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MANAGEMENT OF THE MEDITERRANEAN FRUIT FLY (*CERATITIS CAPITATA* WIED.) USING PHEROMONE TRAPS AND NEEM SEED EXTRACT

By

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DECLARATION

I hereby declare that, except for reference to other peoples' work which have been duly cited, this work is the result of my original research and that this thesis has neither in whole nor in part been presented for a degree elsewhere.

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ABSTRACT

Laboratory and field experiments were conducted to evaluate the effectiveness of neem seed water extracts (NSWE) and pheromone traps to control the Mediterranean fruit fly, *C. capitata* infesting citrus at the University of Ghana Agricultural Research Station (ARS), Kade. Neem seed extract of concentration 15%, 20%, 25% and 30% wt/vol were prepared and left overnight after which the suspensions were seived and used for spraying. Ripened fruits of *Citrus sinensis* cultivar Late Valencia and *Citrus unshiu* cultivar Satsuma were harvested into insect cages containing adult *C. capitata* after which they were sprayed with the neem seed water extract (NSWE) suspensions.

Second and third instar larvae and pupae were removed from untreated rotten fruits into petri dishes. These were exposed to the various suspensions as indicated above. Field experiments were conducted concurrently with the laboratory experiment to determine the seasonal abundance and activity pattern of *C. capitata* using pheromone traps baited with med-call, a Japanese formulated pheromone. To monitor the seasonal abundance of *C. capitata* two rectangular traps each baited with trimedlure (TML) were installed in Satsuma and Late Valencia citrus orchards.

Data were collected every other day between 8:00 – 10:00 am. from September, 1997 to July, 1998. A similar method was used to investigate the diurnal activity and behaviour of *C. capitata* from 6:00 am. – 6:00 pm.

To test the effectiveness of the NSWE under field conditions, field experiment were conducted in two selected citrus orchards i.e. Late Valencia and Satsuma. The two fields were each laid under the Randomised Complete Block Design (RCBD). NSWE of 25kg/ha suspension was sprayed on the trees. Two controls of picking of dropped infested fruits and no picking of dropped

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infested fruits were included. Dimethoate 40EC was also used as the basis for comparing the performance of the NSWE suspension.

Results from laboratory work showed that oviposition of the adult female was significantly reduced when 20%, 25%, and 30% wt/vol NSWE were sprayed on each citrus variety. The anti-ovipositional effect of NSWE was dosage-dependent. The number of oviposition punctures on Late Valencia were significantly fewer than on Satsuma .

The development period from eggs to adult in fruits sprayed with the NSWE suspensions was found to be between 38 – 40 days which was not significantly different among the citrus species examined.

25% and 30% wt/vol. of NSWE were more effective against the larvae removed from fruits. About 79% of 2nd instar larvae died when exposed to 25% and 30% NSWE suspensions. The percent mortalities were 73% and 84% for 25% and 30%wt/vol NSWE respectively when 3rd instar larvae were exposed. Furthermore, the development of pupae into adult was delayed by 6 – 9 days. There was significant difference among the treatments when pupae were sprayed together with soil. However there was no significant difference on the mortality of the pupae when only the soil was sprayed with the NSWE before introducing the untreated pupae.

The results from the seasonal abundance and diurnal activity pattern using the pheromone traps indicated that, the population of *C. capitata* was high during the period of fruit maturation (colour break) and continued till harvesting. The peak periods in Satsuma occurred in September 1997(38.5) and March 1998 (68.5) whereas that of Late Valencia occurred in December 1997 (36.5) and March 1998 (29.5) which are the harvesting periods for Satsuma and Late Valencia respectively. The diurnal activity of *C. capitata* was higher during 8:00-10:00 am. than 3:00-5:00 pm . The mean

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number of 9.1 and 10.0 per trap were recorded between 8:00-10:00 am. for Late Valencia and Satsuma orchards respectively. The value for 3:00-5:00 pm. were 4.4 and 5.7 for Late Valencia and Satsuma respectively.

The management practices adopted in the field to determine the effectiveness of NSWE under field conditions showed that *C. capitata* caused as high as 46% and 32% damage to Satsuma and Late Valencia citrus fruits respectively if not controlled. Damage was high in Trimedlure + picking of dropped infested fruits (TML+P) (46%) treated plots. The neem seed water extract and picking of dropped infested fruits (NSWE+P) treated plants performed significantly better than Control + no picking of infested dropped fruits (CNP), control + picking of infested dropped fruits (CP) and TML+P. Percentage damage recorded in the NSWE+P treated plots was however, significantly higher than the Dimethoate 40 EC and picking of dropped infested fruits (Dim+P) treated plots.

Dim+P reduced fruits damage by more than half compared with the trimedlure and the control plots Dim+P and NSWE+P reduced the population of *C. capitata* by 27% and 10% the Satsuma orchards respectively. Similarly the *C. capitata* population was reduced by 24% and 14% by Dim+P in the Late Valencia orchard respectively. The NSWE sprayed at 25 kg/ha was comparatively better than the control treated plants.

The cost-benefit analysis showed that the cost of spraying the NSWE could be beneficial particularly in the Satsuma orchard. This was however, not the case in the Late Valencia orchard.

The relationship between adult *C. capitata* captured in TML baited traps, the number of damaged fruits, the number of larvae and pupae from damage fruits and the number of oviposition holes on fruits showed significant correlation.

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DEDICATION

To my dear parents

Mr. & Mrs. Akotsen and family

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LIST OF ABBREVIATIONS

a.i.	Active ingredient
ANOVA	Analysis of variance
ARPPIS	African Regional Postgraduate Programme Insect Science
ARS	Agricultural Research Station
CNP	Control (no picking of dropped fruits)
CP	Control (picking of dropped fruits)
CV	Coefficient of variation
DAS	Days after spraying
DBS	Days before spraying
Dim	Dimethoate 40 EC
Dim+P	Dimethoate 40 EC and picking of dropped infested fruits
et.al.	And others
F. tab.	Fiducial value read from table
F. Val.	Fiducial value
GMT	Greenwich meantime
IITA	International Institute of Tropical Agriculture
IPM	Integrated Pest Management
JICA	Japan International Co-operation Agency
Kg/ha	Kilogram per hectare
MS	mean square

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NSWE	neem seed water extract
NSWE+P	neem seed water extract and picking of dropped infested fruit
OPRI	Oil Palm Research Institute
Ppm	parts per million
PROC GLM	Procedure of general linear model
P.val	Probability value
RCBD	Randomised Complete Block Design
s. e.m	Standard error of the mean
SPSS	Statistical Programme for social science
SS	sum of squares
TML	Trimedlure and picking of infested dropped fruits
WAPP	West African Plantain Project

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CHAPTER ONE

1.0 GENERAL INTRODUCTION

The Mediterranean fruit fly, *Ceratitis capitata*, Wiedemann, (Diptera: Tephritidae) is a major agricultural pest in many citrus growing countries world-wide (Health *et al.*, 1990), and has been described as a quarantine pest because they attack a wide range of commercial horticultural crops and inhabit areas in a broad spectrum of climates (Cowley *et al.*, 1992). Currently, the fly causes great losses to citrus plantations throughout Ghana especially around the catchment area of the Agricultural Research Station, Kade (Afreh-Nuamah, 1985). Enkerlin and Mumford (1997) have estimated that if control measures are not applied the loss due to *C. capitata* in the Mediterranean basin is $365 million. The economic importance of this pest will increase as more land is cropped to citrus. Thus, there is the need for a more effective and efficient control strategy against this important pest, which attacks all citrus species.

In Ghana, the main method of control of this pest is the use of insecticides. However, there is now an increasing awareness of the failure of most insecticides to sustain agricultural production, due to the following reasons; (1) the development of resistance by insects to these insecticides, (2) pollution of the environment due to their rather extremely slow rate of degradation (Kiss and Meerman, 1991), (3) their high mammalian toxicity. (4) toxic residues in food, soil and water bodies.

Consequently, there is the need to select environmentally friendly insecticides which would at the same time maintain their biological activity for longer periods even after exposure to ultra violet radiation (Stark *et al.*, 1990). Several commercially produced plant derived compounds have exhibited enormous potential as insecticides. Azadirachtin, a limonoid or tetranortriterpenoid from the neem tree *Azadirachta indica* A.Juss is now well established to have insect repellant, growth disruption and antifeeedant properties (Jotwani and Srivastava, 1981; Reed *et al.*, 1982; Green *et al.*, 1987). Azadirachtin (C_{37} H_{48} O_{13}) has been shown to have

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effect on metamorphosis, reproduction and longevity of *C. capitata*, *Bactrocera dorsalis* (Hendel), and *Bactrocera cucurbitae* (Coquillet) at late 3rd instar larvae and pupae to treated sand (Stark *et al.*, 1990). Water extracts from leaves and seeds of the neem tree are used in many developing countries as protectant against plant pests (Heyde *et al.*, 1984).

Pheromone traps are used to monitor the population of fruit flies to predict pest development and to forecast pest abundance and damage. Newly emerged adult populations of fruit flies can also be detected by monitoring traps set in areas susceptible to fruit fly attack (Somerfield, 1989).

Many workers have demonstrated that localised fruit fly infestation in orchards or grooves can be controlled by proper field sanitation methods (Ceiba-Geigy, 1975; Hagen *et al.*, 1981; Afreh-Nuamah, 1985). This involves the collection and destruction of infested dropped fruits by burying or spraying an appropriate insecticide on them. Black polythene bags could also be spread on the collected damaged fruits to prevent adults from escaping after emergence.

Work done to determine the efficacy of neem against most pests of tree crops appear little and scanty (Adu-Acheampong, 1997) particularly for citrus. However, with current emphasis on use of more ecologically sustainable management strategies in Pest Management strategies, especially for citrus production in Ghana, my interest has been stimulated to evaluate the biological activity of neem and to develop a more sustainable management practices against the Mediterranean fruit fly, *C. capitata* infesting citrus in Ghana.

This research was undertaken with the following objectives:

1. To evaluate different control strategies against *C. capitata*,
2. To reduce the damaging effect of the fly on citrus plantations in Ghana.
3. To determine the most effective and efficient means of controlling the fly.

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CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 The citrus crop

Citrus belongs to the family Rutaceae. The crop is an aromatic, broad-leaved, evergreen tree native to tropical and subtropical regions. The trees vary in size from 3-5 m tall for lime and up to 10 m for grapefruit cultivars. The fruit is a berry with a leathery pericarp, which has numerous oil sacs in its tissue (Rice *et al.*, 1993). The original home of the genus *Citrus* is not known (Leslie, 1957; Ceiba-Geigy, 1975) although the history of its cultivation shows that it must have originated in the South Eastern Asian region.

Very few plantations have been cultivated and maintained for thousands of years particularly in backyards and residential areas but it was only during the last century that the citrus industry has developed with the cultivation of large plantations (Leslie, 1957) as seen presently in major producing countries such as United States of America (USA), Brazil, Israel, Japan etc.

Citrus does well in warm climates where there are suitable soils which are slightly acidic i.e. pH of 6 (Karikari, 1971) and have sufficient moisture to sustain the trees (Ceiba-Geigy, 1975). They grow better in cooler, frost-free (Mediterranean) climate if soils are suitable (Ceiba-Geigy, 1975). The primary species of cultivated citrus are the sweet orange *Citrus sinensis* L. (Osbeck), the lemon *Citrus limoni* (Burmann), the grapefruit *Citrus paradisi* (Macf.), the lime *Citrus aurantifolia* (Swingle) and the mandarin, also known as tangerine *Citrus reticulata* (Blanco). Various hybrids such as the tangor (tangerine x sweet orange) and the tangelo (tangerine x grapefruit) are also cultivated.

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2.2 The economic importance of citrus

Citrus are now the major fruit of the subtropical regions (Rice *et al.*, 1993). The main centres of production in the world are southern Africa, Israel, the U.S.A., Brazil, Spain, Japan, Italy and Mexico. Of these the U.S.A. is the largest producer (Rice *et al.*, 1993).

Production levels for 1994/95 in the major producing countries of the world was estimated at 62.45 million tonnes. This shows an increase of 4% over 1993/94 production but down by 1% from the 1992/93 records. The increase was as a result of significantly larger crops in Brazil and USA, in addition to moderate gains in China and Spain (Kirby-Strzelecki, 1995).

In Africa, most of the citrus is grown in Southern Africa (i.e. in Zimbabwe, South Africa, Mozambique and Swaziland) and the Mediterranean regions of Egypt, Morocco and Tunisia. In most of the remaining regions of Africa production is on a small scale and is primarily for local consumption (Rice *et al.*, 1993).

In Ghana, citrus has been grown since 1913 (Adansi, 1972). However, production levels are still low (Anno-Nyarko, 1998. Until recently the crop was left to grow wherever it germinates (Gyamera-Antwi, 1966).

Currently, however, citrus has become a major cash crop in Ghana (Anno-Nyarko, 1998). The crop is cultivated in the semi-deciduous forest zone which covers the parts of Ashanti, Brong-Ahafo, Eastern, Western, Central and Volta regions (Anno-Nyarko, 1998). The importance of citrus to the country's economy can not be overemphasised. As a non-traditional crop, sweet oranges can be exported to other countries if produced in large quantities to obtain foreign exchange (Gyamera-Antwi, 1966). Both fresh fruits and juice can be exported. No information is currently available on the level of citrus production and the volume of fruits exported in Ghana.

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Over 1 billion cedis was generated from all citrus related activities within the Kwaebibirem district alone where the University of Ghana Agricultural Research Station is located (Osei, personal communication). Marketing potential exists for Ghana to export substantial amounts of fresh citrus fruits and juice to some neighbouring countries like Togo, Burkina Fasso and Cote d'Ivoire as was done in 1997 and 1998 citrus seasons.

Apart from the export of the fruit and juice, the fruits, flowers, leaves and stem of all species of citrus contain several essential oils, differing in composition from species to species and often differing in composition in different parts of the plant (Leslie, 1957). Although certain proportion of the fruit oil is found mixed with the juice, the rind oils are the most important. The various oils obtained from citrus are used for various purposes, especially in the production of fruit cordials, flavourings for soft beverages and liquors in confectionery and in the preparation of perfumes, toilet waters, cosmetics (Leslie, 1957) and insecticides (Taylor and Vickery, 1974). Dropped fruits could also serve as a good source of feed for livestock especially pigs and ruminants like sheep and goats (personal observation).

2.3 Production constraints

Although the total land area under citrus cultivation in Ghana has increased significantly since the past few years especially in the Eastern Region of Ghana, the production of citrus has not reached its potential because of a number of production constraints. Currently, local demands exceed that of supply and within the year there is shortage of sweet oranges. This brings about differential prices within the year. Ghana has comparative advantage in the production of citrus than most African countries because of its location within the tropics. Considerable foreign exchange can therefore be earned from citrus production.

Some of the major constraints associated with citrus production are (a) farmers' reluctance to use improved scientific methods in production (Gyamera-Antwi, 1966; Osei, 1986), (b) marketing and lack of storage facilities resulting in high percentage of post-harvest

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losses and (c) disease and pest damage. Some of the most important diseases which attack citrus are die back (Tristeza), gummosis, psorosis or scaly bark and root rots. These diseases have been reported to have caused significant reduction in yield about 10,000 tons in 1940 (Gyamera-Antwi, 1966). Their incidence have, however, subsided considerably (about 4,000 tons in 1947) with the use of sweet orange varieties on rough lemon as rootstock (Gyamera-Antwi, 1966; Osei, 1989). Currently insect pest damage is one of the major problems facing the citrus industry in Ghana particularly the Kwaebibirem district.

The humid type of climate, the perennial nature of the tree, as well as the vegetation associated with the citrus crop favour the occurrence of large numbers of species of arthropods that form a settled and balanced ecosystem. There are three categories of arthropods, including (1) those that live primarily or solely on the citrus trees, (2) others which live on the associated vegetation that grows in the shade of the trees and (3) a number of predators and parasites that derive their food mainly by attacking the two aforementioned categories of plant feeders. The first and third are of most interest to the citrus grower (Ceiba-Geigy, 1975; Afreh-Nuamah 1985). In an extensive survey conducted in the country, between 1980-1983 a total of 140 species of insects were found to be permanently associated with citrus plantations in Ghana (Afreh-Nuamah, 1985). Three of them i.e. the fruit piercing moth, *Achae spp* (Lepidoptera; Noctuidae), the fruit flies, *C. capitata* Wied. (Diptera: Tephritidae) and black plant bug *Leptoglossus membranaceous* F. (Heteroptera: Coreidae) were found to be serious pests of the fruit. In addition the red ants, *Oecopyhlla longinoda* Lart. (Hymenoptera: Formicidae) and black ant *Tetramorium aculeatum* Mayr. (Hymenoptera: Formicidae) were observed to cause nuisance during harvesting. None of the scale insect pests, which are observed to be most injurious insect pests on citrus elsewhere, were found to be of any importance in Ghana.

