

# Evaluation of Napier grass (*Pennisetum purpureum*) varieties for use as trap plants for the management of African stemborer (*Busseola fusca*) in a push–pull strategy

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## Abstract

We evaluated eight Napier grass [*Pennisetum purpureum* Schumach (Poaceae)] varieties, used in various parts of eastern Africa as fodder, for their potential role as trap plants in the management of the African stemborer, *Busseola fusca* Füller (Lepidoptera: Noctuidae) through a push–pull strategy. Oviposition preference, larval orientation, settling, arrest and dispersal, feeding, mortality and survival, and development were determined for each of these varieties under laboratory and screen house conditions. Two-choice tests showed that only two of the varieties tested (cv. Bana and cv. Uganda Hairless) were preferentially chosen by gravid female moths for oviposition over a susceptible maize variety, cv. Western Hybrid 502. Larval preference was, however, highly variable. Larval feeding by first instars on the maize leaves was more intense and significantly more than on leaves of all the Napier grass varieties evaluated. Food consumed and amounts assimilated by the third instars over a 24-h period were not different among larvae fed on stems of maize and those fed on stems of the various Napier grass varieties. Larval survival was significantly lower on all the Napier grass varieties (below 3%) than on maize (about 44%). Similarly, larval development was about 2–3 weeks longer on majority of the Napier grass varieties. It was concluded that cv. Bana had potential for use as a trap plant in the management of *B. fusca* because it was more preferred by the moths for oviposition, equally preferred as maize by the larvae for orientation, settling, and arrest, and allowed minimal survival of the larvae. It can thus be used with such ‘push’ plants as *Desmodium* spp. (Fabaceae) in a ‘push–pull’ strategy, but the effectiveness of such a strategy would strictly depend on proper establishment and management of these companion plants.

## Introduction

Phytophagous insects often demonstrate preferences for particular plant species, cultivars, or crop stage in response to different cues, a behavior that can be exploited to manage insect pests through the use of trap crops (Hokkanen, 1991; Shelton & Nault, 2004). Host-plant recognition and selection in Lepidoptera is primarily a function of the ovipositing female (Konstantopoulou et al., 2002), and, as

newly emerged larvae are often limited in their dispersal abilities, oviposition is particularly crucial because it determines survival of their progeny (Renwick, 1989). A majority of these insects are, therefore, able to discriminate between plants that are acceptable for oviposition and feeding and those that are not (Otter & Kahoro, 1983; Rebe et al., 2004). Host-plant colonization by cereal stem borers, which are the most injurious insects of maize and sorghum in sub-Saharan Africa (Kfir et al., 2002), is a process based on the interactions between sensory cells and the physical and chemical characteristics of the plant (Waladde et al., 1990).

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The major cereal stemborer species, *Busseola fusca* Füller (Lepidoptera: Noctuidae) and *Chilo partellus* Swinhoe (Lepidoptera: Crambidae), cause significant yield losses and are difficult to manage. This is largely due to the nocturnal behavior of the adult moths and the protection provided to the larvae and pupae by the stems of the host plants (Khan et al., 1997). In sub-Saharan Africa, cereals are grown in small plots surrounded by wild grasses, and as stemborers utilize wild grasses as alternative hosts (Khan et al., 1997; Polaszek & Khan, 1998; Ndemah et al., 2001; van den Berg et al., 2001), it was previously recommended that these grasses should be destroyed to reduce stemborer infestation on the main crops (Ingram, 1958). However, recent studies have demonstrated that some of these grasses can be maintained and used to manage the pests as trap plants. Some of the wild grasses are preferred to maize for oviposition by stemborer moths (Khan et al., 1997, 2006; van den Berg et al., 2001, 2003; van den Berg, 2006). Similarly, other grasses and legumes have been shown to repel gravid moths (Khan et al., 2000). A habitat management strategy, based on stimulo-deterrent or 'push-pull' approach, formalized by Miller & Cowles (1990), has been developed for cereal stemborer management in eastern Africa (Cook et al., 2007). In this strategy, maize is intercropped with a repellent legume such as desmodium, *Desmodium uncinatum* (Jacq.) (Fabaceae), or molasses grass, *Melinis minutiflora* (Beauv.) (Poaceae), with an attractant wild grass such as Napier grass, *Pennisetum purpureum* (Schumach.) (Poaceae), planted around the plot. Studies have shown that stemborer infestation of the main crop is significantly reduced and grain yields significantly enhanced in the 'push-pull' plots when the companion crops are properly established and managed (Khan et al., 1997, 2001; Midega et al., 2005).

Smallholder farmers in sub-Saharan Africa practice mixed farming, with livestock being an important component (Khan & Pickett, 2004). The companion plants used in the habitat management strategy should not only offer an effective and sustainable control of the stemborers, but should also be of additional economic value (Khan et al., 2001), either in terms of fodder or source of income to the farmers. We had previously evaluated various Napier grass varieties for use as trap plants for the management of *C. partellus* (Khan et al., 2006). Here, our objective was to evaluate the Napier grass varieties for their suitability as trap plants in the management of the indigenous stemborer, *B. fusca*. In this study we evaluated various behavioral and physiological responses of *B. fusca*, such as larval orientation and settling, feeding, metabolism of ingested food, mortality, survival, development, and oviposition on various Napier grass varieties grown in eastern Africa.

## Materials and methods

### Study site

The studies were conducted at the International Centre of Insect Physiology and Ecology (ICIPE), Thomas Odhiambo Campus (ITOC), Mbita Point, in western Kenya, 0°25'S, 34°12'E. The center is situated on the eastern shores of Lake Victoria in Suba district of Kenya, where cereal stemborers are a serious limitation to cultivation of maize. The area receives approximately 900 mm of rainfall per annum, has a mean annual temperature of 27 °C, and is located at an altitude of approximately 1200 m above sea level.

### Plants and insects

The Napier grass plants used in these studies were as described by Khan et al. (2006). Eight varieties were taken from a stock of plants supplied by the Kenya Agricultural Research Institute (KARI) collected from various parts of Kenya: Ex-Nyanza, Ex-Machakos, Uganda Hairless, Gold Coast, French Cameroon, Clone 13, Bana grass, and Pakistan hybrid. In addition, a maize variety susceptible to *B. fusca* attack, maize cv. Western hybrid 502, was used as a control.

