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Establishment and spread of *Cotesia flavipes* Cameron: Implication on the local population of *Chilo partellus* (Swinhoe) across three agro-ecological zones in Kenya

Esther Adhiambo Abonyo[1*], George Otieno Ongámo[2], Catherine Wanjiru Lukhoba[3], Gideon Nzioka Nyamasyo[4] and Bruno Le Ru Pierre[5]

[1]College of Biological and Physical Sciences, University of Nairobi, P. O. Box 30197 (00100), Nairobi, Kenya. E-mail: e_abonyo@yahoo.com
[2]Noctuid Stem Borer Biodiversity Project, ICIPE, Nairobi, P. O. Box 30772 (00100), Nairobi, Kenya. E-mail: gongamo@uonbi.ac.ke
[3]College of Biological and Physical Sciences, University of Nairobi, P. O. Box 30197 (00100), Nairobi, Kenya. E-mail: clukhoba@uonbi.ac.ke
[4]College of Biological and Physical Sciences, University of Nairobi, P. O. Box 30197 (00100), Nairobi, Kenya. E-mail: gnyamasyo@uonbi.ac.ke
[5]UMR IRD 247 Laboratoire Evolution, Génomes, Comportement et Ecologie, Diversité, Ecologie et Evolution des Insectes Tropicaux, CNRS, 91198 – Gif-sur-Yvette, France and Université de Paris-Sud, 91405 - Orsay, France. E-mail: bleru@icipe.org

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Abstract

Chilo partellus causes yield losses estimated at 73-100% in maize and 88-100% in sorghum in Kenya. *icipe* spearheaded the importation and release of the larval parasitoid, *Cotesia flavipes,* in moist lowland, dry mid-altitude and moist mid-altitude Agro-Ecological Zones (AEZs) of Kenya against *C. partellus.* However, the establishment of *C. flavipes* is only well documented for the moist lowland AEZ. This study was thus carried out in the three AEZs in order to update establishment, spread and impact of the parasitoid on stem borer populations. Maize and sorghum farms were sampled radiating from parasitoid release points up to a distance of 45 km. Percentage infestation and parasitism were analyzed using one factor ANOVA or Kruskal-Wallis test. Results of this study revealed that *C. flavipes* established and spread beyond release sites across the three AEZs. Overall stem borer infestation was 28.9 ± 3.1, 22.5 ± 7.4 and 2.7 ± 0.4% in the moist lowland, dry mid-altitude and moist mid-altitude AEZ, respectively, showing a significant reduction from parasitoid pre-release levels. Stem borer parasitism levels were 36.1 ± 3.0, 25.3 ± 3.3 and 5.5 ± 2.5% in the moist lowland, dry mid-altitude and moist mid-altitude AEZ, respectively, showing a significant increase from parasitoid pre-release levels. *Cotesia flavipes* is steadily suppressing stem borer population in the three AEZs.

Keywords: *Cotesia flavipes, Chilo partellus, Biological control, Infestation, Parasitism*

1. Introduction

Lepidopteran stem borers constitute one of the most important constraints to maize and sorghum production in Sub-Saharan Africa (Brownbridge, 1991; Odindo, 1991; and Schulthess *et al*., 1997). In Kenya and East Africa in general, the most important cereal stem borer pests are the crambids *Chilo partellus* (Swinhoe) and

* *Corresponding author: Esther Adhiambo Abonyo, College of Biological and Physical Sciences, University of Nairobi, P. O. Box, 30197 (00100), Nairobi, Kenya. E-mail: e_abonyo@yahoo.com*

Chilo orichalcociliellus Strand and the noctuids *Busseola fusca* (Fuller) and *Sesamia calamistis* Hampson (Nye, 1960; Bonhof *et al*., 1997; and Overholt *et al*., 2001). These stem borer pests are indigenous to Africa except for *Chilo partellus* which originated from Asia (Tams, 1932; Nye, 1960; and Bleszynski, 1970; and Van Hamburg, 1979). *Chilo partellus* was accidentally introduced in Africa in the 1930s (Tams, 1932) and owing to its excellent colonizing abilities, has since spread to various countries (Duerden, 1953; Nye, 1960; Ingram, 1983; Harris, 1990; and Overholt *et al*., 1994a). In Kenya, *C. partellus* is the most damaging and important pest (Seshu Reddy, 1983; and Overholt *et al*., 1997) since it was first reported in 1950s (Nye, 1960). Cereal yield losses associated with its infestation are estimated to reach as high as 73-100% in maize and 88-100% in sorghum (Seshu Reddy, 1983; and Seshu Reddy and Walker, 1990).

