RESISTANCE OF SOUTHERN AFRICAN VARIETIES OF SUGARCANE TO *CHILO SACCHARIPHAGUS* (LEPIDOPTERA: CRAMBIDAE) IN MOZAMBIQUE, AND DEVELOPMENT OF A NON-DESTRUCTIVE FIELD RESISTANCE RATING SYSTEM

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Abstract

While completing pre- and post-release surveys of sugarcane fields for release of the pupal parasitoid *Xanthopimpla stemmator* against *Chilo sacchariphagus* in Mozambique, it was noticed that there were differences in infestation levels in similarly aged varieties of sugarcane at Açucareira de Mozambique and Companhia de Sena sugar estates. Both these estates had recently imported numerous South African varieties of sugarcane. It was also noticed that the intensity of infestation of some of these varieties by *C. sacchariphagus* was different to the intensity of infestation of the same varieties by *Eldana saccharina* under South African conditions. These differences are discussed.

In addition, a difference between varieties was also noted in the degree of damage to the top leaves of different varieties of young sugarcane by early instar *C. sacchariphagus*. This led to the development of a promising non-destructive method of evaluating the resistance of these varieties against this borer. Comparison of the non-destructive method with the destructive stalk dissection methods showed some correlation. The method and correlations are described.

Keywords: sugarcane, stalk borer, resistance, assessment, *Chilo sacchariphagus*, leaf feeding index, Mozambique

Introduction

In 2000, the Mozambican sugar estate of Açucareira de Mozambique (A de M) (34° 10' E; 19° 20' S), at Mafambisse, 80 km inland from Beira, requested that the South African Sugar Association Experiment Station (SASEX) investigate biological means of controlling an exotic stalk borer, *Chilo sacchariphagus* Bojer (Lepidoptera: Crambidae) (Conlong and Goebel, 2002). *C. sacchariphagus* was positively identified by Way and Turner (1999), who postulated that the introduction of this Asian borer was probably through infested sugarcane brought into Mozambique from Mauritius, where it is a major pest (Williams, 1983), as it is in Réunion (Goebel, 1999). Subsequent to the commencement of the biological control programme at A de M, *C. sacchariphagus* was identified at another sugar estate in Marromeu, north of A de M, and belonging to Companhia de Sena (Sena) (35° 57' E; 18° 17' S) (Conlong and Goebel, 2002). As a result, biological control programmes commenced at both sugar estates.

A prerequisite for the introduction of a new biological control programme, especially a classical one (Huffaker, 1971) as envisioned for these two sugar estates, is that intensive surveys be undertaken in the crop to determine the impact, if any, of indigenous parasitoids which may have established on the introduced pest, and which may be displaced by an introduced parasitoid (Overholt *et al.*, 1994). During these surveys information was collected on the intensity of pest infestation on the sugarcane varieties recently planted at A de M and Sena. When completing surveys for parasitoids, the age and variety of the sugarcane being surveyed is always recorded to determine any parasitoid preferences.

In South Africa, both variety and age had marked effects on infestation by an indigenous sugarcane borer, *Eldana saccharina* Walker (Lepidoptera: Pyralidae) (Carnegie, 1982; Carnegie and Smaill, 1980; Keeping, 1999). A de M and Sena had fairly recently been rehabilitated, and had imported a number of the newer varieties from South Africa and Zimbabwe for assessment in the very heavy soils occurring on both estates. It was while surveying these varieties for indigenous parasitoids attacking *C. sacchariphagus* that data was obtained which led to the publication of this paper.

Materials and methods

Survey methodology

At both sugar estates, the survey methodology as described by Keeping and Govender (2002) was followed. At Sena, varieties of eight to nine months old in irrigated and dryland fields were surveyed. Fifty stalks were randomly collected from the fields in question. Individual stalk length and stalk damage was measured in cm, and total number of internodes per stalk and total number of internodes bored per stalk were recorded. Immature stages of *C. sacchariphagus* collected were weighed and summed per variety sampled, to determine resistance ratings as described by Keeping and Govender (2002).

