DETERMINATION OF PHYTOTONICITY, PHYTOTOXICITY, SAFETY, AND RESIDUE OF BOOM FLOWER-n (NITROBENZENE 20% v/v) IN OKRA *Abelmoschus esculentus* (L.) MOENCH

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ABSTRACT

DETERMINATION OF PHYTOTONICITY, PHYTOTOXICITY, SAFETY, AND RESIDUE OF Boom Flower®-n (NITROBENZENE 20% v/v) IN OKRA *Abelmoschus esculentus* (*L.*) MOENCH

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Studies were conducted to evaluate the phytotonic and phytotoxic effects Boom Flower®-n (Nitrobenzene 20% v/v) in okra, its residues in harvestable produce, sensitivity to natural enemies and pollinators (honeybees) and non target organisms like fish and earthworms.

Field trials conducted with Boom Flower®-n revealed that @ 3 ml lit^{-1} showed superiority in all growth parameters like plant height, plant canopy (leaf production and leaf area), total chlorophyll content, and quality of fruits. Consequently these attributes inflicted in increasing the yield by 54 and 57 per cent in trial I and trial II respectively.

Natural enemies such as *Trichogramma chilonis* (Ishii), *Chelonus blackburni* (Cameron) and *Chrysoperla carnea* (Stephens), were found to be moderately sensitive to the tests conducted in the laboratory. Generally lower mortalities and higher parasitization / fecundity were observed at lower concentrations. However, Boom

Flower®-n was moderately toxic to the natural enemies. Nevertheless in the field, these parasitoids and predator activities were at lower ebb immediately after the application of Boom Flower®--n, but the population rebound with in a week time.

Likewise, Indian, Italian and dammer bees were found to be less sensitive at recommended dose as approximately a mortality of 30% was recorded. However, in the field, a reduction of honey bee visitation was observed immediately after the application of Boom Flower®--n. But, an increase in the frequency of honey bee visitation was observed with in a week.

The LC_{50} values of Boom Flower®-n for earthworm *Eudrilus eugeniae* (Kingberg) and *Perionyx excavatus* (Perrier) were 7.89 and 6.62 g kg^{-1} of soil respectively, while that for common carp *Cyprinus carp* (L.) and *Tilapia mosambica* (Peters), the LC_{50} values were 70.37 and 165.63 ppm, respectively.

The initial deposits of Boom Flower®-n applied @ 2, 3, 4 and 8 ml lit^{-1} were at 0.111 to 0.167, 0.222 to 0.334, 0.634 to 0.723 and 1.506 to 0.890 $\mu g\ g^{-1}$on okra fruits. In both the trails, Boom Flower-n dissipate from okra fruits with in 2 days at lower concentrations. However, at higher concentration, the rate of dissipation was rather slow and took nearly a week time to reach below detectable level.

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CHAPTER I
INTRODUCTION

Okra, *Abelmoschus esculentus* (L.) Moench, belonging to the family Malvaceae is grown as a vegetable crop in the tropics. In India, it is widely cultivated in Gujarat, Andhra Pradesh, Karnataka and Tamil Nadu. In Tamil Nadu, it is cultivated in an area of 4,750 hectares with an annual production of 47,500 tonnes of fruits and a productivity of 10 tonnes ha $^{-1}$ (Anonymous, 2000). The area under okra cultivation in India is 4.31 lakh hectares, and its production is 40.33 lakh tonnes with an average yield of 9.36 tonnes ha $^{-1}$ It is cultivated mainly for its immature fruits, which are rich in proteins, vitamins, calcium, potassium and other minerals. It is also used in paper industry and medicine.

Like any other malvaceous crop, insect pests right from germination to harvest ravage okra. Sucking pests like aphids, *Aphis gossypii* (Glov.), leaf hopper, *Amrasca biguttula biguttula* (Ishi.), thrips, *Thrips tabaci* (Lind.), whitefly, *Bemisia tabaci* (Genn.) and in the later stage fruit borers cause serious problems, resulting reduction in yield to a level of 69 per cent (Rawat and Suhu, 1973) and the quality of the fruits.

Farmers rely solely on the chemical insecticides for the management of pests of okra because of easy avilability, immediate and spectacular knock down effects. Despite these credentials, continuous use of chemical insecticides found to be ecologically unsafe and indiscriminate use of insecticides has resulted in accumulation of pesticide residues in fruits, resurgence of secondary pests, mortality of predators and parasitoids (Mitra *et al.*, 1999) and environmental pollution (Mahapatra and Gupta, 1998).

On the other side, maximization of yield in any crop is of prime concern, physiological efficiency of a crop should be improved to achieve the potential productivity and expression of the crop (Dhashora and Jain, 1994). This is possible by regulating the biosynthesis of macromolecules, such as starch and protein and by

manipulating source-sink relationship, role played by plant hormones in regulating physiological process of the plant at molecular and sub molecular levels.

Exogenous application of chemicals, either promotory or inhibitory nature might substitute some endogenous substances and activate the metabolic reactions. Application of plant growth regulators to improve crop growth is one of the modern techniques in agriculture. Plant growth regulators help in overcoming certain environmental limitations, such as improving germination percentage, growth of shoots, roots and finally yield as well as quality. But such chemicals should be safer to non target organisms with out leaving any harmful residues.

Boom Flower®–n is one such chemical gaining importance in South Asia as plant growth regulator, although a large quantity of the same is produced for industrial use. In agriculture, nitrobenzene is mostly applied along with pesticide as, it is compatible with many commonly available pesticides. (Lingappa *et al* ., 2001; Dandale, 2001; Reddy and Joshi, 1990; Srinivas and Clement Peter, 1993). Its residue though, not as an end product but as converted metabolite been detected in cotton, tomato and soil (Oleszek and Jurzysta, 1987). Environmental Protection Agency (EPA) has identified seven sites of risk out of the 1,777 sites of national priority lists of risk for nitrobenzene. This emphasizes the importance of nitrobenzene in human health and its effect on non target beneficial organisms. Direct contact of nitrobenzene may cause mild irritation to skin and eyes and repeated exposure may lead to methemoglobinemia, which affect the ability of blood to carry oxygen. There is a very little information available about the long term exposure of human and animals to nitrobenzene particularly against the non target organisms.

Given this background the present investigation were made on Boom Flower®-n (nitrobenzene20% v/v) with the following objectives

1. To evaluate the phytotonic and phytotoxic effects of Boom Flower®-n on okra.
2. To determine the sensitivity of certain natural enemies in okra ecosystem and certain non target organisms to Boom Flower® -n and
3. To estimate the residues of Boom Flower® -n from the harvestable produce of okra.

CHAPTER II

REVIEW OF LITERATURE

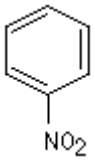
Okra is an important vegetable grown globally over a wide range of climatic conditions. It is one of the important vegetables used in south Indian cusine, having good sources of proteins, vitamins (A, B and C) and minerals such as iodine and calcium. Owing to its closeness to cotton, it is often infested by various groups of pests like sucking pests and borers. To minimise the tremendous loss caused by these pests, several insecticides are used in large quantities. This may led to the destruction of natural enemy fauna in the ecosystem, development of resistance, replacement of pests, resurgence and environmental pollution.

In the recent years apart from pesticides, plant growth regulators are increasingly used to overcome physiological constraints, leading to enhanced production in several crops (Malik *et al.*, 1990). Nitrobenzene is a phytotonic chemical, which is found to stimulate flower production in okra and there by increase the yield. The work done on various aspects about the phytotonic, phytotoxic effects, safety and residue are reviewed here.

2.1. Nitrobenzene

Nitrobenzene is an oily yellow liquid having an almond or shoe polish odour, with a reported threshold of 0.092 mg m^{-3} (0.018 ppm) (Amoore and Hautala, 1983). It dissolves slightly in water, but very easily in some organic solvents. It is a flowering stimulant cum yield booster for many crops.

2.1.1. Structure and chemistry

Common name	Nitrobenzene
Chemical formula	$C_6H_5NO_2$
Chemical structure	

Relative molecular mass	123.11
CAS name	Nitrobenzene
IUPAC name	Nitrobenzene
Synonyms	Nitrobenzol, mononitrobenzol, MNB, C.I. solvent black 6, essence of mirbane, essence of myrbane, mirbane oil, oil of mirbane, oil of myrbane, nigrosine spirit soluble B
Trade name	Boom Flower ®–n(Nitrobenzene 20%v/v)

2.1.2. Chemicals and physical properties of nitrobenzene

Specific gravity	1.2037 at 20°C
	1.205 at 28°C
Melting point	5.7°C
Boiling point	211°C
Vapour pressure	20 Pa (0.15 mmHg) at 20°C
	38 Pa (0.284 mmHg) at 25°C
	47 Pa (0.35 mmHg) at 30°C
Flash point (closed cup)	88°C
Explosive limit (lower)	1.8% by volume in air
Solubility in water	1900 mg l^{-1} at 20°C
	2090 mg l^{-1} at 25°C
Solubility in organic solvents	Freely soluble in ethanol, benzene, acetone, ether and oils
Octanol / water partition coefficient ($\log K_{ow}$)	1.85 (1.6–2.0)
Organic carbon/water partition coefficient ($\log K_{oc}$)	1.56
Henry's law constant (measured)	2.4 Pa m^3/mol (20 °C)
	0.868 Pa m^3/mol (25 °C)

2.2. Phytotonic effects

2.2.1. Phytotonic effects by plant growth regulators

Nitrophenol (ATONIK) is an aromatic nitro compound, which consists of sodium ortho-nitrophenol, sodium para-nitrophenol and sodium nitro-guaiacol as active ingredients. The presence of these nitro groups helps in the easy absorption of nitrophenol by the leaves.

Nitrophenol stimulates plant activity without causing malformation or toxicity to the plants and accelerates the plasma streaming of the cells (Datta *et al*., 1979). The physiological role of phenolic substances is still controversial. Some investigators argue against any role of phenols in plant growth regulation, because these compounds are localized in closed compartment of the cell such as the vacuole and hence remote from control of physiological processes. Other scientists consider that endogenous phenols possess only stimulatory properties and thus can promote growth due to auxin like properties of caffeic acid and ferreulic acid (Kafeli and Kutacek, 1977). Nitrophenol increased the yield, which may be due to increase in the endogenous auxin level by external application (Datta *et al*., 1986). Another reason for increased yield may be attributed to auxin stimulated ethylene production in the plant tissue (Pratt and Goeschl, 1969). Polyphenols like chlorogenic acid and tannic acid all induce floral buds as GA_3 in *Impatiens balsamina* (L.) under strictly non-inductive photoperiods. Monophenols (salicylic acid and β-naphthol) resembled Gibberellic acid (GA_3) in inducing floral buds (Nanda and Surinder Kumar, 1977). It was also inferred that there was a synergistic effect of these phenols with GA_3.

Brassinosteriod enhances cell division, cell elongation and cell differentiation (Mitchell and Gregory, 1972). It promotes DNA polymerase activity, RNA polymerase activity, replication, transcription and translation, however increases enzyme activity, protein pump action and regulates plant metabolism for improved growth.

Gibberellic acid prevents genetic and physiological dwarfism, breaking of dormancy, flowering induction in long day plants, increases amylase activity promotes cell elongation and influences root growth (Metzger, 1990).

Salicyclic acid is a secondary metabolite acting as analogues of growth regulating substances. It plays an important role in the initiation and development of floral buds and yield improvement in cereals (Datta and Nanda, 1985).

Benzyladenine is a synthetic cytokinine ($C_{10}H_9N_{50}$), which helps in promotion of cell division, delaying of senescence, contract apical bud dominance, flowering induction in short day plants, translocation of assimilates and thereby affecting the yield potential of plants. Cylokinin improves crop production through redirecting the metabolic balance of growth, retardation of yellowing leaves, increased photosynthetic carbon dioxide uptake and partitioning of assimilates (Biddington and Thomas, 1978; Van staden *et al.,* 1988; Brault and Maldiney, 1999).

2.2.1.1. Plant height

Plant height has been considered as one of the important morphological characters determining plant growth development. Application of gibberellic acid increased number of internode and length of one or more internode of full and dwarf strains of rice (Srivastava *et al.,* 1979). Kinetin application (10^{-4} M) significantly enhanced the shoot length of maize (Renu Dogra and Thukral, 1989).

Foliar spraying of brassinosteroid on rice seedling with 4^{th}, 5^{th} and 6^{th} leaves expanding stage promoted cell elongation and thereby increase in plant height (Fujii and Saka, 2001), height of the wheat plant was progressively increased by foliar application of gibberellie acid (100 ppm) (Hadolo *et al.,* 2002).

2.2.1.2. Number of leaves

Leaves are the most important functional unit of plant. Tri-iodo benzoic acid application increased the number of leaves of mid-winter and late-winter rice cultivars (Keith Moddy, 1986). Arora *et al.* (1990) reported an increase in the number of leaves due to cycocel treatment in okra.

Application of kinetin increased the number leaves in maize (Renu Dogra and Thukral, 1989). Late spraying of urea (1%) delayed leaf senescence, prolonged leaf survival and increased number of leaves in wheat. (Saad *et al.,*1990). Whereas wheat

grains treated with brassinolide (3 μ M for 8 or 12 h) showed an increase in leaf number (Hayat *et al.,* 2001). Kinetin (25 and 50 ppm) application increased flag leaf area of maize plant and delayed flag leaf senescence by reducing endogenous abscissic acid (Shehata *et al.,* 2001).

2.2.1.3. Leaf area index (LAI)

Leaf area index (LAI) is the leaf area per land area occupied by the plant. LAI is one of the important growth attributes that determines the dry matter production of a crop and yield. Gibberellic acid application significantly enhanced LAI of rice (Chatterjee *et al.,* 1976). Foliar spray of ethrel resulted increment in LAI in soybean (Harshan Singh *et al.,* 1987) while salicylic acid application resulted the highest LAI in rice (Kalpana, 1997).

2.2.1.4. Earliness in flowering

Application of brassinosteroid and salicylic acid induced early flowering in rice plants (Roy stephan, 1998) and pearl millet, respectively (Sivakumar *et al*., 2001) due to induction of indole acetic acid oxidase activity and increased auxin content. Abad *et al.* (1991) reported that application of humic acid at 50 mg l^{-1} increased the number of flower per plant and foliar area in cucumber plant.

2.2.1.5. Chlorophyll content

Total chlorophyll content in leaves at 20 days after flowering was enhanced by spraying of kinetin (10 ppm) in rice (Debata and Murthy, 1981). Shehata *et al.* (2001) reported increase in chlorophyll content of maize due to foliar application of kinetin (25 and 50 ppm). Foliar chlorophyll was increased by foliar application of cycocel in brinjal (Prakash and Ramachandran, 2000).

2.2.1.6. Girth

The girth of pseudostem in banana cv. Nendren was significantly influenced by cytozyme (Vijayalakshmi and Mathan, 2000).

2.2.1.7. Yield

Foliar spraying of 20 ppm benzyladenine increased the yield of rice (Anandhakrishnaveni *et al.*, 2001). Like wise, foliar application of cytokinin (120 ppm) at tillering stage increased rice yield (Pandey *et al.*, 2001). Wheat seed treatment with 24 – epibrassinocide recorded in increased crop yield (Nilovskaya *et al.*, 2001).

Foliar application of mepiquat chloride significantly increased cotton yield (Heliman, 1985). Kumara (1989) reported that treatment with brassinolide increased yield in rice, maize, soyabean and potatoes. Kulkarni *et al.* (1994) stated that application of 1000 ppm of mepiquat chloride in sunflower increased the seed yield, number of filled seeds, 100 seed weight, and seed filling percentage. Foliar application of mepiquat chloride at 150 ppm increased the pod yield in groundnut (Jeyakumar and Thangaraj, 1996; Jeyakumar, 1991).

Phenolics play a significant role in the initiation and development of floral buds of tomato, which in turn increase the fruit number and the yield (Kalarani *et al.*, 2002). Application of cycocel 1000 ppm in potato was found to increase the yield many fold (Pravin Prakash *et al.*, 1999).

