

**STUDY ON THE BIOLOGY AND POPULATION
VARIATION OF *COTESIA FLAVIPES*
(HYMENOPTERA: BRACONIDAE) ON *CHILO
PARTELLUS* (LEPIDOPTERA: CRAMBIDAE)**

BY: Henok Abraha

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List of Abbreviations

EARO	Ethiopian Agriculture Research Organization
MARC	Melkasa Agriculture Research Center
Cf Ziw	<i>C. flavipes</i> collected from Ziway
Cf Mel	<i>C. flavipes</i> Collected from Melkasa
Cp Ziw	<i>C. partellus</i> collected from Ziway
Cp Mel	<i>C. partellus</i> collected from Melkasa
Cf Mel Cp Mel	<i>C. flavipes</i> of Melkasa reared on Cp of Melkasa
Cf Mel Cp Ziw	<i>C. flavipes</i> of Melkasa reared on Cp of Ziway
Cf Ziw Cp Ziw	<i>C. flavipes</i> of Ziway reared on Cp of Ziway
Cf Ziw Cp Mel	<i>C. flavipes</i> of Ziway reared on C.p of Melkasa
HSD	Honestly Significant Difference test (Tukey's studentized test)

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ABSTRACT

Maize and Sorghum are the major staple food crops in the tropics. Of many insects that attack these crops *Chilo partellus* (Swinehoe) (Lepidoptera: Crambidae) is the most notorious species. *Cotesia flavipes* Cameron (Hymenoptera: Braconidae) is a co-evolved parasitoid of *C. partellus* and has been utilized as a biological control of stemborers including *C. partellus* in many countries.

By measuring some life history parameters, the variability of *C. flavipes* from two different geographic locations, which were reared on two different geographic populations of *C. partellus*, was studied. The parameters taken were suitability test, developmental time, realized fecundity and longevity. The suitability test was studied at constant temperature of 28°C, while developmental time, realized fecundity and longevity were studied at constant temperatures of 20°C, 25°C, 28°C and 30°C. All the experiments were studied under 12L: 12D photoperiod and a relative humidity of 50-60%. Data were analyzed using SPSS computer software, when ANOVA results were significant, means were separated by Tukey's studentized test (HSD).

In the suitability test, the best population interaction was when *C. partellus* and *C. flavipes* populations were from the same geographic area. Both populations of *C. flavipes* have shown variability when the hosts were from different geographic populations. The developmental time of *C. flavipes* increased as the temperature increased, however, the total progeny produced per female and longevity increased as the temperature decreased. Effect of temperature and populations were seen in most of the life history of *C. flavipes*, which are bases for successful establishment and efficient parasitism.

1. INTRODUCTION

Cereal crops, particularly maize (*Zea mais L.*) and sorghum (*Sorghum bicolor moech*) are the main staple food crops in eastern and southern African countries (Minja, 1990). In Ethiopia, these crops rank first and second in terms of yield per hectare and total production, respectively. They account for about 41% of the total crop production of the country (CSA, 2000).

Among the various insect pests attacking maize and sorghum, Lepidopterous stem borers are considered to be the most damaging (Nye, 1960; Ampofo, 1986; Seshu Reddy and Sum, 1992). In Africa, more than 18 species of stem borers in the order Lepidoptera cause damage to maize and sorghum (Maes, 1997; Polaszek, 1997). They belong to three families: Crambidae, Noctuidae and Pyralidae (Maes, 1997).

In Ethiopia, four Lepidopteran and two Coleoptera stem borers attack maize and sorghum (Emana Getu, 2001; Emana Getu *et al.*, 2001). Of the various insect pests attacking maize and sorghum in eastern and southern Africa, the maize stalk borer, *Busseola fusca* Fuller (Lepidoptera: Noctuidae) and the spotted stem borer, *Chilo partellus* (Swinhoe) (Lepidoptera: Crambidae) are by far the most important (Kfir, 1998). *Chilo partellus* was introduced accidentally from Asia to Africa in the early 1930's (Tams, 1932) while *B. fusca* is indigenous to Africa (Harris and Nwanze, 1992). *Chilo partellus* is a polyphagous insect pest and has a wider host range which attack sorghum, maize, pearl millet, finger millet, rice, wheat, sugar cane, foxtail, and various wild grass species (Sithole, 1990).

Cotesia flavipes Cameron (Hymenoptera: Braconidae) is important parasitoid of graminaceous stem borers in the Oriental region. It has been utilized for biological control

worldwide. *Cotesia flavipes* is now established in eastern and southern Africa countries including Ethiopia (Emana Getu *et al.*, 2001, Cugala and Omwega, 2001; Matama *et al.*, 2001; Nsami *et al.*, 2001; Songa *et al.*, 2001).

Local parasitoid populations may be adapted to their sympatric major plant-host complex. Parasitoid strains may thus differ in their physiological compatibility with particular host species. Introduction of a parasitoid population can induce artificial allopatric speciation by forcing the parasitoid to locally adapt to its new host or host microhabitat. This scenario could induce the formation of adapted parasitoid races and over time even distinct species. The failure or success of a parasitoid introduction can thus be dependent on the physiological compatibility between the introduced parasitoid and local host population (Potting, 1996). *Cotesia flavipes*, a larval parasitoid used worldwide in biological control of tropical stemborers, has a wide host range in diverse habitats. Potting *et al.* (1997a) reported the existence of *C. flavipes* strains based on differences in physiological compatibility between local parasitoid and host population. However, variability in terms of reproductive success among different populations of *C. flavipes* and *C. partellus* in Ethiopia is not known.

Laboratory investigations indicated that *C. flavipes* performed differently in various stemborer species and at different temperatures (Ngi-Song *et al.*, 1995; Potting, 1996; Mbapila, 1997). Among many other factors that limit the distribution of *C. flavipes*, temperature is the most important. In Ethiopia there is a plan to release *C. flavipes* inundatively in different parts of the country (Emana Getu, *pers. Comm.*). In order, to optimize releases in different parts of Ethiopia, it is useful to investigate if there is variability in terms of reproductive success and effect of temperature on the biology of *C. flavipes* among different geographic populations of *C. flavipes* and *C. partellus*. This study is carried out to investigate whether variability in

terms of reproductive success exist and the possible effect of temperature on the biology of *C. flavipes* on two geographic populations of *C. partellus* where inundative releases are planned.

Objectives of the study

General objective

To study the variation in geographic populations of *C. flavipes* in terms of reproductive success on *C. partellus* by measuring some life history parameters under different temperatures.

Specific Objectives

1. To determine the best interactions of *C. partellus* and *C. flavipes* populations for effective biological control.
2. To study developmental time, longevity and realized fecundity of two populations of *C. flavipes* under different temperatures on *C. partellus*.

2. LITRATURE REVIEW

2.1 Geographical Distribution, Lifecycle and Economic Importance of *C. partellus*

2.1.1 Geographical Distribution of *C. partellus*

Chilo partellus is native to Asia, but became established in East Africa in the 1930s where it was first recorded in Malawi (Tams, 1932). Since then, it has spread to southern and eastern Africa. It has been recorded in Afghanistan, Bangladesh, Cambodia, India, Indonesia, Laos, Nepal, Pakistan, Sri Lanka, Taiwan, Thailand, Vietnam, Yemen, Botswana, Comoros, Ethiopia, Kenya, Malawi, Mozambique, Reunion, Somalia, South Africa, Sudan, Tanzania, Uganda, Zimbabwe, Eritrea, Zambia and Zanzibar (CAB International, 2000).

Using geographic information system (GIS), Overholt *et al.* (2000) predicted the eventual distribution of *C. partellus* in Africa, which include several countries in West Africa. However, until now *C. partellus* has not yet invaded West Africa. Most important factors influencing the distribution of *C. partellus* are precipitation, maximum and minimum temperature regiems, while elevation played non significant role (Emana Getu *et al.*, 2002).

In Ethiopia, areas with high precipitation in the months of March and July, low precipitation in the months of August, high maximum temperature and low minimum temperature in August and low annual maximum temperature were predicted to have a high density of *C. partellus* (Emana Getu *et al.*, 2002).

2.1.2 Biology of *Chilo partellus*

Chilo partellus larvae are medium sized, straw or light brown in color, with numerous shiny brown spots on the forewings margins. The hind wings are properly thin and white. The moth is nocturnal in habit and usually lives for approximately one week. Adults emerge from pupae in the late afternoon or early evening and are active at night. They rest on plants and plant debris during the day and are seldom seen, unless disturbed. Females release a pheromone to attract males. Mating generally takes place soon after emergence and on the 2nd or 3rd subsequent nights (Berger, 1989). On 2nd or 3rd nights after emergence, each female lays a total of 200-600 scale-like overlapping eggs in 10-80 separate batches on the undersides of leaves, mostly near the mid-ribs. Eggs hatch in 5-11 days (Chapman *et al.*, 1983).

Many larvae are carried away on silken threads and infest neighboring plants. The larvae move to the leaf whorl and feed on tender leaves. The larval development is completed in 24-27 days (Ampofo, 1986). *Chilo partellus* has usually six instars with capsule measurement of 0.31, 0.43, 0.68, 1.05, 1.38 and 1.78 mm, respectively; rarely an additional instar is present with a head-capsule measuring 2.09 mm, and a few larvae pupate in the fifth instar. When parasitized by *C. flavipes* the larva doesn't molt unless the host larvae had been about to molt at the time of parasitization (Mohyuddin, 1971).

Larvae pupate in infested stems and adults emerge 5-12 days later. The entire life cycle is completed in 30-40 days when climate and growing conditions are favorable. During the entire cropping season three to four overlapping generation occur in the field. Five or more successive generations may develop in areas with suitable climates such as southern India, but in less favorable areas larvae pass the winter or dry season in diapause in stems and stubble. They may remain inactive for up to six months, before pupating and completing their

development early in the following growing season. Population density is low between cropping season (CAB International, 2000).

2.1.3 Economic Importance of *C. partellus*

Chilo partellus has been recorded attacking maize, sorghum, pearl millet, finger millet, wheat, sugar cane, rice, and various wild grass species including important fodder crops (Sithole, 1990). The original hosts of stemborers in general is believed to be wild grass species however due to cultivation of areas inhabited by wild grass species for cereal crops they have expanded their host range to include cereal crops (Overholt, 1998).

Chilo partellus can inflict severe damage and serious yield losses of maize and sorghum if not managed properly from the early growth stage of the crop. Infestation by *C. partellus* on maize starts with oviposition on the leaves causing lesions. The late third or early-fourth instars bore into the stem, feeding on tissues and making tunnels. When the infestation is severe, the larvae either in the leaf whorl or in the stem can cut the meristematic tissues, the central leaves dry up to produce the 'deadheart' symptom resulting in the death of the plant (Seshu Reddy, 1990).

