

# ANTIBIOTIC RESISTANCE OF SUGARCANE VARIETIES TO *ELDANA SACCHARINA* (LEPIDOPTERA: PYRALIDAE) INDICATED BY DIET-INCORPORATION BIOASSAYS

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Diet-incorporation bioassays were performed to investigate (1) the relative antibiotic effects of five commercial varieties of sugarcane on the survival, development, longevity and fecundity of the sugar-cane borer, *Eldana saccharina* Walker (Lepidoptera: Pyralidae) (eldana), and (2) the potential of such bioassays as a screening method for varietal resistance to this pest. Diet-incorporation bioassays have been used only once previously (Meagher *et al.*, 1996) to determine antibiosis of sugarcane tissue to a stalk borer.

Forty stalks each of the sugarcane varieties N11, N12, N16, N21 and NCo376 were cut from an entomology field trial at the South African Sugar Association Experiment Station (SASEX) farm at La Mercy during September 1996. The material was shredded, dried at 50°C for two days, and powdered for incorporation into the diet mix used in the SASEX eldana rearing unit (Graham and Conlong, 1988). The quantity of chickpea normally included in the diet (100 g/L) was reduced by 50% to augment the proportion of powdered cane and enhance any antibiotic effects it may have. The diet mix, made up separately from each variety, was dispensed into 22, 32-cell rearing trays, the latter constituting replications for each variety. Each cell was inoculated with one, and sometimes two, neonate larva. The trays were placed in larval rearing rooms under the same conditions as the routine larval culture (23,5°C; RH 77%). The NCo376 mix was taken as the control, although some comparisons were also made with the routine culture.

At 14 days, a sub-sample of seven trays per variety was harvested and the number of larvae per instar and total mass of larvae recorded, to assess whether differences in survival and development in each variety mix were apparent at an early stage (first to third instar). At 36 days, the remaining 15 trays for N11, N12 and NCo376 were harvested but, due to the delayed development of larvae in the N16 and N21 diets, only seven trays of each were harvested at 36 days and the balance of eight trays each were harvested at 41 and 45 days, respectively. Larval numbers per instar, and pupal numbers and mass, were recorded for all trays. Longevity and lifetime fecundity of a maximum of 50 mated female moths reared from each variety mix was also determined.

At 14 days, varietal mixes already differed significantly in mean numbers of larvae per tray in the second and third instars and the total mass of larvae per tray (Table 1). Planned comparisons between varieties showed that the N21 mix contained significantly more second instar larvae and significantly fewer third instar larvae than the other variety mixes, with the exception of third instar larvae in the N16 mix (Table 1). However, total numbers of larvae in all mixes did not differ significantly at 14 days. Total larval mass was also significantly lower in the N16 and N21 mixes than in most of the other mixes; although larval weight in the N21 mix was lower than that in the N11 mix, the difference was not significant. Larval mass was significantly greater in the NCo376 mix than in all the other mixes (Table 1).

**Table 1**  
Performance of *E. saccharina* larvae after 14 days in diet containing powdered stalk tissue from five commercial varieties of sugarcane.

Variety	Number of larvae			Total larval mass (mg)
	Instar 2	Instar 3	Total	
N11	18,8 ± 4,1a	18,7 ± 2,6a	37,6 ± 6,0	343,7 ± 48,8ac
N12	15,7 ± 3,1a	29,6 ± 4,1a	45,6 ± 1,9	506,7 ± 52,7ad
N16	18,4 ± 5,6a	6,7 ± 2,6b	25,3 ± 8,3	150,7 ± 42,0b
N21	34,4 ± 2,4b	1,7 ± 1,1b	37,0 ± 3,1	161,3 ± 20,1bc
NCo376	12,1 ± 3,4a	28,8 ± 7,2a	41,3 ± 8,3	628,7 ± 116,5d

Means ±SE followed by the same letter are not significantly different ( $p > 0,05$ , Fisher's LSD test). ANOVA for instar 2:  $F=4,8$ ,  $df=4$ ,  $p < 0,005$ ; for instar 3:  $F=9,5$ ,  $df=4$ ,  $p=0,0001$ ; for total larvae:  $F=1,5$ ,  $df=4$ ,  $p=0,21$ ; for total mass:  $F=10,6$ ,  $df=4$ ,  $p < 0,0001$ .  $N=7$  for all treatments.

