52)	34.00 (35.63)	28.95 (32.38)
Azadirachtin	0.01	15.60 (23.20)	20.50 (26.78)	25.60 (30.33)	29.58 (32.88)	33.80 (35.51)	25.01 (29.74)
	0.02	16.76 (24.11)	25.70 (30.37)	31.80 (34.22)	39.77 (39.05)	35.08 (36.28)	29.83 (32.81)
Thiamethoxam	0.025	38.80 (38.50)	68.36 (55.76)	83.13 (65.76)	89.66 (71.53)	90.00 (71.22)	73.99 (60.56)
Chlorpyrifos	0.04	51.80 (46.02)	74.36 (59.89)	79.20 (62.84)	87.53 (69.34)	89.66 (71.24)	76.51 (61.86)
Control	-	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
Mean		22.47 (27.12)	32.25 (33.37)	42.06 (39.54)	48.02 (43.31)	50.40 (44.89)	

CD_{0.05}
 Treatment (T) : (1.75)
 Days (D) : (0.89)
 T x D : (3.91)
 DAT (Days After Treatment)

*Figures in parentheses are arcsine transformed values



Plate 5. Woolly apple aphid colony after chlorpyrifos treatment

Buprofezin (0.025 %) was the least effective treatment which resulted in only 20.16 per cent reduction in colony count and was at par with its higher concentration (0.05 %) and flupyradifurone (0.017 %) where 25.50 and 20.20 per cent reduction in colony count was recorded.

Among biopesticides, fish oil at its higher concentration (1.0 %) was most effective treatment resulting in 25.90 per cent reduction in colony count which was at par with the higher concentrations of all other biopesticides. The least effective treatment among biopesticides was lower concentration of neem oil which resulted in only 18.30 per cent reduction in colony count after seven days of treatment and was at par with the lower concentrations of all other biopesticides evaluated as well as higher concentration (2×10^{10} cfus/L) of *B. bassiana*.

Spirotetramat (0.015 %) was at par with thiamethoxam (0.025 %) where 68.36 per cent reduction in colony count was recorded, whereas rest all the treatments proved to be significantly inferior to the earlier recommended insecticides thiamethoxam (0.025 %). All the treatments including spirotetramat (0.015 %) were statistically inferior to chlorpyrifos (0.04 %) which resulted in 74.36 per cent reduction in colony count after seven days of treatment. Among earlier recommended insecticides chlorpyrifos proved to be significantly superior over the thiamethoxam at their recommended concentrations.

After fourteen days of treatment the trend remained same i.e. spirotetramat and flonicamid at their higher concentrations proved to be the most effective among new insecticides as well as biopesticides and resulted in 71.80 and 67.40 per cent reduction in colony count, respectively, which was at par with each other. The least effective treatment among new insecticides was flupyradifurone at its lower concentration (0.017 %) which resulted in 25.46 per cent reduction in colony count and was at par with buprofezin (0.025 %) where 29.50 per cent reduction in colony count was observed.

Among biopesticides, azadirachtin at its higher concentration (0.02 %), was most effective resulting in 31.80 per cent reduction in colony count which was at par with the higher concentration of all other biopesticides. The least effective treatment was fish oil at its lower concentration (0.5 %) which resulted in 24.18 per cent reduction in colony count after fourteen days of treatment and was at par with lower concentration of all other biopesticides as well as higher concentration (2×10^{10} cfus/L) of *B. bassiana*.

The earlier recommended insecticides namely thiamethoxam (0.025 %) and chlorpyrifos (0.04 %) proved superior over all the new treatments and resulted in 83.13 and 79.20 per cent reduction in colony count after fourteen days of treatment.

When observations were taken after 21 days of treatment, spirotetramat (0.015 %) proved to be most effective treatment resulting in 81.63 per cent reduction in colony count which was followed by flonicamid (0.05 %) where 70.00 per cent reduction in colony count was recorded. Flupyradifurone at its lower concentration (0.017 %) was the least effective treatment among the new insecticides and resulted in only 32.50 per cent reduction in colony count which was at par with lower concentration (0.025 %) of buprofezin where 36.66 per cent reduction in colony count was recorded.

Among biopesticides, fish oil at its higher concentration (1.0 %) was most effective treatment resulting in 40.60 per cent reduction in colony count which was at par with the higher concentrations of all other biopesticides when observed after 21 days of treatment. The least effective biopesticide was *B. bassiana* at its lower concentration (1×10^{10} cfus/L) where per cent reduction in colony count remained same as was recorded at previous observation i.e. 25.63 per cent which was at par with lower concentrations of all other biopesticides when observed 21 days after treatment.

All the test treatments were statistically inferior to the earlier recommended insecticides i.e. thiamethoxam (0.025 %) and chlorpyrifos (0.04 %) where 89.66 and 87.53 per cent reduction in colony count was observed, respectively, after 21 days of treatment.

On the last day of observation i.e. 28 days after treatment, a slight increase in per cent reduction in colony count was noticed and again spirotetramat (0.015 %) proved to be the most effective treatment resulting in 88.46 per cent reduction in colony count followed by flonicamid (0.05 %) where 73.73 per cent reduction in colony count was recorded. Buprofezin at its lower concentration (0.025 %) was the least effective treatment resulting in 38.30 per cent reduction in colony count and was at par with flupyradifurone at its lower concentration (0.017 %) where 38.35 per cent reduction in colony count was observed.

Among biopesticides, fish oil at its higher concentration (1.0 %) was the most effective treatment resulting in 40.60 per cent reduction in colony count and this was at par with

higher concentrations of all other biopesticides except neem oil where only 34.00 per cent reduction in colony count was observed after 28 days of treatment.