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2.4 The Mediterranean fruit fly (*Ceratitis capitata, Wiedemann*)

2.4.1 Economic importance of fruit flies

Fruit flies including the notorious Mediterranean fruit fly *C. capitata* are among the most serious horticultural pests in the world (National Research Council, 1992). They cause millions of dollars of damage to fruits, and their very presence in the tropics is keeping dozens of delicious fruits from becoming major items of international trade (National Research Council, 1992). Some species have become pests in regions far removed from their native range (White and Elson-Harris, 1992). In the USA for example, no exotic insect raises as much concern among regulatory officials as the detection of the Mediterranean fruit fly (Dowell *et al.*, 1999). This concern is based on the economic and environmental damage the medfly could cause if it becomes established permanently (Dowell *et. al.*, 1999).

Quarantine restrictions have to be imposed to limit further spread of fruit fly pests. However, quarantine regulations imposed by an importing country can either deny producing countries a potential export market, or force the producer to carry out expensive disinfestation treatment (Hagen *et al.*, 1981; vanRanden and Roitberg, 1998). For example, New Zealand suspended shipment of peaches and nectarines, *Prunus persica* (L) Batsch *var. nucipersica*, from California in 1989 because of potential risk that fruits might have been infested with walnut husk fly, *Rhagoletis completa* Cresson (Diptera: Tephritidae) (Somerfield, 1989).

Monetary estimates of fruit production and fruit fly damage are not available for most countries (White and Elson-Harris, 1992). However, in Australia, annual fruit production is estimated over US $850m and potential losses if fruit flies were not controlled are believed to exceed $100 million (Anon., 1986). In California, crop losses were estimated at US $910 million and cost growers an extra 1.4 million pounds of pesticides active ingredient (a.i) at a cost of US $290 million to prevent the Medfly from infesting fruits and vegetables (Dowell

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et al., 1999). The cost of eradicating fruit fly from even a small island is very high. For example, it cost Japan US $32 million and 200,000 man days to eradicate fruit flies from its south-western Islands using sterile insect release method (SIT) (Anon, 1986).

Apart from the loss to crops, Jiron and Zeledon, (1979) have studied and provided information on the larvae of *Anastrepha spp* causing abdominal pain and diarrhoea, particularly in children.

Currently, *C. capitata* is the most important pest causing serious losses to citrus in Ghana (Afreh-Nuamah, 1985) especially at the University of Ghana, Agricultural Research Station (ARS), Kade, which has the largest collection of citrus varieties (which mature in different seasons) in the country.

2.4.2 Behaviour, biology and damage

The adult Mediterranean fruit fly is about the size of a housefly measuring 5 – 6 mm. It has yellowish orange marks on its drooping wings and black spots on its yellowish abdomen. Although it can fly over one mile, it generally remains in trees and bushes near where it has emerged from its puparium in the soil (Hagen *et al.*, 1981). The male moves along with female on leaves or fruits in the morning and mate when the temperature exceeds 28°C (Hagen *et al.*, 1981). The females need food to survive and produce eggs. They are ready to lay eggs after about 1 week at high temperatures (Hagen *et al.*, 1981).

Many workers have studied the behaviour and biology of several fruit flies including *C. capitata* (Hanna, 1947; Bateman, 1972; Prokopy and Roitberg, 1989; Stark *et al.*, 1990). *C. capitata* undergoes complete metamorphosis. After successful mating, the female looks out for fruits that are beginning to ripen. It drills its long ovipositor through the skin, making a cavity just below the skin and then deposits between two to six eggs in the cavity. It can lay up to 40 eggs per day and has the capacity to produce over 1000 eggs in its lifetime of 60

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90 days under optimum conditions (Hagen *et al.*, 1981). Some fruit flies normally attack undamaged fruits. However, *C. capitata* can selectively attack oranges (citrus species) which are already damaged and will oviposit into the wound (Papaj *et al.*, 1989a). The whitish eggs in citrus hatch in 2 - 3 days at 25 - 27°C (Hagen *et al.*, 1981).

There are three larval stages although the first instar is completed before emerging from the egg (White and Clement, 1987). Mature larvae are usually creamy white although some may appear dark due to the gut contents showing through the cuticle (White and Clement, 1987). The second and third instar larvae can be distinguished by the mouth hooks and 'flying' ability. Whereas the 3rd instar larva has a small tapered head with two distinct black dots (mouth hooks) and also show 'flying' ability, the 2nd intar larva does not have these characteristics. The rate of larval development is strongly influenced by the host fruit. It is slowest in apple but progressively faster in citrus, peach, figs and pear (Hagen *et al.*, 1981; Nimrod *et al.*, 1997). Mature larvae leave the fruit while it is still hanging on the tree or has fallen to pupate.

The pupa is immobile, long, brownish and seed-like with blunt rounded ends. It remains in the soil, untill the adult fly breaks open one end of the pupal case and pushes its way through the soil. Pupal development depends largely on the soil moisture, structure and temperature (Hagen *et al.*, 1981). The pupal stage can be as short as 6 days at 38°C but 9 - 15 days at 26°C (Hagen *et al.*, 1981). No development occurs below 18°C and may require 60 days before flies emerge during cold condition.

2.4.3 Natural enemies of *C. capitata*

No comprehensive parasitoid-host catalogue has been compiled for *C. capitata* (White and Clement, 1987; Stark *et al.*, 1990; 1991). However, there is evidence of parasites, parasitoids and predators of *C. capitata*. The larvae and puparia are attacked by a variety of parasitic Hymenoptera, particularly by species of Opiinae, Braconidae (Chritenson and

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Foote, 1960; Wharton and Gilstrap, 1983; Wong *et al.*, 1984b) but Chalcidoidea and other groups are also important. Dehouzie (1989) has reviewed the role of parasitoids in the natural regulation of *C. capitata* population.

Drew (1987) showed that birds and rodents sufficiently consume fruits to account for a far higher level of larval mortality than invertebrate, predators and parasitoids and gave the example that rodents consume larvae in 78% of dropped *Planchonella australis* Pierre (Sapotaceae) fruits. Puparia in the soil are very vulnerable to predators as well as parasitoids White and Elson-Harris, 1992). Ants are of particular importance and 38% mortality has been attributed to them (Wong *et al.*, 1984a), although ants found in some regions are unable to detect or crack puparia (Boller and Prokopy, 1976). Some ground dwelling Coleoptera (Carabiidae) and Hemiptera (Pentatomidae) as predators and spiders (Salticidae) have also been reported to feed on pupae and larvae (White and Elson-Harrison, 1992).

2.4.4 Other Diptera often associated with dropped citrus fruits

Some other families of Diptera are sometimes found in association with fruits damaged by *C. capitata* and other fruit flies. *Drosophila* species (Drosophilidae) live in association with fruit flies and are usually primary micro fungi feeders. There is no evidence of members of this family attacking undamaged fruit, and when they are found to be in association with fruits they are probably attracted by fermentation product (White and Clement, 1987). Louis *et al.*, (1989) suggested that they help in spreading fungal infection among packed fruits.

Muscidae (Subgenus Atherigona) are also important. They are phytophagous and develop in the stems of *Poacea* (Graminae), and are often found in abundance in fruit crops, but when found in fruits, they are usually thought to be only secondary invaders (White and Elson-Harris, 1992). Recent reports, however, have shown that these flies cause primary damage to fruits in Australia, Hong Kong, India and Nigeria (Chughtai *et al.*, 1985; Ogbalu, 1989).

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2.4.5 Alternative host plants

There are 253 fruits, nut and vegetables recorded as hosts of Mediterranean fruit fly many of which are tropical in origin (Hagen *et al.*, 1981). Among these 253 hosts, 40 are considered heavily or generally infested. Liquido *et al.*, (1989), have, however, shown that in Hawaii among the 196 species of fruits collected 60 were host of *C. capitata* under natural field condition.

Those that are important to the Ghanaian agriculture are mangoes, apples, avocados, peppers and papaya. The rest are cotton, egg plants and banana (which are rarely damaged). In Ghana, *C. capitata* has been reported to contribute significantly to fruit drop in pepper (Opoku-Asiamah *et al.*, 1987).

2.4.6 Control of C. *capitata*

The successful management of fruit flies in general and *C. capitata* in particular has been a major problem in countries throughout the world (Aluja and Liedo, 1993; White and Elson-Harris, 1992). The most widely used control strategy has relied heavily on the use of insecticides. Protein hydrolysate bait sprays with malathion or other insecticides are considered the most effective single method for suppressing fruit fly populations (Hagen, *et al.*, 1981). The protein simulates honeydew, attracting the fly from a distance, and the malathion kills the flies when they are in contact with or ingest spray droplets. The use of fenthion to spray soil beneath infested trees has been practised in the United States of America (Hagen, *et al.*, 1981).

Fruit removal and localised pesticide treatment of infested crops are practised in developed countries for the eradication of adventitious populations, however, the choice of chemicals is limited (State of California IPM Manual Group, 1984).

Sterilisation of the male Mediterranean fruit fly in the pupal stage with gamma radiation, which stops the production of sperms, has been used extensively in the USA and Japan. The

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sterilised males when released mate with females, which invariably prevent egg development. In most cases the eggs do not hatch even after they have been laid successfully. The principal success behind this technique is that the female Medfly often mates once and hence no eggs are produced after fertile females mate with sterilised males (Hagen *et al.*, 1981). Recent work in Hawaii suggests that the releases of predominantly sterile male medflies are more effective in producing sterile eggs in population of wild medflies than the releases of both sterile male and female (Dowell *et al.*, 1999).

Biological control using natural enemies has been reported. For example, it is reported that in Hawaii a parasitic wasp accounted for over 40% parasitisation and reduced the Medfly problem, however insecticides still had to be used in controlling increasing fruit fly populations on highly susceptible crops (Hagen *et al.*, 1981). In Greece and former Czechoslovakia a parasitic wasp, *Coptera occidentalis* L. was reported to attack Mediterranean fruit fly. Other parasitoids known to attack *C. capitata* and *Bactrocera dorsalis* (Hendel) are *Psytallia incisi* (Silvestri) (Hymenoptera: Braconidae), *Diachasmimorpha longicaudata* (Ashaead) and *D tryoni* (Camson) (Hymenoptera: Braconidae) (Stark *et al.*, 1991). Very little has, however, been done to use these natural enemies for the control of the Mediterranean fruit.

2.5 Development of Integrated Pest Management (IPM) in citrus production

Current efforts at pest control the world over are directed at developing an integrated pest management system where insecticides are applied only when absolutely necessary (Tanzubil, 1992a). Pest control is thus effected through the integration of several measures such as the use of biological agents, host plant resistance, and appropriate cultural practices (Tanzubil, 1992a). Chemical insecticides have been the backbone of insect pest control since the early 1955 when the organochlorine insecticides were first widely introduced (Dent 1992). However, pest

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problems seem not to have reduced. The reasons are due to the fact that most farmers lack the technical know-how on chemical control and also the components of the technology (sprayers, insecticides etc) are usually beyond the reach of many farmers (Tanzubil, 1992a) especially in the developing countries.

Some problems have become apparent with total reliance on broad-spectrum insecticides. These include:

1 Elimination of beneficial insects whose contribution in pest management is often reduced by indiscriminate use of insecticides (Debach and Rosen, 1991; Prokopy and Powers, 1995). For example, the use of malathion spray on citrus for the control of scale insects was followed by the elimination of some bees and other natural enemies (Hagen *et al.*, 1981)

2 Potential ground surface water contamination which usually leads to accidental human and livestock poisonings and to the decline of local plant and animal population (Pimental *et al.*, 1980, 1992; Ascher, 1993)

3 Resistance to pesticides: the widespread and indiscriminate use of insecticides may lead to accelerated development of resistance in insect populations (Pimental *et. al.*, 1992). This results in increased dosages being used at greater expense and with severe effect on beneficial natural enemies. For example, seven day old adult *Bactrocera tau* (Walker) and *B. cucurbitae* (Coquillet) have been reported to be most resistant to fenvalerate, malathion, and trichlorfon (Areekul, 1986).

Current pest control programmes in fruit production must therefore be geared towards environmentally safe and sustainable strategies for use at farmer level such as the use of baited

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traps and picking of dropped infested fruits under the trees, in citrus and other crops production systems.

Many plant species possess compounds with insecticidal activity, some of which are readily extracted, synthesised and formulated for field control of insect pests. For example, *Bersama* species contain compounds that have antifeedant activity against most phytophagus insect pests (Ahmed *et al.*, 1984; Grainge and Ahmed, 1988). *Gomphrena globosa* Linn. and some species of *Amaranthus* contain compounds with antifeedant activity against a range of insects including, *Leptinotarsa decemlineata* (Say) (Jermy, 1966). *Epilobium hirsutum* (Linn) also contains compounds that deter *Locusta migratoria* Linn. from feeding (Ahmed and Grainge, 1988). The seeds, fruit and leaves of the neem tree, *Azadirachta indica* A. Juss (Meliaceae) contain terpenoids with potent anti-insect activity (Rembold, 1989; Ndiaya, 1992; Schmutterer, 1995) which include repellent and antifeedant activity, growth inhibition, suppression of reproduction, mating disruption and ovicidal activity. The active ingredients contained in neem include azadirachtin and salanin (Reed *et al.*, 1982), nimbin, deacetylnimbin and thionemone (Jotwani and Srivastava, 1981; Simmonds *et al.*, 1995). Azadirachtin, salannin and nimbin all have the same basic Limonoid structure (National Research Council, 1992) and hence function as antifeedants or oviposition deterrents (Jacobson, 1989; Schmutterer, 1990; Ascher, 1993). Of the numerous pesticidal agents isolated so far from kernels, azadirachtin is the most active against insects (Barnby *et al.*, 1989; Rembold, 1989).

Warthern *et al.*, (1978) reported that azadirachtin isolated from ethanolic extract of neem seeds inhibited the feeding of the fall armyworm, *Spodoptera frugiperda* (Smith). By 1998, researchers worldwide had shown that neem extracts could influence over 400 insect species. These include many that are resistant to, or inherently difficult to control with conventional pesticides such as some cowpea pod borers, sweet potato whitefly, green peach aphid, diamondback moth, several leafminers and fruit flies (Pradhan *et al.*, 1962; Jackai and Oyediran,

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1991; Lowery *et al.*, 1993; Schmuterrer 1998). In general, neem products are medium - to broad-spectrum pesticides against plant eating (phytophagous) insects. They affect members of most orders of insects.

Neem has been shown to be effective against the Citrus red mite *Panonychus citri* (McG.) in the USA (Jacobson *et al.*, 1978) and in China (Chiu, 1984). In Sudan, Siddig (1980) showed that damage stored wheat was substantially reduced for 490 days after neem seed powder treatment. In Gambia, Redknap (1980) reported that various leaf suspensions were found to control leaf eating flea beetles *Podagrica uniforma* (Jac.) and *Podagrica sjostedti* on Rosselle *Hibiscus sabdariffa* linn., Author and the leaf eating larva *Epilachna chrysomelina* on cucumber (*Cucumis sativas* L.). In addition, citrus seedlings sprayed with neem suspensions were protected from the larvae of *Papillio demodocus* (Esper) (Redknap, 1980). In Tanzania neem extracts were compared against the synthetic insecticide Lindane in the control of pests on beans. The results showed that seed extract was as effective as lindane against thrips (50% control).

In Burkina Fasso, 25 kg/ha neem seeds per 500 litres of water was found to be as effective as the synthetic insecticide, carbofuran 5G against the Sorghum Shootfly, *Atherigona soccata* Rondani (Zongo *et al.*, 1983), *Helicoverpa armigera* Hubner and the pod sucking bugs, *Acanthomia horrida* (Hongo and Karel, 1986).

Heliothis armigera Hubner has also been effectively controlled by neem extracts in India and the extracts were as effective as many conventional insecticides, such as Pyrethroids, Cypermethrin, Malathion, and Endosulfan (Parmar and Srivastava, 1987).

