NEW TABLE GRAPE POSTHARVEST TECHNOLOGIES

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During the last decade, new postharvest technologies have been adapted in commercial table grape operations. Several cultivars with different colors and quality attributes became available to the industry as results of intense activity of the USDA (Fresno) breeding program. Development of a minimum quality index to satisfy domestic and foreign consumers based on consumer preference test (sensory evaluation) is being supported in California. Table grape marketing has become more active across the world, and grape supplies are available almost year-round in the USA. Because of the attractive export market demand in areas with different ethnic backgrounds, new cultivars with a long market life and different taste components are becoming more important than earlier in the decade.

For example, 'Red Globe' grapes with SSC $\geq$16.1% were liked "very much" by Chinese and domestic consumers (combined). Approximately 85% of consumers liked 'Red Globe' grapes with these quality attributes. There were no significant differences in degree of liking between grapes within the 16.1% and 17.0% SSC range. In our studies with other commodities, the highest levels of consumer acceptance ranged from 75% to 85%. However, 'Red Globe' grapes with SSC <16.5% and a SSC:TA of 20.1-22.5 were always liked, but with different degrees of liking. 'Red Globe' grapes with these quality attributes satisfied only 42% of Chinese consumers and 76% of domestic consumers. The percentage of consumers that disliked these grapes was the same for both ethnic groups (~20%). The differences in the percentages of Chinese and domestic consumers accepting the grapes were due to the "neither dislike nor like" category. Chinese consumers chose the "neither dislike nor like" category in 38% of the cases, while only 6% of domestic consumers chose it. Titratable acidity played an important role on the Chinese consumer acceptance.

These new market conditions also encourage a careful cultivar selection based on flavor, susceptibility to specific export treatment (methyl bromide) and long postharvest market life.

THE USE OF SULFUR DIOXIDE TO LIMIT GRAY MOLD

Gray mold (*Botrytis cinerea*) is being well controlled by careful use of sulfur dioxide gas (SO_2) during storage. By using the UC-Total Utilization System, decay is successfully controlled without increases in SO_2 residues and/or air pollution during cold storage. SO_2 total utilization was recently introduced in the California industry. It has been demonstrated that the amount of sulfur dioxide gas needed to kill *Botrytis* spores, or to inactivate exposed mycelium, is dependent on the concentration and the length of time the fungus is exposed to the fumigant. A cumulative concentration, calculated as the product of the concentration and the time, called "CT product," describes the sulfur dioxide exposure needed to kill the decay organism. A CT of at least 100 ppm-hour is the minimum required to kill spores and mycelium of *Botrytis* at 0°C (32°F) or approximately 30 ppm-hour at 20°C (68°F). The CT-100 dose can be obtained with an average concentration of 100 ppm for 1 hour, or 200 ppm for ½ hour, or 50 ppm for 2 hours, or an equivalent combination of concentration and time. This finding was the basis for the development of the total utilization system.

The total utilization system differs from the traditional system in that there is no excess SO_2 fumigant at the end of the fumigation treatment, thus reducing air pollution and sulfite residues on grape. It can be used with forced-air cooling for initial fumigation and in cold storage for subsequent periodic treatments. Total utilization often uses about half as much sulfur dioxide as the traditional method and improves uniformity and effectiveness of the SO_2 fumigant. In the total utilization system, the first fumigation (initial) is done in conjunction with forced air-cooling. The forced air flows through the boxes and ensures good penetration of SO_2 even to the center boxes within a pallet. In most combinations of boxes and packs, this system produces over 80% penetration, measured as percent of the room air CT product. During storage, fumigation is applied every 7-10 days (passive fumigation). After SO_2 application in the room, fans should run at high speed for over 3 hours. In this way, fruit, packaging materials, and room surfaces absorb nearly all of the SO_2. At the end of fumigation, the concentration of SO_2 in the room air should be less than 2-5 ppm and no venting or scrubbing is needed. In this total utilization system, each cold storage room should be calibrated to determine the amount of SO_2 to use. Center boxes within a pallet have lower SO_2 exposures than corner boxes, and pallets closest to the SO_2 inlet have higher fumigant exposures compared to those farthest away. To check fumigant penetration and distribution, inexpensive SO_2 dosimeter tubes are available. These dosimeters were originally designed for human safety monitoring. There is a large difference in SO_2 penetration according to box materials. For example, SO_2 penetration is higher in EPS boxes than wood-end and corrugated boxes; SO_2 penetration in corrugated is lower than in wood-end boxes.