2.2.2. Phytotonic effects of insecticides

Mote and Pawar (1994) reported that carbofuran 3G as slurry treatment soil application at 1 kg a.i. ha^{-1} and carbofuran 50 SP, carbosulfan (STD) and dimethoate 0.03 per cent increased the plant height, number of leaves per plant, leaf area, chlorophyll and nitrogen content of leaves, total number of fruits per plant and yield ha^{-1} as compared to untreated control.

Seed treatment with carbofuran increased plant height of okra (Shanthakumar *et al.*, 1975). Like wise Narke and Suryawanshi (1987) recorded more number of leaves per plant by treating the seeds with carbofuran 5 per cent. Shanthakumar *et al.* (1975) reported higher

chlorophyll and nitrogen content of okra leaves, early flowering by 2-3 days and more number of fruits/plant by treating the okra seeds with carbofuran 4.5 and 6 per cent. The results in respect of carbofuran 3G for higher okra fruit yield are in conformity with those of Fotedar and Singh (1980), Egwuatu (1984) and Narake and Suryawanshi (1987).

Sreelatha and Divakar (1997) stated that seed treatment with imidacloprid at 7.5 g kg^{-1} increased the plant height, leaf area and yield of okra in addition to the control of aphids and jassids. Siva Veerapandian (2000) and Bhargava and Bhatnagar (2001) observed higher yields of okra, when they are seed treated with imidacloprid 600 FS and 70 WP.

Jarande and Dethe (1994) reported that imidacloprid seed treatment increased plant height and total leaf chlorophyll over untreated plants in brinjal. Kirtisharma *et al.* (1997) reported that imidacloprid as seed treatment had increased leaf area without any phytotoxic effect on plants compared to monocrotophos treated plants.

2.3. Sensitivity of natural enemies and beneficial arthropods to insecticides in okra ecosystem

Though insecticides act as important component of integrated pest management (IPM), biological suppression of insect pests is an additive factor to some extent. Hence, protection and preservation of natural enemies of the pests are essential. Among the various entomophages, coccinellids, spiders and green lacewings are very important in okra ecosystem. Similarly, honey bees play an important role in boosting the yield of bhendi as pollinators. These pollinating bees were exposed directly to insecticides during spraying as well as to the left over insecticide on the crop. The toxicity level of insecticides on predator, parasitoids and honey bees are reviewed in this section.

2.3.1. Parasitoids

Singh and Verma (1986) observed that fenvalerate, deltamethrin and permethrin were relatively safer to *Trichogramma* sp. than other insecticides. Gangathara Rao *et al.* (1990) reported that carbaryl (0.15%) and triazophos (0.15%) were more toxic to *T. chilonis.*

Rajendran and Gopalan (1996) conducted an experiment to test the effect of endosulfan 35 EC, monocrotophos 36 WSC and cypermethrin 10 EC at different doses on the egg parasitoid, *T. chilonis* and observed toxicity to the parasitoids even in lower concentration. Toxicity studies conducted by Tiwari and Khan (2002) revealed that fenobucarb and chlorpyriphos methyl adversely affect the parasitisation.

House *et al*. (1985) reported the harmful effects of synthetic pyrethroids on the egg parasitoids *viz.*, *Trichogramma* spp. and *Telenomus* spp. Phosphomidon was reported by Mandal and Somchoudhury (1992) to be the least toxic to *T. chilonis*. Manisegarane and Kumarasami (1998) reported that endosulfan (700 g a.i. ha^{-1}) was relatively safer to *Chelonus blackburnii* (Cameron). Endosulfan was found to be the least toxic to adults of *Bracon hebetor* (Say) (Sreelatha *et al.*, 1995).

2.3.2. Predators

Krishnamoorthy (1985) studied the effect of endosulfan, monocrotophos, dimethoate, phosphamidon and cypermethrin on the eggs, larvae and adults of *Chrysopa sclestes* (Banks) and found that newly hatched larvae were more susceptible than eggs.

Hassen *et al.* (1985) studied the side effects of 14 pesticides on the second instar larvae of *Chyrsoperla carnea* (Stephens). The insecticides pirimicarb, bromophos, heptenophos and trichlorfos were either harmless or slightly harmful, while diflubenzuron, fenvalerate, dimethoate, methamidophos and chlorpyriphos were harmful. Srinivasan and Sundarababu (2000) observed that endosulfan (0.07%) and abamectin (0.03%) were less toxic to *C. carnea*.

Rajagopal and Kareem (1983) reported fenvalerate (0.05%) and methamidophos (0.05%) were toxic to adults and larvae of *Menochilus sexmaculatus* (Fab.), while dichlorvos (0.05%) was less toxic and phasalone (0.05%) and dimethoate (0.05%) were

safer to adults and larvae. Venugopal Rao (1986) stated that acephate at 0.5 kg a.i. ha^{-1}, methamidophos at 0.5 kg a.i. ha^{-1} and fenvalerate at 50 kg a.i. ha^{-1} had deleterious effect on the predator *M. sexmaculatus*.

Sharma *et al*. (1991) reported that cypermethrin @ 0.04 kg a.i. ha^{-1}, dimethaote @ 0.40 kg a.i. ha^{-1}, monocrotophos @ 0.40 kg a.i. ha^{-1} and fenvalerate @ 0.20 kg a.i. ha^{-1} were toxic to *M. sexmaculatus* on chickpea and lentil.

Under laboratory condition, Dhingra *et al*. (1995) evaluated ten insecticides against adults of *M. sexmaculatus*. The LC_{50} of deltamethrin, cypermethrin, phosphamidon, malathion and fenvalerate were 10.18, 3.10, 1.97, 1.55 and 1.05 times more toxic than monocrotophos, respectively, whereas demeton-methyl, endosulfan and lindane were 0.28, 0.04 and 0.02 times, respectively less toxic.

2.3.3. Spiders

Raman and Uthamasamy (1983) reported that carbosulfan was found to be less toxic to spiders in cotton ecosystem. Fenvalerate 10 EC at 0.3 l ha^{-1} recorded 51 per cent mortality of *Paradosa* spp. (Mansour *et al.*, 1992). Negata *et al*. (1997) reported that diazinon, tenobucarb, carbaryl, dichlorvos and imidacloprid were toxic to spiders in cotton. Spiderlings were found to be sensitive to azadirachtin (Punzo, 1997). Cole *et al.* (1997) observed that lambda-cyhalothrin has little impact on ground spiders. Vandenberg *et al.* (1998) found that monocrotophos at 108 g a.i. ha^{-1} adversely affect populations of *Paradosa* spp. in cotton ecosystem.

2.3.4. Honey bees

Usha and Kandasamy (1986) exposed *Apis cerana indica* F. to several insecticides and found diflubenzuron to be safe with no mortality even at 1000 ppm. Panda *et al.* (1989) found that phosphamidon followed by endosulfan and phasalone were

toxic to the foraging bees. Neem oil having insecticidal properties was found to be the less toxic followed by fenvalerate, decamethrin and malathion.

Toxicity tests conducted by Rana and Goyal (1997) revealed that methyl demeton (0.025%) and dimethoate (0.03%) were significantly toxic to honey bees as they reduced the colony strength in terms of mortality of the foragers.

Abrol and Rajiv (2000) reported that dimethoate and carbaryl were found to be highly toxic inhibiting the development of colonies in the treated area followed by endosulfan, fenvalerate and mancozeb, respectively. Kain and Agnello (2001) found that abamectin was toxic to bees and predatory mites and on contact profenofos was found less toxic to honey bees compared to other insecticide in usage (Renuka, 2001).

2.4. Effect of insecticides on earthworm populations

Earthworm populations are drastically influenced by modern agricultural practices. The pesticides applied as soil treatment may affect the non target earthworms due to the left over residues in crop lands (Edwards, 1984) and in forests (Zachariae and Ebert, 1970).

Among the commonly used insecticides, carbamates are the most toxic and are deadly to earthworms. Even small concentrations at recommended rates of application can severely reduce earthworm population (Finlayson *et al.,* 1975; Martin, 1976; Lebrun *et al.,* 1981; Medts, 1981). The commonly used fungicide benomyl is also very toxic to earthworms (Stringer and Lyons, 1974; Keogh and Whitehead, 1975). There is only a few informations regarding the effects of herbicides on earthworms in tropical soils and the toxicity varies with the species (Edward and Stafford, 1979). Herbicides are more toxic in coarse textured soils of low organic matter content than in clayey soils or those with high organic matter. Some herbicides indirectly affect earthworms (Martin, 1982)

by eliminating the algal food supply. Some other herbicides are reported to have beneficial effect on earthworms (Leibundgut, 1981). The relative toxicity of different pesticides on earthworms are presented in Table 1.

Table1. Relative toxicity of different pesticides on earthworms

Chemicals tested	Relative toxicity	Comments	References
Inorganic chemicals			
■ Copper oxychloride	Relatively non toxic	Persistent toxic at high exposure rates	Haque and Ebing (1983) Rhee Van (1969)
■ Lead arsenate	Relatively non toxic	Persistent toxic at high exposure rates	Devey (1963) Lidgate (1966)
■ Potassium bromide	Moderately toxic	-	Edwards (1984) Heimbach (1984)
Aromatic and organochlorine insecticides			
■ BHC	Relatively non toxic	Non toxic except at high exposure rates	Dawson *et al.* (1978) Edwards and Lofty (1973); Bouche(1984)
■ Endosulfan	Moderately non toxic	Highly toxic at high exposure rates	Heimbach (1984) Roberts and Dorough (1984); Hans *et al.* (1990); Reddy and Reddy (1992)
Organophosphorous insecticides			
■ Chlorfenviphos	Slightly toxic	-	Edwards *et al.* (1971)
■ Dimethoate	Slightly toxic	-	Fayolle (1979)
■ Methyl parathion	Very toxic	Extremely toxic at high exposure rates	Hyche (1956); Dikshith and Gupta (1981); Cathey (1982)
■ Malathion	Moderately toxic	Toxic at normal exposure rates	Martin and Wiggans (1959); Galvyalis and Lugaustas (1978); Roberts and Dorough (1984); Senapati *et al.* (1991, 1992)
■ Phorate	Extremely toxic	-	Edwards *et al.* (1971); Malone and Raichle (1973)

Carbamate insecticides			
■ Aldicarb	Extremely toxic	-	Ruppel *et al.* (1973)
■ Bendiocarb	Extremely toxic	Probably toxic at normal exposure rates	Potter *et al.* (1990)
■ Propaxur	Very toxic	-	Fox (1974); Roberts and Dorough (1984); Kula and Kokta (1992)
Synthetic pyrethroids			
■ Cypermethrin	Slightly toxic	-	Inglesfield (1984) Roberts and Dorough (1984)
■ Fenvalerate	Relatively nontoxic	-	Lofs- Homlin (1982); Roberts and Dorough (1984)
■ Pyrethrins	Insufficient evidence	Probably non toxic at normal exposure rates	Bouche (1984)
Fumigants and nematicides			
■ Chloroplorin	Extremely toxic	-	Blackwaardt and Vander Drift (1961); Rheevan (1969)
■ Formaldehyde	Moderately toxic	Expels earthworm from soil	Blankwaardt and Vander Drift (1961)
Fungicides			
■ Benomyl	Extremely toxic	Probably very toxic at normal exposure rates	Patel (1960); Edwards and Lofty (1973); Stringer and Lyons (1974); Bouche (1984); Heimbach (1984); Edwards (1984)
■ Captan	Slightly toxic	Probably nontoxic at normal exposure rates	Martin and Wiggans (1959); Stringer and Lyons (1974); Cook and Swait (1975); Heimbach (1984)
■ Carbendazim	Very toxic	-	Keogh and Whitehead (1975); Lofs-Homlin (1981)

Herbicides			
■ Atrazine	Relatively non toxic	-	Caseley and Eno (1966); Ghabbour and Imam (1967); Haque and Ebing (1983)
■ Pentachloro-phenol	Extremely toxic	-	Goats (1983); Edwards (1984); Gestalvan and Ma (1988, 1990); Goats and Edwards (1988)
■ Simazine	Moderately toxic	-	Ilijin (1969); Galvyalis and Lugavskas (1978); Martin (1982); Gestelvan and Ma (1988)
■ Trichloro acetic acid	Slightly toxic	-	Fox (1964); Atlavinyte *et al.* (1978); Lofs - Holmin (1980); Edwards (1984)
Other organic compounds			
■ 3 - chlorophenol	Extremely toxic	-	Gestal Van and Ma (1988, 1990)
■ Dichloroaniline	Insufficient evidence	Proably nontoxic at normal exposure rates	Gestal Van and Ma (1990)
■ 3,4,Dichloro-phenol	Extremely toxic	-	Gestal Van and Ma (1998, 1990)
■ P. Nitrophenol	Extremely toxic	-	Randall *et al*. (1972)
■ Nitrobenzene	Insufficient evidence	Probably nontoxic at normal exposure rates	Neuhauser *et al.* (1986)

Neuhauser *et al*. (1986) studied the toxicity of organic compounds to the earthworm (*Eisenia foetida* Kinberg), by exposing via filter paper in glass vials in a 48-h contact test reported an LC_{50} of 16 g cm^{-2}.

2.5. Insecticide residues in okra

Endosulfan residues in the fruits of okra, from plants treated twice with 500 and 1000 g a.i. ha^{-1} of endosulfan were found to be below the safety level for consumption (Bhattacharya *et al.*, 1989). Harvest time residues of imidacloprid applied as seed and

foliar applications were below detectable limit (BDL) on tomato fruits (Tolman *et al.,* 2000) and okra (Indumathi *et al.,* 2001; Dikshit *et al.,* 2002.). Dikshit *et al.* (2000) reported that the residues of imidacloprid in okra fruits were found to be 0.08, 0.10, 0.14 and 0.24 mg kg^{-1} from 3, 5.4, 10.8 and 21.6 g a.i. kg^{-1} seed treatments, respectively, after 55 days of sowing and became non detectable after 60 days of sowing.

The residues of monocrotophos applied at 20.78 to 56.76 ppm concentrations, dissipated to the level of 0.16 to 2.59 ppm at an interval of 21 days (Shetgar *et al.,* 1982).

Pokharkar and Dethe (1981) reported that from initial residues of (10.00 ppm) of phasalone sprayed at 0.05 per cent dissipated by more than 90 per cent within three days on okra. The half-life lives of phosalone at 0.05 and 0.10 per cent on okra were 1.20 and 1.34 days. Hence, four days gap between spraying of monocrotophos (0.5 kg a.i. ha^{-1}) and harvesting of okra fruits appears to be safe (Anonymous, 1976).

2.6. Sensitivity of non target organisms to nitrobenzene

Since nitrobenzene was suspected to have an impact on the environment, its toxicity to the microorganisms in the environment has been reviewed.

2.6.1. Toxicity to bacteria

Blum and Speece (1991) studied the effect of nitrobenzene on bacterial populations. The inhibition of ammonia consumption was used as criterion for Nitrosomonas with an Effective Concentration (EC_{50}) of 0.92 mg lit^{-1}. Inhibition of oxygen uptake was used as the criterion for aerobic heterotrophs with an EC_{50} of 370 mg lit^{-1}. Daneer *et al.* (1989), Kaisor and Palabrica (1991) and Kaiser and Ribo (1985) recorded EC_{50} of 17.8, 28.2 and 34.7 mg lit^{-1} for the bacteria *Vibrio fischeri* (Beijerinde) at various exposure duration (*viz.*, 15, 5, 30 min., respectively).

2.6.2. Toxicity to protozoa

EC_{50} of 7 mg lit^{-1} for *Pseudomonas putida* (Trevisan) at 960 min. was recorded by Bringmann and Kuhn (1980). They also reported a toxic threshold for nitrobenzene 1.9 mg lit^{-1} over a 72 h test period, based on cell multiplication using the protozoan *Entosiphon sullatum* (Bringmann).

Yoshioka *et al*. (1985) exposed the freshwater protozoan *Tetrahymena pyriformis* (Ehrenberg) to nitrobenzene. A 24 h, EC_{50} based on growth rate, was 98 mg lit^{-1}. Schultz *et al*. (1989) reported a 48h - EC_{50} of 106 mg lit^{-1} based on growth rate, using the same species.