*Busseola fusca* moths and larvae used in these studies were raised from larvae collected from the wild, principally sorghum fields, and maintained on seminatural diet with maize leaf powder (6-week-old plants, dried and ground into powder) as a main component (Onyango & Ochieng-Odero, 1994) in the laboratory at ITOC for two generations. The purpose of maintaining them in the laboratory was to build up enough culture for the studies. Studies have shown that larval and adult experiences have no influence on host-plant preference of *B. fusca* and *C. partellus* (Mbugi, 1999; J van den Berg, unpubl.).

### Oviposition preference

*Experiment I: two-choice tests.* Modifications of the methodology of Smith et al. (1994) as described by Khan et al. (2006) were employed in these studies. Two-choice tests were carried out in oviposition cages measuring 80 × 40 × 40 cm covered by a fine wire mesh netting with a cloth access flap. Two potted maize plants, 3–4 weeks old, and two potted plants of a Napier grass variety of the same age and size as the maize, raised from stem cuttings, were placed into each cage. A wad of cotton wool (10 cm in diameter) moistened with water was introduced into the cage for the moths to feed on. The four plants were placed one at each corner of the oviposition cage and three mated female *B. fusca* moths were introduced into the cage. The moths were then allowed to oviposit for 48 h under natural light conditions

of approximately L12:D12. The plants were then removed and the number of eggs on each plant was counted under a light binocular microscope at 6.5 magnification to determine the moths' preference for either maize or a Napier grass variety. This arrangement was replicated 10 times. We took 'preference' in this context to be differential oviposition on a plant when the insect is given a choice between two plant species (Singer, 1986).

#### Larval behavior and feeding

*Experiment II: larval orientation and settling.* We assessed host-plant preference of *B. fusca* larvae for Napier grass varieties or maize using a modification of the methodologies of Khan et al. (1996). In a 15-cm Petri dish lined with a moist filter paper disc, four 3-cm long leaf cuts of a Napier grass variety and maize were laid alternately and radially, two for each plant, with their adaxial sides facing up, in a two-choice test. Ten first instars of *B. fusca* were then introduced at the center of each Petri dish and were allowed to orientate and settle on their preferred leaf cuts. The Petri dishes were placed in a dark room and the number of larvae present on/under each leaf cut was counted after 1 h for orientation and after 24 h for settling preference. The experiment was replicated 10 times. Preference was indicated for the plant variety with higher settling scores (Khan et al., 1996).

*Experiment III: arrest and dispersal of first instars.* A 6-cm long leaf cut of a Napier grass variety or maize, with a moist cotton wad at either end, was placed individually, with its adaxial side up, in the center of a 9-cm Petri dish lined with moist filter paper. Ten first instars of *B. fusca* were then released in the middle of each leaf cut. The number of larvae remaining on the leaf cut was counted at 1 h and 24 h after release. The experiment was conducted in a dark room with 10 replications.

*Experiment IV: larval leaf and stem feeding and food assimilation.* To evaluate feeding of *B. fusca* larvae on the Napier grass varieties, studies were conducted with leaves and stem cuttings of the plants. A piece of leaf (2.5 × 2.5 cm) from a 3-week-old maize plant or Napier grass variety was placed in a small cylindrical container (3 cm in height × 4 cm in diameter), lined with a piece of wet filter paper to prevent wilting. Nine containers were assembled, representing each of the plant varieties. Five just-hatched and unfed neonate (first-instar) *B. fusca* were then placed on each leaf cut in the container and each container was covered and sealed with Parafilm 'M' and was kept in the dark room. The leaf area (mm<sup>2</sup>) consumed by the larvae was measured after 24 h using a graph paper (Mohamed et al., 2007). The difference in the leaf area before and after

feeding indicated feeding levels of the larvae on the plant leaves. The experiment was replicated 10 times.

To assess stem feeding on the Napier grass varieties and the amount of food assimilated by *B. fusca* larvae, a study was conducted in the laboratory using stem segments from 3-week-old potted maize and Napier grass plants. Stem segments (4 cm long) were cut from the maize or Napier grass variety. Each segment was then weighed (S1) and placed in a vial. A newly molted third-instar *B. fusca*, previously starved for 3 h but water satiated, was weighed (W1) on a microbalance (Mettler PM460; Mettler Instrument, Greifensee, Zurich, Switzerland) and was introduced into the vial. The vial was then covered with cotton wool and kept in the dark room. After 24 h, the vials were removed from the dark room and the uneaten parts of the stem segments weighed again (S2) after removing the larvae and excreta. To determine weight loss due to evaporation, 10 stem segments (4 cm) of each treatment were weighed (C1), kept in vials alongside the experimental vials, and weighed again after 24 h (C2). The difference between the initial weight (S1) and the final weight (S2) of the stem after the adjustment of the weight loss from evaporation represented feeding on the plants by the larvae (Khan & Saxena, 1985). Each treatment was replicated 10 times.

To assess the amount of food assimilated, each larva was weighed again (W2). To determine weight loss due to catabolism, 10 larvae were weighed (C1), kept in vials without stem alongside the experimental vials, and weighed again after 24 h (C2). The amount of food metabolized by each larva was determined using the formula adapted from Khan & Saxena (1985): Assimilation of food =  $W1 \times (C1 - C2) / C1 + (W2 - W1)$ , where W1 = initial weight of larva, W2 = final weight of larva, C1 = initial weight of control larva, and C2 = final weight of control larva, the mean difference in weights being 0.0013 g.

*Experiment V: larval survival and development under laboratory conditions.* To evaluate survival and development of *B. fusca* larvae on the different Napier grass varieties, an experiment was carried out in the laboratory with no climate control and poor lighting as described by Khan et al. (2006). This was done within a concrete building where temperatures were cooler by day due to shading (mean of 26.5 °C), but similar to outside temperatures by night (mean of 23.5 °C).