At A de M, a greater variety of South African, and some Zimbabwean, varieties were planted into agronomy trials. These trials were all sprinkler irrigated. Field 574 comprised 10 varieties, which were nine months old when assessed for susceptibility. The varieties were planted in adjacent strips, each 50 m long. Each variety comprised four rows, with a break of two metres between varieties. Twenty-five sticks were taken from each of the end rows of each variety, and assessed as described above. The agronomy trial in field 182 had an additional variety of sugarcane, and was set out and sampled as described for field 574. The cane in field 182 was six months old at the time of sampling.

Resistance ratings

As an initial estimate of resistance, a very rough rating scale was used. The lowest infestation level (the most resistant) in each category got the lowest rating (1), and the highest (least resistant) was assigned the highest rating (3 for the irrigated varieties and 6 for the dryland varieties at Sena, and 10 for the varieties at A de M). The rating scores were summed per variety. Where the values for any of the categories were equal for different varieties, they were given the same rating. However, on returning to SASEX, the data collected was assessed as done for *E. saccharina* varietal assessments (Keeping and Govender, 2002). As no varietal preference for South African varieties by *C. sacchariphagus* was known prior to the publication of this paper, no control varieties could be identified for inclusion in these assessments. However, the weighted means were used to rate the resistance of the varieties, with the lowest being the most resistant, and the highest the least resistant.

Leaf damage assessments

A de M had in 2001 imported a number of South African varieties. These had from July to September 2002 been planted out in bulking-up plots, and were between two and four months old. *C. sacchariphagus* is known to oviposit on and bore into sugarcane this young (Goebel, 1999). The opportunity was taken to survey these varieties to determine whether the varietal preference of *C. sacchariphagus* could be measured, because it was noticed in these fields with younger cane, that variable amounts of leaf damage were evident on the first four green leaves of the leaf whorl. This is common when maize or sorghum is attacked by the closely related *Chilo partellus* Swinhoe (Lepidoptera: Crambidae) and another indigenous maize borer, *Busseola fusca* Fuller (Lepidoptera: Noctuidae) (van den Berg *et al.,* 2000). The maize and sorghum entomologists have used the leaf feeding behaviour of these borers as a non-destructive measure, a 'leaf feeding index', to assess when corrective action against the borers should take place, and also to determine the susceptibility of different maize and sorghum cultivars to the borers (van den Berg *et al.,* 2000; van Rensburg, 1999).

It was decided during a visit to A de M in October 2002, to test this method for *C. sacchariphagus* in sugarcane. In agronomy trials established in field 574 (see above), the middle line of each variety was inspected for leaf damage, to give a value for total plants with leaves showing damage. In addition, where leaf damage was observed, the emerging leaf in the centre of the whorl was counted as leaf one, and the leaves were then numbered consecutively as they got older, from the centre, until four leaves were sampled. On each leaf, the number of feeding 'shot holes' and window scars were counted and recorded, and the total for each of the four leaves for each damaged plant was then summed. The number of leaves showing this damage was also recorded. These two values, the total number of leaves damaged per plant, and the total damage recorded on these leaves, was used to determine a damage rating, as shown in Table 1.

Table 1. Visual damage rating scale, using number of the top four leaves of the sugarcane plant with feeding damage, and extent of feeding damage on these, to determine feeding preferences of young *Chilo sacchariphagus* larvae on different sugarcane varieties (modified from van den Berg *et al.* (2000)).

Rating scale	No. of damaged leaves	Total feeding holes
1	1-2	<10
2	1-2	10-20
3	1-2	21-30
4	2-3	31-40
5	2-3	41-50
6	2-3	51-60
7	3-4	61-70
8	3-4	71-80
9	3-4	>80

A 'Mean Leaf Feeding Damage Rating' for each variety tested was thus derived from the total leaf damage ratings obtained for that variety, divided by the total number of plants showing leaf damage for that variety.

A 'Leaf Feeding Score' was derived from multiplying the mean leaf feeding damage rating of a particular variety, by the ratio obtained from dividing the total number of damaged leaves counted in that variety by the total number of plants of that variety showing leaf feeding damage.

