

# IMPROVING GERMINATION OF SINGLE-BUDDED SUGARCANE SETTS USING THERMOTHERAPY AND FUNGICIDE TREATMENTS

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## Abstract

Experiments to investigate the effect of various thermotherapy and fungicide treatments on the germination of single-budded setts (SBS) of sugarcane are described. Treatment in water at 52°C for 10 minutes significantly increased germination and plant growth. Immersing setts in cold water containing Eria® (62,5 ppm carbendazim + 31,25 ppm difenoconazole; 0,5 ml in 1 L) for 10 minutes also improved germination. Treatment with Eria in hot water (52°C) for 10 minutes was more effective than the untreated control, treatment with Eria in cold water and treatment in hot water alone. The active ingredient difenoconazole has both fungicidal and plant growth regulatory properties; the latter was probably largely responsible for the improved germination. Therefore, an inappropriate hormonal balance in SBS may be an important cause of poor germination in transplant nurseries, indicating that the use of freshly harvested, good quality seedcane is necessary to overcome germination problems.

*Keywords:* thermotherapy, fungicides, germination

## Introduction

In the South African sugar industry approximately ten million transplants are produced and sold annually by nurseries at three sugar mills and several privately owned nurseries. Transplants are mainly used for bulking up healthy material to establish farm nurseries and their use also forms part of the strategy to control ratoon stunting disease. The use of transplants can be constrained by several factors, the main one being poor germination.

In other countries, short heat treatment of setts at 52°C for 10 to 20 minutes before planting has significantly improved germination and growth (Benda, 1972; Anon., 1975; Comstock *et al.*, 1981; Peng, 1984; Farid, 1990). The increase in germination has been ascribed to the establishment of an appropriate hormonal balance for germination within the bud region (Benda, 1972; Peng, 1984; Farid, 1990).

In South Africa, benomyl (Benlate®, Du Pont de Nemours, Fundazol®, Sanachem) and guazatine (Panocrine®, Rhône-Poulenc) are registered as sett treatments against pineapple disease and sett rot (Mitchell-Innes and Thomson, 1973, 1974; Bechet, 1977). However, there are problems associated

with these fungicides. Panocrine is a Group II pesticide (poisonous) and Benlate and Fundazol are wettable powders that are insoluble in water, thus tending to settle out of solution, resulting in poor coating of SBS.

The aim of the experiments reported here was to investigate the value of using fungicides and short hot water treatments in the transplant nursery, and to determine which fungicides were the most efficacious in contributing to increased germination and growth of transplants.

## Methods

Single-budded setts were treated with various heat treatments and fungicides before being planted in 98-celled polystyrene trays containing a commercial potting medium (Grovida). Plots in each experiment consisted of a 20-celled section of the tray. The designs were randomised complete blocks with treatments replicated three times for each variety. To determine the efficacy of the treatments, germination and shoot dry mass data were recorded after 28 days. Data were evaluated using two-way analysis of variance (ANOVA) using Statgraphics (Version 5). All pairwise multiple comparisons were accomplished using least significant differences.

### Experiment 1

Single-budded setts of varieties N16, NCo376 and N12 were treated in hot water at 36, 40, 44, 48, 50, 52, 56 and 60°C for 10 minutes.

### Experiment 2

Single-budded setts of varieties N17 and NCo376 were treated in hot water at 50°C for 120 minutes and then soaked for five minutes in solutions of fungicides (Table 1).

### Experiment 3

The fungicides listed in Table 2 were mixed in either cold or hot water. Single-budded setts of varieties N16, N17 and N22 were treated in the fungicides in cold water for five minutes and SBS of varieties NCo376, N12 and N14 were treated in the fungicides in hot water for 10 minutes. The temperature of the hot water was initially 52°C and reduced to 48-50°C when SBS were submerged.

