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## CARBON DIOXIDE EVOLUTION AS A MEARUSE OF

 dAmage to fresh apples handled in CORRUGATED SHIPPING CONTAINERS presented byJAMES MICHAEL BROWN
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of the requirements for
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# CARBON DIOXIDE EVOLUTION AS A MEASURE OF DAMAGE TO FRESH APPLES HANDLED IN CORRUGATED SHIPPING CONTAINERS 

by
James Michael Brown

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# ABSTRACT <br> CARBON DIOXIDE AS A MEASURE OF DAMAGE TO FRESH apples handled in corrugated shipping containers 

## by

James Michael Brown

An objective, non-destructive method for determining the level of damage to horticultural commodities would be valuable for evaluating the peformance of produce shipping containers. Previous research has indicated that mechanically damaged apples show enhanced $\mathrm{CO}_{2}$ output when compared to non-damaged fruit. ${ }^{1 /}$

Apples (Malus domestica Borkh., cv. 'Empire') were packaged in two types of corrugated shipping containers. Three forces (impact by dropping, compression and vibration) with all combinations of packing and forces were applied to the shipping containers. The apples were removed from the shipping containers and placed in air-tight plastic buckets where carbon dioxide evolution was measured $1,2,3,4$ and 5 hours post treatment. Later, a visible rating of mechanical injury was given to the apples.

Visible injury scores positively correlated with the $\mathrm{CO}_{2}$ evolution of mechanically damaged apples at $99 \%$ confidence limits. The type of 1) force(s) applied, 2) packaging, and 3) fruit position within a container caused significant differences in $\mathrm{CO}_{2}$ output and visible injury scores. A method was developed where the change in $\mathrm{CO}_{2}$ output of damaged apples can be measured for determining the protective characteristics of shipping containers.

[^0]
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## LIST OF SYMBOLS AND ABBREVIATIONS

| $\mathrm{CO}_{2}$ | carbon dioxide |
| :---: | :---: |
| cm | centimeter |
| cV | cultivar |
| $\mathrm{C}_{2} \mathrm{H}_{4}$ | ethylene |
| gm | gram |
| $g$ | gravity |
| hr | hour |
| hz | nertz |
| in. | inch |
| m | meter |
| $\mu \ell$ or $\mu \mathrm{L}$ | microliters |
| ml or mL | milliliter |
| N | Newton |
| $\mathrm{O}_{2}$ | oxygen |
| 1 bf | pounds of force |
| r.h. | relative humidity |
| * | significant at 5\% level of probability |
| ** | significant at $1 \%$ level of probability |

## INTRODUCTION

During distribution, fresh fruit and vegetables are susceptible to physical damage, causing major post-harvest losses at all levels of marketing. Produce packaged in shipping containers is likely to encounter various handling hazards, such as dropping, compressive loads and vibration inputs. Before the performance of a produce shipping container can be determined, individual fruit must be inspected and assigned a subjective rating of physical damage. An objective, nondestructive method for readily assessing damage would be useful for evaluating the effects of transit forces on fresh produce packaged in shipping containers.

Many horticultural crops respond to injury by changes in carbon dioxide output. Work by Klein (15) and unpublished results by Dewey and Parker ${ }^{1 /}$ have suggested that the increased carbon dioxide output of damaged apples, tomatoes and oranges is related to the level of visible bruising on these crops. A method by which this change in carbon dioxide production could be captured and measured after damage to fruits might provide an objective, rapid index of injury.

[^1]Therefore, the purpose of this thesis is fourfold: 1) to determine how the change in $\mathrm{CO}_{2}$ output of bruised apples correlates to a subjective rating of visible damage, 2) to examine the effects of various simulated transit inputs (impact by dropping, vibration, compression and combinations thereof) and fruit position within a container on $\mathrm{CO}_{2}$ output and visual injury scores of apples, 3) determine whether the change in carbon dioxide evolution of apples can be used to evaluate the protective characteristics of shipping containers, 4) to provide a method by which the change in $\mathrm{CO}_{2}$ output of apples after damage can be measured and utilized as an objective method for determining the protective characteristics of shipping containers.

## LITERATURE REVIEW

## PHYSIOLOGICAL RESPONSES OF INJURED FRUITS

Many studies have focused on the enhanced $\mathrm{CO}_{2}$ or ethylene production of crops after mechanical damage. Tomato (1, 15, 17,21,22), apple (22), banana (19), cantaloupe (20), cherry (26) and citrus $(6,14,31)$ show enhanced $\mathrm{CO}_{2}$ output following injury. Similarly, enhanced ethylene following injury has been observed in apple (5,22,27), avocado (33), cantaloupe (20), citrus (36) and tomato (17,22).

MacLeod et al. (17) reported higher levels of $\mathrm{CO}_{2}$ production within 24 hours for tomatoes after bruising by impact. Additionally, an increase in the number of drops correlated with higher levels of $\mathrm{CO}_{2}$ and ethylene production. Six days after damage, increased $\mathrm{CO}_{2}$ production could not be detected.

Nakamura and Ito $(21,22)$ found an increase in respiration rate of tomato fruit after vibration. The increase in respiration was observed after vibrating fruit at 1 and $2 g$ at specified durations from 30 minutes to 5 hours. When vibration times were short, there was a proportional increase in $\mathrm{CO}_{2}$ production with respect to the acceleration level.

Other studies have shown that harder surfaces, higher compressive loads and higher vibration accelerations increased the production of $\mathrm{CO}_{2}$ in citrus (6). Vines et al. (36) reported higher respiration rates and
ethylene levels in grapefruit after fruit was dropped from a height of 4 and 6 feet onto a hard surface. The grapefruit returned to normal respiration rates with time. They concluded that $\mathrm{C}_{2} \mathrm{H}_{4}$ after damage was a stress symptom and not a normal metabolic product in citrus.

Information regarding the biochemical changes that take place in damaged fruit is limited. Many studies refer to the increase in $\mathrm{CO}_{2}$ production of bruised fruit as enhanced respiratory activity. Pollack and Hills (27) observed that following bruising of red tart cherries, "the increase in carbon dioxide output greatly exceeded the increase in oxygen utilization". Oxygen consumption increased $50 \%$, while $\mathrm{CO}_{2}$ output increased 126\%. Hyodo et al. (14) observed almost twice the $\mathrm{CO}_{2}$ output as compared to $\mathrm{O}_{2}$ uptake during the first 5 hours post-treatment after Satsuma mandarin fruit were dropped. Later $\mathrm{O}_{2}$ uptake and $\mathrm{CO}_{2}$ evolution were similar. Robitaille and Janick (28) suggested that an increase in $\mathrm{CO}_{2}$ production after bruising of apples was not the result of increased respiration.

Klein (15) demonstrated that excess $\mathrm{CO}_{2}$ after dropping apple fruit "came exclusively from the bruised tissue." He found cortical tissues .5 cm below the epidermis produced $77 \mu \ell / \mathrm{g} \cdot \mathrm{hr}$ at the bruise site and $35 \mu \ell / g \cdot h r-1$ at the control site on the fruit. Additionally, excised bruised tissues displayed a greater response to damage than whole fruit. Klein concluded that "the increase in $\mathrm{CO}_{2}$ evolution from apples after bruising is not due to enhanced aerobic or anaerobic respiratory activity, but rather due to decarboxylation of malic acid in cortical tissues at the bruise site," but gave litle evidence for this statement.

Whatever the cause of increased $\mathrm{CO}_{2}$ evolution of damaged fruit is, the preceding studies have shown that enhanced $\mathrm{CO}_{2}$ output occurred from
damaged fruits when compared to nondamaged fruits, and increasing the level of damage by increasing the impact force, number of impacts, and the duration or magnitude of vibration resulted in a proportionally increased $\mathrm{CO}_{2}$ evolution in these fruits.

## SIMULATED TRANSIT TESTING OF SHIPPING CONTAINERS

A simulated transportation environment can be designed in one of two ways: 1) to record transit vibrations and reproduce them with a servo hydraulic vibrator, or 2) to reproduce the damage observed in the transportation environment by trial and error with arbitrarily chosen impacts and vibrations $(8,10)$. A knowledge of the transportation environment is necessary to develop meaningful tests; however, no one simulated transit test can be used to determine the performance of a container $(12,13,37)$. The design of tests to simulate the transportation environment and to subsequently reproduce the damage in the laboratory basically consists of three types of forces: impact by dropping, vibration and compression.