At 36 to 45 days, total numbers of third to sixth instar larvae and pupae harvested from the remaining 15 trays were significantly different across variety mixes (Pearson  $X^2=875,7$ ,  $df=16$ ,  $p<0,0001$ ). Although ANOVA on individual stages was not performed, the NCo376 mix produced the largest number of pupae, followed by N12, N11, N16 and N21 (Table 2). Pupal mass and female fecundity were also significantly higher on the NCo376 (intermediate resistance) than on the N16 (susceptible) diet mix (Table 2). The N12 (resistant) diet mix produced significantly heavier pupae and more fecund females than the N16 mix. Longevity of females reared on the N12 diet mix was also higher than that of females reared on the other variety mixes, although differences were not significant (Table 2). However,

the relatively poor development, fecundity and longevity of *eldana* reared on the N21 diet mix was striking (Table 2). Larval development in particular was severely delayed relative to other varieties; the majority (29%) of individuals recovered at final harvest were fourth instar larvae. Pooling results from all varieties gave significant correlations ( $p<0,001$ ) between fecundity and pupal mass, and between longevity and fecundity. Female pupal mass and longevity were not correlated. In those variety mixes with reduced pupal production (N21 and N16), sex ratios were male-biased. Of the three dependent variables analysed at the individual level, pupal mass explained most of the variance in the ANOVA, followed by fecundity and longevity ( $r^2=0,39$ ,  $0,10$  and  $0,04$ , respectively).

**Table 2**  
Adult female pupal mass, fecundity and longevity of *E. saccharina* reared for 36 to 45 days in diet containing powdered stalk tissue from five commercial varieties of sugarcane.

Variety	Total pupae (N)*	Pupal mass (mg)	Fecundity**	Longevity (days)
N11	332 (43)	159,9 ± 3,4ac	423,7 ± 24,8ac	6,5 ± 0,4
N12	426 (48)	148,6 ± 2,4b	464,3 ± 25,8a	7,6 ± 0,3
N16	133 (22)	137,1 ± 3,8c	365,7 ± 40,4bc	7,0 ± 0,5
N21	52 (7)	100,9 ± 5,7d	217,0 ± 30,0b	6,0 ± 0,2
NCo376	617 (45)	168,4 ± 3,1e	428,7 ± 32,7a	6,8 ± 0,4
Culture	– (28)	150,9 ± 7,4ab	–	–

\*N = size of sample taken for determination of female pupal mass, fecundity and longevity. \*\*Fecundity represents total eggs laid over lifetime of female. Means ±SE followed by the same letter are not significantly different ( $p>0,05$ , Fisher's LSD test). ANOVA for pupal mass:  $F=14,2$ ,  $df=5$ ,  $p<0,0001$ ; for fecundity:  $F=4,3$ ,  $df=4$ ,  $p<0,003$ ; for longevity: Kruskal-Wallis  $H=5,9$ ,  $df=4$ ,  $p=0,21$ .

The results suggest that antibiotic factors, efficacious in intact plant material, may be altered or lost in the process of incorporating the plant material into the diet mix, leading to relatively resistant varieties such as N12 showing low antibiosis and susceptible varieties such as N16 showing high antibiosis. NCo376, a variety of intermediate resistance, displayed the lowest degree of antibiosis.

This casts doubt on the usefulness of diet-incorporation bioassays as a screening technique. However, the result obtained from N21 suggests that particularly resistant varieties may be identifiable using this technique. Since nutrients (especially N) in the diet were not limiting, the differences are probably due to other (antibiotic) factors inherent in each variety.

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### REFERENCES

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