ASSESSMENT OF SUITABILITY OF DIFFERENT POPULATIONS OF STEMBORER SPECIES FOR THE DEVELOPMENT OF *COTESIA FLAVIPES* (HYMENOPTERA: BRACONIDAE) AND THE ESTABLISHMENT OF THE LATTER IN ZIMBABWE

BY

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Assessment of suitability of different populations of stemborer species for the development of *Cotesia flavipes* Cameron (Hymenoptera: Braconidae) and the establishment of the latter in Zimbabwe.

Submitted by **KWADZANAI MUSHORE** in partial fulfillment of the requirements for the degree of **MASTER OF SCIENCE IN CROP PROTECTION**.

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ABSTRACT

Maize and sorghum are very important crops in Zimbabwe. They are, however, attacked by several pests, among them stemborers. Host suitability studies using *Cotesia flavipes* Cameron (Hymenoptera: Braconidae) against *Chilo partellus* (Swinhoe) (Lepidoptera: Pyralidae) populations from Bushu, Mamina, Musikavanhu, Muzarabani and Sanyati as well as *Busseola fusca* Fuller (Lepidoptera: Noctuidae) from Harare were conducted. The studies showed that *C. flavipes* successfully developed on all the *C. partellus* populations investigated. The highest parasitism was 77.3 %, which was recorded for Sanyati, and the lowest was 61.9 %, which was recorded for Mamina. Muzarabani had the shortest parasitoid developmental period of 17.4 days while Mamina had the longest of 18.2 days. The progeny was generally female - biased and the highest percentage was 78.1 recorded for Bushu. However, on Muzarabani *C. partellus*, adult female parasitoids comprised 13.3 % of each brood. Muzarabani *C. partellus* also recorded the lowest number of progeny (11.1 adults/brood) while the other sites recorded between 26.5 and 27.2. No development took place on *B. fusca* due to egg encapsulation. Releases of *C. flavipes* were conducted for the first time at Mamina, Coburn Estates (Chegutu). Three different release methods were used. These included the release of mated adults, release as *C. flavipes* cocoons and the release of pre - stung *C. partellus* larvae. A recovery rate of 4 % was recorded at Sanyati in the first season of release. Continued monitoring is needed to confirm establishment. Prospects for the establishment at both sites are high considering the results of host suitability studies and the presence of large numbers of the co - evolved host, *C. partellus*. The composition of stemborer species and their associated natural enemies as well as the establishment of *C. flavipes* Cameron were studied during the cropping seasons of 2003/4 and 2004/5 at various lowveld (< 600 m), middleveld (600 - 1200 m) and highveld (> 1200 m) sites. This involved sampling infested maize and sorghum plants, dissecting them and recording the stemborer species and their natural enemies. Three species were recorded, namely, *C. partellus*, *B. fusca* and *Sesamia calamistis* Hampson (Lepidoptera: Noctuidae. *Chilo partellus* dominated in the lowveld (Muzarabani and Musikavanhu) and middleveld (Bushu and Sanyati) while *B. fusca* dominated the highveld (Mamina). *Sesamia calamistis* occurred in all the agroecological zones under consideration but in very low proportions. It was also noted that *C. partellus* is colonizing the highveld as revealed by its abundance at Mamina. *Cotesia flavipes* was released for the first time in the country between July 1999 and March 2001 before recent releases which were conducted between July 2004 and April 2005. The surveys to determine the establishment of *C. flavipes* have been going on since July 1999 depending on the location. However, establishment was only confirmed at Musikavanhu, Bushu and Muzarabani where it parasitised 22.5 %, 23.2 % and 3 % of *C. partellus* larvae respectively sampled in the 2004/5 season. During the same period, the indigenous *Cotesia sesamiae* (Cameron) (Hymenoptera: Braconidae) was associated with all the three stemborer species. Other indigenous natural enemies recorded included the nematode, *Hexamermis* sp. (Mermithidae) which parasitised 0.7 % *C. partellus* larvae at Bushu and the pupal parasitoid, *Dentichasmiasis busseolae* Heinrich (Hymenoptera: Ichneumonidae) recovered at Musikavanhu and Muzarabani which parasitised 20 % of the field collected pupae from each of the two sites.

DEDICATION

To my sister, Priscilla, brother Lot and dearest friend, Blessing.

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CHAPTER 1

INTRODUCTION AND JUSTIFICATION

1.1 INTRODUCTION

The majority of people in sub - Saharan Africa have maize (*Zea mays* L.) and sorghum (*Sorghum bicolor* (L.) Moench) as their staple food (FAO and ICRISAT, 1996; Pingali, 2001). Of all the cereal crops grown in Zimbabwe, maize and sorghum are the most important (Chinwada, 2002). They constitute the staple diet for the majority of Zimbabwe. However, there are several constraints to their production. Among these are low rainfall, poor nutritional status of the soil and insect pests. Of the insect pests, lepidopteran stemborers are among the most damaging. Mohyuddin and Greathead (1970) stated that lepidopteran stemborers are generally regarded as the most serious pests of maize and sorghum in mainland Africa. The stemborer species which attack maize and sorghum in Zimbabwe are *Chilo partellus* (Swinhoe) (Lepidoptera: Crambidae = Pyralidae) the maize stalkborer, *Busseola fusca* Fuller (Lepidoptera: Noctuidae) and the pink stemborer, *Sesamia calamistis* Hampson (Lepidoptera: Noctuidae) (Sithole, 1995; Chinwada, 2002). In Zimbabwe, (Sithole, 1989) estimated yield losses in sorghum due to *C. partellus* to be between 50 and 60 %. In Mozambique, yield losses of over 50 % due to the spotted stemborer, *C. partellus* have been reported in the smallholder farming sector on maize and sorghum (Cugala, 2002). In Kenya, maize grain yield losses of up to 18 % due to *C. partellus* and *C. orichalcocilielus* (Strand) have been reported (Warui and

Kuria, 1983). Regardless of their different species, signs of damage by the stemborers are generally the same and they have similar life - cycles and they all undergo complete metamorphosis (Chinwada, 2002). Their distribution in Zimbabwe follows a definite pattern with *B. fusca* dominating the highveld (> 1200 m) and *C. partellus* dominating the lowveld (< 600 m) and middleveld (600 - 1200 m). *Sesamia calamistis* is found in all the agroecological zones but in very low proportions (Chinwada, 2002).

Farmers use various methods to control stemborers of which the most commonly employed is the use of synthetic insecticides like trichloforn (Dipterex). However, chemicals may not be available or they may be too expensive for the smallholder farmers (Van Den Berg, 1997; Chinwada, 2002). Stemborer larvae feed inside plant stems and this makes control by conventional contact pesticides and stomach poisons difficult (ICIPE, 2000). The development of insect resistance is another disadvantage of reliance on the use of chemicals (Hill, 1987). Cultural practices like burning crop residues, removal of alternative hosts, intercropping and management of sowing date have been recommended for stemborer control (Seshu Reddy, 1998; Cugala, 2002). Other control options involve crop rotation and the use of resistant crop varieties.

There are a lot of natural enemies of stemborers which can be utilised in biological control of stemborers in Africa (Polaszek, 1998). Biological control through the use of pathogens, parasitoids and entomophilous nematodes is a possible method of controlling stemborers. Kfir (1992; 1998) speculated that the low level of occurrence of *S. calamistis* is a result of natural enemies which are keeping its populations under control thereby

preventing serious outbreaks. However, Overholt (1998) doubted the ability of indigenous natural enemies to keep populations of stemborer below injurious levels. The problem of stemborer control in Africa is further compounded by the advent of *C. partellus*, which originated in the India - Pakistan region (Tams, 1932). This stemborer has spread rapidly in eastern and southern Africa and is displacing the native species thus becoming the most injurious stemborer in Africa (Kfir *et al*., 2002). In its native area, the parasitic wasp, *Cotesia flavipes* Cameron (Hymenoptera: Braconidae) keeps *C. partellus* in check. This justifies the need to introduce the exotic *C. flavipes* (Overholt, 1998). *Cotesia flavipes* was found to have successfully established and is promising to give satisfactory levels of *C. partellus* control in Mozambique (Cugala and Omwega, 2001; Cugala, 2002) in Kenya (Songa *et al*., 2001), in Tanzania (Nsami *et al*., 2001), in Uganda (Matama - Kauma *et al*., 2001) and in Zambia (Omwega, personal communication).

In Zimbabwe, *C. flavipes* have was first released in July 1999 at Irrigation Scheme Musikavanhu (Chinwada *et al.*, 2001). Releases were also conducted at ARDA Muzarabani, Bushu and Birchenough Bridge in 2000 and 2001. Up to 2001, establishment had not occurred (Chinwada *et al., 2001*).

1.2 JUSTIFICATION OF THE STUDY

The successful establishment of *C. flavipes* in coastal Kenya, Uganda, Tanzania, Mozambique, Malawi and Zambia after its introduction by the International Centre of Insect Physiology and Ecology (ICIPE) brought about the need to introduce the parasitoid

into Zimbabwean agroecological regions where its co - evolved host, *C. partellus* occurs. Its indigenous relative, *C. sesamiae,* though often recovered from all the local species of stemborers, does not seem to be very effective in suppressing the populations of stemborers (Chinwada, 2002). It was with this background of low parasitism by native parasitoid species that several releases of *C. flavipes* were made between 1999 and 2002 (Chinwada *et al*., 2001).

For the parasitoid to establish, there must be physiological compatibility between the parasitoid and its host because host unsuitability is one of the reasons for a lack of establishment (Overholt, 1998). It is against this background that host suitability studies were conducted as this helps to explain establishment or a lack of it. Host suitability tests are a prerequisite so as to avoid wasting of resources. In Zimbabwe, some host suitability studies were conducted using *B. fusca* (Harare population) and *C. partellus* (Chisumbanje population) (Chinwada, 2002). Clearly, since releases went on to be conducted in other areas for example, Muzarabani and Bushu, where the suitability of the resident stemborer populations had not been tested, follow - up studies were necessary. Such studies were essential as they may help to explain deviations from expected outcomes on establishment or lack of it. Chinwada (2002) found the Chisumbanje population of *C. partellus* to be suitable for development of *C. flavipes*. Therefore releases need to be conducted in this area since chances of successful establishment are very high.

The importance of maize and sorghum in Zimbabwe and the increase in the amount of damage caused by *C. partellus* made this study important.

1.2.1 Overall objective

The main objectives of this study were to test the suitability of local populations of *C. partellus* resident in areas where releases of *C. flavipes* have already been or are intended to be conducted and to make follow - up surveys to determine success or failure of establishment.

1.2.2 Specific objectives

The specific objectives of the study were:

1. To assess the suitability of different species and populations of cereal stemborers for the development of *C. flavipes* in Zimbabwe.
2. To determine stemborer incidence, species composition and parasitism level at different highveld, middleveld and lowveld locations targeted for *C. flavipes* release.
3. To release *C. flavipes* at locations where *C. partellus* is the predominant stemborer species and conduct follow - up surveys at both new and pre - 2004 release locations to determine the failure/success of habitat colonisation/establishment by the parasitoid.