Field trials in Thailand have shown that piperonyl butoxide added to neem extract increased the efficacy of the neem and the combination was as active as cypermethrin (0.025%) against *Plutella xylostella* (Linn.) and *Spodoptera litura* F. (Sombatsiri and Tigvattanont, 1987). In the Dominican Republic water extracts of the neem seed was reported to effective against *Aphis*

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gossypii (Clov.) on cucumber and okra and also against *Lipaphis erysimi* (Kalt) on cabbage (Schmutterer and Ascher, 1987).

In the savanna zone of Nigeria, neem plantations cropped to peal millet and sorghum had lower grasshopper populations than either grazing land or plantations of *Acacia arabia* L. (Amatobi *et al.*, 1988). Powdered neem seed kernels or leaves have been used in seed stores and warehouses to reduce insect infestation and damage to grain during a three to twelve months storage period (Makanjuola, 1989). Also birch trees sprayed with neem extract against birch leafminer (Fenusa pusilla) performed significantly better than the registered commercial pesticide Diazinon (National Research Council, 1992). Salem (1991) found neem seed oil at 100ppm to be significantly similar to Sevin 10% (Carbaryl) in controlling potato against *Phthorimaea operculella* Zell. Margosan O (a synthetic neem product) has been reported to inhibit larval feeding in the cabbage white butterfly, *Pieris brassicae* (Linn) (Luo *et al.*, 1995).

Laboratory studies of neem products against the European corn borer showed 100% mortality at 10-ppm azadirachtin and 90% mortality at 1-ppm. Lower concentrations (0.1ppm) left the larvae apparently unaffected but adults which, emerged had significantly altered sex ratios in favour of male. The few females, which emerged, laid fewer egg and laid them late (Arnason *et al.*, 1985). In the laboratory, neem seeds and neem extracts were found to be as effective as the pyrethrum for the control of aphids on pepper and strawberry (Lowery *et al.*, 1993). Azadirachtin has been shown to affect metamorphosis, fecundity and reproduction of *C. capitata* in the laboratory (Stark et, al., 1990)

In Ghana, neem extracts were found to effectively reduce damage by insect pests of okra, stored maize, cowpea, eggplant and cocoa (Cobbinah and Osei-Owusu, 1988; Cobbinah and Appiah-Kwarteng, 1989; Tanzubil, 1992b; Afreh-Nuamah, 1995; Adu-Acheampong, 1997)

Water extracts of neem cake have been shown to have nematicidal action (National Research Council, 1992). In India, amending soils with sawdust and neem cake dropped the root knot

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index to zero and of all the treatments tested, neem gave the highest growth of tomatoes (National Research Council, 1992). Neem has also been demonstrated to have anti-fungal activity. In one test, neem oil protected the seeds of chickpea against the serious fungal diseases such as *Rhizoctonia solani* (Kuhn), *Sclerotium rolfsii* (Sacc) and *Sclerotina sclerotiorium* (Lib). It has also been shown that neem can slow the growth of *Fusarium oxysporum* (Schelecht), but did not kill it (National Research Council, 1992).

In spite of these remarkable potentials in pest control, neem has proved to be undetrimental to unintended targets (National Research Council, 1992). For example it has been shown that neem-based compounds are safe against adult European honeybee, *Apis mellifera* L (Naumann, *et al.*, 1994; Schmutterer, 1995).

Vollinger (1995) showed that resistance to neem product is possible, but the development of resistance is slow and unstable because it was suggested that a blend of active constituents in a botanical insecticide such as neem might diffuse the selection process mitigating the development of resistance.

The above review underscores the need to explore the potential in the neem tree for use in sound pest management strategies for the control of *C. capitata* in Ghana.

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CHAPTER THREE

3.0 SEASONAL ABUNDANCE AND ACTIVITY PATTERN OF *C. CAPITATA*

3.1 Introduction

Information on the seasonal population fluctuation and peak periods of *C. capitata* activity patterns are important component of pest management strategies because a warning of the timing and extent of pest outbreak can improve efficiency of control measures (Dent, 1992). Central to any pest monitoring programme is the sampling technique that is used to measure changes in insect abundance (Dent, 1992). Although supplementary information about the insect's history and the influence of weather may be needed to produce a pest forecast (Hill and Walker, 1982), pest sampling technique provides the basic measure by which the state of the system could be assessed. The estimate of pest abundance or change in numbers provide the essential measure by which control decision could be made (Dent, 1992). Hence, it is important that the sampling technique used in any monitoring programme is appropriate.

Traps baited with trimedlure (tert-butyl 4 and 5) - chloro cis and trans-2-methyl cyclohexane-1-carboxylate TML) (McGovern *et al.*, 1986) and other pheromones are used to monitor the population of fruit flies, to predict pest development and to forecast pest damage. Newly emerged adult populations of fruit flies can be detected by monitoring traps set in areas susceptible to fruit fly attack. For example, New Zealand has no fruits associated with any species of Tephritidae, but susceptible areas of the country are covered by a grid of monitoring traps designed to detect any arrival of fruit flies (Somerfield, 1989; Cowley 1990; Barker *et al.*, 1991)

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In monitoring programmes trap catches are used to estimate population density, so limited capacity traps are considered unsuitable (Carde and Elkinton, 1984). An ideal *C. capitata* trap must be easy to service, able to capture or attract and protect captured fruit flies from rainfall and remain intact during windstorm. However, most commercially designed traps do not meet these criteria. This therefore means that different trap designs are effective at different times. Numerous traps have been designed to monitor adult fruit fly populations in orchards (Drew 1982). Moreover, an important criterion for any trap monitoring system used for short term pest forecasting is that the relationship between trap catches and a corresponding field infestation should be consistent (Srivastava *et al.*, 1992)

The objectives of this experiment were;

1 To determine the effectiveness of rectangular paper traps baited with trimedlure in detecting field populations of *C. capitata*

2 To determine the seasonal fluctuation and diurnal activity pattern of *C. capitata* in citrus orchards in Ghana.

3.2 Materials and methods

3.2.1 Pheromone traps, trimedlure and experimental plots

Four rectangular paper traps (Plate 1) one side of which was coloured yellow and the other black were used to determine the seasonal abundance and activity pattern of the flies. The choice of the yellow colour was based on earlier report that yellow colour is highly attractive to Mediterranean fruit fly (Economopoulus, 1989; Uchida *et al.*, 1996).

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Plate 1: Rectagular paper traps baited with TML used for monitoring adult fruit fly population

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The trimedlure (Med-Call) was received from Samkei Chemicals, Japan through Japan International Co-operation Agency (JICA), (Ghana) in sachets of 10. The trimedlure (TML) had been impregnated in a cotton wick. The TML sachets were singly removed and hanged in the traps as shown in plate 1. Two traps each were placed in two commercial orchards of Late Valencia (*Citrus sinensis* (L) (Osbeck) of size 2.25 hectares and Satsuma (*Citrus unshiu* Marc.) also of size 2.58 hectares. The traps were randomly placed within the two fields. These citrus varieties were selected based on preliminary observations by Afreh-Nuamah (1985) that though all citrus varieties were susceptible to *C. capitata* attack the soft skinned types were most susceptible to fly damage. Within each field the traps were placed about 30 meters apart and 3 meters above the ground level. This was done to reduce the possibility of inter-trap interference.

Daily observations were made between 8:00-10:00 am. from September 1997 to July 1998. The time in the day was later extended from 6:00 am-6:00 pm. for two weeks in February 1998 for Late Valencia and April 1998 for Satsuma. The aim was to study the period of high *C. capitata* activity and their behaviour during the day. To study fly activities random observations were made on adult C. capitata in the Late Valencia and Satsuma orchards. The fly activities were categorised according to numbers and behaviour. Fly behaviour included oviposition, resting, short intermittent flights, mating and feeding around oviposition punctures by touching fruit surface with proboscis. Each TML was replaced after every two weeks based on manufacturers recommendation and personal observation in the field.

3.2.2 Statistical analysis

Correlation coefficients and multiple regression analyses were determined using SPSS and GENSTAT procedures for the mean number of *C. capitata* attracted by the pheromone traps in the two citrus orchards i.e. Satsuma and Late Valencia and some climatic factors

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such as maximum and minimum temperatures. Other climatic factors used include relative humidity at 0900 and 1500 hrs GMT, the total amount of rainfall and the number of rain days recorded within the period. With the exception of the maximum and minimum temperatures, which were recorded in the field, all the climatic variables were obtained from a local meteorological station located at ARS, Kade.

Data analyses were done on the transformed data using square root of X+0.5, but actual insect numbers are presented in the tables.

3.3 Results

3.3.1 Number of *C. capitata* attracted by Trimedlure baited traps

Results from this work indicated that *C. capitata* Wied. was present in all the two orchards throughout the period of the monitoring i.e. from September 1997 to July 1998. During this period two peaks of *C. capitata* adult fly abundance were evident in the Satsuma citrus orchard. These peaks occurred in September 1997 and March 1998. The average numbers of *C. capitata* attracted per trap during these peak periods were 38.5 and 68.5 (n = 2) respectively (Fig 1).

Two peaks of fly abundance were also observed in the Late Valencia plot. These occurred in December 1997 and March 1998 at 36.5 and 29.5 (n = 2) respectively (Fig 1).

The peak periods observed coincided with the periods when most citrus fruits were maturing and had attained the orange yellow colour.

The fly population was maintained relatively high even after harvest. This is because complete harvesting is usually not achieved at ARS, Kade. Thus, remaining fruits after harvest continue to serve as oviposition sites for newly emerged adults. There was no significant correlation (r = 0.1765 NS) between the mean number of *C. capitata* attracted by the different traps in the orchard of the different citrus variety.

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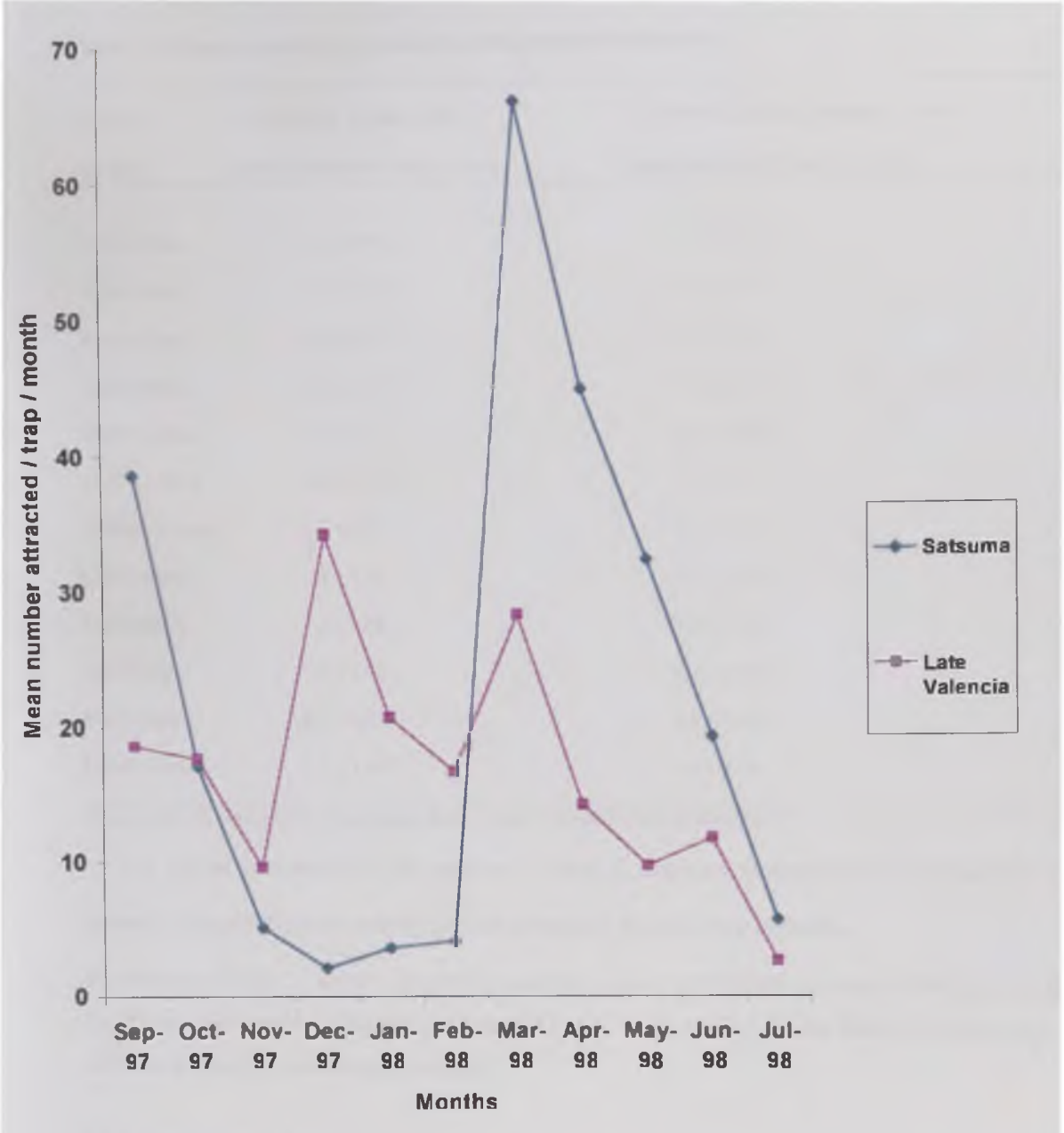

Fig. 1 Number of *C. capitata* attracted by trimedlure baited traps in two citrus orchards

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3.3.2. Periods of high activity of *C. capitata*

Table 1 shows the number of adult *C. capitata* recorded in the two orchards.

Table 1: Mean numbers of *C. capitata* attracted per trap within the day

Period of the day	SATSUMA (April, 1998) mean no. attracted/ trap (± s.e.m)	LATE VALENCIA (February, 1998) mean no.attracted/ trap (± s.e.m)
6.00-7.00am	0.4 ± 0.66	1.00 ± 1.00
7.00-8.00am	2.1 ± 2.66	1.9 ± 2.26
8.00-9.00am	7.0 ± 2.93	5.3 ± 3.58
9.00-10.00am	9.1 ± 5.79	10.0 ± 6.54
10.00-11.00am	2.4 ± 2.11	4.4 ± 3.88
11.00-12.00nn	2.0 ± 2.46	2.1 ± 2.7
12.00nn-1.00pm	1.1 ± 1.37	1.0 ± 1.10
1.00-2.00pm	1.4 ± 1.26	1.1 ± 1.24
2.00-3.00pm	1.3 ± 1.35	1.4 ± 1.28
3.00-4.00pm	1.4 ± 1.28	3.1 ± 2.47
4.00-5.00pm	5.7 ± 8.36	4.4 ± 3.47
5.00-6.00pm	1.7 ± 1.34	1.4 ± 1.96

Means were calculated from 10 sampling dates. s.e.m = standard error of the mean

The results indicated that the number of adult *C. capitata* attracted by the pheromone traps showed a bimodal fly peak activity periods in each of the two citrus orchards.

The periods of high *C. capitata* activities occurred between 8:00-10:00 am. and 4:30-6:30 pm.(Tab. 1) with average numbers attracted per trap of 9.1 and 5.7 respectively in the Satsuma orchard and 10.0 and 4.4 in the Late Valencia orchard.

3.3.3 Orientation behaviour of adult *C. capitata* to pheromone traps

Results obtained from this work showed that 35% of observed adult *C. capitata* showed short intermittent flights around the immediate vicinity of the traps in the Satsuma citrus orchard (Table 2)

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(n=120). This was followed by walking or foraging on ripened citrus fruits whilst probing fruit surfaces. The least behaviour observed in the Satsuma orchard was mating. (Table 2). In the Late Valencia citrus orchard the most predominant behaviour observed was also short intermittent flights (46.67% of observed adult). Feeding at probing centres followed this. The least behaviour observed was oviposition (Table 2).

Table 2: Percent *C capitata* adult showing orientation behaviour to trimedlure pheromone traps from 8:00am. – 10:00am.