2.6.3. Toxicity to fungi

Gershon *et al.* (1971) studied the fungistatic activity of nitrobenzene on *Myrothecium verrucaria* (Alvertini and Sehwein) at 0.9 mg lit^{-1} for *Aspergillus niger* (Van Tighem), *Aspergillus oryzae* (Ahlburg) and *Trichoderma viridi* (Pers.) and recorded no fungistatic effect of nitrobenzene.

2.6.4. Toxicity to algae

Ramos *et al*. (1999) found that the EC_{50} for *Chlorella pyrenoidosa* (Chick) a green algae varied from 28-36 mg lit^{-1}, for the exposure duration of 72 h.

2.6.5. Toxicity to aquatic organisms

For freshwater invertebrates, 24 to 48-h LC_{50} values for nitrobenzene ranged from 24 mg lit^{-1} for the water flea *Daphnia magna* (Ring.) to 140 mg lit^{-1} for the snail *Lymnaea stagnalis* (Linn.). The flatworm *Dugeis japonica* (Jchikawa) was sensitive, with a 168h LC_{50} of 2 mg lit^{-1}. The marine species tested was the mysid shrimp *Mysidopsis bahia* (Jemmlner .F), which was more sensitive than the freshwater species (96 h LC_{50} of 6.7 mg lit^{-1}). In long-term toxicity tests (20 days) using *D. magna*,

Canton *et al*. (1985) reported 20-day values as follows: the LC_{50} was 34 mg lit^{-1}, the EC_{50} based on reproduction was 10 mg lit^{-1}, and the (no-observed-effect concentration) NOEC was 1.9 mg lit^{-1}. Similarly, Deneer *et al*. (1989) found a 21-day EC_{50} (based on immobilization in *D. magna*) to be 24 mg lit^{-1}. The lowest concentration tested that significantly decreased the length of the daphnids was reported to be 17.8 mg lit^{-1}.

The reproductive toxicity of nitrobenzene in the water flea (*D. magna*) was dependent on exposure duration, with NOEC values decreasing from 12 to 2.6 mg lit^{-1} over a 14- to 21-day exposure (Hattori *et al*., 1984).

2.6.6. Toxicity to fish

The 96-h LC_{50} values for nitrobenzene ranged from 24 mg lit^{-1} for *Oryzias latipes* (Temminck and Schlegel) to 142 mg lit^{-1} for guppy *Poecilia reticulate* (Peters). Yoshioka *et al*., (1986) reported that medaka were particularly sensitive, with a 48-h LC_{50} of 1.8 mg $lit^{-1.}$ Little information was available on long-term effects of nitrobenzene. Canton *et al.* (1985) reported an acute 18-day LC_{50} for nitrobenzene for *O. latipes* to be 24 mg $lit^{-1.}$ The NOEC, based on mortality and behaviour was 7.6 mg lit^{-1} .

Black *et al*. (1982) exposed rainbow trout *Oncorhynchus mykiss* (Walbaum) embryo-larval stages subchronic tests from fertilization to 4 days post-hatching to nitrobenzene under flow-through conditions; total exposure time was 27 days. A wide range of concentrations was used in the test 0.001, 0.01, 0.12, 0.36, 0.91 and 11.9 mg lit^{-1}. An LC_{50} of 0.002 mg lit^{-1} for nitrobenzene was reported at the time of hatching and at 4 days post-hatching. However, there is doubt about the validity of this figure, because concentrations below 0.12 mg lit^{1} were nominal values.

2.7. Nitrobenzene residues in environmental samples

Nitrobenzene was detected and analysed from air samples using HRGC (High Resolution Gas Chromatography), GC FID (Gas Chromatography- Flame Ionization Detector), (Harkov *et al.*, 1985; NIOSH, 1984; NIOSH, 1977), from water samples using,

GC- FID and MS (Mass Spectrometry) (Patil and Shinde, 1988; US EPA, 1982a; US UPA, 1982b) and from soil using, GC- MS, FID, HRGC and FTIR (Fourier Transform Infrared Spectrometry) (US EPA, 1986b; US EPA, 1986c; US EPA, 1986d; US EPA, 1986).

The movement of nitrobenzene in soil, water and air is predicted by its physical properties like water solubility, moderate volatility, low octanol water partition co-efficient, soil sediment co-efficient etc. The dimensionless Henry's law constant for nitrobenzene suggests that the transfer from water to air will be significant, but not rapid (US EPA, 1985; Lyman *et al.,* 1982).

CHAPTER III

MATERIALS AND METHODS

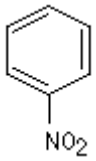
Studies were carried out to evaluate the phytotonic and phytotoxic effects and determine the residue of Boom Flower®–n (Nitrobenzene 20% v/v) in harvestable produce of okra under field conditions. Field and laboratory studies were conducted to determine the sensitivity of natural enemies *viz., Trichogramma chilonis (*Ishii) and *Chrysoperla carnea* (Stephens), *Chelonus blackburni* (Cameron) and Indian bees to Boom Flower ® - n for different experiments was obtained from Devi pesticides Pvt.Ltd., Madurai. The general properties of Boom Flower ® –n formulation are given below.

Chemical name : Nitrobenzene

IUPAC name : Nitrobenzene

Trade name : Boom Flower® – n (Nitrobenzene 20% v/v)

Structure



Formulation : 20% v/v (20% nitrobenzene, 40%surfactant and 40% filler)

3.1.1. Evaluation of phytotonic effect of Boom Flower ® –n

3.1.2. Experimental details

Two field experiments were conducted in a randomized block design with a plot size of 4 x 5 m with three replications. The experimental sites were located in farmers' field at Subramanianagar, Thondamuthur block and Chinnamuthampalayam, Thudiyalur block of Coimbatore district (Plate 1 and 2) during winter (Dec-Feb) 2003 and summer (Mar-May) 2004, respectively, with okra Mahyco hybrid no 10. The crop was maintained well by adapting necessary prophylactic measures against pest and diseases, and other standard agronomic practices as per the recommendations of Tamil Nadu Agricultural

University. Boom Flower® –n was applied at 20 day intervals commencing from 20 days after sowing (20DAS, 40DAS, and 60DAS) at the following concentrations with control plots maintained with water spray.

i.	Boom Flower® –n	2ml lit^{-1}
ii.	Boom Flower® –n	2.5ml lit^{-1}
iii.	Boom Flower® –n	3ml lit^{-1}
iv.	Boom Flower® –n	4ml lit^{-1}
v.	Boom Flower® –n	5ml lit^{-1}
vi.	Boom Flower® –n	8ml lit^{-1}
vii.	Boom Flower® –n	10ml lit^{-1}
viii.	Untreated control (water spray)	-

3.1.2. Observations

The growth parameters and activities of natural enemies were recorded on 0, 1, 3, 5, 7, 10 and 15 days after application. Thus three sets of observations were made; one each during vegetative phase (20 days after sowing), flowering phase (40 days after sowing) and harvesting phase (60 to120 days after sowing). Control plots were maintained with water spray. In each plot ten plants were randomly selected and tagged for taking observations in each treatment.

3.1.2.1. Growth parameters

Height of the plant from the cotyledonary node to the tip of the main stem was measured in ten randomly tagged plants and mean worked out. Like wise, numbers of branches that arise from meristem and leaves produced were recorded. The area of the leaves at three different canopy levels of the plants was found out by feeding the leaves into the portable electronic leaf area meter (CI-203 Area meter, CID Inc., U.S.A.). The area was multiplied with the total number of leaves at corresponding stages.

3.1.2.2. Yield parameters

At each phase of observation, number of flowers bloomed and fruits produced in tagged plants were recorded. At each harvest, fruits from the selected plants were collected, weighed separately and the mean worked out. Further, in order to assertain the quality of fruits, length from the base (stalk end) to the apex and girth at the broadest point in ten randomly selected fruits were measured at the time of harvest. All the fruits harvested from the tagged plants were collected, weighed and yield was calculated.

3.1.2.3. Assessment of natural enemies

The randomly selected tagged plants were thoroughly observed for population of natural enemies. The presence of natural enemies such as coccinellids, chrysopids and spiders present in 10 plants per plot prior to spraying and 1, 3, 7 and 14 days after spraying were recorded.

Honey bees are one of the major pollinators of okra, as it produces large amounts of pollen and often a cross pollinator crop. Hence, the sensitivity of honey bees to Boom Flower ® –n was studied. Observations were made on the pollinators' activity by counting the number of honey bees visiting for flowers in Boom Flower ® –n treated and untreated plots for every 30 min. from 7am to 12 noon. The observations were taken before and 1, 3 and 7^{th} days after treatment.

3.1.2.3. Physiological parameters - Chlorophyll

About 250 mg of the plant sample (leaf bit) was macerated with 80 per cent acetone using pestle and mortar. Macerated sample was centrifuged at 3000 rpm for 10 min. Supernatant solution was poured into 25 ml volumetric flask and the volume made up using 80 per cent acetone. Optical density of the sample solution was observed at 645, 663 and 652 nm in a spectrophotometer (Yoshida *et al*., 1971). Chlorophyll a, chlorophyll b and total chlorophyll contents were calculated using the following formulae

$$\text{Chlorophyll a} = \frac{(12.7 \times \text{OD at } 663) - (2.69 \times \text{OD at } 645)}{1000 \times \text{weight of sample (g)}} \times \text{volume made up}$$

$$\text{Chlorophyll b} = \frac{(22.9 \times \text{OD at } 645) - (4.68 \times \text{OD at } 663)}{1000 \times \text{weight of sample (g)}} \times \text{volume made up}$$

$$\text{Total Chlorophyll} = \frac{\text{OD at } 652 \times 1000 \times \text{volume made up}}{34.6 \times 1000 \times \text{wt}} \text{ mg of chlorophyll } g^{-1}$$

3.2. Phytotoxic effect of Boom Flower ® -n on okra

A field experiment was conducted during winter and summer 2003-2004 in farmers' field at Thoundamuthur and Chinnamathampalayam near Coimbatore to find the phytotoxic effect of different doses of Boom Flower ® -n formulations on okra (Mahyco hybrid no 10). The experiment was conducted in a randomized block design with a plot size of 4 x 5 m replicated thrice. The following concentrations of Boom Flower®–n were tested. Controls plots were sprayed with water.

i.	Boom Flower® –n	2ml lit^{-1}
ii.	Boom Flower® –n	2.5ml lit^{-1}
iii.	Boom Flower® –n	3ml lit^{-1}
iv.	Boom Flower® –n	4ml lit^{-1}
v.	Boom Flower® –n	5ml lit^{-1}
vi.	Boom Flower® –n	8ml lit^{-1}
vii.	Boom Flower® –n	10ml lit^{-1}
viii.	Untreated control (water spray)	-

3.2.1. Observations made

Observations were made on 0, 1, 3, 7 and 14 days after spraying for phytotoxic symptoms like

i. Injury to leaf tip and leaf surface

ii. Wilting

iii. Vein clearing

iv. Necrosis

v. Epinasty and hyponasty

The phytotoxic symptoms were recorded based on the rating scale, described below (Chelladurai, 1999).

Rating	**Phytotoxicity (%)**
0	No Phytotoxicity
1	1 - 10
2	11 - 20
3	21 - 30
4	31 - 40
5	41 - 50
6	51 - 60
7	61 - 70
8	71 - 80
9	81 - 90
10	91 -100

The per cent leaf injury was calculated by the following formula

$$\text{Per cent leaf injury} = \frac{\text{Total grade points}}{\text{Maximum grade x No. of leaves observed}} \text{ x } 100$$

3.3. Laboratory studies

3.3.1 Sensitivity of non target organisms to Boom Flower ® –n

The effect of Boom Flower ® –n formulation on the following beneficial organisms was studied.

i. Egg parasitoid - *Trichogramma chilonis* (Ishii)
ii. Predator - *Chrysoperla carnea* (Stephens)
iii. Egg larval parasitoid - *Chelonus blackburni* (Cameron)
iv. Earth worm - *Eudrilus eugeniae* (Kinberg)
Perionyx excavatus (Perrier)
v. Honey bees
Indian bee - *Apis cerana indica* (Fabricius)
Italian bee – *Apis mellifera* (Linnaeus)
Dammer bee – *Trigona irridipennis* (Smith)
vi. Fishes
Common carp – *Cyprinus carpio* (Linnaeus)
Tilapia - *Tilapia mosambica* (Peters)

3.3.2. Mass rearing of egg parasitoid, *T. chilonis*

3.3.2.1. Culturing of *Corcyra cephalonica* (Staint)

C. cephalonica was reared in the laboratory as per Navarajan Paul (1973). The adult moths were allowed inside an oviposition cage of 21 x 25 cm size, with a wire mesh at bottom and two sides for ventilation. Adults were fed with 50 per cent honey solution. Eggs were collected from the oviposition cages daily upto four days and cleaned. The cleaned eggs were sprinkled over half ground - cumbu grains, at the rate of one cc per 2.5 kg of grains, fortified with ten grams of yeast in a plastic basin of 11 x 37.5 cm size and covered with kada cloth. Care was taken to maintain the culture free of storage mites and diseases by mixing five grams of wettable sulphur (80%) and streptomycin sulphate 0.5 per cent, respectively. The emerged adults were collected and

used again for culturing both host (*C. cephalonica*) and parasitoid (*T. chilonis*). The culture was maintained at room temperature (26 ± 4^{o}C).

3.3.2.2. Culturing of *T. chilonis*

The egg parasitoid *T. chilonis* was mass cultured in biocontrol laboratory on the eggs of rice moth, *C. cephalonica* as per Prabhu (1991). Fresh *C. cephalonica* eggs collected in early morning were sterilized under UV radiation of 15 W capacity for 20 min at a distance of 15 cm to avoid the emergence of *C. cephalonica* larvae. These eggs were pasted on paper cards of 20 x 30 cm size having thirty, 7 x 2 cm rectangles. These egg cards were placed in polythene bags along with nucleus card at 6:1 ratio for parasitization by the egg parasitoids. The parasitised egg cards were cut into one cm^{2} bits and three days old 100 per cent parasitised eggs (eggs appearing black and plumpy) were used for the study.

3.3.3. Mass production of predatory green lacewing, *C. carnea*

Mass rearing of *C.carnea* was done using *C. cephalonica* eggs (Swamiappan, 1996).

3.3.3.1. Larval rearing

Grubs of *C. carnea* were reared in galvanized iron (GI) basins (28 cm dia) at 250 larvae basin^{-1} covered with kada cloth. The eggs of C. *cephalonica* were provided as feed for the grubs in the laboratory. About 25cc of *C. cephalonica* eggs per basin were given on alternate days. After five feedings, the larvae pupated into round white colored silken cocoon. Cocoons were collected and transferred into one litre plastic container with wire mesh window for emergence of adults.

3.3.3.2. Adult rearing

The adults were collected and transferred to GI troughs (30 cm dia x 12 cm ht), wrapped inside with brown sheets for collecting the eggs. The trough was covered with

nylon cloth kept firm with the help of a rubber band. Over the cloth covering, two bits of foam sponge (2.5 cm^2) dipped in water were kept, besides an artificial rich protein diet in the form of semi solid paste was smeared. This diet consisted one part of yeast powder, one part of fructose, one part of honey and one part of Proteinex® mixed in water as a paste. Adults fed the food and laid eggs on the brown sheet wrapped inside the trough. The rearing troughs were changed every day and fresh feed provided.

3.3.4. Mass multiplication of earth worms

3.3.4.1. *E. eugeniae*

The nucleus culture of *E. eugeneae* was obtained from regional research station Paiyur. The culture was mass multiplied in buckets (20 l). Soil (~ 20 kg) collected at TNAU farm, cleaned for pebbles, plant debris and other extraneous materials, was amended with 10 per cent farm yard manure. Pots were maintained in a cool and dark place and moisture was maintained at constant level by periodical sprinkling of water. After six months, medium sized earthworms with well developed clitellum were selected for the bioassays.

3.3.4.2. *P. excavatus*

The nucleus culture of *P. excavatus* was obtained from Department of environmental science, Tamil Nadu Agricultural University, Coimbatore. The culture was mass multiplied in loose compost heaps with high N level. A mixture of dung material (cow, sheep and horse) with kitchen wastes were used as feed material for *Perionyx*. Propagation was done under shady places with sufficient level of moisture condition.