1.2.3 Hypotheses

The hypotheses to be tested were the following:

1. Different species and populations of cereal stemborers in Zimbabwe are equally suitable for development of *C. flavipes*.
2. *Busseola fusca*, *C. partellus* and *S. calamistis* occur in equal proportions at every survey site and have similar within - plant distributions and are associated with the same parasitoid complex at each study site.
3. *Cotesia flavipes* has successfully established at pre - 2004 releases sites and is recoverable within the first season of release at 2004 release sites.

CHAPTER 2

LITERATURE REVIEW

2.1 Economic importance of cereal stemborers in Zimbabwe

Maize and sorghum are very important cereal crops in Zimbabwe. The majority of Zimbabweans rely on them as their staple food crops. The area dedicated to their production shows the importance of the crops to the country. Maize is the mostly widely grown crop in the smallholder sector and is grown for home consumption and for cash income. It constituted about 45 % of all the area under crop production for the 1985/86 to 1988/89 cropping seasons while sorghum occupied about 10 % (Anderson *et al*., 1993). Regardless of their importance, very low yields are produced and this is partly due to insect pests, poor nutritional status of the soil and low rains (Segeren *et al.,* 1995). Larvae of lepidopteran stemborers contribute significantly to the losses. To highlight the economic importance of cereal stemborers in Zimbabwe, Sithole (1989) estimated the reduction in yield of sorghum as a result of damage by *C. partellus* to range between 50 and 60 % in various parts of the country. When the effect of stemborers is combined to that of other factors which lead to production of low yields, for example low rains, the yield will be even lower.

2.2 Major cereal stemborer species in Zimbabwe and their distribution

The cereal stemborer species present in Zimbabwe belong to the Noctuidae and Pyralidae families. The species are *C. partellus*, *B. fusca* and *S. calamistis*. Most often, these borers occur as a complex of species with overlapping spatial and temporal distributions (Chabi - Olaye *et al.*, 2001).

2.2.1 *Chilo partellus*

Chilo partellus, the spotted stemborer (Plate 1), originated in the India - Pakistani region and in Africa it was first recorded in Malawi (Tams, 1932).

Plate 1. *Chilo partellus* larva.

It has spread to countries like Zimbabwe, Kenya, Tanzania, Uganda and South Africa where it is mainly found in areas with an altitude below 900 meters and with high temperatures (Sithole, 1990). In Zimbabwe *C. partellus* is the dominant stemborer species in lowveld areas below 600m. However, in eastern and southern Africa, there is evidence showing that the species is colonising high elevation areas (Kfir *et al*., 2002).

Although *C. partellus* may cause losses in maize, its preferred host is sorghum (Sithole, 1989). Female moths mate soon after emergence from the pupal stage (Plate 2) and lay most of their eggs in two to three consecutive nights. The eggs are laid in batches of 10 - 80 eggs on the underside or upper side of green leaves (Bates *et al*., 1990). The eggs have an overlapping arrangement upon each other (Plate 3).

Plate 2. *Chilo partellus* pupa.

Plate 3. *Chilo partellus* eggs.

A single female can produce a total of 200 - 600 eggs. The eggs hatch in 4 to 8 days after oviposition. The emerging larvae move to the whorls where they will start to feed. Some may move to nearby host plants (Berger, 1992). They may spin silken threads which are launched by wind into the air to infest neighbouring plants (Sithole, 1989). This is an instinctive dispersal mechanism which serves to reduce competition between larvae that hatch from the same egg batch thereby increasing the chances of survival of the larvae. Some larvae have been found along the mid - veins and on maize tassels. Older larvae tunnel into stems where they feed for a period ranging from 2 - 3 weeks before pupating (Plate 2). Adults emerge 5 - 12 days after pupation. The life cycle may be completed in 25 to 30 days. Up to six generations can occur in a single season (Chinwada, 2002).

However, there may be breaks between consecutive growing seasons, when larvae may enter into a diapause state for up to six months (Kfir, 1991).

2.2.2 *Busseola fusca*

Busseola fusca (Plate 4) is native to Africa (Kfir, 1997). It prefers maize as a host plant but it causes serious losses to grain sorghum (Skoroszewski and Van Hamburg, 1987).

Plate 4. *Busseola fusca* larva

Adult female moths lay their eggs on very young maize plants, which are between 3 and 5 weeks old (Van Rensburg *et al*., 1987). Each female lays a total of about 200 eggs, which are laid in batches of 30 - 100 eggs in leaf sheaths and on the outer ear husks. The eggs hatch about a week later. The young larvae, which emerge, move to maize whorls where they feed. Larvae can also feed on maize tassels. The larval stage undergoes six instars before pupation takes place. The larvae feed and tunnel inside the stems and prior to pupation, cut exit holes (pupation windows), through which the moths emerge. This exit is often seen covered by a thin 'membrane' of tissue (Sithole, 1989). The pupal

period lasts for 9 - 14 days. There are usually three generations per season. The third generation is the diapause generation (Smithers, 1960; Harris, 1962). *Busseola fusca* is the dominant stemborer species in areas with an altitude above 900 meters (Cugala, 2002). Chinwada and Overholt (2001) reported *B. fusca* comprising up to 98 % of the stemborers sampled at five highveld (> 1200m) sites in Zimbabwe.

2.2.3 *Sesamia calamistis*

A *S. calamistis* larva is characterised by a pinkish colour and it can have a big body which is similar in size to the other noctuid, *B. fusca* at maturity. Plate 5 shows a *S. calamistis* larva.

Plate 5. *Sesamia calamistis* larva.

Adult female *S. calamistis* can lay up to 350 eggs in batches of 10 - 40 (Ingram, 1958). The batches are arranged in two to four rows and are inserted between the lower leaf sheaths and stems (Cugala, 2002). They hatch in 5 - 7 days. Just after hatching, larvae move from the oviposition site and penetrate directly into stems where they feed (Harris,

1962). Larval feeding on the growing points results in central leaves withering and turning brown (dead hearts) (Plate 6) (Sithole, 1989).

Plate 6. A young maize plant showing a 'dead heart'

There can be entire crop destruction due to 'dead hearts' in young plants. In sorghum, 'dead heart' development can also result in increased tillering (Sithole, 1989). After passing through five to six instars in 4 - 6 weeks, most larvae (Plate 5) pupate in stem tunnels. However others pupate between the stem and leaf sheaths (Cugala, 2002; Chinwada, 2002). Loose silken threads hold pupae in position. Adults emerge between 9 and 13 days after pupation. If conditions are favourable, *S. calamistis* breeds throughout the year without diapause (Chinwada, 2002). This stemborer species is found in very small numbers at low, middle and high altitudes. In southern Benin, the generally low level of *S. calamistis* infestation has been attributed to, among other factors high egg

parasitism by *Telenomus busseolae* (Gahan) and *Telenomus isis* (Polaszek) (both Hymenoptera: Scelionidae) (Chabi - Olaye *et al*., 2001).

2.3 General stemborer damage and larval behaviour

Soon after hatching, larvae crawl over the plant and move into the funnel and feed for a few days before penetrating into the stem. This penetration leads to the formation of 'shot holes' as the plant grows and the leaves in the funnel unroll. Young larvae can also feed on lower palisade cells. This leaves a transparent upper cuticle. This is called 'window panning' (Plate 7). Some of the larvae migrate to nearby plants just after hatching. Larvae can feed on basal meristems of young maize plants resulting in the formation of 'dead hearts' (Plate 6). 'Dead hearts' cause death of plants like maize while sorghum, millets, and rice compensate by tillering (Sithole, 1989).

Plate 7. Maize leaves showing 'window paning'

Older larvae make holes (Plate 8) and tunnels in stems where they feed for 3 - 5 weeks, producing extensive tunnels (Van Rensburg *et al*., 1988).

Plate 8. A maize plant showing a hole made by a stemborer larva

Severe stem tunneling can lead to stem lodging. Other plant parts, which are prone to stemborer damage, are tassels and ears (Van Rensburg *et al*., 1988). *Busseola fusca* can also feed on maize kernels. Stemborer damage can also lead to infection by Fusarium stalk rot (Hill, 1987).

2.4 Host plants

The original host plants of all cereal stemborers are wild grasses (Cugala, 2002). Apart from maize and sorghum, most African cereal stemborers feed on other plants (Chinwada, 2002). However, maize and sorghum (Plate 9) are the most important host plants for stemborers (Harris and Nwanze, 1992).

Plate 9. Sorghum plants damaged by stemborers

Chilo partellus, *B. fusca* and *S. calamistis* have different preferences for host plants. For example, *C. partellus* prefers sorghum to maize and *B. fusca* prefers maize to sorghum (Sithole, 1989; Kfir; 1998). *Sesamia calamistis* was found to be associated with *C. partellus* in the same host plants (Cugala, 2002). *Chilo partellus* has been observed damaging pearl millet, rice, wheat and sugarcane in the field (Sithole, 1990).

Stemborer wild host plants are found in the Graminiae, Cyperaceae and Typhaceae families. Some of the gramineous species which are hosts to *C. partellus*, *B. fusca* and *S. calamistis* in Kenya are *Hyparrenia*, *Panicum*, *Pennisetum*, *Setaria*, *Sorghum* and *Sporobolus* species. (Polaszek, 1998). *Andropogon* spp. is another host for *C. partellus* (Sithole, 1990). Thirteen grass and two sedges' species were found to be hosts of *S. calamistis* in West Africa (Schulthess *et al*., 1997). These alternative hosts serve as reservoirs for stemborers (Ingram, 1958; Harris, 1962; Gebre - Amalak, 1988).

2.5 Control Methods

2.5.1 Cultural control

Cultural measures are an inherent component of the normal crop management practices (Sithole, 1990). These are the regular farm operations, that do not require the use of specialized equipment or extra skills and are designed to kill pests and diseases or to prevent them from causing economic damage (Hill, 1987). This is a prophylactic (preventative) method of control (Dent, 1991). Cultural control is very useful if it succeeds because it combines effectiveness with minimal extra labour and cost.

2.5.1.1 Host plant resistance

Breeding plants, which are resistant to stemborer attack, is a pest management tactic that is economical and requires little or no change in farmer practice (Dabrowski and

Nyangiri, 1983). The mechanisms of resistance are based on antixenosis and antibiosis. These cause stress in insects, which may make them more susceptible to insecticides. Therefore, it is useful in Integrated Pest Management (IPM). IPM is an ecologically based pest control strategy that relies heavily on resistant crops, hygiene, and natural predators and parasitoids, and it tries to disrupt these factors as little as possible by only using appropriate chemical pesticides when necessary (Dobson *et al*., 2002). With antibiosis, the plant resists damage by causing death to the pests or by reducing the rate of reproduction (Hill, 1987). Reduced preference for oviposition, reduced feeding due to the presence of some chemicals in the plant, reduced ability to be tunneled and plant's tolerance to leaf damage, dead heart and stem tunneling are some of the mechanisms of host plant resistance (Seshu - Reddy, 1998). Conventional breeding techniques can be employed in the production of resistant varieties. South African sorghum varieties like Macia, SA 3872 or SDS 2538 are resistant varieties, which reduce moth abundance by suppressing stemborer larvae (Van den Berg, 1997). Of late, the production of transgenic plants by genetic engineering has taken center stage. Examples are '*Bt*' maize hybrids, which were first introduced in the United States of America in 1996 to control the European Corn Borer, *Ostrinia nubilalis.* The *'Bt"* maize hybrids have demonstrated good control of *O. nubilalis* as well as other species like *Diatrea grandiosella* Dyar (Berginvinson *et al*., 1997; Cannon, 2000). However, since the resistance mechanism is based on a single gene, transgenic plants may not offer long - lasting control because the mechanism can easily be overcame by insect pests. Another demerit is that of the unknown effects on human health and biodiversity of beneficial insects.