## IMPACT BY DROPPING

Ostrem and Godshall (25) have indicated that impact damage caused by dropping is affected by the size, weight, contents and shape of the container. During distribution, container bottoms will receive 70\% of all drops; the remaining $30 \%$ of drops will occur on container sides, edges or tops (25). Edge and corner drops occur from greater heights than flat drops. Most containers will be dropped at low levels numerous times, and will experience few drops from high levels. There is a direct correlation between drop height, weight and the size of a
container; the heavier and larger the package, the lower the drop height. Palletized loads experience lower and fewer drops than nonpalletized loads.

Many studies have focused on predicting the level of injury to crops within packages with mathematical expressions after impact ( $12,31,34,35$ ). These mathematical expressions are capable of predicting the level of damaged fruit based on drop height, number of drops, fruit variety and package type. However, in addition to drops, produce containers experience compressive loads and vibration inputs; therefore, these equations fall short of predicting how a particular package system would provide protection from all the hazards experienced in a transportation environment.

Guillou et al. (10) recommends using 50 two-inch flat drops in the laboratory to simulate impact damage to produce containers. Schoorl (30) dropped apple packs from heights of $6^{\prime \prime}$ and $12^{\prime \prime}(1,3,9$ and 27 times), 18" (1, 3 and 9 times), 24" ( 1 and 3 times) and 48" (1 time). American Society for Testing Materials D-4169, Performance Testing of Shipping Containers and Systems (3), suggests 4 drops on each base edge of the container from a height of $3^{\prime \prime}, 6^{\prime \prime}$ or $9^{\prime \prime}$ (depending on the assurance level chosen) to simulate damage to a palletized truckload.

## VIBRATION

Frequencies encountered in the transportation are primarily below 25 Hertz, with 3-15 Hertz being most prevalent (7,9). Vibration inputs are usually from .2 to .8 g at 3 to 10 Hertz for rail transportation and from . 1 to .8 g at 3 to 20 Hertz for trucks (7). In a study of the
causes of fruit damage on transport trucks, it was shown that frequencies can range from 3 to 20 Hertz with accelerations less than .1 g to slightly higher than $1 \mathrm{~g}(24)$.

According to $0^{\prime}$ Brien (23), the vibration of fruit within containers depends on: 1) the depth of the container, 2) the tightness of fill, 3) type of suspension system used in the truck, 4) the magnitude of force exerted to the truck from the roadbed, 5) the vibrating characteristics of the fruit species. Fruit in the upper layers of containers experience greater injury $(9,24)$ since this fruit receives higher levels of acceleration during distribution.

Guillou reported that a test of 12 Hertz at 1 g for 30 minutes satisfactorily reproduced damage in the laboratory to produce in shipping containers (10). ASTM D-4169 (3) recommends testing packages from 5-15 minutes (depending on assurance level chosen) at the resonant frequency of the package system at .5 g to simulate truck transport and .25 g to simulate railroad transport at each possible shipping position of the container, up to four positions.

## COMPRESSION/LOAD FORCES

Fruits within containers are subject to compression bruises when a corrugated shipping container collapses and the fruit is required to carry the weight from other containers (29). Shipping containers are usually tested empty under standard conditions of $23^{\circ} \mathrm{C}$ and $50 \%$ R.H. at deformation rate of $1 / 2^{\prime \prime}$ per minute (2). Ultimately, the load a container can support in the distribution environment is dependent on several factors: moisture content of the board, the way the load is applied, length and rate at which the load is applied, previous handling
of the package (29), length of time in storage, vibration during transport and stacking pattern (2). Additionally, the failure of containers in storage is primarily related to the creep characteristics of the material (8).

The reaction of a stacked load to vibration can produce forces to lower container's useful strength more than the dead-load weight of a stack. Therefore, Godshall (7) recommends utilizing a single container to represent the bottom container with a dead-load mass on top to represent the other containers in a stack. Godshall simulated vertical dynamic loading with dead loads from 10 to $70 \%$ of the compressive strength of the container and applied acceleration inputs from . 2 to .8 g at increments of .1 g with frequencies of $4,6,8$ and 10 Hertz for 1-1/2 hours. With these vibration inputs, it was determined that containers could be loaded to $70 \%$ of their compressive strength and survive the effects of vertical dynamic loading experienced during distribution.

ASTM D-4169 (3) recommends loading containers utilizing the following equation to simulate loads during transport and storage.

$$
L=W \frac{(H-h)}{(h)} \times F
$$

where:
$L=$ minimum required load, lbf or $N$
$W=$ weight of one shipping unit or individual container, lbf or $N$
$h=$ height of shipping unit or individual container, in or $m$
$F=a$ factor to account for the combined effect of the individual factors described above

Balodis (4) suggested the following schedule for compression testing produce containers to simulate the storage environment: two weeks in a cool store with each pack under a 400-1b. load; two days in a conditioned room under 300 lb .; five days in a cool store under a 300-lb. load; and one day in a conditioned room under a 200-lb. load. The conditioned rooms could be set at $20^{\circ} \mathrm{C}$ and $65 \% \mathrm{RH}$ for moderate testing or $30^{\circ} \mathrm{C}$ and $85 \% \mathrm{RH}$ to simulate tropical markets.

## CARBON DIOXIDE RESPONSE OF APPLES DAMAGED IN CORRUGATED SHIPPING CONTAINERS

## MATERIALS AND METHODS

Apples (Malus domestica Borkh., cv. 'Empire') without apparent bruises and defects and relatively uniform in size (2 3/4" in diameter) were taken out of controlled atmosphere storage from the upper layers of fruit orchard bins during April of 1984. The apples were placed in foam trays and corrugated shipping containers and stored under refrigeration at $0^{\circ} \mathrm{C}$ for later use.

Twenty-four hours before conducting each block, fruits were removed from cold storage and repacked into the test containers. This allowed sufficient time for the apples to equilibrate to test temperature and for the possible handling effect of increased $\mathrm{CO}_{2}$ output to subside before testing.

Apples in one-bushel boxes having internal dimensions of $193 / 4$ " x 11 1/2" x $12^{\prime \prime}$ were subjected to various drop, vibration and compressive inputs. After treatment, entire apple boxes were placed in airtight containers having internal dimensions of $201 / 4^{\prime \prime} \times 135 / 8^{\prime \prime} \times 123 / 4$ ". The airtight containers had sampling ports on diagonally opposite sides where gas measurements were taken hourly. There was no significant difference between damaged and nondamaged fruit $\mathrm{CO}_{2}$ responses. It was hypothesized that the $\mathrm{CO}_{2}$ response of the damaged fruit was not detected
because the $\mathrm{CO}_{2}$ within the airtight containers was diluted since all the fruit within the corrugated boxes was not damaged. Therefore, an alternate method where individual sample fruits from damaged corrugated boxes were placed in small airtight buckets was used in this study, and is discussed below.

Before placing the sample fruit in the shipping containers to be tested, they were labeled individually with a permanent ink marker. A total of 60 fruits, 20 for the top, middle and lower layers of the test containers, was labeled (see Figure 1). There were 5 layers of fruit in each container, 125 apples per container. Carbon dioxide response and visible injury scores were taken only from fruit in the top, middle and bottom layers. Sixteen shipping containers was prepared for one replication, and three forces were applied to two types of containers: drop, compression and vibration, with all combinations of packages and forces applied. In total, four replications were run.

Two package types were tested, both of full telescope half-slotted design: 1) tray-pack having internal dimensions of $19-3 / 4$ " $x$ 11-1/2" $x$ 12", the top and bottom corrugated combined board were of 42/33/42 and 69/33/69 construction, respectively; and 2) one-bushel boxes having internal dimensions of $19-7 / 16^{\prime \prime} \times 11-5 / 8^{\prime \prime} \times 10-1 / 4$ ", the top and bottom corrugated combined board were of $42 / 26 / 42$ and $69 / 26 / 69$ construction, respectively. The numbers that describe the construction of the corrugated board refer to the weight of the liners and medium in pounds per 1000 square feet. Containers without fruit were allowed to condition at approximately $50 \%$ R.H. and $20^{\circ} \mathrm{C}$ for one week prior to being tested.

## Placement of Sample Fruit Within Shipping Containers



Bottom Layer


Middle Layer


Top Layer

Figure 1. Placement of sample fruit in shipping containers.

The compressive strength of the two types of containers was determined in accordance with ASTM D-642 (2), Compression Test For Shipping Containers. The pulp spring cushion trays used in the tray packs were capable of holding 25 fruit each, for a total of 125 fruit per container. The one-bushel boxes were hand pattern packed using a pulp tray in the bottom to aid in producing the pattern of successive layers of fruit by the K.P. system (26).