Results

Resistance ratings

Sena sugar estate

Table 2 lists the varieties sampled, and shows how they rank in terms of % stalks bored, % internodes bored, % length bored, *C. sacchariphagus* per 50 stalks and mean *C. sacchariphagus* mass.

Using the very basic rating scheme, it is clearly evident from Table 2 that varieties show different susceptibilities to *C. sacchariphagus* under irrigated and dryland conditions. N19 in irrigated conditions was the most susceptible to this borer, whereas under dryland conditions it was one of the more resistant. In contrast, variety N30 was less susceptible to *C. sacchariphagus* under irrigated conditions.

Similarly, under dryland conditions at Sena, N21 followed by N12 were the most susceptible to *C. sacchariphagus* (Table 2), while N14 and N19 proved to be the most resistant.

Açucareira de Mozambique

Variety N28 was the most resistant to *C. sacchariphagus* at A de M sugar estate, and N25 and N26 the most susceptible (Table 3) in both the younger (field 182) and older (field 574) sugarcane.

Table 2. Basic resistance ratings of South African sugarcane varieties to *Chilo sacchariphagus* in furrow irrigated and dryland conditions at Companhia de Sena sugar estate. The lowest rating value indicates the most resistant, and the highest the least resistant. The actual maximum and minimum values recorded for each category are also given.

Variety	No. stalks sampled	Age (mths)	Sugarcane stalks			*Chilo sacchariphagus*		Total rating
			% stalks bored	% internodes bored	% length bored	Per 50 stalks	Individual mass (g)	
Furrow								
Max			*86*	*13.2*	*0.6*	*11*	*0.13*	*15*
Min			*34*	*1.9*	*4.5*	*1*	*0.02*	*5*
N28	50	9	1	1	1	1	1	5
N30	50	8.5	2	2	2	2	1	9
N19	50	8	3	3	3	3	2	14
Dryland								
Max			*78*	*15.5*	*5.6*	*8*	*0.13*	*30*
Min			*26*	*2.8*	*1.2*	*2*	*0.06*	*5*
N14	50	8	2	2	1	1	3	9
N19	50	9	2	1	1	4	3	11
N33	50	9.5	4	3	2	2	2	13
NCo376	50	9	3	4	3	3	1	14
N12	50	8.5	1	5	4	5	1	16
N21	50	7	5	6	5	5	1	22

Table 3. Basic resistance ratings of South African and Zimbabwean sugarcane varieties to *Chilo sacchariphagus* in irrigated agronomy trials at Acucareira de Mozambique sugar estate. The lowest rating value indicates the most resistant, and the highest the least resistant. The actual maximum and minimum values recorded for each category are also given.

Variety	No. stalks sampled	Age (mths)	Sugarcane stalks			*Chilo sacchariphagus*		Total rating
			% stalks bored	% internodes bored	% length bored	Per 50 stalks	Individual mass (g)	
Field 574								
Max			*98*	*22.0*	*22.2*	*31*	*2.3*	*50*
Min			*38*	*3.6*	*2.9*	*5*	*0.3*	*5*
N28	50	9	1	1	2	1	1	6
N23	50	9	2	2	1	6	4	15
N22	50	9	3	3	7	7	3	23
ZN6	50	9	7	8	3	5	2	25
N32	50	9	4	6	8	4	4	26
N19	50	9	6	5	6	5	6	28
N14	50	9	5	7	9	3	5	29
ZN3	50	9	5	4	4	9	8	30
N25	50	9	8	9	5	2	7	31
N26	50	9	9	10	10	8	9	46
Field 182								
Max			*66*	*9.4*	*10.6*	*19*	*1.2*	*55*
Min			*28*	*3.1*	*2.4*	*1*	*0.2*	*5*
N28	50	6	1	1	1	1	2	6
N32	50	6	2	2	4	4	1	13
N14	50	6	3	4	6	2	4	19
N23	50	6	5	5	3	4	5	22
N22	50	6	4	3	5	5	6	23
ZN5	50	6	7	7	8	2	3	27
N19	50	6	6	9	2	3	9	29
ZN3	50	6	8	8	9	5	4	34
N25	50	6	10	6	11	7	8	42
N26	50	6	8	11	10	6	7	42
ZN6	50	6	9	10	7	8	10	44

Table 4 gives the resistance ratings obtained for the varieties at both estates using the weighted mean method of Keeping and Govender (2002). This reflects very closely the resistance values obtained in Tables 3 and 4.