**Table 1. Rates of fungicides used for the treatment of the varieties N17 and NCo376.**

Commercial name (active ingredient)	Rate (ml/L)	Concentration of active ingredient (ppm)
Eria® (carbendazim+difenoconazole)	0,2	25 + 12,5
	0,5	62,5 + 31,25
	1,0	125 + 62,5
	1,5	187,5 + 94
Punch-Xtra® (carbendazim+flusilazole)	0,5	125 + 62,5
	1,0	250 + 125
	1,5	375 + 187,5
	2,0	500 + 250
Tilt® (propiconazole)	0,02	5
	0,2	50
	1,0	250
	2,0	500

**Table 2. Concentrations of fungicides tested for treatment of single-budded setts.**

Commercial name	Rate	Concentration of active ingredient
Panoptine	2,0 ml/L*	800 ppm guazatine
Benlate	0,5 g./L*	250 ppm benomyl
Previcur	1,0 ml/L	722 ppm propamocarb-HCl
Tilt	0,2 ml/L	50 ppm propiconazole
Punch-Xtra	0,5 ml/L	125 ppm carbendazim + 62,5 ppm flusilazole
Eria	0,5 ml/L	62,5 ppm carbendazim + 31,25 ppm difenoconazole

\*registered at the given concentration for control of sett rots of sugarcane in South Africa (Krause *et al.*, 1996).

*Experiment 4*

Single-budded setts of varieties N11, N12, NCo376, N16 and N17 were treated as follows:

- control (no treatment)
- short hot water treatment (SHWT; 52°C, 10 minutes)
- Eria in hot water (0,5 ml/L, 52°C, 10 minutes)
- Eria in hot water + Previcur/Benlate drench.

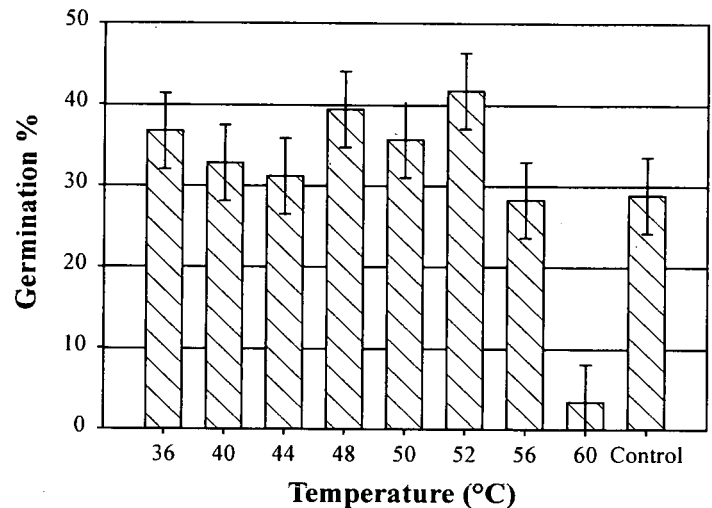
Previcur (1,5 ml/L) and Benlate (0,5 g/L) were mixed together into a paste that was mixed with cold water. After treatment, the SBS were planted into composted bagasse in trays that were drenched with either the fungicide solution or water at a rate of 20 ml per cell.

**Results**

*Experiment 1*

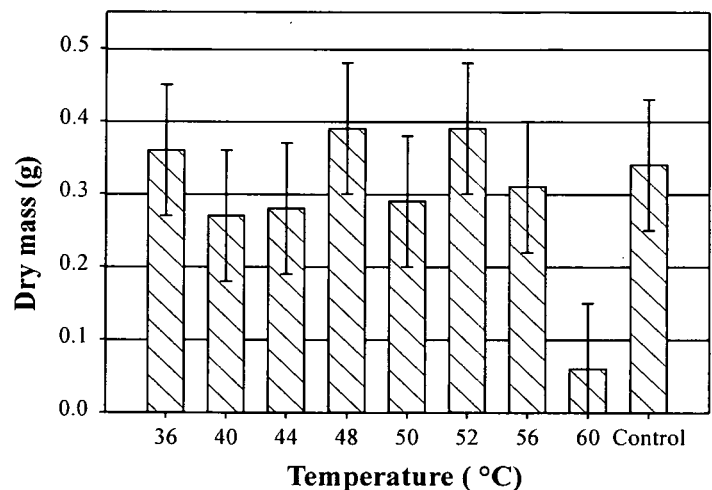
Germination of the untreated SBS was low (28%), indicating that the cane was too old for use as seedcane (Figure 1). Statistical analysis of the data indicated that SHWT had a significant effect on germination (VR=11,686; P<0,01) and

