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www.publish.csiro.au/journals/ajea *Australian Journal of Experimental Agriculture*, 2006, **46**, 107–113

Towards the development of a Cavendish banana resistant to race 4 of fusarium wilt: gamma irradiation of micropropagated Dwarf Parfitt (*Musa* spp., AAA group, Cavendish subgroup)

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Abstract. 'Dwarf Parfitt', an extra-dwarf Cavendish cultivar with resistance to subtropical race 4 *Fusarium oxysporum* f. sp. *cubense* (*Foc*), was gamma irradiated at a dose of 20 Gy and putative mutants were recovered with improved agronomic characteristics. Further screening of putative mutants for improved yield and fruit size, as well as a degree of resistance to fusarium wilt, led to the selection of a line (DPM25) with improved productivity when grown on soils infested with subtropical race 4 *Foc*. DPM25 was equal to the industry standard, 'Williams', in every agronomic trait measured and it consistently showed a lower incidence of fusarium wilt. Further improvement of field resistance to race 4 *Foc* is needed in DPM25 and further cycles of mutation induction and selection is an option discussed.

Additional keywords: mutation breeding, somaclonal variation, tissue culture.

Introduction

The Australian banana industry, and indeed the world export trade, is based on the Cavendish (*Musa* spp. AAA group) subgroup of dessert bananas. Although highly productive in a range of environments, Cavendish is susceptible to a particularly devastating race of *Fusarium oxysporum* f. sp. *cubense* (*Foc*), race 4. Because of the persistent nature of *Foc* in soils and the lack of effective chemical control strategies, the development of race 4 *Foc* resistant cultivars has been a priority in genetic improvement programs for the past 20 years (Moore *et al*. 2001).

Conventional breeding strategies have not delivered a Cavendish cultivar resistant to *Foc* or a suitable replacement largely due to the extreme sterility encountered in this subgroup of bananas. The Cavendish subgroup is composed of triploid (AAA) cultivars that are totally seedless (Robinson 1996). Attention is being given to biotechnology approaches to improve Cavendish cultivars, including genetic transformation (Becker *et al*. 2000), but currently *in vitro* mutation breeding programs have delivered the most promising results (Smith *et al*. 2005).

The Taiwan Banana Research Institute established a Cavendish breeding program in 1985 based on field screening somaclonal variants for resistance to race 4 *Foc* following mass *in vitro* propagation. The program has produced 12 resistant clones; however, a good substitute for 'Giant Cavendish' has not yet been obtained (Hwang 2001). Whereas some resistant clones carry inferior agronomic characters, others have fruit quality defects. Nevertheless a race 4 *Foc* resistant clone, 'Tai Chiao No. 1', was released for commercial planting in 1992 and shows significantly lower incidence of fusarium wilt when planted in race 4 *Foc* infested soils (5.1%–6.5%) compared with the susceptible Giant Cavendish (42.6%–69.0%) (Hwang 2001). Further cycles of *in vitro* propagation and selection of somaclonal variants with improved agronomic characters while retaining race 4 *Foc* resistance have continued with Tai Chiao No. 1 (Tang and Hwang 1998). Recently a new resistant clone superior to the agronomic characters and productivity of Tai Chiao No. 1 has been obtained (Hwang 2001). Other *in vitro* mutation breeding programs based on either somaclonal variation (de Beer *et al*. 2001) or gamma irradiation (Smith *et al*. 1994) have produced more resistant clones but have deleteriously altered the agronomic characters of the original Cavendish cultivar. The work reported in this present paper has taken a different strategy. Instead of starting with a

 10.1071/EA04175 0816-1089/06/010107

commercial Cavendish cultivar susceptible to race 4 *Foc* and attempting to increase its resistance, an extra-dwarf Cavendish cultivar called 'Dwarf Parfitt' resistant to race 4 *Foc* (Pegg *et al*. 1996) was irradiated and putative mutants were screened for improved agronomic features while attempting to retain field resistance to race 4 *Foc*.