Spirotetramat (0.015 %) with 88.46 per cent reduction was at par with thiamethoxam (0.025 %) and chlorpyrifos (0.04 %) which resulted in 90.00 and 89.66 per cent reduction in colony count when recorded after 28 days of treatment. Whereas, rest of the test treatments were significantly inferior to the thiamethoxam (0.025 %) and chlorpyrifos (0.04 %).

The treatment mean for all insecticides and biopesticides at each concentration were calculated which are also presented in Table 3. Data revealed that among the test products, spirotetramat at higher concentration (0.015 %) was found significantly superior over all other insecticides and biopesticides with a mean reduction of 68.48 per cent, followed by flonicamid at higher concentration (0.05 %) which resulted in 61.31 per cent mean reduction in colony count. Spirotetramat at lower concentration (0.007 %) was next best treatment with 39.30 per cent mean reduction, and was at par with flupyradifurone at higher concentration (0.03 %) resulting in 37.75 per cent reduction in colony count. However the earlier recommended insecticides were found more efficacious.

4.2 Observation on colony size

The data presented in Table 4 reveal that after three days of application, flonicamid (0.05 %) and spirotetramat (0.015 %) were the most effective treatments among insecticides as well as biopesticides in reducing colony size of woolly apple aphid with 33.24 and 28.36 per cent reduction in colony size. The least effective treatments among test insecticides were buprofezin (0.025 %) with 15.16 per cent reduction in colony size and flonicamid (0.025 %) with 16.32 per cent reduction, both being at par.

Among biopesticides, azadirachtin at its higher concentration (0.02 %) was numerically better with 25.43 per cent reduction in colony size but was at par with the higher concentrations of *B. bassiana* and fish oil where 22.46 and 21.55 per cent reduction in colony size was observed after 3 days of spray, respectively. The least effective biopesticide was fish oil (0.5%) which resulted in 15.40 per cent reduction in colony size.

Among earlier recommended insecticides, thiamethoxam (0.025 %) resulted in 29.70 per cent reduction in colony size which was at par with the flonicamid (0.05 %) and

spirotetramat (0.015 %), whereas rest all the treatments were statistically inferior to thiamethoxam (0.025 %). Chlorpyrifos (0.04 %), the recommended insecticide resulted in 40.96 per cent reduction in colony size after 3 days of treatment and was found most effective.

After 7 days of treatment, flonicamid (0.05 %) again proved numerically better resulting in 49.67 per cent reduction in colony size but was at par with spirotetramat (0.015 %) treatment which resulted in 48.62 per cent reduction in colony size. Buprofezin at its lower concentration (0.025 %) proved to be the least effective treatment among new chemical insecticides resulting only in 20.60 per cent reduction in colony size when observed after 7 days of treatment.

Amongst different biopesticides, fish oil at its higher concentration (1.0 %) resulted in 30.30 per cent reduction in colony size which was at par with higher concentrations of all other biopesticides except neem oil. Neem oil, at its lower concentration (1.0 %), resulted in 20.10 per cent reduction in colony which was at par with the lower concentrations of all other biopesticides after 7 days of treatment.

Among earlier recommended insecticides, thiamethoxam (0.025 %) resulted in 54.14 per cent reduction in colony size which was at par with flonicamid (0.05 %) but was statistically superior over spirotetramat (0.015 %) where 48.62 per cent reduction in colony size was recorded after 7 days of treatment. Chlorpyrifos (0.04 %) after 7 days of treatment resulted in 61.50 per cent reduction in colony size and proved to be superior among all the new treatments as well as thiamethoxam.

After 14 days of treatment, spirotetramat (0.015 %) resulted in 63.96 per cent reduction in colony size which was at par with flonicamid (0.05 %) where per cent reduction in colony size was recorded to be 59.54. Buprofezin (0.025 %) resulted in 28.26 per cent reduction in colony size and was at par with flupyradifurone (0.017 %) where 31.94 per cent reduction in colony size was recorded after 14 days of treatment.

Among biopesticides, fish oil at its higher concentration (1.0 %) resulted in 38.27 per cent reduction in colony size which was at par with the higher concentrations of all other biopesticides except neem oil. Whereas, neem oil at its lower concentration (1.0 %) resulted in 24.56 per cent reduction in colony size after 14 days of treatment which was at par with its higher concentration (2.0 %) of neem oil as well as lower concentration (0.5 %) of fish oil.

Among earlier recommended insecticides, thiamethoxam (0.025 %) and chlorpyrifos (0.04 %) resulted in 74.63 and 71.86 per cent reduction in colony size, respectively, and proved statistically superior over all the test treatments.

After 21 days of treatment, spirotetramat (0.015 %) was most effective treatment resulting in 78.26 per cent reduction in colony size which was statistically superior over flonicamid (0.05 %) where 66.75 per cent reduction in colony size was recorded. The least effective treatment among chemical insecticide was the buprofezin which resulted in 31.23 per cent and 34.00 per cent reduction in colony size at 0.025 and 0.05 per cent concentrations, respectively.

Among biopesticides, *B. bassiana* (2×10^{10} cfus/L) resulted in 41.20 per cent reduction in colony size which was at par with its lower concentration (1×10^{10} cfus/L) as well as higher concentration (0.02 %) of azadirachtin where 37.30 and 39.30 per cent reduction in colony size was recorded, respectively, after 21 days of treatment. The least effective biopesticides were fish oil (0.5 %) and neem oil (1.0 %) where per cent reduction in colony size was 25.26 and 28.33 per cent, respectively, after 21 days of treatment.

Among earlier recommended insecticides, thiamethoxam (0.025 %) and chlorpyrifos (0.04 %) resulted in 81.40 and 81.20 per cent reduction in colony size, respectively, both being at par with spirotetramat (0.015 %). Whereas, rest of the treatments were found to be statistically inferior to the earlier recommended insecticides.