3.4.4. Experimental protocol

Laboratory experiments were conducted to determine the sensitivity of egg parasitoid, *T. chilonis* and *C. blackburni* and predatory green lacewing, *C. carnea* to Boom Flower ® –n at the following concentrations

The treatments were

i. Boom Flower® –n 2ml lit^{-1}

ii. Boom Flower® –n 2.5ml lit^{-1}

iii. Boom Flower® –n 3ml lit^{-1}

iv. Boom Flower® –n 4ml lit^{-1}

v. Boom Flower® –n 5ml lit^{-1}

vi. Untreated control (water spray)

The experiments were conducted with four replications.

3.3.4.1. Sensitivity of *T. Chilonis* to Boom Flower ® –n

The parasitized egg cards were cut into one cm^2 bits and three days old hundred percent parasitized egg cards (eggs appearing black and plumpy) were sprayed with Boom Flower®–n at different concentrations mentioned alone using an atomizer. For untreated check distilled water was sprayed. The treated egg cards were shade - dried for 10 min. and hen kept in a test tube of 10 x 1.5 cm size (Plate 3). The number of parasitoids emerged from each treatment was recorded after 24 and 48 h of treatment and per cent emergence was worked out. Fresh eggs were provided to these parasitoids at 6:1 ratio and the number of parasitized eggs (eggs appearing black and plumpy) were recorded after 24 and 48 h of treatment and per cent parasitization was worked out.

3.4.4.2. Sensitivity of *C. blackburni* to Boom Flower®–n

a. Effect on adults – Thin film method

C. blackburni wasps, mass breed at Project Directorate of Biological Control (PDBC), Bangalore, on the eggs of *C. cephalonica* were purchased. Experiment with *C. blackburni* adults were carried out as per McCutchen and Plapp (1988) as done for *C. carnea* with modifications.

Glass scintillation vials of 20 ml capacity with 1 mm thickness were evenly coated with 0.5 ml of acetone solution containing Boom Flower®-n formulations and dried by rolling for few seconds. Three day old adults of *C. blackburni* were released into the vials @ 10 per vial, covered with muslin cloth and secured with a rubber band. After 24 h exposure of the wasps, honey solution was provided as feed to the wasps. Per cent mortality of the adults was worked out. Mortality observations were made at 6, 12, 24 and 48 h after treatment.

b. Sensitivity of *C. blackburni* to Boom Flower®–n on adult emergence

Test tubes of size 2.0 x 1.5 cm were coated with 1 ml of acetone solution containing Boom Flower®–n formulations at different concentrations and dried by rolling it for few seconds. One day old *C. cephalonica* eggs were placed inside the test tubes in the ratio of 1:10 (Parasitoid: Host). Three day old adults of *C. blackburni* were released into the vials at 10 per vial and covered with muslin cloth and secured with a rubber band, honey solution was given as feed to the wasps. After 24 h, the egg cards were transferred to plastic container containing broken cumbu grains. The number of parasitoids emerged from each treatment was recorded and per cent emergence worked out.

3.3.4.3. Sensitivity of *C. carnea* to Boom Flower®–n

a. Effect on the eggs

Experiments were conducted under laboratory condition to study the effect Boom Flower®–n formulation on eggs of *C. carnea* as per Krishnamoorthy (1985). Brown paper strips holding one day old eggs with stalk (Plate 4 and 5) were sprayed with different concentrations of Boom Flower®–n mentioned in section 3.3.4 using an atomizer. There were four replications with 20 eggs per treatment. Untreated check was maintained by spraying distilled water. Number of grubs hatched from each treatment was recorded and per cent hatchability was worked out.

b. Effect on the grubs of *C. carnea*

i. Oral feeding method

Eggs of *C. cephalonica* were exposed to UV radiation of 15 W capacity for 15 min. to kill the embryos and sprayed with different concentrations of Boom Flower®–n (3.3.1.) with an atomizer. The treated eggs were shade - dried for 15 min. and then transferred to test tubes (one cc / test tube) of size 15 x 2.0 cm. For untreated check, eggs were sprayed with distilled water. Second instar grubs of *C. carnea* were transferred to these test tubes @ 10 per test tube (Plate 6). After complete feeding of the treated eggs, the grubs were provided with untreated *Corcyra* eggs until pupation. Observations were made on the grub mortality 12, 24 and 48 h after treatment and per cent grub mortality worked out.

ii. Thin film method

The bioassay method described by McCutchen and Plapp (1988) was adopted with modifications. Boom Flower®–n 0.2 , 0.25, 0.3 0.4 and 0.5ml were dissolved in 100 ml analytical grade acetone to obtain required concentrations.

Glass scintillation vials of 20 ml capacity with 1 mm thickness were evenly coated with 1 ml of acetone solution containing Boom Flower®–n formulations at different concentrations and dried by rolling for few seconds. Second instar lacewing grubs were released into the vial at 10 per vial and covered with muslin cloth and secured with a rubber band (Plate 7). After 24 h exposure of the grubs, 1 cc of *C. cephalonica* eggs was provided as feed to the grubs. Observations were made on mortality at 12, 24 and 48 h after treatment and mortality worked out.

C. Effect on the adults of *C. carnea*

Five pairs of *C. carnea* adults were released into individual containers (30 cm dia x 12 cm ht) and allowed to feed ten per cent honey solution containing different concentrations of Boom Flower®–n. Control was maintained by feeding the adult with

honey solution 10%. Eggs laid by adults in each treatment were collected daily by keeping a brown paper sheet of 21 x 6 cm size along the inner side of the container. Observations were made on the number of eggs laid and adult longevity in all the treatments.

3.3.4.4. Sensitivity of honey bees to Boom Flower®–n

A laboratory experiment was conducted to assess the sensitivity of honey bees to Boom Flower®–n .The experiment consisted of treatments as mentioned under Section 3.3.4. One day old Indian, Italian and dammer worker bees were obtained from the Apiary, Department of Agricultural Entomology, Tamil Nadu Agricultural Collage, Coimbatore. Filter paper bits, Whatman No. 1 (7 cm diameter) were impregnated with solutions of different concentrations of Boom Flower®–n, dried and placed in Petri plates along with control. Ten worker bees were allowed in each petriplates. Small paper bits were placed between bottom and covers of petriplates, so as to provide enough ventilation for the bees (Plate 8). After a lapse of 1 hour exposure period , bees were transferred into polybags provided with 40 per cent sucrose solution soaked in cotton wool as food. The bee mortality was observed after 12 and 24 h of treatment and per cent mortality worked out.

3.3.4.5. Sensitivity of Boom Flower®–n on earth worms *E. eugeniae* and *P. excavatus*

The effect of Boom Flower®–n on earthworm *E. eugenae* population was tested by following the artificial soil test method proposed by Biollogische Bundesanstalt fur Land-und Forst Wirsts chaft, Braunschweig (BBA) as reported by Ganesh Kumar (2002).

The test substrate was prepared by mixing fine quartz sand 83.5 percent (84 percent of the sand particle size between 0.06 and 0.2mm), bentonite 5 percent, finely ground and dried sphagnum peat10 per cent (pH 2.6+ 0.5), pulverized calcium carbonate (1 %) and ground dried (0.5 %) cow dung. The pH was adjusted to 7+0.5 and sufficient water was added to bring moisture content to 35 per cent of dry weight of the substrate. The complete mixture was moist enough, but not so wet that water appeared when the artificial soil was compressed.

One kg of conditioned soil in tubular pots (18x 6 cm) was treated with Boom Flower®–n of 7.2, 7.4, 7.6, 7.8, and 8.0 ml for *E. eugeniae* and 6.4, 6.6, 6.8, 7.0 and 7.2 ml for *P. excavatus* and fifteen earthworms washed cleanly in water were placed on the top of the substrate plate (9 and 10). The tubular pots were covered with perforated polythene cover to prevent the worms from crawling out and to avoid evaporation. The set up was kept under shade. After 7 days, 5 g of finely ground dried cow dung was mixed inside the container and water lost by evaporation was replaced. The number of live earthworm was counted. Earthworms were considered dead if they did not respond to a gentle mechanical stimulus (Edwards and Bohlen, 1992). The LC_{50} was calculated by probit analysis (Finney, 1971). Untreated control should also be maintained throughout the experiment.

3.3.4.6 Sensitivity of common carp *C. carpio* and *T. mosambica* to Boom Flower®-n

The acute toxicity of common carp to Boom Flower®-n was done as per Sprague (1969 and 1970). The aquarium tanks of dimensions 15cmx30cmx45cm with 80 litre capacity were provided with artificial aeration facilities (Plate 11). Boom Flower®-n at different concentrations *viz.*, 60, 70, 80, 90, and100 ppm in water were the treatments. Control tanks were maintained without Boom Flower®-n. Each experiment was replicated four times. In each tank a school of twelve hours starved twenty fish of same size was released. Observations on mortality were made on 3, 6, 12, and 48h and LC_{50} and LT_{50} worked out.

Another experiment to asses the acute toxicity of Boom Flower®-n to *T. mosambica* was conducted in the same method. However, the fishes were exposed to 160, 170, 180,190 and 200 ppm of Boom Flower®-n and mortality was observed (Plate 12).

3.4. Estimation of Boom Flower®-n residues in okra

3.4.1. Sampling

Fruit samples of okra were collected on 0, 1, 3, 5, 7, 10 and 15 days after the third round of Boom Flower®-n spraying in both the trial I and trial II. From each treatment, 250 g fruits from each replicate were collected. From these two 50 g of two laboratory

samples were drawn. The samples were placed in a wide mouthed plastic sampling container having 10 percent methanol (10 ml methanol+90 ml distilled water). The bottles were closed tightly with teflon lined cap and stored at -4^0C in deep freezer until extraction of residues.

3.4.2. Extraction

Before extraction process, bottles containing samples in solvents were allowed to thaw to room temperature. Okra fruit samples were shaken intermittently for 2 min. and blended in a homogenizer with 10 percent methanol. The extract was filtered through Buchner funnel with mild suction and the residues re-extracted with two 20 ml portions of 10 percent methanol.

3.4.3. Clean up

3.4.3.1. Liquid liquid clean up

The extract was taken in a 500 ml separating funnel, mixed with 250 ml of saturated sodium chloride solution and partitioned thrice with 25 ml portions of dichloromethane. The resultant organic layers were pooled together and dried by filtering through a funnel plugged with piece of cotton over laid with anhydrous sodium sulphate to remove the traces of water. The filtering funnel and its contents were rinsed with 10 ml portions of the solvent and the resultant solution was condensed to near dryness in a flash vacuum rotary evaporator.

3.4.3.2. Column clean up and estimation of residues

For column chromatography, 50 cm (length) x 1.5 cm (i.d) glass columns were used. The dripping end of the chromatographic column was plugged with cotton wool. The column was filled with the absorbent silica gel (8g of deactivated silica gel) sand witched in between with 2 cm layers of anhydrous sodium sulphate. The silica gel was activated at 135^0C for 6h and cooled in a desiccator. The cooled silica gel was deactivated by adding three per cent water and mixed well.

The condensed dichloromethane extract (10 ml) was loaded into the column and eluted with 50 ml of hexane. About 100 ml of the eluate was collected, condensed to near dryness and redissolved in 5 ml redistilled hexane for final determination in Gas Chromatography (GC) model Chemito - 8610 equipped with Thermionic detector (TID).

3.4.4. Final determination

3.4.4.1. Standards

The reference standard of Boom Flower®-n with 95 per cent purity obtained from Devi Pesticides Pvt. Ltd, Madurai was used for quantification and to determine the detectable limits.

Concentrated stock solution

The analytical grade Boom Flower®-n with 95 per cent purity (105 mg) was dissolved in solvent toluene (100 ml) to get 1000 ppm standard stock solution.

b. Intermediate stock solution

The concentrated stock solution was brought to room temperature and one ml from the concentrated stock solution was transferred to a 100 ml volumetric flask, made up the volume and shaken well to obtain a homogenous solution of 10 ppm of intermediate stock solution. This 10 ppm solution was utilised for fortification of samples.

Working standard

From the intermediate stock solution, after bringing to room temperature, working standards of 0.5 to 5 ppm were prepared by diluting 1 ml of 10 ppm solution to 2-20 times. These working standards were used to find out the retention time of these compounds and for quantitative determination of residues in samples.

3.5.4.2. Recovery studies / Fortification

The okra fruits were fortified at 0.5, 1 and 2 ppm by adding required quantity of 10 ppm standard solution to work out the recovery per cent of analytical methodology.

3.4.4.3. Final determination of residues

3.4.4.3.1. Boom Flower®-n residues

Boom Flower®-n residues were estimated by Chemito model 8610 Gas Chromatograph equipped with Thermoionic detector (TID) fitted with capillary column DB-1701P-30m X 0.32 mm i.d. × 0.25 μfilm thicknesses. The following were the operating parameters.

	:	Column	Injector	Detector
Temperature ^{o} C	:	160	200	250
Current for Rubidium source	:	330		
Gas flow rate (ml/min.)				
N (Carrier)	:	2		
H_2	:	8		
Zero air	:	90		
Make up gas	:	28		
Attenuation	:	8		
Aliquot injected	:	1 μl		

The final quantification was made using the formula:

$$\text{Residues} = \frac{H_s}{H_{std}} \times \frac{W_{std}}{W_s} \times \frac{V_s}{A_{sj}}$$

Where,

H_s - Peak height of the sample in cm

H_{std} - Peak height of standard in cm

W_{std} - Weight of standard injected in ng

W_s - Weight of sample in g

V_s - Volume of sample (final extract in ml)

A_{sj} - Aliquot of sample injected in μl

3.6. Statistical analysis

3.6.1. Laboratory studies

The corrected per cent mortality for laboratory studies was worked out by using Abott's (1925) correction.

$$\text{Corrected percent mortality} = \frac{(Pt - P_c)}{(100 - P_c)} \times 100$$

Where,

Pt - Observed mortality in treatment

P_c - Observed mortality in untreated check

The data of percentage values were transformed to arc sine (angular) values and analysed by completely randomized design. The treatment mean values of the experiment were compared using Duncan's Multiple Range Test (DMRT) (Gomez and Gomez, 1984).

3.5.2. Residue dissipation studies

The residue data were analysed following the methods suggested by Hoskins (1961) and Timme *et al.* (1986). The pesticide decay curves were fitted based on the procedure described by Regupathy and Dhamu (2001) and the half-lives and best fit degradation models arrived at:

$$\text{Half life } (T_{0.5}) = \frac{\log 2}{K_1}$$

Where,

K_1 = Slope or regression co-efficient from a plot of log $\mu g\ g^{-1}$ residues (Y)

3.5.3. Field studies

The natural enemies populations in the field were transformed to $\sqrt{x + 0.5}$ and analysis of variance was carried out by randomized block design (Panese and Sukhatme; 1958) and means were separated by Duncan's Multiple Range Test (DMRT) (Gomez and Gomez, 1984).

CHAPTER IV
EXPERIMENTAL RESULTS

The results of the various experiments conducted in the laboratory and field to evaluate the phytotonic and phytotoxic effects of Boom Flower®-n in okra, its residues in the harvestable produce and sensitivity to natural enemies non target organisms *viz*., pollinators, fish and earthworms are presented in this chapter.

4.1. Phytotonic effect of Boom Flower®-n formulation on okra

4.1.1. Plant height

Applications of Boom Flower®-n on okra increased the plant height in both the trials and the results are presented in Tables 2 &3. In both the cases, all the Boom Flower®-n treatments were superior to control in increasingly plant height.
In the first trial conducted at Subramanianagar during December to February (2003-2004), among all the doses, Boom Flower®-n application at 3 ml lit^{-1} registered maximum plant height. The plants were 27.13, 61.10 and 184.11 cm tall on 30^{th}, 50^{th} and 60 days after sowing or spraying respectively at this concentration. This was followed by application at 4 ml lit^{-1}, where the plants were 25.50, 59.10 and 178.80 cm tall on the same days of observation respectively. Whereas, in control the plants were only 20.80, 47.10 and 130.60 cm tall on the same days of observation. A more or less similar trend was observed in the confirmatory trial conducted at Chinnamathampalayam during March to May, (2004).