Materials and methods

Plant materials and establishment of micropropagated bananas

Conditions for the initiation and growth of micropropagated bananas have been described previously (Hamill *et al*. 1992). Briefly, Williams and Dwarf Parfitt were initiated on a Murashige and Skoog (1962) basal medium supplemented with 10 μmol/L benzylaminopurine, 2% sucrose and 0.8% Difco-Bacto agar. This medium also supported rapid shoot multiplication. Cultures were incubated at 28°C with a 16 : 8 h light : dark photoperiod. Cool white fluorescent tubes provided a photon flux density at the culture surface of about 80 μmol quanta/m^2.s. Before acclimatisation in the glasshouse, plants were subcultured to a hormone-free medium for root initiation and the development of vigorous, single shoots. Plants were established in the field when they reached a height of 20 cm in 2.5-L planter bags.

Mutation induction

The irradiation procedure described by Smith *et al*. (1994) was originally developed for Williams and involved the transfer of explants, containing the shoot tip and ensheathing leaf bases obtained from micropropagated plants, to Petri dishes that contained a multiplication medium. Cultures were incubated for 7 days before exposure to gamma radiation from a ^{60}Co source. The doses used were 0, 10, 20, 30, 40 and 50 Gy. Dosage was calculated using Fricke dosimetry (O'Donnell and Sangster 1970). Plants were subcultured for 2 or 3 cycles on a multiplication medium before root initiation and acclimatisation in the glasshouse.

Establishment and maintenance of field trials

The experimental site has been described previously (Smith *et al.* 1998) and formed part of a commercial Cavendish plantation at Wamuran in subtropical Queensland (27°S, 153°E) that was abandoned because of losses to fusarium wilt caused by subtropical race 4 *Foc*. The soil is classified as a yellow ferrosol (gleyed podzolic soil) and is a heavy clay-clay loam of pH 5.5–6.0. The site was uniformly infested with 3 subtropical race 4 vegetative compatibility groups (VCGs): 0120, 0129 and 01211 (Pegg *et al*. 1996). A series of field trials were carried out over an extended period, commencing in November 1991 and concluding in April 2003 (Table 1). Some were designed to assess the putative mutants' response to race 4 *Foc* whereas others also included measurements of yield, productivity and fruit size. The field trial planted in November 1995 from micropropagated plants was in an area unaffected by fusarium wilt. All other trials were at race 4 *Foc* infested sites as described above. Planting material was either micropropagated plants or 'bits', which are sections of the rhizome containing a well developed growing point and pared to an average weight of 700–800 g. Plants were established at a density of 1600 plants/ha with a spacing of 2.5 by 2.5 m and grown using standard commercial practices (Gall and Vock 1994).

Measurements

Blocks were visited weekly and when banana plants started bunching, the pseudostem height (from the soil surface to the point of intersection of the 2 upper-most leaves) and pseudostem circumference (at 30 cm above ground) were measured, the number of green leaves was counted and the date of bunch emergence recorded. At harvest the following parameters were recorded: date, number of green leaves, total leaf area [calculated using the formula of Robinson and Nel (1985); (leaf length × maximum leaf width) × 0.83 (conversion factor), bunch weight, bunch stalk weight, number of hands, number of fingers in bunch, number of fingers in the third proximal hand and the length of the middle finger of the outer whorl of the third proximal hand. Agronomic traits were not recorded from plants showing obvious symptoms of fusarium wilt.

