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Effect of Phytosanitary Irradiation and Methyl Bromide Fumigation on the Physical, Sensory, and Microbiological Quality of Blueberries and Sweet Cherries

Effect of phytosanitary irradiation and methyl bromide fumigation on the physical, sensory, and microbiological quality of blueberries and sweet cherries

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ABSTRACT

BACKGROUND

The objective of this study was to determine whether irradiation could serve as a suitable phytosanitary treatment alternative to methyl bromide (MB) fumigation for blueberries and sweet cherry and also to determine the effect of phytosanitary irradiation treatment on survival of *Salmonella* spp. and *Listeria monocytogenes* on these fruit. 'Bluecrop' blueberries (*Vaccinium corymbosum*) and 'Sweetheart' cherries (*Prunus avium*) were irradiated at 0.4 kGy or fumigated with methyl bromide and evaluated for quality attributes during storage.

RESULTS

Irradiation caused an immediate decrease in firmness of both fruit without further significant change during storage. Fumigated fruit, in contrast, softened by 11-14% during storage. Irradiation did not adversely affect blueberry and cherry shelf-life and irradiated fruit. MB fumigation did not impact blueberry and cherry quality attributes initially, however, fumigated fruit exhibited greater damage and mold growth than the control and irradiated samples. Irradiation at 400 Gy resulted in a ~1 log CFU/g reduction in *Salmonella* spp. and *Listeria monocytogenes* counts, indicating that this treatment cannot significantly enhance safety.

CONCLUSION

This study indicates that irradiation at a target dose of 0.4 kGy for phytosanitary treatment does not negatively impact blueberry and cherry quality and can serve as an alternative to methyl bromide fumigation.

Keywords: gamma irradiation, methyl bromide, blueberries, sweet cherries, damage, shelf life, *Salmonella*, *Listeria monocytogenes*

INTRODUCTION

The United States is the world's largest producer of blueberries and second largest producer of cherries with 290,344 MT highbush blueberries (*Vaccinium corymbosum*) [1] and 300,000 MT of sweet cherries (*Prunus avium*) produced in 2014. [2] The U.S. also exports significant amounts of both crops. The United States exported 11% of blueberries in 2012 [3] while 45% of sweet cherries produced were exported. [4] Importing countries require a phytosanitary certificate and will often include treatment requirements on their import permits to address insect pests common to these fruit [5] including blueberry maggot (*Rhagoletis mendax* Curran), plum curculio (*Conotrachelus nenuphar*), Mediterranean fruit fly (*Ceratitis capitata*), western cherry fruit fly (*Rhagoletis indifferens*), spider mites (*Tetranychus urticae*), black cherry aphids (*Myzus cerasi*). [2]

Methyl bromide (MB) fumigation is a common phytosanitary treatment that meets most countries' export requirements, [6] however, it is a potent greenhouse gas and is scheduled to be phased out under the Montreal protocol. [7] MB fumigation schedules require the fruit to be at a minimum temperature of 10-16°C for several hours, with the length of the treatment dependent on the temperature and methyl bromide concentration conditions. For example, blueberries and cherries exported to India must be fumigated at 32 g/m^3 for 2 h at 21°C or above under normal atmospheric pressure or equivalent conditions. After the produce is fumigated, the gas must be exhausted, which usually takes several hours, thus exposing the fruit to warm temperatures for an

extended period. Little information is available on the effect of MB fumigation on blueberry quality or shelf-life however, on cherries, fumigation can increase bruising and pitting. [8,9]

Ionizing irradiation is a highly efficacious phytosanitary treatment approved by United States Department of Agriculture-Animal Plant Health Inspection Service (USDA APHIS). Irradiation treatment results in minimal temperature increase, and fruit can be maintained cold if the facility is refrigerated. USDA APHIS has approved fruit to be treated at a minimum generic dose of 0.4 kGy to destroy all insect pests excluding *Lepidoptera* larvae and pupae. [5] At this dose level, blueberries and cherries show minimal changes in quality attributes, [10,11,12] The end of blueberry and cherry shelf-life is often characterized by growth of fungi such as *Monilinia fruticola*, *Rhizopus stolonifer*, and *Botrytis cinerea.* [13] Irradiation at 2-3 kGy has been shown to inhibit growth of these spoilage organisms, [14] but the effect of phytosanitary dose levels on fungal growth, and hence shelf-life, is not known.