4.1.2. Leaf production

Applications of Boom Flower®-n on okra has increased the number of leaves in both the trials and results are presented in Tables 4 and 5. In both the cases, all the Boom Flower®-n treatments were superior to control. Among all the doses evaluated, Boom Flower®-n application at 3 ml lit^{-1} registered maximum number of leaves with 10.60 -11.30, 27.30 - 28.40, and 41.30 - 41.50 leaves on 30, 50 and 90 DAS, respectively. Where as in untreated control, there were only 5.70-6.20, 15.50-16.40, and 29.80-27.60 leaves.

4.1.3. Plant canopy

The data on number of branches and leaf area are presented in Tables 6 and 7. Application of Boom Flower®-n on okra increased number of branches with the maximum number of branches (3.90 per plant) when compared to control (2.00 per plant) in the first trial . More over remarkable increase in the leaf area was recorded in plots, treated with Boom Flower®-n. Consequently number of leaves produced varied. In control plot, on 90^{th} day of application, the leaf area per plant was 1684.70 cm^2, whereas in plots sprayed with Boom Flower®-n at 3 ml lit^{-1}, it was 6896.77 cm^2. A more or less similar trend was observed in the confirmatory trail also.

4.1.4. Chlorophyll

The influences of Boom Flower®-n on chlorophyll content of the leaves are presented in Table 8. In all the stages of observation, significantly more amount of chlorophyll was produced. Among the different treatments of Boom Flower®-n, application at 3 ml lit^{-1} produced more chlorophyll (2.14 mg g^{-1}) during flowering phase), whereas it was 1.70 mg g^{-1} in control. A more or less similar trend was observed in the confirmatory trail also.

4.1.5. Yield parameters

a. Flowers

Application of Boom Flower®-n on okra significantly increased the flower production (Table 9). All the Boom Flower®--n treatments, significantly increased the numbers of flowers produced per plant over control. The highest number of 25.20 flower per plant was recorded in the plot treated with Boom Flower®-n at 3 ml $lit^{-1.}$ It was followed by 4 ml lit^{-1} (21.00 flowers $plant^{-1}$), while the lowest number of flowers (16.40) was recorded in control. A more or less similar trend was followed in the confirmatory trail conducted at Chinnamathampalayam (Table 10).

b. Fruits

Applications of Boom Flower® -n not only increased the fruit production but also improved the quality of the fruits (Table 9). Application of Boom Flower® -n at 3 ml lit^{-1} produced more number of fruits (25.50 per plant), 7.88 cm girth, 23.23 cm length and 20.72 g weight, which were significantly superior to other treatments. Whereas in control the same were 15.20 fruits per plant with 6.20 cm, 10.90 cm and 13.30 g respectively. A more or less similar trend was followed in the confirmatory trial conducted at Chinnamathampalayam (Table 10).

c. Yield

Consequently application of Boom Flower® -n inflicted in increasing the yield and the results on yield increase are presented in Tables 9 and 10. In both the seasons all the Boom Flower® -n treatments significantly increased the yield. Application at 3 ml lit^{-1} recorded the highest yield 7.58 - 7.65 t ha^{-1} when compared to control where as the yield of 4.80 - 4.92 t ha^{-1} was recorded.

4.2. Evaluation of Boom Flower® -n for phytotoxicity on okra

Application of Boom Flower® -n at all concentrations did not cause any phytotoxic symptoms like injury to leaf dip and leaf surface, wilting, vein clearing, necrosis, epinasty and hyponasty on okra in both the field trials conducted at farmers field (Table 11).

4.3. Sensitivity of natural enemies to Boom Flower® -n

Experiments were conducted in the laboratory as well as in the field to assess the sensitivity /safety of Boom Flower® -n to natural enemies

4.3.1. Laboratory studies

4.3.1.1. *Trichogramma chilonis* (Ishii)

The influence of Boom Flower® -n on adult emergence and parasitization of *T.chilonis* was studied under laboratory conditions and presented Table 12. The results revealed that adult emergence was 92.75 and parasitizations 90.00 were the maximum in

untreated check. The adult emergence ranged from 66.25 to 85.85 per cent, and parasitization ranged from 67.00 to 84.75per cent, due to the exposure to Boom Flower® -n. Both these parameters were observed to be reduced as the concentration of Boom Flower® -n increased.

4.3.1.2. *Chelonus blackburni* (Cameron)

The results on the sensitivity of the *C. blackburni* to Boom Flower® -n by dry film method revealed that higher doses of Boom Flower® -n 5ml lit^{-1} recorded higher mortality (72.75%) compared to other doses of Boom Flower® -n and untreated control (9.50 %) at 48 h after treatment (Table 13).

The influence of Boom Flower® -n on the emergence of adults is presented in Table 14. An increase in the concentration of Boom Flower® -n exhibited a decrease in adult emergence. The adult emergence was 97.5 per cent in untreated check, where as it was 82.00, 76.75, 67.50, 61.00 and 57.25 per cent, when Boom Flower® -n was applied at 2, 2.5, 3, 4 and 5ml, respectively (Table 14).

4.3.1.3 *Chrysoperla carnea* (Stephens)

a. Eggs

The effect of Boom Flower® -n on egg hatchability of *C. carnea* was studied under laboratory condition and presented in Table 15. The experimental results revealed that per cent hatchability was maximum (85.50) in untreated check, followed by Flower® -n at 2ml lit^{-1} (81.50%), 2.5 ml lit^{-1} (78.00%)and 3 ml lit^{-1} (75.75 %) with the least in Boom Flower® -n at 5 ml lit^{-1} (69.50%) 24 h. A slight increase in hatchability was observed after 48h.

b. Grubs

(i) Contact toxicity method

The effect of Boom Flower® -n on the grubs of *C. carnea* was studied by contact toxicity method under laboratory condition. The results (Table 16) revealed that Boom Flower® -n at high dose (5 ml lit^{-1}) recorded higher per cent mortality (43.00%) compared

to other doses and untreated check (4.00 %) after 12 h of treatment. However, after 48 h of treatment, except control (15.60 %), all the treatments recorded significantly higher mortalities of 40.00, 54.25, 68.00, 73.00 and 82.00 per cent, when Boom Flower® -n was treated at 2, 2.5, 3, 4 and 5 ml lit^{-1}, respectively.

(ii) Larval feeding method

The results on the effect of Boom Flower® -n to *C. carnea* grubs determined by larval feeding method (Table 17) revealed that all the doses of Boom Flower® -n tested caused significant mortality. The highest mortality (75.50%) in 48 h was observed, when Boom Flower® -n was applied at 5 ml $lit^{-1.}$ This was followed by Boom Flower® -n applied at 4ml lit^{-1} (64.00%), 3 ml lit^{-1} (56.30%), 2.5ml lit^{-1} (40.80%), and 2 ml lit^{-1} (39.50%) as against 18.30 per cent mortality in control.

c. Adults

The results on the sensitivity of *C. carnea* adults to Boom Flower® -n are presented in Table 18. The results revealed that the adult longevity was a maximum of 16 days in untreated check, while it was 6.60, 10.50, 11.50, 12.50 and 13 days when Boom Flower® -n was applied at 5, 4, 3, 2.5 and 2 ml lit^{-1} respectively. Like wise, significantly more number of eggs were laid in untreated check (414.80 eggs per 5 females). However, the fecundity was reduced to 136.50, 131.50, 129.50, 119.50 and 90.60 eggs per 5 females, when the adults were exposed to Boom Flower® -n at 2, 2.5, 3, 4, and 5 ml lit^{-1}, respectively.

43.2. Field studies

a. Chrysopids

Statistical analysis of the data on green lacewings after the application of Boom Flower® -n in field (Table19) revealed that the population of the predator was significantly higher in untreated check (6.33 to 10.67 adults per 10 plants) when

compared to Boom Flower® -n 2ml lit^{-1} (3.30 to 10.00 adults per 10 plants). The activities of green lacewings were very low at higher concentration of Boom Flower® -n (2.63-5.33 adults per 10 plants). It was also interesting to note that generally the activities of green lacewings were lower after the application of Boom Flower® -n and slowly the population built up along with time.

b. Coccinellids

The population of coccinellids varied from 7.00 to 7.33 from the 10 plants before the application of Boom Flower® -n. After a day of application, plots applied with higher concentration of Boom Flower® -n registered significant reduction in coccinellids activity. The reduction was not well pronounced at lower concentrations. However, a slow increase in the population of the predator was noticed along with time. After 14 days of application, the populations of coccinellids in plots applied with lower concentrations of Boom Flower® -n were as good as in control (Table 20).

c. Spiders

The population of spiders varied from 7.30 to 8.30 from 10 plants before the application of Boom Flower® -n. After a day of application, plots applied with higher concentration of Boom Flower® -n registered significant reduction in spiders activity (5.0 spiders per 10 plants). The reduction was not well pronounced at lower concentration. However, a slow increase in the population of the spiders was noticed as the time has gone by. After 14 days after application, the populations of spiders in plots applied with lower concentrations of Boom Flower® -n were as good as number in control (Table 21).

4.3. Sensitivity of honey bees to Boom Flower® -n

Results of experiments conducted to assess the sensitivity of honey bees to Boom Flower® - n are presented in Tables 22, 23 and 24. The results revealed that honey bees were less sensitive to Boom Flower® -n. After 24 h of treatment with Boom Flower®-n at

2, 2.5, 3 and 4ml lit^{-1} recorded 35.00, 39.50, 45.50, 55.00 per cent mortality of adults of *Apis cerana indica* (F.) as against 18 per cent in untreated control (Table 22). Boom Flower® -n at 5 ml lit^{-1} recorded 63 percent mortality after 24 h of treatment (Table 22).

In the case of Italian bees, *Apis mellifera* (L.) after a day of treatment with Boom Flower® -n at 2ml, 2.5ml, 3ml and 4ml lit^{-1} was recorded 36.50, 40.75, 44.50, 55 and 62.50 percent mortality as against 18.50 percent in untreated control (Table 23).

More or less similar trend was observed in the case of dammer bees, *Trigona irridipennis* (Smith) (Table 24). After a day of application of Boom Flower® -n at 2, 2.5, 3, 4 and 5ml lit^{-1}, 29.50, 38.50, 44.50, 55.25, and 63.00 per cent mortalities, respectively were recorded as against 16.50 per cent in untreated check.

In addition, the sensitivity of bees to Boom Flower® -n was recorded by observing their visits to the plots sprayed with the chemical (Table 25). The number of visits made by honeybees ranged from 27.50 to 30.00 per 30 min before spraying Boom Flower® -n. Plots treated with 10 ml lit^{-1} recorded 13.00, 18.33 and 21.00 visits per 30 min after 1, 3, and 7 days after treatments, respectively. Even after 3 days of treatment, the maximum dose of Boom Flower® -n 10ml lit^{-1} recorded only 18.33 visits per 30 min, when compared to control, which recorded 30 visits per 30 min. This was followed by Boom Flower® -n at 2.5ml lit^{-1} (25.33 visits per 10 min.) and 3ml lit^{-1} (26.00 visits per 10 min.) which were on par. However, an increase in the frequency of visits by the honey bees increased with in a week was observed as the chemical is bound to lose its effect in due course.

4.4. Sensitivity of Boom Flower® n against non target organisms

4.4.1. Acute toxicity of Boom Flower® -n to earth worms *Eudrilus eugeniae* (Kinberg) and *Perionyx excavatus* (Perrier)

The LC_{50} values of Boom Flower® -n to *E. euginea* and *P. excavatus* by artificial soil test bioassay method were 7.89 and 6.62 g kg^{-1} of soil, respectively (Table 26).

The results revealed that LT_{50} values of Boom Flower® - n at the concentrations of 8.0, 8.2, and 8.4 g kg^{-1} of soil, were 6.71, 5.62, and 4.21 days for *E. euginea* and 6.6, 6.8, and 7.0 g kg^{-1} of soil, were 6.90, 5.81 and 4.86 days for *P. excavatus* (Tables 27 and 28) (Fig 1 and 2).

4.4.2. Acute toxicity of Boom Flower® -n to *C. carpio* and *T. mosambica*

The LC_{50} of Boom Flower® -n formulation to common carp *C. carpio* and *T. mosambica* by static method was 70.37 and 165.63 ppm, respectively (Table 29). The LT_{50} values of Boom Flower® n to common carp at the concentrations of 70, 80, and 90 ppm were 47.80, 25.21 and 10.55 h, respectively. Whereas, for *T. mosambica*, they were 35.00, 15.63 and 11.88 h respectively at the concentrations of 170, 180 and 190 ppm (Table 30 and 31) (Fig 3 and 4).

4.5. Residues

4.5.1. Residues of Boom Flower® -n

The standard chromatogram of Boom Flower® -n was illustrated in Fig 5. The mean recovery was81.26 per cent from samples fortified at 0.5, 1 and 2μg g^{-1} level. The minimum detection limit of the instrument was 5 ng and the determinability level in the sample was 0.05μg g^{-1} considering the weight of the sample as 50 g and final volume of the extract as 1.0 ml with a infection volume of 2 μl.

4.5.1.1. Dissipation of Boom Flower® -n

In the field trial I, Boom Flower® -n at the concentration of 2, 3, 4, and 8ml lit^{-1} left initial deposits of 0.167, 0.334, 0.723 and 1.506 μg g^{-1}, respectively, on okra fruits. The initial deposits dissipated from 33.53 to 77.73 per cent on first DAT and the residues reached below detectable level (BDL) at doses of 2, 3 ml lit^{-1} on 3 DAT. The rate of dissipation was faster for Boom Flower® -n (Table 32 and 33). After linearization of the

residue data, the Boom Flower® -n concentration at 3,4 and 5 ml lit^{-1} followed first order root function (RF) first order and RF first order with a half life values of 0.193, 1.442 and 0.456 days respectively (Table 34 and 35).

In the field trial II, spraying of Boom Flower® -n at 2, 3, 4, and 8ml lit^{-1} left initial deposits of 0.111, 0.222, 0.634 and 0.890 $\mu g\ g^{-1}$, respectively on okra fruits, which dissipated by 33.78 to 100.0 per cent in 1 DAT and reached below detectable level (BDL) at 7 DAT in all the concentrations. The rate of dissipation was comparatively faster in trial I than that of trial II. The residue decay curve (Fig 6 and 7) followed first order, root function second order and inverse power law for 3, 4 and 8 ml lit^{-1} concentration respectively, with the half life values ranged from 0.924 -2.198 days (Table 34 and 35).

CHAPTER V
DISCUSSION

The results obtained from the field experiments to evaluate the phytotonic and phytotoxic effects of Boom Flower®-n (Nitrobenzene 20% v/v) in okra, its residues in harvestable produce, sensitivity of natural enemies and pollinators and toxicity to fish and earthworms are discussed in this chapter. Since Boom Flower®-n is used in agriculture sparsely, the availability of literature are scanty and hence its performance has been compared with other plant growth regulators.

5.1. Phytotonic effect

Generally Boom Flower®-n positively aided the growth and yield parameters. In all the stages of observation growth parameters seemed to have a positive relationship with the application of Boom Flower®-n. Among the different concentrations applied, 3 ml lit^{-1} recorded maximum increase in growth parameters such as plant height, plant canopy, leaf production, leaf area and chlorophyll content. Datta *et al*. (1986) proved that nitrophenol, a closely related compound to nitrobenzene stimulated plant activity without causing malformation or toxicity to plant and accelerated the plasma streaming. Datta and Nanda (1985) also suspected that a linear positive relationship between growth parameters and brassinosteroid might be due to the increase in the endogenous auxin level by exogenous application.

In the same way Datta and Nanda (1985) and Zhao and Lin (1993) postulated that synergistic interaction of salicylic acid with available endogenous auxins could induce cell wall plasticity and cell elongation in soybean. Likewise benzyladenine, a synthetic cytokinin ($C_{10}H_9N_{50}$) had been proved in promotion of cell division, delaying of senescence, contraction of apical bud dominance, flowering induction in short day plants, translocation of assimilated metabolites and thereby affecting the yield potential of

plants. Based on these circumstantial evidences, it is suspected here also that Boom Flower-n also interfered in physiological function of okra in improving the growth parameters.