Data recorded after 28 days of treatment revealed that per cent reduction in colony size increased to 86.00 per cent in case of spirotetramat (0.015 %) which again proved to be best treatment followed by flonicamid (0.05 %) where 70.60 per cent reduction in colony size was observed. Buprofezin at its lower concentration (0.025%) proved to be least effective treatment where per cent reduction in colony size slightly increased to 33.66 per cent when observed after 28 days after treatment.

Among biopesticides, azadirachtin (0.02 %) resulted in 45.66 per cent reduction in colony size and was at par with *B. bassiana* (2×10^{10} cfus/L). The least effective biopesticide was fish oil (0.5 %) which resulted in only 25.26 per cent reduction in colony size after 28 days of treatment.

Table 4. Bioefficacy of insecticides and biopesticides against woolly apple aphid, *E. lanigerum* infesting apple (based on colony size)

Treatment	Concentration (%)	Per cent reduction in colony size over pre-count (DAT)					Mean
		3	7	14	21	28	
Spirotetramat	0.007	20.25 (26.72)	32.95 (35.00)	34.93 (36.20)	40.40 (39.44)	43.53 (41.26)	34.41 (35.72)
	0.015	28.36 (32.15)	48.62 (44.19)	63.96 (53.15)	78.26 (62.21)	86.00 (68.00)	61.04 (51.94)
Flupyradifurone	0.017	24.61 (29.70)	27.83 (31.80)	31.94 (34.39)	35.22 (36.38)	45.29 (42.27)	32.98 (34.91)
	0.03	26.42 (30.89)	32.87 (34.96)	38.31 (38.20)	40.70 (39.62)	48.34 (44.03)	37.33 (37.54)
Flonicamid	0.025	16.32 (23.75)	26.52 (30.95)	36.32 (37.03)	41.42 (40.03)	50.53 (45.28)	34.22 (35.41)
	0.05	33.24 (35.17)	49.67 (44.79)	59.54 (50.49)	66.75 (54.78)	70.60 (57.16)	55.96 (48.48)
Buprofezin	0.025	15.16 (22.90)	20.60 (26.97)	28.26 (32.09)	31.23 (33.90)	33.66 (35.44)	25.78 (30.26)
	0.05	19.50 (26.19)	26.43 (30.92)	30.50 (33.45)	34.00 (35.64)	40.60 (39.55)	30.20 (33.15)
<i>B. bassiana</i>	1x10 ¹⁰ cfus/lt.	18.23 (25.21)	20.20 (26.62)	30.16 (33.28)	37.30 (37.62)	39.63 (38.99)	29.10 (32.35)
	2x10 ¹⁰ cfus/lt.	22.46 (28.25)	25.60 (30.35)	35.46 (36.52)	41.20 (39.90)	44.81 (42.00)	33.91 (35.40)
Fish oil	0.5	15.40 (23.03)	20.50 (26.83)	25.26 (30.13)	25.26 (30.13)	25.26 (30.13)	22.34 (28.05)
	1.0	21.55 (27.60)	30.30 (33.36)	38.27 (38.19)	31.10 (33.85)	31.10 (33.85)	30.46 (33.37)
Neem oil	1.0	16.46 (23.90)	20.10 (26.55)	24.56 (29.68)	28.33 (32.12)	30.43 (33.45)	23.98 (29.14)
	2.0	20.33 (26.68)	25.23 (30.13)	28.13 (32.01)	32.40 (34.67)	35.46 (36.52)	28.31 (32.00)
Azadirachtin	0.01	20.58 (26.95)	24.36 (29.55)	30.49 (33.47)	35.50 (36.54)	37.43 (37.69)	29.67 (32.84)
	0.02	25.43 (30.23)	28.66 (32.33)	35.83 (36.74)	39.30 (38.79)	45.66 (42.49)	34.98 (36.12)
Thiamethoxam	0.025	29.70 (32.97)	54.14 (47.40)	74.63 (59.77)	81.40 (64.68)	86.06 (68.11)	65.18 (54.59)
Chlorpyrifos	0.04	40.96 (39.76)	61.50 (51.67)	71.86 (57.96)	81.20 (64.39)	86.33 (68.48)	68.37 (56.45)
Control	-	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
Mean		21.84 (26.95)	30.32 (32.33)	37.81 (36.99)	42.15 (39.72)	46.35 (42.35)	
CD_{0.05} Treatment (T) : (1.41) Days (D) : (0.72) T x D : (3.15) DAT (Days After Treatment)							

*Figures in parentheses are arcsine transformed values

Among earlier recommended insecticides, per cent reduction in colony size slightly increased to 86.06 per cent and 86.33 per cent in the case of thiamethoxam (0.025 %) and chlorpyrifos (0.04 %), respectively. Both of the earlier recommended insecticides were at par with each other and also with spirotetramat (0.015 %). When compared with the other treatments, earlier recommended insecticides as well as spirotetramat (0.015 %) proved to be significantly superior.

When per cent mean reduction in colony size obtained from different treatments was compared, it was concluded that spirotetramat at higher concentration (0.015 %) caused maximum reduction i.e. 61.04 per cent in colony size and was significantly superior over other test products. The next best treatment was of flonicamid (0.05 %) which resulted in 55.96 per cent reduction in colony size. However none of the test treatments proved better than the standard treatments (thiamethoxam and chlorpyrifos) where, 65.18 and 68.37 per cent mean reduction in colony size was recorded respectively.

Chlorpyrifos (0.04 %) resulted in 51.80 and 40.96 per cent reduction in colony count and colony size, respectively, just after 3 days of treatment which was comparatively higher than the other test products as well as thiamethoxam (0.025 %) which probably is due to the fact that, chlorpyrifos is a contact insecticide which resulted in immediate mortality of the pest whereas most of the test insecticides and earlier recommended thiamethoxam are systemic in nature which become effective after entering into the plant sap, thereby taking more time in reducing the population.