Management of *C. partellus* populations is considered an important step towards increasing maize and sorghum production and has thus been the focus of various management initiatives (Overholt *et al*., 1997). Two classical biological control attempts targeting *C. partellus* have been undertaken in Eastern Africa. The first one was undertaken by the Commonwealth Institute for Biological Control (now International Institute of Biological Control) and it involved introduction of nine parasitoid species from Rawalpindi, Pakistan into Kenya, Uganda and Tanzania. These organisms failed to establish (CIBC, 1968-1972). The second attempt was initiated in Kenya by the International Centre of Insect Physiology and Ecology (*icipe*) Biocontrol Programme in 1991 (Overholt, 1993). Following a foreign exploration in the pest's native range during this attempt, *Cotesia flavipes* was selected from Sindh region of Pakistan and introduced in Kenya (Overholt *et al*., 1994a). The parasitoid was released along the moist lowland AEZ (Kilifi and Kwale counties), in July 1993 (Overholt *et al*., 1994a). Surveys to assess its establishment were carried out later and results showed that *C. flavipes* had established (Overholt *et al*., 1994b and 1997; and Omwega *et al*., 1995). Stem borer parasitism by *C. flavipes* progressively increased reaching levels of 0.03 to 7.1 in 1997 (Overholt *et al*., 1997). These figures were however relatively low and at the time, *C. flavipes* did not appear to be an important mortality factor for stem borers (Overholt *et al*., 1997). A subsequent assessment demonstrated an increase in parasitism to 13% by 1999 (Zhou *et al*., 2001). In dry mid-altitude AEZ, *C. flavipes* was released in experimental fields at three sites (Katumani, Ithookwe and Kiboko) in 1997 / 1998. Post release surveys carried out within the same season and year of release led to recovery of the parasitoid at release fields (Songa *et al*., 2001). However, the time between releases and post release surveys conducted in the region was too short to deduce establishment or measure spread. In moist mid-altitude AEZ, releases were made at three sites in Kisumu in 2000 (Kenya Agricultural and Livestock Research Organization (KALRO) and *icipe*, unpublished data). Further to this, additional releases were made on farmer's fields in dry mid-altitude AEZ (in Masii, Mulutu and Kitui) in 2002 (KALRO and *icipe* unpublished data). No post release surveys have been carried out following the latter two releases.

Maximum pest suppression can only be achieved once the parasitoid colonizes host populations in all suitable habitats and thus spread is an important aspect in parasitoid impact. It was thus projected that a characteristic *C. flavipes* 'equilibrium' density needed to be reached before its impact on stem borer pest population is measured (Overholt *et al*., 1997). However, no additional surveys have been carried out at the coast during the last seventeen years. In this study, plant infestation levels in the three Agro-Ecological Zones (AEZs) *vis a vis* rates of parasitism attained by *C. flavipes* were assessed. Such information is needed in order to fully assess the impact of this biological control agent as a pest management measure.

2. Materials and methods

2.1. Description of study area

This study was carried out in three different AEZs as classified by the Kenya Maize Database Project (Corbett, 1998). Transects along which surveys were conducted ran across moist lowland, dry mid-altitude and moist mid-altitude AEZs in Kenya (Figure 1). These were areas where *C. flavipes* had previously been released.

2.2. Moist lowland agro-ecological zone

Moist lowland AEZ is characterized by diurnal temperatures ranging between 20 and 31°C, receives around 500-1,000 mm of rainfall annually, with most areas situated below 400 masl (Corbett, 1998). The main maize growing season falls between March and August while the short rains come in October with a decrease being observed in January and February. The amount, reliability and duration of rainfall are the main factors limiting agricultural production in this zone. A variety of crops including pulses, industrial and root crops, fruits and cereals are cultivated with maize being the most important food crop.

2.3. Dry mid-altitude agro-ecological zone

In the dry mid-altitude zone, diurnal temperatures range from 14 to 33°C with annual precipitation varying between 300 to 550 mm. This amount is considered low and unreliable (Bernard *et al*., 1989). Long rains are received between March and May while the short rains are received between October and December (Moore, 1979). The region is predominantly semi-arid and lies at an altitude of 700-1,400 masl (Corbett, 1998). Production systems in the area have moved from agropastoral to mixed farming systems (Rocheleau *et al*., 1995). Bananas, sweet potatoes, green grams, cow peas, pigeon peas, pumpkins, beans and maize are the most common food crops grown in the area. Dry forest and savannah ecosystems have also been converted to agricultural lands besides being burnt to produce charcoal (Rocheleau *et al*., 1995).

2.4. Moist mid-altitude agro-ecological zone

This zone is characterized by diurnal temperatures ranging from 13 to 30°C, receives more than 550 mm of rainfall annually with most areas situated between 1,110 and 1,500 masl (Corbett, 1998). There is commercial farming of sugarcane while maize, sorghum and pearl millet are mainly grown as food crops. This area has two maize growing seasons, March to June / July (long rainy season) and September to December (short rainy season).