Chilo partellus is considered to be the most important stemborer in most of low to medium elevation areas of eastern and southern Africa. Yield losses of 18% is attributed to *C. partellus* and *Chilo orichalcocilielus* in the southern coastal area of Kenya (Warui and Kuria, 1983). 88% yield losses in sorghum due to *C. partellus* was reported by Seshu Reddy (1988) in other area of Kenya.

In Ethiopia, Assefa Gebre-Amlak (1985), and Emanu Getu and Tsedeke Abate (1999) reported 10 to 100% yield losses due to stem borers. Since *C. partellus* is the 2nd most abundant and widely distributed stem borer in Ethiopia (Emanu Getu *et al*, 2001), it contributes for substantial amount of yield loss.

Recent evidences suggest that *C. partellus* is becoming increasingly a pest in higher elevations as well (Kfir, 1997; Emanu Getu, 2001). Kfir (2001) estimated yield losses due to *C. partellus* in maize and sorghum in western Transvaal and northwestern Orange Free State in South Africa to be more than 50%. Sithole (1989) in Zimbabwe reported a yield loss of 50-60% in sorghum due to *C. partellus*.

C. partellus has proved to be a highly competitive colonizer in many areas it has invaded often becoming the predominant and most important stem borer species in maize and sorghum (Seshu Reddy, 1990). There are evidences of partial displacement of native stem borers by *C. partellus*. In coastal areas of Kenya *C. partellus* has displaced *C. orichalcociliellus*. Similar studies in South Africa by Kfir (1992) indicated that the displacement of *B. fusca* over a period of 5-7 years where the proportion of *C. partellus* increased from 3% in 1986 to 91 % in 1992.

2.2 Biological Control of *C. partellus*

There are various control options of *C. partellus*. These include biological control, chemical control, botanical control, host plant resistance, cultural control and habitat management. All of this control options have limitations and are not able to bring a total control of stem borers in general individually. However, using these control options as a component of integrated pest management (IPM) might result in a sustainable control of this pest. Agricultural research and development institutions with a mandate to improve cereal production

techniques and insect pest management in developing countries have directed efforts towards an integrated approach, particularly one which incorporates biological control (Betbeder, 1989).

In general, the efficiency of natural enemies in particular farming system is not known. The short cropping period and the lack of continuous habitats limit the scope of successfully controlling stemborers with natural enemies (Nwanze and Muller, 1989). Combination of the different approaches of classical biological control, conservation of indigenous natural enemies, application of commercially produced micro-organisms and redistribution of locally important natural enemies may provide a suitable management strategy for the sustainable control of *C. partellus* in Africa (Bonhof *et al.*, 1997).

2.2.1 Biological Control of *C. partellus* Using Entomopathogens

Few diseases have been identified on cereal stemborers and their value, as a biological control agent is generally unknown. Pathogens that were recorded to infect *C. partellus* include *Aspergillus* sp., *Beauveria bassina*, *Matarhizium* sp., *Baculoviridae* (granulosis virus), Microsporidae, gregarinae, *Nosema* sp. attacking larvae of *C. partellus*. From Nematods, *Panagro lamimus*, *Hexamemis* sp. *Steinernema intermedia*, *Heterorhabditis* sp. were recorded (Bonhof *et. al.*, 1997).

In Africa many *Nosema* diseases destroy populations of *C. partellus* but their epidemiology and their relative efficiency have not been sufficiently studied (Betbeder, 1989). *Nosema maruca* reduced *C. partellus* population in sorghum significantly and increased yield by 34-80%. *N. maruca* was as effective as carbofuran and trichlorophan. This combined with its

non-toxicity to natural enemies makes *N. maruca* a potential component in the IPM of stemborers (Odindo, 1989).

The impact of naturally occurring fungi, bacteria, protozoa, viruses and nematodes is often mitigated by long dry periods between cropping seasons, and by lack of physical contact between stemborer larvae in a stem. Lack of physical contact between stemborer larvae in a stem is a serious constraint for the application of commercially produced microorganisms such as *B. thuringiensis* (Bonhof *et al.*, 1997).

Some laboratory and field experiments have been carried out to evaluate the pathogenicity of fungi, bacteria and virus. The effect of chromogenic strain of *Serratia marcescens* was studied on *C. partellus* in India. A single drop of the bacterium prevented eggs from hatching and killed newly hatched larvae (Chakravorty *et al.*, 1983). Sinha and Prasad (1975) reported that toxin of *Fusarium aleyrodis* could be used against *C. partellus*. But, very few pathogens have been tested under field conditions so far (Betbeder, 1989).

2.2.2 Biological Control of *C. partellus* Using Entomophagus Insects

Little information is available on the predators of stemborers. Many species of ants are predators of eggs and newly hatched larvae. A number of predators are found in the order Dermaptera and Hymenoptera. Sharma and Sarup (1979) reported the role of different spiders and Seshu Reddy (1985) reported the contribution of ants, ladybird beetle, and earwigs in reducing the population of *C. partellus*.

Grathead (1990) reported that four species of ants (*Tetramorium guineense*, *Phediole megacephala*, *Cardiocondyla badonei* and *Cardiocondyla emeryi*) destroyed almost 90% of

eggs and first instar larvae of *C. partellus* and *B. fusca*. Neonate larvae are vulnerable to predation, especially when migrating from the egg batch (often on the lower leaves of the plant) to the leaf whorl. Later instars which, feed in the more protected environment of the interior of the stem, are probably less vulnerable to predation. However, Predators of adults have not been observed (Bonhof *et al.*, 1997).

Although numerous general predators and diseases cause mortality, parasitoids have been the primary targets as biological control agents. Possibly because of their ecological diversity, host specificity and ability to attack hosts that feed cryptically within the plant. Many parasitoids of different families have been recorded in Asia, Africa and the Americas from *C. partellus*. Three families of larval parasitoids predominate: Braconidae, Ichneumonidae and Tachinidae. Braconids account for 35% of parasitoids recorded on *C. partellus* (Jepson 1954, Mohyddin & Greathead 1970).

Indigenous parasitoids may have a greater impact on stemborer populations residing in wild-grass communities than on populations that periodically invade annual crops. In perennial-grass habitats as opposed to annual cultivated grasses there is no need for natural enemies to migrate periodically to re-colonize the habitat, and thus stemborers and their natural enemies may be able to approach stability (Overholt, 1998).

Additionally, because stemborer survival is much lower in wild grasses than cultivated cereals, natural enemies may be better able to maintain stemborer populations below the economic injury level. There is also some evidence that stemborer larval growth is slower in wild plants which would result in a wider temporal window of susceptibility to stage-specific parasitoids (Smith and Widenmann, 1997). Finally, natural enemies may be more effective in

wild grasses due to physical or chemical constraints to host finding in cultivated grasses. Ectoparasitoids that attack concealed hosts by drilling through the stems of small diameter wild grasses may be unable to parasitize stemborers in the relatively thicker-stemmed maize. Olfactory response to volatiles emanating from wild plants and cultivated grasses may also differ (Overholt, 1998).

The impact of parasitoids and predators could be enhanced by managing native habitat adjacent to maize and sorghum fields. Wild grass species may play an important role in the conservation of parasitoids and predators. Stem borers migrate from maize and sorghum to wild grasses when the crops senesce. The presence of stemborers in wild grasses may allow parasitoids to survive dry periods between cropping seasons. Wild grasses and other plants may also provide shelter and alternative food for predators and parasitoids (Polaszek & Khan, 1998).

Because of the economic importance of *C. partellus*, and its status as an introduced pest, it has been the target of three classical biological control attempts in Africa. The Commonwealth Institute of Biological Control (CIBC) imported nine species of parasitoids of *C. partellus* from India and released these in Uganda, Tanzania, and Kenya from 1968-1972. In South Africa, 13 exotic parasitoids were introduced from 1977 to 1993. No establishments were reported in either of the programs (Kfir, 1994).

A third attempt to introduce exotic parasitoids for control of *C. partellus* was initiated in Kenya in 1991 by the International Center of Insect Physiology and Ecology (ICIPE). This program has focused on the introduction of *Cotesia flavipes* Cameron (Hymenoptera: Braconidae), which was also released in the earlier programs against *C. partellus* mentioned

above. *Cotesia flavipes* is native to the Indo-Australian region, but has been widely introduced against various stemborers in the neotropics, several Indian Ocean Islands and also been redistributed within Asia (Overholt, 1993).

2.3 *Cotesia flavipes* as a Bio-Control Agent of *C. partellus*

Classical biological control which is the introduction of co-evolved natural enemies from a pest aboriginal home into an area where the pest has invaded, is considered to be potentially effective pest management strategy against exotic pests. In addition, it can also provide an opportunity for reducing the damaging impact of native pests as well (Smith *et al.*, 1993). Indigenous parasitoids of African stemborers have expanded their host ranges to include the exotic stemborer, *C. partellus* but do not appear to effectively regulate densities at acceptable levels (Overholt, 1998). *Cotesia flavipes* was selected as the first candidate for introduction because of its history of success outside Africa, and its importance as a parasitoid of stemborers in its aboriginal home (Overholt *et al.*, 1997).

2.3.1 Release and establishment of *C. flavipes* in Africa

In Kenya, *C. flavipes* was released in three sites in four different seasons from 1996-1998. From the four stemborers that occur in these sites *C. flavipes* was recovered only from *C. partellus* and *S. calamistis*. It was found that *C. flavipes* had successfully colonized the stemborers in the three sites just after one season of release (Songa *et al.*, 2001). *Cotesia flavipes* is only likely to establish in areas where the predominant stemborer species available are suitable for its development (Overholt *et al.*, 1994a).

Cotesia flavipes has been released in Zimbabwe and prospects for its establishment appear to be good (Chinwanda *et al.*, 2001). In Mozambique and Uganda it was released in two

localities in 1996 and was recovered from *B. fusca*, *C. partellus*, *S. calamistis* (Cugala and Omwega, 2001; Mattama *et al.*, 2001).

Emana Getu *et al.* (2001) made a survey to know the species composition of stemborers and their natural enemies in Ethiopia. From this survey, *C. flavipes* was found to be the dominant parasitoid contributing for about 66% of the total parasitism and it was recovered from *C. partellus*, *B. fusca* and *S. calamistis*. It has never been released in Ethiopia and the speculation was that, it invaded Ethiopia may be from Somalia where it was released in 1997. In Tanzania, Nsami *et al.* (2001) have also reported the recovery of *C. flavipes* from two districts where no releases were made and it was speculated that it may have spread into this region from Kenya where it was established.