In addition to impact of irradiation on shelf-life, growers are interested in knowing if the dose levels used for phytosanitary purposes can also enhance safety of berries. In general, berries are not common carriers of bacterial pathogens but there have been incidents of foodborne illnesses linked to fresh berries. [15] In 2003, an outbreak of *Salmonella enterica* in California was linked to strawberries resulting in 13 illnesses and two hospitalizations. [16] In June 2009, there was a multistate outbreak of *Salmonella Muenchen* which caused 14 illnesses and was linked to blueberries. [16] *Listeria* has not been associated with berries, but recent outbreaks related to cantaloupes and apples suggest that contamination can occur in packing houses. [17] While cherries are hydrocooled with chlorinated water, blueberries are not washed prior to packing and neither fruit receives a lethal treatment to kill microorganisms. Thus, it would be beneficial if phytosanitary irradiation treatment could also effectively reduce pathogen counts.

The primary objective of this study was to determine whether irradiation could serve as a suitable phytosanitary treatment alternative to MB fumigation by comparing the effects of irradiation and MB on blueberry and sweet cherry quality and shelf-life. The second objective was to determine the effect of phytosanitary irradiation treatment on the survival of *Salmonella* spp. and *Listeria monocytogenes* on these fruits.

MATERIALS AND METHODS

2.1 Study Design and Sample Procurement

Fresh 'Bluecrop' blueberries (*Vaccinium corymbosum*) were harvested from Pan American Berry Growers farm (Salem, OR) on July 8, 2013 and August 5, 2013, cooled in a forced-air cooler and packed in 340 gram (12 oz) clamshells, 12 clamshells per tray. The blueberries were shipped to Chapman University in a refrigerated truck, a distance of 1,537 km, one week following harvest. Fresh 'Sweetheart' cherries (*Prunus avium*) were harvested on July 23, 2013 and August 13, 2013 by Stemilt Growers LLC in Wenatchee, WA were hydrocooled with chlorinated water and packed in 0.90 kg bags, 8 per box. Each box was lined with a low-density polyethylene (LDPE) liner (Freshlok, Yakima, WA). The cherries were transported via refrigerated truck to Chapman University, a distance of 1,880 km.

The fruit was stored in Chapman University's refrigerated unit at 0-1° C and 90-95% relative humidity prior to and following treatments. LogTag ® data loggers (Auckland, New Zealand) were placed in the fruit trays and cases to monitor the temperature and relative humidity during the storage study. The fruit was subjected to irradiation and methyl bromide treatments on the day following receipt.

2.2 Gamma Irradiation

Irradiation treatment was conducted at Sterigenics, Inc. (Tustin, CA). Dose rate was determined using dummy trays/cartons placed in exactly the same configuration as the trays used for sampling. The trays/cartons were stacked in rows of four and columns of six on top of wooden racks. Twelve dosimeters were distributed at various locations in the trays to monitor the maximum and minimum absorbed doses. The trays were placed at a fixed distance from the Cobalt-60 source and irradiated at a dose rate of 0.95 kGy per hour for the blueberries and 0.74 kGy per hour for the sweet cherries. Halfway through the treatment, the sample trays were rotated 180° for a more uniform dose treatment. The fruit received a target dose of 0.4 kGy with a Dmax/Dmin ratio of 1.33. Once the irradiation process was complete, the fruit was transported to Chapman University and stored at 0-1°C.