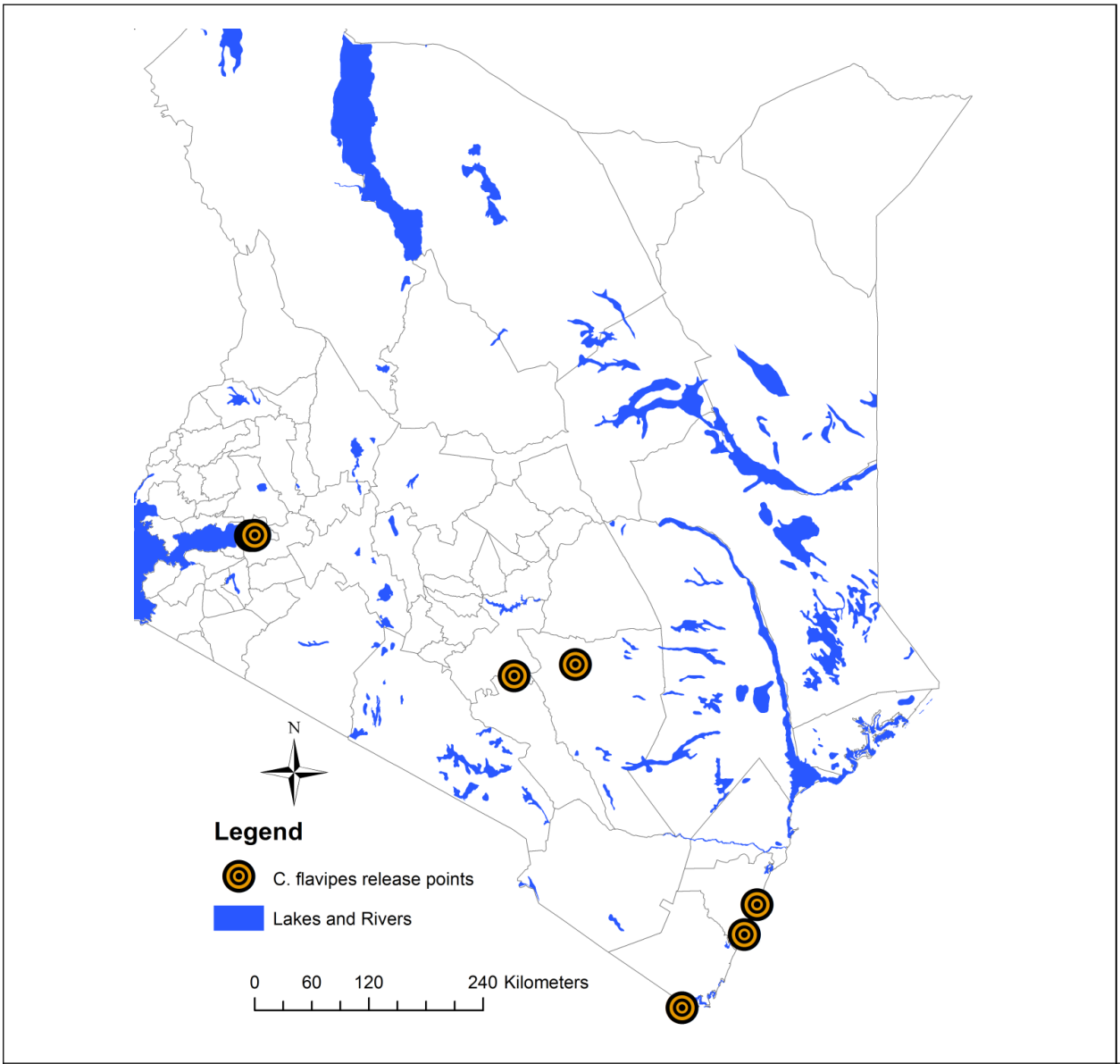

Figure 1: *Cotesia flavipes* release points in three agro-ecological zones in Kenya

2.5. Study design and stem borer sampling protocol

Benchmark sites were marked in farms at which *C. flavipes* had previously been released (Figure 1). Survey farms were subsequently sampled in the four cardinal compass points at every 15, 30 and 45 km (Figure 2). A total of 52, 55 and 42 farms were sampled in moist lowland, dry mid-altitude and moist mid-altitude AEZs respectively. Sampling was done on maize and sorghum farms that had plants at early maturity and mature stages of growth during the long rainy season of 2015 across the three AEZs. In each farm, a total of 100 maize plants were inspected for infestation by stem borers. Ten maize stems which showed symptoms of infestation such as exit holes on stems, frass deposits, window panes on leaves and dead heart were destructively sampled and dissected. Immature stem borer stages were collected, identified and categorized (as small {1st and 2nd instars}, medium, {3rd and 4th instars} and large {5th instars}). Identified larvae were placed individually in glass vials containing artificial diet (Onyango and Ochieng-Odero, 1994) and transported to the laboratory at *icipe* where they were reared at ambient temperatures of 24-25°C and a relative humidity of 55-65%, with a 12:12 light: dark photoperiod. Samples were inspected daily for parasitoid cocoons, pupal development, pupal parasitoid and adult moth emergence. Pupae were transferred into plastic jars lined with wet paper towels. Humidity in the jars was maintained by moistening the soft paper towels once every two days using a few drops of distilled water. Larval parasitoids and adult stem borer moths were identified and recorded.