Dosimeters designated for SO_2 fumigation doses at marked levels from 0-150, 0-100 and 600 ppm-hr are available. These passive dosimeter tubes work well for measuring the SO_2 CT product inside packed grape boxes. The glass dosimeter tubes are placed in the center of the boxes inside tissue wraps of cluster bags if these are present, and usually in boxes located in the center of the pallets. After fumigation the tubes are removed promptly and the ppm-hr exposure to SO_2 is directly recorded. The tubes should be read promptly because some can overestimate the dosage if examining their color reaction is delayed. A dose of 100 ppm-hr is the minimum adequate dose. This allows the operator to adjust the amount of fumigant applied to insure that most boxes are adequately protected from decay but not exposed to fumigant levels that might cause excessive residues and bleaching. Details on this work are available in the University of California DANR, Publication 1932 (Luvisi et al., 1995).

THE USE OF SO_2 PADS FOR EXPORT MARKETS

Improvement in the packaging (box liners, cluster bags etc.) and SO_2 release pad to limit gray mold development during long-term shipment without inducing bleaching or increasing SO_2 residues is being investigated. It is important to point out that the ideal packaging/SO_2 pad system should be developed for each cultivar and for specific industry conditions.

The role of even small amounts of SO_2 within a box during storage/shipment is being studied by our group. For this, individual 'Red Globe' table grape berries, with the pedicels attached, were surface disinfected by dipping them for one minute in 0.5% HOCl solution and then allowed to dry. Berries were then placed on a plastic grid in an 8.3 liter plastic container and sprayed with *Botrytis cinerea* (20,000 spores per ml) to the point of runoff. The berries were allowed to dry at room temperature, then the boxes moved to a 0°C room and attached to a flow-through fumigation system. Berries were exposed to a continuous flow of 0, 0.25, 0.50, 1.0, 2.0 or 3.0 ppm SO_2 (inlet concentrations) under high relative humidity conditions for four weeks. After treatment, berries were evaluated for presence or absence of decay (mycelium or slip-skin). To study nesting formation, individual 'Red Globe' table grape berries were wound inoculated then held at 20°C until covered with mycelium. These decayed fruit were placed in the center of petri dishes and surrounded by six healthy berries that had been disinfected as previously described. Seven petri dishes were placed in each 8.3 liter plastic container and treated at 0°C with a continuous flow of 0, 0.25, 0.50, 1.0, 2.0 or 3.0 ppm SO_2 (inlet concentrations) under high relative humidity conditions for four weeks. Each week berries were evaluated and the number to which mycelium had spread was counted. Outlet SO_2 concentrations were 0.0, 0.0, 0.0, 0.1, 0.2 and 0.2 ppm for the 0, 0.25, 0.50, 1.0, 2.0 or 3.0 ppm inlet SO_2 treatments, respectively. This means that much of the SO_2 introduced into our experimental containers was absorbed into the fruit and experimental containers. The relationship between inlet and outlet SO_2 concentrations requires more study before we can make a final conclusion on the minimum effective SO_2 concentration inside small containers over time.

As a preliminary conclusion of this work in progress, inlet sulfur dioxide concentrations ≤ 3 ppm are not effective in preventing *Botrytis* spore germination. Inlet sulfur dioxide concentrations >0.50 ppm are effective in suppressing mycelium growth and the spread of decay from one berry to another within this time period. It appears that low concentrations can, however, suppress decay nesting during storage and transportation within this time period.