Behaviour	Satsuma (%)	Late Valencia (%)
1. Short intermittent flights	35.0	46.7
2. Oviposition	17.5	1.8
3. Feeding at probing punctures	10.8	25.0
4. cleaning of proboscis	11.7	7.5
5. Moving on fruit surfaces with probing activities	21.7	14.0
6. Mating	3.3	5.0

n = 120

3.3.4 Effect of climatic factors on population of *C. capitata* in citrus orchards

Mean monthly temperatures recorded in the Late Valencia and Satsuma orchards ranged from 22-28°C during the cooler months (June-September) and 25-32°C during the hot season (November-February). The highest and lowest rainfall values of 234.2 and 30.0 were recorded in the months of May and January 1998 respectively. These were distributed within 12 and 4 days, respectively.

Multiple regression analysis of the effect of some climatic factors on the field population of *C. capitata* in the Satsuma citrus orchard revealed that only relative humidity at 0900hrs GMT had a significant negative correlation with the population of *C. capitata* attracted by the traps (Table 3).

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Table 3: Multiple regression analysis of the effect of some climatic factors on the field population of *C. capitata* attracted by pheromone traps in Satsuma citrus orchard

Variable		Partial regression Coefficient	standard error	t value	Prob.of t
X_1	Max. temp.	5.250	16.575	0.317	0.7569
X_2	Min. temp	20.969	25.220	0.831	0.422
X_3	Rel Hum. (0900hr)	7.530	2.438	-3.089	0.009*
X_4	Rel. Hum (1500hrs)	2.777	2.473	1.123	0.283
X_5	Total mon. rainfall	0.618	0.358	1.731	0.109
X_6	no. of rain days	7.140	5.523	1.293	0.220
A = -421.050		496.657			

Equation; $Y = -421.050+5.25X_1 +20.97X_2 -7.53X_3+2.78X_4 +0.618X_5+7.14X_6$

sig t = probability level of t value $R^2 = 0.5740$

For every increase in relative humidity, *C. capitata* population was reduced by 7.53 (Table 3). Taken collectively, all the climatic factors influenced *C. capitata* population by 57.40% which is given by the value of the coefficient of determination ($R^2 = 0.5740$).

Table 4: Multiple regression analysis of the effect of some climatic factors on the field population of *C. capitata* attracted by pheromone traps in late Valencia citrus orchard

Variable	Partial regression Coefficient	standard error	t value	Prob.level of t
X_1 Max. temp.	-27.77	16.37	-1.17	0.12
X_2 Min. temp	-7.70	24.90	-0.31	0.76
X_3 Rel Hum (0900hrs)	-1.64	2.41	-0.68	0.51
X_4 Rel. Hum (1500hrs	-1.52	2.44	-0.62	0.55
X_5 Total mon. rainfall	-0.10	0.33	-0.28	0.02*
X_6 no. of raindays	1.66	5.45	-0.30	0.77
	A = 1259.5159	490.3912		

Equation; $Y = 1259.516-27.77X_1-7.70\ X_2 -1.64X_3-1.52X_4 -0.098X_5-1.65X_6$

probability level of t value $R^2 = 0.4004$

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This suggests that climatic factors were important in determining the population of *C. capitata* in Satsuma orchards. In the Late Valencia orchard the multiple regression analysis also showed significant effect of total monthly rainfall on the population of *C. capitata*. For every increase in total monthly rainfall the population of *C. capitata* was reduced by 0.10. On the whole climatic factors contributed to 40.04% ($R^2 = 0.4004$) in reducing *C. capitata* population in the Late Valencia orchard (Table 3). This means that ther was some correlation between the climatic factors and *C. capitata* development and population in the Late Valencia orchard.

3.4 Discussion

The results from this work showed that *C. capitata* is present at the Agricultural Research Station, Kade throughout the year as long as citrus varieties, which mature at different times, are present. Fly abundance within the year coincides with the period when fruits are about to ripen. This result confirms earlier report by Afreh-Nuamah (1985) that the peaks of the fly occurred in September to November, and also April and May, when most fruits were ripened. Matiola *et al.*, (1990) found that in Brazil significantly higher numbers of tephritid flies were captured in pheromone traps during the ripening period in October to February. Also Barker *et al.*, (1990) working in mango and apple orchards found that significantly higher numbers of *C. capitata* females were captured in traps baited with trimedlure during their ripening periods.

The present study also confirms observations by earlier researches working on other genera of fruit flies. For example, Zahler (1991) showed that *Anastrepha obliqua* Macquart another species of fruit fly in an unripe mango orchard was low but population increased as fruits ripened and declined as the number of ripened fruits declined. It was recommended that monitoring of mango orchards in Brazil should be done during the fruiting and maturation periods (October-February). Satsuma which has two maturing periods within a year (May and June and September-Mid October) had fly peaks coinciding with these periods (Fig 1).

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The population of fly captured in the traps in fig. 1 give an indication of the movement of the fly within plots. Between December and February, the population was high in the Late Valencia plot but just after the fruit harvest, they moved to the adjacent Satsuma plot where fruits were maturing. Fly population in the Satsuma, therefore started rising in March. The rise in population in the Satsuma could also be due to emerging flies from eggs laid in Late Valencia fruits before harvest.

Many researchers working with other fruit fly genera have reported the population flux between commercial orchards and other native vegetation adjacent to each other. Adult females of *B.* (Dacus) *frontalis* (Becker) in the Cape Verde Island was found to move into cucurbit plantations from adjacent plants (*Citrus spp*, *Zea mays* L.; and *Cajanus cajan* L) used for resting in the late afternoon hours (Steffens, 1983). A similar observation was made by Aluja *et al*., (1996) working with commercial mango orchards in southern Mexico that most flies of *A. obliqua* were captured in traps placed at the periphery of those orchards.

The number of flies attracted by the traps during the day showed high insect activity between the hours of 8:00 – 10:00 am and 3:00 –5:00 pm in all the varieties. This may suggest that insects came out from their hiding places to feed when the sun was up but not so hot and hence their numbers were high in the morning than hot afternoons. Also during the day when it was very hot, insects were observed resting or hovering with intermittent short distant flights on the trees and weeds under the plantations. On the average, it took between 40 - 60 minutes for the first adult fly to appear on the traps during the period of high insect activity. The time lag between trap placement and attraction of first adult fly to the pheromone trap could be due to the time required for the pheromone to diffuse within the field. Similar behaviour of *C. capitata* has been shown for other traps. For example, in Egypt it was observed that about 90% of females adult *C. capitata* attracted to standard TML-baited traps remained within 1 m radius on the surrounding foliage without getting into contact with the traps. Similarly, about 30% of the males which aggregated around the traps did not fly away but waited to compete for mating with approaching females (Hendrichs *et al*., 1989)

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The bimodal daily activity pattern clearly exhibited by *C. capitata* in the citrus groves has also been observed by some workers working on other genera of fruit flies and other arthropods. For example, Aluja *et al.*, (1997) working with papaya fruit fly, *Toxotrypana curvicauda* Gerstaecker, in Mexico found that the adult flies showed two distinct peaks.

Throughout the period of the experiment the most important factors, which contributed to population increase in the Satsuma orchard were host availability and relative humidity whilst in the Late Valencia citrus orchard host availability and total monthly rainfall were important in the development of *C. capitata*. This confirms earlier observation (Hanna, 1947; Hagen *et al.*, 1981). In addition daily temperatures of 24^0C - 30^0C were reported to be of greatest potential for population growth of *C. capitata* whilst temperatures below 18^0C inhibited development of *C. capitata* (Hagen *et al.*, 1981).

The effects of meteorological factors have also been reported for other fruit flies such as the Oriental fruit fly, *B. dorsalis*. Serit and Keng-Hong (1990) observed significant positive correlation of maximum and minimum temperatures and maximum relative humidity on trap catches of *B. dorsalis* in mango orchards in India. Also Yokoyama and Miller, (1993) found that diapause and low field temperatures are the main factors that slow pupal development and retard adult emergence of some tephritid flies in the field.

Although the population of *C. capitata* seemed to have correlated significantly with some climatic factors recorded, their effect was not very strong. This may be attributed to the fact that most of the climatic factors were recorded outside the field. It is therefore possible that fly behaviour might have been influenced by micro-habitat rather than conditions outside the field. The reason is that in a close canopy like what was observed in the orchards, the micro-habitat conditions in shelter sites could have been an important factor in determining when the flies moved in and out of the citrus trees thereby been attracted by the pheromone traps. This was evident in the field because most of the flies were observed to be very inactive during period when the weather was

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cloudy hence resulting in few insects been attracted during the wet season even though the Satsuma was then in season (fig. 1). Since most of the climatic factors except temperature were not recorded at the level of confinement possibly needed the correlation obtained appears weak.

Aluja and Birke (1993) stressed the importance of microhabitat conditions on *Anastrepha obliqua* Macquart presence and diurnal patterns of activity in a highly ephemeral and diversified environment.

The difference in the number of *C. capitata* attracted by trimedlure baited traps per month in both citrus orchards confirmed the seasonality of *C. capitata* and clearly established the seasonal abundance and period within the day when activities were high. Srivastava *et al.*, (1992) showed that correlations of such monitoring work is better where the difference between seasons are marked.

This work is consistent with the results obtained by earlier workers that trimedlure is a powerful lure for fruit fly males (Beroza *et al.*, 1961; Cunningham and Couey 1986; Liquido *et al.*, 1993). Liquido *et al.*, (1993) showed however, that, a combination of trimedlure and ammonia resulted in a significantly higher numbers of *C. capitata* males successfully caught in Jackson traps than with trimedlure only. The relatively higher capture efficiency of pheromone traps proved useful in surveillance and detection programs.

However, on their own, the results of the abundance of *C. capitata* in the two citrus orchards have limited value unless it can be related to other variables like levels of damage. The work was further extended to establish the levels of damage due to *C. capitata* damage in two citrus orchards.

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CHAPTER FOUR

4.0 LABORATORY EVALUATION OF NEEM SEED WATER EXTRACT ON THE OVIPOSITION AND IMMATURE STAGES OF *C. CAPITATA*.

4.1 INTRODUCTION

Immature stages of *C. capitata* are very difficult to control with insecticides because they are embedded in the fruits or soil. The eggs and larvae are completely concealed in the fruit while the pupae are formed in the puparium in the soil. Control of the insect is directed mainly against the adult fly (Hagen *et al.*, 1981; Stark *et al.*, 1990; 1991).

Because of the unique properties and great potential of neem as an insecticide, a series of laboratory experiments were conducted to determine the effect of different neem concentrations on the oviposition and development of *C. capitata* at the University of Ghana, Agricultural Research Station, Kade.

4.2 Materials and methods

4.2.1 Collection of neem seeds

Neem seeds used in the study were obtained from Accra, around 37 Military Hospital areas and Kordiabe near Dodowa in the Greater Accra Region of Ghana. The berries were collected from the trees and seeds were depulped from them. Seeds were subsequently air-dried and then stored in jute sacks. These were used as and when needed.

4.2.2. Preparation of neem seed water extract

The dried neem seeds were ground into fine powder using an electric blender (Model 32BL79, Waring Products, USA) obtained from the West African Plantain Project (International Institute of Tropical Agriculture) laboratory at ARS, Kade. 15, 20, 25, and 30 g of the powder were weighed

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separately into 1000 ml capacity beakers and 100 mls of distilled water was added to each beaker to give the estimated 15%, 20%, 25% and 30% wt/vol suspensions. These were left overnight to ensure that there was adequate infusion of the active ingredient of the neem seed into a fine linen cloth. The filtrates were then used as treatments. The moisture content of the ground neem was 22.2 ± 2.8%. The moisture content (MC) was based on earlier report that the efficiency of neem seed was highest at higher MC with diminishing effects as MC declined (Adu-Acheampong, 1997). At low moisture levels, the bulk of the active compounds is contained in oil component of the seeds and that most of these oils were not water soluble hence very little amount of the active compounds could be extracted by the water (Adu-Acheampong, 1997).

The MC was estimated from five groups of 50-g samples of neem seeds taken from the seed stock which were weighed before and after sun drying for 72 hours. The %MC (fresh weight basis) was calculated from the formula

$$\%MC = \frac{\text{Initial weight (g)} - \text{final weight (g)}}{\text{Initial weight (g)}} \times 100$$ (Adu-Acheampong, 1997)

4.2.3. Collection and culturing of test insects

Adults of *C. capitata* were collected from ARS experimental plot Ci 10 and Ci 13 using mouth aspirators (2) into wooden-framed fine nylon mesh cages of approximately 90 cm long by 90 cm wide by 100 cm high consisting of three chambers (Plate 2). Two of the chambers contained 100 females and 100 males separately and the other contained 20 males and 80 females. All weak insects were discarded. The culture was established in November 1997.

The chambers where females were kept contained sand and citrus fruits to serve as oviposition sites. The insects were fed on sugar solution (i.e. 10% sugar solution), water and citrus juice from the different citrus varieties. This was done by mopping the sugar solution and ripened citrus juice in cotton wool and then hanged in the chambers. Some of the sugary solution and citrus juice were also smeared on small rectangular plastic rubbers so that the flies could get access to food when they

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land on them. The cotton wool was changed every other day in order to prevent decay and rot to avoid infection by micro-organism. The insects were maintained at 27 ± 2^{0}C and a photoperiod of 12:12 L:D. Humidity was not controlled.

4.2.4 Oviposition of *C. capitata* on citrus

Fifteen adult females and five adult males were collected from the insectary into 6 insect cages measuring 30 cm long by 30 cm wide by 30 cm high (plate 2). Two each of Late Valencia, Frost Valencia, Satsuma, Lake Tangelo, Subi and Anomabo were harvested and put into the cage containing the insects. The experiment was left for three days after which the fruits were collected and the number and position of punctures were counted and recorded.

Insects were fed on water, citrus juice and 10% sugary solution. The experiment was replicated six times. Flies, which failed to oviposit when presented with the uninfested fruits were discarded and the experiment repeated. Punctured fruits were transferred into different insect cages containing some amount of sand. The experiment was monitored until adults emerged. The time taken for adults to emerge and the sex ratio were recorded.

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Plate 2: Insect cages used for the laboratory experiment

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4.2.5 Ovipositional preference

Choice and no choice experiments were conducted in the laboratory using Satsuma and Late Valencia in wooden-framed fine nylon mesh cages measuring 30 cm x 30 cm x 30 cm. Each cage was large enough to accommodate two separate fruits.

4.2.5.1 Choice experiment

Twenty adult female *C. capitata* were collected from the insectary and put into an insect cage. One ripened Satsuma and Late Valencia fruits each were harvested and put together in the cage containing the insects. Care was taken to ensure that fruits had not been damaged already. The experiment was left for 96 hours after which the fruits were collected, and the number and position of puncture were counted and recorded. The experiment was replicated five times. Punctured fruits were transferred to different insect cages containing some amount of sand to serve as pupation medium for the developing larvae. These procedures followed that of AliNiazee and Brown, (1977) and Cowley *et al.*, (1992).

The experiment was monitored untill the adults emerged. The time taken for adults to emerge and the sex ratio were recorded.

4.2.5.2 No choice experiment

Two separate cages were used. The harvested Satsuma and Late Valencia fruits were put separately into the cages containing 20 adult female flies. After 96 hours the fruits were removed and the number of punctures per fruit was recorded. Punctured fruits were transferred separately into two insect cages and monitored untill the adults emerged. Five replications were used. The time of emergence and the sex ratio were recorded.

4.2.6. Ovipositional bioassay

Twenty adult insects consisting of 15 females and five males were collected form the insectary using a mouth aspirator into 20 different insect cages each. Two Satsuma fruits with similar colouration, size and shape (Papaj *et al.*, 1989b; Cowley *et al.*, 1992) were harvested from the field and put into the five insect cages each representing a replicate. Prior to the introduction of the fruits,

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15%, 20%, 25% and 30% wt/vol of neem seed water extract (NSWE) were prepared as described previously. The treatment were therefore used:

T1 Citrus fruits and adult *C. capitata* were sprayed together with the different NSWE suspensions in the cages;

T2 Fruits were treated with the NSWE suspensions before introducing them into the cages containing unsprayed adult *C. capitata*.

T3 Adult *C. capitata* were sprayed with the NSWE suspensions in the cages before introducing citrus fruits, which had not been treated with NSWE.

T4 Adult flies and citrus fruits were sprayed with only water to serve as control.

Spraying was done with a hand-held mist applicator at a flow rate of 0.50 mls/min. The experiment was left for 96 hours to provide enouugh time for the insects to access the fruits. The insects were fed on sugary solution and citrus juice. The experiment was repeated using Late Valencia.

The number of punctures on the fruits at the end of the 96 hours was recorded. Punctured fruits were transferred into different insect cages containing sand. These were monitored untill the adults emerged. The number of adults that emerged, the days taken for adults to emerge and the sex ratio were recorded. Each treatment was replicated six times.