2.3 Methyl Bromide Fumigation

Fruit cases to be fumigated were kept under ambient conditions overnight to allow the temperature of the cherries and blueberries to reach a minimum of 21°C, as required by India's fumigation protocol. [18] The liners in the cherry cartons were opened and cases of both fruit were exposed to methyl bromide at a concentration of 32 g/m^3 in a chamber operated by Global Pest Management, Inc. (Long Beach, CA) for a duration of 2 h at 21°C. Following fumigation, the chamber was aerated for four hours to exhaust the gas. Fruit cases were cooled to 1°C in a forced air cooler, then transported back to Chapman University where the LDPE liners in the cherry cases were fastened using rubber bands and fruit stored at 0-1°C.

Fruit quality was measured at three points during storage. The first data point is for measurements made on the day following treatments corresponding to day 11 after harvest for

blueberries and day 6 after harvest for cherries, and the second data point is for measurements at approximately the midpoint of shelf-life, and the third data point is for measurements made towards the end of shelf life of the two fruit.

2.4 Yeast and Molds

Duplicate 25 g of blueberries or pitted sweet cherries were stomached at 230 rpm for 90 sec (Stomacher 400 Circulator, Seward, United Kingdom) in a filter bag with 250 mL sterile buffered peptone water (Thermo Fisher Scientific, Massachusetts, USA). Serial 10-fold dilutions were prepared from the stomached samples with peptone water. One mL aliquots of the diluted samples were plated in triplicate onto Yeast and Mold Petrifilms (3M, Minnesota, USA). The petrifilms were incubated at 25 ºC for 5 days and colonies counted.

2.5 Damage Evaluation

All blueberries contained in three random clamshells, each containing between 200-250 blueberries, were evaluated weekly to count the number of berries that were crushed, shriveled, leaking, split, or moldy. For cherries, 100 berries from each treatment were evaluated for noticeable pitting, bruising, cracking, or moldiness. The ratio of damaged fruit to the total number of fruit was used to determine the percentage of damaged fruit.

2.6 Fruit firmness

Blueberry firmness was measured using a Kramer Shear Press with five blades (TA-91) attached to a Stable Micro System Texture Analyzer (Model TA-XT2, Texture Technology Corp. Scarsdale, N.Y., U.S.A., and Stable Microsystems, Godalming, Surrey, U.K.). The five

flat-plate plunger was moved at a speed of 4.0 mm/s through 100 g blueberries with a post-test speed of 10.0 mm/s. Exponent software recorded the maximum force (N) required to shear through the blueberries. Six replicates were made for each treatment.

Cherry firmness was measured using a Firmtech II (Bioworks, Inc., Wamego, KS). This instrument records the force required to compress each cherry by one mm at a load cell speed of 15 mm/s. The compression firmness of 100 fruit was measured for each treatment.

2.7 Soluble Solids Content (SSC)

Blueberries and pitted cherries were juiced (Elite Gourmet Maxi-matic Juice Extractor TS-738, City of Industry, CA) and filtered through 2-3 layers of cheesecloth. A pipette was used to transfer several drops of clear, fruit juice on the prism of a Digital "Pocket" Refractometer PAL (Atago Co. Ltd., Tokyo, Japan) to determine the percent total soluble solids content. Measurements were made in triplicate for each treatment.

2.8 Titratable Acidity (TA)

Five mL of filtered blueberry juice or six g of cherry juice were combined with 50 mL of carbon dioxide-free water. The solution was titrated to pH 8.2 with 0.1 N NaOH. Initial and final pH values were measured using the pH200 Hannah Instruments (Woonsocket, RI). Measurements were done in triplicate for each treatment and % acid was calculated using the following equation with 0.064 as the acid factor for citric acid (for blueberries) or 0.067 for malic acid (for cherries):

% acid = (mL NaOH) x (0.1 N NaOH) x (acid factor) x (100)/mL or g of sample

2.9 Cherry Color and Gloss

Color expression was based on CIE L*, a*, b*, C* and hue angle values using a white tile calibrated spectrophotometer (CR-700d, Minolta, Tokyo, Japan). For fruit color, a single measurement was made on the lateral side of each fruit and averaged over 20 replications per treatment.