Table 4. Resistance to *Chilo sacchariphagus* of the varieties planted at Companhia de Sena and Acucareira de Mozambique, determined by the method of Keeping and Govender (2002).

Companhia de Sena				Acucareira de Mozambique			
Irrigated		Dryland		Field 574		Field 182	
Variety	Weighted Mean	Variety	Weighted Mean	Variety	Weighted Mean	Variety	Weighted Mean
N28	10 548	N14	6 434	N28	8 130	N28	7 116
N30	12 382	N33	14 558	N23	12 151	N32	15 806
N30	45 073	NCo376	18 495	ZN6	14 593	ZN5	16 654
N19	49 723	N19	26 008	N22	16 920	N14	17 341
		N21	34 094	N14	17 038	N23	21 081
		N12	34 773	N25	20 921	N22	21 849
				N32	21 012	N19	27 730
				N19	21 100	ZN3	29 788
				ZN3	44 399	N26	34 486
				N26	45 422	N25	35 264
						ZN6	45 836

Leaf damage assessments

Table 5 summarises the results of this method of assessment completed at A de M.

Table 5. Use of leaf feeding to assess varietal preference of *Chilo sacchariphagus* for varieties planted at Acucareira de Mozambique.

Variety	Total plants with damage	Leaf			
		Total damage	Mean damage rating	Feeding score	Feeding index
South African varieties					
N26	81	184	4.1	9.3	30.2
N25	52	110	3.5	7.4	15.5
N22	79	136	2.6	4.4	13.9
N14	38	80	3.7	7.8	11.9
N28	33	59	3.0	5.4	7.1
N19	25	52	3.3	6.9	6.9
N23	8	16	3.8	7.5	2.4
N32	15	22	2.0	2.9	1.8
Zimbabwean varieties					
ZN3	62	108	2.7	4.7	11.6
ZN6	60	101	1.7	2.8	6.8

Using the leaf feeding index as a measure of varietal preference, it is clear from Table 5 that the leaves of varieties N25 and N26 were most preferred by young *C. sacchariphagus* larvae, whereas N23 and N32 were least preferred.

Discussion

Resistance ratings

Plant resistance is a basic part of an integrated pest management system, especially against stalk borers in sugarcane (Brader, 1979; Meagher *et al.*, 1996). SASEX has recognised this in its programme against *E. saccharina*, and in 1979 instituted the first plant resistance trials against this borer (Bond, 1988). However, already in 1939, *E. saccharina* was known to attack the soft variety POJ 2725. *E. saccharina* 'disappeared' from sugarcane a few years later, which coincided with the large-scale planting of Co 281 (Dick, 1950), only to reappear again in NCo376 in 1970 (Nuss *et al.*, 1986). In recent years, SASEX plant resistance work has become much more refined (Keeping, 1999), with new varieties being assessed against *E. saccharina* on a routine basis (Keeping and Govender, 2002). Figure 1 shows the current resistance ratings of the varieties grown in the South African sugar industry against *E. saccharina*.

