Use of Gamma Irradiation for Control of Postharvest *Botrytis cinerea* Bunch Rot of Table Grapes in Cold Storage*

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The feasibility of employing gamma irradiation for the control of postharvest *Botrytis* bunch rot of table grapes in cold storage was studied. Table grape cultivars from commercial vineyards in the Paarl and Hex River Valley areas were packed as for export in vented corrugated cartons. The cartons were irradiated at 0; 1,5; 2,0 or 3,0 kGy. After irradiation, grapes were kept for 4 weeks at -0,5°C, followed by another week at 10°C. Less decay was observed when table grapes were irradiated soon after packing than after a storage period. Irradiation at 1,5; 2,0 and 3,0 kGy reduced the effect of *Botrytis cinerea* on Barlinka grapes stored without sulphur dioxide. Irradiation of Waltham Cross and Barlinka grapes at a dose of 2,0 kGy, combined with reduced SO_2 treatments, resulted in similar control as with the standard practice of enclosing an SO_2 generator. Browning of Waltham Cross berries and bacterial and yeast growth occurred on the surface of berries irradiated at a dose of 3,0 kGy. Irradiation had no adverse effect on other aspects of quality.

Botrytis cinerea, the main decay pathogen of table grapes in storage (Hewitt, 1974; Nelson, 1985), can be responsible for annual losses of more than R5 million in South Africa (Marais, 1985; Lourens, 1986). The decay is due largely to spores present on bunches at harvest, or to the formation of late season latent infections (De Kock, 1989; De Kock & Holz, 1991). Fungicide application to grapevines at defined stages of bunch development is advocated for the control of *B. cinerea* (Eckert & Ogawa, 1988), but cannot prevent postharvest decay (De Kock, 1989; De Kock & Holz, 1991). Although postharvest fumigation with sulphur dioxide (SO_2) eradicates spores and prevents contact spread (nesting) effectively (Gentry & Nelson, 1968; Nelson & Nelson & Ahmedullah, 1972; Nelson, 1983; Peiser & Yang, 1985; Kokkalos, 1986), the fungus still causes spoilage (Marios *et al.*, 1986; Eckert & Ogawa, 1988; De Kock, 1989; De Kock & Holz, 1991). Alternative control methods must therefore be found to minimize losses due to *Botrytis* decay during storage and export.

The ability of gamma radiation to penetrate fruits and to inactivate pathogens in established lesions deep in the host tissues (Beraha *et al.*, 1961; Eckert & Ogawa, 1988) offers a potential for therapeutic treatment of established infections. A study was therefore made to determine the feasibility of employing gamma irradiation for the control of postharvest *Botrytis* bunch rot of table grapes in cold storage.

MATERIALS AND METHODS

Three trials were conducted during the 1986, 1987 and 1989 seasons on table grapes of different cultivars from commercial vineyards in the Paarl and Hex River Valley areas. Fungicide and pesticide applications in the vineyards, prior to harvest, were made according to the recommendations of De Klerk (1985). Grapes were harvested and bunches, free from visible symptoms of *Botrytis* bunch rot, were packed as for export in vented corrugated cartons (Patent no. RSA 75/6116) with polyethylene bags as lining. Each carton contained 5 kg fruit and an SO_2 generator (0,3 - 0,55 g sodium metabisulfite affixed to a 250 x 320 mm paper sheet [Laszlo *et al.*, 1981; Nelson, 1983]) was enclosed where appropriate.

Trial 1: During the 1986 season grapes of different cultivars were harvested in four experiments. Experiment (Expt.) 1 of cultivars Waltham Cross, Dan Ben Hannah and Bien Donné, harvested in January 1986 in the Paarl area. Experiment 2 consisted of cultivars Dan Ben Hannah and Bien Donné, harvested in January 1986 in the Hex River Valley area. Experiment 3 consisted of cultivars Alphonse Lavallée and Waltham Cross, harvested in February 1986 in the Hex River Valley area. Experiment 4 consisted of cultivars Waltham Cross and Barlinka, harvested in April 1986 in the Hex River Valley area. A quarter of the standard SO_2 generator was enclosed in all the cartons to suppress the development of *Botrytis* bunch rot during storage and transit. The cartons were kept in cold storage (-0,5°C) for 5-19 d before being sent by road (cold transit for 3 d) to Iso-Ster (Pty) Ltd (PO Box 3219, 1620 Kempton Park) in Johannesburg for gamma irradiation. Cartons of each cultivar were divided into four equal groups and irradiated at

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TABLE 1

The percentage *Botrytis cinerea* postharvest decay of table grapes[a] after gamma irradiation and cold storage[b] conducted during the 1986 season.