2.5.1.2 Utilisation of wild gramineous plants

Sudan grass (*Sorghum vulgare sudanese*) was observed to attract maize stemborer females for oviposition resulting in significant yield increase in Kenya (Khan *et al*., 1997). Oviposition away from maize leads to reduced larval damage. Sudan grass around a maize field may also increase the efficiency of natural enemies because it enables natural enemies to colonise the crop in large numbers (Khan *et al*., 1997).

2.5.1.3 Intercropping

The practice is not suitable for large - scale production but can be employed successfully by small - scale communal farmers. The effects of intercropping are believed, among other factors to be due to increased diversity in the agroecosystem (Nwanze, 1997). Intercropping reduces pest population on a crop by reducing the visual and olfactory stimuli which attract pests onto a crop (Hill, 1987). Another effect of intercropping is oviposition on non - host crop plants of the system. Larvae emerging from eggs on non - hosts die as a result of starvation and this reduces the number of larvae migrating to host plants (Ampong - Nyarko *et al.,* 1995). However, one of the problems associated with intercropping as a stemborer control measure is predicting the cropping systems which will best reduce pest abundance. In Kenya, Amoako - Atta and Omolo (1983), showed that intercropping maize, cowpea and sorghum gives good control of *B. fusca* while Adesiyum (1983) working in Nigeria reported that an intercrop of pearl millet and sorghum in alternate stands within the same row can reduce *B. fusca* infestation.

2.5.1.4 Crop residue management

The removal of plant residues helps clean fields from pests. Plant material infested with various stages of insects must be removed before pests spread to the desired crop. Stemborers are known to overwinter in maize stalks. Many noctuids and pyralids pupate in the lower parts of cereal stems and as such will be left in the stubble even if the main parts of the stem are removed (Hill, 1987). Ploughing in of residues may not kill all insects but burning is the most effective method (Harris and Nwanze, 1992). However, the practice of completely burning maize residues may not be feasible in some parts of West Africa because the stalks are sometimes used as fencing poles and for building huts (Harris and Nwanze, 1992).

2.5.1.5 Tillage practices

A large number of larvae of many lepidopterans (especially Noctuidae, Sphingidae and Geometridae), Coleoptera and Diptera are found in the soil where they pupate. Most are found in the upper 10 cm. Deep ploughing will bring the larvae and pupae to the soil surface. They will then be exposed to heat from the sun and predators like cattle ergots (van den Berg *et al*., 1998). The relatively dry conditions on the earth's surface will desiccate the larvae and pupae. Consequently, they will die. Deep ploughing may also destroy and bury volunteer plants and other alternative hosts, which would otherwise harbour pests. Deep ploughing can also control stemborers by burying pupae and stemborer moths do not emerge from great depths (Harris and Nwanze, 1992). Leaving a

bare fallow may be effective in pests and disease control. However, this is not always possible due to land shortages. Zero tillage may provide insect pests with shelter from plant materials. This may lead to an increase in the number of pests and must not be done if stemborer numbers are to be lowered (Kfir, 1990).

2.5.1.6 Crop rotation

The alternation of crops from completely different families in a field has obvious advantages in pest control. For effective insect pest control, crops must be planted a long distance away from the previous field. Crop rotations are effective against mono and oligophagus pests (Wright, 1984). The working principle is that the following generations of a pest will find a different host from the one they prefer. This will make the pests migrate or they will starve to death. Planting the preferred host after starvation or migration of pests will minimise chances of attack by pests. However, crop rotations are not very effective against polyphagous and migratory pests (Dent, 1991). Ovipositing stemborer moths may easily be dispersed by wind and this may cancel out the benefits of crop rotations (Harris and Nwanze, 1992).

2.5.1.7 Manipulation of planting dates

By careful planning, it may be possible to have a crop pass through the vulnerable stage in the absence of damaging insect pest numbers (Hill, 1987). Warui and Kuria (1983), working in Kenya, reported a lower incidence of *C. partellus* and *C. orichalcociliellus*

on maize which was planted at the beginning of the rainy season than on that planted 3 - 8 weeks later. Similar findings were also reported in Zimbabwe (Sithole, !987) and in Ethiopia (Gebre - Amlak*,* 1989). However, Swaine (1957) and Chinwada (*et al.,* 2001) reported high infestation levels of *B. fusca* on early planted maize. It is important to note that the choice of planting dates may be influenced by other factors, for example rainfall.

2.5.1.8 Optimal growing conditions

Healthy plants have considerable tolerance to pests and diseases. Healthy plants are a result of good genetic stock and optimal growing conditions. Pest damage is more serious when a plant is suffering from water stress, unfavourable temperature, nutrients imbalance or nutrient deficiency (Hill, 1987). It is therefore imperative that optimal conditions for growing vigourous plants be provided. Supplying the correct levels of water and nutrients are practices that in general promote rapid growth and shorten the time at which the plant is very susceptible to insect pest damage (Coaker, 1987).

2.5.1.9 Removal of alternative and volunteer host plants

Most insect pests are not monophagous (Hill, 1987). They can survive on a wide range of host plants. Stemborers have alternative hosts in the Cyperaceae, Graminiae and Typhaceae families. Examples of gramineous species which are hosts to *C. partellus*, *B. fusca* and *S. calamistis* in Kenya are *Hyparrenia*, *Panicum*, *Pennisetum*, *Setaria*, *Sorghum* and *Sporobolus* species (Polaszek, 1998). *Andropogon* spp. is another host for

C. partellus (Sithole, 1990). These alternative hosts serve as reservoirs for stemborers (Ingram, 1958; Harris, 1962; Gebre - Amlak, 1988). Weeds may also provide oviposition sites for stemborers. Removal of the alternative hosts will starve and deny the insects of oviposition sites. Thus their removal from the vicinity of the crop field is a recommended stemborer cultural control practice (Lawani, 1982). However, the removal of volunteer plants is only effective against monophagous pests.

2.5.2 Chemical control

Chemicals are sometimes used to control stemborers. In general, all stemborer control strategies involving the use of chemicals target the larval stage. Dusts and sprays are applied down the funnel of young plants to kill the emerging and feeding first instar larva. Dusts of endosulfan, malathion, and carbaryl are effective (Van den Berg and Nur, 1998). Sprays of endosulfan can also be used. Deltamethrin alone or in a mixture with endosulfan gives effective control against *C. partellus* in maize and sorghum when applied 10 - 14 days after crop emergence. In commercial farming systems, foliar sprays of endosulfan, monocrotophos and pyrethroids are common for the control of *C. partellus*. Granular application of insecticides in the whorls of maize and sorghum plants is the most effective and economic way of controlling stemborer species which feed in the whorls (Van den Berg and Nur, 1998). In the smallholder sector, whorl - applied granular insecticides like trichloforn (Dipterex) are the most widely used because of their ease of application and lack of the requirement for special application equipment (Chinwada, 2002). They are easy and safe to handle, can be applied by hand, and can be

applied accurately in a controlled manner (Van den Berg and Nur, 1998). They are also retained there under conditions of rain and wind. Soil applied granular systemic pesticides such as carbofuran applied at planting have been shown to be effective in controlling stemborers (Van den Berg and Nur, 1998).

Despite their proven effectiveness, the use of chemicals has some disadvantages. Hill (1987) states a danger of the development of resistance to the pesticides. Also, chemicals may not be available or they may not be affordable to peasant farmers. Because of the low profit margin of maize and sorghum, peasant farmers cannot afford the cost of chemical control against stemborers (Kfir, 1998). Another problem is caused by the fact that stemborers are physically protected from contact pesticides when they are feeding inside stems (Overholt, 1998). Another disadvantage of the use of synthetic pesticides is that the world is making frantic efforts to reduce synthetic pesticide use, an example being the banning of endosulfan use. This makes chemical control less attractive. Also the exclusive use of pesticides by resource limited small - scale farmers is uneconomical and not practical (Seshu - Reddy, 1998). Against this background, it is necessary to find a lasting and self - sustaining way of controlling the pests. Effective and sustainable control should involve as many methods as possible in IPM systems.

2.5.3 Biological control and its agents

Biological control is the use of predators, parasitoids, nematodes and pathogens to maintain density of a species at a lower than would occur in their absence (DeBach and

Rosen, 1991). The main attraction of biological control is that it lowers the need for using chemicals and there is therefore no environmental pollution, which may affect non - targeted flora and fauna. When it succeeds, it offers a lasting solution of stemborer control from one introduction and this is very helpful to both small - holder and large - scale farmers (Wiedenmann and Smith, 1997).

For biological control to be successful, care must be taken to minimize the disturbance of the natural balance of mortality factors at play in controlling pests. This is achieved by sparingly using chemicals, and when there is real need, using selective pesticides.

In the field, all stages in the life - cycle of *C. partellus* are subject to attack by predators, parasites, viruses, fungi and bacteria (Sithole, 1990). *Trichogramma* spp.(Hymenoptera: Trichogrammatidae) and *Platytelenomous busseolae* are egg parasitoids, which contribute to natural mortality of stem borers (Van Rensburg and Drinkwater, 1987).

2.5.3. 1 Predators

Predators are valuable components of IPM. Ants (Hymenoptera: Formicidae) are the most important predators of stem borers in maize fields (Bonhoff, 2000). Ants attack all stages of stemborers, and are among the few predators preying on larvae and pupae (Bonhoff, 1998). *Componotus* spp. and *Pheidole* spp appear to be the most important and common species. Ants of the genus *Lepisiota* can prey on stemborer eggs and pupae (Bonhoff, 2000).

2.5.3.2 Pathogens

Control of insects by pathogens is also called microbial control (Hill, 1987). Entomophagous viruses, bacteria and fungi are used to control insect pests. Doom, a product based on *Bacillus* spp. was the first microbial control agent to be registered and this was in 1948 (Dent, 1991). *Bacillus thuringiensis* was reported to significantly lower the population of stemborers in Kenya with a consequent increase in the yield (Brownbridge, 1991). The interest in these bacteria as bio - control agents and their subsequent commercial success can be attributed to their non - hazardous nature as well as to the fact that they can store well and they can be applied using standard equipment (Dent, 1991). *Bacillus thuringiensis* is fast acting and it acts as a stomach poison (Dent, 1991). However, it is non - persistent (Dent, 1991). Infection of *Chilo sacchariphagus* Bojer (Lepidoptera: Pyralidae) by viruses was found to be very high in Reunion (Fournier and Etienne, 1981) and in India (Mehta and David, 1980). Thus the viruses can be used against *C. sacchariphagus*. For an entomopathogen to be useful as a microbial agent, it must possess the following characteristics: virulence, predictability of control, ease of application, ease of production, be of low cost, good storage properties, safe, aesthetically acceptable and it must be able to reduce pest populations to sub-economic levels (Dent, 1991). However, some virus - based chemicals take long to kill insects. This is not desirable.