dry mass (VR=2,73; P=0,0134). Compared with the control, all treatments at temperatures less than 56°C improved germination, and treatments at 48 and 52°C significantly improved germination by 37 and 45% respectively (P<0,01). Treatment at 60°C had an adverse effect on germination.



**Figure 1. Effect of short heat treatment of single-budded setts for 10 minutes on the mean germination of varieties N12, N16 and NCo376 (means where 95% comparison intervals do not overlap are significantly different).**

Treatment of SBS at 36, 48 and 52°C for 10 min improved dry mass by 6-15% and treatment at 60°C significantly decreased dry mass by 74% compared with the control (Figure 2). Lack of a significant interaction between SHWT and variety for both germination (VR=1,317; P=0,2211) and dry mass (VR=0,558; P=0,9011) indicated that results were consistent for all varieties. Therefore, the mean germination of the three varieties was significantly improved by heat treating SBS at 48 and 52°C for 10 minutes, and both these treatments improved plant growth.



**Figure 2. Effect of short heat treatment of single-budded setts for 10 minutes on the mean dry mass of varieties N12, N16 and NCo376 (means where 95% comparison intervals do not overlap are significantly different).**

Experiment 2

The mean germination of the untreated SBS of both varieties was high (78%). Analysis of the data indicated that fungicide treatment did not have a significant effect on germination (VR=1,937; P=0,0543). Compared with the control, germination was increased by Eria (0,5 ml/L) (+7%), Punch-Xtra (0,5 ml/L) (+6%) and Tilt (0,2 ml/L) (+4%) (Figure 3).

Fungicide Treatment (ml/l)

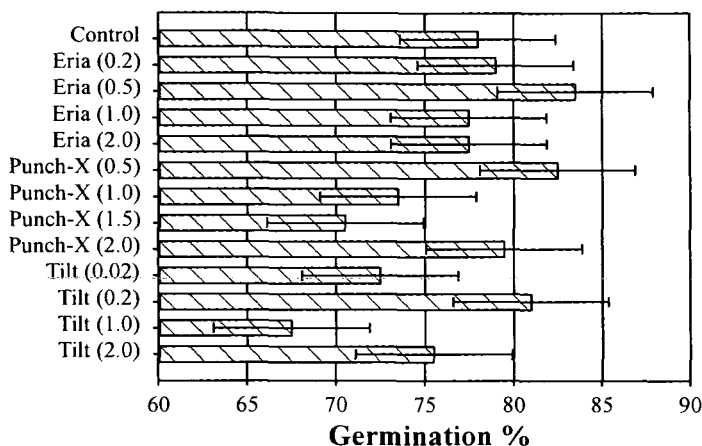


Figure 3. Effect of fungicide treatment of single-budded setts on the mean germination of varieties N17 and NCo376 (means where 95% comparison intervals do not overlap are significantly different).

The germination results were not consistent for both varieties, as indicated by the significant interaction between fungicide treatment and variety (VR=2,628; P=0,0092). Compared with the untreated control, treatment with Eria, Tilt and Punch-Xtra at most concentrations improved germination of N17, the treatments with Tilt (0,2 ml/L) and Punch-Xtra (0,5 and 1 ml/L) significantly so (+15-18%, P<0,05) (Table 3). Compared with the untreated control, treatment with Eria at three concentrations slightly increased germination and treatment with both Tilt and Punch-Xtra decreased germination of NCo376. These results indicated that the varieties responded differently to treatment with both Punch-Xtra and Tilt, but not to Eria.