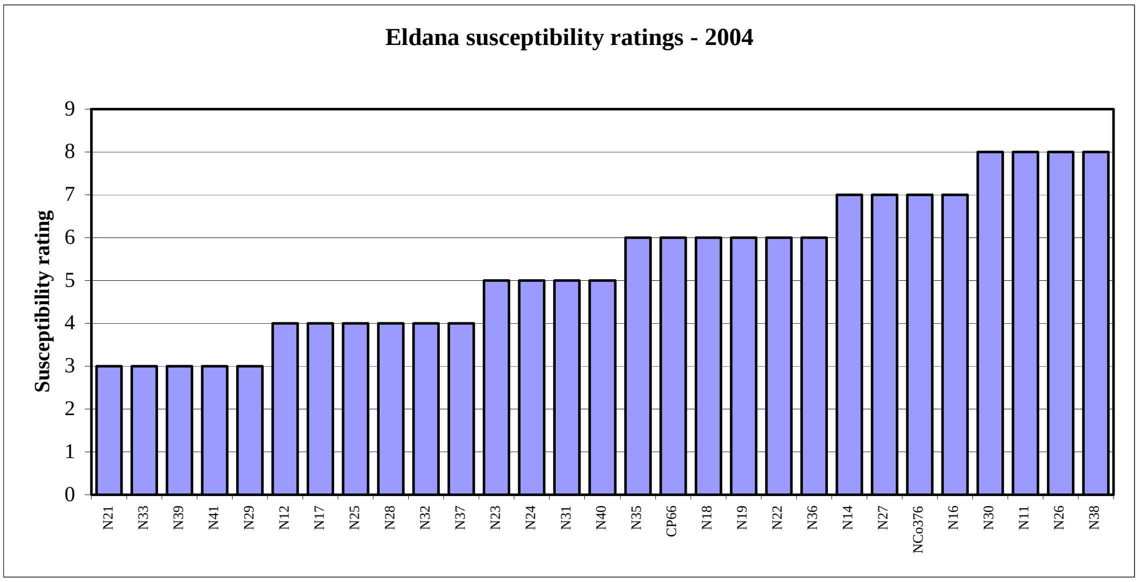

Figure 1. Current susceptibility ratings of commercial South African varieties to *Eldana saccharina*. (1 = very highly resistant; 9 = very highly susceptible) (source: South African Sugar Association Experiment Station).

These ratings were obtained by comparing the newer varieties with a number of control varieties known to be susceptible or resistant to *E. saccharina* (Keeping and Govender, 2002). This comparison is very important in any resistance trial before final decisions are made regarding varietal reaction to insect damage (van den Berg *et al.*, 2000). The results presented in this paper are not designed to give conclusive resistance ratings to *C. sacchariphagus*, as the susceptibility or resistance of the varieties tested was not known. Rather, the results show (i) that there are varietal preferences shown by *C. sacchariphagus*, and this could change with the same variety grown under irrigated or dryland conditions, and (ii) that there are differences in the susceptibility of the same varieties to different sugarcane stalk borers. The estates concerned should thus consider the establishment of a well-structured variety selection programme for their estates, to assess the suitability of varieties available for planting in areas known to harbour *C. sacchariphagus*.

The former is demonstrated by the results obtained from Sena. N19 in irrigated conditions was the most susceptible to this borer, while under dryland conditions it was one of the more resistant varieties (Tables 2 and 4). In South Africa, N19 is moderately susceptible to *E. saccharina* (Figure 1). In contrast, N30 was less susceptible to *C. sacchariphagus* under irrigated conditions (Tables 2 and 4). Under South African conditions, this variety is regarded as one of the most susceptible to *E. saccharina* (Figure 1).

The latter is also demonstrated by results obtained from Sena. In South Africa, N21 is the most resistant to *E. saccharina*, closely followed by N12 (Figure 1). Conversely, N14 and N19, considered susceptible to *E. saccharina* (Figure 1), seem the most resistant to *C. sacchariphagus* under dryland conditions. N33, considered highly resistant to

E. saccharina (Figure 1), is only moderately resistant to *C. sacchariphagus* (Tables 2 and 4). Clearly the two borers respond to different resistance mechanisms in the same sugarcane varieties.