Gloss was measured using a spectrophotometer (CR-700d, Minolta, Tokyo, Japan) equipped with a shield having a circular 15 mm-diameter aperture and expressed as gloss units (GU) at an angle of 60°. For each treatment, 20 fruit were used with 20 measurements per treatment. [19]

2.10 Gas concentrations

Oxygen and carbon dioxide concentrations in the cherry cases were monitored during storage using a gas analyzer (Pac Check 650, Mocon, Minneapolis, MN) through a septum attached to the liner bags to prevent tears.

2.11 Weight Loss

Two blueberry and cherry trays from each treatment were used to track weight during storage using a SB32000 scale (Mettler Toledo, Columbus, OH) to determine weight loss.

2.12 Sensory Evaluation: Consumer Affective Testing

Approximately 75 consumers evaluated the blueberries and cherries on each testing day. One h prior to consumer testing, the fruit were pulled from refrigeration, brought to room temperature, lightly rinsed with cold water and patted dry with paper towels. Two cherries and

six blueberries selected at random from each treatment were placed in 2 oz. plastic soufflé cups with lids labeled with random 3-digit codes produced by SIMS 2000 Sensory Evaluation Software (Berkeley Heights, NJ). Consumers rated the degree of liking for the fruits' appearance, flavor, texture, and overall liking on a 9-point hedonic scale. [20] Tests were administered through SIMS 2000 Sensory Evaluation software with samples and their corresponding questions were presented on a randomized blind incomplete block system to prevent positional bias. Consumers were prompted to cleanse their palate with an unsalted soda cracker and filtered water between samples.

2.13 Survival of Salmonella and Listeria monocytogenes

Since fumigation was conducted at a commercial facility and because the treatment requires the product to be exposed to allow penetration of methyl bromide, the effect of fumigation on inoculated pathogen counts was not tested. Thus, only the impact of phytosanitary irradiation on pathogen counts was evaluated in this study.

2.13.1 Salmonella and Listeria Inoculum Preparation

Five strains of *Salmonella enterica* serovars Muenchen, Newport, Typhimurium, Saintpaul, and Agona), and two strains of *Listeria monocytogenes* (*L. monocytogenes* T1 and *L. monocytogenes* T4) were obtained from the FDA Pacific Regional Laboratory Southwest (Irvine, CA). One loopful (~10 μL) of bacteria from each slant was streaked on each of three Tryptic Soy Agar petri plates containing 0.6% yeast extract (TSAYE), (Thermo Fisher Scientific, Massachusetts, USA) and incubated at 37°C for 24 h. An isolated colony from each incubated plate was inoculated into 25 mL of Tryptic soy broth (TSB) (Thermo Fisher Scientific,

Massachusetts, USA) in individual tubes and incubated at 37°C for 24 h. The tubes were then centrifuged (Thermo Fisher Scientific, Massachusetts, USA) for 2 minutes at 3000 rpm at 4°C. The supernatants of both blueberry and cherry samples were discarded and 45 mL of autoclaved TSB was added to the tubes and the pellet was re-suspended by vortexing the tubes. The tubes were incubated for 24 h at 37°C. The process of centrifugation, resuspension and incubation explained above was performed three times. Five mL of final inoculum from each of the five *Salmonella* strain tubes were mixed to achieve a concentration of approximately 10^9 CFU/mL for Salmonella and 5 mL of final inoculum from each of the two *L. monocytogenes* strain tubes were mixed to achieve a concentration of approximately 10^7 CFU/mL for *L. monocytogenes*.

2.13.2 Salmonella and Listeria Inoculation Procedure

Triplicate 25 g samples of blueberries and cherries were prepared separately for the inoculation study. Each sample was spot inoculated with 75 µL of *Salmonella* or *Listeria* inoculum on sterile trays in a biosafety cabinet, then allowed to dry for 2 h at 21°C. The dry samples were placed aseptically in sterile filter bags and sealed. Each filter bag was placed in a Ziplock™ bag, and all the Ziplock™ bags were placed in a single cardboard box and sent for irradiation treatment as described previously. Inoculated samples that were not irradiated were used as the control samples for the experiment.