Several factors could be responsible for the success of *C. flavipes*. In its area of origin, it attacks several species of Crambid, Pyralid and Noctuid stemborers. The relatively wide taxonomic range of suitable hosts, coupled with a narrow habitat specificity, may favor its establishment. Stemborers often occur as species complexes, and a parasitoid that can exploit more than one of the hosts may be able to colonize a new area better than a monophagous parasitoid, due to a more constant availability of hosts. Another factor may be the high reproductive potential of *C. flavipes* in relation to most stemborers. *Cotesia flavipes* have short generation time (16-18 days) in comparison to the stem borer (30-50 days) and fairly high fecundity (30-40 progeny) per oviposition with a female-biased sex ratio (Overholt *et al.*, 1997)

Cotesia flavipes has high host searching ability due to its behavior of entering tunnels in plant stems to attack stemborer larvae. Many other larval parasitoids of stemborers remain on the

outside of the stem and parasitize larvae by drilling or locating breaches through the stem with their ovipositor. This strategy may be effective in wild grasses, but in a relatively larger stemmed cultivated cereals the length of the ovipositor may limit the number of hosts susceptible to attack (Overholt, 1998).

In Kenya, Ngi- Song et al. (1995) and in West Africa Hailemichael et al. (1997) studied the acceptance and suitability of some stemborers occurring in Africa including *C. partellus* *B. fusca* and *S. calamistis* for oviposition and development of *C. flavipes*. All were accepted by *C. flavipes* for oviposition, but development occurred only on *C. partellus* and *S. calamistis*. The eggs of *C. flavipes* were encapsulated in *B. fusca* indicating that *B. fusca* was not a suitable host.

The recovery of *C. flavipes* from *B. fusca* in Mozambique, Uganda and Ethiopia seems to contradict with the above finding. Cugala and Omwega (2001) gave two possible explanations. First, the immune system of *B. fusca* was significantly compromised by the attack of a pathogen or other parasitoid to allow the development of *C. flavipes*. Second, populations in these countries are physiologically different from Kenyan or West African population.

In a laboratory experiment in Ethiopia *C. flavipes* was developed from two different geographic population of *B. fusca* while it was encapsulated in other eight different populations of *B. fusca* indicating the existence of geographic variation in immune response of *B. fusca* in Ethiopia (Emana Getu, 2001).

2.3.2 Biology of *C. flavipes*

Cotesia flavipes adults start mating soon after emergence, especially in bright light. Mating lasted for about one minute. Unmated females produce males by parthenogenesis. Females start ovipositing on the day of emergence (Mohyuddin, 1971). *Cotesia flavipes* female enters through holes in stems excavated by stemborers, traverses the tunnel to locate the stemborer and then directly injects eggs into the host. Oviposition lasts only few seconds.

Eggs hatch within 3-4 days in its host larvae and first-instar parasitoid larva begins feeding internally. The immature parasitoids develop through three larval instars in the host body, the larval period on average is 11 days. After finishing its development the larvae emerge from the host by chewing through the integument. After emergence from the host, the last-instar larvae immediately spin cocoons and pupate. Pre-pupal and pupal period being 4-5 days after which adults emerge. In nature, the cocoons are found inside host feeding tunnels in graminaceous plants. Development is completed in 16 days at 30°C (larval period 11.5 days, pre-pupal and pupal period 4.5 days) (Mohyuddin, 1971; CAB International, 2000; Smith *et al.*, 1993).

The adults are small wasps, 3-4 mm in length, which live only a few days. The life span of *C. flavipes* is approximately 34 hours at 25°C if adults are not fed. The mean longevity of unfed female is about 24 hours at 24°C. Provision of a 20% honey/water solution prolongs the life span to about 51 hours. Females have an initial egg load of around 150 eggs and deposit around 40 eggs in a host and the highest reproductive success is on the later instars (4th-6th). In the first-encountered hosts 35-45% is allocated to each host, and a female *C. flavipes* is equipped with an egg load to parasitize 3-4 hosts only (Potting, 1997). The sex ratio is

usually female biased (60-70%) and males mate repeatedly (Kajita and Drake, 1969; Mohyuddin, 1971).

Cotesia flavipes and most of its hosts breed almost continually in equatorial humid climate, but in areas with a summer-rain climate their hosts undergo a resting period in response to dry conditions, low temperatures or combination of the two. These periods coincide with periods of slower development within their hosts. In larvae that had been collected from dry stems during the dry period, exposed to parasitoids and then fed in fresh stems, development was extended to 35 days. In areas where the hosts of *C. flavipes* are forced into hibernation due to cold weather, it overwinters in the cocoon stage and also as larvae in the diapausing host larvae (Mohyuddin, 1971).

However, the developmental time of *C. flavipes* in aestivating & non-aestivating larvae was not different, suggesting that the endocrine of the host did not induce diapause in *C. flavipes*. The result of this study suggests that *C. flavipes* must locate non aestivating larvae in wild host plants in order to survive during off season (Overholt *et al.*, 1997).

Mating experiments revealed that all-possible combination of male and female of the three species, which belong to *Cotesia flavipes* complex, interbreed in the laboratory. However, the only crosses that produce female offsprings are the monogametic cross and cross between male *Cotesia sesamiae* and female *Cotesia Chillonis*. Crosses between *C. sessamiae* and *C. flavipes* did not produce female progeny. Indiscrimination of interspecific mating in the field could theoretically contribute to failure in classical biocontrol programs. However, gregarious parasitoids mate with their sibling soon after emergence. Investigation on male-female attraction demonstrated that *C. flavipes* males are attracted to conspecific females, but not to

females of other two species, suggesting that opportunity for interbreeding in nature would be rare (Ngi-Song and Overholt, 1997).

2.3.3 Host Location and Parasitism by *C. flavipes*

Studies have shown that chemical compounds (mainly volatile), termed as infochemicals convey information on the likely presence of suitable hosts to parasitoids (Dick and Sabelis, 1988). Infochemical is any chemical that, in the natural context, conveys information in an interaction between two individuals, evoking in the receiver a behavioral or physiological response. Chemical information can originate from the herbivore, from its food, from organisms associated with herbivore presence or from interactions between these resources (Vet and Dick, 1992).

Plants provide information to foraging parasitoids in the form of volatile chemical cues, which aids parasitoids locate their hosts at some distance. In single choice olfactometric test *C. flavipes* was attracted to 8-10 week old undamaged maize sorghum and Napir grass. However, volatiles from undamaged plants are poor indicators of herbivore presence to the foraging parasitoid (Vet and Dick, 1992). More useful for parasitoid host selections are those volatiles produced by plants in defense to herbivore attack, which are commonly termed, as herbivore induced synomones (Turling *et al.*, 1995).

Cotesia flavipes was reported to be more attracted to volatiles emanating from maize, sorghum and napir grass infested with larvae of *C. partellus*, *C. orichalcociliellus*, *S. calamistis* and *B. fusca* than uninfected plants (Potting *et al.*, 1995; Ngi- Song *et al.*, 1996).

Having successfully located a suitable habitat the parasitoids must then find their hosts. A number of parasitoids utilize Kairomones to locate the hosts (Rabb *et al.*, 1984). Cues from host by-products such as frass, or silk are generally the most reliable source of information on the presence, identity, availability and suitability of the host to the foraging parasitoid. Although these cues are highly reliable they are limited by their low detectability because they are generally produced in small amounts and are not very volatile. Once *C. flavipes* has found the habitat of its host, it locates the host by using the larval frass, caterpillar regurgitate and holes in the plant stem (Potting *et al.*, 1997a).

In a dual choice test, *C. flavipes* responded more strongly to frass than to stem borers (Potting *et al.*, 1995). Ngi-Song and Overholt (1997) have also reported that *C. flavipes* and *C. sessamiae* can't use host frass to discriminate among *C. partellus* and *B. fusca* infesting a maize plant. From this study there was no evidence that *C. flavipes* discriminated herbivore species that were suitable or unsuitable for successful reproduction.

After locating the exit hole of the stem borer tunnel, where larval frass has accumulated, the parasitoid female tries to enter the stem borer tunnel. This can take a long time because the tunnel is often blocked by larval frass and the female sometimes has to squeeze through small holes (Potting *et al.*, 1997b; Takasu and Overholt, 1997). It has been suggested that dorso-ventrally flattened body shape, which is typical of the *Cotesia* species belonging to *Cotesia flavipes* complex is morphological adaptation to facilitate this behavior (Walker, 1994). Most larvae are often stung near the head or near the posterior end (Ngi-Song and Overholt, 1997). Time spent inside the tunnel is highly variable and depends on the position of the larvae and the amount of frass inside the tunnel (Potting *et al.*, 1997b).

Attacking a concealed stemborer larvae in the confined space is not only time consuming but also risky for the parasitoid. However, the majority of the female parasitoids parasitize the host before being killed. A female *C. flavipes* needs only a few seconds to lay about 45 eggs into it's host (Potting, 1997).

Potting *et al.* (1995) reported that 31% of foraging *C. flavipes* that entered stem inhabited by *C. partellus* were found dead when the plants were dissected, and suspected that the parasitoid had been killed by biting and spitting of the host. *Chilo partellus* larvae spit when they are attacked by *C. flavipes* and the saliva frequently contaminates the parasitoids. In a laboratory experiment application of saliva of *C. partellus* significantly increased mortality of *C. flavipes* (Takasu and Overholt, 1997). The relatively high mortality risk as each host encounter in combination with the short life span results in a very few host encounters of *C. flavipes* (Potting, 1997).

3. MATERIALS AND METHODS

3.1 Insect collection

Chilo partellus larvae and pupae were collected from Melkasa Agricultural Research Center (MARC) experimental field (Cp Mel) and Ziway farmers' field (Cp Ziw). The Altitude for Melkasa and Ziway are 1550m and 1678m above sea level respectively. The annual maximum and minimum temperature for Melkasa are 28.4⁰C and 14⁰C, while for Ziway they are 27.8⁰C and 13.9⁰C. The average relative humidity for Melkasa and Ziway are 56% and 64% respectively. Maize and sorghum plants, which showed signs of stemborer infestation, were dissected using a knife and larvae of *C. partellus* were collected from the infested plants. Collected larvae were given a fresh piece of maize or sorghum stem and kept in a plastic container (20×10×6cm) covered with fine mesh screen, labeled and taken to the entomology laboratory of MARC. In the laboratory, the larvae collected from both areas were separated into a petri dish of 9cm diameter and 2cm deep and given fresh piece of maize stem.