Over 500 species of fungi are known to infect insects (Dent, 1991). The most useful entomopathogenic fungi are from the Deuteromycetes (imperfect fungi) namely

Beauvaria bassiana, Metarhizium anisopliae, Verticillium lecanii and *Hirsutella thompsonii* (Dent, 1991). *Verticillium lecanii* and *Hirsutella thompsonii* are registered and marketed as microbial pesticides in the United States of America and Europe. *Metarhizium anisopliae* has diverse insect hosts including species of Coleoptera, Lepidoptera, Orthoptera, Hemiptera and Diptera (Hall and Pierock, 1982). *Beauveria bassiana* is known to attack *C. partellus* (Sithole, 1990). Fungi infect insect hosts through skin penetration. This makes them readily able to kill piercing and sucking pests which may not be killed by stomach poisons. However, fungi require high humidity for germination. They are also difficult to produce and have a limited shelf life. Fungal pesticides may therefore not be useful for dry areas but they are useful in glasshouses where conditions are easily controlled.

2.5.3.3 Parasites

Protozoa can also be used in the control of insects. Studies have shown that *Vairimorpha necatrix* which affects Lepidoptera particularly noctuids can cause the death of the host within six days. Before death, there is reduced feeding by the stemborer. The nematode can therefore be used as a microbial pesticide (Dent, 1991). Its mode of action is septicaemia caused by bacteria that enter the insect haemocoel from the midgut through punctures caused by protozoal spores (Maddox *et al*., 1981). *Nosema marucae* was found to be as effective as synthetic insecticides like carbofuran and trichlorphan in reducing *C. partellus* in Kenya (Odindo *et al*., 1991). It is effective and non toxic to natural enemies (Odindo *et al*., 1991). However, these microbial pesticides have low pathogenicity,

causing chronic rather than acute infections, and difficulty of large - scale production makes them unattractive.

In agricultural systems, the searching ability of nematodes, although limited, makes them ideally potential candidates for use in situations where chemical and microbial formulations cannot target effectively, for instance the cryptic habitats of stem - boring insect pests (Poinar, 1983). The three families of nematodes, which kill their hosts in a relatively short time, are the Steinernematidae, the Heterorhabditidae and the Mermithidae (Dent, 1991). The first two are terrestrial nematodes that are associated with symbiotic gut bacteria that kill the pest by septicaemia. The mermithidae are aquatic nematodes that kill their hosts upon exit through the cuticle (Dent, 1991). The mermithid *Hexamermis* has been recovered from *B. fusca* (Chinwada and Overholt, 2001). However, the survival and successful use of nematodes is limited by the need for damp conditions, which may not be available always (Dent, 1991).

2.5.3.4 Parasitoids

A parasitoid is an insect whose larval stage feeds exclusively on another insect, its host, and eventually killing it (Godfray, 1994). Adults of parasitoids are free- living (Pedigo, 1989). Only a single host is required for the completion of development and often a number of parasitoids develop gregariously on the same host. All parasitoids undergo complete metamorphosis (Godfray, 1994). The adult female who lays her eggs either directly on the host or in the vicinity of the host usually locates hosts. In the latter case,

larvae then actively look for hosts. Chemical cues from the host or its microhabitat help the parasitoid to locate its host. Hymenopteran parasitoids like *Cotesia* spp. (=*Apanteles* spp.) have highly specialized ovipositors for stinging and depositing eggs in the host. The sting may cause permanent paralysis in the host (Godfray, 1994).

Some parasitoids attack eggs, some attack larva, while some attack pupae. According to Cugala (2002), *Trichogramma* spp parasitize eggs of stemborers while *Cotesia* spp. are larval parasitoids. In India, Philipines and Thailand *Trichogramma* species were used to control *Chilo* and *Diatrea* species (Li, 1994). Egg parasitism offers a good solution in that it stops the emergence of the damaging larval stage. Thus damage to the crop is avoided. *Dentichasmiasis busseolae* (Heinrich) (Hymenoptera: Ichneumonidae) (Plate 10), *Pediobus furvus* (Gahan) (Hymenoptera: Eulophidae) and *Lepidoscelio* spp. are pupal parasitoids of stemborers (Sithole, 1990).

Plate 10. *Dentichasmiasis busseolae* adult

Another pupal parasitoid of stemborers is *Xanthopimpla stemmator* (Thunberg) (Hymenoptera: Ichneumonidae) (Kfir, 1997). Pupal parasitism of up to 100 % on *B. fusca*

by *Procerochasmiasis nigromaculatus* Cameron (Hymenoptera: Ichneumonidae) was observed in South Africa (Kfir, 1997). Kfir (1997) also recorded up to 100 % pupal parasitism of *C. partellus* by *D. busseolae* in South Africa. Some parasitoids can pupate within the eaten out body of the host. Parasitoids of hosts which feed in exposed situations usually pupate in protective silken cocoons produced by the larvae themselves.

2.5.3.5 *Cotesia flavipes* biology and ecology

The most common species of *Cotesia* which are known to parasitise stemborers are *Cotesia sesamiae* Cameron (Hymenoptera: Braconidae), which is native to Africa and *C. flavipes* which is of Asian origin. *Cotesia sesamiae* is the most commonly recovered larval parasitoid of several stemborer species in maize and sorghum in many areas of sub -Saharan Africa (Overholt, 1998). These parasitoids are morphologically similar and both attack medium to large larval stages of cereal stemborers (Omwega and Overholt, 1997). Examination of male genitalia is a reliable way of distinguishing between the two species (Van Achterberg and Walker, 1998; Polaszek and Walker, 1991; Kimani and Overholt, 1997). Males of the two species are easily distinguished from females because they have longer antennae. The male genitalia of *C. flavipes* are slender and elongated while those of *C. sesamiae* are robust and short and are about half the length of *C. flavipes* male genitalia. However, it is very difficult to distinguish between the species when there are no males produced in the progeny. Rao and Nagaraja (1967) and Nagaraja (1971) stated that identification is possible using the punctuation and pubescence on the mesosoma and metasoma as well as the shape of the scutellum. They stated that *C. flavipes* has a

sparsely punctuate mesonotum and its scutellum and propodeum are narrow, while *C. sesamiae* has an enlarged propodeum and a uniformly punctuate mesonotum. Mohyuddin (1971) stated that cocoons (Plates 11 and 12) could also be used to differentiate between the two species because those of *C. flavipes* (Plate 11) are closely packed while those of *C. sesamiae* are loosely packed (Plate 12).

Plate 11. *Cotesia flavipes* cocoons

Plate 12. *Cotesia sesamiae* cocoons.

Apart from morphological structures, the parasitoids can also be distinguished from each other by using allozyme frequencies and mating experiments with laboratory populations of *C. flavipes* (Omwega *et al*., 1995).

The method of host attack used by *Cotesia* spp. is called "ingress and sting" (Smith and Wiedenmann, 1997). With this method, the mated adult female parasitoid enters the tunnels made by stemborers in plants and actively searches for and stings hosts and oviposit in the process. The success of biological control has been attributed in part to the high searching efficiency of natural enemies for their hosts (Waage, 1991). A natural enemy must be able to locate its host if it is to establish in a new environment. Unlike

other parasitoids which do not enter tunnels to look for hosts, the "ingress and sting" mechanism helps in easy colonisation of a habitat (Overholt, 1998). When stem openings are blocked by frass, the dorso - ventral flattened shape of the *Cotesia* complex helps them to squeeze through very small openings (Walker, 1994). *Cotesia flavipes* uses olfactory stimuli to locate host - infested plants (Potting, 1997). The parasitoids follow cues produced by infochemicals which are produced by the host plant after stemborer attack. These volatile chemicals are commonly termed Herbivore Induced Synomones (HIS). Examples of these are heptanal, linalool and (Z)-3-hexenyl acetate (Ngi - Song *et al*., 2000; Gohole and Ngi - Song, 2001). Host frass is another cue (Ngi - Song and Overholt, 1997). Holes on host plant stems are also used in host location (Potting *et al.*, 1997). Caterpillar regurgitate is also used in host location by *C. flavipes* (Van Leerdam *et al*., 1985). The stemborers try to defend themselves from attack by the parasitoids inside the stem tunnel. They do that by spitting against and biting the parasitoid (Takasu and Overholt, 1997; Potting *et al*., 1997).

Between 12 and 120 *Cotesia* progeny can emerge from a single larval host. Cugala (2002) recorded 30.9 and 50.5 adults of *C. sesamiae* emerging from single *C. partellus* and *S. calamistis* larva, respectively, and between 0.0 and 33.5 *C. flavipes* from *S. calamistis* and *C. partellus* larvae respectively. However, *C. flavipes* produces significantly more progeny on large - sized *C. partellus* larvae than on small and medium sized larvae (Omwega and Overholt, 1997). In laboratory experiments, they also found a larger clutch size and more female progeny from large size larvae than from medium and small sized larvae. The differences in progeny production between different host sizes

could be due to lower density dependant mortality in larger hosts especially in the early life stages (Omwega and Overholt, 1997). The emerging larvae immediately spin cocoons in which they pupate. Adults emerge after 5 - 7 days and mate immediately after emergence. High light intensity increases activity and ensures mating. The egg - larval period lasts for 12 - 15 days.

2.5.3.6 The practice of Biological Control

The practice of biological control has three major components. The components are conservation, augmentation (inundation and innoculation) and introduction (Classical Biological Control). Conservation entails the maximisation of the effectiveness of the natural enemies, which are already present in the local ecosystem for the control of pests. The practices stresses the use of pesticides sparingly and when very necessary. It also encourages the use of selective pesticides to avoid harming non - targeted beneficial insects and other natural enemies, for example arachnids (Thomas and Waage, 1996). Granular insecticides and systemic insecticides must be used in conservation practices (De Bach and Rosen, 1991). Augmentation is the deliberate release of artificially reared natural enemies into an ecosystem to increase their number for effective control of insect pests. Inoculation is a type of augmentation whereby small numbers of the natural enemy are released with no expectation of immediate but long - term control. The natural enemy is expected to reproduce over time so as to give effective control of the pests. This is in sharp contrast with inundation (flooding) which entails the release of large numbers of natural enemies with the intention of having an immediate control of the pest (De Bach

and Rosen, 1991). Classical Biological Control (CBC) is a technique of introducing native parasitoids, parasites, pathogens, or predators to foreign lands with the intention of forming an old host association with their native hosts (Greathead, 1986). The use of the gregarious braconid larval endoparasitoid, *C. flavipes* to control *C. partellus* is an example of CBC.