Treatment	Postharvest decay (%)[c]								
	Expt 1			Expt 2		Expt 3		Expt 4	
	WX	DbH	BD	DbH	BD	AL	WX	WX	Bar
1,5 kGy	9,6	73,3	41,6	36,8	25,9	70,4	15,3	14,0	4,9
2,0 kGy	11,9	64,8	28,8	24,4	17,4	38,9	10,9	10,0	2,9
3,0 kGy	13,0	64,6	40,1	22,8	11,7	36,0	7,3	3,7	3,1
Control	18,4	67,7	48,4	31,0	26,3	71,9	6,2	23,1	21,7
D-value ($p \leq 0,05$)	NS	NS	NS	NS	NS	29,52	NS	15,45	13,91

[a]Table grape cultivars: WX = Waltham Cross; DbH = Dan Ben Hannah; BD = Bien Donné; AL = Alphonse Lavallée; Bar = Barlinka.
[b]Grapes were packed in cartons with a quarter of an SO_2 generator and stored for 35 d after irradiation.
[c]Percentage decay was calculated according to the formula of Kremer & Unterstenhöfer (1967).

doses of 0; 1,5; 2,0 or 3,0 kGy respectively. Each treatment was replicated five times with one carton (8-10 bunches) per replication. After irradiation, grapes were kept for 4 wks at -0,5°C, followed by another week at 10°C and then assessed for *Botrytis* bunch rot.

Trial 2: Waltham Cross grapes were harvested in February 1987 in the Paarl area. Cartons were divided into three equal groups of 24 cartons each. The first group was packed without SO_2 generators, but with grapes inoculated after packing by spraying with a spore suspension of *B. cinerea* as described by De Kock & Holz (1991); the second group was packed without SO_2 generators and the third group with standard SO_2 generators enclosed. Cartons were kept in cold storage for 24 h before being sent by road (cold transit) for gamma irradiation to Hepro (Pty) Ltd (High Energy Processing, P O Box 141, 7435 Milnerton), Cape Town. The same treatments were applied to grapes harvested in the Hex River Valley in March 1987. Cartons were kept in cold storage for 7 d before being sent by road to Iso-Ster, Johannesburg. At the irradiation facilities, each group of cartons was then divided into four separate groups and the cartons irradiated at 0; 1,5; 2,0 or 3,0 kGy. Grapes were kept in cold storage after irradiation and assessed for *Botrytis* decay as described. The trial was repeated with Barlinka grapes, harvested in the Paarl area in March 1987. Cartons were kept in cold storage for 24 h, before being sent by road to Hepro or 6 d before being sent by road to Iso-Ster.

Trial 3: Waltham Cross grapes were harvested in February 1989 in the Paarl area and were packed without SO_2 generators, or with 1/8, 1/4, 1/2, 3/4 or a complete SO_2 generator enclosed. Each treatment was replicated six times with one carton per replication. Cartons were kept in cold storage for 24 h and sent by road (cold transit) to Hepro for gamma irradiation. All treatments, except the control, were irradiated at 2,0 kGy. After irradiation, grapes were kept for 4 wks at -0,5°C, followed by another 2 wks at 10°C before being assessed for *Botrytis* decay. The trial was repeated with Barlinka grapes harvested in March 1989 in the Paarl area.

Disease and quality assessment: Postharvest *Botrytis* decay was assessed according to the rating proposed by Unterstenhöfer (1963) for the infection of berries by *Plasmopara viticola* (Berk. & Curt.) Berl, & de Toni. The decay of each replicate was calculated using the formula of Kremer & Unterstenhöfer(1967). In some instances the decay of each bunch was determined on a mass basis and the average decay per treatment calculated. The quality (defined as percentage loose berries, berry texture and dryness or browning of stems) of the irradiated treatments was compared to that of the untreated control.