The lengthy period of time required to apply the treatments prevented all 16 treatment combinations and replications from being performed on the same day. Therefore, the treatments were split into two blocks (see Table 1).

Since the 16 treatment combinations were split into two blocks, the Drop $x$ Vibrate x Compression x Package (tray) interaction was confounded due to incomplete blocks by day. Four variables were studied: drop, vibrate, compression and package type each having two levels. With time all 16 treatment combinations $\left(2^{4}=16\right)$ were replicated 4 times. Additionally, each container was sampled at 6 positions within each container. Therefore, the experimental design was a split plot with main plot treatments represented by the $2^{4}$ factorial and subplot treatments by position.

Force treatments were applied first by dropping followed by vibration and then compressing. Drops were performed with an MTS (MTS Systems Co., Minneapolis, MN) shock machine which had a guided table and a 2 ms impact programmer to produce a repeatable shock which was transmitted to the package on the table. Dropping consisted of two 25 cm flat drops. To ensure reproducibility, containers were securely fastened to the table.

Table 1. Application of force treatments by incomplete blocks on K.P. and tray-pack shipping containers with cv. 'Empire' apples.

Time 1
Treatment Type of Force Package Type12 CompressNone
K.P.
Compress Tray
VibrateTray4Vibrate x CompressDropDrop x CompressDrop x VibrateDrop x Vibrate x CompressK
,
Drop $x$ Vibrate
Drop x Vibrate x Compress
Time 2
Treatment

| 9 | Drop | K.P. |
| ---: | :--- | :--- |
| 10 | Drop $\times$ Compress | Tray |
| 11 | Drop $\times$ Vibrate | Tray |
| 12 | Drop $\times$ Vibrate $\times$ Compress | K.P. |
| 13 | None | Tray |
| 14 | Compress | K.P. |
| 15 | Vibrate | K.P. |
| 16 | Vibrate $\times$ Compress | Tray |

Compression testing was performed with a Baldwin-Emery compression tester. The load was applied in accordance with ASTM D-642 (2) with a continuous motion of $.5+.1 \mathrm{in} . / \mathrm{min}$. to 1600 lbs .

Vibration testing was performed on a MTS servo hydraulic vibration table. Shipping containers were securely fastened to the table with two $3 / 8^{\prime \prime}$ steel rods which screwed into the table and a 1 " $x 1^{\prime \prime}$ piece of wood. Vibration testing consisted of brining the table up sinusoidally from 3 Hertz to 9.5 Hertz at a constant $g$ level of .8 g and maintaining the vibration level at .8 g and 9.5 Hz for 30 minutes. All simulated transit testing was performed at $50 \%$ R.H. and $23^{\circ} \mathrm{C}$.

After the treatments had been applied, fruit samples were removed from the packages and placed in plastic 6000 ml buckets. Ten fruit were placed in each bucket, equaling two buckets per layer and six buckets per corrugated container. The lids for the buckets were modified with two $1 / 4^{\prime \prime}$ holes where gas samples could be taken at $1,2,3,4$ and 5 hours post treatment. The tubs were closed and sealed with airtight lids in the same order and time sequence at which $\mathrm{CO}_{2}$ samples were obtained. The gas samples were taken with an ADC (Analytical Development Company) analyzer utilizing an infrared detector with a built in recirculating pump and digital percentage readout. Gas samples were taken by inserting the two needles of the analyzer through the lid holes covered with gas-proof tape. Fifteen to 20 seconds were required to obtain a stable $\mathrm{CO}_{2}$ reading. After each reading, a fresh piece of tape was immediately placed over the holes to prevent the escape of the atmosphere from the bucket.

After the fifth-hour reading, lids were removed from each bucket and the fruit was weighed. The apples were then held 4 days at $20^{\circ} \mathrm{C}$ and
rated for visible damage. A rating system of from 1 to 5 was utilized: $1=$ no damage; $2=$ slight but noticeable; $3=$ moderate, affects marketing; $4=$ severe, reduces value; 5 = very severe, unmarketable.

The microliters of $\mathrm{CO}_{2}$ evolved per gram of fruit per hour were calculated as follows:

$$
\mathrm{CO}_{2}(\mu \ell / \mathrm{gm} \cdot \mathrm{hr})=\frac{\left(\mathrm{CO}_{2}\right)(.01)(\Delta \text { volume })}{\text { (fruit weight })(\text { time })}
$$

| $\mathrm{CO}_{2}=$ | reading from analyzer |
| :--- | :--- |
| $\Delta$ volume $=$ | gross volume of containers (me) - fruit weight (grams) |
| time $=$ | length of time the tubs were sealed (hourly reading) |

## RESULTS

## RELATIONSHIP BETWEEN $\mathrm{CO}_{2} \quad \mu \ell / \mathrm{gm} \cdot \mathrm{hr}$ AND INJURY SCORES OF 'EMPIRE' APPLES

The highly significant positive correlation of $\mathrm{CO}_{2}$ evolution of the apples at the fourth hour following treatment to the subsequently measured injury scores is shown in Figure 2. These 4 th-hour $\mathrm{CO}_{2}$ readings yielded the highest $r$-square value to the injury scores and the 1st-hour readings the lowest (Table 2). Therefore, only the 4 th-hour $\mathrm{CO}_{2}$ readings are presented in the results and discussion sections of the text. The $\mathrm{CO}_{2}$ reading for all other hours (1, 2, 3 and 5) are tabulated in the appendix.

Each variable tested (drop, compress, vibrate, package type and position) correlated at $99 \%$ confidence limits with the visual injury scores of the apples. However, r-square values varied from a high of .6039 for Position $F$ to a low of .2801 for dropping (Table 3); the overall r-square value for the 4 th-hour reading was . 4568.

## SPLIT-PLOT ANALYSIS OF VARIANCE

The data were analyzed two ways: 1) as a split plot by factorial effects where all 6 variables were included (drop, vibrate, compress, package type, position and replication (blocks) - see Tables 4 and 5), and 2) as a split plot by treatment combination where 16 treatments ( 2 drop $\times 2$ vibrate $\times 2$ compress $\times 2$ package type $=16$ ) with the $\mathrm{CO}_{2}$ output and injury scores for the 6 positions within each container grouped together as one mean.

Table 2. Relationship between $\mathrm{CO}_{2}$ production and injury score rating of 'Empire' apples at $1,2,3,4$ and 5 hours post treatment

| Hour | r-square | F |
| :---: | :---: | :---: |
|  |  |  |
| 1 | .3335 | $191.18^{* *}$ |
| 2 | .4521 | $315.23^{* *}$ |
| 3 | .4472 | $309.02 * *$ |
| 4 | .4568 | $321.22^{* *}$ |
| 5 | .4528 | $316.06 * *$ |

Table 3. Relationship between $\mathrm{CO}_{2}$ production, treatment and injury score rating of 'Empire' apples 4 hours post treatment

| Treatment | r-square |  |
| :--- | :---: | :---: |
|  |  |  |
| Drop | .2801 | $73.93^{* *}$ |
| Vibrate | .4648 | $164.98^{* *}$ |
| Compress | .3957 | $124.39^{* *}$ |
| Package Type | .5440 | $226.66^{* *}$ |
| Position | - |  |
| A | .5611 | $79.28 * *$ |
| B | .4099 | $43.07 * *$ |
| C | .3735 | $36.96 * *$ |
| D | .3448 | $32.63^{* *}$ |
| E | .5903 | $89.32^{* *}$ |
| F | .6039 | $94.54 * *$ |



Figure 2. The relationship between $\mathrm{CO}_{2}$ evolution and injury scores of 'Empire' apples at 4 hours post treatment.