2.5.4 Classical Biological Control

Cotesia flavipes has been introduced into more than 40 countries of the world to control native and exotic stemborers with variable success (Polaszek and Walker, 1991). For introductions to be successful, the numbers of the host must be more than those of the biocontrol agent (Knipling, 1992). Compared with new associations, old host - parasitoid associations are responsible for a larger share of known successful establishments (Overholt, 1998).

Cotesia sesamiae has been recorded as the most common and widely distributed parasitoid of *B. fusca* larvae throughout most of Africa south of the Sahara, particularly in Eastern and Southern Africa (Overholt, 1998). Greathead (1971) reported several successful introductions of the species from the mainland to the Indian Ocean islands of Madagascar, Mauritius and Réunion against *S. calamistis*. Within Africa, *C. sesamiae* has been imported into Benin from Kenya to control *Sesamia* spp. (Hailemichael *et al.*, 1997). However, introduction of *C. flavipes* from Asia into Africa by ICIPE into the southern coastal region of Kenya in 1993 is the most notable example of the successful stemborer CBC (Omwega *et*

al., 1997). *Cotesia flavipes* has become well established on the coast and is spreading further inland and is having an impact on stemborer populations. In the coastal area of Kenya, stemborer density decreased by about 50% from the second growing season of 1997/98 to the first growing season of 1999 and this was attributed to the increase of the *C. flavipes* population (Zhou and Overholt, 2001). However, the parasitoid was found not be very effective at high altitude (Western Kenya) and it was speculated that this is largely due to differences in stemborer species composition. *Busseola fusca* which is not a suitable host is the dominant species at this level (Zhou and Oveholt, 2001). *Cotesia flavipes* has now established in Mozambique (Cugala and Omwega, 2001), Uganda (Matama - Kauma, *et al*., 2001), Malawi and Zambia (Omwega, pers. communication). In mainland Tanzania, *C. flavipes* first became established not from any local releases but apparently as a result of natural spread from areas in neighbouring Kenya where the parasitoid had earlier on been released and confirmed established (Omwega *et al*., 1995, 1997; Nsami *et al*., 2001).

In 1960 and 1961, *C. flavipes* was introduced into Madagascar and it established and reportedly provides some control of *C. sacchariphagus* in sugarcane (Betbeder - Matibet and Malinge, 1967). Both *C. partellus* and *C. flavipes* are native to Asia, hence the success of bio - control because of the old parasitoid and host association. *Cotesia flavipes* was recovered in Eastern Uganda where it caused parasitism of 4 - 32.9 % in *C. partellus* and it was also recovered from the indigenous *B. fusca* and *S. calamistis* (Matama - Kauma *et al*. 2001). This confirms establishment of the parasitoid in Eastern Uganda (Matama - Kauma *et al*., 2001). One of the factors which enables easy

colonisation of a habitat by *C. flavipe* is its high reproductive potential in relation to most stemborer species. *Cotesia flavipes* is pro - ovigenic and has about 150 eggs available for oviposition and these are distributed over 3 or 4 stemborer hosts (Potting *et al*., 1997). *Cotesia* spp. have short generational times (16 - 18 days) as compared to their stemborer hosts (30 - 50 days) and a fairly high fecundity (30 - 50 progeny per oviposition with a female biased sex ratio) (Overholt, 1998; Omwega and Overholt, 1997; Chinwada and Overholt, 2001; Cugala, 2002).

However, not all releases of *C. flavipes* have succeeded. In Tanzania, releases were made in the 1960s but establishment failed (CIBC, 1968 - 72). In South Africa, establishment failed as a result of harsh climatic winter conditions, lack of wild host plants to sustain *C. partellus* during the cropping off - season and the long diapause of stemborers in the highveld region of South Africa (Kfir, 1997). In such circumstances, it may be prudent to try and employ new parasitoid - host associations through the use of stemborer parasitoids from temperate areas. These parasitoids normally diapause and are adapted to the harsh winters (Kfir, 1997). A possible candidate in this respect is *Macrocentrus grandii* Goidanich (Hymenoptera: Braconidae) which was imported into the United States of America from Europe for the control of *O. nubilalis* (Kfir, 1997).

To avoid failure of biological control programmes, sole reliance on natural enemy - host compatibilities reported in literature must be avoided since biotypes of the host or natural enemy may exist and these may not be able to make old or new associations (Mochia *et al*., 2001). Before any releases of parasitoids to establish new or old associations between

the parasitoid and the host stemborer species are conducted, it is therefore imperative that host suitability studies on stemborer populations from the intended release sites be carried out first (Ngi - Song *et al*., 1998). This helps in the prediction of the possibility of successful establishment of the parasitoid as well as explaining why establishment of the parasitoid does not happen in some areas. Wiedenmann and Smith (1993) stated that before any releases of a parasitoid are conducted, physiological compatibility between the target host and the natural enemy needs to be evaluated. When there is compatibility, the parasitoid will be able to complete its life cycle. Host suitability studies will not only increase the understanding of success or failure of biological control programmes, but also avoid wasted effort and resources in rearing and releasing parasitoids incompatible with the targeted host (Cugala, 2002). Cugala (2002) reported high levels of compatibility between *C. flavipes* and *C. partellus* in Mozambique. Other workers who have found compatibility between *C. flavipes* and *C. partellus* are Chinwada (2002) working in Zimbabwe and Omwega and Overholt (1997) working in Kenya. Ngi - Song *et al.*, (1995) also reported compatibility between *C. flavipes* and some *C. partellus* populations from Kenya. Studies have shown very little compatibility between *C. flavipes* and *B. fusca*. Laboratory experiments in Kenya have shown that *C. partellus* is a suitable host for development while *B. fusca* is only acceptable for oviposition but subsequent development fails due to egg encapsulation (Ngi - Song *et al*., 1995). Cugala (2002), working in Mozambique reported very low compatibility between *C. flavipes* and *B. fusca*. Factors which affect the development of a parasitoid inside a host larva are diverse and they include among others, the host defence system, for example egg encapsulation; competition among parasitoids; the presence of toxins detrimental to the parasitoids and

host nutritional inadequacy (Vinson and Inwantsch, 1980). However, parasitoids have a way of getting around the obstacles. Most ichneumonid and braconid parasitoids suppress the immune system of the host by injecting some substances during oviposition which interfere with the immune system of the host (Fleming, 1992).

CHAPTER 3

MATERIALS AND METHODS

3.1 HOST SUITABILITY STUDIES OF *COTESIA FLAVIPES*

3.1.1 Rearing procedures for stemborers

Rearing of the stemborers and the experiments were conducted at the Plant Protection Research Institute, Harare during the 2003/4 and 2004/5 cropping seasons. Six populations, one of *B. fusca* and five of *C. partellus* were used in the study. *Busseola fusca* was collected from Harare Research Station while *C. partellus* populations were collected from Muzarabani, Bushu, Sanyati, Mamina and Musikavanhu (Table 3.1). Laboratory colonies of each species and population were set up using the field - collected stemborer larvae and pupae. Collection involved dissection of plants showing signs of stemborer damage and removal of stemborer larvae and pupae. From the field, larvae were brought into the laboratory and reared in ventilated plastic jars at room temperature ($25 \pm 2^{o}C$) until pupation. Pieces of fresh maize stems were supplied as food. Stems were dissected after every two days to check for larval mortality, natural parasitism or pupation. Pupae present at each checking interval were removed and placed in petri dishes inside small wooden cages (40 x 30 x 60 cm) for adult emergence (Plate 13). Maize plants (5 - 6 week old) with intact leaf sheaths and leaves were provided for oviposition. To prevent rapid plant desiccation, the base of each stem rested in water

inside a small vial and the mouth was plugged with cotton wool to prevent moths from falling into the water and drowning. Distilled water (on soaked cotton wool) was supplied to the moths inside each cage (Plate 13).

Plate 13. Oviposition cage for *Chilo partellus*

Leaves and sheaths were examined each morning for eggs. Egg batches were removed and placed in petri dishes for hatching. Early instar larvae were reared in groups inside 350 ml jars and supplied with tender leaves. After two weeks of rearing, the leaf diet was replaced with stems. All rearing was conducted at room temperature conditions in the insect rearing unit.

3.1.2 Rearing procedures for *Cotesia flavipes*

Rearing of *C. flavipes* up to cocoon stage was conducted at ICIPE. Cocoon masses were then sent to Zimbabwe upon request. Upon receipt, cocoons were placed in glass vials (2.5 cm dia. x 7.5 cm high) for incubation. The top ends of the vials were plugged with cotton wool to prevent the escaping of adult *C. flavipes* and at the same time enabling good ventilation. Some batches of cocoons were incubated at 25^{o}C while some were

incubated at 15^{o}C so as to stagger adult emergence. This was done to make sure that parasitoid adults emerged when there is a sufficient number and right size of larvae for oviposition. Once adult emergence started, cocoons were placed in perspex cages for mating. Artificial light was used to enhance mating between adult parasitoids. Adults were fed on 20 % honey/water solution (soaked on cotton wool) and left to mate for about 24 hours.

3.1.3 Experimental procedures

The relative suitability of each stemborer population for development of the parasitoid was studied by assessing the subsequent development of the parasitoid on each stung host. Stinging (ovipositing) involved holding 3rd - 4th (medium - sized) larvae with soft forceps and putting them into a cage, containing mated 24 - hour old mated *C. flavipes* adult females. This hand - stinging procedure was described in detail by Overholt *et al*. (1994a). As soon as the larvae were placed in the cage, mated females oviposited on them (Plate 14). Each larva was permitted to be stung only once and by a single parasitoid.

Plate 14. Mated adult *Cotesia flavipes* ovipositing on a *Chilo partellus* larva.

Soon after being stung, the larvae were removed from the cage and were placed individually in plastic vials (4 cm dia x 10 cm high) with perforated tops (Plate 15). Fresh maize stems were supplied as food. Vials were incubated at 25^{o}C. A total of 100 larvae of each stemborer population were exposed to mated parasitoids by *C. flavipes*. Of these, 20 larvae were the control. These were not stung. Sodium hypochlorite was used to clean the working surfaces to maintain hygiene so as to avoid stemborer larvae deaths due to diseases.

Plate 15. Plastic vial for stemborer rearing

The maize stem diet was changed after every two days under hygienic conditions. Dead larvae and pupae were removed from the vials.

Ten days after stinging, larvae were removed from the diet and placed individually in clean vials so that parasitoid emergence would occur outside the diet. Each larva rested on a piece of clean white paper placed at the base of the vial. This was to ensure that all the emerging parasitoid larvae were accounted for as well as ensuring that cocoon emergence took place outside the diet media (Chinwada *et al.,* 2003). One day after parasitoid emergence, the paper was removed.