THE USE OF SO_2 PADS FOR EXPORT CONTAINERS

The use of SO_2 pads without liners, in a container shipment with vents closed, on decay and fruit quality has been tested several times. Shipping containers overseas with the vents closed provided an excellent fruit temperature control (Fig. 1), accumulation of SO_2, a slight increase in CO_2 and C_2H_4, and decrease in O_2 without affecting grape quality. In four of the containers, carbon dioxide concentrations ranged from 0.4% to 1.4%, oxygen concentrations decreased from 21% to 17%, and ethylene concentrations reached 32 ppb to

52 ppb during the 21 day simulated shipment. Fruit flesh temperature was maintained at -0.6°C to 0.6°C. These temperatures were very close to the container set point (0°C). The use of slow release SO_2 pads without box liners limited *Botrytis* nest development during shipment in a non-vented container.

RELATIONSHIP BETWEEN ETHYLENE AND GRAY MOLD DEVELOPMENT

Individual 'Red Seedless' table grapes were surface disinfected and inoculated with *Botrytis cinerea* and placed into plastic containers. The containers were connected to an ethylene flow-through system and placed in a cold storage room at 0°C and 95% RH for 2 months. Six ethylene concentrations were assayed: 0 (control), 0.125, 0.25, 0.50, 1.0, and 2.0 ppm. Incidence of decayed spray-inoculated berries and number of decayed berries surrounding the central inoculated berry (nest formation) were checked monthly. There were no significant differences found in either decay incidence (90-100%) or nesting (30-40% of infected berries surrounding the inoculated berry) between ethylene concentrations after 2 months of storage at 0°C.

DECAY PREDICTION

Several gray mold detection techniques such as Harvey's modified, freezing and PCR are being examined to predict and segregate grape lots based on their potential decay. In this way, "strong" lots can be identified and segregated for long-term storage and shipment. On the other hand, "weak" lots can be marketed earlier in the season. In the application of these methods, sampling is becoming a critical step in the success of the technique.

CALIBRATING SO_2 CONTAINER FUMIGATION

One-half pound of "Fruit Doctor" SO_2 was introduced into the containers through a plastic tube inserted through the rear door bottom seal. The concentration of SO_2 measured in the container was 1,000 ppm five minutes after one-half pound gas introduction (Fig. 2). This concentration decreased to 200 ppm after 70 minutes. At this time the container was vented. After the SO_2 was exhausted, the container was unloaded and the passive dosimeters recovered. At all of the sampling positions the Gastec 5D (0-100 ppm-h) tubes were completely saturated. The Gastec 5DH tubes (50-600 ppm-h) measured from 400-250 ppm-h depending upon pallet and box position. The calculated C x T from the one-half pound fumigation in the 2,366 ft^3 container was 360 ppm-h.

PACKAGE AND PACKAGING CHANGES

Changes in containers and packaging to reduce shatter, improve cooling process, and protect grape quality have also been made. A complete evaluation on the effect of container size on grape quality was performed by the UC group (UC DANR Publication 1934, Luvisi et al., 1995).

The use of a restricted cluster bag has been tested in several cultivars. 'Ruby Seedless' grapes were packed by using the commercial cluster bag (with 60% perforation) or the restricted cluster bag (with 1.4% perforation) in foam boxes. Five boxes (10 kg) were field packed for each treatment/evaluation date and stored at 32°F with 90% relative humidity. Forced-air cooling and initial fumigation were done at the same time. SO_2 penetration was measured initially and weekly during the storage period. Grapes were removed after 3, 6, and 9 weeks of cold storage for evaluation. Fruit were inoculated with a *Botrytis* solution before cold storage (32°F/90%RH). Decay, stem condition (stem browning and dryness), SO_2 phytotoxicity, shattering incidence, and buyer opinion grade were measured on each evaluation date. After 3 weeks of cold storage, the use of the restricted cluster bag with a 1.4% perforation reduced stem browning and increased the buyer opinion grade without affecting decay and phytotoxicity as compared to grapes stored in bags with 60% perforation. Grapes packed in the bags with 1.4% perforation were categorized as "good" according to the buyer opinion grade. After 6 weeks of cold storage, grapes from the bags with 1.4% perforation showed better stem condition (browning and dryness) than grapes from the control. In both treatments, decay incidence was low. By the 9-week storage evaluation date, stem dryness was classified as "severe" in the control (commercial cluster bag, 60% perforation) fruit, but "moderate" in the restricted cluster bag. According to the buyer opinion grade, grapes packed in the restricted cluster bag were categorized as "fair" while grapes packed in the commercial cluster bag were categorized as "poor."