Cocoons of *C. flavipes* were collected from MARC experimental field and Ziway farmers' field using a soft forceps from infested plants. The collected cocoons were kept in a plastic container (20×10×6cm) labeled and taken to the entomology laboratory of MARC. The laboratory in which the insects were reared had 29⁰C maximum day temperature and 14⁰C minimum day temperature and a relative humidity of 50-60%.

3.2 Rearing of test insects

3.2.1 *Chilo partellus*

The collected larvae from the two geographic populations were reared separately on section of maize stem of approximately 6cm length. In all experiments each stem was changed every two days. The pupae were kept in a 30×30×30cm perspex sleeve cages. Glossy paper stripes

(10×45cm) were each folded longitudinally and placed diagonally in the cages as an oviposition substrate. Two cotton balls soaked with 20% sugar-water solution were placed in small vials (2.3×4.4cm) and put at the corners of the cage to serve as source of food for the moths. The glossy papers were removed daily and checked for egg masses (Emana Getu *Pers. Comm.*)

Glossy papers with mass of eggs were cut into smaller sections and kept in an incubator adjusted to $25\pm 1^{\circ}\text{C}$ and was observed daily till black head formation. New glossy papers were placed in the cages to replace the removed ones. Enough sections of the papers containing approximately 300 black head stage of the same egg batches were placed in rearing plastic containers (20×10×6cm) containing young seedlings of maize as food for the first instar *C. partellus* larvae. The plastic containers were covered with tight fitting lids with fine mesh screen lid to prevent the newly hatched larvae from escaping. The first three instars were fed on leaf whorl for 2-3 weeks. The later instars were fed with maize stems (variety Melkasa-1) from plants at tasseling and silking stage individually in a petri dish.

Adult females of the stemborer were also able to lay eggs on the walls of the rearing cage. To collect the first instar larvae from this location, young maize shoots were placed in a large vial (4×11cm) half filled with water. The young shoots were placed in upright position to simulate the natural condition. These shoots were removed and dissected to remove the larvae. The larvae collected in this way were not used for the experiments, since the exact hatching day could not be easily recorded. However, they were maintained for rearing. Fifteen days old larvae (4th instar) were used in all the experiments. New pupae were removed daily from the rearing petri dishes and separated by the date of collection to give the approximate age of the pupae. Mature pupae were then placed in the oviposition cage a day or two days before the

predicted moth emergence date. The process of rearing was repeated for a number of generations for the two geographic populations (Cp Mel and Cp Ziw) until the end of the experiments.

3.2.2 *Cotesia flavipes*

Cocoons of *C. flavipes* from Melkasa (Cf Mel) and Ziway (Cf Ziw) were kept separately in a 20×20×20cm perspex sleeve cage. As soon as the cocoons turn to black color, two cotton balls soaked with 20% honey-water solution were placed in small vials (2.3×4.4cm) and put in the corners of the cage to serve as food source for the emerging wasps as described by Overholt, 1994b & Potting 1996.

Fourth instar larvae of *C. partellus* were exposed to one day old mated female of *C. flavipes* using a hand stinging method described by Overholt (1994b). Two or three hour before stinging the female wasp was placed under incandescent light to activate them for stinging in the cage. Each host larva was held using soft forceps and offered individually to a female parasitoid. Oviposition of *C. flavipes* females was noticed as the larvae reacted violently when the ovipositor of the parasitoid was inserted. Generally, the whole interaction between the wasp and its host larvae lasted for few seconds. The stinging process continued until the parasitoids lost interest in oviposition or enough number of host larvae were stung. After oviposition each parasitized larva were removed from the cage and kept on a petri dish and given maize stem.

The stem was changed every two days for the stung larvae so that fungal infection was minimized. The parasitized *C. partellus* larvae were observed for cocoon formation daily. The

cocoons were removed and transferred to the perspex sleeve cage for adult emergence. The rearing process continued up to the end of the experiments.

3.3 Laboratory Bioassays

Developmental time, longevity and realized fecundity of *C. flavipes* were studied in incubators adjusted to temperature levels of 20⁰C, 25⁰C, 28⁰C and 30⁰C. For suitability test only 28⁰C was used. All the experiments were studied under 12L: 12D photo period and 50-60% relative humidity. For all experiments one day old mated females of Cf Mel and Cf Ziw were used to parasitize fourth instar Cp Mel and Cp Ziw using a hand stinging method described by Overholt (1994b). As a result there are four population interactions (four treatments). Each stung larva was placed separately in a petri dish (9cm diameter and 2cm deep). Observations were made daily until cocoon formation, host death or pupation. For all stung larvae maize stem was changed once every two days until cocoon formation or death of the larvae. For suitability test and developmental time ten test insects were used for each treatment and the whole experiment was replicated four times. Hence, a total of 160 insects (4 treatments ×10 test insects ×4 replication) were used.

3.3.1 Suitability Test

Cocoons from each petri dish were transferred into glass vials covered with nylon organdy to avoid escaping of adult wasps. Date of exposure, date of cocoon formation, date of adult emergence, number of larvae died inside the host, number of progeny and sex of progeny were recorded. To count number of larvae which died inside the host, the stung larvae were dissected using a dissecting blade one day after cocoon formation. Counting was done under microscope. Sexes of progeny were discriminated using the length of the antennae. The experiment was layed in a complete randomized design (CRD).

3.3.2 Developmental Time

As mentioned for suitability test, cocoons from each petri dish were transferred into glass vial covered with nylon organdy to avoid escaping of adult wasps. Date of exposure, date of cocoon formation and date of adult emergence were recorded. The experiment was layed in a complete randomized design (CRD).

3.3.3 Realized Fecundity

When the cocoons described under 3.3 (laboratory bioassay) turned to black they were transferred to a 20×20×20cm perspex sleeve cage with 20% honey water-solution. After emergence, the adults were allowed to mate for about 12 hours. Then 30 adult females from each treatment and temperature regime were randomly selected and provided with a 4th instar larvae of *C. partellus*. Stung larvae were placed in a petri dish provided with a maize stem, and placed back in the temperature regime. After stinging, *C. flavipes* female were kept individually in a 2.5×7cm glass vial covered with nylon organdy. A cotton ball soaked with 20% honey-water solution was placed in each vial. Each female parent was provided with a *C. partellus* larvae twice a day until her death. After cocoon formation for all stung larvae per female lifetime, the stung larvae were dissected and the parasitoid larvae, which failed to develop to cocoons (dead) were counted under microscope. Number of stung larvae per female, total number of progeny produced from all stung larvae per female, sex ratio of the total progenies and dead cocoons from all the stung larvae per female were also recorded. The experiment was layed in a complete randomized design (CRD).

3.3.4 Adult longevity of *C. flavipes*

When the cocoons described under 3.3 (laboratory bioassay) turned to black they were transferred to a 20×20×20cm perspex sleeve cage with 20% honey water-solution. Five to six hour after emergence of adult wasps, 30 male and 30 female C.f Mel and C.f ZiW were placed individually in 2.5×7cm glass vials covered with nylon organdy to keep the vial aerated and to avoid escaping of the wasp. Vials were then placed back in the temperature regiems. Mortality was recorded twice a day (8 a.m. and 5 p.m.) until the death of all parasitoids. The experiment was layed in a complete randomized design (CRD).

3.4 Data Analysis

Data was analyzed using SPSS software. One way ANOVA was run for suitability test and longevity, whereas General Linear Model (GLM) procedure was used to see the interaction of temperature and population interaction for developmental time and realized fecundity. Where ANOVA is significant means were separated by Tukey's studentized test (HSD).

4. RESULTS

4.1 Suitability test

The ANOVA results on number of dead larvae inside the host, number of dead cocoons, total progeny per host and percent of female progeny per host are shown in Appendix Table 1. Population interaction was significant with respect to the number of dead larvae inside the host ($P < 0.0001$), number of dead cocoons ($P < 0.0001$) and total progeny ($P < 0.0001$). However, population interactions was not significant in respect to percent of female progeny per host ($P = 0.653$).

Comparison of the four-population interactions for mean number of dead larvae inside the host, dead cocoon and total progeny is given in Table 4.1. The number of dead larvae inside the host for Cf Ziw reared on Cp Ziw was significantly lower than Cf Ziw reared on Cp Mel and Cf Mel reared on Cp Ziw. Cf Mel had significantly lower number of dead larvae inside the host when the host was Cp Mel than Cp Ziw. Significantly higher number of cocoons were dead for Cf Mel reared on Cp Ziw and Cf Ziw reared on Cp Mel. No significant difference in number of dead cocoon and total progeny was observed when *C. flavipes* of both geographic populations were reared on the same geographic population of *C. partellus*. Significantly higher number of total progeny was found for Cf Mel reared on Cp Mel and Cf Ziw reared on Cp Ziw than the other two population interactions.

Table 4.1. Mean number of dead parasitoid larvae inside its host larvae, dead cocoons and total emerged progeny under variable population interactions.

Population type	Dead larvae	Dead cocoon	Total progeny
Cf Ziw Cp Ziw	0.35 ± 0.1a	1.27±0.23a	33.37±1.1a
Cf Mel Cp Mel	0.77 ± 2.38ab	2.65 ± 0.67a	35.87±2.1a
Cf Ziw Cp Mel	1.52 ± 0.27b	7.62 ± 0.84b	24.05±1.37b
Cf Mel Cp Ziw	2.62 ± 0.27c	9.77 ± 0.57b	20.37±0.94b

Mean (means±SE) followed by the same letter within a column are not significantly different from each other, $\alpha=5\%$ (HSD).

Cf Ziw Cp Ziw = *C. flavipes* Ziway reared on *C. partellus* Ziway

Cf Mel Cp Mel = *C. flavipes* Melkasa reared on *C. partellus* Ziway

Cf Ziw Cp Mel = *C. flavipes* Ziway reared on *C. partellus* Melkasa

Cf Mel Cp Ziw = *C. flavipes* Melkasa reared on *C. partellus* Ziway

4.2 Developmental Time

The ANOVA results for the developmental time are given in Appendix Table 2, 3 & 4. Egg to cocoon developmental time was significantly affected by temperature ($P < 0.0001$), population interaction ($P < 0.0001$) and the interaction of temperature and population ($P < 0.0001$) (Appendix Table 2). Similar results were obtained for cocoon to adult developmental time (Appendix Table 3) and egg to adult developmental time (Appendix Table 4).