Our results are in line with the findings of Nauen *et al.* (2008), who found that when apple seedlings infested with woolly apple aphid were treated with spirotetramat (20-100 µg a. i. per leaf) resulted in 70-90 per cent reduction in woolly apple aphid population after 28 days of application.

Reduction in colony count was observed till last day of observation in an increasing trend, which indicates that spirotetramat persisted till 28th day after spray and caused significant reduction in colony count as also recorded by Kumar *et al.* (2009) where foliar application of spirotetramat against cotton white fly, *B. tabaci* revealed that spirotetramat @ 75 gm a.i. /ha persisted up to 25 days. Long lasting effect of spirotetramat is also reported by Pasqualini and Scannavini (2015) against *M. persicae*.

Results also find support from Misra (2009), who reported upto 90.30 per cent mortality in the population of brown plant hopper, *Nilaparvata lugens* with flonicamid, though the crop and pest are different but damage is caused by sucking of sap.

Our findings however differ from that of Patel *et al.* (2010), who observed up to 95 per cent mortality in the cotton mealy bug *P. solenopsis* population with buprofezin. probably due to the period of application of the chemical. Findings also differs from Jat and Jayekumar (2006) who recorded up to 39.7 to 72.6 per cent reduction in whitefly population with *B. bassiana*. Chand *et al.* (2010) also recorded 84.18 per cent reduction in *L. erysimi* population with *B. bassiana*, when applied @ 3.0 g/L of water. This is probably due to the fact that WAA is covered with waxy strands and hence the fungus could not pierce the barrier efficiently.

Fish oil at both the concentrations was found to have less persistence, as per cent reduction in colony count was stabilized after fourteen days of spray in case of both the concentrations. However in a study carried out by Singh and Rao (1979) in Karnataka, India, fish oil insecticidal rosin soap (4.0 %) effectively controlled soft green scale, *Coccus viridis* (Green) on citrus. Similarly, Karuppuchamy *et al.* (1998) also reported fish oil rosin soap (2.5 %) highly effective in controlling the pomegranate aphid, *Aphis punicae* population.

The present findings are partially supported by Jat and Jeyakumar (2006) who recorded up to 50 per cent reduction in whitefly population with neem oil on cotton. Anitha and Nandihalli (2008) also reported that neem oil (2%) was found effective in controlling the leafhopper, *A. bigutulla bigutulla* population (1.41 and 4.11 leafhopper/ 3 leaves) after 15 days of first and second spray, respectively.

Both of the neem products viz. neem oil and azadirachtin were not able to provide desirable level of control against woolly apple aphid population. Findings are supported by Caldwell *et al.* (2013), who stated that effectiveness of insecticides with semi systemic activity, such as azadirachtin or neem oil based products is usually insufficient against woolly apple aphid. The reasons for this could be a naturally occurring dilution of the active substance(s) in plant tissues caused by the growth of the plant, and/or differences in the concentration of active substance(s) among the test products, which are obtained by using different extraction and manufacturing processes.

4.3 Toxicity of test insecticides and biopesticides to natural enemies

4.3.1 Effect on *A. mali*

For observing effect of test insecticides and biopesticides on *A. mali*, parasitized aphids were collected from field 3, 7 and 14 days after foliar application on experimental apple trees and were brought to laboratory for observing per cent emergence of adult parasitoids. The data (Table 5) revealed that, a particular trend in adult parasitoid emergence was recorded where moderate emergence of parasitoid was recorded from the samples collected 3 days after treatment followed by low emergence from the mummified aphids collected after 7 days of treatment. Higher emergence was recorded from the parasitized aphids collected after 14 days of treatment.

However, in case of fish oil, neem oil and chlorpyrifos emergence of adult parasitoid increased from the mummified aphids collected after 7 days of treatment in comparison to 3 days after treatment, which further recorded slight increase in the samples of 14 days after treatment.

The parasitized aphids were collected from the field in the month of October and were put under observation till the month of March. Emergence of adult parasitoids was noticed till last week of November and thereafter in December and January no adult emergence was recorded which again resumed in the month of February. This is due to the fact that parasitoid overwintered during winter months and resumed its activity with the warming of the season as reported by Gupta *et al.* (2007).

Among new insecticides, lower concentration (0.025 %) of flonicamid proved to be the safest insecticide to the *Aphelinus mali* and resulted in 75.00, 60.00 and 80.00 per cent emergence of adult parasitoids from the parasitized aphids collected after 3, 7 and 14 days of treatment respectively. The second best treatment was flupyradifurone (0.017%), which resulted in 65.00, 60.00 and 65.00 per cent emergence of adult parasitoids respectively from the three collections of parasitized aphids as shown in Table 5. Buprofezin at higher concentration (0.05 %) resulted only in 45.00, 40.00 and 50.00 per cent emergence of adult parasitoids from first, second and third collection of parasitized aphids, respectively, indicating thereby moderate safety to the parasitoid.