Table 4. Split plot by factorial effects for $\mathrm{CO}_{2} \mu \ell / \mathrm{gm} \cdot \mathrm{hr}$ - hour 4 of cv. 'Empire' apples damaged in corrugated shipping containers

| Source | DF | Anova SS | F Value | PR>F |
| :---: | :---: | :---: | :---: | :---: |
| BLOCK | 3 | 27.21 | 2.19 | . 1019 |
| TREATMENT | 15 | 390.97 | 6.30 | . 0001 ** |
| DROP | 1 | 102.61 | 24.81 | .0001** |
| VIBRATE | 1 | 151.28 | 36.58 | .0001** |
| COMPRESS | 1 | 10.25 | 2.48 | . 1224 |
| PACKAGE | 1 | 2.22 | 0.54 | . 4672 |
| DROPxVIBRATE | 1 | 4.17 | 1.01 | . 3208 |
| DROPxCOMPRESS | 1 | 8.40 | 2.03 | . 1609 |
| DROPxPACKAGE | 1 | 0.49 | 0.12 | . 7322 |
| VIBRATExCOMPRESS | 1 | 4.49 | 1.09 | . 3030 |
| VIBRATExPACKAGE | 1 | 36.90 | 8.92 | . $0045^{* *}$ |
| COMPRESSxPACKAGE | 1 | 13.94 | 3.37 | . 0730 |
| DROPxVIBRATExCOMPRESS | 1 | 23.00 | 5.56 | .0228* |
| DROPxVIBRATExPACKAGE | 1 | 0.72 | 0.17 | . 6786 |
| DROPxCOMPRESSxPACKAGE | 1 | 22.34 | 5.40 | .0247* |
| VIBRATExCOMPRESSxPACKAGE | 1 | 4.36 | 1.05 | . 3102 |
| DROPxVIBRATExCOMPRESSxPACKAGE | 1 | 5.80 | 1.40 | . 2424 |
| BLOCKxTREATMENT (Error 1) | 45 | 186.08 | 4.43 | .0001** |
| POSITION | 5 | 55.00 | 11.77 | .0001** |
| DROPxPOSITION | 5 | 53.04 | 11.35 | .0001** |
| VIBRATExPOSITION | 5 | 112.87 | 24.16 | .0001** |
| COMPRESSxPOSITION | 5 | 6.37 | 1.36 | . 2401 |
| PACKAGExPOSITION | 5 | 78.74 | 16.85 | .0001** |
| DROPxVIBRATExPOSITION | 5 | 7.73 | 1.65 | . 1475 |
| DROPxCOMPxPOSITION | 5 | 4.81 | 1.03 | . 4005 |
| DROPxPACKAGExPOSITION | 5 | 30.19 | 6.46 | .0001** |
| VIBRATExPACKAGExPOSITION | 5 | 4.30 | 0.92 | . 4686 |
| VIBRATExPACKAGExPOSITION | 5 | 79.66 | 17.05 | .0001** |
| COMPRESSXPACKAGExPOSITION | 5 | 8.81 | 1.89 | . 0967 |
| DROPxVIBRATExCOMPRESSxPOSITION | 5 | 3.76 | 0.80 | . 5506 |
| DROPxVIBRATExPACKAGExPOSITION | 5 | 0.50 | 0.11 | . 9900 |
| DROPxCOMPRESSxPACKAGExPOSITION | 5 | 4.72 | 1.01 | . 4124 |
| VIBRATExCOMPRESSxPACKAGExPOSITION | 5 | 8.25 | 1.77 | . 1197 |
| DROPxVIBRATExCOMPRxPACKxPOSITION | 5 | 5.68 | 1.22 | . 3003 |
| (Error 2) | 240 | 224.24 |  |  |
| TOTAL | 383 |  |  |  |

Table 5. Split plot by factorial effects for injury scores of cv . 'Empire' apples damaged in corrugated shipping containers

| Source | DF | Anova SS | F Value | PR>F |
| :---: | :---: | :---: | :---: | :---: |
| BLOCK | 3 | 1.17 | 1.43 | . 2456 |
| TREATMENT | 15 | 276.34 | 151.63 | . $0001^{* *}$ |
| DROP | 1 | 161.20 | 591.23 | .0001** |
| VIBRATE | 1 | 44.15 | 161.91 | .0001** |
| COMPRESS | 1 | 10.21 | 37.43 | .0001** |
| PACKAGE | 1 | 17.68 | 64.85 | .0001** |
| DROPxVIBRATE | 1 | 1.58 | 5.78 | .0204* |
| DROPxCOMPRESS | 1 | 4.46 | 16.37 | .0002** |
| DROPxPACKAGE | 1 | 7.26 | 26.63 | .0001** |
| VIBRATExCOMPRESS | 1 | 0.92 | 3.38 | . 0728 |
| VIBRATEXPACKAGE | 1 | 14.81 | 54.30 | .0001** |
| COMPRESSxPACKAGE | 1 | 8.46 | 31.03 | .0001** |
| DROPxVIBRATExCOMPRESS | 1 | 1.04 | 3.82 | . 0569 |
| DROPxVIBRATExPACKAGE | 1 | 0.08 | 0.28 | . 6003 |
| DROPxCOMPRESSxPACKAGE | 1 | 4.13 | 15.13 | .0003** |
| VIBRATExCOMPRESSxPACKAGE | 1 | 0.09 | 0.34 | . 5605 |
| DROPxVIBRATExCOMPRESSxPACKAGE | 1 | 0.28 | 1.03 | . 3149 |
| BLOCKxTREATMENT (Error 1) | 45 | 12.27 | 2.24 | .0001** |
| POSITION | 5 | 2.07 | 3.40 | .0056** |
| DROPxPOSITION | 5 | 42.33 | 69.68 | .0001** |
| VIBRATExPOSITION | 5 | 33.49 | 55.12 | .0001** |
| COMPRESSXPOSITION | 5 | 1.52 | 2.50 | .0314* |
| PACKAGExPOSITION | 5 | 18.29 | 30.11 | .0001** |
| DROPxVIBRATExPOSITION | 5 | 0.72 | 1.19 | . 3147 |
| DROPxCOMPxPOSITION | 5 | 1.40 | 2.30 | . 0457 |
| DROPxPACKAGExPOSITION | 5 | 0.64 | 1.05 | . 3890 |
| VIBRATExPACKAGExPOSITION | 5 | 1.61 | 2.65 | .0236* |
| VIBRATExPACKAGExPOSITION | 5 | 23.94 | 39.41 | .0001** |
| COMPRESSXPACKAGExPOSITION | 5 | 2.12 | 3.49 | . 0046 |
| DROPxVIBRATExCOMPRESSXPOSITION | 5 | 0.93 | 1.53 | . 1811 |
| DROPxVIBRATExPACKAGExPOSITION | 5 | 0.46 | 0.76 | . 5794 |
| DROPxCOMPRESSxPACKAGExPOSITION | 5 | 0.37 | 0.61 | . 6923 |
| VIBRATExCOMPRESSxPACKAGExPOSITION | 5 | 0.31 | 0.51 | . 7686 |
| DROPxVIBRATExCOMPRxPACKxPOSITION | 5 | 0.14 | 0.23 | . 9492 |
| (Error 2) | 240 | 29.16 |  |  |
| TOTAL | 383 | , ' |  |  |

Split Plot by Factorial Effects - Main Effects
The variables drop, vibrate and position had a statistically significant effect on $\mathrm{CO}_{2}$ evolution, Table 6. Vibration had the greatest effect on $\mathrm{CO}_{2} \mu \ell / \mathrm{gm} \cdot \mathrm{hr}$, followed by position, dropping, compression and package type, respectively. Dropping had the greatest effect on injury scores, followed by vibration, package type, compression, and position, respectively.

Dropping
Dropping containers significantly enhanced $\mathrm{CO}_{2}$ evolution by $1.03 \mu \ell / \mathrm{gm} \cdot \mathrm{hr}$ and injury scores by 1.30 units compared to not dropping containers. The significant increase in $\mathrm{CO}_{2}$ evolution for dropped fruit amounted to $9 \%$ over that of fruit which was not dropped but otherwise exposed to all other treatments; the increase in visible damage was $61 \%$.

## Vibrating

Vibrating containers significantly enhanced $\mathrm{CO}_{2}$ evolution by $1.26 \mu \ell / \mathrm{gm} \cdot \mathrm{hr}$ and injury scores by .68 unit compared to not vibrating containers. The significant increase in $\mathrm{CO}_{2}$ evolution for vibrated fruit amounted to $10 \%$ over that of fruit which was not vibrated but otherwise exposed to all other treatments; the increase in visible damage was $28 \%$.