Analysis of the data indicated that fungicide treatment had a significant effect on dry mass (VR=2,628; P<0,01). The mean dry mass was significantly increased by treatment with Eria (0,2 ml/L) (+100%), Eria (0,5 ml/L) (+69%) and Eria (1 ml/L) (+55%) (Figure 4). The dry mass results were consistent for both varieties, as indicated by the lack of a significant interaction between fungicide treatment and variety (VR=0,596; P=0,8351). Overall, Eria at 0,2 and 0,5 ml/L was the most effective fungicide.

Experiment 3

Cold-fungicide treatments

Analysis of the data indicated that fungicide treatment had no significant effect on germination (VR=0,917; P=0,4891) or dry mass (VR=1,702; P=0,1353). Compared with the control, germination was increased after treatment with Eria (+6%)

and Tilt (+5%); treatment with Panoctine decreased germination by 5% and dry mass by 21% (Figures 5 and 6). The germination results were consistent for all varieties, as indicated by the lack of a significant interaction between fungicide treatment and variety (VR=0,676; P=0,7675). However, the dry mass results were not consistent for all varieties, as indicated by the significant interaction between fungicide treatment and variety (VR=1,928; P=0,0475). Treatment of N16 with Panoctine and Tilt significantly decreased dry mass (P<0,05), but these treatments had little effect on the dry mass of N17 and N22.

Fungicide Treatment (ml/l)

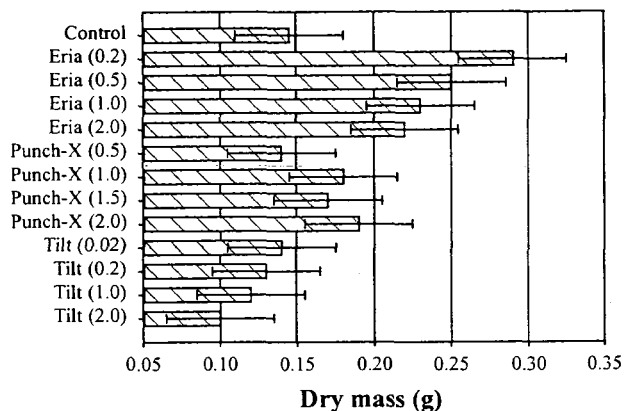


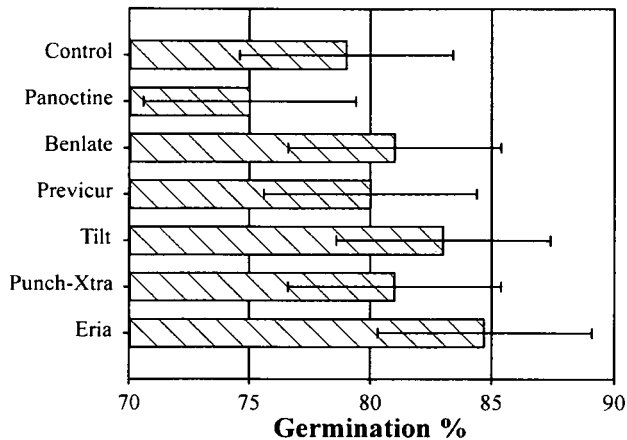
Figure 4. Effect of fungicide treatment of single-budded setts on the mean dry mass of varieties N17 and NCo376 (means where 95% comparison intervals do not overlap are significantly different).

Table 3. Effect of fungicides on the germination and dry mass of varieties N17 and NCo376.