3.1.3.1 Experimental Design and Data Collection

The experiment was set up as a Randomized Complete Block Design with 6 treatments (stemborer populations) and 4 blocks (time). The proportion of stung larvae that produced cocoons, parasitoid egg - larval developmental period, pupal period, the brood size and sex ratio, the number of parasitoid larvae that exited the host but failed to pupate and the number of parasitoids that died inside cocoons were recorded (Appendix 2.1.).

3.1.3.2 Data Analysis

Data were subjected to analysis of variance (ANOVA) (Proc GLM, SAS Institute, 1981) followed by the Student - Newman - Keul mean separation test when the ANOVA was significant ($P < 0.05$). Insect counts were log - transformed before being subjected to analysis (Sokal and Rohlf, 1981). Percentage stemborer parasitism was arcsine square root - transformed before being subjected to the ANOVA (Sokal and Rohlf, 1981). After analysis, the data were detransformed (Sokal and Rohlf, 1981). All larvae which pupated

or died shortly after exposure were not included in the analysis. The effect was felt not to be due to parasitoid exposure.

3.2 RELEASES OF *C. FLAVIPES*

3.2.1 Release sites

The sites for *C. flavipes* releases and post - release surveys are shown in Table 3.1.

Table 3.1. Sites for *C. flavipes* releases and post - release surveys.

Ecological Zone	Coordinates	Elevation (m)
Lowveld (< 600 m)		
Muzarabani	31° 10' 01s E 16° 20' 00s S	441
Musikavanhu	32° 18' 41s E 20° 25' 52s S	430
Birchenough Bridge	32° 20' 00s E 19° 50' 00s S	525
Chisumbanje	32° 15' 00s E 20° 48' 00s S	421
Middleveld (600 - 1200m)		
Bushu	31° 40' 25s E 17° 06' 44s S	866
Sanyati	29° 22' 57s E 17° 56' 29s S	841
Highveld (> 1200)		
Mamina	30° 42' 32s E 18° 37' 38s S	1316

Releases were made in 1999 at Musikavanhu and in 2000 at Muzarabani. About 50000 cocoons were released on dry season maize at Musikavanhu while 1000 were released in Muzarabani. In 2001, about 28 500 and 69 000 *C. flavipes* cocoons were released at Birchenough Bridge and Bushu respectively (Chinwada *et al.*, 2001). An additional 1500 were indirectly released at Bushu through *C. partellus* larvae that were exposed to day old mated female *C. flavipes.*

Sites for releases (Table 3.1) conducted in 2004 and 2005 were selected basing on preliminary survey data as well as Chinwada *et al.,* (2001) work. The sites were Bushu, Sanyati, Chisumbanje, Musikavanhu, Coburn Estates (Chegutu) and Mamina. Of these

sites, *C. flavipes* had earlier on been released at Musikavanhu in July 1999 and Bushu in March and December, 2001 (Chinwada *et al*, 2001; Chinwada, pers. communication).

3.2.2 Methods of release

To increase chances of establishment, three different methods of parasitoid release were used (Table 3.2). One method involved release of mated adult parasitoids. The other method was an indirect release of *C. flavipes* through infesting plants using pre - stung host larvae. With this method, *C. partellus* larvae were exposed to mated female *C. flavipes* adults which were put directly into stems through holes made using sharpened sticks. The third method involved release as darkened cocoons, which were placed in funnels or were placed into small vials, which were tied onto stems of plants.

Table 3.2. *Cotesia flavipes* release locations in Zimbabwe and approximate numbers released per location in 2004 and 2005.

Location	Date of release	Methods of release	Approximate number released
Lowveld			
Chisumbanje	23.7.2004	Mated adults	1000
Middleveld			
Bushu	30.7.2004	Mated adults	40 000
		Cocoons	50 000
	12.8.2004	Mated adults	20 000
Sanyati	13.12.2004	Mated adults	10 000
		Pre - stung larvae	103
	7.2.2004	Pre - stung larvae	82
Highveld			
Coburn Estates	8.2.2004	Pre - stung larvae	25
Mamina	9.2.2005	Pre - stung larvae	15

3.3 POST - RELEASE SURVEYS

3.3.1 Sampling Method

This study involved sampling of infested maize and sorghum plants which were six weeks and older at all locations on which *C. flavipes* had been release since 1999 (Table 3.1). However, sampling was not conducted at Birchenough Bridge because there was no maize in the farmers' fields at the time when sampling was conducted. Plants were sampled along two or more diagonal transects in a field and selecting a plant, or the nearest infested plant (Overholt *et al*., 1994b), every 5 - 10 steps. At least ten fields, each measuring approximately 40 x 20 m were sampled at each location. At least twenty plants were sampled from each field. Selected plants were examined for the presence of eggs under leaf sheaths and on leaf surfaces. All egg batches found on plants were taken to the laboratory for incubation to determine their parasitism status. Plants showing larval damage and feeding symptoms were dissected to collect stemborer larvae, pupae and parasitoid pupal stages inside tunnels.

3.3.2 Stemborer identification

All stemborers were identified to species level using taxonomic keys outlined by Polaszek (1998). Larval stages were reared in plastic vials (Plate 15) on a diet of maize stems which was changed after every two days until death, pupation or parasitoid emergence. The vials were incubated at room temperature in the insect rearing unit.

Parasitoid cocoons obtained in the field as well as those emerging in the laboratory were incubated at room temperature for adult emergence.

3.3.3 Parasitoid identification

Adult parasitoid males from each clutch were preserved in 70 % alcohol and were sent to ICIPE for identification to species level. This identification was conducted through microscopic examination of male genitalia (Polaszek, 1998). After identification to species level, parasitism attributable to *C. flavipes* or *C. sesamiae* was then determined.

3.3.4 Data collection

The stemborers species, number of parasitoid cocoons, numbers and the sex of emerging parasitoid adults were recorded on a data sheet (Appendix 2.2.).

CHAPTER 4

RESULTS

4.1 HOST SUITABILITY STUDIES

All the *B. fusca* used in the experiment did not produce cocoons due to egg encapsulation. They were dissected and encapsulated eggs were observed under a microscope. They were therefore not included in the analysis. Stung *C. partellus* larvae, which produced cocoons, were considered as suitable for the development of *C. flavipes*.

Chilo partellus populations from Bushu, Mamina, Musikavanhu, Muzarabani and Sanyati were suitable for development of *C. partellus* (Table 4.1). There were no significant differences in percentage parasitism among the five *C. partellus* populations ($F = 0.20$; $df = 4, 16$; $P = 0.9343$).

Table 4.1. Percentage parasitism of different *Chilo partellus* populations (±SE)

Host population	N	% host successfully parasitised
Lowveld		
Musikavanhu	49	72.1±3.7a
Muzarabani	39	65.1±11.9a
Middleveld		
Sanyati	53	77.3±5.2a
Bushu	48	73.3±1.7a
Highveld		
Mamina	55	61.9±14.6a

Numbers followed by the same letter in a column are not significantly different (P>0.05)
N=number of larvae exposed

Table 4.2 shows that there were significant differences among the populations for egg - larval period ($F = 6.93$; $df = 4, 238$; $P < 0.0001$). The longest egg - larval period was 12.3 days recorded for Mamina and the shortest was 11.6 days, which was recorded for Muzarabani. However, there were no significant differences for pupal periods ($F = 0,69$; $df = 4, 237$; $P = 0.6026$). The same trend was recorded for total developmental period though Mamina recorded 18.2 days while Muzarabani recorded 17.4 days.

Table 4.2. Egg - larval, pupal and total developmental periods (days) of *C. flavipes* (±SE)

Population	**N**	**Egg-larval period (days)**	**N**	**Pupal period (days)**	**N**	**Total developmental period (days)**
Lowveld						
Musikavanhu	49	12.0±0.90a	49	6.0±0.08a	49	18.0± 0.66a
Muzarabani	38	11.6±0.09b	37	5.8±0.30a	37	17.4±0.20a
Middlewveld						
Bushu	48	12.2±0.12a	49	5.9±0.08a	49	18.1±0.69a
Sanyati	53	11.7±0.08b	52	6.0±0.07a	52	17.7±0.57a
Highveld						
Mamina	55	12.3±0.14a	55	5.9±0.08a	55	18.2±0.35a

Numbers followed by the same letter in a column are not significantly different ($P>0.05$); N=number of larvae exposed.

The total parasitoid progeny per borer larvae and percentage female parasitoid progeny produced per stemborer larvae are shown in Table 4.3. The highest progeny per stemborer larva was 27.2.3 recorded for Sanyati while the lowest was 11.1 recorded for Muzarabani. All the populations, except Muzarabani, had a female biased progeny percentage. The highest was 78.1, which was for Bushu, and Muzarabani recorded 13.3 %.

Table 4.3. Total *C. flavipes* progeny and percent adult female progeny (±SE) produced per oviposition.

Population	N	Total progeny	N	% female progeny
Lowveld				
Musikavanhu	48	25.6±0.02a	48	77.5±2.8a
Muzarabani	39	11.1±0.1b	33	13.3±4.0b
Middleveld				
Bushu	49	26.3±0.02a	49	78.1±2.2a
Sanyati	53	27.2±0.02a	53	73.8±3.0a
Highveld				
Mamina	55	26.9±0.02a	55	77.5±2.8a

Numbers followed by the same letter in a column are not significantly different (P>0.05); N=number of larvae exposed.

Table 4.4 shows the total number of parasitoid larvae failing to pupate and the number which died inside cocoons. Only Muzarabani had a significantly higher number of parasitoid larvae which failed to pupate and that which died inside cocoons.

Table 4.4. Number of parasitoid larvae failing to pupate and number dying in cocoons (±SE) per host larva.

Population	N	No failing to pupate	N	No dying inside cocoons
Lowveld				
Musikavanhu	49	0.19±0.22a	48	0.13±0.02a
Muzarabani	39	0.80±0.06b	39	2.73±0.07b
Middleveld				
Bushu	49	0.06±0.01a	49	0.15±0.02a
Sanyati	53	0.15±0.02a	53	0.14±0.02a
Highveld				
Mamina	55	0.18±0.02a	55	0.09±0.01a

Numbers followed by the same letter in a column are not significantly different (P>0.05); N=number of larvae exposed.

4.1.1 *COTESIA FLAVIPES* RELEASES AND POST - RELEASE SURVEYS

4.2.1 Stemborer species composition at release locations

Table 4.5 shows stemborer composition at release locations. Three species namely *C. partellus*, *B. fusca* and *S. calamistis* were recovered at *C. flavipes* release locations during surveys conducted in 2004 and 2005. *Chilo partellus* was the dominant species at Chisumbanje, Musikavanhu and Muzarabani, accounting for over 97 % of the total number of stemborers sampled at each location. It was also dominant at Bushu and Sanyati. At Mamina (highveld), it had slight dominance over *B. fusca* on 10 December 2004 and 9 February 2005. However, on 8 April 2005, the percentage of *B. fusca* was higher than *C. partellus*. *Sesamia calamistis* was found at all sites except Muzarabani though in low proportions while *B. fusca* occurred in four. Surveys conducted during winter at Bushu on 30 July 2004 had a lower number of infested plants as well as stemborer larvae when compared to surveys conducted during the summer season, i.e. 5 February 2004 and 13 January 2005.