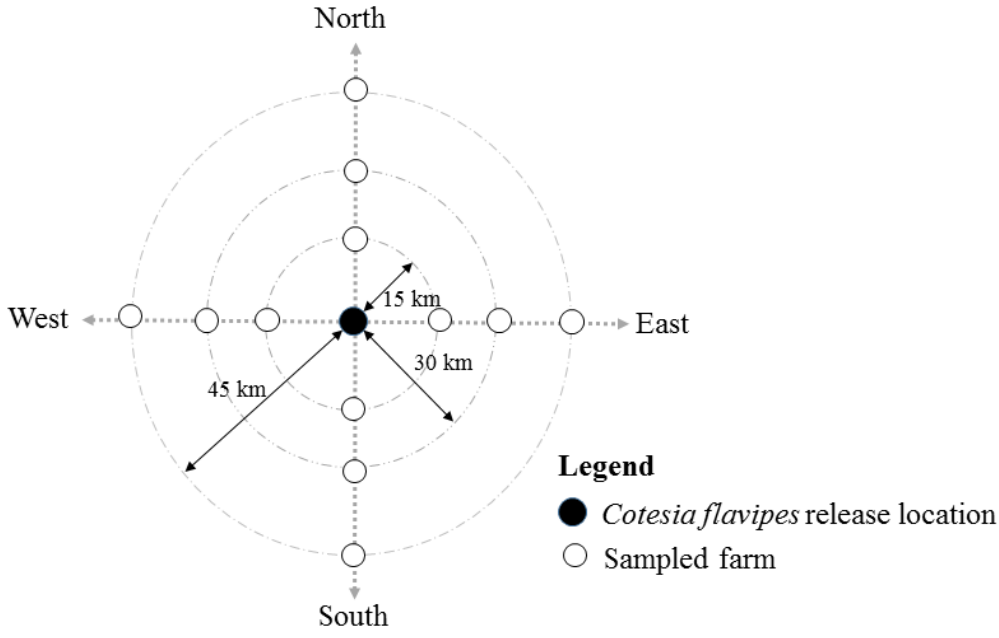

Figure 2: *Cotesia flavipes* release point and sampling transect radiating in four main compass point directions at 15, 30 and 45 km distances

3. Statistical analyses

Farms sampled at predetermined distances along different transects were treated as replicates and resulting data used to estimate means of infestation and parasitism for respective distance intervals. Percentage infestation was computed by expressing the number of infested plants as a percentage of the total plants inspected in respective farms. To estimate larval densities in respective farms, the total number of stem borer larvae collected from dissected plants was expressed as a proportion of the number of infested plants. Parasitoid cocoons that were spun from appropriate larval stages were expressed as a proportion of respective field densities in order to compute percentage parasitism. Percentage infestation, larval density and parasitism were subjected to Shapiro-Wilk test for normality upon which abnormal data were appropriately transformed. Normal data were analyzed using One-Way ANOVA and significantly different means separated using Tukey's HSD test. Data which failed the normality test was subjected to Kruskal-Wallis rank sum test and significantly different means separated using Nemenyi post-hoc test ($p < 0.05$). One sample *t*-test and Wilcoxon rank sum test were

used to compare mean infestation and parasitism levels obtained before and after the release of parasitoids. In order to determine which climatic parameters prevailing in sampled localities highly correlated to plant infestation and stem borer parasitism, data was extracted from BIOCLIM and subjected to multiple regression analysis using the vegan package in R.

4. Results

4.1. Stem borer composition, diversity and larval density

In moist lowland AEZ, stem borer species recovered were *Chilo spp.* (*Chilo* species) and *S. calamistis*. *Chilo spp.* were dominant and constituted 98.5% of total stem borers collected with the exotic *C. partellus* constituting 96.4% while the indigenous *C. orichalcociliellus* constituted 1.7%. *Sesamia calamistis* constituted 1.8% of total stem borers collected (Table 1). Mean larval densities significantly varied among the stem borer species (χ^2 = 100.58, $p < 0.05$; Table 1). *Chilo partellus* exhibited a significantly higher larval density (14.41 ± 3.16) compared to both *Chilo orichalcociliellus* (0.23 ± 0.07) and *S. calamistis* (0.15 ± 0.05). Mean larval density of *C. orichalcociliellus* and *S. calamistis* were not significantly different.

In dry mid-altitude AEZ, stem borer species recovered were *C. partellus, S. calamistis* and *B. fusca*. *C. partellus* dominated the stem borer pest community by constituting 71.2%. *Sesamia calamistis* constituted 26% while *B. fusca* constituted 2.8% of the stem borers collected (Table 1). Mean larval density varied significantly among the three stem borer species (χ^2 = 74.92, $p < 0.05$). Highest larval density was exhibited by *C. partellus* (2.4 ± 0.37) followed by *S. calamistis* (0.80 ± 0.19) and *B. fusca* (0.06 ± 0.03). *Sesamia calamistis* and *B. fusca* were not significantly different in their mean larval densities (Table 1).

Table 1: Percentage composition and density of stem borer species recovered in the three agro-ecological zones

Agro-Ecological Zone	Stem borer species	% composition	Larval density ($\bar{x}$ ± SE)
Moist lowland	*Chilo partellus*	96.4	14.4 ± 3.2^{b}
	Sesamia calamistis	1.7	0.2 ± 0.1^{a}
	Busseola fusca	1.8	0.2 ± 0.1^{a}
	χ^2 value		100.58
	df		2
	p-value		2.20E-16
Dry mid-altitude	*Chilo partellus*	71.2	2.4 ± 0.4^{b}
	Sesamia calamistis	26	0.8 ± 0.2^{a}
	Busseola fusca	2.8	0.1 ± 0.0^{a}
	χ^2 value		74.92
	df		2
	p-value		2.20E-16
Moist mid-altitude	*Chilo partellus*	58.0	0.2 ± 0.1^{b}
	Sesamia calamistis	21.4	0.1 ± 0.0^{ab}
	Busseola fusca	19.1	0.1 ± 0.0^{ab}
	Eldana saccharina	1.5	0.005 ± 0.005^{a}
	χ^2 value		10.47
	df		3
	p-value		0.015

Note: Larval density within columns followed by the same lower case superscripts are not significantly different ($p > 0.05$).