It is a well known tenet that any improvement in the growth parameters, could directly inflict in improving the yield. The increases in plant height, plant canopy, leaf area and chlorophyll content in okra certainly have enhanced positively the photosynthetic ability. This has resulted in improved yield. For example, in the present study, Boom Flower®-n applied at the concentration of 3 ml lit.$^{-1}$ has increased the plant height (Fig 8.), number of branches, leaf production and chlorophyll content, which had a direct impact on the production of quality fruits (Plate 12). Consequently the yield of okra has been increased by 54 and 59 per cent in trial I and II yield, respectively (Fig. 9).

5.2. Phytotoxic effect

The results of the field experiment on the foliar application of Boom Flower®-n indicated that even at the concentration of 10.0 ml lit.$^{-1}$, which is approximately three to four times more than the normal dose, did not cause any phytotoxic symptoms such as injury to leaf tip and leaf surface, wilting, vein clearing, necrosis, epinasty and hyponasty on okra. The central insecticides Board insisted to generate data for any new chemicals at 4x dose for phytotoxicity evaluation. This Boom Flower®-n is safer, if farmers used this chemical at higher doses.

5.3. Sensitivity of natural enemies to Boom Flower-n

Experiments were conducted both in the laboratory as well as in the field to evaluate the sensitivity of natural enemies and beneficial insect fauna to Boom Flower®-n. Generally Boom Flower®-n is not toxic to the natural enemies and beneficial insects. In the case of the egg parasitoid, *T. chilonis* as well as *C. blackburni*, adult emergence and parasitization efficiency (Figs. 10 and 11) were more than 80 per cent at recommended doses, although a

mortality of 50 per cent was noticed due to continuous exposure of the chemical in laboratory in a closed container. However, small amount of Boom Flower®-n released in the air incidentally after application is not expected to hinder the activities of these parasitoid. Nevertheless, predatory green lacewing (Fig. 12 and 13) was found to be moderately sensitive to Boom Flower-n.

Generally nitrobenzene was found to be less toxic to aquatic insects such as water fleas, *Daphnia magna* Straus and *Ceriodaphina dupia* and midge, *Culex pipens* Linnaeus, where the insects happen to be continuously exposed to nitrobenzene (Bringmann and Kuhn, 1980; Canton *et al*., 1985). In the present study, the probability of insects to come in contact Boom Flower®-n was very less and hence in the field Boom Flower®-n is not expected to hinder the normal life cycle of the natural enemies and beneficial insects. A perusal of data presented in Tables 19, 20, 21 and 25 support this argument where the activities of chrysopids, coccinellids, spiders and honey bees (Fig.14) were not as hindered as by an insecticide. Immediately after application of Boom Flower®-n, the activities of these natural enemies were found to be reduced. Nevertheless, the activities of these natural enemies and the visitation of pollinator honeybees rebounded with in a few days of application. This proved that Boom Flower®-n did not affect the natural enemy complex and beneficial insect fauna in the field.

5.4. Sensitivity to non target organisms

Environment Protection Agency (EPA) has identified seven sites to be risky out of the 1770 national priority sites of risks. Hence, toxicity of Boom Flower®-n against certain non target organisms were determined.

Earthworms are known to improve the quality of the soil continuously and fish is one of the components of integrated farming in certain parts of the country. LC_{50} of Boom Flower®-n against the two species of earthworms tested were very high (7.89 and

6.62 g kg^{-1} of soil); likewise the LC_{50} of two fish tested were also very high and the possibilities of such high concentrations of nitrobenzene to occur in the environmental samples such as soil and water are highly improbable. Even if, such high concentrations of nitrobenzene occur they would breakdown very fast. Although Rickart *et al*. 1983 implicated in rats that nitrobenzene in pure form at continuous exposure to cause irritation to skin and eyes, methemoglobinemia in blood and reduced level of sperm due to long hours of exposure, which may not occur in reality. The ability of nitrobenzene to breakdown in the environment rather quickly is proved in one of the experiments investigated in the present study explained *passim*.

5.5. Residues

Boom Flower®-n dissipates in the environment very fast. A perusal of the data recorded in the terminal harvestable produce revealed that at recommended dose of application such as 2 to 3 ml $lit.^{-1}$ Boom Flower®-n dissipated in less than three days. However, at the higher concentration (8 ml $lit.^{-1}$) terminal residues were observed upto five days. The residues may be diluted due to the faster growth of okra fruits. Hastings and Hasten (1948) reported that exposure of nitrobenzene to ultraviolet rays in air hassens the photodegradation. It is reported that photochemical decomposition to be the most important route of removal of nitrobenzene in the environmental (EPA, 1989). Tabak *et al*. (1989) was of the opinion that biodegradation of nitrobenzene from industrial and domestic waste water occur in 7 days. These supporting evidences along with the present investigations reemphasise the quick degradation of nitrobenzene.

CHAPTER VI

SUMMARY

The results of laboratory and field experiments carried out to determine the phytotonic and phytotoxic effects of Boom Flower®-n (Nitrobenzene 20% v/v), its safety to non target organisms, and residues in the harvestable produce in okra are summarised in this chapter.

- Boom Flower® -n as foliar application has significantly increased the plant growth and yield parameters such as plant height, number of leaves produced, leaf area, fruit weight, fruit length, fruit girth and fruit production per plant. Maximum increase was observed at a dose of 3 ml lit^{-1}. Other treatments had significant increase, when compared to control.
- All the Boom Flower® -n treatments although resulted in improving the growth and yield parameters, did not cause any phytotoxic symptoms in both the trials.
- The influence of Boom Flower® -n on adult emergence and per cent parasitisation of *Trichogramma chilonis* Ishii experimented by direct spraying on egg card revealed that there was no significant adverse effect at lower concentration. After 24 h of treatment, Boom Flower -n at different doses recorded 66.25 to 85.85 per cent adult emergence and 67.00 to 84.75 per cent parasitisation. In the case of *Chelonus blackburni*(Cameron), application at 5 ml lit^{-1} resulted higher mortality (75.75%) when compared to application at 3 ml lit^{-1} (35.50%) and control (9.50%) after 48h of application. The rate of parasitization also varied from 57.25 per cent at higher concentration (5 ml lit^{-1}) and 76.75 % at lower dose (3 ml lit^{-1}).
- The studies on the egg hatchability of *Chrysoperla carnea* Stephens indicated that the ovicidal action was less at 2 ml lit^{-1} (13.00%) medium at 2.5 ml lit^{-1} (20.75%) and maximum at 5 ml lit^{-1} (29.25%) after 48 h of treatment.
- In the field, population of green lacewing and coccinellids reduced initially, after the application of Boom Flower® -n. But, it rebounded back at lower dose with in a

week. Spiders on the other hand, remained insensitive to Boom Flower -n at lower concentration; but, at higher concentration, after an initial reduction, thus rebounded with in a week.

- *C. carnea* grubs were found to be moderately sensitive to Boom Flower® -n. After 48 h of treatment at 2 ml lit^{-1}, 40 per cent mortality was observed when tested, either by larval feeding and thin film method. Longivity of the adults was affected only at high concentration, but fecundity was drastically reduced by 66-80 per cent.
- Generally, all the species of honey bees were observed to be sensitive to Boom Flower® -n at recommended dose as approximately a mortality of 30 per cent was noticed when compared to control (16.00 %). In the field, a reduction in the frequency of honey bee visitation was observed on the intial days after treatment. However an increase was observed with in a week.
- The LC_{50} of Boom Flower® -n to *Eudrilus eugeniae* (Kinberg) and *Perionyx excavatus* (Perrier) were 7.89 and 6.62 g kg^{-1} of soil.
- The LC_{50} values of Boom Flower® -n to *Cyprinus carpio* (L.) and *Tilapia mosambica* (Peters) were 70.37 and 165.53 ppm.
- The LT_{50} of Boom Flower® -n to *E. eugeniae* are 6.71, 5.62 and 4.21 days at a concentration of 8.0, 8.2 and 8.4 g kg^{-1} of soil, respectively and while that for *P. excavatus* the LT_{50} values were 6.90, 5.81 and 4.86 days at the concentrations of 6.6, 6.8 and 7.0 g kg^{-1} of soil, respectively.
- The LT_{50} values of Boom Flower -n to *C. carpio* were 47.80, 25.21 and 10.55 h at 70, 80 and 90 ppm respectively and while the same were 35.10, 15.63 and 11.88 h for *T. mosambica* at 170, 180 and 190 ppm respectively.
- In both the field trials, Boom Flower -n dissipates from okra fruits with in two days at lower concentration. However, at higher concentration, the rate of dissipation was rather slow and took nearly a week to reach below detectable level.

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Table 2. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) on plant height of okra (Field experiment I)

Rate of application (ml lit^{-1})	I Spray	II spray		III Spray	
	PTC on 20 DAS	30 DAS	40 DAS	50 DAS	60 DAS	90 DAS
2.0	8.70 [a]	24.03 [d]	35.60 [f]	58.50 [d]	114.57 [b]	178.67 [bc]
2.5	8.77 [a]	25.63 [b]	35.13 [f]	57.30 [f]	114.80 [b]	178.57 [c]
3.0	8.80 [a]	27.13 [a]	38.40 [a]	61.10 [a]	117.20 [a]	184.11 [a]
4.0	8.80 [a]	25.50 [b]	37.37 [bc]	59.10 [b]	114.73 [b]	178.80 [b]
5.0	8.77 [a]	25.50 [b]	36.87 [bc]	58.80 [c]	113.80 [bc]	178.53 [c]
8.0	8.80 [a]	24.61 [c]	36.37 [cd]	58.10 [e]	113.00 [c]	168.83 [d]
10.0	8.77 [a]	22.80 [e]	36.20 [de]	57.20 [f]	109.57 [d]	167.40 [e]
Untreated control	8.83 [a]	20.86 [f]	24.23 [g]	47.10 [g]	80.80 [e]	130.60 [f]

PTC – Pre treatment count; DAS – Days after treatment
Observations are made in 10 tagged plants in three replications each
In a column means followed by common alphabet are not significantly different by DMRT (p=0.05).

Table 4. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) on number of leaves of okra (Field experiment I)

Rate of application (ml lit^{-1})	I Spray		II spray		III Spray	
	PTC on 20DAS	30DAS	40DAS	50DAS	60DAS	90DAS
2.0	4.20 [a]	10.20 [bc]	14.50 [b]	26.50 [b]	32.10 [f]	40.20 [b]
2.5	4.30 [a]	10.30 [bc]	14.50 [b]	26.60 [b]	32.50 [e]	39.40 [c]
3.0	4.30 [a]	10.60 [a]	16.00 [a]	27.30 [a]	33.80 [a]	41.30 [a]
4.0	4.20 [a]	10.50 [ab]	14.50 [b]	26.70 [b]	33.10 [b]	40.20 [b]
5.0	4.20 [a]	9.80 [d]	13.70 [b]	25.70 [c]	32.80 [c]	38.30 [b]
8.0	4.30 [a]	9.70 [d]	13.70 [b]	24.70 [d]	32.60 [d]	37.20 [e]
10.0	4.20 [a]	9.50 [e]	14.50 [b]	25.50 [e]	32.40 [e]	36.30 [f]
Untreated control	4.30 [a]	5.70 [e]	11.50 [c]	15.50 [f]	19.60 [g]	29.80 [g]

PTC – Pre treatment count; DAS – Days after treatment
Observations are made in 10 tagged plants in three replications each
In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)

Table 6. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) on plant canopy (Field experiment I)

Rate of application (ml lit^{-1})	Number of branches/ plant on 90 DAS	Leaf area /plant (cm^2)		
		30DAS	60DAS	90DAS
2.0	2.70[e]	1041.17[g]	4397.40[d]	5831.80[c]
2.5	2.80[de]	1177.60[e]	4493.80[c]	5996.73[b]
3.0	3.90[a]	1733.40[a]	5336.40[a]	6896.77[a]
4.0	3.30[b]	1378.53[b]	4611.43[b]	5946.73[d]
5.0	3.20[b]	1309.50[c]	4430.50[c]	5152.43[e]
8.0	3.10[c]	1207.50[d]	3829.30[g]	4943.40[f]
10.0	2.90[d]	1138.67[f]	3957.43[f]	4876.00[g]
Untreated control	2.00[f]	578.50[h]	1476.63[h]	1684.70[h]

DAS – Days after treatment
Observations are made in 10 tagged plants in three replications each
In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)

Table 8. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) on total chlorophyll content (mg/g)

Rate of application (ml lit^{-1})	Vegetative stage (30 DAS)		Flowering stage(40DAS)		Maturity stage (90DAS)	
	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2
2.0	1.50 ^{b}	1.47^{bc}	2.01 ^{b}	2.00^{b}	1.83 ^{ab}	1.77^{d}
2.5	1.51 ^{b}	1.49^{b}	1.94 ^{c}	1.91^{d}	1.81 ^{abc}	1.81^{c}
3.0	1.58 ^{a}	1.57^{a}	2.14 ^{a}	2.25^{a}	1.92 ^{a}	1.94^{a}
4.0	1.49 ^{bc}	1.48^{b}	1.90 ^{c}	1.97^{c}	1.78 ^{bc}	1.83^{b}
5.0	1.52 ^{b}	1.46^{c}	1.85 ^{d}	1.84^{e}	1.70 ^{cd}	1.80^{c}
8.0	1.46 ^{c}	1.44^{c}	1.78 ^{e}	1.75^{f}	1.62 ^{de}	1.73^{e}
10.0	1.47 ^{c}	1.36^{e}	1.75 ^{f}	1.72^{g}	1.51 ^{e}	1.62^{f}
Untreated control	1.42 ^{d}	1.20^{f}	1.70 ^{g}	1.61^{h}	1.34 ^{f}	1.31^{g}

DAS– Days after treatment.

Observations are made in 10 tagged plants in three replications each

In a column means followed by common alphabet are not significantly different by DMRT (p=0.05).

Table 9. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) on yield parameters of okra (Field experiment I)

Rate of application (ml lit^{-1})	Number of flowers per plant	Number of fruits per plant	Fruit girth(cm)	Fruit length(cm)	Fruit Weight (g)	Yield kg ha^{-1}
2.0	19.40 ^{ef}	18.60^{f}	7.41 ^{c}	17.45 ^{bc}	16.43 ^{e}	5703.00 ^{g}
2.5	19.90 ^{cd}	19.10 ^{d}	7.56 ^{b}	17.93 ^{bc}	17.62 ^{d}	5800.33 ^{f}
3.0	25.50 ^{a}	25.50 ^{a}	7.88 ^{a}	20.23 ^{a}	20.72 ^{a}	7578.33 ^{a}
4.0	21.00 ^{b}	21.50 ^{b}	7.20 ^{d}	18.41 ^{b}	18.52 ^{b}	6826.00 ^{b}
5.0	20.00 ^{c}	19.80 ^{c}	6.52 ^{d}	14.67 ^{d}	18.00 ^{c}	6500.33 ^{c}
8.0	19.60 ^{de}	18.90 ^{e}	6.91 ^{e}	17.22 ^{bc}	17.48 ^{d}	5926.00 ^{d}
10.0	19.20 ^{f}	18.60 ^{f}	6.78 ^{f}	16.87 ^{c}	16.30 ^{e}	5807.67 ^{e}
Untreated control	16.40 ^{g}	15.20 ^{g}	6.20 ^{g}	10.90 ^{e}	13.30 ^{f}	4916.00 ^{h}

In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)
Observations are made in 10 tagged plants in three replications each

Table 3. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) on plant height of okra (Field experiment II)

Rate of application (ml lit^{-1})	I Spray		II spray		III Spray	
	PTC on 20DAS	30DAS	40DAS	50DAS	60DAS	90DAS
2.0	9.00[a]	26.40[b]	36.57[cd]	59.37[bc]	115.30[c]	177.27[d]
2.5	8.77[a]	26.96[c]	36.63[cd]	58.80[bc]	115.63[b]	177.70[c]
3.0	9.00[a]	28.50[a]	38.37[a]	63.60[a]	119.80[a]	186.20[a]
4.0	9.03[a]	27.57[b]	37.43[b]	60.40[b]	115.20[c]	178.40[b]
5.0	8.97[a]	26.67[cd]	37.10[bc]	59.13[bc]	113.20[d]	176.47[c]
8.0	9.03[a]	25.70[e]	36.37[d]	58.10[c]	112.06[e]	175.67[f]
10.0	9.00[a]	24.77[f]	35.17[e]	54.73[e]	101.13[f]	169.10[g]
Untreated control	9.03[a]	20.20[g]	23.03[f]	46.91[e]	81.40[g]	132.30[h]

PTC – Pre treatment count; DAT – Days after treatment
In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)
Observations are made in 10 tagged plants in three replications each

Table 5. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) on number of leaves of okra (Field experiment II)

Rate of application (ml lit^{-1})	I Spray		II spray		II Spray	
	PTC on 20DAS	30DAS	40DAS	50DAS	60DAS	90DAS
2.0	4.50 [a]	10.00 [bc]	16.30 [d]	26.60 [c]	33.40 [c]	39.10 [d]
2.5	4.50 [a]	10.50 [b]	16.50 [c]	27.40 [b]	34.47 [b]	39.70 [c]
3.0	4.50 [a]	11.30 [a]	18.30 [a]	28.40 [a]	35.80 [a]	41.50 [a]
4.0	4.40 [a]	10.50 [b]	17.60 [b]	27.30 [b]	34.80 [b]	40.10 [b]
5.0	4.50 [a]	10.20 [c]	16.30 [e]	26.30 [d]	34.60 [b]	40.00 [b]
8.0	4.40 [a]	9.80 [d]	16.10 [e]	26.70 [d]	34.10 [b]	38.70 [e]
10.0	4.50 [a]	9.60 [d]	15.70 [f]	25.70 [e]	32.10 [d]	37.53 [f]
Untreated control	4.40 [a]	6.20 [e]	12.20 [g]	16.40 [f]	20.20 [e]	27.60 [g]

PTC – Pre treatment count; DAS – Days after sowing.
Observations are made in 10 tagged plants in three replications each
In a column means followed by common alphabet are not significantly different by DMRT (p=0.05).