Table 4.5. Stemborer species composition at different *C. flavipes* release locations.

Location	Sampling date	No of larvae	No of pupae	Stemborer species composition (%)		
				C. partellus	*B. fusca*	*S. calamistis*
Lowveld						
Chisumbanje	23.07.2004	60	0	90.0	0.0	10.0
Musikavanhu	04.03.2004	681	20	99.4	0.0	0.6
	15.12.2004	339	4	97.3	2.7	0.0
	10.02.2005	750	0	100.0	0.0	0.0
Muzarabani	20.02.2004	1000	10	100.0	0.0	0.0
	27.01.2005	295	1	99.7	0.3	0.0
Middleveld						
Bushu	05.02.2004	1312	6	100.0	0.0	0.0
	30.07.2004	44	0	97.7	0.0	2.3
	13.01.2005	490	5	99.0	1.0	0.0
Sanyati	27.03.2004	27	1	100.0	0.0	0.0
	13.12.2004	646	2	100.0	0.0	0.0
	08.02.2005	87	0	82.8	0.0	17.2
Highveld						
Mamina	10.12.2004	524	1	51.6	47.1	1.3
	09.02.2005	164	0	58.5	41.5	0.0
	08.04.2005	231	0	41.1	54.1	4.8

4.2.2 Stemborer larval densities at *Cotesia flavipes* release sites

Table 4.6 shows the densities of stemborer larvae at *C. flavipes* release sites. There were variations in the densities of stemborer species at various locations. A density of 8.2 larvae per plant for *C. partellus* was recorded at Sanyati on 13 December 2004. This was followed by 7.5 for Bushu recorded on 05 February 2004. For *B. fusca*, the highest density, which was 3.1, was recorded at Mamina on 08 April 2005. On the same date, Mamina recorded the highest density for *S. calamistis* which was 0.3.

Table 4.6. Stemborer larval densities (number of larvae/infested plant) at *C. flavipes* release locations.

Location	Sampling date	Total no of infested Plants	Stemborer species density/infested plant.		
			C. partellus	***B. fusca***	***S. calamistis***
Lowveld					
Chisumbanje	23.07.2004	31	1.7	0.0	0.2
Musikavanhu	04.03.2004	175	3.9	0.0	0.0
	15.12.2004	198	1.7	0.0	0.0
	10.02.2005	174	4.3	0.0	0.0
Muzarabani	20.02.2004	195	5.1	0.0	0.0
	27.01.2005	137	2.1	0.0	0.0
Middleveld					
Bushu	05.02.2004	175	7.5	0.0	0.0
	30.07.2004	32	1.3	0.0	0.0
	13.01.2005	97	5.0	0.0	0.0
Sanyati	27.03.2004	15	1.8	0.0	0.0
	13.12.2004	78	8.2	0.0	0.0
	08.02.2005	68	1.0	0.0	0.2
Highveld					
Mamina	09.12.2004	110	2.4	2.2	0.0
	09.02.2005	58	1.7	1.2	0.0
	08.04.2005	40	2.4	3.1	0.3

4.2.3 Stemborer parasitism at *C. flavipes* release locations in 2004 and 2005

Table 4.7 shows the detailed recovery rates of various natural enemies from *C. flavipes* release locations. A percentage parasitism of 23.2 by *C. flavipes* was recorded on *Chilo partellus* on 13 January 2005 at Bushu while Muzarabani had 3.0 % on 27 January 2005 (Table 3.2). On 8 January 2005, *C. partellus* parasitism due to *C. flavipes* at Sanyati was 4.2%. At Musikavanhu on 4 March 2004, 2.0 % parasitism by *C. flavipes* was recorded on *C. partellus*. Musikavanhu also recorded 0.5 and 22.5 % parasitism on 15 December 2004 and 10 February 2005 respectively. A percentage of 1.3 on *C. partellus* attributed to

C. flavipes was also recorded at Chisumbanje (1.3 %) on 23 July 2004. However no previous releases were made at Chisumbanje. A percentage parasitism of 4.2 *C. flavipe*s on *C. partellus* was recorded at Sanyati in the same cropping season. At Mamina *Cotesia* spp. was recovered on *C. partellus, B. fusca* and *S. calamistis*. However, species determination is yet to be conducted. *Cotesia sesamiae* was recovered at Mamina, Musikavanhu and Sanyati. The highest percentage was 8.3 and was recorded at Mamina on 9 December 2004 where it was recovered from *S. calamistis* while 3.8 was recovered from *C. partellus*. *Chilo partellus*, *B. fusca* and *S. calamistis* larvae sampled on 8 April 2005 were also parasitised by *Cotesia* spp. However the actual species is not yet known since *C. flavipes* releases were done at Mamina on 9 February 2005. Parasitism by *Cotesia* spp. of 4.6 % was also recorded at Bushu during the 2003/4 season on *Chilo partellus*. However, the determination of the actual species was not done. Other natural enemies recovered were the mermithid nematode, *Hexamermis* sp. from Bushu where it parasitised 0.7 % of *C. partellus* larvae on 13 January 2005. The pupal parasitoid, *Dentichasmiasis busseolae* was also recovered from Muzarabani on 20 February 2004 where it parasitised 20 % of pupae. At Bushu it also parasitised 20 % on 13 January 2005.

Table 4.7. Stemborer larval parasitism recorded at *C. flavipes* release locations in 2004 and 2005.

Location	Sampling date	Borer Species	N	% parasitism		
				C. flavipes	***C. sesamieae***	***Hexamermis* sp.**
Lowveld						
Chisumbanje	23.07.2004	*C. partellus*	78	1.3	0.0	0.0
Musikavanhu	04.03.2004	*C. partellus*	491	2.0	0.2	0.0
	15.12.2004	*C. partellus*	182	0.5	0.5	0.0
	10.02.2005	*C. partellus*	463	22.5	0.2	0.0
Muzarabani	20.02.2004	*C. partellus*	134	3.0	0.0	0.0
Middleveld						
Bushu	30.07.2004	*C. partellus*	40	0.0	0.0	0.0
	13.01.2005	*C. partellus*	254	23.2	0.0	0.7
		B. fusca	5	0.0	0.0	0.0
Sanyati	27.03.2004	*C. partellus*	27	0.0	7.4	0.0
	13.12.2004	*C. partellus*	305	0.0	0.3	0.0
	08.02.2005	*C. partellus*	72	4.2	0.0	0.0
Highveld						
Mamina	09.12.2004	*C. partellus*	133	0.0	3.8	0.0
		S. calamistis	12	0.0	8.3	0.0
	08.04.2005	*C. partellus*	75	0.0	32.0	0.0
		B. fusca	46	0.0	23.9	0.0
		S. calamistis	7	0.0	14.3	0.0

4.2.4 Clutch sizes and sexes of *Cotesia flavipes* adults

The results obtained for sexes of *C. flavipes* adults were biased towards females (Table 4.8). All the progeny had over 67 % females. Locations with very low levels of parasitism, Muzarabani and Sanyati also produced low mean numbers of progeny per clutch i.e. 5.7 and 11.5 respectively as compared to Musikavanhu and Bushu which had 16.8 and 21.9 respectively.

Table 4.8. Clutch sizes and sexes of *Cotesia flavipes* adults emerging from parasitised *Chilo partellus* larvae at *C. flavipes* release sites sampled during the 2004 - 2005 cropping season.

Site	**Sampling date**	**Number of cocoon masses***	**Number of emerging parasitoid progeny/clutch (Mean ± S.D)**	**% female progeny (Mean ± S.D)**
Lowveld				
Musikavanhu	10.2.2005	104	16.8±9.9	70.4±24.2
Muzarabani	27.1.2005	4	11.5±9.2	83.3±13.1
Middleveld				
Bushu	13.1.2005	59	21.9±12.3	67.5±25.9
Sanyati	8.2.2005	3	5.7±2.8	72.2±25.9

* total includes cocoons found in empty stem tunnels in the field.

CHAPTER 5

DISCUSSION

Cotesia flavipes was able to develop on *C. partellus* from Bushu, Mamina, Musikavanhu, Muzarabani and Sanyati. Previous work done in Zimbabwe by (Chinwada, 2002) also showed that it can successfully develop on *C. partellus* from Chisumbanje. It was also shown that it can develop on *C. partellus* from Mozambique (Cugala, 2002) and from coastal Kenya (Ngi - Song *et al*., 1995). The results obtained for percent parasitism do not vary widely with 74.6 % which was recorded in Kenya (Ngi - Songa *et al*., 1995).

The shortest developmental period (egg - adult period) was 17.4 days which was recorded for Muzarabani and the longest recorded was 18.2 days for Mamina. These results compare well with a developmental period of 18.8 days which was recorded by Omwega and Overholt (1997). Ngi - Song *et al*. (1995) recorded 17.9 days. Chinwada (2002) recorded 21.9 days while Cugala (2002) recorded 17.9 days. A short developmental period is advantageous for the rapid colonisation of a habitat since the parasitoid can multiply quickly in a short space of time.

For total progeny, the highest obtained was 27.2 adults for stemborers from Sanyati. This compares well with 27.4 recorded by Chinwada (2002) and 26.8 recorded by Cugala (2002). However, this is less than 36.5 recorded by Ngi - Song *et al*. (1995). This low level of progeny production may partly explain the long time it can take for the parasitoid

to establish in the country. The production of large numbers of progeny can lead to quick colonisation and establishment of the parasitoid and this is very advantageous in biological control programmes.

The progeny were female - biased for Bushu, Mamina, Musikavanhu and Sanyati with over 70 % of the progeny being females. This compares well with Cugala (2002) who recorded 68 % and Ngi - Song *et al*. (1995) who recorded 66 and 72.6 % for 3rd and 4th instar *C. partellus* larvae respectively. However, for the Muzarabani population, females accounted only for 13.3 %. Chinwada (2002) also recorded male - biased adult *C. flavipes* progeny for the Chisumbanje population which produced 41.3 % females. This may result in a reduced rate of colonisation of new habitats because it is the females which lay eggs after mating.

The successful development of *C. flavipes* on all the *C. partellus* populations suggests that it has a mechanism of overcoming the immune system of these stemborer populations. *Cotesia flavipes* injects substances into the stemborer body on laying eggs, which interfere with the immune system of host (Edson *et al*., 1981; Stoltz and Guzo, 1986; Fleming, 1992). These results show that there are chances for successful establishment of the parasitoid because there is physiological compatibility between the stemborer populations and *C. flavipes*.

Releases of *C. flavipes* were conducted at Sanyati (middleveld) as well as at the highveld locations of Mamina and Coburn Estates (Chegutu) for the first time. The releases at highveld locations were a marked deviation from the norm in which all releases conducted before where at low and middleveld locations. The releases were made possible because of the presence of high numbers of the co - evolved host, *C. partellus* at these locations and this is a factor which enhances chances of successful establishment.. Another factor, which can enhance the chances of establishment at Mamina and Sanyati is the year - round cultivation of maize at the sites. This ensures a continuous supply of stemborer hosts for the parasitoid. This further enhances its chances of establishment.