In moist mid-altitude AEZ, four stem borer species were recovered; *C. partellus, S. calamistis, B. fusca* and *Eldana saccharina* Walker. *C. partellus* was dominant (58.02%), followed by *S. calamistis* (21.37%), *B. fusca* (19.08%) and *E. saccharina* (1.53%). There was a significant difference in mean larval densities among the four stem borer species (χ^2=10.47; $p < 0.05$) (Table 1). *C. partellus* had the highest larval density (0.21 ± 0.08) followed by *S. calamistis* (0.07 ± 0.02) and *B. fusca* (0.06 ± 0.02). Mean larval densities of these three species were significantly higher than *E. saccharina* (0.005 ± 0.005).

4.2. Stem borer parasitoid composition and diversity

In moist lowland AEZ, four larval and three pupal parasitoids were recovered (Table 2). The larval parasitoids included *C. flavipes*, *Cotesia sesamiae* (Cameron), *Chelonus curvimaculatus* (Cameron) and Tachnidae. Pupal parasitoids were *Pediobius furvus* (Gahan), *Dentichasmias busseolae* Heinrich and *Xanthopimpla stemmator*

Table 2: Percentage composition of parasitoids and stem borer species attacked in different agro-ecological zones

Parasitoid species	Guild	Moist lowland AEZ		Dry mid-altitude AEZ		Moist mid-altitude AEZ	
		% Composition	Stem borer species	% Composition	Stem borer species	% Composition	Stem borer species
Cotesia flavipes	Larval	75.9	*Chilo partellus*	76.5	*C. partellus, S. calamistis, B. fusca*	69.6	*Chilo partellus*
Cotesia sesamiae	Larval	18.3	*Chilo partellus*	7.4	*C. partellus, S. calamistis, B. fusca*	-	-
Chelonus curvimaculatus	Larval	0.3	*Chilo partellus*	0.4	*C. partellus, B. fusca*	-	-
Tachnidae	Larval	0.05	*Chilo partellus*	-	-	-	-
Dolichogenidea polaszeki	Larval	-	-	0.1	*S. calamistis*	-	-
Sturmiopsis parasitica	Larval / pupal	-	-	0.03	*B. fusca*	-	-
Pediobius furvus	Pupal	0.7	*Chilo partellus*	-	-	28.3	*Chilo partellus*
Xanthopimpla stemmator	Pupal	0.05	*Chilo partellus*	-	-	-	-
Dentichasmias busseolae	Pupal	0.05	*Chilo partellus*	0.2	*C. partellus*	-	-
Psilochalsis soudanensis	Pupal	-	*Chilo partellus*	-	-		
Aphanogmus fijiensis	Hyper-parasitoid	4.6	*Chilo partellus*	15.1	*C. partellus, S. calamistis*	0.4 1.8	*Chilo partellus* *Chilo partellus*

Thunberg which is exotic and was also introduced to manage stem borer pests. The larval parasitoid community was dominated by the exotic *C. flavipes* (75.87%) followed by *C. sesamiae* (18.32%), *C. curvimaculatus* (0.33%) and Tortricidae (0.02). Pupal parasitoid community was dominated by *P. furvus* (0.7%) followed by *D. busseolae* (0.05%) and *X. stemmator* (0.05%). The hyperparasitoid *Aphanogmus fijiensis* (Ferriére) was also recovered (4.63%). Parasitoids were recovered from *C. partellus* only.

In dry mid-altitude AEZ, six larval parasitoids, one pupal parasitoid and one hyperparasitoid were recovered (Table 2). The larval parasitoids were *C. flavipes* (76.47%), *C. sesamiae* (7.64%), *C. curvimaculatus* (0.41%), *Dolichogenidea polaszeki* Walker (0.14%) and *Sturmiopsis parasitica* (Curran) (0.03%). The pupal parasitoid was *D. busseolae* which constituted 0.17% of the total parasitoids collected while the hyperparasitoid, *A. fijiensis,* constituted 15.1%.

In moist mid-altitude AEZ, *C. flavipes* was the only larval parasitoid collected and it constituted 69.57% of the parasitoids. Two pupal parasitoids, *Psilochalsis soudanensis* (Steffan) and *P. furvus*, were collected and they constituted 28.26% and 1.81% of the parasitoid community respectively. All these parasitoids emerged from *C. partellus* only (Table 2).