Table 5. Toxicity of insecticides and biopesticides on aphelinid parasitoid, *Aphelinus mali* parasitizing woolly apple aphid

Treatment	Concentration (%)	Per cent emergence of parasitoids from parasitized aphids collected from field at different interval (DAT)		
		3	7	14
Spirotetramat	0.007	60.00 (50.83)	55.00 (47.86)	65.00 (53.74)
	0.015	55.00 (47.86)	50.00 (44.98)	60.00 (50.76)
Flupyradifurone	0.017	65.00 (53.74)	60.00 (50.83)	65.00 (53.84)
	0.03	55.00 (47.89)	55.00 (47.89)	60.00 (50.83)
Flonicamid	0.025	75.00 (60.29)	60.00 (50.83)	80.00 (63.90)
	0.05	60.00 (50.83)	55.00 (47.89)	65.00 (53.74)
Buprofezin	0.025	55.00 (47.89)	50.00 (44.98)	60.00 (50.76)
	0.05	45.00 (42.07)	40.00 (39.19)	50.00 (44.98)
<i>B. bassiana</i>	1x10 ¹⁰ cfus/lt.	60.00 (50.83)	55.00 (47.86)	65.00 (53.74)
	2x10 ¹⁰ cfus/lt.	55.00 (47.89)	50.00 (44.98)	65.00 (53.84)
Fish oil	0.5	60.00 (50.83)	70.00 (56.97)	75.00 (60.05)
	1.0	55.00 (47.89)	70.00 (56.81)	70.00 (56.81)
Neem oil	1.0	60.00 (50.83)	75.00 (60.05)	80.00 (63.90)
	2.0	55.00 (47.89)	70.00 (56.97)	75.00 (60.29)
Azadirachtin	0.01	60.00 (50.74)	55.00 (47.86)	60.00 (50.76)
	0.02	55.00 (47.89)	50.00 (44.98)	60.00 (50.76)
Thiamethoxam	0.025	55.00 (47.86)	50.00 (44.98)	60.00 (50.83)
Chlorpyrifos	0.04	15.00 (22.58)	20.00 (26.44)	20.00 (26.44)
Control	-	90.00 (71.92)	95.00 (77.04)	90.00 (71.92)
CD_{0.05}		(8.19)	(7.59)	(8.08)

*Figures in parentheses are arcsine transformed values
DAT (Days After Treatment)

Among biopesticides, neem oil (1.0 %) was safest to the parasitoid and resulted in 60.00, 75.00 and 80.00 per cent emergence of adult parasitoids, respectively, from first, second and third collection. This treatment was followed by fish oil (0.5 %) which resulted in 60.00, 70.00 and 75.00 per cent emergence of parasitoids from the first, second and third collections of parasitized aphids, respectively. Azadirachtin (0.02 %) proved to be most toxic among biopesticides to *A. mali* resulting in 55.00, 50.00 and 60.00 per cent emergence of adult parasitoids from the three collections of parasitized aphids, respectively. This treatment was at



Plate 6. Parasitized aphids with exit hole



Plate 7. Adult parasitoids emerged from the field collected parasitized aphids

par with higher concentration (2×10^{10} cfus/L) of *B. bassiana* which resulted in 56.66 per cent mean parasitoid emergence.

Overall, lower concentration (0.025 %) of flonicamid with maximum adult emergence proved to be the safest treatment followed by lower concentration (1.0 %) of neem oil. When compared to the standard treatments, all the test insecticides and biopesticides were safer as compared with chlorpyrifos (0.04 %) which resulted in 15.00, 20.00 and 20.00 per cent emergence only, of adult parasitoids, respectively, from first, second and third collection. Whereas another standard treatment thiamethoxam (0.025 %) resulted in 55.00, 50.00 and 60.00 per cent emergence, respectively, from first, second and third collection and was comparatively safer than chlorpyrifos as well as higher concentration (0.05 %) of buprofezin.

The results of the present study are in line with Gupta *et al.* (2013), who recorded 59.70 per cent emergence of *A. mali* adults in thiamethoxam (0.025 %) from the parasitized aphids collected 5 days after the spray. Further in chlorpyrifos (0.04 %), only 12.5 per cent adult emergence was recorded, which again corroborate the findings of the present study where adult emergence of parasitoid recorded was 15.00, 20.00 and 20.00 per cent from the parasitized aphid samples, collected after 3, 7 and 14 days of chlorpyrifos (0.04 %) spray, respectively.

Toxicity of chlorpyrifos to *A. mali* has also been reported by Khajuria *et al.* (2010) from Kullu, H.P. which again corroborate the current findings.

Our findings are in contrast to Frewin *et al.* (2012), who reported flonicamid toxic in comparison to *B. bassiana* and spirotetramat against *A. certus*, a parasitoid of soybean aphid.

Cohen *et al.* (1996) reported chlorpyrifos highly toxic to adult wasps of *A. mali* and safe to immature stages of the parasitoid which is contrary to the results obtained in present study, where only 15.00-20.00 per cent adult emergence was recorded from mummified aphids, revealing thereby its toxicity to immature stages of parasitoid.

4.3.2 Effect on coccinellids

The evaluation of safety of different treatments to coccinellids namely *Coccinella septempunctata* and *Hippodamia variegata* was done in the laboratory by following residual film method. The data so gathered are presented in Tables 6 and 7.

C. septempunctata

The data (Table 6) revealed that among various chemical insecticides, spirotetramat at lower concentration (0.007 %) was safest to the adults of *C. septempunctata* causing only 11.66, 15.00 and 15.00 per cent mortality after 24, 48 and 72 hours of treatment, respectively. Lower concentration (0.025 %) of buprofezin resulted in 11.11, 15.00 and 16.66 per cent mortality after 24, 48 and 72 hours of treatment, respectively, and was statistically at par with spirotetramat (0.007 %).

The next safest insecticide was flonicamid at its lower concentration (0.025%) which resulted in 15.00, 20.00 and 23.33 per cent mortality and was followed by higher concentration (0.05%) of buprofezin which resulted in 15.00, 20.00 and 23.33 per cent mortality when recorded after 24, 48 and 72 hours of treatment, respectively. Flupyradifurone at higher concentration (0.03%) resulted in 26.66, 30.00 and 31.66 per cent mortality of *C. septempunctata* after 24, 48 and 72 hours of treatment, respectively, and was statistically at par with lower concentration (0.017 %) of flupyradifurone and higher concentration (0.015 %) of spirotetramat as shown in Table 6.