Treatment	Germination (%)		Dry mass (g)	
	N17	NCo376	N17	NCo376
0,2 ml/L Eria	78 bc	80 abc	0,34 a	0,24
0,5 ml/L Eria	85 abc	82 a	0,24 abc	0,25
1,0 ml/L Eria	75 c	80 abc	0,25 ab	0,20
2,0 ml/L Eria	82 abc	73 abcd	0,26 ab	0,18
0,5 ml/L Punch-Xtra	82 abc	63 bcd	0,14 bcd	0,13
1,0 ml/L Punch-Xtra	90 a	72 abcd	0,12 cd	0,13
1,5 ml/L Punch-Xtra	78 bc	57 d	0,12 cd	0,12
2,0 ml/L Punch-Xtra	85 abc	66 abcd	0,09 d	0,11
0,02 ml/L Tilt	92 a	73 abcd	0,15 bcd	0,14
0,2 ml/L Tilt	92 a	55 d	0,19 bcd	0,17
1,0 ml/L Tilt	78 bc	63 cd	0,20 bcd	0,14
2,0 ml/L Tilt	87 ab	72 abcd	0,19 bcd	0,19
Control	78 bc	78 abc	0,17 bcd	0,12
LSD (P=0,05)	10,88	16,28	0,12	NS
LSD (P=0,01)	14,72	22,13	0,16	

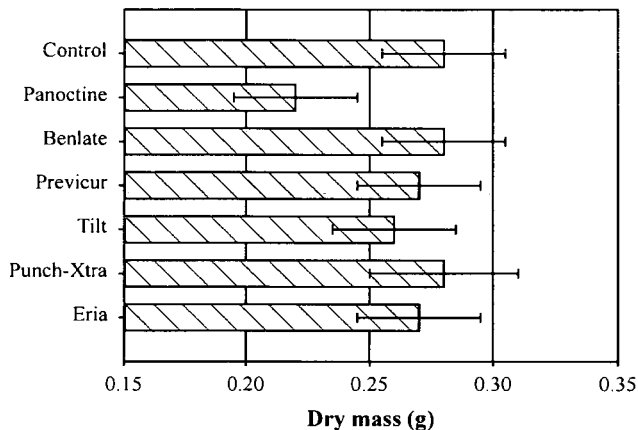
Means in a column with a letter in common are not significantly different at the 5% level.

**Fungicide Treatment**



**Figure 5.** Effect of cold-fungicide treatment of SBS on the mean germination of varieties N16, N17 and N22 (means where 95% comparison intervals do not overlap are significantly different).

**Fungicide Treatment**



**Figure 6.** Effect of cold-fungicide treatment of SBS on the mean dry mass of varieties N16, N17 and N22 (means where 95% comparison intervals do not overlap are significantly different).

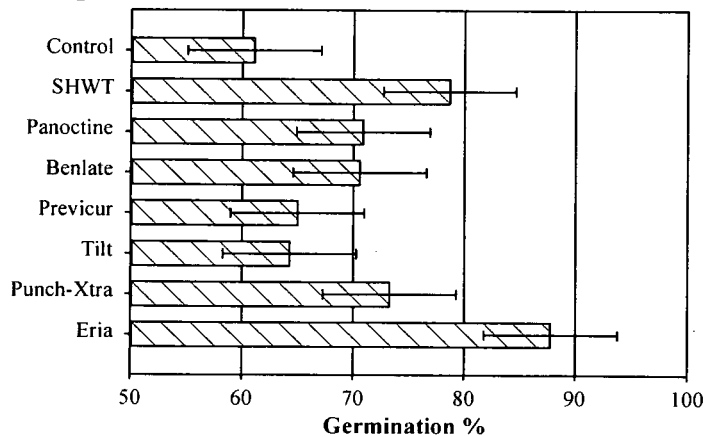
*Hot-fungicide treatments*

Analysis of the data indicated that fungicide treatment had a significant effect on mean germination (VR=7,469; P<0,01) and dry mass (VR=4,656; P<0,01) of varieties NCo376, N12 and N14, indicating that the fungicides were more effective when applied in hot water than in cold water. Compared with the untreated control, the mean germination was significantly increased at the 1% level by treatment with Eria (+44%), SHWT (+29%) and Punch-Xtra (+20%), and significantly increased at the 5% level by treatment with Benlate (+16%) and Panoctine (+16%) (Figure 7). Dry mass was significantly increased by treatment with Eria (+92%) (P<0,01) and SHWT (+46%) (P<0,05), and increased by Punch-Xtra (+38%) (Figure 8).

