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Blackheart resistance in three clones of pineapple [*Ananas comosus* (L.) Merr.] in subtropical Queensland

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Summary. Smooth Cayenne Queensland clone 13, and intergroup hybrids 53-116 and 73-50 were harvested in mid July 1994 and evaluated for blackheart resistance. Relative to C13, hybrids 73-50 and 53-116 had some level of field resistance to blackheart but only 53-116 was without blackheart after storage at 10ºC for 14 days followed by 20ºC for 8 days. Hybrid 53-116 had a high vitamin C content compared with clone 13. However, 73-50 had the highest level of vitamin C but still developed blackheart, although the symptoms were not as severe as C13. This indicates that factors other than vitamin C may play a role in blackheart resistance. The level of field resistance to blackheart shown by 73-50 may be sufficient for winter production in some sites in south-east Queensland. Field testing is needed to determine the value of this resistance.

Introduction

The fresh pineapple industry in Queensland has a national wholesale value of about $\$30 \times 10^6$/year (Swete-Kelly and Bagshaw 1993). Smooth Cayenne is the predominant cultivar. While it is acceptable as a fresh fruit in the summer months, fruits ripening in the winter have several major quality faults. They are high in acid, low in sugars and prone to the internal chilling disorder, blackheart, also known as internal browning, endogenous brown spot and internal brown spot (Teisson *et al.* 1979; Swete-Kelly and Bartholomew 1993).

Consumer and market surveys have indicated blackheart as a major deterrent to repeat sales. During market surveys conducted in Melbourne and Sydney in winter, 45% of fruit displayed blackheart symptoms. Field-induced blackheart, as opposed to that induced by storage, was found to be the most prevalent (Swete-Kelly and Bagshaw 1993).

A pineapple hybridisation program was initiated in 1991 to develop hybrids for subtropical Queensland that are more suited for the fresh fruit market. Blackheart resistance is a major criterion for selection. Importation and evaluation of hybrids developed in others regions of the world is an important part of the breeding and selection program.

The Pineapple Research Institute of Hawaii (PRI) hybrid 73-50 was imported into Queensland in 1987. Current trials indicate that it possesses many of the qualities required in a fresh market fruit and it has been used extensively as a parent in the Queensland fresh market pineapple hybridisation program. Grower observation suggests it may have some resistance to blackheart but there are no published studies on this aspect. Another PRI hybrid, 53-116, is reputed to have good resistance to blackheart (Wassman 1983; Swete-Kelly and Bartholomew 1993) and was used in this trial, along with the more susceptible Smooth Cayenne clone 13 (C13), to examine the relative blackheart resistance of 73-50.

Materials and methods

In mid July 1994, 20 fruit each of C13, and intergroup hybrids 53-116 and 73-50 were harvested from a property at Wamuran, near Brisbane. Fruit as close to eating ripe as could be visually judged were selected. This was achieved by cutting a small number of fruit at different stages of shell colour. The cultivars were all grown within a 100 m radius of each other in a similar soil type and subject to similar cultural practices. There were no obvious site differences.

The incidence of blackheart at harvest was assessed by cutting 10 fruit of each cultivar into longitudinal quarters. The spots were then counted, and ratings (Table 1) given for the darkness of the spots and the percentage of the total area covered by them. Vitamin C was measured using the dichlorophenol titration method (Anon. 1990) on juice extracted from a horizontal centre slice of fruit using a hand-operated juice press.

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Table 1. Ratings to describe blackheart incidence of pineapple

Darkness of spots	Area of spots (%)
0, No spots	0, none
1, Slight (small faint spots often more translucent than brown)	1, 1–10
2, Moderate (mainly brown, some translucent)	2, 11–30
3, Severe (very dark brown to black)	3, 31–60
	4, 61–100

The remaining 10 fruit of each cultivar were placed in a coolroom at 10°C for 14 days followed by a further 8 days in an incubator at 20°C. This temperature regime has been shown to induce severe blackheart in Smooth Cayenne and Queen pineapple (Smith 1982). Upon removal, the fruit were cut as described previously and rated for the incidence of blackheart.

Treatments were compared using analysis of variance for a completely randomised design, followed by the protected least significant difference procedure. Blackheart ratings were compared with the constant zero using Student's *t*-test. Spot numbers were $\sqrt{(x + 0.5)}$ transformed before analysis. Both transformed and back-transformed data are presented.

Results and discussion

When data were collected immediately after harvest (Table 2) only C13 exhibited any symptoms. There were mild symptoms in 50% of C13 fruit.

After storage, C13 exhibited severe blackheart symptoms in all fruit (Table 3) and in most the spots had coalesced. In 73-50, all fruit had discrete spots of slight to moderate darkness. Hybrid 53-116 was free of blackheart symptoms after storage. While reference has been made to the blackheart resistance of 53-116 (Wassman 1983; Swete-Kelly and Bartholomew 1993) this is the first time this has been documented experimentally to the authors' knowledge.