Five sections of a 3-week-old Napier grass variety were placed in a screw-top glass jar, 20 cm in height × 8 cm in diameter. Each of these sections, measuring between 0.5 cm and 1.5 cm diameter at the base and approximately 15 cm long, consisted of stem, leaf, and sheath regions for the larvae to feed on (Khan et al., 2006). Into each jar, 25 newly hatched and healthy larvae were introduced via fine

camel hair paint brush. Two replicates were established for each Napier grass variety and maize. The lids of the jars were tightly screwed shut, and paper towelling was used to make a tighter seal, such that the larvae could not escape from the jars (Khan et al., 2006). The larvae were then allowed to feed for 5 days, the least number of days taken between instars (Päts, 1992), in a shaded position in the laboratory. The plants were then removed and carefully dissected to recover the larvae. Each larva recovered was assessed on the basis of its developmental stage.

The remaining larvae were then placed into a similar jar containing five freshly cut sections of the same Napier grass variety or maize as that from which they were removed. They were then left to feed for another 5 days and then the plants were dissected and the larvae were reassessed. As the larvae reached the third instar, stem sections were introduced to replace some of the whorl sections and larvae separated into their own jars to avoid competition for food. White absorbent paper towelling, used as a pupation medium, was introduced to the bottom of the jar when the larvae were nearing pupation (Khan et al., 2006). Thus, a time sequence of the development and mortality was recorded at 5-day intervals. In addition, the time to pupation of the individual larvae was recorded.

*Experiment VI: larval mortality and plant damage.* This experiment was conducted over a 25-day period under uninterrupted, seminatural conditions, using modifications of suggestions of Smith et al. (1994) as described by Khan et al. (2006). Four-week-old potted Napier grass varieties and the maize control were placed on inverted bucket lids filled with water, which was regularly topped up. This acted as a moat to prevent ant infestation of the plants and pots (Khan et al., 2006). These pots were arranged in a randomized block design within a 15 × 5 × 4 m screen house with a sloping roof and central ridge running lengthwise. The plants were thinned to two best tillers in each pot. Five newly hatched and healthy *B. fusca* larvae were then introduced to the whorl region of each of the plants using fine brushes. The pots were placed approximately 80 cm apart to, together with the moats, prevent dispersal of the larvae between pots.

At 25 days after infestation, data were collected on leaf feeding and larval mortality. The former was assessed qualitatively for each individual plant in each pot via a visual scoring system of between 1 (little or no leaf feeding) and 5 (heavy leaf feeding). Larval mortality was assessed by counting the larvae recovered dead or missing and expressed as a percentage of the total larvae introduced per plant. The larvae recovered dead and missing were grouped under 'mortality' (Midega et al., 2004) and expressed as a percentage of the total larvae introduced

per plant. Additionally, data on the number of surviving individuals and their life stages were collected for other studies.

#### Data analysis

Student's t-test (SAS Institute, 2001) was used for any differences between maize and each Napier grass variety with regards to the number of eggs laid in each plant and larvae orientated or settled. One-way analysis of variance (ANOVA) (SAS Institute, 2001) using the generalized linear model was used for any differences between the Napier grass varieties with regards to leaf feeding scores, larval recoveries, and percentage of larval survival and mortality. The data on proportion of larvae surviving (percentage of survival) and larval mortality were subjected to square root and arcsine transformation, respectively, and conformed to the assumptions of ANOVA as indicated by tests of normality (SAS Institute, 2001) prior to analysis. Similarly, the data on larval arrest and settlement were subjected to log transformation ( $\log x+1$ ) prior to analyses. Thereafter, Tukey's studentized test was used to separate the means at  $P<0.05$ . Means of non-transformed data are presented in figures and tables.

## Results

#### Oviposition preference

*Experiment I: two-choice tests.* Significantly higher number of eggs were laid on cultivars Bana grass and Uganda Hairless than on maize ( $P<0.05$ ) by *B. fusca* moths in the two-choice tests. The number of eggs laid on the rest of the test plants however did not significantly differ from those laid on maize ( $P>0.05$ ), except for cv. Ex-Machakos where significantly more eggs were laid on maize ( $P<0.05$ ) (Table 1).

#### Larval behaviour and feeding

*Experiment II: larval orientation and settling.* When given a choice of leaf cuts of maize and those of a Napier grass variety, most of the larvae orientated to those of maize in most of the tests, although the differences were only significant in cv. Bana grass after 1 h of larval release (Table 2). Similarly, most of the larvae settled on leaf cuts of maize than those of the Napier grass varieties in most of the tests although the differences were not significant in all cases (Table 2).

*Experiment III: arrest and dispersal of first instars.* Significantly more first-instar *B. fusca* larvae were arrested (retained) on maize leaf cuts than on those of the Napier grass varieties within the first hour after release ( $F_{8,81} = 4.92$ ,  $P<0.01$ ), except in cv. Bana grass and cv. Ex-Machakos where these numbers did not significantly differ from

**Table 1** Average ( $\pm$  SE) number of *Busseola fusca* eggs laid on Napier grass varieties and maize in two-choice tests

Napier grass variety vs. maize	Mean ( $\pm$ SE) number of eggs per plant	t-value	P-value
Bana grass Maize	525.9 (37.8) 252.5 (22.9)	-6.18	<0.01
Clone 13 Maize	352.9 (46.5) 405.1 (40.0)	0.85	0.41
Ex-Machakos Maize	281.7 (29.2) 412.9 (23.1)	3.52	<0.01
Ex-Nyanza Maize	367.4 (26.2) 356.2 (42.9)	-0.22	0.83
French Cameroon Maize	304.7 (28.9) 359.4 (32.5)	1.26	0.22
Gold Coast Maize	435.1 (15.8) 413.5 (47.4)	-0.43	0.67
Pakistan hybrid Maize	412.0 (24.3) 380.5 (28.8)	-0.83	0.41
Uganda Hairless Maize	467.7 (41.1) 236.9 (17.0)	-5.19	<0.01

maize. However, significantly more larvae were retained 24 h after release on the leaf cuts of maize than on the leaf cuts of cv. Clone 13, cv. Gold Coast, and cv. Uganda Hairless ( $F_{8,81} = 9.59, P < 0.01$ ) (Table 3).