Table 7. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) on plant canopy (Field experiment II)

Rate of application (ml lit^{-1})	Number of branches/ plant on 90 DAS	Leaf area /plant (cm^2)		
		30DAS	60DAS	90DAS
2.0	2.70[e]	1045.27[g]	4675.30[f]	5930.93[c]
2.5	2.90[d]	1178.53[d]	4982.93[d]	5947.20[c]
3.0	3.90[a]	1379.33[a]	5343.03[a]	6872.21[a]
4.0	3.60[b]	1302.53[b]	5276.07[b]	6274.37[b]
5.0	3.30[c]	1208.13[c]	4986.17[c]	5835.97[c]
8.0	3.40[c]	1174.40[e]	4971.57[e]	5488.97[d]
10.0	2.80[c]	1138.93[f]	4432.10[g]	4934.91[e]
Untreated control	2.20[f]	588.37[h]	1878.33[h]	1749.07[f]

PTC-Pre treatment count; DAS-Days after sowing.
Observations are made in 10 tagged plants in three replications each
In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)

Table 10. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) on yield parameters (Field experiment II)

Rate of application (ml lit^{-1})	Number of flowers per plant	Number of fruits per plant	Fruit girth(cm)	Fruit length (cm)	Fruit Weight (g)	Yield kg ha^{-1}
2.0	19.20 [b]	19.10 [d]	7.37 [c]	16.83 [b]	17.25 [cd]	5981.33 [f]
2.5	19.80 [b]	19.20 [d]	7.60 [b]	16.33 [b]	18.48 [b]	6201.33 [e]
3.0	23.60 [a]	24.70 [a]	8.21 [a]	19.06 [a]	22.58 [a]	7650.33 [a]
4.0	19.90 [b]	21.70 [b]	7.63 [b]	17.01 [b]	17.52 [cd]	6901.00 [b]
5.0	20.10 [b]	19.80 [c]	7.43 [c]	15.83 [b]	18.11 [bc]	6703.33 [c]
8.0	19.20 [b]	18.60 [e]	7.23 [d]	15.63 [b]	16.72 [de]	6603.33 [d]
10.0	19.10 [b]	18.30 [f]	6.87 [e]	15.53 [b]	15.92 [e]	5983.00 [f]
Untreated control	15.18 [c]	16.10 [g]	5.87 [f]	10.77 [c]	13.86 [f]	4801.00 [g]

Observations are made in 10 tagged plants in three replications each
In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)

Table 11. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) on okra (visual phytotoxicity in 0-10 % grade)

Rate of application (ml lit^{-1})	Phytotoxicity rating*									
	Injury to, leaf tip and leaf surface		Wilting		Vein clearing		Necrosis		Epinasty and Hyponasty	
	Trial I	Trial II	Trial I	Trial II	Trial I	Trial II	Trial I	Trial II	Trial I	Trial II
2.0	0	0	0	0	0	0	0	0	0	0
2.5	0	0	0	0	0	0	0	0	0	0
3.0	0	0	0	0	0	0	0	0	0	0
4.0	0	0	0	0	0	0	0	0	0	0
5.0	0	0	0	0	0	0	0	0	0	0
8.0	0	0	0	0	0	0	0	0	0	0
10.0	0	0	0	0	0	0	0	0	0	0
Untreated control	0	0	0	0	0	0	0	0	0	0

*Observed on 1, 3, 5, 7, 10 and 20 days after spraying; 0- No phytotoxicity

Table 12. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) on egg parasitoid *Trichogramma chilonis* (Ishii)

Rate of application (ml lit^{-1})	Adult emergence (%)	Parasitisation (%)
2.0	85.85 (67.85)[b]	84.75 (67.03)[b]
2.5	81.50 (64.55)[c]	83.00 (65.66)[b]
3.0	81.00 (64.18)[c]	82.25 (65.01)[b]
4.0	69.75 (56.64)[d]	76.50 (61.02)[c]
5.0	66.25 (54.48)[e]	67.00 (54.95)[d]
Untreated control	92.75 (74.46)[a]	90.00 (71.62)[a]

In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)
Figures in parentheses are arc sine transformed values.

Table 15. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) on the eggs of *Chrysoperla carnea* (Stephens)

Rate of application	24 HAT		48 HAT	
(ml lit^{-1})	Hatchability %	Mortality %	Hatchability %	Mortality %
2.0	81.50 (64.54)[b]	18.50 (25.47)[b]	87.00 (68.90)[b]	13.00 (21.09)[b]
2.5	78.00 (62.03)[c]	22.50 (27.97)[c]	79.25 (62.91)[c]	20.75 (27.75)[c]
3.0	75.75 (60.51)[d]	24.25 (29.49)[d]	78.75 (62.55)[c]	21.25 (27.45)[c]
4.0	72.00 (58.06)[e]	28.00 (31.94)[e]	73.25 (58.57)[d]	26.75 (31.13)[d]
5.0	69.50 (56.48)[f]	30.50 (33.52)[f]	70.75 (57.26)[d]	29.25 (32.74)[e]
Untreated control	85.50 (67.62)[a]	14.50 (22.38)[a]	91.50 (73.08)[a]	9.50 (17.95)[a]

HAT-Hours after treatment

In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)

Figures in parentheses are arc sine transformed values.

Table 16. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) on the grubs of *C. carnea* by dry film method

Rate of application (ml lit^{-1})	Mortality %		
	12 HAT	24 HAT	48 HAT
2.0	23.10	27.25	40.00
	(28.03) [b]	(31.46) [b]	(39.23) [b]
2.5	27.00	43.00	54.25
	(31.29) [c]	(40.97) [c]	(47.44) [c]
3.0	31.50	58.00	68.00
	(34.14) [d]	(49.60) [d]	(55.55) [d]
4.0	37.80	68.00	73.00
	(37.91) [e]	(55.55) [e]	(58.69) [e]
5.0	43.00	70.00	82.00
	(40.97) [f]	(66.27) [f]	(64.90) [f]
Untreated control	4.00	13.00	15.50
	(11.46) [a]	(21.12) [a]	(23.16) [a]

HAT-Hours after treatment
In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)
Figures in parentheses are arc sine transformed values.

Table 17. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) on the grubs of *C. carnea* by larval feeding method

Rate of application (ml lit^{-1})	Mortality %		
	12 HAT	24 HAT	48 HAT
2.0	23.10 (23.96) [b]	22.30 (28.14) [b]	39.50 (38.44) [b]
2.5	22.30 (28.14) [c]	33.00 (35.06) [c]	40.80 (39.67) [c]
3.0	27.00 (31.30) [d]	37.80 (37.91) [d]	56.30 (48.59) [d]
4.0	33.00 (35.06) [e]	43.00 (40.97) [e]	64.00 (53.13) [e]
5.0	40.75 (39.61) [f]	56.30 (48.59) [f]	75.50 (60.33) [f]
Untreated control	3.50 (10.64) [a]	12.00 (20.24) [a]	18.30 (25.57) [a]

HAT-Hours after treatment
In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)
Figures in parentheses are arc sine transformed values.

Table 18. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) on the adults of *C. carnea*

Rate of application (ml lit^{-1})	Adult longevity	No of eggs laid per 5 female
2.0	13.00 (3.67) [b]	136.50 (11.70) [b]
2.5	12.50 (3.60) [c]	131.50 (11.49) [c]
3.0	11.50 (3.40) [d]	129.50 (10.40) [d]
4.0	10.50 (3.31) [e]	119.50 (10.95) [e]
5.0	6.60 (2.66) [f]	90.60 (9.54) [f]
Untreated control	16.00 (4.06) [a]	414.80 (20.38) [a]

In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)
Values in the parentheses are $\sqrt{x+0.5}$ transformed values.

Table 13. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) to *Chelonus blackburni* (Cameron)

(Mean of four observations)

Rate of application (ml lit^{-1})	Mortality (%)				Corrected mortality (%)		
	6HAT	12 HAT	24 HAT	48 HAT	12 HAT	24 HAT	48 HAT
2.0	5.50 (13.55) [b]	11.25 (19.59) [b]	17.25 (24.53) [b]	35.00 (36.26) [b]	8.97	13.80	28.18
2.5	7.50 (15.89) [c]	16.50 (23.95) [c]	27.25 (31.46) [c]	49.75 (44.86) [c]	14.36	24.22	44.48
3.0	8.50 (16.94) [d]	26.00 (30.65) [d]	26.00 (30.65) [d]	55.75 (48.30) [d]	24.10	32.81	51.10
4.0	10.75 (19.13) [e]	37.50 (37.70) [e]	37.50 (37.70) [e]	68.00 (55.55) [e]	35.90	38.80	64.64
5.0	14.00 (21.97) [f]	41.50 (40.10) [f]	41.50 (40.10) [f]	72.75 (58.53) [f]	40	52.08	69.89
Untreated control	0.00 (0.14) [a]	2.50 (9.05) [a]	2.50 (9.05) [a]	9.50 (17.92) [a]	-	-	-

HAT-Hours after treatment.
In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)
Figures in parentheses are arc sine transformed values.

Table 14. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) on parasitisation of C*helonus blackburni*

(Mean of four observations)

Rate of application (ml lit^{-1})	Adult emergence (%)
2.0	82.00 (64.91)[b]
2.5	76.75 (61.18)[c]
3.0	67.50 (55.29)[d]
4.0	61.00 (51.36)[e]
5.0	57.25 (49.17)[f]
Untreated control	97.50 (80.95)[a]

In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)
Figures in parentheses are arc sine transformed values.

Table 22. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) on Indian bees *Apis cerana indica* F.

(Mean of four observations)

Rate of application (ml lit^{-1})	Mortality (%)		Corrected mortality (%)	
	12HAT	24 HAT	12HAT	24HAT
2.0	26.75 (31.14)[b]	35.00 (36.26)[b]	16.761	20.732
2.5	29.10 (32.58)[c]	39.50 (38.94)[c]	19.432	26.219
3.0	33.00 (35.06)[d]	45.50 (42.42)[d]	23.864	33.536
4.0	40.75 (39.67)[e]	55.00 (47.87)[e]	32.670	45.122
5.0	50.50 (45.29)[f]	63.00 (52.54)[f]	43.75	54.87
Untreated control	12.00 (20.24)[a]	18.00 (25.09)[a]	-	-

HAT-Hours after treatment

In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)

Figures in parentheses are arc sine transformed values.

Table 23. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) on Italian bees *Apis mellifera (L.)*

(Mean of four observations)

Rate of application (ml lit^{-1})	Mortality (%)		Corrected mortality (%)	
	12HAT	24 HAT	12HAT	24HAT
2.0	26.25 (30.81) [b]	36.50 (37.16) [b]	16.66	22.09
2.5	31.50 (34.10) [c]	40.75 (39.67) [c]	22.60	27.30
3.0	34.25 (35.81) [d]	44.50 (41.84) [d]	25.71	31.90
4.0	43.00 (40.97) [e]	55.00 (47.87) [e]	35.59	44.76
5.0	50.50 (45.29) [f]	62.50 (52.24) [f]	44.07	53.99
Untreated control	11.50 (19.80) [a]	18.50 (25.47) [a]	-	-

HAT-Hours after treatment

In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)

Figures in parentheses are arc sine transformed values.

Table 24. Effect of Boom Flower® -n (Nitrobenzene 20% v/v) on dammer bees *Trigona irridipennis* (Dal.)

(Mean of four observations)

Rate of application (ml lit^{-1})	Mortality (%)		Corrected mortality (%)	
	12HAT	24 HAT	12HAT	24HAT
2.0	25.50 (30.32) [b]	29.50 (32.89) [b]	16.76	15.57
2.5	29.50 (32.89) [c]	38.50 (38.35) [c]	21.23	26.35
3.0	34.75 (36.12) [d]	44.50 (41.84) [d]	27.09	33.53
4.0	44.25 (41.70) [e]	55.25 (48.01) [e]	37.71	46.41
5.0	47.75 (43.71) [f]	63.00 (52.54) [f]	41.62	55.69
Untreated control	10.50 (18.88) [a]	16.50 (23.95) [a]	-	-

HAT-Hours after treatment

In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)

Figures in parentheses are arc sine transformed values.

Table 25. Sensitivity of honey bees to Boom Flower® -n (Nitrobenzene 20% v/v) in okra eco system

Rate of application (ml lit^{-1})	No per plot per 30 min.			
	PTC	1 DAT	3 DAT	7 DAT
2.0	28.05	23.00	25.33	26.33
	(5.34) [a]	(4.84) [b]	(5.08) [bc]	(5.18) [b]
2.5	28.30	22.00	25.33	27.00
	(5.36) [a]	(4.74) [bc]	(5.08) [bc]	(5.24) [b]
3.0	29.30	21.00	26.00	26.33
	(5.47) [a]	(4.64) [c]	(5.15) [bc]	(5.18) [b]
4.0	29.30	18.00	24.33	24.67
	(5.47) [a]	(4.30) [d]	(4.98) [c]	(5.01) [bc]
5.0	30.00	16.00	21.00	22.67
	(5.52) [a]	(4.06) [e]	(4.64) [d]	(4.81) [cd]
8.0	29.30	15.33	18.00	22.00
	(5.47) [a]	(3.98) [e]	(4.30) [e]	(4.74) [d]
10.0	27.50	13.00	18.33	21.00
	(5.59) [a]	(3.67) [f]	(4.34) [e]	(4.63) [d]
Untreated control	30.00	29.33	30.00	30.00
	(5.54) [a]	(5.46) [a]	(5.54) [a]	(5.54) [a]

DAT-Days after treatment; PTC-Pre treatment count.
In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)
Values in the parentheses are $\sqrt{x+0.5}$ transformed values.