Table 2. Vitamin C content and blackheart ratings of fruit of hybrids 73-50 and 53-116 and the Smooth Cayenne C13 taken immediately after harvest

Values represent the mean of 10 fruit
s.e.m. is the standard error of the mean; for vitamin C it applies to all 3 treatment means, while for blackheart ratings it applies only to the C13 mean
Vitamin C means followed by the same letter are not significantly different at P = 0.05

Characteristic	Hybrid or clone			s.e.m.
	73-50	53-116	C13	
Vitamin C (mg/100 mL)	64.6a	35.8b	14.6c	1.8
Number of fruit affected	0	0	5	
Number of spots[A]	0	0	3.3 (2.0)[B]	(0.5)
Darkness of spots	0	0	0.5[B]	0.2
Rating of total area of spots	0	0	0.5[B]	0.2

[A] $\sqrt{(x + 0.5)}$ transformed. The back-transformed mean is presented, with the transformed value in parentheses.
[B] Significantly greater than 0 (P<0.05).

Table 3. Blackheart ratings after storage of fruit of the pineapple hybrids 73-50 and 53-116 and the Smooth Cayenne C13 at 10°C for fourteen days and at 20°C for eight days

Values represent the mean of 10 fruit
s.e.m. is the standard error of the mean; for number of spots it applies only to the 73-50 mean, while for darkness of spots and rating of area affected it applies to the 73-50 and C13 means
Values within each row followed by the same letter are not significantly different at P = 0.05

Characteristic	Hybrid or clone			s.e.m.
	73-50	53-116	C13	
Number of fruit affected	10	0	10	
Number of spots[A]	20 (4.5)[B]	0	Coalesced	(0.4)
Darkness of spots	1.9b[B]	0	2.6a[B]	0.2
Rating of total area of spots	1.7b[B]	0	3.1a[B]	0.3

[A] $\sqrt{(x + 0.5)}$ transformed. The back-transformed mean is presented, with the transformed value in parentheses.
[B] Significantly greater than 0 (P<0.05).

The data for vitamin C content of Smooth Cayenne is in general agreement with that quoted by Miller (1951) at 17.8 mg/100 mL. Hybrid 53-116 was high in vitamin C compared with C13 and this is generally considered to play a significant role in blackheart resistance in pineapple (Miller and Heilman 1952; Van Lelyveld and De Bruyn 1977; Teisson *et* al. 1979; Hassan *et al*. 1985; and Hassan 1993). However, 73-50 had a high vitamin C content but still developed blackheart, although the symptoms were not as severe as C13. This indicates that factors other than vitamin C content play a role in blackheart resistance. The large number of spots that developed in 73-50 suggests a significant amount of cold-damaged tissue. However, the spots were not as dark as those in C13 suggesting that darkening was inhibited to some extent, possibly by the high vitamin C content.

There are several fruit characteristics that could determine the level of blackheart resistance. While the physiological process that results in browning has been proposed (Van Lelyveld and De Bruyn 1977; Rohrbach and Paull 1985), there are no published studies that quantify the physiological differences between blackheart-resistant and blackheart-susceptible varieties. For instance, peroxidase activity is known to differ between some pineapple cultivars (Miller 1951) but measurements have not been made in resistant and susceptible cultivars. In apples, the degree of browning has been related to the ratio of phenols to ascorbic acid (Weurman and Swain 1955). Polyphenol oxidase is considered the principal enzyme involved in blackheart development in pineapple and has been shown to increase 30 times in pineapple fruit in response to chilling (Paull and Rohrbach 1985). The ratio of polyphenol oxidase activity to antioxidant level in blackheart-resistant and susceptible cultivars of pineapple is presently unknown but could be important.

The only antioxidant which has received any attention in pineapple is vitamin C. There are many plant compounds that possess antioxidant properties. As an example, vitamin E is an antioxidant in plant tissues that in some cases may be more potent than vitamin C (Packer *et al*. 1979; Fryer 1992), but no published data on the vitamin E content of pineapple were found. Wang and Baker (1979) postulated that ethoxyquin and sodium benzoate decreased the oxidation of cucumber membrane lipids by scavenging free radicals of fruits when exposed to low temperature. Membrane characteristics are also considered to be involved in cold tolerance in many plant species (Lyons 1973). The membrane characteristics of blackheart-susceptible and tolerant pineapple cultivars have not been studied.

The blackheart resistance of 73-50 may be sufficient for production in some sites in south-east Queensland but further field testing is needed. However, 53-116 appears to be the only hybrid in Australia with very good, if not complete, resistance to blackheart.

While genetic resistance to blackheart has been identified the mechanism for this resistance is unknown. It is important that the physiological basis for this resistance is determined if this characteristic is to be used in hybridisation programs where blackheart resistance is a selection criterion.

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