Table 6. Comparison of treatment main effects on $\mathrm{CO}_{2}$ production and injury scores for cv. 'Empire' apples damaged in corrugated shipping containers

| Treatment <br> (Variable) |  | N | $\mathrm{CO}_{2}$ Evolution $\mu \mathrm{l} / \mathrm{gm} \cdot \mathrm{hr}$ |  | Injury Score Rating |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  |  | Mean | Duncan Grouping | Mean | Duncan Grouping |
| Dropping | (+) | 192 | 13.17 | A | 3.44 | A |
|  | (-) | 192 | 12.13 | B | 2.14 | B |
| Vibrating | (+) | 192 | 13.28 | A | 3.13 | A |
|  | (-) | 192 | 12.02 | B | 2.45 | B |
| Compressing | (+) | 192 | 12.81 | A | 2.95 | A |
|  | (-) | 192 | 12.49 | A | 2.62 | B |
| Package Type | (tray) | 192 | 12.73 | A | 3.00 | A |
|  | (K.P.) | 192 | 12.57 | A | 2.57 | B |
| Position: |  |  |  |  |  |  |
| Top Layer | a | 64 | 12.42 | C | 2.79 | ABC |
|  | b | 64 | 12.51 | C | 2.85 | A |
| Middle Layer | c | 64 | 12.29 | C | 2.71 | BC |
|  | d | 64 | 12.37 | C | 2.81 | AB |
| Bottom Layer | e | 64 | 12.95 | B | 2.68 | C |
|  | f | 64 | 13.35 | A | 2.88 | A |

Means with the same letter are not significantly different at $\alpha=.05$.

## Compressing

Compressed containers did not significantly enhance $\mathrm{CO}_{2}$ evolution of the apples; however, injury scores were significantly increased . 33 unit compared to non-compressed containers. The increase in $\mathrm{CO}_{2}$ evolution for compressed fruit amounted to $3 \%$ over that fruit which was not compressed but otherwise exposed to all other treatments; the increase in visible damage was $13 \%$.

## Package Type

$\mathrm{CO}_{2}$ evolution was higher for apples in the K.P. container; however, there was no significant difference between the two types of containers tested. Injury scores were significantly higher by .43 unit for apples in the K.P. container.

## Position

The position of apples in a package had a significant effect on their $\mathrm{CO}_{2}$ output and injury scores. Positions E and F in the top layer yielded significantly higher $\mathrm{CO}_{2}$ values than one another and positions A, B, C and D, while positions A, B, C and D were not significantly different from one another with respect to $\mathrm{CO}_{2}$ output. Positions A, B, D and F did not produce significantly different injury scores than each other, but damage to apples in these positions was significantly higher than in positions C (middle layer) and E (top layer).

## Split Plot by Factorial Effect - Significant Two-way Interactions

The highly statistically significant interactions of Drop $x$ Position, Vibrate x Package, Vibrate x Position, and Package x Position on $\mathrm{CO}_{2} \mu \ell / \mathrm{gm} \cdot \mathrm{hr}$ and injury scores are presented in Figures 3 through 10. Additionally, Drop x Vibrate, Drop x Compress, Drop x Package, Compress x Package and Compress x Position were significant treatment interactions for injury scores but not $\mathrm{CO}_{2}$ evolution.

## Drop x Position (Figures 3 and 4 and Table 7)

When containers were not dropped, $\mathrm{CO}_{2}$ evolution was the lowest for positions A and B (bottom layer) followed by positions C, D, E and $F$, see Figure 3. Injury scores for non-dropped containers followed a similar trend, see Figure 4 and Table 7 . When containers were dropped, position D (middle layer) had the lowest $\mathrm{CO}_{2}$ output, followed by positions C, E, A, B and F - fruit in the middle of the container had the lowest $\mathrm{CO}_{2}$ output.

Apples in positions $A, B, C$ and $D$ when dropped had a higher $\mathrm{CO}_{2}$ output when compared to non-dropped apples. Carbon dioxide evolution for positions $E$ and $F$, both located in the upper layer of the containers, were least affected by dropping. Injury scores for fruit in dropped containers were the highest for apples in the bottom of containers, followed by those in the middle and top, respectively. Injury scores for all of the positions were significantly affected by dropping except positions $E$ and $F$ (top layer). Therefore, when containers were dropped, apples located near the bottom of the containers were injured most, and apples located near the top, the least.

## DROP x POSITION

Carbon Dioxide ( $\mu \mathrm{L} / \mathrm{gm} \cdot \mathrm{hr}$ )


Figure 3. Interaction effect between dropping and position on $\mathrm{CO}_{2}$ $\mu \ell / g m \cdot h r$ evolved from 'Empire' apples measured 4 hours post treatment.


Figure 4. Interaction effect between dropping and position on injury score of 'Empire' apples.

Table 7 Treatment comparisons within the drop x position treatment interaction on $\mathrm{CO}_{2}$ production and injury scores for cv . 'Empire' apples damaged in corrugated shipping containers.

| Treatment Comparison | Mean Diff. $\mathrm{CO}_{2} \mu \ell /$ $\mathrm{gm} \cdot \mathrm{hr}$ | F | Mean Diff. In jury Score | F |
| :---: | :---: | :---: | :---: | :---: |
| ( $\mathrm{d}^{-}$, position ${ }^{\text {A }}$ ) vs $\left(\mathrm{d}^{+}\right.$, position ${ }^{\text {A }}$ ) | 1.98 | 3.89** | 1.82 | 13.94** |
| ( $\mathrm{d}^{-}$, position ${ }^{\text {B }}$ ) vs ( $\mathrm{d}^{+}$, position ${ }^{\text {B }}$ ) | 1.80 | 3.54** | 1.80 | 13.79** |
| ( $\mathrm{d}^{-}$, position ${ }^{\text {C }}$ ) vs ( $\mathrm{d}^{+}$, position ${ }^{\text {C }}$ ) | 1.29 | 2.54** | 1.53 | $11.72^{* *}$ |
| ( $\mathrm{d}^{-}$, position ${ }^{\text {D }}$ ) vs ( $\mathrm{d}^{+}$, position ${ }^{\text {D }}$ ) | . 92 | 1.81** | 1.41 | 10.80** |
| ( $\mathrm{d}^{-}$, position ${ }^{\text {E }}$ ) vs ( $\mathrm{d}^{+}$, position ${ }^{\mathrm{E}}$ ) | . 05 | 0.10 | . 54 | 4.14 |
| $\left(d^{-}\right.$, position ${ }^{\text {F }}$ ) vs ( $d^{+}$, position ${ }^{\text {F }}$ ) | . 15 | . 30 | . 31 | 2.37 |

Vibrate x Package Type (Figures 5 and 6 and Table 8)
Vibration increased $\mathrm{CO}_{2}$ output in the tray containers by 1.87 $\mu \ell / \mathrm{gm} \cdot \mathrm{hr}$ ( $16 \%$ ) and injury scores by 1.07 units ( $52 \%$ ). Vibrating K.P. containers increased $\mathrm{CO}_{2}$ output by $.63 \mu \mathrm{l} / \mathrm{gm} \cdot \mathrm{hr}(5 \%)$ and injury scores by . 28 unit ( $10 \%$ ). When containers were not vibrated, the K.P. container had a higher $\mathrm{CO}_{2}$ and injury score reading; however, when containers were vibrated, the tray-pack container had a higher $\mathrm{CO}_{2}$ output and a similar injury score reading when compared to the K.P. container. Therefore, the effect of vibrating was greater for tray-pack containers than for K.P. containers.

## Vibrate x Position (Figures 7 and 8 and Table 9)

Vibration resulted in the highest $\mathrm{CO}_{2}$ output and injury scores in the top layer of the shipping containers, positions $E$ and $F$. Vibration caused no significant effect on $\mathrm{CO}_{2}$ output or injury scores for fruit in positions A and B, located in the bottom layer of the containers but did significantly increase $\mathrm{CO}_{2}$ evolution for apples located in the top (positions $E$ and F). Non-vibrated containers displayed higher $\mathrm{CO}_{2}$ output and injury scores for fruit located toward the bottom and middle of the containers.

Package Type x Position (Figures 9 and 10 and Table 10)
Positions E and F (top layer) in the tray-pack container had the highest $\mathrm{CO}_{2}$ output, while positions A and B (bottom layer) had the highest $\mathrm{CO}_{2}$ output for the K.P. container.

## VIBRATE x PACKAGE



Figure 5. Interaction effect between vibrating and package type on $\mathrm{CO}_{2}$ $\mu \ell / \mathrm{gm} \cdot \mathrm{hr}$ evolved from 'Empire' apples measured 4 hours post treatment.

## VIBRATE x PACKAGE



Vibrate
Figure 6. Interaction effect between vibrating and package type on injury score of 'Empire' apples.

