

Fig. 2. Survival of 'Gala' on 18 dwarfing rootstocks to a severe fire blight infection in their third leaf. Mean separation using logistic regression (P = 0.05).

The ultimate solution to the loss of trees due to fire blight infection in the rootstock is to use resistant rootstocks coupled with resistant scion cultivars. It is hoped that some of the Vineland, Geneva, or other rootstock series will prove to have the necessary desirable horticultural characteristics, as well as resistance so that this problem can be avoided in the future.

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# Concentration by Time Product and Gas Penetration after Marine Container Fumigation of Table Grapes with Reduced Doses of Sulfur Dioxide

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**ADDITIONAL INDEX WORDS.** Vitis vinifera, export markets, gray mold, Botrytis cinerea, sulphur dioxide, total utilization SO, fumigation

SUMMARY. Reduced doses of sulfur dioxide (SO<sub>2</sub>) were evaluated for the fumigation of marine containers with respect to the concentration × time (CT) product and gas penetration. Two commercial export containers were loaded at 32 °F (0 °C) with 20 metric pallets  $[40 \times 48 \text{ inches } (102.5$  $\times$  123.1 cm) comprised of 72 expanded polystyrene foam boxes (12 tiers, 6 boxes/tier) of table grapes (Vitis vinifera) and fumigated with 1.0 and 0.5 lb (0.454 and 0.227 kg) SO<sub>2</sub>, respectively. A third marine container was loaded with 20 metric pallets comprised of 84 plastic boxes of table grapes (14 tiers, 6 boxes/tier) and fumigated with 0.25 lb (0.113 kg) SO<sub>2</sub>. The boxes contained 16 lb (7.3 kg) of table grapes distributed in nine polyethylene cluster bags

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We thank the California Table Grape Commission for financial support and Gerawan Farming (Reedley, Calif.) for providing the fruit and the export containers and allowing us to use their facilities.

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enclosed in a perforated polyethylene box liner. Fumigations were performed through the bottom seal of the rear door from pre-weighed compressed SO<sub>2</sub> cylinders. CT product was calculated after taking samples of the atmosphere in the container every 5 to 10 min and measuring the ambient SO, concentration with a gas sampling pump and colorimetric dosimeter tubes. Pallet and box penetration of the gas was assessed by placing passive colorimetric SO, dosimeters inside the cluster bags in boxes located in both the third and ninth center boxes from the top of pallets located in the front, center, and rear of the load. Fumigations with 1.0, 0.5, and 0.25 lb SO<sub>2</sub>, with calculated CT products at 32 °F of 925, 360, and 40 ppm-h (μL·L<sup>-1</sup>·h<sup>-1</sup>) respectively, were found to provide excessive, adequate, and insufficient SO, doses.

xport markets for California table grapes are often ✓ located at least 10 d away by overseas transport and most of the produce is shipped in refrigerated marine containers. Shipments in 1999 totaled 84.1 million 21-lb (9.53-kg) carton-equivalent units (Cook, 2000). Gray mold, caused by *Botrytis cinerea*, is one of the most significant problems limiting quality of shipped produce (Capellini et al., 1986; Nelson, 1991). Control of gray mold during cold storage of table grapes is achieved by fumigations with sulfur dioxide (SO<sub>2</sub>), a practice that has been used in California for more than 70 years (Nelson, 1985). An initial sulfur dioxide treatment can kill fungal inoculum present on the fruit surface, but subsequent periodic fumigations are needed to prevent gray mold nesting caused by mycelial spread from infected berries to adjacent healthy berries (Nelson, 1985).

Usual handling of table grapes for shipment is as follows: fruit are selected, classified into quality categories, packed in boxes, and palletized in the field, then transported to the packinghouse, precooled as soon as possible, treated with gaseous SO<sub>2</sub>, and held at about 32 °F and high relative humidity (RH) until the container is loaded. Pallets are normally refumigated weekly during the cold storage period. It has been recommended to perform, when possible, precooling and initial SO, fumigation simultaneously in a forced-air precooling room under the total utilization system. This method uses about 75% less SO, than traditional initial fumigations (Luvisi et al., 1992). In most cases, loaded containers are gassed again at the loading point (shipping) in a last effort to assure a good arrival. In other cases, grapes may be packed with a two-phase in-package SO<sub>2</sub> generating pad and shipped without an initial fumigation. However, recent studies (Crisosto et al., 1994, 2000) showed that in California this procedure is not as effective in controlling decay as the application of an initial fumigation to fruit packed with a vented plastic box liner and a slow-release generator.