**Table 2** Orientation and settling of first and third instars of *Busseola fusca* on different Napier grass varieties and maize under two-choice tests

Napier grass varieties and maize	Mean ( $\pm$ SE) number of larvae orientated and settled on each leaf cut					
	Orientation			Settling		
	1 h	t-value	P-value	24 h	t-value	P-value
Bana Maize	3.0 (0.4) 5.4 (0.3)	-4.6	<0.01	3.7 (0.5) 4.2 (0.5)	-0.72	0.48
Clone 13 Maize	4.2 (0.4) 4.0 (0.4)	0.35	0.73	3.4 (0.4) 4.4 (0.5)	-1.56	0.13
Ex-Machakos Maize	4.3 (0.5) 4.5 (0.4)	-0.16	0.88	3.1 (0.5) 4.1 (0.5)	-1.47	0.16
Ex-Nyanza Maize	3.6 (0.6) 4.9 (0.5)	-1.67	0.11	3.6 (0.5) 4.0 (0.5)	-0.53	0.60
French Cameroon Maize	3.9 (0.6) 5.1 (0.4)	-1.55	0.14	3.6 (0.3) 4.0 (0.4)	-0.74	0.47
Gold Coast Maize	4.3 (0.4) 4.5 (0.5)	-0.31	0.76	4.4 (0.5) 4.7 (0.4)	-0.46	0.65
Pakistan hybrid Maize	3.6 (0.5) 4.6 (0.4)	-1.52	0.14	3.9 (0.5) 3.8 (0.4)	0.15	0.88
Uganda Hairless Maize	3.4 (0.5) 4.7 (0.5)	-1.85	0.08	4.1 (0.4) 4.2 (0.4)	-0.16	0.88

*Experiment IV: larval leaf and stem feeding and food assimilation.* *Busseola fusca* larvae consumed a significantly larger area of leaf cuts from maize plants than they did on those from the Napier grass varieties ( $F_{8,81} = 5.85, P < 0.01$ ; Table 4). On the other hand, the amount of material from stem pieces of Napier grass varieties and maize consumed by the larvae did not significantly differ ( $F_{8,81} = 1.75, P = 0.10$ ), although relatively more, in terms of weight, was consumed from maize than from the Napier grass varieties (Table 4). Similarly, the amount of food assimilated by larvae fed on stem pieces of the different test plants did not significantly differ from the amount assimilated by the larvae fed on maize ( $F_{8,81} = 1.24, P = 0.2896$ ), although more food was assimilated by the larvae fed on the latter (Table 4).

*Experiment V: larval survival and development under laboratory conditions.* Survival of *B. fusca* larvae on the different Napier grass varieties over the 70-day period showed that the greatest mortality (approximately 90%) occurred within the first 15 days after egg hatch (Figure 1). This was followed by a period of considerably reduced mortality, reaching approximately 95% at 50 days in most of the varieties. In maize on the other hand, the initial mortality was much less (approximately 45% at 15 days after egg hatch) and generally reaching a maximum of about 55% after 50 days (Figure 1).

The time taken to pupation by *B. fusca* larvae on the Napier grass varieties was on average 62.5 days, with an

**Table 3** *Busseola fusca* larval arrest on leaf cuts of different Napier grass varieties and maize in no-choice tests

Napier grass varieties and maize	Mean ( $\pm$ SE) number of larvae arrested on each leaf cut	
	1 h	24 h
Bana	3.4 (0.5)ab	6.3 (0.9)a
Clone 13	1.8 (0.2)b	1.5 (0.3)bc
Ex-Machakos	2.6 (0.5)ab	4.4 (0.9)ab
Ex-Nyanza	1.8 (0.4)b	5.7 (1.0)a
French Cameroon	2.3 (0.4)b	3.6 (0.7)ab
Gold Coast	1.6 (0.5)b	2.3 (0.9)bc
Pakistan hybrid	1.4 (0.4)b	4.9 (0.7)a
Uganda Hairless	1.5 (0.3)b	0.9 (0.3)c
Maize	4.4 (0.6)a	7.5 (1.0)a
F <sub>8,81</sub>	4.92	9.59
P-value	<0.01	<0.01

Means within a column followed by same letters are not significantly different ( $P>0.05$ ): Tukey's studentized range test.

average survival rate of 1.8% (Table 5). On the other hand, an average time to pupation on maize was about 37.2 days, with a mean survival rate of 44% (Table 5). Among the Napier grass varieties, the greatest survival rate (2.4%) occurred on cv. Clone 13, cv. Bana grass, cv. Ex-Machakos, and cv. Gold Coast, with the rest having survival rates of 1.6%. However, no larvae survived to pupation in cv. Pakistan hybrid. For the purposes of analysis and comparison, a very small percentage survival (0.00001%), technically 0, and a very large time to pupation were theoretically used to obtain a result from the calculations (Khan et al., 2006). Pupation occurred most rapidly on cv. French Cameroon (50 days) and Uganda Hairless (55 days), with the others being in the range of 62.7–65.0 days (Table 5). Larval growth index was considerably higher on

maize (1.2) than on all the individual Napier grass varieties and the overall mean for the Napier varieties (Table 5).

*Experiment VI: larval mortality and plant damage.* Significantly more *B. fusca* larvae were recovered from maize than from the Napier grass varieties ( $F_{8,81} = 14.43$ ,  $P<0.01$ ) 25 days after infestation (Table 6). There were no significant differences among the Napier grass varieties with regards to the number of *B. fusca* larvae recovered. The Napier grass varieties had mortality rates ranging from about 88% to about 96%, except in cv. Pakistan hybrid where there was 100% mortality. On the other hand, mortality of *B. fusca* larvae was only 24% on maize, significantly lower than in the Napier grass varieties ( $F_{8,81} = 22.7$ ,  $P<0.01$ ) (Table 6).

Leaf feeding was significantly more intense, exemplified by higher leaf damage scores, on maize than on the Napier grass varieties ( $F_{8,81} = 7.91$ ,  $P<0.01$ ), except for cv. Clone 13. The Napier grass varieties generally supported only slight leaf feeding with scores ranging between 1.2 (Bana, Pakistan hybrid, Ex-Machakos) and 1.9 (Uganda Hairless), except for cv. Clone 13 where a mean damage score of 2.2 was recorded (Figure 2).