Comparison of population interactions and level of temperatures for mean number of days taken from egg to cocoon developmental time is given in Table 4.2. Cf Ziw reared on both *C. partellus* populations at 20⁰c had significantly shorter egg to cocoon developmental period than Cf Mel reared on both *C. partellus* populations at the same temperature. At 25⁰C, Cf Ziw has shown a significant variation when the host was

from Ziway or Melkasa. When the host was Cp Ziw, it had the shortest developmental time of all the interactions and temperature regimes (9.90 days) but when the host was Cp Mel the developmental time significantly increased to 12.53 days. Cf Ziw reared on Cp Ziw at 28⁰C had significantly shorter egg to cocoon developmental time than the other population interactions at 28⁰C. At 30⁰C Cf Ziw reared on Cp Mel had the largest number of days for egg to cocoon developmental time than the rest of the interactions at the same temperature.

Egg to cocoon developmental time was significantly longer at 20⁰C for all population interactions than the rest of the temperature regimes. The longest average egg to cocoon developmental time recorded was 19.0 days for Cf Mel reared on Cp Ziw, while the shortest average egg to cocoon developmental period recorded was 9.9 days at 25⁰C for Cf Ziw reared on Cp Ziw. For Cf Ziw reared on Cp Ziw, only 20⁰C significantly affect the egg to cocoon developmental time while developmental time for egg to cocoon at temperatures of 25⁰C, 28⁰C, and 30⁰C are not significantly different from each other.

Table 4.2 Mean number of days required by *C. flavipes* to develop from egg to cocoon on its host at different temperature and population interaction

Population type	Temperatures			
	20 ⁰ C	25 ⁰ C	28 ⁰ C	30 ⁰ C
Cf Mel Cp Mel	18.68 ± 0.2a(a)	12.53 ± 0.11a(b)	12.08 ± 0.22a(b)	10.38 ± 0.14a(c)
Cf Mel Cp Ziw	18.68 ± 0.2a(a)	11.78 ± 0.07b(b)	12.05 ± 0.06a(b)	10.20 ± 0.06a(c)
Cf Ziw Cp Ziw	17.25 ± 0.08b(a)	9.90 ± 0.12c(b)	10.18 ± 0.09b(b)	10.20 ± 0.06a(b)
Cf Ziw Cp Mel	17.25 ± 0.08b(a)	12.53 ± 0.11a(b)	11.08 ± 0.04c(c)	12.25 ± 0.08b(b)

Means (mean days±SE) followed by (a) the same letter within a column (b) the same letter (in bracket) across a row are not significantly different from each other, $\alpha=5\%$ (HSD).

Cf Mel Cp Mel = *C. flavipes* Melkasa reared on *C. partellus* Melkasa

Cf Mel Cp Ziw = *C. flavipes* Melkasa reared on *C. partellus* Ziway

Cf Ziw Cp Ziw = *C. flavipes* Ziway reared on *C. partellus* Ziway

Cf Ziw Cp Mel = *C. flavipes* Ziway reared on *C. partellus* Ziway

Comparison of population interactions and level of temperatures for mean number of days taken from cocoon to adult is given in Table 4.3. At 20⁰C Cf Mel reared on the two *C. Partellus* populations had the largest cocoon to adult developmental time than Cf Ziw reared on both population of *C. partellus*. At 28⁰C Cf Ziw reared on both *C. partellus* populations had the largest cocoon to adult developmental time than Cf Mel reared on both *C. partellus* populations. The longest cocoon to adult developmental time was for Cf Mel reared on Cp Mel at 20⁰C (11.58 days) and the shortest was For Cf Ziw reared on Cp Mel at 30⁰C (4.93 days).

Table 4.3 Mean number of days required by *C. flavipes* to develop from cocoon to adult at different temperature and population interaction

Population type	Temperatures			
	20 ⁰ C	25 ⁰ C	28 ⁰ C	30 ⁰ C
Cf Mel Cp Mel	11.58±0.2a(a)	6.50±0.08a(b)	5.48±0.09a(c)	5.83±0.08a(c)
Cf Mel Cp Ziw	11.15±0.09a(a)	7.98±0.06b(b)	5.25±0.07a(c)	5.93±0.05a(d)
Cf Ziw Cp Ziw	10.28±0.07b(a)	8.63±0.1c(b)	6.03±0.02b(c)	5.93±0.05a(c)
Cf Ziw Cp Mel	9.78±0.13c(a)	6.50±0.08a(b)	5.98±0.02b(c)	4.93±0.04b(d)

Means (mean days±SE) followed by (a) the same letter within a column (b) the same letter (in bracket) across a row are not significantly different from each other, $\alpha= 5%$ (HSD).

Cf Mel Cp Mel = *C. flavipes* Melkasa reared on *C. partellus* Melkasa

Cf Mel Cp Ziw = *C. flavipes* Melkasa reared on *C. partellus* Ziway

Cf Ziw Cp Ziw = *C. flavipes* Ziway reared on *C. partellus* Ziway

Cf Ziw Cp Mel = *C. flavipes* Ziway reared on *C. partellus* Ziway

Comparison of population interactions and level of temperatures for mean number of days taken from egg to adult is given in table Table 4.4. The egg to adult developmental time ranged from 30.25 days at 20⁰C for Cf Mel reared on Cp Mel to 16.13 days at 30⁰C, for Cf Ziw reared on Cp Ziw and Cf Mel reared on Cp Ziw. The egg to adult developmental time for Cf Ziw reared on both *C. partellus* population at 20⁰C was significantly shorter than Cf Mel reared on both *C. partellus* populations at the same temperature regime.

For Cf Mel reared on both *C. partellus* populations egg to adult developmental time significantly decreased as the temperature increased from 20⁰C -30⁰C. For Cf Ziw reared on both *C. partellus* populations the egg to adult developmental time significantly increased up to 28⁰C. However, there was no significant difference between 28⁰C and 30⁰C.

Table 4.4 Mean number of days required by *C. flavipes* to develop from egg to adult at different temperature and population interaction

Population type	Temperatures			
	20 ⁰ C	25 ⁰ C	28 ⁰ C	30 ⁰ C
Cf Mel Cp Mel	30.25 ± 0.2a(a)	19.02 ± 0.07a(b)	17.55 ± 0.25a(c)	16.20 ± 0.07a(d)
Cf Mel Cp Ziw	30.15 ± 0.17a(a)	19.75 ± 0.07b(b)	17.30 ± 0.07a(c)	16.13 ± 0.06a(d)
Cf Ziw Cp Ziw	27.7 ± 0.94b(a)	18.50 ± 0.1a(b)	16.20 ± 0.08b(c)	16.13 ± 0.06a(d)
Cf Ziw Cp Mel	27.03 ± 0.15c(a)	19.02 ± 0.07a(b)	17.05 ± 0.03a(c)	17.18 ± 0.06b(c)

Means (mean days±SE) followed by (a) the same letter within a column (b) the same letter (in bracket) across a row are not significantly different from each other, $\alpha= 5\%$ (HSD).

Cf Mel Cp Mel = *C. flavipes* Melkasa reared on *C. partellus* Melkasa

Cf Mel Cp Ziw = *C. flavipes* Melkasa reared on *C. partellus* Ziway

Cf Ziw Cp Ziw = *C. flavipes* Ziway reared on *C. partellus* Ziway

Cf Ziw Cp Mel = *C. flavipes* Ziway reared on *C. partellus* Ziway

Figures, 1-3 show the linear regression of developmental times as a function of temperature for all population interactions. The regression equation for egg to cocoon, cocoon to adult and egg to adult developmental time was $Y= 31.83-0.73X$, $Y=21.08-0.53X$ and $Y= 52.90-1.27X$ respectively. Where Y represents the developmental time and X represents the temperature in ⁰C. The R² value for all the developmental times is greater than 0.7.

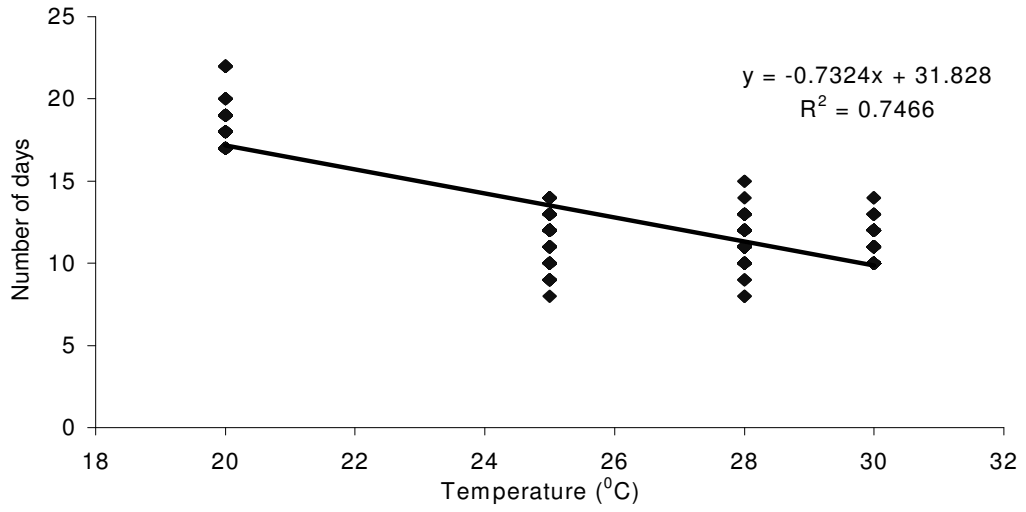


Figure 1, Regression of egg to cocoon developmental time (days) as a function of temperature.