Assessment of fusarium wilt infection

External symptoms of fusarium wilt were recorded at the final, destructive harvest when internal symptoms were also recorded. Plants were judged to have external symptoms of disease if they displayed any sign of wilting, yellowing of foliage, petiole buckling or splitting of the pseudostem base. Severity ratings were on a scale of 1–3 that was developed by the International Network for the Improvement of Banana and Plantain (Jones 1994): 1, no symptoms; 2, mild symptoms; and 3, severe symptoms. Internal symptoms were recorded from plants that were removed from the soil and cut transversely through the rhizome about one-quarter of the way above the rhizome's base. The cut surface of the rhizome was rated for discolouration on a scale of 1–6 (Jones 1994): 1, no vascular discolouration; 2, isolated points of vascular discolouration; 3, less than one-third of the vascular tissue discoloured; 4, one- to two-thirds of the vascular tissue discoloured; 5, greater than two-thirds of the vascular tissue discoloured; and 6, total vascular discolouration or discolouration of leaf bases or both.

Experimental design and statistical analysis

Each trial used a completely randomised design and had unequal replication for each mutant line and cultivar due to differences in availability of planting material. For the yield data from the field trials planted in November 1991 and 1995, mutant lines were compared using analysis of variance; whereas, for the field trial planted in October 1998, line comparisons were made using residual maximum likelihood with lines as a fixed effect (tested using the Wald statistic), and rows and columns as random effects. For these trials, the protected LSD test was used to compare treatment means using the significance level of $P = 0.05$. Severity of race 4 *Foc* infection was analysed using the Kruskal–Wallis test. Incidence of fusarium wilt was compared using generalized linear models for a binomial distribution with logit link followed, where significant, by pairwise *t*-tests. Again testing was at $P = 0.05$.

Table 1. Outline of experiments conducted to assess performance of irradiated Dwarf Parfitt mutant lines

Planting material	Planting date	Completion date	No. of lines	Assessment
Bits[A]	Nov. 1991	Nov. 1993	22	Plant crop data; *Foc*[B]
Micropropagated	Nov. 1992	June 1993	22	*Foc*
Micropropagated	Nov. 1992	Oct. 1993	22	*Foc*
Micropropagated	Nov. 1995	Oct. 1998	7	Plant and ratoon crop data
Micropropagated	Nov. 1996	Oct. 1997	7	*Foc*
Bits	Oct. 1998	Apr. 2003	1	Plant and ratoon crop data; *Foc*

[A]Bits, sections of the rhizome.

[B]*Foc*, plants were assessed for symptoms of fusarium wilt.

Results

The LD_{50} for micropropagated Williams was about 30 Gy but shoot multiplication and general vigour of these plantlets was poor. The optimal dose was set at 20 Gy and used for mutation breeding trials. At this dosage, visual changes were apparent and plant survival, at 73%, was sufficiently high to make the technique practical on a larger scale. The radiosensitivity of the Cavendish clones used in these experiments compares well with that found by Novak *et al*. (1990) for 'Grande Naine'.

Following irradiation of about 500 Dwarf Parfitt explants at a dose of 20 Gy, 35 plants were observed in the field planting that possessed improved agronomic features. Plants were significantly taller than the extra-dwarf mother plants, they bunched earlier, their yield was greater and severe choking was eliminated (Smith *et al*. 1994). Bits of rhizome containing a growing point were removed from plants displaying no symptoms of fusarium wilt and were replanted in an adjacent *Foc* infested site, together with planting material from Williams and Dwarf Parfitt cultivars. Plant crop data of the M_1V_4 generation (i.e. the fourth vegetative cycle following 1 mutation induction treatment) revealed large differences in yield (Table 2) and assessments of micropropagated plants revealed differences in susceptibility to fusarium wilt between different mutant lines (Table 3).

There was a large group of mutant lines whose fruit bunch weight and finger length were comparable with those of Williams cultivars, as well as several lines with significantly poorer bunch weights and finger lengths. Dwarf Parfitt was significantly poorer, from an agronomic perspective, than all mutant lines selected. However, it had a very low incidence of fusarium wilt (an overall incidence of 8%, consisting of 1 in 5 plants with external symptoms in August 1993 and 1 in 21 plants with internal symptoms in October 1997), whereas there was a great range of incidence of fusarium wilt in the mutant lines (0–80%). Lines DPM2, 22, 25, 15 and 16 suggested fusarium wilt resistance was possible without a large loss in productivity and these, as well as several more resistant lines, were established as micropropagated plants for further tests and evaluation.