Statistical analysis: All data were subjected to a standard analysis of variance, and significance of differences between treatments was determined by means of a D-value based on the Studentized Q-test (Snedecor & Cochran, 1967).

RESULTS

Trial 1: The postharvest decay observed in the four experiments is given in Table 1. Gamma irradiation caused no consistent reduction in decay of the different cultivars. Significant control of decay at all the doses was obtained only with Barlinka in Expt. 4 where 7d elapsed between packing and irradiation.

No meaningful difference in quality was observed between irradiated and non-irradiated grapes (data not shown). As with decay, gamma irradiation had a differential effect on some of the cultivars. In the first experiment, irradiation at 2,0 and 3,0 kGy caused a change of colour in Bien Donné berries. The yellow-green berries turned brown with dark longitudinal stripes. In the second experiment, no change in berry colour was observed. Irradiation at 3,0 kGy resulted in a high proportion of soft Waltham Cross berries, but only in the first experiment.

Trial 2: The postharvest *Botrytis* decay on inoculated and uninoculated grapes is given in Table 2. Gamma irradiation caused a significant reduction in the effect of inoculum on inoculated Barlinka bunches, but decay was still unacceptably high. On uninoculated Barlinka grapes, stored with or without SO_2 generators, irradiation significantly

reduced *Botrytis* bunch rot. The best control was achieved when Barlinka grapes were irradiated at 2,0 kGy at Hepro and stored with SO_2 generators.

No meaningful difference in quality was observed between irradiated and non-irradiated grapes (data not shown). Some berries treated at 2,0 kGy and 3,0 kGy developed cracks and were covered with bacteria and yeasts, whereas some Waltham Cross berries irradiated at Hepro turned caramel-brown.

Trial 3: The postharvest *Botrytis* decay on irradiated grapes exposed to different SO_2 treatments is given in Table 3. On Waltham Cross, the best control was achieved when irradiated grapes were stored with either a 3/4 or a complete SO_2 generator enclosed. However, reduction in decay was not significant when compared with the non-irradiated control. Irradiation of Barlinka grapes stored with a portion of an SO_2 generator resulted in similar decay as in the non-irradiated control.

Irradiation had little effect on the quality of the grapes. As in the previous trial, some berries developed cracks and some berries of Waltham Cross turned caramel-brown.

DISCUSSION

High incidences in the decay of control table grapes in the first trial indicate that the inclusion of a quarter of the standard SO_2 generator was not able to suppress. *B. cinerea* development during the 8-22 d storage and transit period preceding irradiation. The resulting *Botrytis* decay masked the effect of irradiation. In the subsequent trial, application of irradiation to table grapes soon after packing resulted in less decay than treatments applied after a storage period of 9d. Although gamma irradiation caused a reduction in the effect of *B. cinerea* inoculum on Barlinka bunches stored without SO_2, decay was still unacceptably high. Gamma irradiation combined with SO_2 was able to control decay of table grapes in cold storage more effectively than the standard practice of including SO_2 alone (Combrink *et al.*, 1978; Laszlo *et al.*, 1981). Irradiation at 2,0 kGy gave the best control on Barlinka grapes and had little effect on the quality of Waltham Cross. Shirzad & Langerak (1984) observed the same effect with a similar combination, but they stored grapes at a constant temperature of 10°C during the experiment. Our findings also confirmed those of Bramlage & Couey (1985) (according to Thomas, 1986) who showed that gamma irradiation alone was less effective in controlling decay than the standard SO_2 treatment. Gamma irradiation at 2,0 kGy, combined with reduced SO_2 treatments, of Waltham Cross and Barlinka grapes resulted in similar decay as with the standard practice.

Browning of Waltham Cross berries at a dose of 3,0 kGy was similar to that observed by Matthee & Marais (1963) at higher doses. Although better control was

TABLE 2

The percentage *Botrytis cinerea* postharvest decay of table grapes after gamma irradiation by different institutions[a] and cold storage[b] during the 1987 season.