The results from A de M do not show the marked differences obtained at Sena. At A de M, results were obtained from planted agronomy trials, rather than from different fields on the estate, so the local environmental conditions within the trial sites were very uniform in terms of soil conditions, watering and fertilising regimes, and microclimate. N28 and N32 are regarded as only moderately resistant to *E. saccharina*, yet they showed the most resistance to *C. sacchariphagus.* Also growing in irrigated conditions as at Sena, N19 at A de M was susceptible to *C. sacchariphagus* attack, as was N14 (Tables 3 and 4). N25 and N26 proved to be the least resistant to *C. sacchariphagus* (Tables 3 and 4), and are also the least resistant to *E. saccharina* (Figure 1).

It is worthy of note that South African varieties such as N14 and N19, and Zimbabwean varieties ZN3 and ZN6 show different susceptibility to *C. sacchariphagus* at different ages (Table 3). This warrants further investigation. However, for management recommendations, it may be more practical to use the resistance ratings of the older sugarcane varieties.

Leaf damage assessments
In any variety evaluation programme, the assessment method should be simple, accurate and efficient, as very often many hundreds of plants have to be evaluated (van den Berg *et al.*, 2000). However, these methods are clearly related to the biologies and behaviours of the insects on the respective crop plants. Destructive sampling methods such as those used for varietal evaluation against *E. saccharina* (Keeping and Govender, 2002) are determined by this insect's cryptic behaviour at all stages in its life cycle. Although this type of evaluation will have to be made at least initially for *C. sacchariphagus*, its oviposition habits and feeding behaviour of the neonate larvae (Goebel, 1999; Conlong and Goebel, 2002) may lead to an easier and simpler method of evaluation, as described in this paper and used by maize and sorghum entomologists (van den Berg *et al.*, 2000). Table 6 lists the most resistant and most susceptible varieties as measured by leaf inspection when the sugarcane in the agronomy trial sites was at chest height (two to three months old), and then by stalk dissection when the varieties were six and nine months old.

Table 6. List of the sugarcane varieties planted in agronomy trials at Acucareira de Mozambique showing most, moderate and least resistance to *Chilo sacchariphagus* at different ages and using different measures of assessment.

Type of survey (Age of cane)	Cane variety resistance		
	Most	Moderate	Least
Leaf index (3 mths)	N32; N23	N28; N14	N25; N26
Stalk survey (6 mths)	N28; N32	N22; N23	N25; N26
Stalk survey (9 mths)	N28; N23	N32; N14/N19	N25; N26

It thus seems likely that leaf damage assessments may prove the most efficient, simple and accurate method of assessing sugarcane varieties for resistance to *C. sacchariphagus.* In all methods tested, the most susceptible varieties were shown to be N25 and N26. This was picked up in the leaf damage assessments. Varieties N32 and N23 indicated the least susceptibility in the leaf damage assessments, and N23 proved the most resistant when destructive stalk samples were taken when the variety was nine months old. N32 showed moderate resistance at that stage. The converse was true with N28, which showed moderate resistance when leaf damage was assessed, but became the most resistant variety at nine months of age.

This method thus holds promise for being suitable for assessing resistance of sugarcane varieties to *C. sacchariphagus*. However, it will have to be calibrated more accurately in properly designed plant resistance trials before it can become a useful tool for routine use by plant breeders and entomologists.

Conclusion

Until now, there was no information available on the performance of southern African varieties to the introduced *C. sacchariphagus*. However, the results presented in this paper show how important it is to collect as much relevant information as possible when assessing field trials. Initial surveys were designed to collect as much information as possible on the performance of indigenous and introduced parasitoids on populations of *C. sacchariphagus*. It soon became evident, however, that the varieties newly introduced to the estates were attacked to different degrees by *C. sacchariphagus*. More detailed surveys were then undertaken.

This paper thus provides the first evidence that:
- The same southern African varieties differ in their susceptibility to *C. sacchariphagus*, depending on whether they are planted in irrigated or dryland conditions (e.g. N19).
- Varieties that show high resistance to *E. saccharina*, show low resistance to *C. sacchariphagus* (e.g. N21 and N12).
- The biologies and behaviours of different stalk borers on the same variety may determine the amount of susceptibility of that variety.
- A leaf damage rating system may be an efficient, simple, quick and accurate varietal assessment tool against *C. sacchariphagus.*

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