During this trial, fruit packed in the top of the box had a higher shattering incidence than the fruit packed in the bottom of the box. However, fruit packed with the restricted cluster bag (1.4% perforation) had less shattering than fruit packed with the commercial bag (60% perforation), 16.7% compared to 21.3%.

During the storage period, the restricted and commercial cluster bags did not show any excessive condensation. SO_2 penetration was adequate in both types of cluster bags during the initial treatment and weekly fumigations.

The results indicate that the restricted cluster bag was more effective to reduce water loss and maintain stem freshness without interfering with the SO_2 penetration than the current cluster bag commercially used. Shatter was always higher in the top position than bottom position within the box, regardless of the type of cluster bag used.

Because a high incidence of bruising has been observed in 'Thompson Seedless', we decided to look at this situation. A small test was designed to understand bruising development. Position of the bags in the box was not related to bruising damage. The bruising damage was random in the box, suggesting that the packers do not produce damage. In general, bruised berries were located in the bottom position of the bag touching the bottom of the box. Crew handling during hauling was an important factor in bruising damage incidence. The gentle handling crew had an average of 30 bruised berries in six boxes, while the normal handling crew had an average of 68 bruised berries in six boxes. The use of the bottom pad (BP) reduced the number of bruised berries by nearly one half (from 64 berries to 34 berries in six boxes). Bruising incidence was almost double in the boxes toward the bottom of the pallet (65 berries) than toward the top of the pallet (32 berries). This indicates that the bruising damage is associated with pallet loading. Among all the treatments, grapes (gentle x top W-BP) located toward the top of the pallet had only 4 bruised berries. Grapes normally packed without a BP located toward the bottom of the pallet had 113 bruised berries. A large reduction in the number of bruised berries can be achieved if a BP and gentle handling are used during pallet loading.

SEARCHING FOR THE SULFUR DIOXIDE REPLACEMENTS

Several works on SO_2 replacement and or substitutes such as ethanol, chlorine dioxide, chlorine pads, pulsated ultraviolet, and carbonates are undergoing in different countries without too much success. Lack of contact between chemicals and organisms and potential damage have been the main difficulties. More work in this area is strongly recommended. High carbon dioxide exposure during low temperature storage has shown promising results on limiting *Botrytis* development and potential insect control. However, losses on grape taste, development of internal browning and stem browning should be carefully investigated. Also, preliminary information indicates that cultivars react in different ways to high CO_2 levels.

Carbonates: Smilanick's group evaluated the control of postharvest gray mold of table grapes by treatment with carbonate and bicarbonate salt solutions. Sodium carbonate (SC), potassium carbonate (PC), sodium bicarbonate (SBC), potassium bicarbonate (PBC) and ammonium bicarbonate (ABC) were tested without control of pH for their toxicity to spores of *Botrytis cinerea* in vitro, and the EC_{95} concentrations were 16, 17, 36, 58, and 163 mM, respectively. When bicarbonate solutions were tested at pH 7.2 (±0.2), the EC_{50} concentrations of ABC, SBC, and PBC were 26, 46, and 48 mM, respectively. In practical tests to control gray mold on grapes, among the bicarbonates, each applied at 500 mM, ABC was significantly more effective than SBC and PBC. It was also superior to PC (100 mM) and equal in effectiveness to SC (100 mM) and ethanol (70% wt/vol). The addition of 200 μg/ml chlorine to the bicarbonate salts significantly decreased gray mold incidence. Among all the treatments, berry condition was an important factor; there was significant decrease in control when wounded berries were treated compared to unwounded berries. The quality of grapes after treatment with ABC, SBC, ethanol, and chlorine was acceptable. Severe injuries, mostly brown spots on berries, occurred after SC, PC, and PBC treatments.