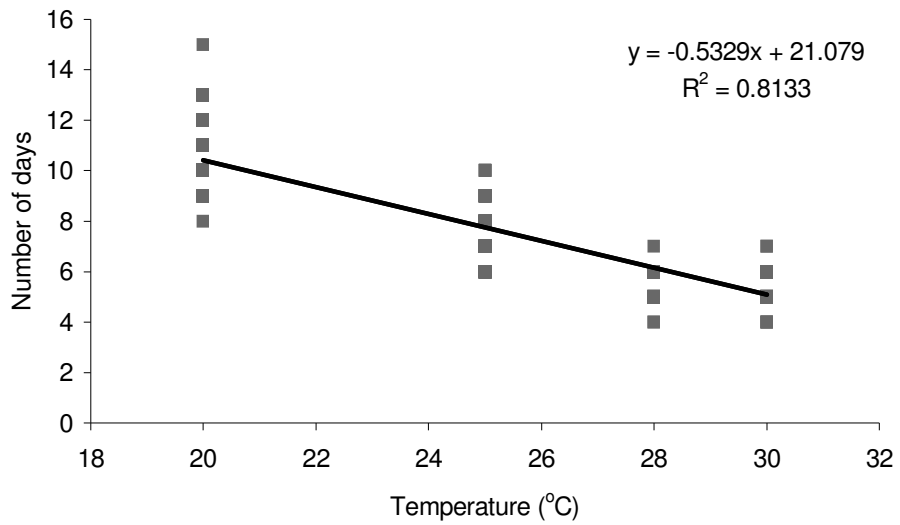


Figure 2, Regression of cocoon to adult developmental time (days) as a function temperature

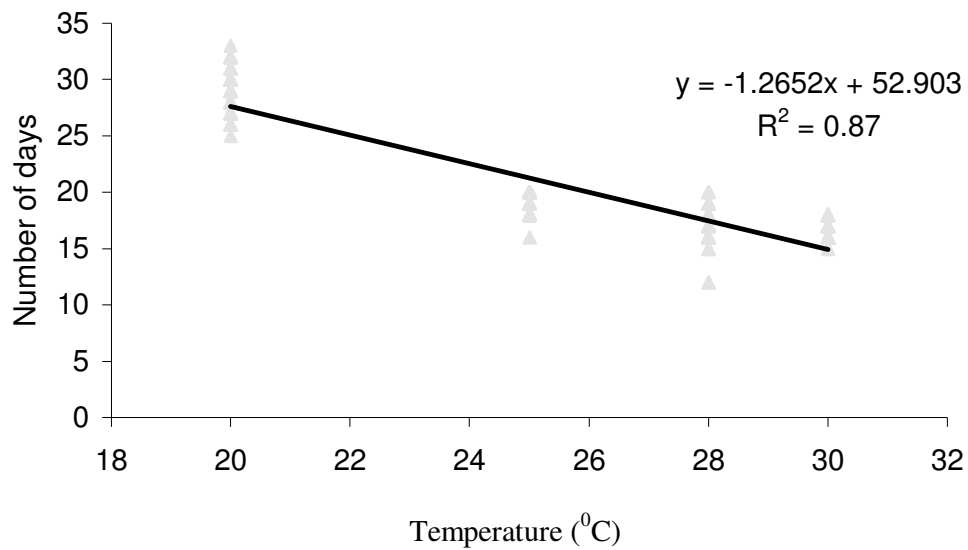


Figure 3, Regression of egg to adult developmental time (days) as a function of temperature

4.3 REALIZED FECUNDITY

Number of parasitoid larvae dead inside its host larvae for all host attacked per female, dead cocoons, number of stung larvae per female were analyzed, because they contribute for the total progeny produced per female from all hosts attacked. The ANOVA outputs for number of dead larvae inside the host, dead cocoon, total progeny, percent female progeny and number of stung larvae per female are given in Appendix Tables 5-9. The number of dead larvae inside the host was significantly affected by population ($P=0.001$) and temperature ($P<0.0001$), but it was not significantly affected by the interaction of temperature and population ($P=0.907$) (Appendix Table 5). The number of dead cocoon inside the host was significantly affected by population ($P<0.0001$), temperature ($P<0.0001$) and the interaction of temperature and population ($P<0.0001$) (Appendix Table 6).

Similar results were obtained for the total progeny produced per female (Appendix Table 7) and percent female progeny (Appendix Table 8). However, the number of larvae stung by a single female of *C. flavipes* was significantly affected by temperature only $P < 0.0001$ (Appendix Table 9).

Comparison of population interaction and temperature for mean number of dead larvae inside the host is given in Table, 4.5. The lowest number of dead larvae inside the host was 0.33 for Cf Ziw on Cp Mel at 20°C and the highest was 5.67 for Cf Mel reared on Cp Ziw at 28°C. At 28°C and 30°C, there was no significant difference in number of dead larvae inside the host among population interactions within the same temperature regime. At 25°C Cf Mel reared on Cp Ziw had the largest number of dead larvae inside the host than other interactions.

For Cf Mel reared on Cp Mel temperatures of 20°C, 25°C and 28°C do not differ in number of larvae found dead inside the host, but when the temperature is at 30°C significantly more number of larvae died inside the host than when it was at 20°C or 25°C. For Cf Mel reared on Cp Ziw at 20°C significantly less number of larvae were found dead inside the host than at 25°C, 28°C and 30°C. For Cf Ziw reared on Cp Ziw temperatures of 20°C, 25°C and 28°C do not differ in number of larvae dead inside the host, but when the temperature was at 30°C significantly more number of larvae died inside the host than it was at 20°C or 25°C.

Comparison of population interactions and temperature for mean number of dead cocoon is given in Table 4.6. The highest mean number of dead cocoon was from Cf Mel reared on Cp Mel (14.37) at 30°C while the smallest was at 20°C (1.43) for the same population interaction. At 20°C there was no significant difference among all population interactions. However, At

30⁰C Cf Mel reared on both *C. partellus* population had significantly large number of dead cocoon than Cf Ziw reared on both *C. partellus* populations at 30⁰C. Cf Mel reared on Cp Mel at 30⁰C had significantly higher number of dead cocoon than other temperature regimes. For Cf Mel reared on Cp Ziw, at 30⁰C and 28⁰C significantly higher number of dead cocoons were recorded than at 20⁰C or 25⁰C.

Comparison of population interaction and temperature for mean number of total progeny per female is given in Table 4.7. The highest total progeny per female was recorded for Cf Mel reared on Cp Mel at 20⁰C (71.73) and the lowest was for Cf Ziw reared on Cp Mel at 30⁰C (15.97). At 28⁰C and 30⁰C, there was no significant difference among population interaction for total progeny per female within the same temperature regimes. Temperature at 20⁰C resulted in significantly higher total progeny produced than other temperature regiems except for Cf Mel reared on Cp Mel.

Mean comparison for percent female for all population interactions within the same temperature regime is given in Table 4.8. The highest percent female progeny was Cf Mel reared on Cp Mel at 25⁰C (78.73%) while the lowest was Cf Mel reared on Cp Ziw at 30⁰C (53.47%). The only significant difference observed among population interactions for percent female progeny was at 25⁰C. At this temperature, Cf Mel reared on Cp Mel had the highest percent female progeny than the rest population interactions at 25⁰C. Cf Mel reared on Cp Mel at 20⁰C, 28⁰C, and 30⁰C did not have significant difference, but Cf Mel reared on Cp Mel at 25⁰C had significantly higher percent female than the other temperatures.

Comparison of temperature levels for mean number of stung larvae is given in Table 4.9. The highest mean number of stung larvae was two while the lowest was one. Only population

interactions at 20⁰C were found to have significantly higher mean number of stung larvae than the other temperature and population interactions.

Table 4.5 Mean number of dead parasitoid larvae in its host larvae at different population interaction and temperature levels

Population type	Temperatures			
	20 ⁰ C	25 ⁰ C	28 ⁰ C	30 ⁰ C
Cf Mel Cp Mel	0.47 ± 0.11a(a)	0.47 ± 0.21a(a)	2.07 ± 0.41a(ab)	5.00 ± 0.84a(b)
Cf Mel Cp Ziw	1.33 ± 0.19b(a)	2.67 ± 0.27b(b)	2.97 ± 0.35a(b)	5.67 ± 1.0a(b)
Cf Ziw Cp Ziw	0.50 ± 0.14abc(a)	0.37 ± 0.12a(a)	1.90 ± 0.38a(ab)	4.87 ± 0.94a(b)
Cf Ziw Cp Mel	0.33 ± 0.09ac(a)	0.37 ± 0.13a(a)	2.00 ± 0.31a(b)	4.60 ± 1.01a(b)

Means (means±SE) followed by (a) the same letter within a column (b) the same letter (in bracket) across a row are not significantly different from each other, α= 5% (HSD).

Cf Mel Cp Mel = *C. flavipes* Melkasa reared on *C. partellus* Melkasa

Cf Mel Cp Ziw = *C. flavipes* Melkasa reared on *C. partellus* Ziway

Cf Ziw Cp Ziw = *C. flavipes* Ziway reared on *C. partellus* Ziway

Cf Ziw Cp Mel = *C. flavipes* Ziway reared on *C. partellus* Ziway

Table 4.6 Mean number of dead cocoons at different population interaction and temperature levels

Population type	Temperatures			
	20 ⁰ C	25 ⁰ C	28 ⁰ C	30 ⁰ C
Cf Mel Cp Mel	1.43 ± 0.25a(a)	1.93 ± 0.4a(a)	4.27 ± 0.85a(a)	14.37 ± 1.50a(a)
Cf Mel Cp Ziw	1.83 ± 0.29a(a)	7.07 ± 0.52b(b)	9.77 ± 0.64b(c)	13.20 ± 1.22a(c)
Cf Ziw Cp Ziw	1.57 ± 0.27a(a)	1.57 ± 0.27a(a)	4.43 ± 0.77a(ab)	5.33 ± 1.08b(b)
Cf Ziw Cp Mel	1.70 ± 0.28a(a)	1.90 ± 0.56a(a)	7.33 ± 0.85ab(b)	5.67 ± 1.14b(ab)

Means (means±SE) followed by (a) the same letter within a column (b) the same letter (in bracket) across a row are not significantly different from each other, α= 5% (HSD).

Cf Mel Cp Mel = *C. flavipes* Melkasa reared on *C. partellus* Melkasa

Cf Mel Cp Ziw = *C. flavipes* Melkasa reared on *C. partellus* Ziway

Cf Ziw Cp Ziw = *C. flavipes* Ziway reared on *C. partellus* Ziway

Cf Ziw Cp Mel = *C. flavipes* Ziway reared on *C. partellus* Ziway

Table 4.7 Mean number of total progeny produced per female at different population interaction and temperature levels

Population type	Temperatures			
	20 ⁰ C	25 ⁰ C	28 ⁰ C	30 ⁰ C
Cf Mel Cp Mel	71.13 ± 3.90a(a)	65.20 ± 5.42a(a)	40.17 ± 2.78a(b)	20.70 ± 1.88a(c)
Cf Mel Cp Ziw	49.60 ± 3.20b(a)	28.80 ± 2.49b(b)	26.83 ± 1.95a(b)	18.70 ± 1.01a(b)
Cf Ziw Cp Ziw	67.57 ± 5.14a(a)	49.33 ± 3.75ac(b)	37.70 ± 2.87a(c)	25.97 ± 1.24a(c)
Cf Ziw Cp Mel	67.07 ± 4.69a(a)	42.87 ± 4.10bc(b)	31.77 ± 3.23a(c)	15.97 ± 1.08a(c)

Means (means±SE) followed by (a) the same letter within a column (b) the same letter (in bracket) across a row are not significantly different from each other, α= 5% (HSD).