Treatment and irradiation dose	Postharvest decay (%)[c]			
	Waltham Cross		Barlinka	
	Hepro	Iso-Ster	Hepro	Iso-Ster
Inoculated, stored without SO_2				
1,5 kGy	63,4	100,0	57,3	97,7
2,0 kGy	63,1	99,5	44,1	72,5
3,0 kGy	54,5	100,0	53,3	98,9
Control	70,9	99,6	90,9	100,0
D-value ($p \leq 0,05$)	NS	NS	12,43	5,61
Uninoculated, stored without SO_2				
1,5 kGy	32,0	37,6	45,7	53,8
2,0 kGy	13,5	29,7	32,8	47,1
3,0 kGy	33,8	27,1	43,1	51,0
Control	22,7	50,7	78,4	72,5
D-value ($p \leq 0,05$)	16,77	NS	15,14	12,60
Uninoculated, stored with SO_2				
1,5 kGy	0,2	10,4	7,4	37,4
2,0 kGy	0,2	7,6	5,2	39,0
3,0 kGy	0,3	5,5	8,5	37,9
Control	0,9	14,6	19,0	62,2
D-value ($p \leq 0,05$)	NS	NS	7,08	22,56

[a]Hepro in Cape Town, Iso-Ster in Johannesburg.
[b]Grapes were stored for 35 d after application of irradiation.
[c]Percentage postharvest decay of grapes treated by Hepro was determined on a mass basis. Percentage postharvest decay of grapes treated by Iso-Ster was calculated with the formula of Kremer & Unterstenhöfer (1967).

TABLE 3

The percentage *Botrytis cinerea* postharvest decay of table grapes after gamma irradiation at 2,0 kGy and treated with different levels of SO_2 during cold storage[a].

Treatment and SO_2[c]	Postharvest decay (%)[b]	
	Waltham Cross	Barlinka
Irradiated		
One eighth	35,3	7,7
One quarter	21,9	3,6
Half	6,8	3,4
Three quarter	3,0	4,4
Complete	5,2	3,9
None	60,3	30,1
Non-irradiated		
Complete	13,2	4,8
D-value ($p \leq 0,05$)	11,57	8,75

[a]Grapes were stored for 42 d after irradiation application.
[b]Percentage postharvest decay was determined on a mass basis.
[c]Portion of a standard SO_2 generator enclosed in polyethylene bags.

achieved at 3,0 kGy in some of the experiments, bacterial and yeast growth on the surface of berries irradiated at this dose indicated that some micro-organisms survived the high irradiation dose (Nelson, Maxie & Eukel, 1959). Irradiation at 3,0 kGy would therefore be impractical.

The change in colour of Bien Donné and Waltham Cross berries might also be due to the gibberellic acid applied while the berries were pea-size to ensure a subsequent uniform berry-size (Combrink *et al.,* 1974). No loss of other quality aspects such as berry texture was observed. The cultivar Waltham Cross is known to have loose berries, but berry fall was not promoted by irradiation in this investigation, as recorded elsewhere (Kim *et al.,* 1969, according to Thomas, 1986).

The irradiation of packed table grapes, combined with a sulphur dioxide treatment, offers the potential of controlling postharvest *B. cinerea* decay. Investigations into the postharvest handling of table grapes and the economic feasibility are needed before commercial implementation. Current regulations, however, do not permit the export of irradiated food from the RSA.

LITERATURE CITED

BERAHA, L., RAMSEY, G.B., SMITH, M.A., WRIGHT, W.R., & HEILIGMAN, F., 1961. Gamma radiation in the control of decay in strawberries, grapes and apples. *Food Technol.* **15,** 94-98.

COMBRINK, J.C., LASZLO, J.C., TRUTER, A.B. & BOSCH, P.J.C., 1978. Can the pre-cooling time for table grapes be shortened? *Decid. Fruit Grow.* **28,** 166-171.

COMBRINK, J.C., MALHERBE, W.S., GINSBURG, L. & TRUTER, A.B., 1974. Treatment of Waltham Cross grapes with growth regulators. *Decid. Fruit Grow.* **24,** 146-149.

DE KLERK, C.A., 1985. Chemical control programme for vine diseases and pests. Farming in South Africa. Viticultural and Oenological Series 200. VORI. Private Bag X5026, 7600 Stellenbosch, Republic of South Africa.