Chlorine Dioxide (CD): Studies to evaluate the effect of chlorine dioxide on the table grape decay *Botrytis cinerea* were done at the F. Gordon Mitchell postharvest laboratory. 'Thompson Seedless' and 'Ruby Seedless' table grapes were grown and packaged using standard commercial practices. The 'Thompson Seedless' grapes were packaged in plastic Stretch-Vent cluster bags in 19 lb (8.6 kg) corrugated boxes (6.5" H x 16" L x 13.5" W). The 'Ruby Seedless' grapes were packaged in plastic Stretch-Vent cluster bags in 19 lb (8.6 kg) TKV boxes (7" H x 16" L x 13.5" W). Table grapes were inoculated by placing a *Botrytis* inoculated berry within each cluster during packaging. For each cultivar, ten boxes per each treatment were commercially harvested and packed. The potential beneficial effect of chlorine dioxide during storage and simulated shipment was evaluated by placing one 20 g and one 5 g (25 g total per box) Engelhard Scentrex^{TM} sachet in the table grape boxes designated for ClO_2 treatment. In all of the experiments, to test the role of the initial SO_2 fumigation, half of the grapes were initially fumigated (200 CT).

Fruit were removed after 3 weeks storage and postharvest quality evaluated for the different treatments. Shatter, stem condition (color and flexibility), decay incidence, and ClO_2 and SO_2 phytotoxicity were measured as previously described (Crisosto et al., 1994). A scoring system of 1 = healthy, 2 = slightly brown (capstems), 3 = moderately brown (capstems + laterals) and 4 = brown (capstems + laterals + rachis) was used to evaluate stem color.

(capstems + laterals) and 4 = brown (capstems + laterals + rachis) was used to evaluate stem color. Shatter, decay, and phytotoxicity are expressed as the percentage by weight of the fruit affected.

The chlorine dioxide sachets did not significantly reduce the incidence of decay during storage of 'Thompson Seedless' or 'Ruby Seedless' table grapes at 32° or 38°F. The incidence of decay from the field was very high this season. For both cultivars, the incidence of decay was higher at 38°F than 32°F. Shatter and phytotoxicity were not affected by the chlorine dioxide treatment. If any testing is done in the future, we recommend combining the chlorine dioxide treatment with a perforated box liner. By enclosing the chlorine dioxide generator and fruit within a liner, the ClO_2 concentration may accumulate to reach a minimum effective concentration. A large chlorine dioxide pad that fits the size of the box is also suggested.

Controlled Atmosphere and Modified Atmosphere (MAP): High carbon dioxide (CO_2) and low oxygen (O_2) levels, controlled atmosphere conditions (CA), have been used to extend the postharvest life of many fruits and vegetables. CA has been demonstrated to reduce metabolic activity, softening, flesh browning, incidence of physiological disorders and ethylene susceptibility, thus extending postharvest life of apples, artichokes, Asian pears, Brussels sprouts, cabbage, cauliflower, cherries, citrus, honey melon, kiwifruit, lettuce, peaches, and pears. In strawberries, high CO_2 levels during postharvest storage were shown to maintain or even increase fruit firmness. Optimal combinations of low O_2 and high CO_2 levels have been developed for different species and even cultivars within the same species. The optimal CO_2 and O_2 levels may vary according to exposure period and temperature conditions. Although CA conditions during storage and/or shipment are recommended for many different commodities, their use are not commercially suggested for table grapes in California. Table grape quality and changes during postharvest life under CA conditions have not been studied in detail. Several potential advantages such as delaying senescence, decreasing stem and berry respiration, reducing stem browning, maintaining berry firmness, and retarding decay development have been suggested. Formation of "off flavors" and internal browning are a concern, however. Unfortunately, most of the in-depth work that has been done utilized the 'Alphonse Lavallee', 'Rakazi', 'Emperor', and 'Ribier' cultivars. There has been a little work done on 'Thompson Seedless' (Coachella Valley), but none on the other cultivars that are currently economically important such as 'Flame Seedless', 'Crimson Seedless', 'Ruby Seedless', or 'Red Globe'. Nelson (1969) found that berry internal browning incidence overcame the potential benefits of CA on his early trials with 'Thompson Seedless' from the Coachella Valley. Yahia et al. (1983) studied the effect of the addition of carbon monoxide to air or CA storage atmospheres on the postharvest quality and storage life of 'Thompson Seedless' table grapes. Decay and internal browning development were the main limitations beyond eight weeks storage. Unfortunately, the only CA treatment tried was 5% CO_2 + 2% O_2. As the formation of "off-flavors" and internal browning have been related to low O_2 and high CO_2 storage, respectively, these two parameters in addition to a taste score can be used to develop optimum CA concentrations (low O_2 and high CO_2 concentration thresholds) for different table grape cultivars.