Compared with the SHWT, treatment with Eria significantly increased germination by 12% (P=0,01) and dry mass by 32% (P=0,05). Treatment with Panoctine, Benlate and Punch-Xtra decreased germination and dry mass, but not significantly so.

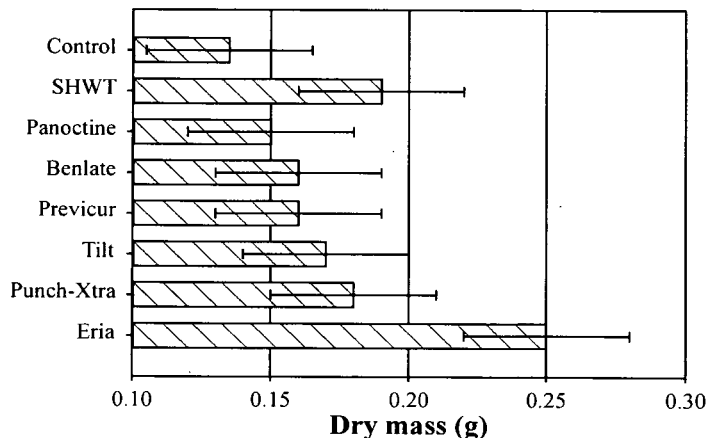
Treatment with Previcur and Tilt significantly decreased germination and decreased dry mass compared with the SHWT (P=0,01) (Figures 7 and 8). Therefore, these fungicides used in hot water were not as effective as heat treatment alone.

**Fungicide Treatment**



**Figure 7.** Effect of hotfungicide treatment of single-budded setts on the mean germination of varieties NCo376, N12 and N14 (means where 99% comparison intervals do not overlap are significantly different).

**Fungicide Treatment**



**Figure 8.** Effect of hotfungicide treatment of single-budded setts on the mean dry mass of varieties NCo376, N12 and N14 (means where 99% comparison intervals do not overlap are significantly different).

Lack of a significant interaction between fungicide treatment and variety for both germination (VR=1,401; P=0,1896) and dry mass (VR=0,645; P=0,8140) indicated that results were consistent for all varieties.

These results indicated that the highest germination and plant growth occurred after treatment with Eria when fungicides were applied in both cold and hot water, and after the SHWT at 52°C for 10 minutes. Addition of the other fungicides to hot water was not as effective as the SHWT alone.

*Experiment 4*

The mean germination of the untreated SBS of the five varieties was poor (58%), indicating that fungal pathogens

possibly inhibited germination. Analysis of the data indicated that treatments had a significant effect on germination (VR=22,313; P<0,01) and dry mass (VR=52,370; P<0,01). Compared with the control, the heat treatment significantly increased germination by 28% (P=0,01) (Figure 9) and dry mass by 43% (P=0,05) (Figure 10).

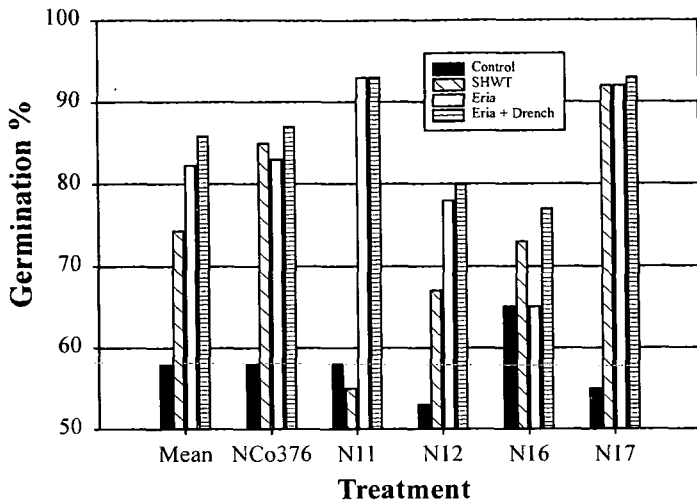


Figure 9. Effect of single-budded sett treatment on the germination of varieties NCo376, N11, N12, N16 and N17.