Evaluation of these more resistant lines grown on several more sites and during various seasons confirmed the

Table 2. Bunch weight and finger length of Williams, Dwarf Parfitt and putative mutants of Dwarf Parfitt at M_1V_4 generation following gamma irradiation

Cultivars and mutant lines were ranked according to mean bunch weight and finger length in the plant crop. Field trial was planted in November 1991 using bits (sections of rhizome) Means within each column followed by the same letter are not significantly different at $P = 0.05$

Cultivar or mutant line	No. of plants	Bunch weight (kg)	Cultivar or mutant line	No. of plants	Finger length[A] (cm)
DPM25	6	34.8a	Williams	18	23.1a
Williams	18	33.5a	DPM22	6	23.0ab
DPM2	7	32.8ab	DPM8	5	22.7abcd
DPM5	9	32.5ab	DPM15	9	22.6abc
DPM16	4	32.1abcd	DPM2	7	22.4abcde
DPM1	10	32.0abc	DPM16	4	22.1abcdef
DPM19	6	31.5abcd	DPM18	7	22.1abcdef
DPM15	9	31.3abcd	DPM13	8	21.9abcdef
DPM12	7	30.7abcd	DPM11	9	21.8abcdef
DPM27	11	30.5abcd	DPM19	6	21.8abcdef
DPM13	8	30.0abcde	DPM27	11	21.6bcdef
DPM24	8	29.2bcde	DPM24	8	21.6bcdef
DPM11	9	29.1bcde	DPM12	7	21.4bcdefg
DPM3	12	28.6bcde	DPM21	11	21.4bcdefg
DPM22	6	28.6bcdef	DPM25	6	21.4bcdefg
DPM18	7	28.6bcdef	DPM9	7	21.1cdefg
DPM8	5	28.1bcdefg	DPM1	10	21.1defg
DPM23	9	27.7cdefg	DPM4	14	20.9efg
DPM4	14	27.2defg	DPM5	9	20.8efg
DPM21	11	25.4efg	DPM10	8	20.7fgh
DPM10	8	23.9fgh	DPM3	12	20.6fg
DPM14	6	22.7gh	DPM23	9	19.9gh
DPM9	7	20.0h	DPM14	6	18.9h
Dwarf Parfitt	5	12.1i	Dwarf Parfitt	5	13.0i
		s.d. = 5.06			s.d. = 1.69

[A]Finger length was measured from the middle finger of the outer whorl of the third hand from the proximal end.

agronomic superiority of DPM25 compared with the other mutant lines (Table 4). Williams and DPM25 consistently had better yields and fruit size than other lines, whereas Dwarf Parfitt and DPM10 had consistently poor yields and fruit size. The majority of the DPM lines, including DPM2, DPM15, DPM16 and DPM22 also had a tendency to choke, whereas DPM25 did not. This is a condition when the bunch fails to emerge normally from the pseudostem and typically occurs when temperatures remain below 15°C for long periods in subtropical climates (Stover and Simmonds 1987). DPM25 also showed greater resistance to subtropical race 4 *Foc* than Williams (Table 3) and was therefore selected for further evaluation with Williams.

Bits were collected and a field trial established and maintained in a subtropical race 4 *Foc* infested block from 1998–2003. Bits were used owing to their greater resistance to *Foc*, compared with micropropagated plants (Smith *et al.* 1998), and were grown using commercial practices. A large number of traits were measured in both plant and ratoon crops of Williams and DPM25 and there were no significant differences in measured traits between Williams and DPM25 (Table 5). According to the agronomic traits studied, it was not possible to distinguish DPM25 from Williams in these trials.