Table 8. Treatment comparisons within the vibrate x package type treatment interaction on $\mathrm{CO}_{2}$ production and injury scores for cv. 'Empire' apples damaged in corrugated shipping containers.

| Treatment Comparison | $\begin{gathered} \text { Mean } \\ \text { Diff. } \\ \mathrm{CO}_{2} \mu \ell / \\ \mathrm{gm} \cdot \mathrm{hr} \\ \hline \end{gathered}$ | F | Mean Diff. Injury Score | F |
| :---: | :---: | :---: | :---: | :---: |
| ( $\mathrm{v}^{-}, \mathrm{p}^{\text {tray }}$ ) vs ( $\mathrm{v}^{+}, \mathrm{p}^{\text {tray }}$ ) | 1.87 | 6.37** | 1.07 | 14.20** |
| ( $\mathrm{v}^{-}, \mathrm{p}^{\text {tray }}$ ) vs ( $\mathrm{v}^{+}, \mathrm{p}^{\mathrm{K} . \mathrm{P} \cdot}$ ) | 1.4 | 4.77* | 1.10 | 14.60** |
| ( $\mathrm{v}^{-}, \mathrm{p}_{\mathrm{K}}^{\text {tray }}$ ) vs ( $\mathrm{v}^{-}, \mathrm{p}^{\mathrm{K} . \mathrm{P} .}$ ) | 0.77 | 2.62 | 0.82 | 10.88** |
| ( $\mathrm{v}^{-}, \mathrm{p}_{\mathrm{K}}^{\mathrm{K}, \mathrm{P} .}$ ) vs ( $\mathrm{v}^{+}, \mathrm{p}_{\mathrm{K}}^{\text {tray }}$ ) | 1.10 | 3.75 | 0.25 | 3.32 |
|  | 0.63 | 2.15 | 0.28 | 3.72 |
| ( $\mathrm{v}^{+}, \mathrm{p}^{\text {tray }}$ ) vs ( $\mathrm{v}^{+}, \mathrm{p}^{\text {K.P. }}$ ) | 0.47 | 1.60 | 0.03 | . 40 |

VIBRATE x POSITION

Carbon Dioxide ( $\mu \mathrm{L} / \mathrm{gm} \cdot \mathrm{hr}$ )


Figure 7. Interaction effect between vibrating and position on $\mathrm{CO}_{2}$ $\mu \ell / \mathrm{gm} \cdot \mathrm{hr}$ evolved from 'Empire' apples measured 4 hours after treatment.

## VIBRATE x POSITION



Figure 8. Interaction effect between vibrating and position on injury score of 'Empire' apples.

Table 9. Treatment comparisons within the vibrate $x$ position treatment interaction on $\mathrm{CO}_{2}$ production and injury scores for cv . 'Empire' apples damaged in corrugated shipping containers.

| Treatment Comparison | Mean Diff. <br> $\mathrm{CO}_{2} \mu \mathrm{hl}$ <br> $\mathrm{gm} \cdot \mathrm{hr}$ | F | Mean Diff. Injury Score | F |
| :---: | :---: | :---: | :---: | :---: |
| ( $\mathrm{v}^{-}$, position ${ }^{\text {A }}$ ) vs ( $\mathrm{v}^{+}$, position ${ }^{\text {A }}$ ) | . 52 | 1.02 | . 12 | . 92 |
| ( $\mathrm{v}^{-}$, position ${ }^{\text {B }}$ ) vs ( $\mathrm{v}^{+}$, position ${ }^{\text {B }}$ ) | . 16 | . 31 | . 01 | . 08 |
| ( $\mathrm{v}^{-}$, position ${ }^{\text {C }}$ ) vs ( $\mathrm{v}^{+}$, position ${ }^{\text {C }}$ ) | . 61 | 1.20 | . 55 | 4.21* |
| ( $\mathrm{v}^{-}$, position ${ }^{\text {D }}$ ) vs ( $\mathrm{v}^{+}$, position ${ }^{\text {D }}$ ) | . 71 | 1.40 | . 44 | 3.37 |
| $\left(\mathrm{v}^{-}, \operatorname{position}{ }^{\mathrm{E}}\right)$ vs ( $\mathrm{v}^{+}$, position ${ }^{\mathrm{E}}$ ) | 2.62 | 5.15* | 1.48 | 11.34** |
| $\left(\mathrm{v}^{-}\right.$, position $\left.{ }^{\mathrm{F}}\right)$ vs ( $\mathrm{v}^{+}$, position ${ }^{\mathrm{F}}$ ) | 2.91 | 5.72* | 1.47 | 11.26** |

## PACKAGE x POSITION

Carbon Dioxide ( $\mu \mathrm{L} / \mathrm{gm} \cdot \mathrm{hr}$ )


Figure 9. Interaction effect between package type and position on $\mathrm{CO}_{2}$ $\mu \ell / \mathrm{gm} \cdot \mathrm{hr}$ evolved from 'Empire' apples measured 4 hours after treatment.

## PACKAGE x POSITION

Injury Score


Figure 10. Interaction effect between package type and position on injury score of 'Empire' apples.

Table 10 Treatment comparisons within the package type $x$ position treatment interaction on $\mathrm{CO}_{2}$ production and injury scores for cv. 'Empire' apples damaged in corrugated shipping containers.

| Treatment Comparison | $\begin{aligned} & \text { Mean } \\ & \text { Diff. } \\ & \mathrm{CO}_{2} \cdot \mu \mathrm{l} / \\ & \mathrm{gm} \cdot \mathrm{hr} \end{aligned}$ | F | Mean Diff. In jury Score | F |
| :---: | :---: | :---: | :---: | :---: |
|  | 0.99 | 1.95 | 0.48 | 3.68 |
|  | 0.78 | 1.53 | 0.72 | 5.52* |
|  | 0.35 | 0.69 | 0.63 | 4.83* |
|  | 1.0 | 1.97 | 1.03 | 7.89** |
|  | 1.1 | 2.16 | . 12 | . 92 |
| ( $\mathrm{K}^{\mathrm{K} \cdot \mathrm{P} \cdot}$, pos. ${ }^{\text {F }}$ ) vs ( $\mathrm{tray}^{\text {tray }}$, pos. ${ }^{\text {F }}$ ) | 1.08 | 2.12 | . 17 | 1.30 |

Split Plot by Factorial Effects - Significant Three-way Interactions
The Drop x Compress x Package and the Vibrate x Package x Position three-way interactions were significant for $\mathrm{CO}_{2}$ evolution and injury scores, while the Drop $x$ Vibrate $x$ Compress and the Drop $x$ Package $x$ Position three-way interactions were significant for $\mathrm{CO}_{2}$ evolution but not for injury scores; and the Vibrate $x$ Compress $x$ Position and the Compress $x$ Package $x$ Position were signficant for injury scores but not for $\mathrm{CO}_{2}$ evolution.

Drop x Vibrate x Compress (Figures 11-12)
The effect of vibrating on fruit $\mathrm{CO}_{2}$ output when containers were not dropped or compressed was to increase $\mathrm{CO}_{2}$ by $2.17 \mu \mathrm{l} / \mathrm{gm} \cdot \mathrm{hr}$ (20\%) and to increase injury scores 1.01 units (74\%). The effect of vibrating containers that were compressed and dropped was to increase $\mathrm{CO}_{2} 1.32$ (11\%) $\mu \mathrm{l} / \mathrm{gm} \cdot \mathrm{hr}$ and injury scores . 56 unit ( $17 \%$ ). The effect of dropping on $\mathrm{CO}_{2}$ output when containers were not vibrated or compressed was to increase $\mathrm{CO}_{2}$ by $2.02 \mu \mathrm{l} / \mathrm{gm} \cdot \mathrm{hr}$ (19\%) and injury scores by 1.75 units (129\%). The effect of dropping when containers were vibrated and compressed was to increase $\mathrm{CO}_{2} 1.02$ $\mu \ell / \mathrm{gm} \cdot \mathrm{hr}$ ( $8 \%$ ) and injury scores by 1.06 units (39\%). The effect of compressing when containers were not dropped or vibrated was to increase $\mathrm{CO}_{2} 1.33 \mu \mathrm{l} / \mathrm{gm} \cdot \mathrm{hr}$ (12\%) and injury scores . 75 unit (55\%). The effect of compressing when containers were dropped and vibrated was to increase $\mathrm{CO}_{2}$ by $.30 \mu \ell / \mathrm{gm} \cdot \mathrm{hr}$ (2\%) and injury scores . 12 unit (3\%). Containers dropped, not vibrated and not compressed had .24 higher $\mathrm{CO}_{2} \mu \mathrm{l} / \mathrm{gm} \cdot \mathrm{hr}(2 \%)$ than containers dropped, not vibrated and compressed.