Among biopesticides, fish oil at both the concentrations (0.5 and 1.0 %) proved the safest treatment causing no mortality and was at par with the control treatment where in only water was sprayed. Neem oil (1.0 %) with 10.00, 15.00 and 15.00 per cent mortality, and azadirachtin (0.01 %) with 8.33, 15.00 and 18.33 per cent mortality after 24, 48 and 72 hours, respectively, were next in order of safety to predator.

Azadirachtin (0.02 %) resulted in 20.00, 23.33 and 26.66 per cent mortality of *C. septempunctata* and was statistically at par with neem oil (2.0 %) which resulted in 16.66, 23.33 and 25.00 per cent mortality, when recorded 24, 48 and 72 hours after treatment, respectively.

Overall, among all the test treatments, fish oil (0.5 and 1.0 %) proved to be the safest with nil mortality followed by neem oil (1.0 %), which was statistically at par with azadirachtin (0.01 %), spirotetramat (0.007%) and buprofezin (0.025 %).

Table 6. Toxicity of the insecticides and biopesticides on coccinellid predator, *Coccinella septempunctata*

Treatment	Concentration (%)	Per cent mortality of <i>C. septempunctata</i> adults (HAT)		
		24	48	72
Spirotetramat	0.007	11.66 (19.87)	15.00 (22.77)	15.00 (22.77)
	0.015	23.33 (28.76)	26.66 (31.05)	30.00 (33.19)
Flupyradifurone	0.017	23.33 (28.76)	26.66 (31.05)	30.00 (33.19)
	0.03	26.66 (30.98)	30.00 (33.19)	31.66 (34.21)
Flonicamid	0.025	15.00 (22.58)	20.00 (26.55)	23.33 (28.84)
	0.05	20.00 (26.55)	25.00 (29.98)	26.66 (31.05)
Buprofezin	0.025	11.66 (19.87)	15.00 (22.77)	16.66 (24.03)
	0.05	15.00 (22.58)	20.00 (26.55)	23.33 (28.84)
<i>B. bassiana</i>	1x10 ¹⁰ cfus/lit.	10.00 (14.99)	16.66 (24.03)	20.00 (26.55)
	2x10 ¹⁰ cfus/lit.	13.33 (17.70)	20.00 (26.55)	20.00 (26.55)
Fish oil	0.5	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
	1.0	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
Neem oil	1.0	10.00 (18.42)	15.00 (22.77)	15.00 (22.77)
	2.0	16.66 (24.03)	23.33 (28.84)	25.00 (29.98)
Azadirachtin	0.01	8.33 (13.73)	15.00 (22.77)	18.33 (25.29)
	0.02	20.00 (26.55)	23.33 (28.84)	26.66 (31.05)
Thiamethoxam	0.025	26.66 (30.98)	30.00 (33.19)	33.33 (35.23)
Chlorpyrifos	0.04	86.66 (68.82)	100.00 (89.39)	100.00 (89.39)
Control	-	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
CD_(0.05)	-	(10.05)	(1.68)	(2.10)

*Figures in parentheses are arcsine transformed values
HAT (Hours After Treatment)

Flupyradifurone resulted in 26.66, 30.00 and 31.66 per cent mortality to *C. septempunctata* at higher concentration (0.03 %) and 23.33, 26.66 and 30.00 per cent mortality at lower concentration (0.017 %) after 24, 48 and 72 hours, respectively. Both the concentrations

were statistically at par with one another and also with higher concentration (0.015 %) of spirotetramat (Table 6).

All the treatments except fish oil proved toxic as compared to control where nil mortality was recorded. When compared with standard treatments, thiamethoxam where 30.00 per cent mean mortality was recorded was statistically at par with both the concentrations of flupyradifurone and higher concentration of spirotetramat. Rest of the treatments were significantly inferior to the thiamethoxam (0.025 %) as far as safety to predators is concerned. In comparison to the recommended chlorpyrifos (0.04 %), all the treatments were found safe as, cent per cent mortality of adults was recorded in chlorpyrifos (0.04 %) treated Petri plates.

It was revealed that in the first 24 hours, the mortality in all the treatments recorded was higher which later increased gradually, thereby indicating that the toxicity of test insecticides is more initially.

H. variegata

Among test insecticides, buprofezin proved safest treatment causing 20.00, 26.66 and 30.00 per cent mortality @ 0.025 per cent and was at par with higher concentration (0.05 %) which resulted in 23.33, 28.33 and 31.66 per cent mortality after 24, 48 and 72 hours of treatment, respectively. Higher concentration (0.03 %) of flupyradifurone proved to be the most toxic insecticide resulting in 55.00, 61.66 and 65.00 per cent mortality, respectively, after 24, 48 and 72 hours as shown in Table 7.

Among different biopesticides, fish oil proved to be the safest causing only upto 10.00 and 15.00 per cent mortality at lower and higher concentrations, respectively, when recorded after 72 hours of treatment. This treatment was followed by lower concentrations of azadirachtin and neem oil where 15.00, 18.33, 23.33 and 18.33, 20.00, 23.33 per cent mortality was recorded after 24, 48 and 72 hours of treatment, respectively. The higher concentration of *B. bassiana* proved to be the most toxic biopesticide to *H. variegata* causing 30.00, 35.00 and 38.33 per cent mortality followed by lower concentration of the same biopesticide where 26.66, 30.00 and 35.00 per cent mortality was recorded after 24, 48 and 72 hours of treatment.