During a period of high disease pressure prior to fusarium wilt assessments made in June 1993, there was a significant difference between Williams and DPM25 for severity and incidence of internal ratings of fusarium wilt ($P<0.05$). However, when disease pressure was less severe, no significant differences were observed even though Williams had consistently worse infection and incidence of disease (Table 6). 'Bluggoe', a cultivar that is highly susceptible to subtropical race 4 *Foc*, was significantly worse ($P<0.05$) than these 2 cultivars for all comparisons made in April 2003 (internal and external for severity and incidence). Dwarf Parfitt, as expected, had a lower overall incidence of fusarium wilt than Williams and DPM25, and had significantly less internal symptoms ($P<0.05$).

Discussion

Mutation breeding is a viable strategy for the improvement of bananas and plantains. The development of fusarium wilt resistant Cavendish bananas with acceptable yield and fruit quality is possible but has involved a considerable investment of time and resources as demonstrated by de Beer *et al*. (2001) and Hwang (2001). Their strategy was to develop subtropical race 4 *Foc* resistance in a commercially

Table 3. Incidence of fusarium wilt for Williams, Dwarf Parfitt and putative mutants of Dwarf Parfitt (obtained by gamma irradiation)

Cultivars and mutant lines were ranked according to the percentage of plants showing symptoms of fusarium wilt in the plant crop. The incidence of external fusarium wilt was assessed on August 1993 using plants established from bits (sections of rhizome) 21 months after planting. The incidence of internal fusarium wilt was assessed on October 1997 using micropropagated plants 11 months from planting

	Aug. 1993			Oct. 1997	
Cultivar or mutant line	No. of plants	External incidence of wilt (%)	Cultivar or mutant line	No. of plants	Internal incidence of wilt (%)
DPM2	7	0	Dwarf Parfitt	21	5
DPM22	7	0	DPM22	14	7
DPM10	9	11	DPM16	20	10
DPM24	9	11	DPM10	19	21
DPM19	9	11	DPM2	13	23
DPM14	7	14	DPM25	20	30
DPM3	13	15	DPM18	17	35
DPM25	10	20	DPM15	22	36
Dwarf Parfitt	5	20	Williams	8	50
DPM16	5	20	—	—	—
DPM18	9	22	—	—	—
DPM13	8	25	—	—	—
DPM15	11	27	—	—	—
DPM27	11	27	—	—	—
DPM4	14	29	—	—	—
DPM9	10	30	—	—	—
Williams	22	32	—	—	—
DPM1	12	33	—	—	—
DPM12	9	33	—	—	—
DPM23	10	40	—	—	—
DPM21	13	46	—	—	—
DPM5	11	55	—	—	—
DPM11	10	60	—	—	—
DPM8	5	80	—	—	—

accepted cultivar. Our strategy also demonstrates that it is possible to start with an agronomically inferior but race 4 *Foc* resistant cultivar and select for lines with improved yield, productivity and fruit quality following mutation induction. However, both strategies involve trade-offs. In the program of Hwang (2001), it involved sacrificing improved agronomic characteristics for greater resistance to the pathogen. The results presented in this present paper have shown that mutant lines (e.g. DPM25) can be selected that are equal to the industry standard, Williams; however, the high degree of resistance to fusarium wilt shown by Dwarf Parfitt has been diminished in the process. Nevertheless, DPM25 has been shown to be significantly less affected by subtropical race 4 *Foc* than Williams and the incidence of infection has consistently been lower. Trials are currently underway in the Northern Territory to assess the reaction of DPM25 to tropical race 4 *Foc*, a far more devastating race of *Foc*.