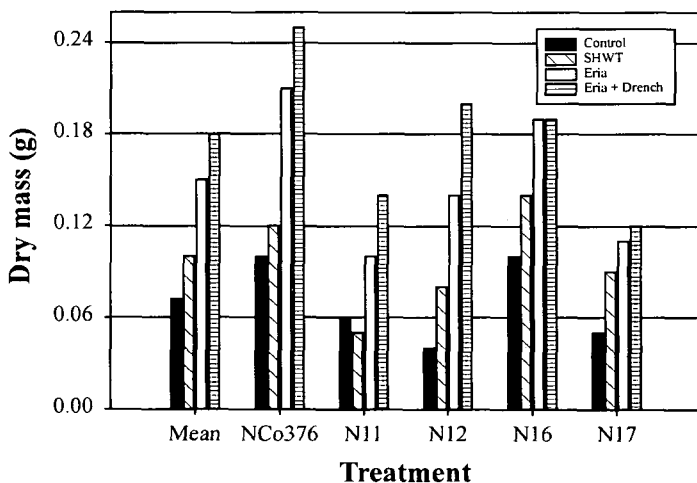


Figure 10. Effect of single-budded sett treatment on the dry mass of varieties NCo376, N11, N12, N16 and N17.

Addition of Eria to the hot water significantly increased both germination and growth compared with the heat treatment alone (P=0,05), and significantly increased germination by 41% (P=0,01) and dry mass by 114% (P=0,01) compared with the control.

Drenching trays containing Eria-treated SBS with Previcur/Benlate further improved germination and significantly improved dry mass compared with the Eria treatment, and significantly improved germination and plant growth compared with the SHWT (P=0,01). Compared with the untreated control, this treatment significantly increased germination by 48% (P=0,01) and dry mass by 157% (P=0,01).

A significant interaction between variety and treatment for both germination (VR=2,783; P=0,0075) and dry mass (VR=2,092; P=0,0410) indicated that results were not consistent for all varieties. Germination of SBS treated with the SHWT ranged from 55 to 92%. The SHWT significantly increased germination of NCo376 by 47% and N17 by 67% (P<0,01), increased germination of N12 by 26% and N16 by 12% and had no effect on germination of N11 (Figure 9).

Germination of SBS treated with Eria ranged from 65 to 93% and germination after treatment with Eria and the fungicide drench ranged from 77 to 93%. These treatments significantly increased germination of N11 by 43 to 50% (P<0,05), NCo376 by 60% (P<0,01), N12 by 47 to 51% (P<0,01) and N17 by 92 to 93% (P<0,01) compared with the untreated control, and significantly increased germination of N11, and increased germination of N12 compared with the SHWT.

Compared with the control, the SHWT increased dry mass of N12, N16 and N17, and the hot-Eria treatment significantly increased dry mass of NCo376 by 110% (P<0,01), N12 by 250% (P<0,05), N16 by 90% (P<0,01) and N17 by 120% (P<0,01) (Figure 10). Drenching Eria-treated SBS significantly increased dry mass of all the varieties by 90 to 400% (P<0,01). Compared with the SHWT, the hot-Eria treatment significantly increased dry mass of NCo376 (P<0,01) and N11 (P<0,05) and increased dry mass of N12, N16 and N17.

### Discussion

The results show that short heat treatments alone can be used to increase both germination and plant growth. They adjust the hormone balance of the buds to an appropriate level for germination by the loss of indole-3-acetic acid (IAA) and indole-3-butyric acid (IBA) into the water (Benda, 1972; Peng, 1984; Farid, 1990). They also probably control systemic fungi, increasing the rate of germination and allowing shoots to develop faster than those grown from untreated SBS, resulting in increased plant growth. These treatments do not control *Clavibacter xyli* subsp. *xyli*.