Recent work carried out in England with 'Thompson Seedless' from Egypt (Berry and Aked, 1997) suggested a *Botrytis cinerea* inhibition by the CA treatments (5% O_2 + 15% CO_2) as well as the presence of sulfur dioxide pads. The reduction in stem browning has been thought to be a consequence mainly of the reduction in stem respiration. Stem respiration is almost 15 times higher than berry respiration (Gardea and Crisosto, unpublished data). Mitcham's group, using 'Thompson Seedless', is studying the benefits of a 45% CO_2 atmosphere insect quarantine treatment of up to two weeks.

Modified Atmosphere Packaging (MAP) technology, because of its low cost, is being developed with the goal of replacing CA for many commodities. With this new technology, targeted CO_2 and O_2 concentrations are reached mainly by using plastic liners with different gas permeabilities in conjunction with fruit respiration at a given temperature. By using this technology MAP conditions can be inexpensively achieved in the box or in a pallet unit. One problem is that some species, and even cultivars within the same species, can tolerate high CO_2 and/or low O_2 concentration better than others. The development of specific CA concentration requirements for different cultivars for long-term storage or long distance overseas shipment is essential for the application of this technique in the table grape industry. Dr. Retamales is developing plastic liners (MAP) for the Chilean table grape industry.

Ethanol: Work in Israel demonstrated that dipping of detached berries and grape clusters in ethanol almost completely eradicated the fungal and bacterial populations on the berry surface. Smilanick's group has demonstrated the efficacy of ethanol on decay control in lemons and stone fruit in controlling decay. The use of ethanol dipping could be well adapted to the stemless berry packed industry.

Pulsated Ultraviolet Light (PUV): Work done evaluating the use of pulsated ultraviolet light (PUV) on postharvest decay development has not been promising. PUV ($1 J/cm^2$) treated fruit had earlier and more severe *Botrytis cinerea* decay than the postharvest fungicide commercially treated fruit. The same results were obtained with wounded and not wounded inoculated fruit. A blemish developed on the surface of the peach fruits as a consequence of the PUV treatment. These results indicate that more detailed work on the effectiveness and safety of PUV as a fungicide replacement must be done before this technique can be used commercially.

Ozone: The potential effect of air ozone exposures to gray mold development on table grapes during storage was tested under controlled conditions. For the evaluation of gray mold incidence on grapes, a conidia suspension containing $2x10^4$ spores ml^{-1} of *B. cinerea* was uniformly sprayed for about 5 s on clusters of 'Thompson Seedless' table grapes. For the evaluation of *B. cinerea* nesting ability, 10 μl of a suspension of $2x10^6$ spores ml^{-1} of *B. cinerea* were injected in the flesh of one central single berry per cluster using a Hamilton syringe. Inoculated grapes were placed on wooden trays and stored for 7 weeks at 5°C and 90% RH under 0 (control room) or 0.3-ppm ozone. In every storage treatment, spray inoculation was applied to six replicates (trays) of two clusters each and syringe inoculation to six replicates of four clusters each. The number of infected berries per cluster and the number of infected berries surrounding the inoculated berry were recorded weekly. There were no significant differences on the incidence of gray mold on