Table 19. Sensitivity of chrysopids to Boom Flower® -n (Nitrobenzene 20% v/v) in okra eco system

(Mean of four observations)

Rate of application (ml lit^{-1})	Green lacewings per 10 plants				
	PTC	1 DAT	3 DAT	7 DAT	14 DAT
2.0	4.33 (2.20) [a]	3.30 (1.95) [bc]	6.67 (2.68) [b]	7.00 (2.73) [b]	10.00 (3.24) [a]
2.5	4.33 (2.20) [a]	3.67 (2.04) [bc]	6.67 (2.68) [b]	7.00 (2.73) [b]	8.33 (2.97) [b]
3.0	4.33 (2.20) [a]	4.33 (2.20) [b]	6.33 (2.61) [c]	7.00 (2.73) [b]	7.67 (2.85) [bc]
4.0	4.00 (2.11) [a]	3.67 (2.04) [bc]	5.33 (2.41) [bcd]	6.67 (2.68) [bc]	7.33 (2.80) [bc]
5.0	4.33 (2.20) [a]	3.33 (1.95) [bc]	5.00 (2.34) [bcd]	5.67 (2.48) [bc]	6.67 (2.68) [cd]
8.0	4.33 (2.20) [a]	4.00 (2.11) [bc]	4.67 (2.27) [cd]	5.33 (2.41) [bc]	5.67 (2.48) [de]
10.0	4.33 (2.20) [a]	2.67 (1.77) [c]	4.00 (2.11) [d]	5.00 (2.34) [c]	5.33 (2.41) [e]
Untreated control	4.00 (2.11) [a]	6.33 (2.61) [a]	8.67 (3.02) [a]	11.67 (3.48) [a]	10.67 (3.34) [a]

DAT-Days after treatment; PTC-Pre treatment count.
In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)
Values in the parentheses are $\sqrt{x+0.5}$ transformed values.

Table 20. Sensitivity of coccinellids to Boom Flower® -n (Nitrobenzene 20% v/v) in okra eco system

(Mean of four observations)

Rate of application (ml lit^{-1})	Beetles per 10 plants				
	PTC	1 DAT	3 DAT	7 DAT	14 DAT
2.0	7.33 (2.80) [a]	6.67 (2.68) [ab]	7.67 (2.86) [b]	9.33 (3.13) [b]	11.67 (3.49) [ab]
2.5	7.33 (2.80) [a]	5.67 (2.48) [b]	7.00 (2.73) [bc]	8.67 (3.03) [bc]	10.33 (3.29) [bc]
3.0	7.00 (2.73) [a]	5.67 (2.48) [b]	6.33 (2.61) [cd]	8.00 (2.91) [bc]	9.33 (3.13) [bcd]
4.0	7.00 (2.73) [a]	6.00 (2.34) [b]	6.33 (2.61) [cd]	8.00 (2.91) [bc]	8.67 (3.02) [cd]
5.0	7.67 (2.86) [a]	5.33 (2.41) [bc]	6.33 (2.61) [cd]	7.67 (2.85) [c]	8.00 (2.91) [cd]
8.0	7.00 (2.73) [a]	5.00 (2.34) [bc]	5.67 (2.48) [d]	7.67 (2.85) [c]	8.00 (2.91) [cd]
10.0	7.00 (2.73) [a]	4.00 (2.11) [c]	5.67 (2.48) [d]	7.33 (2.80) [c]	7.33 (2.80) [d]
Untreated control	7.00 (2.73) [a]	8.33 (2.99) [a]	9.00 (3.08) [a]	12.33 (3.58) [a]	14.00 (3.81) [a]

DAT-Days after treatment; PTC-Pre treatment count.

In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)

Values in the parentheses are $\sqrt{x+0.5}$ transformed values.

Table 21. Sensitivity of spiders to Boom Flower® -n (Nitrobenzene 20% v/v) in okra eco system

(Mean of four observations)

Rate of application (ml lit^{-1})	Spiders per 10 plants				
	PTC	1 DAT	3 DAT	7 DAT	14 DAT
2.0	8.30	7.00	7.67	8.00	11.33
	(2.95) [a]	(2.73) [ab]	(2.86) [b]	(2.91) [ab]	(3.44) [ab]
2.5	7.30	6.00	7.33	7.67	10.67
	(2.78) [a]	(2.54) [bc]	(2.30) [bc]	(2.85) [b]	(2.85) [bc]
3.0	8.00	6.00	6.67	7.00	10.33
	(2.91) [a]	(2.54) [bc]	(2.68) [bcd]	(2.73) [bc]	(3.29) [bcd]
4.0	8.00	5.33	6.33	6.33	10.33
	(2.91) [a]	(2.40) [bc]	(2.61) [bcd]	(2.61) [bc]	(3.29) [bcd]
5.0	8.00	5.67	6.33	6.67	9.67
	(2.91) [a]	(2.48) [bc]	(2.61) [bcd]	(2.68) [bc]	(3.19) [cd]
8.0	7.30	5.67	6.00	6.67	9.33
	(2.78) [a]	(2.48) [bc]	(2.54) [cd]	(2.68) [bc]	(3.73) [d]
10.0	8.00	5.00	5.67	5.67	6.33
	(2.91) [a]	(2.33) [cd]	(2.48) [d]	(2.48) [c]	(2.61) [e]
Untreated control	8.30	8.33	9.00	10.80	12.33
	(2.95) [a]	(2.93) [a]	(3.08) [a]	(3.24) [a]	(3.88) [a]

DAT-Days after treatment; PTC-Pre treatment count

In a column means followed by common alphabet are not significantly different by DMRT (p=0.05)

Values in the parentheses are $\sqrt{x+0.5}$ transformed value

Table30. LT_{50} values of Boom Flower® -n (Nitrobenzene 20% V/V) to *Cyprinus carpio* (L.)

Concentration (ppm)	LT_{50} (h)	95 per cent fiducial limit		Regression equation	χ^2 at P = 0.05
		UL	LL		
70	47.80	187.76	12.17	Y = 3.56+0.86 X	1.68
80	25.21	65.38	9.72	Y = 3.75+0.89 X	1.30
90	10.55	17.54	6.34	Y = 3.63+1.34 X	3.01

Table 31. LT_{50} values of Boom Flower® -n (Nitrobenzene 20% V/V) to *Tilapia mosambica* (peters)

Concentration (ppm)	LT_{50} (h)	95 per cent fiducial limit		Regression equation	χ^2 at P = 0.05
		UL	LL		
170	35.00	118.78	10.31	Y = 3.73+0.82 X	8.20
180	15.63	34.59	7.07	Y = 3.94+0.89 X	0.24
190	11.88	23.37	6.04	Y = 3.95+0.98 X	0.33

Table 26. LC_{50} values of Boom Flower® -n Nitrobenzene 20% V/V) to earth worms

Organisms	LC_{50} (g/kg)	95 per cent fiducial limit		Regression equation	χ^2 at P = 0.05
		UL	LL		
Eudrilus eugeniae	7.89	8.08	7.71	Y = 22.93+31.13x	1.26
Perionyx excavatus	6.62	6.84	6.41	Y = 14.94+24.28x	0.31

Table 27. LT_{50} values of Boom Flower® -n (Nitrobenzene 20% V/V) to *Eudrilus eugeniae*(Kinberg)

Concentration (g/kg)	LT_{50} (days)	95 per cent fiducial limit		Regression equation	χ^2 at P = 0.05
		UL	LL		
8.0	6.71	9.74	4.62	Y = 2.31+3.25X	1.68
8.2	5.62	7.74	4.09	Y = 2.75+3.00 X	1.30
8.4	4.21	5.17	3.42	Y = 2.11+4.21X	3.01

Table 28. LT_{50} values of Boom Flower® -n (Nitrobenzene 20% V/V) to *Perionyx excavatus* (Perrier).

Concentration (g/kg)	LT_{50} (days)	95 per cent fiducial limit		Regression equation	χ^2 at P = 0.05
		UL	LL		
6.6	6.90	11.59	4.10	Y = 2.96+2.43 X	0.23
6.8	5.81	8.81	3.82	Y = 3.18+2.38 X	0.16
7.0	4.86	6.35	3.73	Y = 2.71+3.33 X	0.26

Table 29. LC_{50} values of Boom Flower® -n (Nitrobenzene 20% V/V) to *Cyprinus carpio* (L.) and *Tilapia mosambica* (Peters).

Organisms	LC_{50} (ppm)	95 per cent fiducial limit		Regression equation	χ^2 at P = 0.05
		UL	LL		
Cyprinus carpio	70.37	78.60	62.88	Y = 6.06+5.98X	1.27
Tilapia mosambica	165.63	178.11	154.03	Y = 19.82+11.19X	0.35

Table 32. Dissipation of Boom Flower® -n (Nitrobenzene 20% v/v) in/on okra (Experiment I)

Location: Thondamuthur

Days	2 ml lit^{-1}		3 ml lit^{-1}		4 ml lit^{-1}		8 ml lit^{-1}	
	Residues in $\mu g\ g^{-1}$	% Dissipation	Residues in $\mu g\ g^{-1}$	% Dissipation	Residues in $\mu g\ g^{-1}$	% Dissipation	Residues in $\mu g\ g^{-1}$	% Dissipation
0	0.167	-	0.334	-	0.723	-	1.506	-
1	0.111	33.53	0.161	51.80	0.167	77.73	0.612	59.36
3	BDL	100	BDL	100	0.139	80.77	0.500	66.80
5	BDL	-	BDL	-	BDL	100	0.112	62.56
7	BDL	-	BDL	-	BDL	-	BDL	100

BDL - Below detectable level

Table 33. Dissipation of Boom Flower® -n (Nitrobenzene 20% v/v) in/on okra (Experiment II)

Location: Chinnamathampalayam

Days	2 ml lit^{-1}		3 ml lit^{-1}		4 ml lit^{-1}		8 ml lit^{-1}	
	Residues in $\mu g\ g^{-1}$	% Dissipation	Residues in $\mu g\ g^{-1}$	% Dissipation	Residues in $\mu g\ g^{-1}$	% Dissipation	Residues in $\mu g\ g^{-1}$	% Dissipation
0	0.111	-	0.222	-	0.634	-	0.890	-
1	BDL	100	0.147	33.78	0.250	60.56	0.806	9.44
3	BDL	-	BDL	100	0.195	69.24	0.506	43.14
5	BDL	-	BDL	-	BDL	100	0.085	90.45
7	BDL	-	BDL	-	BDL	-	BDL	100

BDL - Below detectable level

Table 34. Correlation co-efficients for Boom Flower® -n (nitrobenzene 20% v/v) in/on okra by different methods of linearization of residue data (Experiment I and II)

Function	Experiment I (Location: Thondamuthur)						Experiment II (Location: Chinnamathampalayam)					
	3 ml lit^{-1}		4 ml lit^{-1}		8 ml lit^{-1}		3 ml lit^{-1}		4 ml lit^{-1}		8 ml lit^{-1}	
	r	modified r^2	r	modified r^2	r	modified r^2	r	modified r^2	r	modified r^2	r	modified r^2
First order	-0.998	0.988	-0.819	0.661	-0.955	0.897	-0.988	0.911	-0.872	0.754	-0.931	0.673
1.5^{th} order	0.984	0.306	0.843	0.591	0.930	0.796	0.974	0.243	0.897	0.756	0.894	-1.694
2^{nd} order	0.968	-41.660	0.869	0.486	0.893	-82.246	0.962	-236.64	0.922	0.743	0.865	-19.617
RF 1^{st} order	-0.944	0.780	-0.945	0.933	0.925	0.915	-0.907	0.615	-0.973	0.964	-0.813	0.205
RF 1.5^{th} order	0.897	-1.845	0.959	0.924	0.851	-0.727	0.874	-0.0947	0.984	0.975	0.764	-5.109
RF 2^{nd} order	0.867	-13.082	0.972	0.898	0.782	-11.381	0.850	-190.010	0.993	0.984	0.728	-11.607
Inverse power law function	-	-	-	-	0.813	0.394	-	-	-	-	0.861	0.775

RF - Root function

Table 35. Dissipation pattern for Boom Flower® -n (Nitrobenzene 20% v/v) in/on okra with statistical parameters.

Function		Experiment I (Location: Thondamuthur)			Experiment II (Location: Chinnamathampalayam)		
		3 ml lit^{-1}	4 ml lit^{-1}	8 ml lit^{-1}	3 ml lit^{-1}	4 ml lit^{-1}	8 ml lit^{-1}
		1^{st} order	RF 1^{st} order	RF 1^{st} order	1^{st} order	RF 2^{nd} order	Inverse power law function
Intercept	(a)	3.566	0.129	5.140	3.235	0.617	-0.217
	UL	4.723	-0.436	6.732	-5.953	0.0453	10.289
	LL	2.410	-0.178	3.548	0.517	-0.012	-10.723
Slope	(b)	-0.874	-0.089	-1.027	-0.751	0.021	1.243
	UL	-0.240	0.427	0.298	0.738	0.052	10.581
	LL	-1.410	-0.248	0.255	-2.239	-0.011	-10.723
Half life	$T_{(0.5)}$	0.193	1.442	0.456	0.924	2.198	1.747
	UL	1.369	14.324	1.593	2.756	13.127	1.069
	LL	0.218	-11.440	-0.682	-0.909	-8.831	-5.575

RF - Root function

Plate 1. Field experiment I - Thoundamuthur

Plate 2. Field experiment II - Chinnamathampallayam

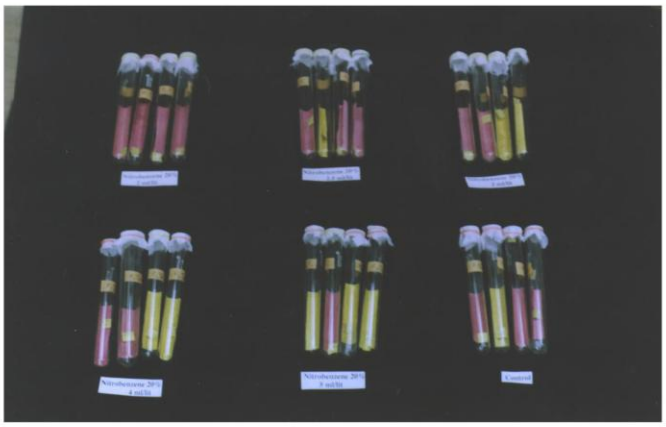

Plate 3. Safety evaluation setup - *T. chilonis*

Plate 4. Safety evaluation setup - Eggs of *C. carnea* (closed condition)

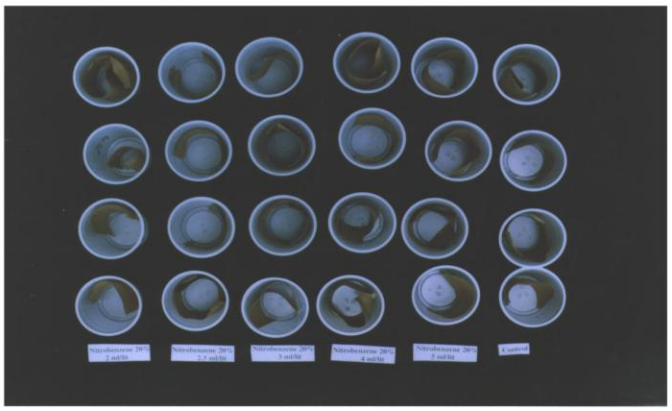

Plate 5. Safety evaluation setup - Eggs of *C. carnea* (opened condition)

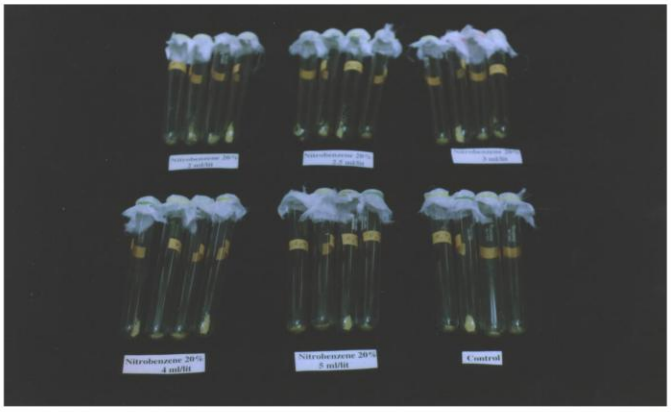

Plate 6. Safety evaluation setup - Grubs of *C.carnea* (Larval feeding method)

Plate 7. Safety evaluation setup - Grubs of *C . carnea* (Thin film method)

Plate 8. Safety evaluation setup -Honey bees

Plate 9. Acute toxicity experimental setup - *E. eugeniae*

Plate 10. Acute toxicity experimental setup - *P. excavatus*

Plate 11. Experimental setup for evaluation of acute toxicity to *C. carpio* and *T. mosambica*

Plate 12. Phytotonic effect of BOOM FLOWER - n on okra fruits