The action of SO<sub>2</sub> during fumigation is described in terms of the toxic concentration and the amount of time that the gas remains in contact with the target organism. These factors are multiplied and the gas doses are expressed by the CT product (concentration × time, typically measured in ppm-h) (Luvisi et al., 1992; Smilanick and Henson, 1992).

Sulfite residues and phytotoxicity (bleaching of fruit color and hairline splits) are the main problems associated with sulfur dioxide fumigations. In 1986, a residue tolerance of 10 ppm (μg·g<sup>-1</sup>) sulfite on table grapes was established by the US Environmental Protection Agency (EPA) (EPA, 1986). Bleaching occurs when the gas is released at excessive concentrations and penetrates into the stem end or through lenticels or skin wounds, causing bleached or sunken areas (Ryall and Harvey, 1959). Hairline splits on the berry surface appear to be related to excessive sulfur dioxide doses. Symptoms are microscopic longitudinal splits often followed by exudation of pulp juice (Santiago and Hanke, 2000; Zoffoli et al., 2000a).

Cylinders of compressed SO<sub>2</sub> (pressurized liquid gas) are used in California for storage and container fumigation. The rates for SO<sub>2</sub> use in containers in commercial use today were adapted from those originally developed for railcars (Jacob, 1929). These very high rates were developed to overcome densely packed wood boxes, and poor ventilation. For example, Uota and Harvey (1964) stated that 3 to 5 lb (1.36 to 2.27 kg) of SO<sub>2</sub> were commonly used commercially to fumigate railcars of 2,360 ft<sup>3</sup> (66.83 m<sup>3</sup>) capacity typically containing about 1000 boxes of 20 to 24 lb (9.07 to 18.14 kg) each. They found that even at these high fumigation rates, high rates of air movement and ventilation channels were needed

among the boxes. No similar research characterizing SO<sub>2</sub> distribution, residues, or bleaching injury with modern refrigerated containers has been done. We believe that a correct balance between decay control, SO<sub>2</sub> residues and fruit injury could be maintained with the application of lower SO<sub>2</sub> doses whenever the gas is uniformly distributed within the container. Our objective was to measure actual CT and SO<sub>2</sub> distribution following marine container fumigations with low amounts of SO<sub>2</sub>.

## **Materials and methods**

Three trials with commercial marine containers were conducted during the 1999 and 2000 table grape seasons in the San Joaquin Valley (California). In every case, loading and fumigation techniques followed common export commercial procedures in use today in California. In the first trial, a 2,377 ft<sup>3</sup> (66.14 m<sup>3</sup>) refrigerated export container (Maersk Container Industri AS, Tingley, Denmark) was loaded with 20 metric pallets comprised of 72 expanded polystyrene foam boxes of table grapes (12 tiers, 6 boxes/tier) and fumigated with 1 lb SO<sub>2</sub>. In the second trial, a 2,366 ft<sup>3</sup> container (65.84 m³; Maersk Container Industri AS, Tinglev, Denmark) was loaded with 20 metric pallets comprised of 72 expanded polystyrene foam boxes (12 tiers, 6 boxes/tier) and fumigated with 0.5 lb SO<sub>2</sub>. And in the third trial, a 2,352 ft<sup>3</sup> container (65.45 m<sup>3</sup>; Maersk Container Industri AS) was loaded with 20 metric pallets comprised of 84 plastic boxes (14 tiers, 6 boxes/tier) and fumigated with 0.25 lb SO<sub>2</sub>. The following methodology was common to all three trials.

CONTAINER LOADING. The metric pallets were pinwheeled into the container in the packinghouse cold storage facilities. An additional tier of boxes was added to the top of the pallets to bring the load to within 6 inches (15.4 cm) of the limit line. The packages were commercial 16-lb (7.3 kg) boxes containing nine polyethylene cluster bags of 'Redglobe', 'Ruby Seedless' and/or 'Red Seedless' table grapes with a slowrelease SO, pad, all enclosed in a microperforated polyethylene box liner (1.2% vented area). The pinwheeled loading pattern left no more than 1 inch (2.6 cm) of clearance between pallets or between the pallets and sidewalls. There were about 10 inches (25.6 cm) of uncovered floor at the rear of the container. The container drain holes and air