2.13.3 Enumeration of Salmonella and Listeria in Blueberries

Peptone water, 225 mL, was added into each of the sterile filter bags (containing 25 g samples) and stomached for 3 minutes. Serial dilutions prepared with peptone water were surface plated on XLD agar plates overlayed with 10 mL of TSAYE for *Salmonella* and on PALCAM

Agar (Thermo Fisher Scientific, Massachusetts, USA) plates overlayed with 10 mL TSAYE for *Listeria*. The selective media diffuses into the TSAYE and helps suppress the background micro flora, while TSA allows the injured cells to resuscitate and form colonies. [21] The agar plates were incubated for 48 h at 37°C. For *Salmonella*, isolated typical (appear as pink colonies with/without black center) and atypical colonies (yellowish with/without black center) were counted. [22] Isolated *Listeria* colonies that appeared as greyish-green were counted. [237]

2.14 Statistical Analysis

A longitudinal randomized treatment design with repeated measurements was employed and the R statistical software package (R Development Core Team 2012, Vienna, Austria) was used for model building, estimated mean generation, and interaction effects. Linear mixed effect models with random effects were applied to analyze the effects of treatment and time on the various quality attributes as well as sensory data. The model was used to compare the changes of every treatment across time and also compare all treatments at each time point. We used Bonferonni correction for multiple comparisons in order to keep Type-1 error rate at the nominal level.

RESULTS AND DISCUSSION

3.1 Mold growth

Initial yeast and mold counts on blueberries were approximately 3 logs CFU/g and irradiation at 0.4 kGy did not significantly affect yeast and mold counts (Fig. 1A). During storage, the counts remained unchanged for the control and irradiated blueberries. Fumigated

samples maintained similar counts up to day 37 after which the counts increased by about 1 log as compared to the control ($P<0.05$).

Mold levels in cherries were very low initially and increased gradually during refrigerated storage to a little over 3 log CFU/g after 34 days (Fig. 1B) after which they remained in the 3-4 log range. Irradiation did not impact mold growth as compared to the control, however fumigated cherries consistently had higher counts ($p<0.05$).

Monilinia fruticola, *Rhizopus stolonifer*, and *Botrytis cinerea* are the common fungi found on berries and cherries. [13] Kim and others [24] studied the effects of gamma irradiation on peaches and found that a dose of 1 kGy was sufficient to inactivate a number of molds including *B. cinerea*, *R. stolonifer*, and *M. fruticola*. The D values for each of these species on peaches was 0.15 kGy, 0.16 kGy, and 0.16 kGy, respectively. In our study, however, 0.4 kGy did not lower mold counts as compared to the control suggesting that the D values for these organisms on cherries is higher than 0.4 kGy. In fact, Jeong and others [14] determined D values for pure cultures to be between 3 and 4 kGy for *B. cinerea* and 1 and 2 kGy for *P. expansum* and *R. stolonifera* and several studies show that decay in blueberries is not impacted even at 3.0 kGy. [10, 25, 26, 27] Drake and Neven [28] saw increased defects and softening in Bing cherries irradiated at 0.9 kGy so it is likely that at the doses required for significant mold reduction, quality may be compromised.

Mold growth occurred earlier in the fumigated fruit as compared to control and irradiated samples. MB can act as a fungicide, [29] but at the treatment levels for phytosanitary purposes, it was not only insufficient to control mold, but seemed to enhance mold growth, most likely as a result of the high temperature exposure.

3.2 Damage Evaluation

Soft blueberries were the primary reason for categorizing blueberries as damaged, but any berries that were shriveled, split, leaking were also deemed as damaged. The percent of blueberries that were initially soft was high, with percentages of almost 50%, most likely because these were harvested late in the season (Fig. 2A). By day 31, the control sample had the least ($P<0.05$) number of damaged berries, 69.5%, as compared to more than 87% of the MB and 81.5% of the IRR blueberries. White mold spots became visible in fumigated blueberries at approximately day 31 after harvest. Control and irradiated berries developed mold closer to day 37.