4.3. Plant infestation by stem borers across the three agro-ecological zones

Plant infestation by stem borers in moist lowland AEZ was estimated at 28.94 ± 3.11% (Figure 3). Mean infestation varied significantly across distances at which sampling was conducted (χ^2_3 =10.72; $p < 0.05$). The least infestation levels were detected at parasitoid release points (11 ± 3.46%) while the highest (35.58 ± 9.8) were at farms sampled farthest from release points. Infestation progressively increased from release points to the furthest points sampled. Infestation levels significantly reduced between 1983 (92%-pre-release period) (Seshu Reddy, 1983) and 2015 (current study) ($V = 3$; $p < 0.05$) (Table 4).

In dry mid-altitude AEZ, plant infestation by stem borers was 22.47 ± 7.42% (Figure 3). There was no significant difference in infestation levels across sampling distances ($F_{3,51} = 0.4$; $p > 0.05$). Infestation levels were 22.4 ± 6.7%, 24.8 ± 2.1%, 20.6 ± 2.6% and 20.7 ± 3.3% at 0, 15, 30 and 45 km respectively (Table 3). A comparison between infestation levels recorded before *C. flavipes* was released (92% in 1997) (Songa *et al*., 2001) revealed a significant decrease from pre-release figures (t_{54}=41.63; $p < 0.05$) (Table 4).

Overall plant infestation by stem borers in moist mid-altitude AEZ was relatively low (2.71 ± 0.44%) (Figure 3). There was no significant difference in plant infestation levels across distances sampled (χ^2_3 =1.53; $p >$ 0.05) (Table 3). Results obtained showed a significant decrease in plant infestation levels when compared to levels obtained in 2006 (Ongámo *et al*., 2006) (V=0, $p < 0.05$) (Table 4).

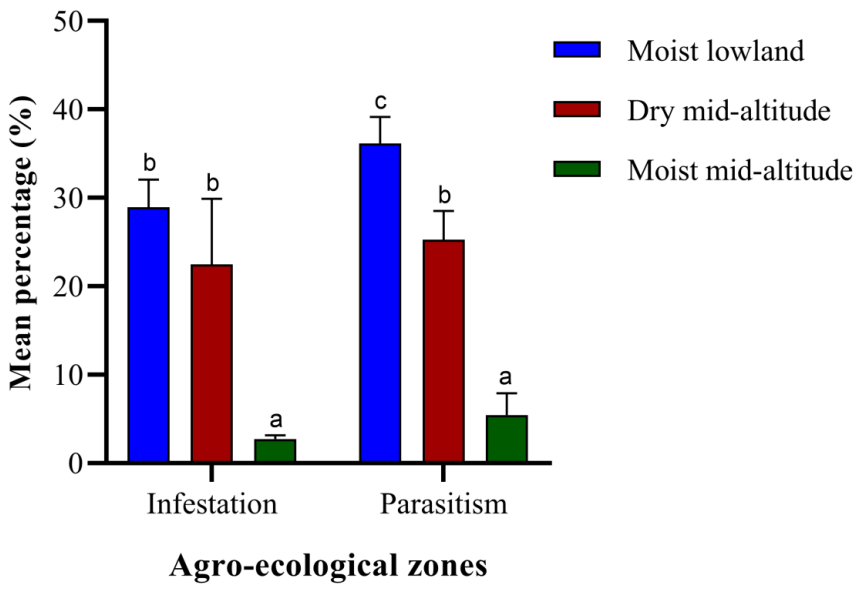

Figure 3: Percentage infestation and parasitism across different AEZ. Percentage infestation with same letters above the bars are not significantly different ($p > 0.05$)

Table 3: Mean plant infestation and stem borer parasitism ($\bar{x}$+*SE*) in different AEZs

Distance from release points	Sampled Agro-Ecological Zones (AEZ)					
	Moist lowland		Dry mid-altitude		Moist mid-altitude	
	Infestation	Parasitism	Infestation	Parasitism	Infestation	Parasitism
0 km	11.0 ± 3.5[a]	52.0 ± 10.9[b]	22.4 ± 6.7[a]	26.6 ± 7.6[a]	3.0 ± 3.0[a]	0.0 ± 0.0[a]
15 km	28.6 ± 5.2[a]	49.1 ± 4.2[b]	24.8 ± 2.1[a]	31.9 ± 6.0[a]	3.1 ± 0.6[a]	3.2 ± 2.4[a]
30 km	32.7 ± 3.4[a]	24.1 ± 3.1[a]	20.6 ± 2.6[a]	23.0 ± 5.8[a]	2.6 ± 0.9[a]	9.9 ± 6.8[a]
45 km	35.6 ± 9.8[a]	28.5 ± 5.4[a]	20.7 ± 3.3[a]	14.9 ± 6.7[a]	2.0 ± 1.1[a]	6.3 ± 6.3[a]
df	3, 48	3, 48	3, 51	3, 51	3, 37	3, 37
F-value	-	7.44	0.4	-	-	-
χ^2 value	10.72	-	-	4.34	1.53	0.86
p-value	0.013	0.00034	0.75	0.23	0.67	0.84

Note: Percentage infestation and parasitism ($\bar{x}$+*SE*) within columns followed by the same lower case superscripts are not significantly different ($p > 0.05$).