Table 7. Toxicity of the insecticides and biopesticides on coccinellid predator, *Hippodamia variegata*

Treatment	Concentration (%)	Per cent mortality of <i>H. variegata</i> adults (HAT)		
		24	48	72
Spirotetramat	0.007	28.33 (32.12)	36.66 (37.24)	40.00 (39.21)
	0.015	45.00 (42.10)	50.00 (44.98)	53.33 (46.89)
Flupyradifurone	0.017	51.66 (45.93)	55.00 (47.86)	56.66 (48.81)
	0.03	55.00 (47.86)	61.66 (51.73)	65.00 (53.70)
Flonicamid	0.025	40.00 (39.21)	46.66 (43.07)	50.00 (44.98)
	0.05	50.00 (44.98)	55.00 (47.86)	56.66 (48.81)
Buprofezin	0.025	20.00 (26.55)	26.66 (31.05)	30.00 (33.19)
	0.05	23.33 (28.84)	28.33 (32.12)	31.66 (34.21)
<i>B. bassiana</i>	1x10 ¹⁰ cfus/lt.	26.66 (31.05)	30.00 (33.19)	35.00 (36.25)
	2x10 ¹⁰ cfus/lt.	30.00 (33.19)	35.00 (36.25)	38.33 (38.22)
Fish oil	0.5	0.00 (0.00)	10.00 (18.42)	10.00 (18.42)
	1.0	0.00 (0.00)	11.66 (19.87)	15.00 (22.77)
Neem oil	1.0	18.33 (25.29)	20.00 (26.55)	23.33 (28.84)
	2.0.	21.66 (27.69)	25.00 (29.98)	30.00 (33.19)
Azadirachtin	0.01	15.00 (22.77)	18.33 (25.29)	23.33 (28.84)
	0.02	23.33 (28.84)	28.33 (32.12)	33.33 (35.23)
Thiamethoxam	0.025	53.33 (46.90)	70.00 (56.76)	73.33 (58.90)
Chlorpyrifos	0.04	100.00 (89.39)	100.00 (89.39)	100.00 (89.39)
Control	-	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
CD_{0.05}		(2.88)	(2.59)	(2.04)

*Figures in parentheses are arcsine transformed values
HAT (Hours After Treatment)

Overall, among all the treatments including insecticides and biopesticides, fish oil proved to be the safest treatment causing only 10.00 and 15.00 per cent mortality at lower and higher concentrations, respectively, when recorded after 72 hours. Whereas flupyradifurone at higher concentration proved to be the most toxic treatment causing upto 65.00 per cent mortality in *H. variegata* population after 72 hours of treatment.

The standard treatments namely thiamethoxam (0.025 %) and chlorpyrifos (0.04 %) resulted in 73.33 and 100.00 per cent mortality of *H. variegata* adults after 72 hours of treatment, respectively. These two standard treatments were more toxic to the coccinellid in comparison to the test insecticides used in the present study.

H. variegata was more sensitive to the test treatments in comparison to *C. septempunctata*; fish oil was safest with nil mortality after 24 hours of treatment which later increased to 10.00 per cent in next 48 hours. Whereas in *C. septempunctata*, no mortality was recorded. Similar pattern was recorded in other treatments also, indicating high sensitivity of *H. variegata*.

Fonicamid (0.025 %) resulted in 15.00, 20.00 and 23.33 per cent mortality of *C. septempunctata* adults 24, 48 and 72 hours after the treatment, respectively, and was comparatively safe to the predator. Jalali *et al.* (2009) while evaluating toxicity of insecticides against two spotted lady bird, *Adalia bipunctata* also found fonicamid the safest with no lethal effects on larvae and female adults, thus corroborating the present findings.

Singh and Gupta (2016) evaluated some insecticides and biopesticides against *C. septempunctata*, previously fed on *Brachycaudus helichrysi*, in the laboratory by using residue film method. They observed neem oil (1.0 %) as safe to the coccinellid as only 2.5 per cent mortality was recorded. The neonicotinoid thiamethoxam (0.025 %) was moderately toxic with 25.00 per cent mortality of adults. These results are partially in line with the findings of the present study. Maximum mortality occurred within 24 hours of treatment in the present study which find support from Singh and Gupta (2016) who also recorded maximum mortality of *C. septempunctata* adults within 24 hours.

Chapter- 5

SUMMARY AND CONCLUSIONS

The present investigations entitled “**Management of woolly apple aphid, *Eriosoma lanigerum* (Hausmann) using new insecticide molecules and biopesticides**” was divided into two parts i.e. testing the efficacy of new group insecticide molecules against woolly apple aphid and to study the effect of tested products on the natural enemies of woolly apple aphid. The first part of the research was conducted at farmer’s field in Matiana area of Shimla district during October-November 2016 and second part was conducted under laboratory conditions in Dr. Y S Parmar University of Horticulture and Forestry Naini, Solan. The results obtained are summarized as below:

- Spirotetramat (0.015 %) was the best among the test insecticide treatments resulting in 68.48 per cent reduction in colony count of woolly apple aphid and was followed by flonicamid (0.05 %) which resulted in 61.31 per cent reduction in colony count.
- When the data on colony size were recorded, it followed the same trend i.e. spirotetramat (0.015 %) proved most efficacious causing 61.04 per cent reduction in aphid colony size followed by flonicamid (0.05 %) with respective figure of 55.96 per cent.
- Among the test biopesticides, the higher concentration of each i.e. *B. bassiana* (2×10^{10} cfus/L), fish oil (1.0 %), neem oil (2.0 %), azadirachtin (0.02 %), resulted in 29.17, 31.32, 28.95 and 29.83 per cent reduction in colony count, respectively, and were at par. Similar trend was recorded in colony size.
- When the safety to natural enemies of *E. lanigerum* was evaluated, neem oil (1.0 %) and flonicamid (0.025 %) proved safest to the parasitoid, *A. mali* with 60.00-80.00 per cent and 75.00-80.00 per cent adult emergence, respectively.
- Spirotetramat (0.015 %) and flonicamid (0.05 %) were also moderately safe (55.00-60.00 and 60.00-65.00 % adult emergence, respectively) and were at par with the earlier recommended insecticide, thiamethoxam (0.025 %) where adult emergence recorded was 55.00-60.00 per cent. Chlorpyrifos proved most toxic to *A. mali* where only 15.00-20.00 per cent adult emergence was recorded. Rest of the treatments were moderately safe as far as safety to *A. mali* is concerned.