DROP x VIBRATE x COMPRESS
Drop (-)


DROP x VIBRATE $x$ COMPRESS
Drop ( + )
Corton Dioxide (uL/gm•hr)

Figures 11 and 12. Interaction effect between dropping, vibrating and compressing on $\mathrm{CO}_{2} \mu \ell / \mathrm{gm} \cdot \mathrm{hr}$ evolved from 'Empire' apples measured 4 hours post treatment.

Drop x Compress x Package Type (Figures 13-16)
When containers were dropped and compressed, the K.P. package had $.12 \mathrm{CO}_{2} \mu \ell / \mathrm{gm} \cdot \mathrm{hr}(1 \%)$ and $.80(26 \%)$ injury score unit higher than the tray-pack container. K.P. containers compressed and not dropped had $.95 \mathrm{CO}_{2} \mu \mathrm{l} / \mathrm{gm} \cdot \mathrm{hr}(8 \%)$ and $.66(32 \%)$ injury score unit higher than the tray-pack container. Tray-pack containers dropped and not compressed had $.32 \mathrm{CO}_{2} \mu \mathrm{l} / \mathrm{gm} \cdot \mathrm{hr}$ (2\%) and . 62 (20\%) injury score unit lower than K.P. containers. Therefore, the combined effect of dropping and compressing was greater for the K.P. than the tray-pack container on $\mathrm{CO}_{2}$ output and injury scores.

## Drop x Package Type x Position (Figures 17-18)

In the K.P. containers that were not dropped, fruit in positions A and B (bottom layer) evolved the lowest $\mathrm{CO}_{2}$, or $2.56 \mathrm{CO}_{2}$ $\mu \ell / \mathrm{gm} \cdot \mathrm{hr}$ lower than in K.P. containers that were dropped. Top-layer fruit (positions $E$ and $F$ ) evolved the highest $\mathrm{CO}_{2}$ in K.P. containers that were not dropped and the lowest $\mathrm{CO}_{2}$ in containers that were dropped. Positions C and D (middle layer) in the K.P. container evolved moderate levels of $\mathrm{CO}_{2}$ for both dropped and not dropped containers. Positions E and F evolved the highest $\mathrm{CO}_{2}$ in the traypack containers whether they were dropped or not dropped. Positions $A$ and $D$ evolved the lowest $\mathrm{CO}_{2}$ in the tray-pack containers whether the containers were dropped or not dropped. Therefore, dropping affected the ordering of the positions in the K.P. containers more than in tray-pack containers.


Figures 13 and 14. Interaction effect between dropping, compressing and package type on $\mathrm{CO}_{2} \mu \ell / \mathrm{gm} \cdot \mathrm{hr}$ evolved from 'Empire' apples measured 4 hours post treatment.


Figures 15 and 16. Interaction effect between dropping, compressing and package type on injury score of 'Empire' apples.

DROP x PACKAGE x POSITION
Drop (-)


DROP $\times$ PACKAGE $\times$ POSITION Drop ( +1


Figures 17 and 18. Interaction effect between dropping, package type and position on $\mathrm{CO}_{2} \mu \mathrm{l} / \mathrm{gm} \cdot \mathrm{hr}$ evolved from 'Empire' apples measured 4 hours post treatment.

Positions F and E (top layer) evolved the lowest levels of $\mathrm{CO}_{2}$ $\mu \ell / \mathrm{gm} \cdot \mathrm{hr}$ and injury scores for non-vibrated tray-pack containers. Vibrating tray-pack containers caused Positions F and E to evolve the highest levels of $\mathrm{CO}_{2} \mu \ell / \mathrm{gm} \cdot \mathrm{hr}$ and injury scores. Vibrating K.P. containers did not change the ordering of positions for injury scores when compared to nonvibrating K.P. containers. Vibrating K.P. containers did change the ordering of positions for $\mathrm{CO}_{2}$ $\mu \ell / \mathrm{gm} \cdot \mathrm{hr}$, but the effect of vibrating and position was not as great as for vibrating tray-pack containers.


Figures 19 and 20. Interaction effect between vibrating, package type and position $\mathrm{CO}_{2} \mu \ell / \mathrm{gm} \cdot \mathrm{hr}$ evolved from 'Empire' apples measured 4 hours post treatment.


Figures 21 and 22. Interaction effect between vibrating, package type and position on injury score of 'Empire' apples.

The data obtained from this study was also analyzed as a split plot by treatment combination. For this analysis, the mean $\mathrm{CO}_{2}$ output and injury scores of the six positions in each container were grouped together. Therefore, there was one $\mathrm{CO}_{2}$ output and injury score reading for each container tested instead of six readings as in the split plot by factoral effects discussed previously. The split plot by treatment combination showed which treatment forces for a specific package type yielded the highest or lowest $\mathrm{CO}_{2}$ output or injury scores without taking position into consideration.

The split plot by treatment combination analysis of the data yielded significant differences among treatment combinations at $95 \%$ confidence limits by Duncan's multiple range test (see Figures 23 and 24). All forces applied (drop, vibrate and compress) produced the highest $\mathrm{CO}_{2}$ output for the tray-pack container, and were significantly different than eight other treatment combinations; injury scores followed a similar trend. When forces were not applied, the lowest $\mathrm{CO}_{2}$ output and injury score was obtained for the tray-pack container. The tray-pack container with no forces applied was significantly different than any container with a force applied, except for the compressed tray-pack container and the non-treated K.P. container. The dropped or compressed K.P. containers were not significantly different from one another. Mean $\mathrm{CO}_{2}$ values differed from a high of $14.15 \mu \ell / \mathrm{gm} \cdot \mathrm{hr}$ for the drop x vibrate x compress tray container to a low of $10.63 \mu \mathrm{l} / \mathrm{gm} \cdot \mathrm{hr}$ for the tray container with no forces applied, while injury score varied from a high of 3.91 for the drop $x$ vibrate $x$ compress K.P. container to 1.35 units for the tray with no forces applied.

TREATMENT vs. CARBON DIOXIDE ( $\mu \mathrm{L} / \mathrm{gm}-\mathrm{hr}$ at Hour 4)


Figure 23. Mean $\mathrm{CO}_{2}$ production of 'Empire' apples 4 hours post treatment. Means with the same letter are not significantly different by Duncan's multiple range test, $5 \%$ level.

TREATMENT vs. INJURY SCORE


Figure 24. Mean injury scores of 'Empire' apples examined 4 days after treatment. Means with the same letter are not significantly different by Duncan's multiple range, 5\% level.

1. The $\mathrm{CO}_{2}$ response of apples subsequent to simulated transit testing provided an objective method for readily identifying and measuring the visual level of injury to apples.
2. The effects of dropping, compressing and vibrating were greater on $\mathrm{CO}_{2}$ evolution and injury scores when each were applied individually than when any combination thereof was applied to a particular container. Vibrating resulted in the largest increase in $\mathrm{CO}_{2}$ production following by dropping and compressing, respectively, when compared to non-damaged fruit.
3. The $\mathrm{CO}_{2}$ evolution of apples post treatment detected the effects of different forces (dropping, vibrating and compressing) and on different shipping containers and how the effects of these forces interact. The $\mathrm{CO}_{2}$ output of apples also provided an index which indicated the least injurious position for an apple within a particular container for a specific force or combination of forces.
4. Visual injury scores were significant for all the main effects (drop, vibrate, compress and package) and drop and vibrate were the only significant main effects for $\mathrm{CO}_{2}$ evolution. Additionally, analysis of the data resulted in 9 and 4 significant two-way interactions for injury scores and $\mathrm{CO}_{2}$ evolution, respectively.

## DISCUSSION

The 4 th-hour $\mathrm{CO}_{2}$ reading yielded the highest $r$-square value for injury scores by the bucket method. There was a significant correlation between $\mathrm{CO}_{2}$ evolution and injury scores; but, r-squares were not particularly high. The low correlation coefficients might have occurred because it was difficult to see all the damaged cells on the apple fruit or the increased $\mathrm{CO}_{2}$ output may be detecting a different type of injury than simply mechanical damage. More study would be beneficial to determine the response of individual apple fruits to different destructive forces. The highest $\mathrm{CO}_{2}$ outputs were obtained for the 1sthour after-treatment and declined steadily through the 5 th-hour reading (see Appendix). Klein (31) indicated that $\mathrm{CO}_{2}$ output was highest between 3 and 6 hours post damage to apples. Pollack and Hills (27) showed a linear $\mathrm{CO}_{2}$ response of bruised red tart cherry through the 6-hour post treatment, while Hyodo, Hasegawa, Iba and Manago (14) showed that $\mathrm{CO}_{2}$ production was greatest immediately following damage of Satsuma Mandarin. Other studies $(17,20,36)$ have shown the greatest $\mathrm{CO}_{2}$ response after damage occurs within the first 24 hours following damage to horticultural crops.