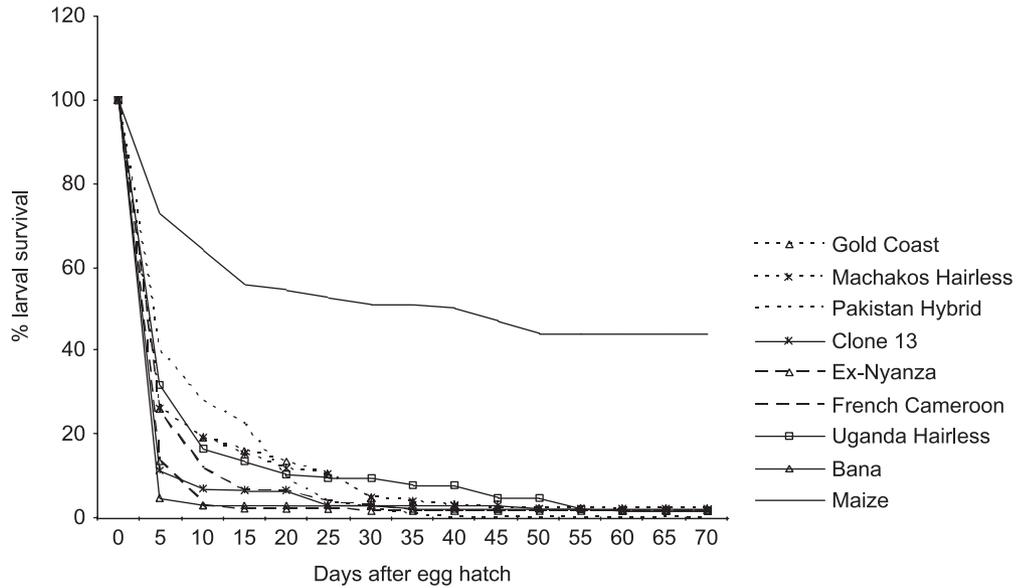
## Discussion and conclusions

In Lepidoptera, host-plant recognition and selection, primarily a function of the ovipositing female, is mediated by many factors, with chemical cues (green leaf volatiles) unambiguously playing a major, if not decisive, role (Udayagiri & Mason, 1995). Results of our study showed that although most of the Napier grass varieties were not preferentially chosen over maize by *B. fusca* moths for oviposition, cv. Bana grass and Uganda Hairless were significantly more preferred. The Bana grass variety has

**Table 4** Feeding and food assimilation by *Busseola fusca* on different Napier grass varieties and maize

Napier grass varieties and maize	Mean ( $\pm$ SE) leaf area (mm <sup>2</sup> ) consumed by five first instars	Mean ( $\pm$ SE) weight of stem weight (mg) consumed/third instar	Mean ( $\pm$ SE) weight of food assimilated/third instar
Bana	0.8 (0.4)b	0.21 (0.06)a	0.005a
Clone 13	4.1 (1.4)b	0.09 (0.02)a	0.004a
Ex-Machakos	1.1 (0.4)b	0.21 (0.06)a	0.002a
Ex-Nyanza	4.1 (1.8)b	0.15 (0.02)a	0.004a
French Cameroon	0.2 (0.2)b	0.22 (0.04)a	0.005a
Gold Coast	3.5 (0.8)b	0.15 (0.04)a	0.004a
Pakistan hybrid	3.6 (0.9)b	0.10 (0.03)a	0.004a
Uganda Hairless	2.3 (1.2)b	0.18 (0.03)a	0.004a
Maize	10.3 (2.3)a	0.26 (0.05)a	0.006a
F <sub>8,81</sub>	5.85	1.75	1.24
P-value	<0.01	0.100	0.2896

Means within a column followed by same letters are not significantly different ( $P>0.05$ ): Tukey's studentized range test.



**Figure 1** Percentage survival of *Busseola fusca* larvae on different Napier grass varieties and maize at different times following egg hatch under laboratory conditions.

**Table 5** Survival and development of *Busseola fusca* larvae in different Napier grass varieties and maize under laboratory conditions

Variety	Percentage of survival to pupation (P)	Mean ( $\pm$ SE) days to pupation (D)	Growth index (P/D)	Rank
Pakistan hybrid	0.00001	10 000	$1 \times 10^{-9}$	1
Ex-Nyanza	1.6	63.5 (3.5)	0.02	2
Uganda Hairless	1.6	55.5 (1.5)	0.03	3
French Cameroon	1.6	50.0 (2.0)	0.03	4
Ex-Machakos	2.4	64.0 (2.0)	0.04	5
Gold Coast	2.4	65.0 (1.1)	0.04	6
Bana	2.4	62.7 (1.8)	0.04	7
Clone 13	2.4	64.0 (2.0)	0.04	8
Napier (mean)	1.8	62.5	0.03	
Maize (mean)	44.0	37.2	1.2	

been shown to produce significantly more amounts (almost 100-fold) of the green leaf volatiles than maize within the first hour of the scotophase, the period at which *C. partellus* and *B. fusca* moths seek host plants (Chamberlain et al., 2006) for oviposition. Previous studies have shown clear preference of *C. partellus* moths for various Napier grass varieties for oviposition (van den Berg et al., 2003; Khan et al., 2006; van den Berg, 2006). Moreover, results obtained in the field have also shown a similar trend, where the Napier grass (largely cv. Bana) planted as a trap plant around a plot of maize in the push-pull system significantly reduced *C. partellus* and *B. fusca* infestation in the maize crop (Khan et al., 2001; Midega et al., 2005). The results of the current study thus