Given that yield and fruit quality of Williams and DPM25 are similar, significantly improved productivity in soils infested with race 4 *Foc* is possible, as DPM25 has consistently shown a lower incidence of infection than Williams (Table 6). Even small improvements in resistance (e.g. a 5% reduction in disease incidence) will ensure an overall higher yield per hectare. However, we believe a greater level of resistance is needed to obtain industry acceptance of this line. That DPM25 can be further improved is demonstrated by Tang and Hwang (1998) where recurrent mutation breeding and selection within the Tai Chiao No. 1 cultivar succeeded in decreasing incidence of infection from 11–20% to 3–12%, and in significantly improving agronomic traits, such as plant height. Other strategies may

Table 4. Across years comparison of Williams, Dwarf Parfitt and putative mutants of Dwarf Parfitt (obtained by gamma irradiation)

Cultivar and mutant lines were ranked according to pseudostem height, bunch weight and finger length in the plant crop

Cultivar or mutant line	Plant crop[A] Nov. 1991–Nov.1993	Plant crop[B] Nov.1995–Oct.1998	Ratoon crop[B] Nov.1995–Oct.1998
	Pseudostem height (m)		
Williams	2.92	2.28	2.53
DPM25	2.63	2.31	2.41
DPM16	2.88	1.79	1.79
DPM2	2.64	1.71	1.79
DPM22	2.45	1.68	1.81
DPM18	2.23	1.72	1.78
DPM15	2.39	1.67	1.72
DPM10	1.89	1.62	1.74
Dwarf Parfitt	1.32	1.18	1.10
	Bunch weight (kg)		
Williams	33.5	25.6	24.2
DPM25	34.8	24.2	23.3
DPM16	32.1	20.5	20.4
DPM2	32.8	20.2	20.4
DPM22	28.6	20.0	20.0
DPM15	31.3	20.8	18.6
DPM18	28.6	21.1	19.4
DPM10	23.9	14.8	15.9
Dwarf Parfitt	12.1	9.0	6.5
	Finger length[C] *(cm)*		
Williams	23.1	20.9	21.5
DPM25	21.4	20.8	20.8
DPM15	22.6	20.2	19.8
DPM22	23.0	19.5	19.7
DPM2	22.4	18.9	19.7
DPM16	22.1	19.1	19.0
DPM18	22.1	19.1	19.0
DPM10	20.7	17.7	18.0
Dwarf Parfitt	13.0	11.4	9.8

[A]Field trial was planted using bits (sections of rhizome).
[B]Field trial was planted using micropropagated plants.
[C]Finger length was measured from the middle finger of the outer whorl of the third hand from the proximal end.

need to be used to lower incidence of infection in Cavendish plantations, such as crop rotation practices that are known to reduce soil inocula levels (Moore *et al*. 2001).

The resistance of Dwarf Parfitt to race 4 *Foc*, compared with the susceptible Williams, was believed to have resulted from its greater retention of leaf area during winter, its higher chlorophyll concentration and more effective photosynthetic activity during winter (Moore *et al*. 1993; Pegg *et al*. 1996). Because host defence reactions, such as the formation of gums, gels and tyloses used to block the invading pathogen, are presumably driven by photoassimilates, retention of the crop's high photosynthetic capabilities during winter was believed to effectively prevent widespread epidemics of fusarium wilt from occurring during spring and summer. In this present study, there was no difference in leaf number or leaf area between DPM25 and Williams at any stage

Table 5. A comparison of agronomic traits of Williams and DPM25 in a plant and ratoon crop

Values are means of 23–42 replicates. Field trial was established in October 1998 from bits. No significant differences between Williams and DPM25 were found for any measured parameter at $P = 0.05$

Cultivar or mutant line	Pseudostem height (m)	Pseudostem circumference (cm)	No. of leaves at harvest	Leaf area (cm^2)	Days to harvest	Bunch weight (kg)	Fruit weight (kg)	No. of hands	No. of fingers	Finger length[A] (cm)	Yield (t/ha)
					Plant crop						
DPM25	2.34	76.0	5.8	59149	727	21.8	20.1	9.7	157	19.6	36.7
Williams	2.36	77.3	5.6	58818	746	23.1	21.2	10.0	163	19.4	38.9
s.d.	0.19	4.3	0.7	2078	106	3.7	3.4	1.2	25	1.0	6.3
					Ratoon crop						
DPM25	2.66	81.1	6.0	68465	1115	27.3	25.5	10.1	172	21.0	45.9
Williams	2.63	81.2	6.1	70877	1099	27.6	25.6	9.9	167	21.1	46.4
s.d.	0.28	6.0	0.7	3237	85	5.5	5.2	1.3	31	1.5	9.2

[A]Finger length was measured from the middle finger of the outer whorl of the third hand from the proximal end.