4.2.7 Effect of neem seed water extract (NSWE) on larval development.

Five groups of twenty-second instar larvae aged 5 – 7 days were obtained from infested dropped citrus fruits collected in the field. Each group of twenty were put into five separate petri dishes and each topically sprayed with one of five dosages of NSWE ie 15%, 20%, 25% and 30% wt/vol. and a control of water only. The larvae were made to stay in the spray and petri dishes for 10 minutes to ensure that considerable amount of active ingredient had been absorbed through the insect cuticle into the body. Each group of treated larvae was transferred into one sterilised citrus fruit for further

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larval development. The sterilisation of the fruits was done by putting the fruits in water at temperature of 30^0C for two days to kill all eggs and larvae hidden in the fruits. The sterilization process followed that of Eskafi and Fernandez (1990) who observed that larvae *C. capitata* submerged under water survived up to three days and four days, respectively at room temperature.

Each containing the treated larvae was placed separately in different insect cages containing some sand. Larvae, collected from Satsuma, were put in sterilized Satsuma fruits. Similarly, sterilized Late Valencia fruits were used for larvae collected from Late Valencia, to eliminate any error that could result from varietal differences. Different citrus varieties have different fruit characteristics at different stages of their development. The pH, titrable acidity, total soluble solids differ from fruit to fruit and variety to variety (Davies and Albrigo, 1994). The experiment was monitored until the adults emerged. Adults which emerged from treated larvae were exposed to fresh fruits to study their fertility after emergence The same procedures were followed using third instar larvae. The number of adult *C. capitata* that emerged and the number of days taken for adults to emerge after NSWE treatment and sex ratio were recorded.

4.2.8 Effect of neem seed water extract on pupal development.

Pupae were collected from a breeding stock in the laboratory. Five groups of twenty pupae each were separately placed into five petri dishes. The pupae were treated with 15%, 20%, 25% and 30% wt/vol of neem seed extract and control of water only. This constituted one set in which the pupae and soil were sprayed together. In the other set the sand was sprayed before introducing the untreated pupae to the treated sand. Each of the treatments were used to determine whether neem seed extract could be used as a substitute for synthetic insecticides applied to the soil against the pupae of some fruit flies. The experiment was monitored until the adults emerged.

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4.2.9 Statistical analysis

Data obtained were transformed using square root of (x+0.5) where appropriate before they were subjected to analysis of variance. The Abbot's (1925) formula was used to correct mortality due to natural factors Means were separated with Least Significant difference at (P = 0.05). A chi-square analysis was determined for the sex ratio of the emerged adult flies Correlation coefficient was determined for the number of adult *C. capitata* that emerged from punctured fruits and the number of oviposition punctures formed on the citrus fruits used.

4.3 Results

4.3.1 Oviposition of *C. capitata* on citrus species

The results showed that the number of punctures found on the citrus varieties used was not significant (F = 1.8 NS, df = 5, 30 P = 0.05) (Table 5) (appendix 3). However fewer numbers of punctures were consistently formed on the sweet orange (*Citrus sinensis*) and the local varieties (*Citrus sinensis)* than the mandarins (*Citrus unshiu*).

The results showed that it took between 38-41 days for the fly to complete its development in the laboratory. The difference in number of days taken for the adult to emerge from infested fruits was not significant for each treatment (F = 1.93 NS, df = 5,30 P = 0.05) (Table 5) (Appendix 4). The small difference of about 2 days between the sweet oranges and the hybrids may be due to the fact that the insects took a little more time in successfully ovipositing on the Late Valencia which is hard skinned compared with Satsuma (Table 5).

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The small difference of about 2 days between the sweet oranges and the hybrids may be due to the fact that the insects took a little more time in successfully ovipositing on the Late Valencia which is hard skinned compared with Satsuma (Table 5).

The number of adults that emerged from Late Valencia was three times lesser than that of Satsuma (Table 5). The number of adult *C. capitata* emerging from Late Valencia consistently, had fewer adults than the Satsuma fruits.

Table 5: Oviposition of *C. capitata* on citrus fruits

Variety	position of puncture	mean no. of punct./fruit ± s.e.m	days for adults to emerge	sex ratio male:female
Sweet orange(exotic)				
(Citrus sinensis) cv Late Valencia	Distal	0.83 ± 0.98	39.8 ± 1.21	8:0(4.00)*
Frost Valencia	Proximal	1.17 ± 1.17	40.2 ± 0.71	4:8(1.33)NS
Hybrid				
(Citrus unshiu) cv Satsuma	Distal	2.17 ± 0.75	38.6 ± 1.76	14:11(0.036)NS
Lake Tangelo	Distal	2.50 ± 1.38	38.2 ± 2.10	18:16(0.00)NS
Sweet orange (Local)(*Citrus sinensis cv*)				
Subi	Proximal	1.50 ± 1.21	41.4 ± 1.36	9:4(3.06)NS
Anomabo	Distal	1.83 ± 1.17	40.6 ± 1.44	5:8(1.67)NS
		NS	**NS**	

NS: not significant at (P=0.05). Numbers in parentheses are Chi-square homogeneity test values, df = 1 * significant at (P = 0.05) cv = cultivar

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The correlation between the mean number of punctures formed on the varieties tested and the total number of adults that emerged from the fruits was highly significant ($r = 0.914^{**}$). This suggests that the total number of adult *C. capitata* that may emerge from punctured fruits will depend on the number of ovipositional punctures.

4.3.2 Ovipositional preference *of C. capitata* to Satsuma and Late Valencia citrus fruits

Results showed that in both choice and no choice tests fewer numbers of punctures were found on Late Valencia than Satsuma. There was, however, no significant difference between the varieties i.e. ($F = 2.41$, $df = 1,8$ $P = 0.05$) for choice test and ($F = 0.987$, $df=1,8$, $P = 0.05$) for no choice test (Table 6).

This means that there is no varietal preference for oviposition. Each of the varieties has equal chance of being damage by the adult fly. However, because of the hard skin of Late Valencia fruits it tend to provide some physical barrier against the insect from drilling it long ovipositor through. This resulted in fewer number of punctures formed on Late Valencia (Table 6).

Table 6: Oviposition preference of *C. capitata* Late Valencia and Satsuma citrus fruits

Variety	Choice Mean no. of punct. Per fruit ± s.e.m	No choice Mean no.of punct. per fruit ± s.e.m
Late Valencia	1.40 ± 0.54	1.80 ± 0.44
Satsuma	2.40 ± 0.32	2.20 ± 0.84
	NS	NS

NS : not significant at (P=0.05).

4.3.3 Effect of neem seed water extract on oviposition of *C. capitata*

Results obtained from this work showed that neem seed water extract reduced to a greater extent the number of punctures made by the flies on the two citrus varieties. Whether the citrus fruits and adult fruit flies were sprayed together or only citrus fruits were sprayed before introducing insects, had no significant effect on the number of punctures created on the fruits ($F = 1.27$, $df = 3,132$,

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p=0.05) (Table 7). However, neem seed water extract significantly reduced the number of punctures made on both Satsuma and Late Valencia ($F= 10.53, df = 9,132$ $p< 0.005$) (Table 7).

The performance of the control and 15% wt/vol was significantly the same (Table 7). There was significantly less number of punctures on Late Valencia than Satsuma (Table 7). The number of days taken for adults to emerge from NSWE treated fruits was significant among treatments. 30% wt/vol of NSWE significantly delayed the emergence of adults from NSWE treated Late Valencia fruits.

The sex ratio of adult *C. capitata* that emerged from punctured fruits did not differ significantly from 1:1 except when 20% NSWE was sprayed on late Valencia (Table 7).

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Table 7: Effect of neem seed water extract on oviposition of *C. capitata* in the laboratory

Citrus variety	NSWE dosage (% wt/vol)	Mean no.of punctures/fruit ± s.e.m	Days to emerge ± s.e.m	Sex ratio M:F
	Control	2.44 ± 0.86^{a}	38.6 ± 1.14^{de}	28:25 (0.08)NS
	15	2.31 ± 0.98^{abc}	38.2 ± 1.30^{e}	18:28 (2.17)NS
Satsuma	20	1.50 ± 0.87^{a-d}	39.2 ± 0.84^{c-e}	5:10 (1.66)NS
	25	0.81 ± 0.88^{b-d}	39.8 ± 1.31^{b-e}	10:8 (0.22)NS
	30	0.63 ± 0.86^{cd}	41.0 ± 1.58^{ab}	3:6 (1.00)NS
	Control	1.88 ± 0.93^{a-d}	38.6 ± 1.67^{de}	16:12(0.44) (NS)
	15	0.29 ± 0.96^{d}	40.8 ± 1.64^{b-e}	6:6 (0.00)NS
Late Valencia	20	1.06 ± 0.75^{a-d}	40.2 ± 2.14^{b-e}	4:17 (8.02)*
	25	0.56 ± 0.86^{d}	42.0 ± 1.90^{ab}	9:13(0.76)NS
	30	0.56 ± 0.70^{d}	42.6 ± 1.74^{a}	7:8 (0.07)NS

Means followed by same letter in the same column are not significant from each other at P=0.05
Numbers in parentheses are Chi-square homogeneity test values , df = 1 * significant at P = 0.05, NS= not significant

4.3.4. Effect of neem seed water extract on larval development of *C. capitata*

Table 8 shows the effect of neem seed water extract on the larval development of *C. capitata* The result showed that the neem seed water extract had a highly significant effect on mortality of second and third instar larvae of *C. capitata* (F = 19.34 df = 9,40, p=0.001) (Appendix 9). The mortality of both second and third instar larvae increased with the dosages of the neem seed water extract (NSWE) (Table 8). Significant number of third instar larvae exposed died due to the neem seed water extract. Beyond 15% concentration all NSWE treatments lengthened the developmental periods of second and third instar larvea to the adult stage when compared with the control (Table 8).

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The NSWE also delayed larval development by between 1 - 7 days for 2nd instar and 3-6 days for 3rd instar larva. The delay in larval development was also dosage dependent (Table 8). Sex ratio of the adults that emerged showed that sex differences did not differ significantly.

Table 8: Effect of neem seed water extract on 2nd and 3rd instar larvae of *C. capitata* in the laboratory

Stage of deve't	NSWE dosage (% wt/vol)	Mean no.of dead larvae ± s.e.m	Days to emerge ± s.e.m	Sex ratio M:F
	Control	6.20 ± 2.23 [b]	31.2 ± 2.32 [b]	30:39 (1.17)NS
	15	13.0 ± 2.00 [a]	33.2 ± 2.48 [b]	10:25 (2.17)NS
2nd instar	20	15.6 ± 1.74 [a]	37.0a ± 1.26 [b]	17:5(0.00)NS
larvae	25	15.8 ± 1.73 [a]	39.8 ± 1.31 [cd]	12:12 (0.00)NS
	30	15.8 ± 1.47 [a]	39.6 ± 1.62 [a]	9:12 (0.43)NS
	Control	4.60 ± 2.06 [b]	16.6 ± 3.00 [d]	48:29 (4.69)*
	15	11.2 ± 1.47 [a]	15.8 ± 2.93 [d]	24:20 (0.36)NS
3rd instar	20	14.4 ± 2.24 [a]	19.80 ± 2.23 [c]	9:19 (3.57)*
larvae	25	14.5 ± 2.65 [a]	22.8 ± 2.24 [c]	12:20(2.00)NS
	30	16.8 ± 1.94 [a]	22.4 ± 3.00 [c]	10:6 (1.00)NS

Means followed by same letter in the same column are not significant from each other at P=0.05 Numbers in parentheses are Chi-square homogeneity test values, df = 1 * significant at P = 0.05

4.3.5 Effect of neem seed water extract on pupal development

On pupal development, the results obtained showed significant differences among the treatments when only the soil was spayed before introducing the pupae and when both pupae and soil were sprayed together (Table 9).

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Table 9: Effect of neem seed water extract on pupal development in the laboratory

Technique Of Applic.	NSWE dosage (% wt/vol)	Mean no.of emerged adults ± s.e.m	Days to emerge ± s.e.m	Sex ratio M:F
	Control	13.8 ± 1.60 [a]	13.6 ± 0.98 [b]	48:31(3.65)NS
Only soil	15	13.4 ± 2.30 [a]	13.4 ± 1.34 [b]	13:54 (24.54)**
Sprayed before	20	11.0 ± 2.10 [b]	13.2 ± 0.87 [b]	29:26(0.16)NS
Pupae added	25	11.2 ± 1.94 [b]	14.0 ± 1.34 [b]	24:32 (1.14)NS
	30	13.8 ± 1.60 [a]	13.6 ± 0.98 [b]	48:31 (3.36)NS
	Control	14.0 ± 2.10 [a]	9.8 ± 0.68 [c]	39:31(1.04)NS
Both pupae	15	9.4 ± 2.73 [c]	8.4 ± 1.24 [c]	20:17 (0.36)NS
And soil	20	7.4 ± 3.14 [d]	19.6 ± 2.30 [a]	18:19 (0.03)NS
sprayed	25	5.6 ± 1.50 [e]	19.4 ± 2.3 [a]	8:20 (5.14)**
together	30	4.8 ± 1.33 [ef]	21.6 ± 1.4 [a]	14:10 (0.67)NS

Means followed by same letter in the same column are not significant from each other at (P=0.05). Numbers in parentheses are Chi-square homogeneity test values, df = 1 * significant at (P = 0.05) ** significant at (P=0.09)

The number of days taken for the adults to emerge showed that 20%, 25% and 30% wt/vol of NSWE delayed the time taken for adults to emerge when both soil and pupae were sprayed (Table 9). The results show that if the neem seed water extract is applied to the soil there is the possibility of the NSWE reducing the larvae and pupae which are already in the soil before the NSWE application. However, the NSWE will not have any significant effects on larvae and pupae which will later come from dropped fruits.

Sex ratio of the adults that emerged from exposed pupae was not significant except for 15% (only soil sprayed) and 25% wt/vol of NSWE (soil and pupae sprayed together) in which more adult females of *C. capitata* emerged.

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4.5 Discussion

The results from the laboratory work showed that the ovipositional behaviour of *C. capitata* does not differ among the sweet orange (exotic and local) varieties and the mandarins grown at ARS, Kade. However, damage of the mandarins may always be higher than Late Valencia because of the thickness of the sweet oranges. Late Valencia has thick skin hence flies find it difficult to oviposit or even if they are able to puncture some of the eggs do not get to the pulp for them to hatch. Oi and Mau (1989) working with avocados showed the effect of fruit skin thickness in preventing oviposition of *C. capitata*. They observed in the laboratory that the hard skin of avocados served as physical barrier against the oviposition of *C. capitata* and Oriental fruit fly, *B. dorsalis*.

The observation from the laboratory experiment which showed that less oviposition occured at the proximal ends of the fruits where the tissues are more compact than the distal end confirms the above.

Another factor, which could have contributed to more punctures at the distal end may be due to that, female flies exploited successful oviposition(s) made by other females since in most cases a fluid-like liquid comes out after successful oviposition. Females trying to feed on this in tend laid their eggs around those punctured areas. Similar observations were made by Papaj *et al.*, (1989a) who showed that *C. capitata* females are more likely to landed on oranges that were artificially wounded than unwounded control oranges, and that having landed, they more likely attempted oviposition into a wounded orange than unwounded oranges. They also observed that females, which attempted oviposition into wounded, did so directly into or very near the wound.

Neem seed extract showed promising in preventing oviposition of *C. capitata* in the laboratory as shown in many other tephritid flies (Singh and Srivastava, 1983 ; Chen *et. al.*, 1996) who showed that extracts prepared directly from neem seeds significantly deterred oviposition of tephritids. They attributed this to the widely varied levels of bioactive compounds in the extracts prepared in this

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manner suchs as the levels of azadirachtin and other limonoids in the neem (Isman *et. al.*, 1990; Addae-Mensah, 1998).

The results showed that neem seed extract retards development of larvae of *C. Capitata* into pupal or adult stage. This contrasts with the findings of Stark *et. al.*, (1990) who reported that azadirachtin at 14 ppm exposed to larvae did not affect the formation of puparia and that 95% of adults which emerged from the treated puparia appeared normal.