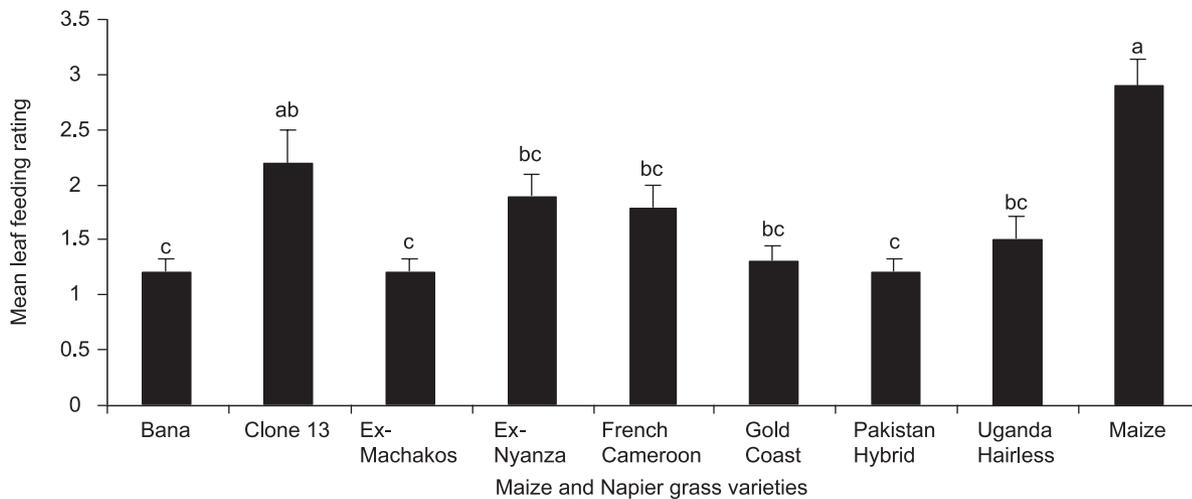
confirmed what we have observed under field conditions. In our experimental plots, we have observed that planting three rows of the trap crop around a  $50 \times 50$  m plot of maize, without the 'push' plants, offers effective control of stemborers. Otherwise the strategy becomes less effective on larger plots.

While host-plant experience has been found to influence oviposition preference in some insect species, such as *Plutella xylostella* (L.) (Lepidoptera: Plutellidae) (Zhang & Liu, 2006) and *Helicoverpa armigera* (Hübner) (Lepidoptera: Noctuidae) (reviewed by Jallow et al., 2004), none has been observed in *B. fusca* and *C. partellus* (Mbugi, 1999; J van den Berg, unpubl.). Additionally, the use of such a trap crop in a habitat-management strategy is often combined

Maize and Napier grass variety	Mean ( $\pm$ SE) number of larvae recovered per plant	Mean ( $\pm$ SE) percentage of larval mortality per plant
Pakistan hybrid	0.0 (0.0)b	100 (0.0)a
Ex-Nyanza	0.6 (0.3)b	88.0 (5.3)a
Uganda Hairless	0.2 (0.1)b	96.0 (2.7)a
French Cameroon	0.3 (0.2)b	94.0 (4.3)a
Ex-Machakos	0.3 (0.2)b	94.0 (4.3)a
Gold Coast	0.4 (0.3)b	92.0 (5.3)a
Bana	0.3 (0.2)b	94.0 (4.3)a
Clone 13	0.6 (0.3)b	88.0 (5.3)a
Maize	3.8 (0.4)a	24 (8.3)b
F <sub>8,81</sub>	14.43	22.7
P-value	<0.01	<0.01

**Table 6** Mean ( $\pm$  SE) recovery and mortality of *Busseola fusca* larvae in different Napier grass varieties and maize under seminatural conditions in the screen house

Means within a column followed by same letters are not significantly different ( $P>0.05$ ): Tukey's studentized range test.



**Figure 2** Average ( $\pm$  SE) larval leaf feeding score in different Napier grass varieties and maize under seminatural conditions in the screen house after 25 days of larval release. Bars marked with different letters are significantly different ( $P<0.05$ ): Tukey's studentized range test; 1 = little or no feeding, 5 = heavy leaf feeding.

with a repellent plant intercropped with the target crop, which serves to 'push away' the moths from within the target crop (Khan et al., 2000).

Dispersal of larvae of most Lepidoptera is usually restricted to neonates (Zalucki et al., 2002; Moore & Hanks, 2004), and is largely linked to low-quality, non-preferred or unsuitable host species (Ramachandran, 1987), or when food is not available (van der Linde, 1971). Our results show that in general, first instars of *B. fusca* preferred leaf cuts of maize to those of most of the Napier grass varieties for orientation and settling, although differences were not significant except in orientation response between maize and Bana grass. Similar trends were observed with regards to larval arrest within the first hour of release. These results are similar to those of Mohamed

et al. (2004) who reported preference of maize leaf cuts over those of wild grasses by larvae of *C. partellus* for settling and arrest. They speculated that this observation could be linked to the suitability of the plants for larval growth and development, a plausible explanation for the trends in the current study. This observation therefore calls for a careful layout of the trap crop so that any dispersing larvae do not end up on the target crop. Positioning the first innermost row of the trap crop at 1 m from the first row of maize has been shown to effectively prevent larval dispersal from the trap to the target crop (Khan et al., 1997).

Once oviposition preference has been determined for a candidate trap plant, the suitability of the plant for larval feeding and development is the next step. It is essential that a successful trap crop should not allow any significant

larval survival and/or development (Shelton & Nault, 2004). Host-plant quality is known to largely determine the biological performance of herbivores (Awmack & Leather, 2002). Previous studies have shown poor *C. partellus* larval survival on some of its wild hosts such as Napier grass varieties (van den Berg et al., 2001; Rebe et al., 2004; Khan et al., 2006; van den Berg, 2006) probably due to poor nutritional status and/or antibiotic effects. Some herbivores, such as *Anticarsia gemmatalis* (Hübner) (Lepidoptera: Noctuidae) (see review of Awmack & Leather, 2002), are known to feed more (compensatory feeding) when exposed to host plants of low nutrition (Slansky & Wheeler, 1989). In the current study, however, no such compensatory feeding was observed. Instead, first instars fed more on the leaf cuts of maize than on those of the Napier grass varieties. Similarly, there was more intense feeding by the larvae on the leaves of maize (whole plant) than most of the Napier grass varieties causing more damage on the former. The third instars, however, did not significantly differ with regards to the amount of food consumed and assimilated between the maize and Napier grass varieties. This may have been due to the limited time (24 h) the larvae were left to feed.

Our results show that all the Napier grass varieties supported low survival and a much longer developmental period for the larvae, implying that the plant material was either of poor nutritional value, or had an antibiotic effect on the larvae (Khan et al., 2006), or had improper nutrient proportions (Hoo & Fraenkel, 1966). Previous studies showed no improvement in survival rates of the surviving larvae of both *C. partellus* and *B. fusca* continuously reared on Napier grass (cv. Bana) up to the second generation (Mbugi, 1999). Most of the *B. fusca* larvae (approximately 90%) on Napier grass varieties died within the first 15 days of egg hatch, comparable to the mortality reported for *C. partellus* larvae on the plants within the same period after egg hatch (van den Berg et al., 2003; Khan et al., 2006; van den Berg, 2006).