Vibration had the largest effect on $\mathrm{CO}_{2}$ evolution when compared to dropping or compressing. When the apples rolled within the containers during vibration, this may have damaged more cells than dropping or compressing, especially in the layers of fruit tissue closest to the
epidermis. Additionally, a 2nd or 3 rd injury (bruise) to tissue that had already sustained impact damage by vibration would likely show no great $\mathrm{CO}_{2}$ response. However, vibration may have been a more severe treatment than either dropping or compressing. Since dropping was the first force treatment applied to the apples, followed by vibration and compression, the overall $\mathrm{CO}_{2}$ response of dropping and vibration may not have been captured, thus suggesting a lower $\mathrm{CO}_{2}$ output for dropping or vibration than what was actually observed.

The effect of compressing was decreased when containers were dropped, and the effect of dropping was decreased when containers were compressed. Compressing reduced the effect of dropping, but not to the extent that dropping reduced the effect of compressing. Possibly, dropping permitted settling of fruit within the containers; therefore, when containers were compressed, the opportunity for fruit to be damaged was likely reduced since the height of the fruit within the shipping container was reduced. The fact that compressing reduced the effect of dropping on $\mathrm{CO}_{2}$ output and injury scores may indicate once again that a 2nd or 3rd bruise to tissue that had already been damaged may show no great $\mathrm{CO}_{2}$ response or increased visible damage.

When containers were dropped, apples located near the bottom of the containers were injured the most, and apples located near the top sustained the least damage. Holt, Schrool and Lucas $(12,13)$ showed that bruising was more severe on the bottom of shipping containers than on the top layers of fruit after dropping by impact. They claimed that fruit in the lower layers of shipping containers receive multiple impacts, whereas apples in the top layers receive only one during dropping. Additionally, apples in the lower portion of a container support the weight of the apples above.

The effect of vibrating on $\mathrm{CO}_{2}$ production of apples was greater for tray-pack containers than for K.P. containers. Simulated vibration testing conducted prior to this study indicated that fruit within the K.P. containers resonated at approximately 8 Hz , while fruit in the K.P. containers resonated at approximately 12 Hz . During this preexperimental study, it was noted that the trays in the tray-pack container acted as one large spring mass system, which permitted sustained bouncing of fruit. Movement of fruit in the K.P. container was less noticeable and occurred over a smaller range of frequencies than for the tray-pack container. Therefore, there was greater opportunity for apples in the tray-pack container to experience impacts due to bouncing from vibration, thus increasing $\mathrm{CO}_{2}$ evolution and injury scores.

Holt and Schoorl (30) showed fruit located in the bottom portion of shipping containers experienced lower acceleration levels than fruit in upper levels. $0^{\prime}$ 'Brien (23) quantified the level of injury in various layers within produce containers and found upper layers of fruit had more injury after vibration testing. Similarly, in this study vibrating resulted in the highest $\mathrm{CO}_{2}$ output and injury scores in the top layer of the shipping containers, positions E and F.

The effect of compressing on injury scores was greater for the K.P. container than for the tray-pack container because the K.P. container did not prevent the compressive load force from coming in contact with the fruit. Compression strength testing of the two types of containers yielded mean compression strength values of 1002 and 2645 for the K.P. and tray-pack corrugated shipping containers, respectively.

The effect of positions differed depending on the type of container tested. Positions E and F (top layer) in the tray-pack container had the highest $\mathrm{CO}_{2}$ output, while positions A and B (bottom layer) had the highest $\mathrm{CO}_{2}$ output for the K.P. container. This interaction effect occurred because positions $E$ and $F$ (top layer) were affected more by vibration in the tray-pack container and positions A and B being greatly affected by dropping in the K.P. container.

The effect of vibrating on $\mathrm{CO}_{2}$ evolution and injury scores was less if containers were dropped and compressed; the effect of dropping on $\mathrm{CO}_{2}$ evolution and injury scores was less if containers were vibrated and compressed; the effect of compressing on $\mathrm{CO}_{2}$ evolution and injury scores was less if containers were dropped and vibrated. Vibrating and dropping permitted settling of fruit within the containers and may account for the reduced effect of compressing when containers were vibrated and dropped. Similarly, dropping containers settled fruit, which resulted in a denser pack, thus lessening the effects of vibration and compression.

The combined effect of dropping and compressing was greater for the K.P. than the tray-pack container on $\mathrm{CO}_{2}$ output and injury scores, since: 1) the K.P. container had a lower mean compressive strength value ( 1002 lbs ) compared to the tray-pack container ( 2645 lbs ), and 2) dropping had a smaller effect in the tray-pack container because the trays probably provided shock-absorbing material during impact. These trends were seen in the significant Drop $x$ Compress and Drop x Package interactions for injury scores and the significant Drop $x$ Compress $x$ Package Type interaction for $\mathrm{CO}_{2}$ evolution and injury scores.

Dropping affected the ordering of the positions in the K.P. containers more than in tray-pack containers. The pulp trays within the tray-pack container provided protection against impact by dropping, thus, the position of an apple in a K.P. package was more critical than in a tray-pack container. This was evident when K.P. containers were dropped, causing $\mathrm{CO}_{2}$ evolution to be highest for apples in the bottom of the container, while the $\mathrm{CO}_{2}$ evolution from apples in the tray-pack containers was only negligibly affected by dropping.

Vibrating the K.P. container did change the ordering of positions for $\mathrm{CO}_{2}$ evolution, but not to the extent that vibrating altered the ordering in the tray-pack container. Vibrating affected fruit located in the upper layer of the tray-pack containers more than in the K.P. containers since the trays amplified the simulated vibration inputs, resulting in sustained bouncing of the fruit. Additionally, apples located in the top layer of the tray-pack container were affected most, where the highest acceleration is.

## $\mathrm{CO}_{2}$ EVOLUTION AS METHOD TO DETERMINE THE PROTECTIVE CHARACTERISTICS OF SHIPPING CONTAINERS

Simulated transit handling of apples in shipping containers resulted in an increase in $\mathrm{CO}_{2}$ evolution similar to those applied to individual fruits by previous investigators. Therefore, a damage detector system for assessing the protective characteristics of produce shipping containers which utilizes the objective increase in carbon dioxide of mechanically injured apples could be carried out as follows:

1. Select fruit of similar variety free from obvious physical damage and physiological disorders, uniform in size and maturity. An entire shipping container can be filled with optimum fruit or the fruit $\mathrm{CO}_{2}$ response will be measured from can be carefully placed among less than optimum filler fruit of similar size and maturity.
2. Fruit should be pre-conditioned to the test temperature upon removal from cold storage.
3. Various forces (e.g., dropping, compression and/or vibration) are applied in a designated order to the different shipping containers the experimentor wishes to evaluate, or the $\mathrm{CO}_{2}$ response could be measured from apples after a truck, rail or air ride.
4. Following damage treatment, the fruit is carefully removed and placed in an airtight container which provides a minimum amount of headspace.
5. Gas samples are analyzed from the airtight containers using a gas chromatograph or infrared $\mathrm{CO}_{2}$ analyzer four to five hours after sealing the containers.
6. After sampling the accumulated $\mathrm{CO}_{2}$ within the airtight containers the fruit is weighed.
7. The level of $\mathrm{CO}_{2}$ detected by a gas chromatograph or infrared $\mathrm{CO}_{2}$ analyzer is calculated based on fruit weight and headspace volume of the airtight containers.
8. The rates of $\mathrm{CO}_{2}$ outputs from the apples that were located in different locations and/or shipping containers with various forces applied can then be compared to one another, and to nondamaged fruit of similar variety, size and maturity in the same test.

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## LIST OF REFERENCES

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## APPENDIX

$\mathrm{CO}_{2}$ Evolution 1, 2, 3, 4 and 5 Hours
Post Treatment and Injury Score
cv. 'Empire' Apples Damaged in

Corrugated Shipping Containers
for Each Treatment Combination




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[^0]:    1/ Klein, J. D. 1983. Physiological causes for changes in carbon dioxide and ethylene production by bruised apple fruit tissues. Ph.D. dissertation, Michigan State University, East Lansing.

[^1]:    $\overline{1 /}$ D. H. Dewey, Ph.D., Professor Emeritus and Michael L. Parker, Graduate Student, Michigan State University, East Lansing, MI 48824.