exchange vent were closed for fumiga-

**FUMIGATION.** The targeted amount of SO, was introduced into the container from pre-weighed compressed gas cylinders (Fruit Doctor; Snowden Enterprises Inc., Fresno, Calif.) through a plastic tube inserted under the rear door bottom seal. The temperature in the container during loading and fumigation was set at 32 °F. A fan located in the front bulkhead distributed the gas in the container. Cold air was delivered through T-channels in the floor of the container, forced up through the boxes of fruit, and then returned to the refrigeration unit at the top of the front bulkhead. The introduction of the gas lasted no more than 5 min. About 70 or 95 min after fumigation, the container was vented by opening the air exchange vent as well as the rear door. Cold air circulation was not stopped during venting and SO<sub>2</sub> concentration in the container dropped to near zero after 10 to 15 min of venting.

**DETERMINATION OF FUMIGATION EFFICIENCY.** The SO<sub>2</sub> concentration in the container was measured by withdrawing air samples through plastic tubing placed at the bottom rear pallet position, then fed out under the rear door seal. Sampling was performed first just after introduction of the gas and every 5-10 min thereafter. The samples were analyzed using a gas-sampling pump (model 8014-400A; SE certified model 42CFR84; Matheson Kitagawa, East Rutherford, N.J.) and various colorimetric dosimeter tubes with detection limits from 1.25 to 3,600 ppm (1 ppm = 2.62 mg·m<sup>-3</sup>) SO<sub>2</sub> (Gastec-Sensidyne No. 5L, 5D, and 5M; Gastec Corp., Fukaya, Ayase-City, Japan). Data on SO, concentration over time were used to calculate the container's CT according to the procedure described by Luvisi et al. (1992).

The amounts of SO<sub>2</sub> surrounding the grapes were measured by placing passive colorimetric dosimeter tubes (Gastec-Sensidyne Nos. 5D and 5DH; Gastec Corp.) within the cluster bags that contained the fruit in boxes placed in the front (second row from bulkhead), center (fourth row from bulkhead), and rear (eighth row from bulkhead, third from rear doors) of the load. Two different passive tubes were used in each sampled bag. One 5D tube (detection limits of 0 to 100 ppm-h) and one 5DH tube (detection limits of 10 to 600 ppm-h) were placed in both the

third and ninth center box from the top of each pallet (including the additional top box). Passive dosimeters were placed in center boxes as recommended for inbox CT determination by Luvisi et al. (1992). After venting, the container was unloaded and the passive dosimeters recovered. The container was then loaded again and shipped to destination.

### Results

**1-LB SO<sub>2</sub> FUMIGATION.** The concentration of SO<sub>2</sub> measured in the container was 1,500 ppm 5 min after gas introduction. This concentration decreased to 250 ppm after 90 min (Fig. 1). Based on the measurements performed 5, 15, 20, 30, 40, 50, 70, and 90 min after the injection of the gas (Fig. 1), the calculated container's CT from the 1-lb fumigation in the 2,377-ft<sup>3</sup> container was 925 ppm-h.

After 95 min of exposure, the container was vented, unloaded and the passive dosimeters checked. At all of the sampling positions the 5D tubes (detection limits of 0 to 100 ppm-h) were completely saturated. With the exception of the rear top and rear bottom pallet positions, the 5DH tubes (detection limits of 10 to 600 ppm-h) measured a dose higher than 600 ppm-h. The rear top and rear bottom pallet positions measured 400 and 350 ppm-h, respectively (Table 1).

**O.5-LB SO<sub>2</sub> FUMIGATION.** The concentration of SO<sub>2</sub> measured in the container was 1,000 ppm 5 min after gas introduction. This concentration decreased to 200 ppm after 70 min (Fig. 1). Based on the measurements performed 5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 55, 60, and 70 min after

the injection of the gas (Fig. 1), the calculated container's CT from the 0.5-lb fumigation in the 2,366-ft<sup>3</sup> container was about 360 ppm-h.

After 70 min of exposure, the container was vented, unloaded and the passive dosimeters checked. At all of the sampling positions the 5D tubes were completely saturated. With the exception of the rear top and rear bottom pallet positions, the 5DH tubes measured from 400 to 250 ppm-h depending upon pallet and box position. The rear pallet position, bottom box recorded the lowest dose at 250 ppm-h (Table 1).