Treatment at 52°C for 10 minutes significantly improved the germination of SBS and is highly recommended with or without the addition of a fungicide. Where heat treatment tanks are not available, the use of hot tap water in large plastic containers is possible. The use of the SHWT has been used successfully at SASEX for both commercial and unreleased varieties. However, some unreleased varieties are sensitive to the treatment, indicating that all new varieties must be tested before large-scale treatments are carried out.

The use of Eria in hot water is recommended for treatment of SBS that are to be planted into polystyrene trays for transplant production. Eria is a persistent systemic fungicide, available as a suspension concentrate. It is not as expensive or toxic as Panoptine and constant agitation is not required to keep the active ingredients in solution.

The improved germination and growth responses after treatment with Eria could have been due to the activity of both carbendazim and difenoconazole against Ascomycetes, Basidiomycetes and Deuteromycetes, as well as the plant growth regulator properties of difenoconazole. Difenocona-

zole is a triazole that inhibits sterol biosynthesis, resulting in blocking of the gibberellin biosynthesis pathway of plants, promoting growth of lateral buds (Werbrouck and Debergh, 1996; Werbrouck *et al.*, 1996). Combining Eria treatment of SBS with a Previcur/Benlate drench allows the developing roots to absorb both Previcur and Benlate, resulting in control of diseases for a long period after planting.

The use of Eria in hot water should result in increased germination of transplants, thereby decreasing production costs. This would make the transplant option more viable for farmers intending to establish their own seedcane nurseries. Some transplant growers have already used Eria and reported increases in germination and plant vigour compared with other fungicides, particularly when used as a cold treatment after setts have been heat treated at 50°C for 120 minutes.

### Conclusions

Treatment of single-budded setts in water at 52°C for 10 minutes and treatment with Eria (0,5 ml/L) improved germination and growth. When the seedcane from which the SBS are taken is heat treated before planting and found to be free of ratoon stunting disease, these treatments should result in lower production costs and more vigorous transplants that can be used to establish seedcane nurseries. Using seedcane from these nurseries would result in healthier and higher yielding cane in commercial fields and would form part of the strategy to control ratoon stunting disease.

### REFERENCES

- Anon (1975). Factors affecting seed germination. *Ann Rep Haw Sug Plant Ass* pp 54-56.
- Bechet, GR (1977). Further evaluation of fungicides for control of pineapple disease of sugarcane. *Proc S Afr Sug Technol Ass* 51: 51-54.
- Benda, GTA (1972). Hot water treatment for mosaic and RSD control. *Sug J* 34: 32-39.
- Comstock, JC, Ferreira, SA and Osgood, RV (1981). Effect of heat treatment on germination. *Ann Rep Haw Sug Plant Ass* pp 33-34.
- Farid, G (1990). Studies on the effect of hot water treatment and duration of treatment on the sprouting of sugarcane setts. *Pak Sug J* 4(2): 5-7.
- Krause, M, Nel, A and van Zyl, K (1996). *A Guide to the Use of Pesticides and Fungicides in the Republic of South Africa*. Published by the National Department of Agriculture.
- Mitchell-Innes, L and Thomson, GM (1973). A new fungicide for the pre-planting treatment of sugarcane setts. *Proc S Afr Sug Technol Ass* 47: 181-184.
- Mitchell-Innes, L and Thomson, GM (1974). Tests with some additional non-mercurial fungicides for the control of pineapple disease. *Proc S Afr Sug Technol Ass* 48: 85-87.
- Peng, SY (1984). The growing of sugarcane. pp. 13-14; 17-24 In: *The Biology and Control of Weeds in Sugarcane. Developments in Crop Science 4*. Elsevier, Amsterdam. 366 pp.
- Werbrouck, SPO and Debergh, PC (1996). Imidazole fungicides and paclobutrazol enhance cytokinin-induced adventitious shoot proliferation in Araceae. *J Plant Growth Regul* 15: 81-85.
- Werbrouck SPO, Redig, P, Vanonckelen, HA and Debergh, PC (1996). Gibberellins play a role in the interaction between imidazole fungicides and cytokinins in Araceae. *J Plant Growth Regul* 15: 87-93.