Crude water extracts of neem have been reported to contain between 60-5000 ppm azadirachtin (Addae-Mensah, 1998). It is possible that the neem seed water extrac used for the laboratory work contained more azadirachtin than the 14 ppm tested by Stark *et al.*, (1990). It is also possible that other limonoids in the neem seed extract contributed to the larval and pupal mortality recorded in this work.

Other significant observations made in the experiment were that most adult flies which emerged from the second instar larvae and pupae exposed could not fly properly. A number of the flies died shortly after emergence. Others were observed to have crumpled wings shortly after emergence. Similar observation was made by Steffens and Schmuterrer (1982) that adult Mediterranean fruit fly which developed from the larvae exposed to a crude methanolic neem seed extracts had significantly reduced flight ability, female chemotactic response and mating propensity. Fecundity of adults declined with time since there was consistently fewer numbers of adults emerging from fruit exposed to neem seed water extract. This may be probably due to the fact that the NSWE inhibited embryo formation hence resulting in fewer egg production. The ovicidal effect of neem extracts have been observed in many insect pests (Schmutterer 1990;1995).

Neem seed water extract could bring about a considerable decrease in the number of oviposition punctures on citrus at least within 3 days. Also when applied to the soil, the neem seed water extract at the dosage of 30% wt/vol could delay and prevent pupae emergence.

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CHAPTER FIVE

5.0 FIELD EVALUATION OF NEEM SEED WATER EXTRACT AGAINST *C. CAPITATA*

5.1 INTRODUCTION

Although *C. capitata* is considered to be one of the most important pest of citrus in Ghana (Afreh-Nuamah, 1985), there is relatively very little information available relating to their control. Many citrus farmers maintain an intensive management regime that primarily involves regular application of insecticides aimed at decreasing adult fruit fly population (VanRanden and Roitberg, 1998). The over-dependence of synthetic chemical insecticides has brought about a number of problems to the natural ecosystem. Several practical Mediterranean fruit fly management programmes such as the use of baited traps impregnated with pesticides, are however available particularly in the United States of America.

Interest in the use of biopesticides such as neem to control insect pests is recently receiving serious attention worldwide.

Following the success of earlier laboratory investigation in the use of neem seed water extract against *C. capitata*, further field studies were planned to

1. To evaluate the effectiveness of neem seed water extract in controlling *C. capitata* under field conditions
2. . To determine the relative importance of other control strategies such as picking of damaged citrus fruits under citrus plantations and insecticide application against the adult fruit fly in Ghana.

5.2. Materials and methods

5.2.1.Calibration of spray equipment

Two spray equipment 'Urgent' (GmbH) and 'Solo' (GmbH) knapsack mistblowers of engine capacity 35 cc and 50 cc, respectively which are normally used in spraying plantation crops in

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Ghana were used. Each of the equipment was fitted with the standard air shear (gaseous) nozzle. The weight of the 'Solo' with and without load was 20 and 8 kg whereas the 'Urgent' was 18 kg with load and 8 kg without load. The fuel and spray tank capacity for the Solo is 2 and 12 litres, respectively while that of the Urgent is 1 and 12 litres, respectively (Afreh-Nuamah, 1992).

The 'Solo' has a fuel consumption rate of 38.5 mls/min and a vertical projection of 10 m. The 'Urgent' on the other hand has a fuel consumption rate of 33.3 mls/min and a vertical projection of 8 m. The 'Solo' mistblower has a separate cut-off valve and a restrictor. The 'Urgent' mist blower, however, has no separate cut-off valve. The restrictor acts as the cut-off valve when set at zero position (Afreh-Nuamah, 1992). The two equipment were interchanged during each time of the field spraying.

A neem seed water extract at a rate of 25 kg/ha was prepared as described in the laboratory bioassay and left overnight. The prepared suspension was sieved using a fine linen cloth before it was used for the calibration. The tank of each of the motorised mist blowers (i.e. Urgent GmbH and 'Solo' mist blowers) fitted with the standard air shear nozzle was each filled to capacity with the prepared neem seed water extract suspension. At full throttle the time taken to discharge the 12 litres spray liquid was recorded at restrictor selections 2 and 3 for the Urgent mist blower and fully and half-opened cut-off points for the 'Solo' mist blower. Each observation was repeated three times and the average time recorded. Water only was used as control for comparison with the neem seed water extract.

5.2.2 Study site and experimental design

The field experiments were conducted in two commercial citrus orchards at the University of Ghana Agricultural Research Station, Kade between August 1998 and May 1999. This period falls within the peak season of many of the citrus varieties at the Station. One of the orchards is planted solely to Late Valencia (*Citrus sinensis(L)* Osbeck) and the other to Satsuma (*Citrus unshiu* Marc). The Satsuma orchard (Ci 10) of about 2.32 hectares was

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planted some 30 years ago. It is bounded on the east by a mixed variety citrus plot, on the south by an undisturbed forest, on the west by a kola plot (about 50 m away) and on the north by an almost open field with few scattered Washington navel trees.

The Late Valencia orchard (Ci 9) about 2.19 hectares was also planted some 20 years ago. It is bounded on the north by a mixed variety mango plot, on the south by an open field, on the west by one-year fallow field and on the east by a mixed variety citrus plots (both early and late season crops).

Each of the fields was laid under a randomised complete block design (RCBD) with three replications. A plot consisted of 3 x 3 citrus trees with a row of two buffer trees separating experimental plots and replications. Six trees in each plot were selected and tagged for subsequent data collection. Average tree height was about 6 m and 6 m apart. Weed management programmes adopted by ARS, Kade i.e. monthly weeding during the rainy seasons and bimonthly weeding during the dry seasons was followed.

5.2.3 Experimental treatments

Five treatments including two untreated checks were investigated. They were;

i) Control (no picking of dropped fruits, no insecticide application) (CNP)

ii) Control (picking of dropped fruits, no insecticide application) (CP)

iii) Application of Dimethoate 40 EC at a rate of 1.5 l/ha and picking of dropped fruits (Dim+P)

iv) Instalment of trimedlure (TML) baited traps and picking of dropped fruits (TML+P)

v) Application of neem seed water extract at 25 kg/ha and picking of dropped fruits (NSWE+P).

Treatments were randomly assigned. Insecticide treatments were applied by means of motorised knapsack Urgent (GmbH) and 'Solo' (GmbH) misthlowers at a rate of 25 kg/ha for neem seed water extract and 1.5 l/ha for Dimethoate 40 EC (Sumitomo Chemical Co. Ltd). Insecticide treatments in the Satsuma plot commenced on 5th August 1998 and subsequently on 2nd and 30th September 1998.

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In the Late Valencia, insecticide treatment began on the 4th December 1998 and subsequently on 12th January and 23rd February 1999.

5.2.4. Sampling method

Before each treatment five traps each baited separately with trimedlure (female sex pheromone) and methyl eugenol (female sex pheromone) were installed along the periphery of each field to detect the presence and the possible movement of *C. capitata* from adjacent fields. The trimedlure treatment consisted of three traps (Plate 3) placed at three randomly selected points on the field.

A modified Steiner's trap was used for this experiment. These traps could stay in the field without being removed throughout the experiment (Plate 3). Different trap designs were used because of the fact that the traps used for the seasonal and the diurnal activity pattern work shown in plate 1 required some labour to monitor them as long as they remained in the field. It was also possible to catch the adult fly unlike traps used in the seasonal abundance experiment.

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Plate 3 : A modified Steiner's trap with trimedlure and some captured *C. capitata*

5.2.5 Monitoring of *C. capitata* population in the two citrus orchards

A pre-treatment assessment of the Mediterranean fruit fly was carried out every two days before each spraying in both the Satsuma and Late Valencia orchards to determine the presence /absence of *C. capitata* in the field. The aim was to determine the population levels of the fly before and after each spraying time. An average of not less than four minutes was spent on each tagged tree to take all insect count data.

The overall location of the flies within the tree canopy and the vegetation under the trees were also investigated to establish the distribution of the flies within the citrus plant. The position of the flies sighted on each of the tagged plants was recorded. The locations of the fly were categorised as those on fruits, those on branches and those on the weeds growing under the citrus trees.

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Treatment effects was assessed by recording the number of the flies on tagged trees in each plot 2, 7 and 14 days after each spraying and thus the percent adult population reduction compared with untreated plots. The percent adult population reduction was calculated using a modified Abbots' formular (Fuller *et al.*, 1991) given by

$$\% \text{ Population reduction} = \frac{\text{No. recorded on control} - \text{No. recorded on treatment}}{\text{No. recorded on control plots}} \times 100)$$

5.2.6 Damage and yield assessment

Damaged fruits were collected every two weeks until final harvest. During each time damaged fruits were grouped according to those due to *C. capitata* and other factors such as physiological and rainstorms. During the final harvest all the punctured fruits from the tagged plants of the same plot were grouped and a sample of thirty fruits were selected at random to determine the number of punctures per fruit. The fruits were dissected using a knife and with the aid of a magnifying glass the number of all developing *C. capitata* larvae were counted and recorded.

The percent cumulative damage was calculated from the following formula;

$$\% \text{ Cumulative Damage} = \frac{\text{Total no.of fruits damaged by } C. \text{ capitata or others}}{\text{Potential yields per tree (Total fruits set by each plant).}} \times 100$$

The potential yield per tree was obtained by adding all the harvested fruits, the total number of fruits damaged by *C. capitata* and those damaged by other factors per each tagged tree throughout the period for each citrus variety. The percentage of damage due to *C. capitata*, other factors and marketable yield were determine by dividing each of the total number of fruits damaged for each factor by the potential yield per plot.

Air temperatures within the field were recorded at 0900 and 1500 hours GMT every other day using three laboratory thermometers. These thermometers were installed within the canopy of three randomly selected trees within the field.

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5.2.7 Cost- benefit analysis of control strategies

The experiment in the two orchards was valued to determine the cost-benefit ratio of each control strategy. At the end of each experiment i.e.3rd November 1998 for Satsuma and 30th March 1999 for Late Valencia, fruits were harvested and yield obtained per plot was determined.

The values of yield loss and marketable fruits were determined at the wholesale market price prevailing at ARS, Kade at the time of final harvesting for each variety. At ARS, Kade yield of citrus is determined in terms of numbers that is 10,000 fruits representing one box is sold at the prevailing market price.

The cost of control was determined by adding all the cost components which include the cost of labour for spraying, the cost of insecticides including the neem seeds, the cost of fuel and engine oil and the cost of picking dropped fruits. Other cost items determined included depreciation on the spraying machines calculated from the straight-line method (Johnson, 1990) and interest on the cost of spraying machine assuming the spraying equipment used were hired (Urgent' GmbH" and Solo 'GmbH' mistblowers).

The cost–benefit ratio of each treatment was determined by converting the total number of fruits damaged by *C. capitata* and marketable yields in each treatment per hectare in monetary terms. The cost of damage in each treatment was subtracted from the total revenue generated from each treatment to obtain the net revenue. The percent yield gain over control for each treatment was obtained by subtracting the net revenue obtained from each treatment from the control (no picking, no insecticide applied) (CNP) and then dividing the value by the control (no picking, no insecticide application)

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5.2.8 Alternative host plants

To identify some alternative hosts of *C. capitata* in Ghana and particularly at ARS, Kade, collections of some fruit crops reported to be infested by *C. capitata* were made. Ten fruits each were placed in five insect cages containing some sand to serve as pupation medium for the developing larvae. The cages were placed under shade to ensure that conditions are quite favourable for adults to emerge if they occur in those fruits. Each insect cage measured about 30 cm x 30 cm x 30 cm.

Monitoring was done to determine whether any adult fruit flies would emerge. The fruits collected included, mangoes (*Mangifera indica* L.), cashew (*Anacardium occidentalis* L.), papaya (*Carica papaya* L.), avocado (*Persea americana* Mill) and guava (*Psidium* spp). Coffee (*Coffea arabica* L. *and Coffea robusta* Linden). were also included. The coffee was collected from an abandoned coffee plot at ARS, Kade. These crops were chosen because they have all been reported to be the preferred host for oviposition by *C. capitata* (Waikwa, 1979; Liquido *et. al.*, 1994). Mangoes (*Mangifera indica* L.) were collected from three different mango plots located at ARS, Kade. The mango varieties collected were Jaffna, Kent and Haden. The papaya (*Carica papaya* L.), guava and cashew (*Anarcardium occidentalis* L.) were also collected from individual plants within the vicinity of ARS, Kade.

5.2.9 Effect of treatment on other arthropods

The short-term benefits from the use of insecticides are immense, with reduction in the losses from field and orchard crops. As a result of these benefits, insecticides have proved popular as a means of pest control. But there are also indirect costs associated with their use. These effects include influence of the insecticides on non-targeted arthropods etc. As a result it was decided to determine the effects of the neem seed water extracts and Dimethoate 40 EC on the insect spectrum found under citrus plantation ecosystem. Four 3m by 3m jute sacks were spread under each tagged

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tree sprayed with neem seed water extract and Dimethoate 40 EC. The sheets were held close against the trunk and were intended to collect all arthropods that dropped from the treated trees 2 and 7 days after treatment. All insects collected were later identified where possible.

5.2.10 Statistical analysis

Analysis of variance (ANOVA) (PROC GLM SAS, 1994) was used to compare all the data collected. Where appropriate, data were subjected to square root transformation to ensure that they conform to the uniformity of ANOVA. Means with significant ANOVA were separated with the LSD test, (SAS Institute, 1994). Correlation coefficients were determined for the mean number of punctures per fruit and the mean number of developing *C. capitata* larvae in treated and untreated fruits

5.3 RESULTS

5.3.1 Calibration of spray equipment

The results obtained from the experiment indicated that there was no significant difference in all the time taken for the NSWE and the water to be discharged completely from the tank of each machine separately (Table 10). On the average it took about 5 and 12 minutes for both the water and NSWE to be discharged when the cut off point of the 'Solo' was fully opened and half-opened, respectively. This indicates that a significant difference in the position of the cut-off for the 'Urgent' ($F = 9.41$, $df = 7,16$ $P=0.001$) (Appendix 13).

The results suggest there was no significant difference in the flow rate using the NSWE as spray liquid. This contrasts with the findings of (Adu-Acheampong, 1997) who observed that neem seed extract significantly caused erratic flow when the restrictor of the Urgent mistblower was set at 2. The difference may be due to how well the neem seeds were ground which can affect the viscosity of the NSWE.

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Table 10: Mean time taken to discharge spray liquid from tank equipment

Equipment		cut off point/ restrictor selection	mean time (min) ± s.e.m
	Water	fully opened	5.00 ± 0.00 [d]
		½ opened	12.00 ± 0.00 [b]
SOLO	NSWE	fully opened	4.97 ± 0.06 [d]
		½ opened	12.00 ± 0.12 [b]
	Water	restrictor 2	16.00 ± 0.00 [a]
		restrictor 3	9.51 ± 0.01 [c]
URGENT	NSWE	restrictor 2	16.07 ± 0.12 [a]
		restrictor 3	9.50 ± 0.01 [c]

Means followed by the same letters in the same column are not significantly different from each other using LSD at P=0.05.

5.3.2 Effect of treatments on field population of *C. capitata* in Satsuma orchard

Table 11 shows the effect of the treatments on the field population of adult fruit flies during each time or phase of spraying. The results indicated that the population of *C. capitata* at the beginning of the data collection in the Satsuma orchard was low but increased progressively with time (Table 11) (Fig 2).