Some of the Napier grass varieties produce a sticky gum around the leaf sheath region in reaction to stemborer larval penetration, which has been observed to restrict larval movement into the stem (Khan & Pickett, 2004; Khan et al., 2006), particularly observable in cv. Bana and cv. Ex-Machakos, when the plants are attacked or damaged to seal the wound (Hill, 1983). The plant sap therefore acts as a mortality factor for the larvae by restricting their movements, physically trapping the larvae, killing them or exposing them to the other mortality factors such as natural enemies (Hill, 1983; Khan & Pickett, 2004).

*Busseola fusca* larvae had a mean growth index of 0.03 on the Napier grass varieties relative to 1.2 on maize,

comparable to 0.1 reported for *C. partellus* on the Napier grass varieties (Khan et al., 2006). However, because not all the varieties were more preferred to maize for oviposition by *B. fusca* moths, only cv. Bana can be recommended for use in regions where *B. fusca* and *C. partellus* are occurring together. Uganda Hairless is more preferred to maize for oviposition by both *C. partellus* moths (Khan et al., 2006) and *B. fusca* (current study) and was found to support higher survival of *C. partellus* larvae and its leaves equally acceptable to the larvae for feeding as those of maize (Khan et al., 2006). Its usage as a trap plant would thus require an additional pest control method, such as application of insecticides on the trap plant to reduce the pest population within it, because of the danger of the pest multiplying on it and eventually attacking the main crop, either through larval dispersal or mixing of stubble. We had earlier recommended cv. Bana grass as one of the Napier grass varieties with potential as a trap plant for the management of *C. partellus* (Khan et al., 2006). The results of the current study therefore indicate that the variety can be used as a trap plant for both stemborer species, highly attractive for oviposition but only minimally supporting larval survival, useful characteristics of a suitable trap crop (Shelton & Badenes-Perez, 2006). Additionally, Bana grass is equally preferred as maize by the larvae for orientation, settling, and arrest, and therefore rates of dispersal that may lead to the larvae getting to the maize under field conditions are low. Its preference by the smallholder farmers who practice mixed farming as fodder for their livestock over the other Napier grass varieties (ZR Khan, unpubl.) makes it a preferable trap plant in the push-pull strategy. However, the effectiveness of the push-pull strategy largely depends on proper establishment and management of the companion crops.

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### References

- Awmack CS & Leather SR (2002) Host plant quality and fecundity in herbivorous insects. *Annual Review of Entomology* 47: 817–844.

- van den Berg J (2006) Oviposition preference and larval survival of *Chilo partellus* (Lepidoptera: Pyralidae) on Napier grass (*Pennisetum purpureum*) trap crops. *International Journal of Pest Management* 52: 39–44.
- van den Berg J, Midega CAO, Wadhams LJ & Khan ZR (2003) Can Vetiver grass be used to manage insect pests on crops? Proceedings of the Third International Conference on Vetiver and Exhibition, Guangzhou, China, October 2003 (ed. by RG Grimshaw), pp. 254–264. China Agriculture Press, Beijing, China.
- van den Berg J, Rebe M, de Bruyn J & van Hamburg H (2001) Developing habitat management systems for gramineous stem borers in South Africa. *Insect Science and its Application* 21: 381–388.
- Chamberlain K, Khan ZR, Pickett JA, Toshova T & Wadhams LJ (2006) Diel periodicity in the production of green leaf volatiles by wild and cultivated host plants of stem borer moths, *Chilo partellus* and *Busseola fusca*. *Journal of Chemical Ecology* 32: 565–577.
- Cook SM, Khan ZR & Pickett JA (2007) The use of ‘push-pull’ strategies in integrated pest management. *Annual Review of Entomology* 52: 375–400.
- Hill DS (1983) *Agricultural Insect Pests of the Tropics and their Control*, 2nd edn. Cambridge University Press, Cambridge, UK.
- Hokkanen HMT (1991) Trap cropping in pest management. *Annual Review of Entomology* 36: 119–138.
- Hoo SCF & Fraenkel G (1966) The consumption and digestion and utilization of food plants by phytophagous insects; *Prodenia eridania* (Cramer). *Journal of Insect Physiology* 12: 711–730.
- Ingram WR (1958) The lepidopterous stalk borers associated with Graminae in Uganda. *Bulletin of Entomological Research* 49: 367–383.
- Jallow MFA, Cunningham JP & Zalucki MP (2004) Intra-specific variation for host plant use in *Helicoverpa armigera* (Hübner) (Lepidoptera: Noctuidae): implications for management. *Crop Protection* 23: 955–964.
- Kfir R, Overholt WA, Khan ZR & Polaszek A (2002) Biology and management of economically important lepidopteran cereal stem borers in Africa. *Annual Review of Entomology* 47: 701–731.
- Khan ZR, Abenes MLP & Fernandez NJ (1996) Suitability of graminaceous weed species as host plants for rice leaf folders, *Cnaphalocrocis medinalis* and *Marasmia patnalis*. *Crop Protection* 15: 121–127.
- Khan ZR, Chiliswa P, Ampong-Nyarko K, Smart LE, Polaszek A et al. (1997) Utilisation of wild gramineous plants for the management of cereal stem borers in Africa. *Insect Science and its Application* 17: 143–150.
- Khan ZR, Midega CAO, Hutter NJ, Wilkins RM & Wadhams LJ (2006) Assessment of the potential of Napier grass (*Pennisetum purpureum*) varieties as trap plants for management of *Chilo partellus*. *Entomologia Experimentalis et Applicata* 119: 15–22.
- Khan ZR & Pickett JA (2004) The ‘push-pull’ strategy for stem borer management: a case study in exploiting biodiversity and chemical ecology. *Ecological Engineering for Pest Management: Advances in Habitat Manipulation for Arthropods* (ed. by GM Gurr, SD Wratten & MA Altieri), pp. 155–164. CABI Publishing, CABI, Wallingford, Oxon, UK.
- Khan ZR, Pickett JA, van den Berg J, Wadhams LJ & Woodcock CM (2000) Exploiting chemical ecology and species diversity: stem borer and striga control for maize and sorghum in Africa. *Pest Management Science* 56: 957–962.
- Khan ZR, Pickett JA, Wadhams LJ & Muyekho F (2001) Habitat management strategies for the control of cereal stem borers and striga in maize in Kenya. *Insect Science and its Application* 21: 375–380.
- Khan ZR & Saxena RC (1985) Behavioral and physiological responses of *Sogatella furcifera* (Homoptera: Delphacidae) to selected resistant and susceptible rice cultivars. *Journal of Economic Entomology* 78: 1280–1286.
- Konstantopoulou MA, Krokos FD & Mazomenos BE (2002) Chemical stimuli from corn plants affect host selection and oviposition behavior of *Sesamia nonagrioides* (Lepidoptera: Noctuidae). *Journal of Economic Entomology* 95: 1289–1293.
- van der Linde VR (1971) The sailing flight of the gypsy moth (*Lymantria dispar* L.) and the effect of the food plant on this phenomenon. *Zeitschrift für Angewandte Entomologie* 67: 316–324.
- Mbugi JP (1999) Comparative Studies on the Adult Dispersal Behaviour of *Busseola fusca* (Füller) (Lepidoptera: Noctuidae) and *Chilo partellus* Swinhoe (Lepidoptera: Pyralidae). MSc Dissertation, Kenyatta University, Kenya.
- Midega CAO, Khan ZR, van den Berg J & Ogol CKPO (2005) Habitat management and its impact on maize stem borer colonization and crop damage levels in Kenya and South Africa. *African Entomology* 13: 333–340.
- Midega CAO, Ogol CKPO & Overholt WA (2004) Effect of agroecosystem diversity on natural enemies of maize stem borers in coastal Kenya. *International Journal of Tropical Insect Science* 24: 280–286.
- Miller JR & Cowles RS (1990) Stimulo-deterrent diversion: a concept and its possible application to onion maggot control. *Journal of Chemical Ecology* 16: 3197–3212.
- Mohamed HM, Khan ZR, Mueke JM, Hassanali A, Kairu E & Pickett JA (2007) Behaviour and biology of *Chilo partellus* (Swinhoe) on *Striga hermonthica* (Del.) Benth. infested and uninfested maize plants. *Crop Protection* in press.
- Mohamed HM, Khan ZR, Overholt WA & Elizabeth DK (2004) Behaviour and biology of *Chilo partellus* (Lepidoptera: Pyralidae) on maize and wild gramineous plants. *International Journal of Tropical Insect Science* 24: 287–297.
- Moore RG & Hanks LM (2004) Aerial dispersal and host plant selection by neonate *Thyridopteryx ephemeraeformis* (Lepidoptera: Psychidae). *Ecological Entomology* 29: 327–335.
- Ndemah R, Schulthess F, Poehling M & Borgemeister C (2001) Spatial dynamics of lepidopterous pests on *Zea mays* (L.) and *Pennisetum purpureum* (Moench) in the forest zone of Cameroon and their implications for sampling schemes. *Journal of Applied Entomology* 125: 1–8.
- Onyango FO & Ochieng-Odero JPR (1994) Continuous rearing of the maize stem borer *Busseola fusca* on an artificial diet. *Entomologia Experimentalis et Applicata* 73: 139–144.