In studies to determine the species composition at release sites, *C. partellus* was found to dominate the lowveld and middleveld locations. These results were similar to the findings of (Chinwada, 2002). However, at the highveld location of Mamina, the numbers of *B. fusca* and *C. partellus* did not vary widely with the former dominating the third sampling on 08 April 2005 when it accounted for 54.1 % of the total stemborer composition. For sampling done at the same location on 10 December 2004 and 09 February 2005, *C. partellus* showed dominance over *B. fusca* when it accounted for 51.6 % and 58.5 % respectively of all the stemborer larvae sampled. Previous studies by Chinwada and Overholt (2001) did not record a presence of this species at Mamina. This shows that the species is capable of adapting to a wide range of agroecological zones. The ability of this pest to colonise high altitude areas is a serious threat to production of host crop plants in the highveld. This justified the parasitoid releases conducted at Mamina. *Sesamia*

calamistis was found in low proportions at all the sites considered. This is similar to the findings of Chinwada (2002).

In post - release surveys, recoveries of *C. flavipes* at Musikavanhu, Muzarabani, and Bushu confirmed establishment of the parasitoid whose releases commenced at Musikavanhu in July 1999, Muzarabani in March 2000 and Bushu in March 2001 (Chinwada *et al.*, 2001). This was expected since the areas have the parasitoid's co - evolved host species, *C. partellus* as the dominant species (Cugala, 2002). Results of host suitability studies also showed that *C. flavipes* can successfully develop in *C. partellus* from Musikavanhu, Muzarabani and Bushu. Muzarabani and Musikavanhu are lowveld areas which have *C. partellus* as the dominant species (Sithole, 1990). Previously, the parasitoid was not recovered at Bushu, Muzarabani and Musikavanhu. This may be attributed to inadequate sampling or to an extremely low number of the parasitoid during the initial stages of habitat colonisation.

The delay in recovering the parasitoid some years after its release is not unique to Zimbabwe alone. In Kenya where the parasitoid was released in 1993, the levels of parasitism remained very low for the first four years and then increased to over 30 % (Overholt, 1998). This was also observed in Barbados after release on *D. saccharalis* in sugarcane where recoveries were not made for at least a year despite intensive surveys. The recovery rate then increased steadily over the following few years (Alam *et al*., 1971). The population simply needs time to build up (Cugala, 2002). However, for quick

benefits to the farmer, it is preferred that the parasitoid builds up quickly as this will immediately lower the losses caused by stemborer damage.

At Chisumbanje where releases had not been made before 2004, *C. flavipes* was recovered from one *C. partellus* larva. This may be attributed to dispersion from Musikavanhu, which is 60 km away and where releases were made in July 1999 on winter irrigated maize. A similar scenario was also observed in mainland Tanzania where the parasitoid established partly as a result of migration from neighbouring Kenya where the parasitoid had earlier on been released (Omwega *et al*., 1995, 1997; Nsami *et al*., 2001). In Kenya, the parasitoid is spreading and is now being recovered from sites further away from the original sites of release (Overholt *et al*., 1997). However, the concentration of the parasitoid in the first three years after release (1994 - 96) was around the release sites (Zhou and Overholt, 2001). In Mozambique, Cugala (2002) reported *C. flavipes* recoveries at locations more than 10 km from the release sites. The ability of the parasitoid to disperse from the release locations is advantageous as it lessens costs which would have to be incurred if releases are to be conducted at all sites where the target pest species occurs.

At Sanyati, *C. flavipes* was recovered from three *C. partellus* larvae in the same season of release (2004/5). Unlike, Musikavanhu, Muzarabani and Bushu where releases were first conducted between 1999 and 2001, recoveries simply indicate successful habitat colonisation rather than establishment. However, chances of successful establishment are

very high because its co - evolved host is the dominant species in the area and the results of host suitability studies give an indication of this.

The recovery of *C. sesamiae* from *B. fusca* and *C. partellus* at Mamina (Highveld) agrees with reports by Chinwada and Overholt (2001) who recovered the parasitoid from stemborers sampled at highveld locations. *Cotesia sesamiae* is the most widely recorded larval parasitoid of *B. fusca* in sub - Saharan Africa (Overholt, 1998). The surveys also showed that there are various natural enemies, which contribute to the mortality of stemborers. The indigenous *C. sesamiae* was recovered from *C. partellus*, *B. fusca* and *S. calamistis* and was recorded at all the sites sampled with the exception of Muzarabani. The absence of *C. sesamiae* from Muzarabani was also reported by Chinwada *et al.,* (2001). This shows that it is generally widely distributed. This is expected since it has adapted to the local conditions. The presence of this indigenous parasitoid and the high levels of maize and sorghum infestation by stemborers confirm the fact that it is not a very effective mortality factor. This brings about the need to try and introduce exotic species of parasitoids so as to have old and new host associations, which may result in the significant lowering of stemborer populations thereby benefiting the farmer.

The low mean number of *C. flavipes* adult progeny per *C. partellus* larva sampled in the field (5.7 - 21.9) may have contributed to the slow rate of establishment in the country. This is very low when compared to between 21.7 and 33.5 recorded by Cugala (2002). Ngi - Song *et al*. (1995) reported 36.5 from *C. partellus* and 35.2 *C. sesamiae* from *S.*

calamistis. The adult progeny from all the sites in Zimbabwe were female - biased. Females constituted over 67 % of the total progeny at all the four sites under consideration. Cugala (2002) reported between 68 and 78 % female progeny. This is also in agreement with the findings of Omwega and Overholt, (1997) and Chinwada and Overholt (2001) who reported female - biased progeny from *Cotesia* spp. This bias towards female progeny and the presence of males is an advantageous survival strategy, which helps, in the rapid colonisation of a habitat since females are the ones which oviposit after mating. Unmated females can also reproduce but the progeny will be all be males. If the progeny was male dominated, there will be reduced multiplication of the parasitoid and hence reduced colonisation of habitats which leads to reduced control of stemborers.

Winter sampling at Bushu on 30 July 2004 had a low number of stemborer larvae. This is expected since low temperatures are not favourable to the development of insects and they were possibly diapausing and this reduces the number of larvae available for parasitisation. When some larvae are diapausing, a small fraction of the stemborer larvae remain continuing with development thereby providing the parasitoid with very small numbers of stemborer hosts for parasitisation. There will therefore be little perpetuation of the parasitoid.

CHAPTER 6

CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

Many conclusions can be made from the results of the study. It can be concluded that *C. flavipes* can successfully develop on *C. partellus* populations from Bushu, Mamina, Musikavanhu, Muzarabani and Sanyati. Since releases were made over three seasons before the surveys at Muzarabani, Bushu, and Musikavanhu and recoveries of *C. flavipes* were made this year, it can be concluded that the parasitoid has established in Zimbabwe. It can also be concluded that establishment takes at least three seasons for it to rise to significant levels. Results from the study can also lead to the conclusion that *C. partellus* is the dominant stemborer species at low altitudes and it is spreading towards middleveld and highveld areas while *S. calamistis* is found at low, middle and high altitude though in very low numbers. *Busseola fusca* is found in large numbers in highveld locations. Another conclusion to be made is that *C. sesamiae*, *Hexamermis* spp. and *D. busseolae* are indigenous parasitoids which are contributing to the natural mortality of stemborers in Zimbabwe.

6.2 Recommendations

1. Host suitability studies using *C. flavipes* need to be conducted with populations of stemborers where parasitoid releases are to be conducted.

2. Since *Hexamermis* spp. has been found to contribute to the mortality of stemborers at Bushu, further studies to assess its suitability and impact need to be conducted.

3. Biological control of stemborers must target all stages of the insect. This brings the need to consider multiplying, conducting host suitability studies and releasing pupal parasitoids like *X. stemmator* and egg parasitoids like *Telenomus isis.*

4. Surveys to determine the level of parasitism by *C. flavipes* must continue so as to determine its impact on stemborer populations. The surveys must also cover areas where the parasitoid was not released so as to determine its rate of spread.

5. Studies must be conducted to determine the overwintering mechanisms of *C. flavipes.*

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APPENDICES

Appendix 1. ANOVA tables for host suitability studies

Appendix 1.1. Percent *C. partellus* larvae parasitised by *C. flavipe*

Source	DF	SS	MS	F	P
Population	4	0.07321077	0.01830269	0.20	0.9343
Error	16	1.45930591	0.09120662		
Total	20	1.53251668			

Appendix 1.2. Egg - larval period of *Cotesia* on different *C. partellus* populations

Source	DF	SS	MS	F	P
Population	4	17.29153509	4.32288377	6.93	<.0001
Error	238	148.4450904	0.6237189		
Total	242	165.7366255			

Appendix 1.3. *Cotesia flavipes* pupal period

Source	DF	SS	MS	F	P
Population	4	2.16643272	0.54160818	0.69	0.6026
Error	237	187.2385260	0.7900360		
Total	241	189.4049587			

Appendix 1.4. Total *Cotesia flavipes* progeny produced per oviposition on different *C. partellus* populations

Source	DF	SS	MS	F	P
Population	4	4.16024570	1.04006143	13.86	<.0001
Error	239	17.92840608	0.07501425		
Total	243	22.08865178			

Appendix 1.5. Percent female *Cotesia flavipes* adult progeny produced per oviposition on different *C. partellus* populations

Source	DF	SS	MS	F	P
Population	4	11787.21506	2946.80376	47.92	<.0001
Error	237	14574.35519	61.49517		
Total	241	26361.57025			

Appendix 1.6. Number of *Cotesia flavipes* larvae failing to pupate

Source	DF	SS	MS	F	P
Population	4	1.37943214	0.34485804	8.55	<.0001
Error	240	9.68243366	0.04034347		
Corrected Total	244	11.06186580			

Appendix 1.7. Number of *Cotesia flavipes* progeny dying inside cocoons

Source	DF	SS	MS	F	P
Population	4	8.89393317	2.22348329	53	<.0001
Error	239	9.95825370	0.04166633		
Total	243	18.85218687			

Appendix 2. Recording sheets for post-release surveys and host suitability studies

Appendix 2.1. Recording sheet for host suitability studies

Cotesia flavipes host suitability studies recording sheet

Stemborer host_______________________ Population_______________________

Vial no.	Host no.	Exposure date	Fate of exposed host	Fate Date	Parasitoid cocoon emergence date	Parasitoid adult emergence date	No of parasitoid adults emerged			No of parasitoids not forming cocoons	No of parasitoids dead inside cocoons	Egg-larval period (days)	Pupal period (days)
							females	males	Total				

Appendix 2.2. Recording sheet for post - release surveys

STEMBORER PARAMETERS

Date ______________________________ Longitude____________________
Location ___________________________ Latitude______________________
Cropping system _____________________ Elevation_____________________

Plant number	**Stemborers**							**Parasitism**	
	Borer species	Total number	Stage	**Location**				Host sp.	Parasitoid sp.
				Stem	Leaf	Funnel	Tassel		