Table 6. Incidence and severity of fusarium wilt infection of Williams and DPM25, a Dwarf Parfitt mutant (obtained by gamma irradiation)

Micropropagated plants were assessed for external and internal symptoms of fusarium wilt 7–11 months from planting and plants established using bits (sections of rhizome) were assessed at harvest
Severity values given are average ranks and analysed using the Kruskal–Wallis test. Means for each assessment date within each column followed by the same letter are not significantly different at $P = 0.05$

Planting material	Cultivar or mutant line	No. of plants	*Foc* external infection (%)	*Foc* internal infection (%)	External rank[A]	Internal rank[B]
			June 1993			
Micropropagated	Williams	12	67a	100a	14.8a	16.2a
	DPM25	13	38a	69b	11.4a	10.1b
			Aug. 1993			
Bits	Williams	22	32a	—	—	—
	DPM25	10	20a	—	—	—
	Dwarf Parfitt	5	20a	—	—	—
			Oct. 1993			
Micropropagated	Williams	12	92a	—	—	—
	DPM25	16	75a	—	—	—
			Oct. 1997			
Micropropagated	Williams	8	—	50a	—	31.3a
	DPM25	20	—	30ab	—	27.0a
	Dwarf Parfitt	21	—	5b	—	20.7b
			Apr. 2003			
Bits	Williams	31	3a	10a	43.0a	40.4a
	DPM25	44	2a	7a	42.5a	38.8a
	Bluggoe	15	40b	87b	59.5b	75.8b

[A]External severity ratings on a scale of 1–3: 1, no symptoms; 3, severe symptoms.
[B]Internal severity ratings on a scale of 1–6: 1, no symptoms; 6, total vascular discolouration of rhizome (Jones 1994).

(Table 5), although more work is needed to compare photoassimilation rates and relative cold tolerance between these plants.

This study has focussed on the role of mutation breeding for developing fusarium wilt resistant Cavendish cultivars; however, whether the putative mutants created were the result of somaclonal variation or physical mutagens is not known. Certainly gamma irradiation of meristems has been successfully used to develop a Cavendish line that flowers significantly earlier (Novak *et al*. 1990). It has been subsequently released as 'Novaria', and was entered into commercial production in Malaysia in 1993 (Mak *et al*. 1996). Somaclonal variation has also been exploited in micropropagated bananas and plantains with a range of desirable characteristics being uncovered (Smith *et al*. 2005). For instance, Daniells *et al*. (1999) noted a variant of 'Mons Mari' (AAA, Cavendish) with fruit 2–3 cm longer than usual for all hands and with the potential to boost profitability through increased sales of extra-large fruit. This clone has subsequently been released to industry as 'J.D. Special'. Until genetic transformation of Cavendish is routine and effective genes for fusarium resistance have been found and validated in field trials, future improvement in the Cavendish subgroup of dessert bananas will rely mainly on mutation breeding approaches.

Acknowledgments

We thank L. Pedwell for the use of his land as a race 4 *Foc* experimental site. The technical assistance of A. Hardwick and J. Hertrick is gratefully appreciated. Thanks also to J. Daniells for his constructive comments during his review of the manuscript. We acknowledge the financial support of the Australian Centre for International Agricultural Research, the Queensland Banana Industry Protection Board and Horticulture Australia Limited.

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Received 16 August 2004, accepted 23 December 2004