**0.25-LB SO<sub>2</sub> FUMIGATION.** The concentration of SO<sub>2</sub> measured in the container was 100 ppm 5 min after gas introduction. This concentration decreased to 5 ppm after 70 min (Fig. 1). Based on the measurements performed 5, 8, 15, 20, 38, 47, 60, and 70 min after the injection of the gas (Fig. 1), the calculated container's CT from the 0.25-lb fumigation in the 2,352-ft<sup>3</sup> container was about 40 ppm-h.

After 70 min of exposure, the container was vented, unloaded and the passive dosimeters checked. At all of the sampling positions the 5D tubes measured from 40 to 7 ppm-h depending upon pallet and box position. The top boxes at the center and rear pallet positions recorded the lowest doses at 7 ppm-h (Table 1).

Fig. 1. Sulfur dioxide ( $SO_2$ ) concentrations [ppm ( $\mu$ L·L<sup>-1</sup>)] measured in air samples from a marine container loaded with table grapes withdrawn during fumigation with 1.0, 0.5, or 0.25 lb (0.454, 0.227, or 0.113 kg) of gas. Air samples were taken from the bottom rear pallet position.

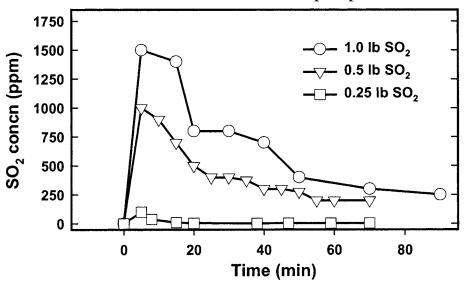


Table 1. Sulfur dioxide ( $SO_2$ ) doses (concentration  $\times$  time) in expanded polystyrene foam or plastic boxes containing fruit in cluster bags enclosed in perforated box liners that had been palletized and loaded into a marine container and fumigated with 1.0, 0.5, or 0.25 lb (0.454, 0.227, or 0.113 kg) of gas.

Pallet position	Box position	SO, dose [ppm-h ( $\mu \mathbf{L} \cdot \mathbf{L}^{-1} \cdot \mathbf{h}^{-1}$ )]		
		1.0 lb SO <sub>2</sub> <sup>z</sup>	0.5 lb SO <sub>2</sub> <sup>z</sup>	0.25 lb SO <sub>2</sub> <sup>y</sup>
Rear	Тор	400	400	7
Rear	Bottom	350	250	10
Center	Тор	>600	350	7
Center	Bottom	>600	350	15
Front	Тор	>600	400	30
Front	Bottom	>600	375	12

 $<sup>^{</sup>z}$ Expanded polystyrene foam boxes;  $SO_2$  measured using Gastec-Sensidyne 5DH passive dosimeter tubes (Gastec Corp., Fukaya, Ayase-City, Japan).

### **Discussion**

The purpose of the SO<sub>2</sub> container fumigation of table grapes is to maximize decay control while minimizing losses due to SO<sub>2</sub> damage in order to adequately preserve the fruit during shipping and offer high quality produce in the arrival markets. At 32 °F, a CT product of at least 100 ppm-h in the air spaces surrounding the berries kills both the spores and the mycelia of B. cinerea (Smilanick and Henson, 1992; Luvisi et al., 1992). This CT dose can be obtained with an average concentration of 100 ppm for 1 h, 200 ppm for 0.5 h, or 25 ppm for 4 h, or an equivalent combination of concentration and time. However, gaseous SO, should be applied at higher doses to ensure that at least such a minimum effective dose reaches the atmosphere surrounding the grapes. Sorption of the gas by fruit and packaging materials and resistance of the load to gas penetration force the use of higher SO, doses. Gas dose, however, has to be low enough to cause minimal phytotoxicity and to keep sulfite residues under regulatory tolerances. These problems have led investigators to search for fumigation technologies that can effectively use reduced levels of SO, (Luvisi et al., 1992; Marois et al., 1986). Alternatives to the use of SO, such as chlorine dioxide, carbonates, ethanol or ozone are also being evaluated (Crisosto and Smilanick, 2000; Palou et al., 2001).