'Thompson Seedless' table grapes inoculated by spraying their surface and stored at 5°C under 0 or 0.3 ppm ozone. After 7 weeks storage, decay incidence on air and ozone exposed clusters reached about 25% and 19%, respectively. However, gray mold nesting was strongly prevented under 0.3 ppm ozone. The percentage of decayed berries surrounding the inoculated berry in control and ozone treated clusters after 7 weeks storage at 5°C was 65% and 1%, respectively. *Botrytis* nests quickly developed on ozone treated clusters that were placed at 20°C after the 7-week exposure period.

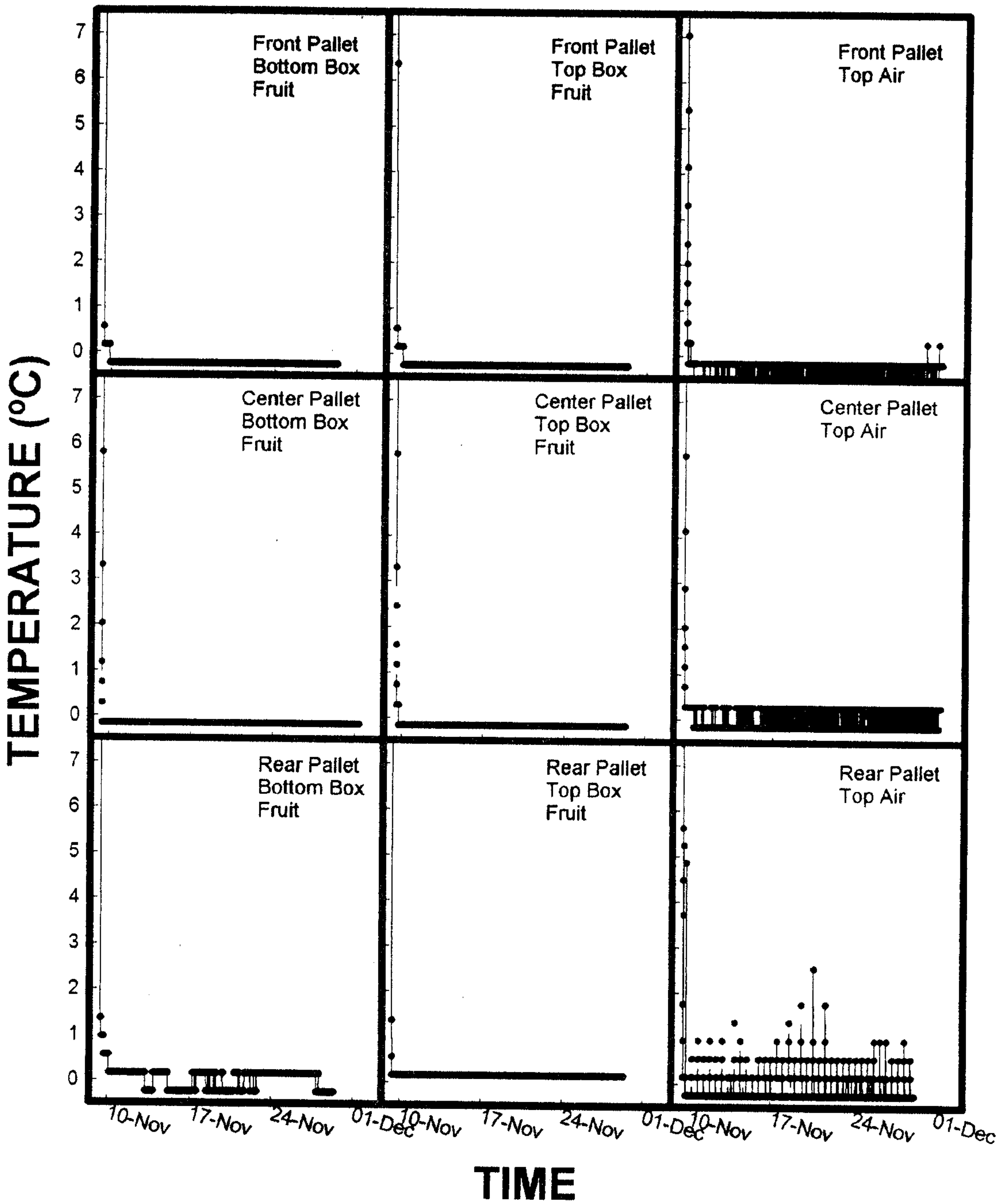

Fig. 1. Table grape pulp and air temperatures in a closed vent container during a 23-day simulated shipment.

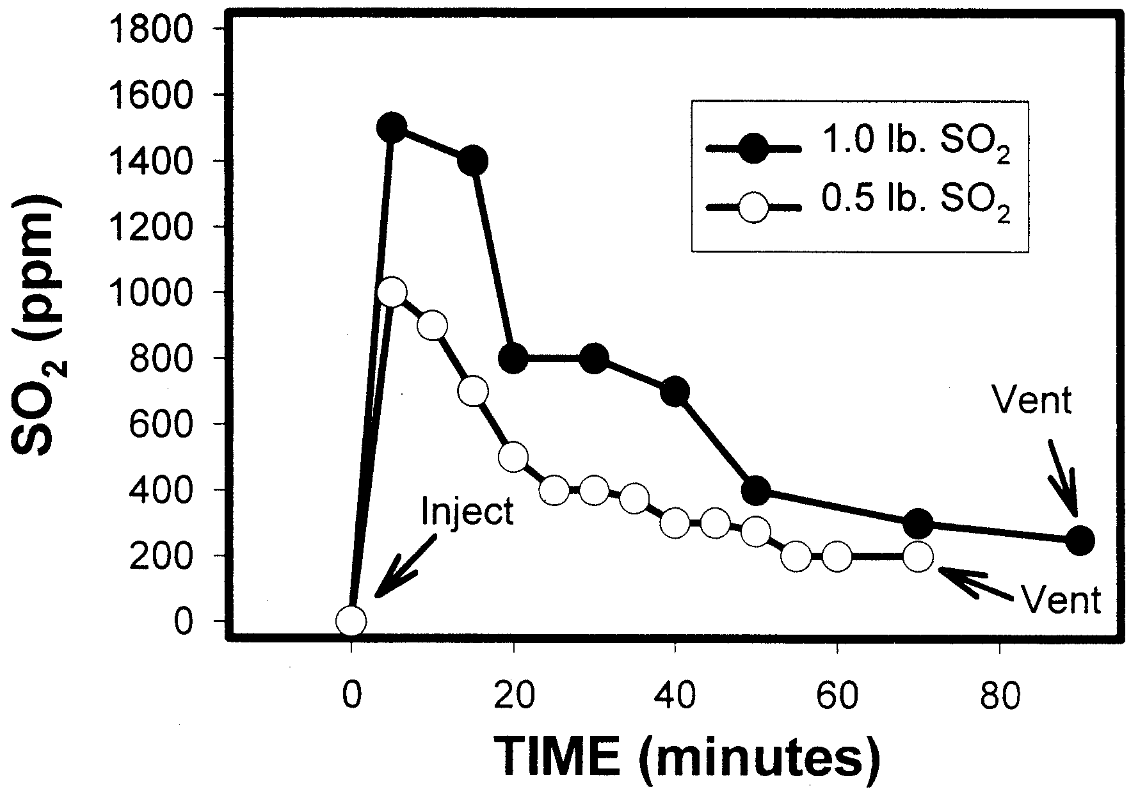

Fig. 2. Sulfur dioxide concentration during container fumigation using one and one-half pound application.

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