Table 4: Plant infestation by stem borers and parasitism levels before and after *C. flavipes* release in moist lowland, dry mid-altitude and moist mid-altitude AEZs in Kenya

AEZ	Period	Infestation	Period	Parasitism
Moist lowland	Pre-release (1983)	92[a]	Post-release (1994)	3
	2014	28.9 ± 3.1[b]	2014	36.1 ± 3.0[b]
	V-value	3	*t*-value	10.96
			df	51
	p-value	4.26E-10	*p*-value	5.22E-15
Dry mid-altitude	Pre-release (1997)	92[a]	Pre-release (2001)	10[a]
	2014	22.5 ± 7.4[b]	2014	25.3 ± 3.3[b]
	t-value	41.63	*V*-value	1213
	df	54		
	p-value	2.20E-16	*p*-value	0.0002
Moist mid-altitude	Post-release (2006)	37.4 ± 3.8[a]	1999	4[a]
	2014	2.7 ± 0.4[b]	2014	5.5 ± 2.5[b]
	V-value	0	*V*-value	195
	p-value	2.24E-08	*p*-value	0.0009

Note: Percentage infestation and parasitism ($\bar{x}$ ± *SE*) within columns followed by the same lower case superscripts are not significantly different ($p > 0.05$).

4.4. Stem borer parasitism across the three agro-ecological zones

Stem borer parasitism in moist lowland AEZ, was 36.1 ± 3.0% (Figure 3). Parasitism levels varied significantly with sampling distance ($F_{3,48}$=7.44; $p < 0.05$) (Table 4). The highest rate of parasitism was recorded at parasitoid release sites (52.0 ± 10.9%) and this was not significantly different from parasitism at farms situated 15 km from release points (49.1 ± 4.2%). Parasitism in farms located 30 km (24.1 ± 3.1%) and 45km (28.5 ± 5.4%) away from release points did not differ significantly with each other though there was significant variation with farms at 0 and 15 km from release points (Table 3). There was a significant increase in parasitism levels compared to results recorded during the last post release survey (3% in 1994) (t_{51}=10.96; $p < 0.05$) (Table 4).

In dry mid-altitude AEZ, stem borer parasitism was estimated at 25.3 ± 3.3% (Figure 3). There was no significant difference in parasitism levels across distances sampled (χ^2_3 =4.34; $p > 0.05$) (Table 4). Parasitism levels were 26.6 ± 7.6, 31.9 ± 6.0, 23 ± 5.8 and 14.9 ± 6.7% at 0, 15, 30 and 45 km respectively (Table 3). A significant variation was observed when parasitism results from this study were compared to pre-release figures (10% in 1998) (V = 1213, $p < 0.05$) (Table 4).

Parasitism levels in the moist mid-altitude AEZ were low (5.5 ± 2.5%) compared to results from moist lowland and dry mid-altitude AEZs (Figure 3). There was no significant difference in parasitism levels across distances sampled (χ^2_3 = 0.86; $p > 0.05$). No parasitoids were recovered at release points (0.0 ± 0.0%). Parasitism progressively increased with distance, that is, 3.2 ± 2.4, 9.9 ± 6.8 and 6.3 ± 6.3% at 15, 30 and 45 km respectively (Table 3). There was significant variation in parasitism rates when results of this study were compared to pre-release rates obtained in 1999 (4%) (V=195, $p < 0.05$) (Table 4).

4.5. Climatic variables affecting infestation and parasitism

Climatic variables which showed highest correlation to infestation and parasitism are shown in Table 5. Mean diurnal range, temperature seasonality and annual precipitation were highly correlated to infestation ($p < 0.05$). Annual mean temperature, mean diurnal range and isothermality were highly correlated to performance of the parasitoid ($p < 0.05$).

Table 5: Correlation of climatic parameters to infestation and parasitism

BIOCLIM variable	df	*F*-value	*p*-value
Infestation			
Annual mean temp	1	8.76	0.004**
Mean diurnal range	1	48.53	1.607e-10***
Isothermality	1	8.00	0.005**
Temperature seasonality	1	20.03	1.683e-05***
Maximum temperature of warmest month	1	7.62	0.007**
Annual precipitation	1	15.91	0.0001***
Precipitation of coldest quarter	1	3.63	0.059 ·
Parasitism			
Annual mean temp	1	20.56	1.326e-05***
Mean diurnal range	1	12.47	0.00058***
Isothermality	1	18.96	2.740e-05***
Temperature seasonality	1	3.81	0.053 ·
Precipitation of wettest quarter	1	5.86	0.017*
Precipitation of driest quarter	1	7.65	0.007**

5. Discussion

In classical biological control, natural enemies are released and relied upon to establish and spread beyond release points. *Cotesia flavipes* was released in moist lowland AEZ in 1993 (Overholt *et al*., 1994a). Releases were also made in moist mid-altitude AEZ in 2000 and central and eastern parts of dry mid-altitude in 2002 (KALRO and *icipe*, unpublished data). It is important to note that before 2002, release of *C. flavipes* had been carried out in experimental fields in southern part of dry mid-altitude AEZ (Songa *et al*., 1999). Following the afore-mentioned releases, establishment and spread of the parasitoid had only been documented for the moist lowland AEZ (Overholt *et al*, 1994b; Omwega *et al*., 1997; and Overholt *et al*., 1997). This study is hereby confirming the establishment and spread of *C. flavipes* in dry mid-altitude and moist mid-altitude AEZs. However, it is not clear whether the *C. flavipes* population in the moist mid-altitude AEZ originated from official releases in 2000 or accidental escape from *icipe*, Mbita point field station where they had been quarantined for pre-release studies as the presence may be from both sources (Omwega *et al*., 1995).