- Fish oil proved safest to *C. septempunctata* adults with no mortality, when recorded in laboratory, even after 72 hours of treatment. Rest of the test treatments were moderately safe (13.33 to 29.69 per cent mortality).
- In case of *H. variegata*, fish oil again proved safest among the test insecticide/biopesticide treatments with only 10.00 and 15.00 per cent mortality after 72 hours at lower (0.5 %) and higher (1.0 %) concentrations respectively. The earlier recommended insecticides thiamethoxam (0.025 %) with 73.33 per cent mortality and chlorpyrifos (0.04 %) with 100 per cent mortality were toxic to *H. variegata* adults.
- Buprofezin was the safest among new test insecticide molecules (30.00 and 31.66 per cent mortality) in comparison to 40 to 65 per cent mortality in rest of the test insecticide treatments.

On the basis of the above study it is concluded that spirotetramat (0.015 %) and flonicamid (0.05 %) proved superior among the test insecticides evaluated in this study. Though the earlier recommended insecticides namely thiamethoxam and chlorpyrifos were more effective but they were found toxic to the predators and parasitoid in comparison to the spirotetramat and flonicamid. The biopesticides did not prove much effective in managing *E. lanigerum* but were found comparatively safe to natural enemies. Hence these new molecules can be further evaluated for incorporation in the management programme.

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Name of the student : Vineet Kumar
Admission Number : H-2015-14-M
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ABSTRACT

The bioefficacy study revealed that, spirotetramat (0.015 %) proved best among the test insecticide treatments resulting in 68.48 per cent reduction in colony count of woolly apple aphid and was followed by flonicamid (0.05 %) which resulted in 61.31 per cent reduction in colony count. When the data on colony size were recorded, it followed the same trend i.e. spirotetramat (0.015 %) proved most efficacious causing 61.04 per cent reduction in aphid colony size followed by flonicamid (0.05 %) with a reduction of 55.96 per cent. The higher concentration of each test biopesticides i.e. *B. bassiana* (2×10^{10} cfus/L), fish oil (1.0 %), neem oil (2.0 %) and azadirachtin (0.02 %) resulted in 29.17, 31.32, 28.95 and 29.83 per cent reduction in colony count being at par. Similar trend was recorded in colony size. When the safety to natural enemies of *E. lanigerum* was noticed, neem oil (1.0 %) and flonicamid (0.025 %) proved safest to the parasitoid, *A. mali* with 60.00-80.00 per cent and 75.00-80.00 per cent adult emergence, respectively. Spirotetramat (0.015 %) and flonicamid (0.05 %) were moderately safe with 55.00-60.00 and 60.00-65.00 per cent adult emergence, respectively, and were at par with the earlier recommended insecticide, thiamethoxam (0.025 %) where adult emergence was 55.00-60.00 per cent. Chlorpyrifos proved most toxic to *A. mali* where only 15.00-20.00 per cent adult emergence was recorded. In case of *C. septempunctata* and *H. variegata*, fish oil at both the concentrations with no or very low mortality, even after 72 hours of treatment proved safest followed by buprofezin; whereas, chlorpyrifos (0.04 %) was most toxic to the coccinellid species causing upto 100 per cent mortality. *H. variegata* was more sensitive to all the test insecticides and biopesticides as compared to *C. septempunctata*.

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APPENDIX-I
Analysis of variance (ANOVA)

Table 3. Bioefficacy of insecticides and biopesticides against woolly apple aphid, *E. lanigerum* infesting apple (based on colony count)

Source of Variation	DF	Sum of Squares	Mean Squares	F-Calculated	Significance
Replication	2	51.418			
Factor A	18	53,839.938	2,991.108	506.404	0.00000
Factor B	4	10,520.856	2,630.214	445.304	0.00000
Interaction A X B	72	2,765.441	38.409	6.503	0.00000
Error	188	1,110.433	5.907		
Total	284	68,288.086			

Table 4. Bioefficacy of insecticides and biopesticides against woolly apple aphid, *E. lanigerum* infesting apple (based on colony size)

Source of Variation	DF	Sum of Squares	Mean Squares	F-Calculated	Significance
Replication	2	184.048			
Factor A	18	40,044.147	2,224.675	580.043	0.00000
Factor B	4	8,546.828	2,136.707	557.107	0.00000
Interaction A X B	72	3,358.262	46.643	12.161	0.00000
Error	188	721.048	3.835		
Total	284	52,854.333			

Table 5. Toxicity of insecticides and biopesticides on aphelinid parasitoid, *Aphelinus mali* parasitizing woolly apple aphid

Source of Variation	DF	MS		
		3 DAT	7 DAT	14 DAT
Replication	2			
Treatment	18	241.31	298.70	250.60
Error	36	24.28	20.85	23.62
Total	56			

Table 6. Toxicity of the insecticides and biopesticides on coccinellid predator, *Coccinella septempunctata*

Source of Variation	DF	MS		
		After 24 hrs	After 48 hrs	After 72 hrs
Treatment	18	690.81	1046.24	1061.25
Error	38	35.67	1.025	1.60
Total	56			

Table 7. Toxicity of the insecticides and biopesticides on coccinellid predator, *Hippodamia variegata*

Source of Variation	DF	MS		
		After 24 hrs	After 48 hrs	After 72 hrs
Treatment	18	1259.30	1014.88	994.80
Error	38	3.02	2.45	1.51
Total	56			

BRIEF BIO-DATA

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