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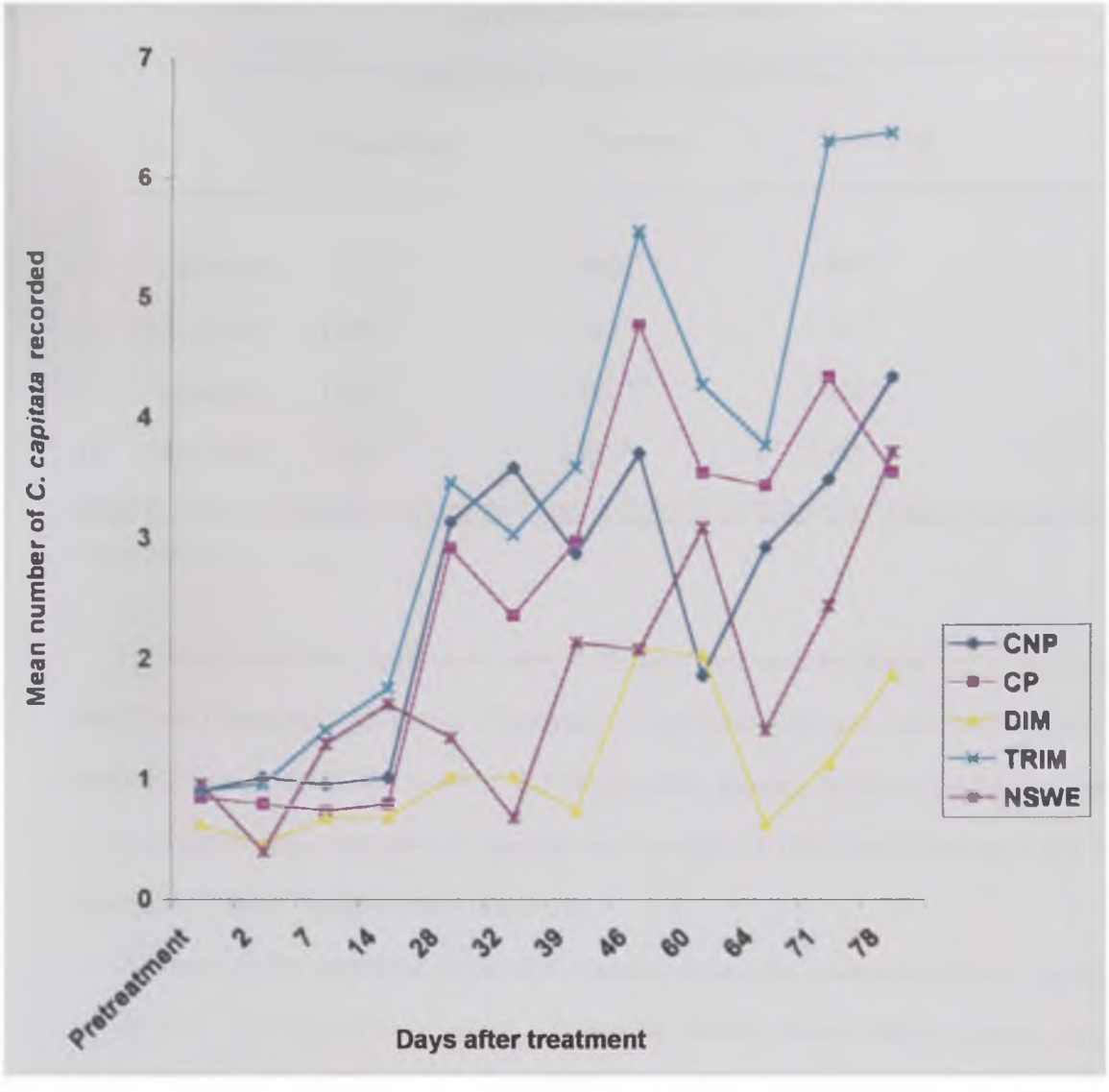

Fig. 2 Population fluctuation of *C.capitata* recorded before and after treatment of Satsuma citrus trees

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Table 11: Effect of treatments on the population of *C capitata* before and after each time of spraying in Satsuma orchard

		Mean no. of *C. capitata* recorded Ps tree		
		1st spraying	2nd spraying	3rd spraying
2	days before	1.173 f	1.662 de	1.840 bc
2	days after	1.085 f	1.514 e	1.711 dc
7	days after	1.209 f	1.673 cde	1.918 ab
14	days after	1.261 f	1.987 ab	2.025 a

Means followed by same letter (s) taken all the columns together are not significantly different from each other using LSD at P=0.05

The results show that significantly more *C. capitata* were recorded during the 3rd spraying than the 2nd and 1st spraying. During the 1st spraying no significant difference were observed 2 days pre-treatment insect count (DBS) and 2, 7 and 14 days after spraying (DAS) in the Satsuma orchard. However significantly more adult *C. capitata* were recorded 14 DAS than 2DBS and 2 and 7 DAS during the 2nd and 3rd spraying times (Table 11)

Difference in the population levels of *C. capitata* among the treatments differed significantly (Table 12). The trimedlure + picking of dropped infested fruits (TML+P) treated plots had significantly more *C. capitata* than all the treatments (15.16% over (CNP).

Dimethoate 40 EC and picking of dropped infested fruits (Dim+P) and NSWE and picking of dropped infested fruits (NSWE+P) treated plots recorded significantly the least number of *C. capitata* than the controls showing population reduction of 27.17% and 9.66%, respectively. Dim+P was, however, significantly better than NSWE+P (Table 12).

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Table 12: Effect of treatments on the population of *C. capitata* in Satsuma citrus orchard

Treatment	Mean number of *C. capitata* recorded	% reduction
CNP	1.656 [b]	---
CP	1.696 [b]	(2.42)
Dimethoate	1.206 [d]	27.17
Trimedlure	1.907 [a]	(15.16)
NSWE	1.496 [c]	9.66

Means followed by the same letter in the same column are not significantly different from each other using LSD P= 0.05. Number in parentheses shows increase in population.

5.3.3. Effect of treatments on damage and yield of Satsuma citrus fruits

Yields from treated and untreated Satsuma trees were significantly different at the end of harvest (Table 13). The analysis of variance of the percentage cumulative damage due to *C. capitata* among the treatments during some of the periods of damage assessment (Appendices 15-22). The cumulative percentage damage showed that significantly more damage were recorded on the trimedlure and picking of dropped infested fruits (TML+P) treated plots than all the other treatments (Table 13). Damage due to *C. capitata* to Satsuma fruits was 45.65% in the TML+P) treated plots. The CNP recorded the second highest percentage damage (Table 13). The cumulative damage showed a progressive increase in damage with time in the Satsuma orchard (Fig. 3). Damage in the TML+P and CNP plots was over two times higher than that in the Dim+P treated plots.

Other damage such as physiological stress and windstorm also contributed between 12-13% fruit drop (Table 13).

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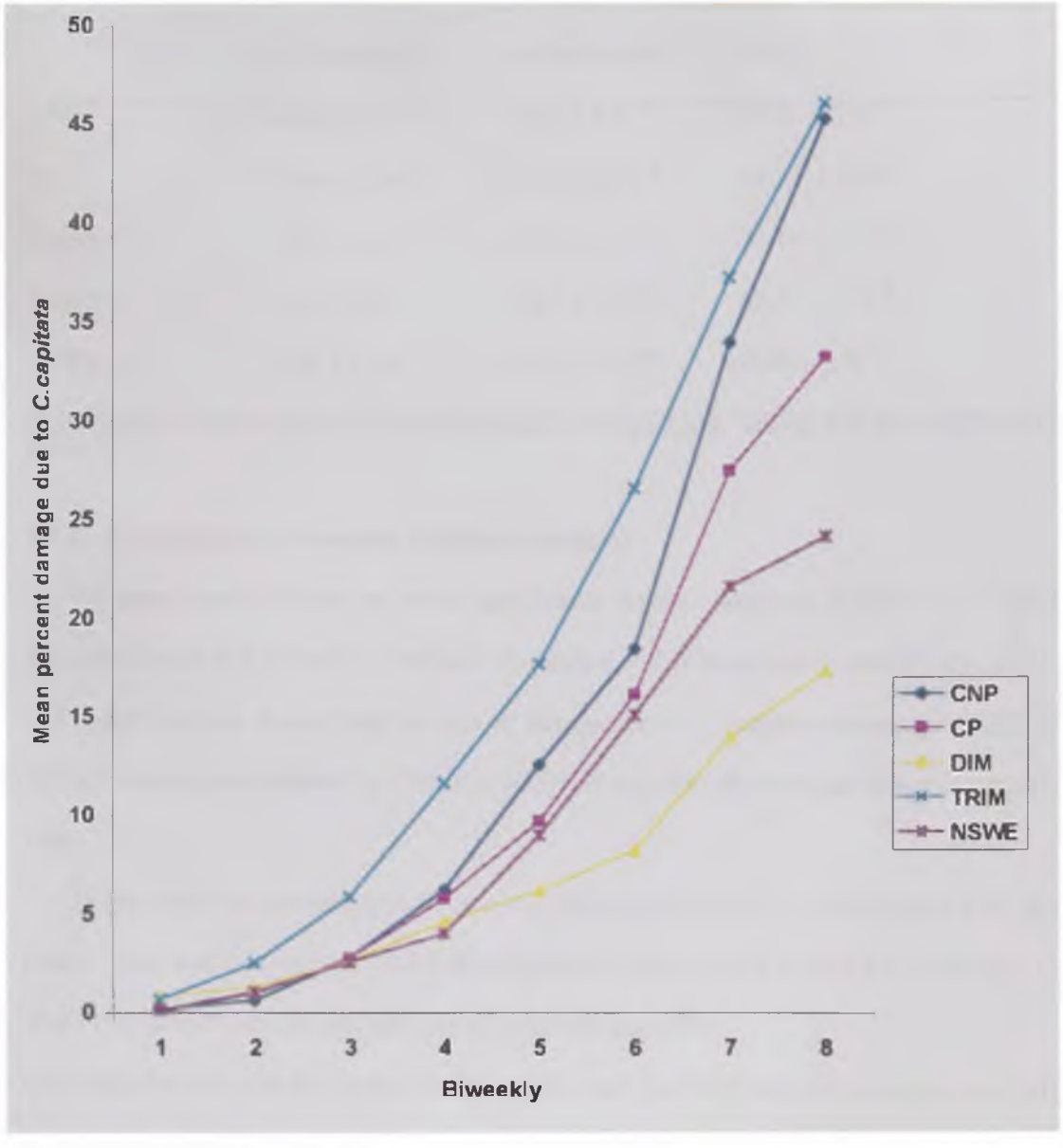

Fig. 3 Percent cumulative damage due to *C. capitata* in Satsuma citrus orchard

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Table 13: Effect of treatments on the damage and yield of Satsuma fruits

Treatment	% damage due to *C. capitata*	% damage due to other factors	% marketable yield
CNP	44.88 ± 1.97 [a]	12.03 ± 3.03 [a]	43.09 ± 5.97 [d]
CP	32.94 ± 1.29 [b]	12.82 ± 2.92 [a]	54.24 ± 3.68 [c]
Dim+P	17.13 ± 2.46 [d]	12.38 ± 0.56 [a]	70.49 ± 2.39 [a]
Trim+P	45.65 ± 2.38 [a]	12.37 ± 1.32 [a]	43.35 ± 3.39 [d]
NSWE+P	23.96 ± 1.94 [c]	13.18 ± 1.17 [a]	62.86 ± 1.91 [b]

Means followed by the same letters in the same column are not significantly different from each other using LSD at P=0.05

5.3.4. Cost-benefit of treatments in Satsuma orchard

The mean plant yield per ha varied significantly among treatments (Table 14a). The high damage recorded in the TML+P, CNP and CP resulted in significant loss in yield (Table 14a). The cost-benefit analysis showed that the cost of damage due to *C. capitata* damage was high in the TML+P treated plots followed by CNP, CP, NSWE+P and Dim+P treated plots, respectively (Table 14a).

On the whole the percentage yield gain was highest for Dim+P i.e. 60.69% over CNP treated plants. This was followed by NSWE+P recording 47.98% and CP recorded 11.25% over CNP. TML+P, which consistently had high insect population than CNP,

had a negative gain over the control (-1.06). This means that CNP even did better than the TML+P treated plants.

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Table 14a: Cost -benefit analysis of various treatments to control *C. capitata* in the Satsuma orchard

Treatment	Mean % damage at harvest	loss (¢000) per ha	cost of (¢000) control / ha (a)
CNP	44.88 ± 1.97 [a]	3400.0	---
CP	32.94 ± 1.29 [b]	2271.2	108.86
Dimethoate	17.13 ± 2.46 [d]	1414.4	752.72
Trimedlure	45.65 ± 2.38 [a]	3780.8	360.86
NSWE	23.96 ± 1.94 [c]	1972.0	582.62

Table 14b: Cost-benefit analysis of treatments for Satsuma orchard:

	Total rev (¢000) per ha (b)	net revenue per ha (¢000) (b-a)	% yield gain over control
CNP	3264.00	3264.00	-
CP	3740.00	3631.14	11.25
Dim.	5997.60	5244.88	60.69
Trim.	3590.40	3229.54	(1.06)
NSWE	5412.80	4830.18	47.98

10000 fruits = 500,000 cedis as at harvest. cost of picking / tree = 50 cedis $ 1= 2470 cedis ; Cost of labour to spray/acre =15000 cedis; cost of dimethoate / litre= 35,000 cedis: 1 ha = 272 trees. Number in bracket shows percentage loss over the control

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5.3.5 Effect of treatments on field population of *C. capitata* in Late Valencia orchard

Table 15 shows the effect of the treatments on the field population of *C. capitata* in the Late Valencia citrus orchard. The results showed similar population trends as recorded in the Satsuma orchard (Fig 4). Like the Satsuma orchard, the population of *C. capitata* at the beginning of the experiment was low in almost all the plots. However, significantly higher numbers of *C. capitata* were recorded 2 days before spraying (DBS) and 14 days after spraying (DAS) during the 1st spraying and the 2nd spraying (Table 15). This suggests that the flies reinfested the treated Late Valencia plots 14 days after the 1st spraying till the 2nd spraying was made before the population reduced again. However, the reduction of the population was not significantly low compared with the initial population.

This means that the adult fly population increased with time as was observed in the Satsuma citrus orchrd (Table 15).

Table 15: Effect of treatments on the population of *C. capitata* recorded before and after spraying in Late Valencia orchard

		Mean No. Of *C. capitata* recorded during		
		1st spraying	2nd spraying	3rd spraying
2	days before	1.239 ^{e}	1.557^{bc}	1.669^{ab}
2	days after	0.968 ^{g}	1.376^{d}	1.464^{cd}
7	days after	1.058 ^{fg}	1.466^{cd}	1.565^{bc}
14	days after	1.158 ^{e}	1.709^{a}	1.721^{a}

Means followed same letters taken all the columns together are not significantly different from each other using LSD at P=0.05

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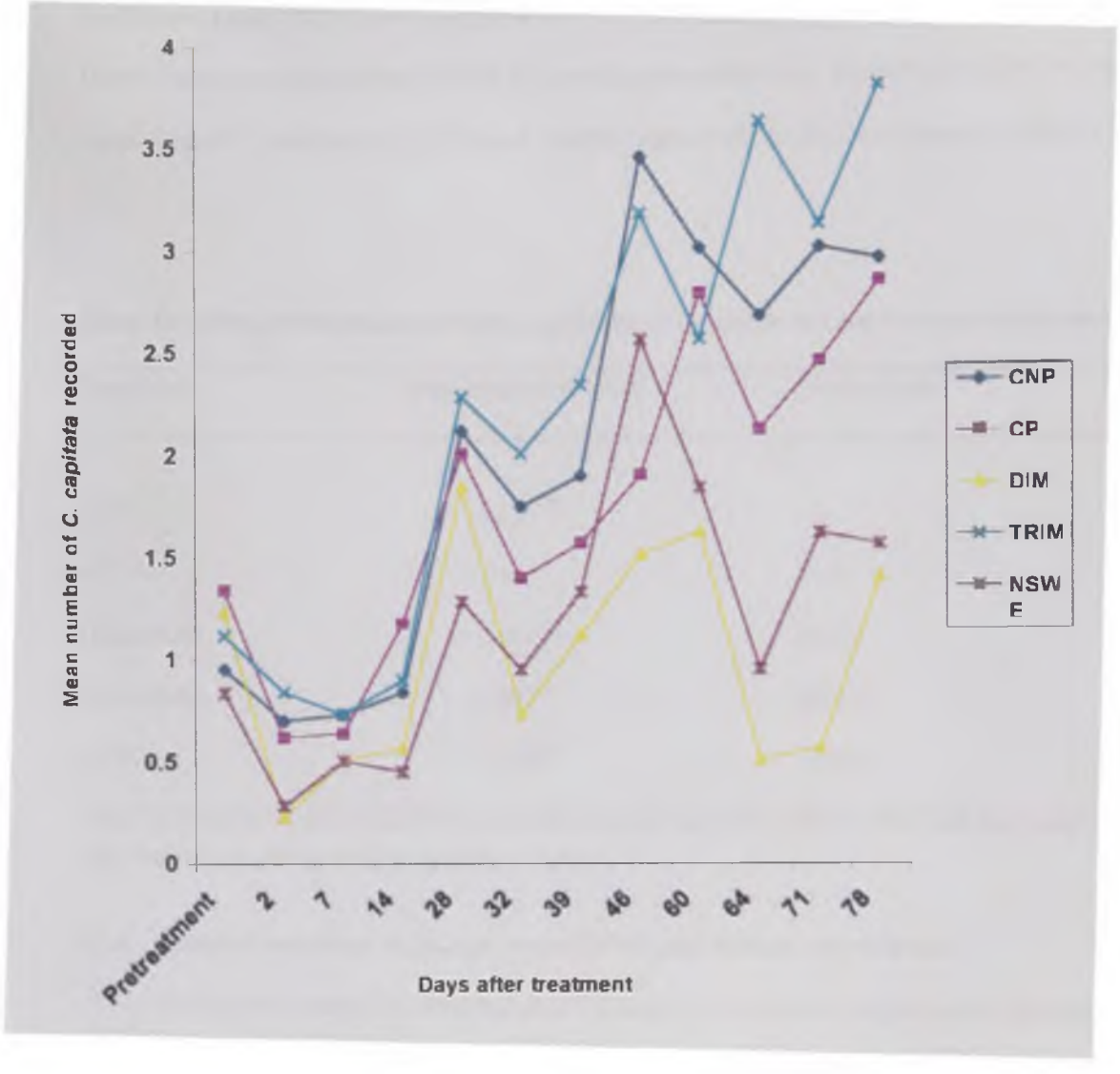

Fig. 4 Population fluctuation of *C. capitata* recorded before and after treatment of Late Valencia citrus trees

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The main reason attributed to this is that by the time the 3rd spraying was made most of the citrus had begun to attain the orange yellow colour, which attracted more insects to the field. Significantly more adult *C. capitata* were recorded in TML+P treated plots than all the other treatments (Table 16). Dim+P and NSWE+P recorded the least numbers of *C. capitata* however, Dim+P was more effective than NSWE in controlling the adult flies. Dim+P and NSWE+P reduced population of *C. capitata* by 24.10% and 14.45% respectively in the Late Valencia orchard (Table 16).