Cf Mel Cp Mel = *C. flavipes* Melkasa reared on *C. partellus* Melkasa

Cf Mel Cp Ziw = *C. flavipes* Melkasa reared on *C. partellus* Ziway

Cf Ziw Cp Ziw = *C. flavipes* Ziway reared on *C. partellus* Ziway

Cf Ziw Cp Mel = *C. flavipes* Ziway reared on *C. partellus* Ziway

Table 4.8 Mean percent female progeny produced per female *C. flavipes* at different population interaction and temperature levels.

Population type	Temperatures			
	20 ⁰ C	25 ⁰ C	28 ⁰ C	30 ⁰ C
Cf Mel Cp Mel	59.56 ± 1.29a(a)	78.73 ± 1.50a(b)	61.13 ± 2.37a(a)	58.13 ± 3.47a(a)
Cf Mel Cp Ziw	62.70 ± 0.94a(a)	57.99 ± 1.30b(a)	62.43 ± 1.17a(a)	53.47 ± 1.91a(a)
Cf Ziw Cp Ziw	62.24 ± 1.54a(a)	65.08 ± 1.33b(a)	64.19 ± 1.44a(a)	56.72 ± 1.43a(a)
Cf Ziw Cp Mel	65.36 ± 1.74a(a)	67.60 ± 2.65b(a)	68.31 ± 2.30a(a)	54.68 ± 3.64a(a)

Means (means±SE) followed by (a) the same letter within a column (b) the same letter (in bracket) across a row are not significantly different from each other, α= 5% (HSD).

Cf Mel Cp Mel = *C. flavipes* Melkasa reared on *C. partellus* Melkasa

Cf Mel Cp Ziw = *C. flavipes* Melkasa reared on *C. partellus* Ziway

Cf Ziw Cp Ziw = *C. flavipes* Ziway reared on *C. partellus* Ziway

Cf Ziw Cp Mel = *C. flavipes* Ziway reared on *C. partellus* Ziway

Table 4.9 Mean number of stung larvae per female *C. flavipes* at different population interaction and temperature levels.

Population type	Temperatures			
	20 ⁰ C	25 ⁰ C	28 ⁰ C	30 ⁰ C
Cf Mel Cp Mel	2.03 ± 0.14a	1.40 ± 0.12b	1.13 ± 0.06b	1.0 ± 0.00b
Cf Mel Cp Ziw	2.03 ± 0.14a	1.37 ± 0.10b	1.17 ± 0.07b	1.0 ± 0.00b
Cf Ziw Cp Ziw	2.07 ± 0.13a	1.43 ± 0.09b	1.17 ± 0.07b	1.0 ± 0.00b
Cf Ziw Cp Mel	2.13 ± 0.14a	1.43 ± 0.09b	1.17 ± 0.07b	1.0 ± 0.00b

Means (means±SE) followed by the same letter across a row of population interaction are not significantly different from each other, $\alpha=5\%$ (HSD).

Cf Mel Cp Mel = *C. flavipes* Melkasa reared on *C. partellus* Melkasa

Cf Mel Cp Ziw = *C. flavipes* Melkasa reared on *C. partellus* Ziway

Cf Ziw Cp Ziw = *C. flavipes* Ziway reared on *C. partellus* Ziway

Cf Ziw Cp Mel = *C. flavipes* Ziway reared on *C. partellus* Ziway

4.4. Longevity

The ANOVA results for the effect of temperature, population interaction and sex on longevity of *C. flavipes* is given in Appendix Table 10. The longevity of adult *C. flavipes* was significantly affected by temperature ($P < 0.0001$) and sex ($P = 0.001$), but not by population interactions ($P = 0.562$).

The effect of temperature on longevity of *C. flavipes* is given in Table 4.10. The highest mean longevity of *C. flavipes* went upto to 97 hours for Cf Ziw reared on Cp Mel male at 20⁰C, while the lowest was 18.8 hours for Cf Mel reared on Cp Mel male at 30⁰C. Comparison of the longevity between males and females of the same population interaction is given in Table 4.10. Females of Cf Mel reard on both *C. partellus* populations at 28⁰C lived significantly longer than males of the same population interaction at the same temperature. Females of Cf

Ziw reared on both *C. partellus* population at 20⁰C had lived significantly longer than males of the same population interactions at the same temperatures.

Table 4.10 Mean number of adult longevity of *C. flavipes* in hours at different population interaction and temperature levels

		Temperatures			
Population type	Sex	20 ⁰ C	25 ⁰ C	28 ⁰ C	30 ⁰ C
Cf Mel Cp Mel	M	79.0 ± 3.13a	44.9 ± 2.06b	43.7 ± 3.32b(a)	18.8 ± 1.35c
	F	77.9 ± 2.64a	51.3 ± 1.90b	58.9 ± 2.05b(b)	21.4 ± 1.65c
Cf Mel Cp Ziw	M	79.0 ± 3.13a	44.73 ± 2.06b	43.53 ± 3.32b(a)	18.8 ± 1.35c
	F	76.3 ± 2.51a	51.3 ± 1.90b	58.23 ± 2.20b(b)	21.7 ± 1.69c
Cf Ziw Cp Ziw	M	78.40 ± 3.84a(a)	61.3 ± 2.02b	36.0 ± 1.23c	19.8 ± 1.25d
	F	97.40 ± 3.69a(b)	67.5 ± 1.37b	33.9 ± 1.19c	20.5 ± 1.24d
Cf Ziw Cp Mel	M	78.1 ± 3.84a(a)	61.3 ± 2.02b	36.0 ± 1.23c	19.8 ± 1.25d
	F	97.4 ± 3.69a(b)	67.5 ± 1.37b	34.3 ± 1.12c	20.1 ± 1.30d

Means (mean hours ± SE) followed by (a) the same letter across a row of population interaction for each sex and (b) the same letter within a column of each temperature for each population interaction are not significantly different from each other, α=5% (HSD).

Cf Mel Cp Mel = *C. flavipes* Melkasa reared on *C. partellus* Melkasa

Cf Mel Cp Ziw = *C. flavipes* Melkasa reared on *C. partellus* Ziway

Cf Ziw Cp Ziw = *C. flavipes* Ziway reared on *C. partellus* Ziway

Cf Ziw Cp Mel = *C. flavipes* Ziway reared on *C. partellus* Ziway

4. DISCUSSION

In the suitability test of *C. flavipes*, the two geographic locations have shown variability in terms of reproductive success when the host insects were different in geographical location. Both geographic populations have shown more reproductive success on the *C. partellus* population of the same geographic location than *C. partellus* from different geographic location. The highest total progeny and lowest dead cocoon were recorded when Cf Ziw reared on Cp Ziw and Cf Mel reared on Cp Mel indicated that the two *C. flavipes* populations are best when the host is from similar geographical area.

Potting *et al.*, (1997a), have also reported the occurrence of variation in terms of total progeny production among different geographical populations of *C. flavipes* that differed in the plant host-complex they were obtained from, but there was no significant difference in a sex ratio of emerged progeny between the populations. The current work has also found variation in total progeny between *C. flavipes* collected from Ziway and Melkasa when the hosts were from different geographical location. But, in the current work the two populations of *C. flavipes* did not differ in host plant complex they were obtained from, they only differ in geographic location. The two geographical areas have different climatic property, Melkasa represents a dry hot area while, Ziway is a humid hot area and these differences in climatic property may have contributed to the variation.

Emana Getu (2001) reported variability in mean number of larvae died inside the host and dead cocoon among five different geographical populations of *C. flavipes* in Ethiopia which were reared on *C. partellus* from Melkasa. The current study has also confirmed variability in mean number of dead larvae inside the host, dead cocoon and total progeny among *C. flavipes*/ *C. partellus* population interactions tested. However, there was no significant

difference in percent female produced, which agrees with the findings of Potting *et al.* (1997a) and Emanu Getu (2001).

Difference in physiological compatibility between different populations of *C. flavipes* may reflect a difference in efficacy of the parasitoid population, but it may also reflect differences in immune response between different populations of *C. partellus*. However, since *C. flavipes* is introduced recently intraspecific variation is not expected. Therefore, the most likely reason would be a difference in immune response of the hosts. The higher number of dead larvae inside the host for *C. flavipes* from Melkasa reared on *C. partellus* from Ziway and low number of dead larvae inside the host for *C. flavipes* from Ziway reared on *C. partellus* from Ziway, indicated a difference in immune response of *C. partellus* from Ziway when the parasitoids are from different geographic locations.

Cotesia sessamiae, close relative to *C. flavipes* was unable to develop in *B. fusca* populations of coastal area of Kenya because their eggs are encapsulated, but they successfully develop in other populations of *B. fusca* in Kenya. *Cotesia flavipes* is also unable to develop in most of *B. fusca* populations but it can develop in some populations (Cugala and Omwega, 2001; Emanu Getu 2001; Mattama *et al.*, 2001). This indicates variation in virulence between parasitoid populations and variation in encapsulation ability between host populations.

In studying the developmental time of the two population of *C. flavipes*, it was observed that the two populations of *C. flavipes* had different developmental time at different temperature and even within the same temperature they have shown different developmental time when the hosts were from different geographical locations.

Many authors have reported that the developmental time of insects to be dependent on the temperature of their environment and have determined the optimum temperature requirement of many insects (Vincent *et al.*, 1997; James and Ying Hong, 1998; Kirk *et al.*, 1998). In the current study, the egg to adult developmental time of the two populations of *C. flavipes* was inversely related to the temperature regimes they were exposed to, no matter on which population host they were reared. The largest was at 20⁰C and the shortest was at 30⁰C. This finding agrees with most of the works done on the effect of temperature on the developmental time of *C. flavipes* (Overholt *et al.*, 1994b; Mbapila, 1997; Emanu Getu, 2001).

In the current experiment, the significance of the interaction of the population interaction and temperature in the developmental time of *C. flavipes* indicated that developmental time of *C. flavipes* varied with temperatures and in different geographic population of *C. partellus*. Emanu Getu (2001), has also studied the developmental time of Indian and Pakistan population under different conditions than used in the current study and suggested that the developmental time of *C. flavipes* varied with temperature, relative humidity and geographic population.

The establishment and efficiency of parasitoids depend on the population increase which is the function of length of generation time, survival and fecundity (Rosh, 1990). If the two populations of *C. flavipes* tested are to be released in a cooler area which has a temperature of less than or equal to 20⁰C, population growth may be very slow which in turn would reflect low initial parasitism. Emanu Getu (2001) has suggested the slow growth rates of *C. flavipes* at lower temperatures to be the reason why establishment of *C. flavipes* takes some times to be realized in some areas.