Multiple factors influence the distribution and penetration of SO<sub>2</sub> during fumigation. The type of container, number and position of fans, and location of channels where the air is allowed to circulate influence how the

gas is distributed in and around the load (Luvisi et al., 1995; Uota and Harvey, 1964). An additional amount of fumigant would be required when there is water condensation in the container because considerable proportions of the gas are readily absorbed by free water (Uota and Harvey, 1964). We did not observe water condensation in these trials. Concentration of SO, also depends on the position of the pallet in the container and on the position of the box in the pallet. In our trials, penetration at the rear bottom position was in general lower than at the center or the front. This may be due to leaving the small portion of floor near the rear doors uncovered. The packaging materials also have a great influence on SO, penetration. Luvisi et al. (1992) observed that SO, penetration was higher in foam boxes than in wood-end [Technical Kraft Veneer (TKV)] and corrugated boxes, and higher in wood-end than in corrugated boxes. In their experiments, gas penetration in boxes with fruit in plastic cluster bags (without box liners) was not retarded when compared to gas penetration in boxes with plainpacked grapes. In laboratory experiments, grapes packed in expanded polystyrene foam boxes sorbed ca. 50% less SO, as did those in wood-end boxes; grapes in fiberboard boxes sorbed ca. 50% more SO<sub>2</sub> than did those in wood-end boxes (Harvey et al., 1988). We used polystyrene and plastic boxes, which are materials that sorb little SO, (Harvey et al., 1988). For that reason, we assume that CT and SO<sub>2</sub> distribution with either material would not differ substantially.

Our results showed that the 1-lb SO<sub>2</sub> container fumigation, with a cal-

culated CT of 925 ppm-h and a dose higher than 600 ppm-h in most of the box positions, under the experimental conditions, was an excessive dose for the purposes of the treatment. This rate may induce berry bleaching and rachis pitting in susceptible cultivars. A level of sulfite residues higher than the tolerance of 10 ppm may also be of concern. Therefore, worse consequences on fruit quality should be expected from container fumigations of 3 to 5 lb, and even worse when the produce have already received an initial fumigation and multiple SO, gassings during the cold storage period prior to this container fumigation. Furthermore, because the grapes were packed with plastic bags and box plastic liners and these materials certainly reduced the amount of gas that contacted the fruit, the negative effects of the treatment at these rates would be even greater on plain-packed grapes. In contrast, the 0.25 lb SO, fumigation did not generate a sufficient dose (40 ppm-h) in the wellvented plastic boxes tested. This dose is lower than the minimum acceptable level of 100 ppm-h necessary to effectively control gray mold (Smilanick and Henson, 1992) and thus cannot be recommended. A dose of about 360 ppm-h was obtained after fumigating a container of foam boxes with 0.5 lb of SO<sub>2</sub>. Under the conditions of our trial, this dose could be appropriate to maintain a balance between decay control and SO<sub>2</sub> damage and residues.

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# Legume and Grass Cover Crops for Seedless Watermelon Production

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Additional index words. crimson clover, hairy vetch, rye, Secale cereale, Trifolium incarnatum, Vicia villosa, nitrogen fertilizer

SUMMARY. Although there is increasing interest in reducing the use of nitrogen (N) fertilizers due to the potential of unused N causing pollution of surface and groundwater, N is a major nutrient for plant growth. Our objective was to determine the potential of using winter legume cover crops to meet the N needs of seedless watermelon (Citrullus lanatus), a potential cash crop for farmers in Virginia. Fruit number, fruit weight, fruit yield, and fruit quality traits (flesh to rind ratio, water content, total soluble solids, sugar content, and pH) of seedless watermelons were evaluated in replicated experiments in Virginia at three locations during 1997-98 and two locations during 1998-99 following cover crop treatments consisting of crimson clover (Trifolium incarnatum), hairy vetch (Vicia villosa), crimson clover + rye (Secale cereale), hairy vetch + rye, and a bareground control treatment that received 100 lb/acre (112 kg·ha<sup>-1</sup>) of N. At all five locations, the bareground control treatment resulted in fewer fruit [1803 fruit/acre (4454 fruit/ha)], lower fruit weight [9.8 lb (4.5 kg)], and lower fruit yield [8.9 tons/acre (20.0 t·ha-1)] compared to the four cover crop treatments. The crimson clover + rye and hairy vetch treatments resulted in highest number

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Contribution of Virginia State University, Agricultural Research Station, journal article series 221. The use of trade names or vendors does not imply approval to the exclusion of other products or vendors that may also be suitable.

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