Table 16: Effect of treatments on field population of *C capitata* in Late Valencia citrus orchard

Treatment	Mean number recorded	% reduction
CNP	1.531 [ab]	----
CP	1.467 [b]	4.18
Dimethoate	1.162 [b]	24.10
Trimedlure	1.907 [a]	(24.56)
NSWE	1.310 [b]	14.45

Means followed by the same letter in the same column are not significantly different from each other using LSD P= 0.05. Number(s) in parenthesis shows population increase

5.3.6 Effect of treatment on damage and yield of Late Valencia citrus fruits

Yields from treated and untreated Late Valencia trees were also significantly different at the end of the harvest. (Table 17). The analysis of percentage cumulative damage showed significant differences in the damage due to *C. capitata* among the treatments during some of the periods of fruits damage assessment (Fig. 5) (Appendices 26-37).

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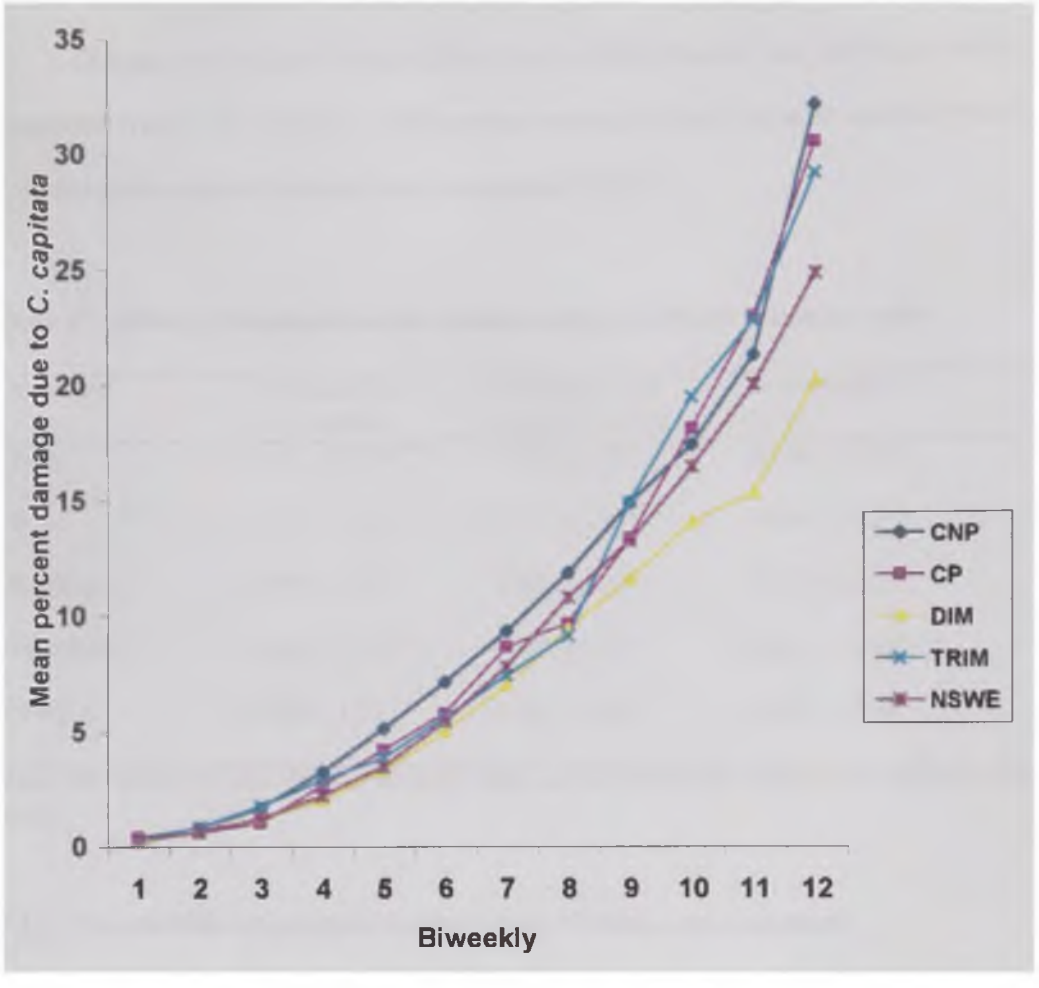

Fig. 5 Percent cumulative damage due to *C. capitata* in Late Valencia citrus orchard

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Damage due to *C. capitata* to Late Valencia fruits was highest i.e. 31.82% in the CNP plots. This was followed by CP, TML+P, NSWE+P and Dim+P recording 30.20%, 28.89%, 24.64% and 29.99%; respectively. NSWE+P was significantly better than CNP, CP and TML+P. However, Dim+P was significantly better than all the treatments (Table 17).

Damage to the Late Valencia fruits due to other factors was not significant among the treatments (Table 17). Percentage yield per tree was significantly higher in the Dim+P and NSWE + P treated plots compared with the other treatments (Table17).

Table 17: Effects of treatment on the damage and yield of Late Valencia fruits

Treatment	% damage due to *C capitata*	% damage due to other factors	% marketable yield
CNP	31.82 ± 2.39 [ab]	5.78 ± 0.72 [a]	62.40 ± 2.26 [c]
CP	30.20 ± 3.51 [a]	5.75 ± 2.32 [a]	64.05 ± 3.70 [c]
Dimethoate	19.99 ± 1.87 [d]	6.49 ± 0.43 [a]	73.52 ± 0.32 [a]
Trimedlure	28.89 ± 1.34 [b]	6.86 ± 1.11 [a]	64.25 ± 1.56 [c]
NSWE	24.64 ± 1.21 [c]	6.58 ± 1.02 [a]	68.78 ± 2.74 [b]

Means followed by the same letters in the same column are not significantly different from each other using LSD at P=0.05

5.3.7 Cost-benefit of control strategies in Late Valencia citrus orchard

The results indicate that highest yields were obtained from plots treated with Dim+P which resulted in higher economic returns showing percentage yield increase of 12.07 over CNP. The cost-benefit analysis showed that CNP was economically better than the CP, TML and even NSWE+P (Table 18b). Even though the damage caused to NSWE+P treated Late Valencia fruits was significantly lower than the control (Table18a), the cost-benefit showed significantly less revenue. The reason may be probably be due to the high cost of the neem seeds which resulted in high cost of

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spraying the NSWE, hence when the cost of application was deducted from the revenue generated its effect on the total revenue per hectare was significant (Table 18b).

Table 18a: Cost -benefit analysis of various treatments for Late Valencia orchard

Treatment	Mean % damage Per tree at harvest	loss (¢000) at harvest/ha	cost of control per ha (¢000) (a)
CNP	31.82 ± 2.39 [ab]	2856.0	—
CP	30.20 ± 3.51 [a]	2502.4	163.2
Dimethoate	19.99 ± 1.87 [d]	190.4	735.0
Trimedlure	28.89 ± 1.34 [a]	2148.8	411.0
NSWE	24.64 ± 1.21 [c]	2158.0	618.6

Table 18 b:Cost- benefit analysis of treatments for Late Valencia orchard:

	Total rev(¢000). Per ha (b)	net (¢000) revenue/ha (b-a)	% yield gain over control
CNP	5589.6	5589.6	--
CP	5304.0	5140.8	(0.08)
Dim+P	6999.1	6264.1	12.07
TML+P	5225.3	4814.3	(0.14)
NSWE+P	6024.8	5406.2	(0.033)

10000 fruits = 500,000 cedis as at harvesting: cost of picking / tree = 50 cedis $ 1= 2500 cedis ; Cost of labour to spray/acre =15000 cedis; cost of dimethoate / litre= 35,000 cedis Means followed by same letters are not significantly different from each other using LSD at P=0.05. Numbers in parentheses shows percent decrease over control

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Plate 4: Adult *C. capitata* attacking citrus fruits

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Table 19: Distribution of *C. capitata* in Satsuma and Late Valencia citrus orchards;

Within plant distribution	Satsuma plants (% adults)	LateValencia plants (%adults)
Fruits (both ripened and unripened)	94.50	89.00
Weeds under plantation	3.50	5.00
Leaves and branches	2.00	6.00

n=120

5.3.9 Effect of treatments on the number of oviposition punctures and developing *C. capitata* larvae in citrus fruits

Table 20 shows the effect of the different treatments on the number of oviposition punctures formed by *C. capitata* and the number of developing *C. capitata* larvae which developed in the citrus fruits (Table 20). Except TML+P treated fruits which had significantly more larvae developing in the fruits, the remaining treatments had significantly the same number of developing larvae in the fruits in Satsuma variety (Table 20). There was evidence of Dimethoate 40 EC showing some systemic action against the developing larvae inside citrus fruits EC (Table 20).

Correlation coefficient between the number of punctures formed and developing *C. capitata* larvae per fruit was significant (r = 0.681*) for Late Valencia treated fruits but was not significant (r = 0.433 NS) for Satsuma treated fruits.

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Table 20: Effect of treatments on the number of punctures and developing *C capitata* larvae in Satsuma and Late Valencia citrus fruits

Variety	Treatment	Mean no. of punctures/fruit	Mean number of developing *C capitata* larvae / fruit
Satsuma	CNP	6.45 ± 1.58[a]	10.27 ± 2.09[b]
	CP	6.72 ± 1.89[a]	9.56 ± 2.56[b]
	Dimethoate	3.55 ± 0.57[b]	6.91 ± 2.70[b]
	Trimedlure	5.39 ± 1.11[ab]	15.41 ± 2.67[a]
	NSWE	4.18 ± 2.66[ab]	7.96 ± 1.55[b]
Late Valencia	CNP	3.59 ± 0.59[ab]	8.85 ± 2.12[b]
	CP	3.62 ± 0.43[ab]	6.66 ± 1.92[b]
	Dimethoate	2.47 ± 0.57[b]	6.55 ± 3.28[b]
	Trimedlure	3.85 ± 0.24[ab]	8.73 ± 0.38[b]
	NSWE	2.57 ± 0.26[b]	6.91 ± 1.77[b]

Means followed by the same letters in the same column are not significantly different using LSD at P= 0.05

5.3.10 Alternative host plants of *C. capitata*

Results from the identification of alternative host plants of *C. capitata* indicated that no alternative host of *C. capitata* exists at ARS, Kade based on the fruits collected.

No fruit fly adult emerged from any of the fruits collected for this work, a few larvae were however, observed in dropped guava fruits collected under three guava plants at ARS, Kade, but none of these could develop to adult stage.

Further investigation is required to confirm this observation since all the fruits which were collected have been reported to be highly susceptible to *C. capitata* (Hagen et al., 1981; Liquido *et. al.*, 1989).

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5.3.11 Effect of treatment on other arthropods

Table 21 and 22 show the number of other unintended arthropods found dead after treatment.

Table 21: Number of other arthropods recorded 2 and 7 days after spraying the Satsuma citrus orchard

Species	Family	Order	Dim.	NSWE
Dysdercus spp	Pyrrhocoreidae	Heteroptera	34(0)	0(10)
Acanthomia spp	Coreidae	Heteroptera	6(23)	8(11)
Unidentified	Lygaeidae	Heteroptera	5(4)	0(11)
Unidentified	Reduvidae	Heteroptera	9(6)	2(3)
Oecophylla spp	Formicidae	Hymenoptera	**(*)	23(6)
Tetramorium spp	Formicidae	Hymenoptera	**(*)	6(6)
Apis mellifera	Vespidae	Hymenoptera	27(25)	0(2)
Crematogaster spp	Formicidae	Hymenoptera	8(4)	0(4)
Unidentified	Apidae	Hymenoptera	15(2)	2(5)
Unidentified	Chrysomelidae	Coleoptera	25(9)	12(5)
Unidentified	Carabidae	Coleoptera	8(6)	0(2)
Unidentified	Pentatomidae	Heteroptera	12(8)	6(0)
Unidentified	Coccinellidae	Coleoptera	24(6)	3(2)
Unidentified	Spiders	----	42(13)	6(2)
Musca domestica	Muscidae	Diptera	19(12)	2(10)
Drosophila spp	Drosophilidae	Diptera	**(12)	11(6)
Larvae		Lepidoptera	75(13)	39(59)
Achae spp	Noctuidae	Lepidoptera	19(7)	8(12)
Papillio spp	Papillionidae	Lepidoptera	5(2)	0(2)
Zonocerus sp	Pyrgomorphidae	Orthoptera	22(6)	2(13)
Unidentified	termites	Isoptera	48(3)	0(3)
Brachytrupes sp	Gryllidae	Orthoptera	14(1)	0(0)

Numbers in parentheses are insects collected 7 days after spraying. *** represents many insects which could not be easily counted.

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Table 22: Number of other arthropods recorded 2 and 7 days after spraying the Late Valencia citrus orchard

Species	Family	Order	Dim.	NSWE
Dysdercus spp	Pyrrhocoreidae	Heteroptera	21(5)	2(8)
Unidentified	Lygaeidae	Heteroptera	9(5)	2(0)
Unidentified	Pentatomidae	Heteroptera	15(8)	8(11)
Unidentified	Reduvidae	Heteroptera	9(6)	2(3)
Acanthomia spp	Coreidae	Heteroptera	19(8)	5(6)
Unidentified	Aphididae	Homoptera	**(*)	*(*)
Oecophylla spp	Formicidae	Hymenoptera	**(*)	23(6)
Tetramorium spp	Formicidae	Hymenoptera	**(*)	6(6)
Apis mellifera	Vespidae	Hymenoptera	27(9)	1(2)
Crematogaster spp	Formicidae	Hymenoptera	21(13)	0(1)
Unidentified	Apidae	Hymenoptera	23(7)	2(5)
Unidentified	Chrysomelidae	Coleoptera	25(9)	12(5)
Unidentified	Carabidae	Coleoptera	8(6)	0(4)
Unidentified	Coccinellidae	Coleoptera	24(6)	3(2)
Unidentified	Spiders	----	42(13)	6(2)
Musca domestica	Muscidae	Diptera	19(12)	2(10)
Drosophila spp	Drosophilidae	Diptera	**(12)	12(7)
Larvae	___	Lepidoptera	55(13)	39(28)
Achae spp	Noctuidae	Lepidoptera	19(7)	8(12)
Papillio spp	Papillionidae	Lepidoptera	33(12)	10(2)
Zonocerus sp	Pyrgomorphidae	Orthoptera	20(6)	2(13)
Unidentified	termites	Isoptera	36(9)	8(3)
Brachytrupes sp	Gryllidae	Orthoptera	21(8)	0(0)

Numbers in parentheses are insects collected 7 days after spraying. *** represents many insects which could not be easily counted.