At 20⁰C, Cf Ziw had the shortest egg to adult developmental time than Cf Mel, which suggests its better adaptation to cooler areas. The largest developmental time of Cf Ziw on C.p Mel at 30⁰C than the other interactions at this temperature, the largest developmental time of Cf Mel reared on Cp Ziw at 25⁰C than the other interactions at this temperature and the shortest developmental time of Cf Ziw at 28⁰C than other interactions at this temperature suggested that temperature acted differently for different population interactions. Therefore, it may be useful to select populations based on the climate where they are intended for release.

In studying the realized fecundity of the two populations of *C. flavipes* at different temperature regimes, number of dead larvae inside the host at 28⁰C for all population interaction was not significantly different from each other. This result deviates from the suitability test result, which was conducted at 28⁰C. In the suitability test result, there was a significant difference in a number of dead larvae inside the host for population interactions (see Table, 4.1.). The deviation in the two results might be due to differences in experimental procedures. In the suitability test, the female *C. flavipes* tested were reared in the laboratory at room temperature (29⁰C maximum day temperatures and 14⁰C minimum day temperature), but in the realized fecundity female *C. flavipes* tested were reared at 28⁰C constant temperature.

At higher temperatures, more number of dead cocoons and more number of dead larvae were found inside the host than at lower temperatures. This suggests that temperature is an important mortality factor for the immature stages of *C. flavipes*. Cf Ziw reared on Cp Ziw and Cf Ziw reared on Cp Mel significantly smaller numbers of cocoons were found dead than Cf Mel reared on Cp Ziw at 25⁰C. Cf Ziw on both populations of *C. partellus* at 30⁰C had also significantly smaller number of dead cocoons than Cf Mel on both *C. partellus* populations at

the same temperature. This suggested that temperature acted differently for mortality at cocoon stage for both populations of *C. flavipes*.

The realized fecundity measured as the total number of progeny allocated to all attacked hosts was greater at 20⁰C than it was at 30⁰C. This suggested that temperature is an important physical factor for total progeny production of *C. flavipes* populations. Cf Mel reared on Cp Ziw at 20⁰C had the smallest total progeny production than the other population interactions at the same temperature regimes. At 25⁰C Cf Mel reared on Cp Mel had significantly higher total progeny than Cf Mel reared on Cp Ziw and Cf Ziw reared on Cp Mel. This suggested that temperature had different effects on different population interactions.

The total number of progeny recorded for all population interactions was 16-72, which is quite similar to 23-70 reported by Emanu Getu (2001). But, it is very different from that by Potting (1996) who reported realized fecundity measured as the total number of progeny per female to be 155. The explanation for this deviation could be differences in experimental procedures, he dissected all parasitized larvae per female to count the progeny of the parasite. Lower longevity of female *C. flavipes* in the current work and higher relative humidity (70-80%) in Potting (1996) can also be the reasons for the deviation.

Total number of progeny at all temperature regimes for all population interactions is found to be female biased, the highest being 78.7%. A female biased progenies of *C. flavipes* have been reported by other authors (Mohuddin, 1971; Overholt, 1994b; Potting, 1997b; Emanu Getu, 2001). Waage (1990) reported that total progeny and percent female are important criteria for selection of a biological control agent. The higher percent female progeny of the

two populations of *C. flavipes* at all temperatures tested indicated their effectiveness as a bio-control agent in areas, which has similar environmental conditions.

Females of *C. flavipes* only stung one time at 30°C. The number of stung larvae per female was found to be higher (more than two) at 20°C than the rest of temperature regimes. The lifetime host encounter of *C. flavipes* females at 20°C is in agreement with Mohuddin (1971) and Smith and Widenmann (1993), who reported 2-3 encountered host per female lifetime.

The longevity of adult *C. flavipes* is longer at 20°C than it is at 30°C for all population interactions. The higher number of stung larvae per female and the higher number of total progeny to all attacked hosts per female at lower temperatures than at higher temperatures might be due to a relatively longer adult longevity at lower temperatures than at higher temperatures.

Potting (1996) indicated that lower temperatures, humid environment and the availability of honey as food increased the life span of adult *C. flavipes*. He reported the average life span of fed females in a humid environment at 22°C to be 6.1 days whereas fed parasitoids at dry environment at 31°C lived on average for 8 hours. In the current study, longevity for both populations at 20°C was much more less than that reported by Potting (1996) at 22°C. The explanation for this could be that, the level of relative humidity was much higher in the Potting's experiment (70-80%) than in the current work (50-60%). Similarly at 30°C the current experiment had a longer longevity than reported by Potting (1996) at 31°C, this again could be due to population difference or differences in relative humidity, he tested the longevity at 35-40% RH which is lower than the current experiment (50-60% RH). Females of *C. flavipes* from Melkasa lived longer than males at 28°C and females of Ziway *C. flavipes*

lived longer than males at 20⁰C. Emanu Getu (2001) has also reported that sex significantly affected the longevity of adult *C. flavipes*.

6. CONCLUSION AND RECOMMENDATIONS

6.1 Conclusion

From this work, it can be concluded that the interactive effect of temperature and population was seen in most of the life history parameters of *C. flavipes*, which are bases for successful establishment and efficient parasitism. The practical application of studying variation in reproductive success is to select the appropriate parasitoid population. If inundative release of both *C. flavipes* populations studied is to be made it should be released on the geographic area from which it originated. This study showed the need for selecting populations based on their reproductive success and the climate they are intended for release. In cool or hot environments it may be useful to identify populations better adapted to these environments. Cf Ziw seems to be adapted to cooler areas than Cf Mel. Hence, if *C. flavipes* is to be released inundatively these factors should be considered to optimize the release strategy.

The current findings are based on constant relative humidity (50-60%) for all temperatures tested. However, temperature exerts a more severe limiting effect on organisms when there is either very high moisture or very low than when there are moderate conditions (Odum, 1983). Therefore, if the two *C. flavipes* populations tested are to be released under diverse field conditions, the interaction of different relative humidity with temperature and population should also be considered.

6.2 Recommendations

- ◆ The two geographic populations should be studied genetically, physiologically and ecologically to understand the mechanisms involved in variability.
- ◆ If releases of *C. flavipes* are planned for cooler areas, Cf Ziw is recommended.
- ◆ A temperature of 25⁰c is recommended for laboratory rearing. Rearing the two populations on *C. partellus* originated from the same geographic area is also recommended.
- ◆ If inundative releases are to be carried out in Melkasa and Ziway, the parasitoids should be laboratory reared parasitoid of similar population (i.e. for Melkasa Cf Mel and for Ziway Cf Ziw).
- ◆ Studies on other populations of *C. flavipes* and *C. partellus* in Ethiopia under different physical factor which mimic the climate of the area in which the releases is intended to be made should be carried out.

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APPENDICES

Table 1. Analysis of variance for number of dead larvae inside the host, number of dead cocoon, total progeny per host and percent of female progeny.

Variable	Source	DF	SS	MS	F	P
Number of dead larvae inside the host	Pop	3	119.319	39.773	18.388	<0.0001
Number of dead cocoon	Pop	3	1946.019	648.673	40.846	<0.0001
Total progeny	Pop	3	6557.919	2185.973	26.025	<0.0001
Percent female	Pop	3	317.660	105.887	0.544	0.653

Pop = population interaction

Table 2. Analysis of variance for egg to cocoon developmental time

Source	DF	Type III SS	MS	F value	P
Temp	3	5673.450	1891.15	3318.553	< 0.0001
Pop	3	231.963	77.321	135.681	< 0.0001
Temp * Pop	9	262.363	29.151	51.154	< 0.0001

Temp = temperature, Pop = population interaction, Temp * Pop = interaction of temperature and population

Table 3. Analysis of variance for cocoon to adult developmental time

Source	DF	Type III SS	MS	F value	P
Temp	3	2697.238	899.079	2675.371	< 0.0001
Pop	3	78.613	26.204	77.925	< 0.0001
Temp * Pop	9	185.225	20.581	61.241	< 0.0001

Temp = temperature, Pop = population interaction, Temp * Pop = interaction of temperature and population

Table 4. Analysis of variance for egg to adult developmental time

Source	DF	Type III SS	MS	F value	P
Temp	3	15907.680	5302.560	9027.481	< 0.0001
Pop	3	156.892	52.297	89.035	< 0.0001
Temp * Pop	9	276.952	30.772	52.389	< 0.0001

Temp = temperature, Pop = population interaction, Temp * Pop = interaction of temperature and population

Table 5. Analysis of variance for number of dead larvae inside the host

Source	DF	Type III SS	MS	F	P
Pop	3	141.840	47.280	5.777	0.001
Temp	3	143.0956	476.985	58.285	<0.0001
Temp * Pop	9	33.119	3680	.450	0.907

Pop = population interaction, Temp = temperature, Temp * Pop = interaction of temperature and population

Table 6. Analysis of variance for number of dead cocoon

Source	DF	Type III SS	MS	F	P
Pop	3	1502.167	500.722	27.437	<0.0001
Temp	3	4664.567	1554.856	85.197	<0.0001
Temp * Pop	9	1766.900	196.322	10.757	<0.0001

Pop = population interaction, Temp = temperature, Temp * Pop = interaction of temperature and population

Table 7. Analysis of variance for total progeny

Source	DF	TypeIII SS	MS	F	P
Pop	3	1502.167	500.722	27.437	<0.0001
Temp	3	124039.773	41346.591	126.657	<0.0001
Temp * Pop	9	11115.085	1235.009	3.785	<0.0001

Pop = population interaction, Temp = temperature, Temp * Pop = interaction of temperature and population

Table 8. Analysis of variance for percent female progeny

Source	DF	Type III SS	MS	F	P
Pop	3	2058.275	686.092	5.534	0.001
Temp	3	8561.053	2853.684	23.016	<0.0001
Temp * Pop	9	6383.223	709.247	5.720	<0.0001

Pop = population interaction, Temp = temperature, Temp * Pop = interaction of temperature and population

Table 9. Analysis of variance for number of stung larvae per female *C. flavipes*

Source	DF	TypeIII SS	MS	F	P
Pop	3	0.150	0.0050	0.193	0.901
Temp	3	79.517	26.506	102.374	<0.001
Temp * Pop	9	0.167	0.001852	0.072	1

Pop = population interaction, Temp = temperature, Temp * Pop = interaction of temperature and population

Table 10. Analysis of variance for adult longevity of *C. flavipes*

Source	DF	SS	MS	F	P
Temp	3	495228.4	165076.126	768.844	<0.0001
Pop	3	1500.188	500.063	0.683	0.562
Sex	1	8027.267	8027.267	11.105	0.001

Temp = temperature, Pop = population interaction