The main objective of *icipe*'s Biological Control program was to reduce stem borer pest populations in maize and sorghum crop by introducing *C. flavipes* in areas where *C. partellus* was the main pest (Overholt *et al*., 1994a). Across the three AEZs, *C. partellus* dominated the stem borer pest community corroborating reports from moist lowland (Nye, 1960; Van Hamburg, 1979; and Overholt *et al*, 1994a) and dry mid-altitude AEZ (Songa *et al*., 1999; and Ongamo *et al*., 2006). However, this is contrary in moist mid-altitude AEZ where previous reports indicated that *B. fusca* dominated (Zhou *et al.,* 2001; and Ongamo *et al.,* 2006). Formerly known to be a lowland tropical and dry mid-altitude species, the dominance demonstrated by *C. partellus* from these results validate findings that the pest's distribution has expanded into mid and high altitude areas. Further to this, in two of the zones, *C. partellus* was the only stem borer host from which parasitoids were recovered. This corroborates reports that the exotic stem borer has a larger number of parasitoids attacking it in comparison to native borers (Zhou *et al*., 2003). This is attributable to *C. partellus'* competitive superiority which has led to displacement of native stem borers and thus their unavailability for parasitization. Native parasitoids have therefore expanded their host ranges to include the dominant and readily available, *C. partellus.*

This study was carried out to assess the current stem borer and parasitoid levels in order to discern whether the introduced parasitoid has succeeded or failed in suppressing pest population. Observed plant infestation levels were significantly lower compared to figures recorded prior to release of parasitoid in all the three AEZs. In the moist lowland AEZ, levels of infestation on maize farms were estimated at 92% (Seshu Reddy, 1983) before the parasitoid was released. Surveys carried out in 2005 yielded lower infestation levels (77.2%, Ongamo *et al*., 2006). The sampling protocol used in both previous surveys were repeated during this study. Maize plants sampled were at early maturity and mature stages of growth for the three surveys. Plant infestation levels recorded from this study are significantly lower. Together, these data show that the rate of plant infestation by stem borers has been decreasing steadily over the years. A similar trend was observed in dry mid-altitude AEZ where infestation levels were higher (82-92%) before release of *C. flavipes* (Songa *et al*., 2001) but reduced to 62% later after release and to the current level of 22.47%. In the moist mid-altitude AEZ, previous reports of infestation levels were estimated to vary between 8-100% (Seshu Reddy, 1983) and later to 33.72% (Ongámo *et al*., 2006a) and reduced further to the current level of 2.71%. Reduction in levels of plant infestation by stem borers is evident across all the three AEZs, an indication that *C. flavipes* is steadily suppressing the stem borer pest population as sites with lower infestation levels exhibited higher parasitism.

Parasitism by *C. flavipes* following release has been assessed through different studies. In moist lowland AEZ, post release parasitism rates were recorded at 0.05 to 3 (Overholt *et al*., 1997), 7.1% in 1997 (Overholt *et al*., 1997) and 13% in 1999 (Zhou *et al*., 2001). This study demonstrated a significant increase in parasitism levels. Similarly, in dry mid-altitude AEZ, a rise in parasitism level was observed. Parasitism levels recorded in the central and eastern parts of this zone were significantly higher than levels observed in the southern part (Songa *et al*., 2001). In moist mid-altitude, parasitism rates were previously recorded at 3.5% (Khan *et al.,* 1997), 2.2% (Ogeda, 1999) and 4% in surveys carried out in 1999 (Zhou and Overholt, 2001). Current parasitism levels demonstrated a significant increase. This study thus confirms a rise in parasitism levels which in turn have suppressed pest populations in all the three AEZs.

Temperature and precipitation showed high correlation to both infestation and parasitism levels, results that corroborate reports by Van Lenteren *et al*. (2006) and Jiang *et al*. (2004). These parameters are among abiotic factors considered important in exotic biocontrol agent establishment in new environment. They influence key processes that dictate if an exotic biocontrol agent will establish and spread (Van Lenteren *et al*., 2006). Climate

matching (of native and new environment) is an important aspect in classical biological control as this can influence establishment or failure of biocontrol agents. Results of this study can be used in more informed choice of release localities both within and across Kenya's borders.

6. Conclusion

Generally, despite a rise in parasitism levels by *C. flavipes* across the surveyed regions, observed levels were still low compared to what is in the pest's native range. In India, 80% parasitism by *C. flavipes* is observed in maize (Singh *et al.,* 1975) with 0-43% being recorded in maize-sorghum intercrops (Subba Rao *et al*., 1969). Nonetheless, this scientific investigation has birthed some knowledge regarding the success of *C. flavipes* across various habitat types.

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