- Otter DCJ & Kahoro HM (1983) Taste cell responses of stemborer larvae, *Chilo partellus* (Swinhoe), *Eldana saccharina* Walker and *Maruca testulalis* (Geyer) to plant substances. *Insect Science and its Application* 4: 153–157.
- Päts P (1992) Reproductive Biology of the Cereal Stemborer *Chilo partellus*. PhD Dissertation, University of Uppsala, Sweden.
- Polaszek A & Khan ZR (1998) Host plants. African Cereal Stemborers: Economic Importance, Taxonomy, Natural Enemies and Control (ed. by A Polaszek), pp. 3–10. CABI Publishing, CABI, Wallingford, UK.
- Ramachandran R (1987) Influence of host plants on the wind dispersal and the survival of an Australian geometrid caterpillar. *Entomologia Experimentalis et Applicata* 44: 289–294.
- Rebe M, van den Berg J & McGeoch MA (2004) Growth and development of *Chilo partellus* (Swinhoe) (Lepidoptera: Crambidae) on cultivated and indigenous graminaceous host plants. *African Entomology* 12: 253–258.
- Renwick JAA (1989) Chemical ecology of oviposition in phytophagous insects. *Experientia* 45: 223–228.
- SAS Institute (2001) SAS software. Statistics, Release 8.1. SAS Institute, Cary, NC, USA.
- Shelton AM & Badenes-Perez FR (2006) Concepts and applications of trap cropping in pest management. *Annual Review of Entomology* 51: 285–308.
- Shelton AM & Nault BA (2004) Dead-end trap cropping: a technique to improve management of diamondback moth, *Plutella xylostella* (Lepidoptera: Plutellidae). *Crop Protection* 23: 497–503.
- Singer MC (1986) The definition and measurement of oviposition preference in plant-feeding insects. *Insect-Plant Interactions* (ed. by JR Miller & TA Miller), pp. 65–69. Springer-Verlag, New York, NY, USA.
- Slansky F & Wheeler GS (1989) Compensatory increases in food consumption and utilization efficiencies by velvetbean caterpillars mitigate impact of diluted diets on growth. *Entomologia Experimentalis et Applicata* 51: 175–188.
- Smith CM, Khan ZR & Pathak MD (1994) Techniques for Evaluating Insect Resistance in Crop Plants. CRC-Lewis Press, Boca Raton, FL, USA.
- Udayagiri S & Mason CE (1995) Host plant constituents as oviposition stimulants for a generalist herbivore: European corn borer. *Entomologia Experimentalis et Applicata* 76: 59–65.
- Waladde SM, Kahoro HM & Ochieng SA (1990) Sensory biology of *Chilo* spp. with special reference to *C. partellus*. *Insect Science and its Application* 11: 593–602.
- Zalucki MP, Clarke AR & Malcolm SB (2002) Ecology and behavior of first instar larval Lepidoptera. *Annual Review of Entomology* 47: 361–393.
- Zhang P & Liu S (2006) Experience induces a phytophagous insect to lay eggs on a nonhost plant. *Journal of Chemical Ecology* 32: 745–753.