DE KOCK, P.J., 1989. *Botrytis cinerea* bunch rot of table grapes: colonization and timing of fungicide application. M.Sc. Agric. thesis. University of Stellenbosch, 7600 Stellenbosch, Republic of South Africa.

DE KOCK, P.J. & HOLZ, G., 1991. Colonization of table grapes by *Botrytis cinerea* in the western Cape Province. *Phytophylactica* **23,** 73-80.

ECKERT, J.W. & OGAWA, J.M. 1988. The chemical control of postharvest diseases: Deciduous fruits, berries, vegetables and root/tuber crops. *Ann. Rev. Phytopathol.* **26,** 433-469.

GENTRY, J.P. & NELSON, K.E., 1968. Further studies on control of decay of table grapes by two-stage generation of sulfur dioxide within unvented containers. *Am. J. Enol. Vitic.* **19,** 70-81.

HEWITT, W.B., 1974. Rots and bunch rots of grapes. California Agricultural Experimental Station Bulletin 868.

KOKKALOS, T.I., 1986. Postharvest decay control of grapes by using sodium metabisulfite in cartons enclosed in plastic bags. *Am. J. Enol. Vitic.* **37,** 149-151.

KREMER, W. & UNTERSTENHÖFER, G., 1967. Computation of results of crop protection experiments by the method of Townsend and Heuberger. *Pflanzenschutz-Nachr. Bayer* **20,** 625-628.

LASZLO, J.C., COMBRINK, J.C., EKSTEEN, G.J. & TRUTER, A.B., 1981. Effect of temperature on the emission of sulphur dioxide from gas generators for grapes. *Decid. Fruit Grow.* **31,** 112-119.

LOURENS, P., 1986. Parasitism by *Botrytis cinerea. Decid. Fruit Grow.* **36,** 133-137.

MARAIS, P.G., 1985. Infection of table grapes by *Botrytis cinerea. Decid. Fruit Grow.* **35,** 166-170.

MAROIS, J.J., BLEDSOE, A.M., GUBLER, W.D. & LUVISI, D.A., 1986. Control of *Botrytis cinerea* on grape berries during postharvest storage with reduced levels of sulfur dioxide. *Plant Dis.* **70,** 1050-1052.

MATTHEE, F.N. & MARAIS, P.G., 1963. Preservation of food by means of gamma rays. *Food Irradiat.* **4,** A10-A17.

NELSON, K.E., 1983. Effects of in-package sulfur dioxide generators, package liners, and temperature on decay and desiccation of table grapes. *Am. J. Enol. Vitic.* **34**, 10-16.

NELSON, K.E., 1985. Harvesting and handling California table grapes for market. University of California, Division of Agriculture and Natural Resources, Bulletin 1913.

NELSON, K.E. & AHMEDULLAH, M., 1972. Effect of in-package sulfur dioxide generator and packaging materials on quality of stored table grapes. *Am. J. Enol. Vitic.* **23,** 78-85.

NELSON, K.E., MAXIE, E.C. & EUKEL, W., 1959. Some studies in the use of ionizing radiation to control *Botrytis* rot in table grapes and strawberries. *Phytopathology* **49,** 475-480.

PEISER, G.D. & YANG, S.F., 1985. Metabolism of sulfur dioxide in 'Thompson Seedless' grape berries. *J. Am. Soc. Hort. Sci.* **110,** 224-226.

SHIRZAD, B.M. & LANGERAK, D.Is., 1984. Gamma radiation technological feasibility of increasing shelf-life of table grapes. *Acta Aliment.* **13,** 47-64.

SNEDECOR, G.W. & COCHRAN, W.G., 1967. Statistical methods. Sixth Ed. The Iowa State University Press, Ames.

THOMAS, P., 1986. Radiation preservation of foods of plant origin. Part IV. Subtropical fruits: citrus, grapes and avocados. *CRC Crit. Rev. Food Sci. Nutr.* **24**, 53-89.

UNTERSTENHÖFER, G., 1963. The basic principles of crop protection field trials. *Pflanzenschutz-Nachr. Bayer* **16,** 81-164.