For cherries, pitting, bruising, and dark spots were the most common defects with the highest incidence in the fumigated cherries (Fig. 2B). Cherries started to soften and sliminess was noticeable about 39 days after harvest, but mold growth was not always obvious until about day 45 after harvest. Irradiated and control cherries were slimy but did not show mold even until day 56 following harvest

The number of soft blueberries correlates with the Kramer Shear values (Tables 1-2), with the fumigated samples showing higher ($P<0.05$) number of soft berries and lowest firmness values on day 31 as compared to the control. Moisture loss causes corrugation and thickening of the epidermal and parenchyma cell walls in the outer skin layer of the blueberries and reduced turgor which can result in softening. [30] In cherries, Drake and Neven [9] saw no effect on visual quality of Bing and Ranier cherries irradiated at 0.3 or 0.6 kGy but at 0.9 kGy, Drake and Neven [28] saw increased bruising and pitting. Drake et al. [8] found that MB-treatment increased pitting in Bing and Rainier cherries, but they did not report on sliminess or mold development.

3.3 Firmness

Irradiated blueberries were 11% softer than control samples during storage (Fig. 3A). In cherries, irradiation caused immediate softening as evident by lower values for compression firmness as measured by the Firmtech (14-17%) but the differences between treatments were not significant at any point during storage (Fig. 3B). Irradiation degrades cell carbohydrates, particularly pectin, and ruptures the cell wall, decreasing turgor pressure and firmness of plant cells. [31] Irradiation-induced loss in firmness in blueberries and cherries has been documented previously. [32, 27, 28] While loss in firmness was measurable, consumer scores for texture were not impacted by irradiation in either fruit (Tables 1-2).

Fumigated blueberries were 14% softer than the control samples ($P<0.05$) at the end of storage and in cherries, fumigation lowered compression firmness by 11% ($P<0.05$). Commodities treated with methyl bromide fumigation could be expected to soften because of the warm temperature during treatment that can increase respiration rate and accelerate ripening, resulting in loss of firmness and enhanced decay. [33] Neven and Drake [12] saw a decrease of 9% and 11% in hardness of MB-treated Bing cherries (1.13 $kg.m^{-3}$, 6°C) after 7 and 14 d of storage, respectively when compared to control cherries, but in an earlier study, Drake and Neven [9] found that fumigation with a lower concentration of MB (56 $g.m^{-3}$, 6°C) did not affect firmness.

3.4 Soluble Solids Content

The SSC values for blueberries and cherries were not impacted by treatment or storage (Tables 1-2). Briggita blueberries that were irradiated up to 1.0 kGy [34] and Rabbiteye blueberries irradiated up to 1.25 kGy [25] also showed no significant effect of dose or storage on SSC. Jessup [35] found that irradiation up to 0.6 kGy did not affect SSC of cherries and neither did MB

fumigation (6°C, 56 g m^{-3}) [9] however, a higher concentration of MB, 1.13 kg m^{-3}, lowered SSC in MB-treated cherries compared to control and irradiated cherries. [12]

3.5 Titratable Acidity

There was no impact of irradiation or fumigation on TA values of blueberries or cherries (Tables 1-2). Previous studies have also noted negligible changes in TA levels of fresh blueberries irradiated up to 3.2 kGy [32] and cherries treated up to 0.6 kGy. [9] However, fumigation has been shown previously to increase TA in Bing cherries. [9, 8]

3.6 Color and gloss

Neither treatment nor storage affected color of cherries (Table 2). Irradiation generally does not impact color of cherries. [28, 9] but Drake and Neven [9] observed that MB fumigation caused darkening of the fruit as seen as a decrease in L* value and an increase in hue values (less red). Gloss values declined gradually during storage ($P>0.05$). Products that are freshly harvested tend to have a bright, glossy surface that can be affected by storage, weight loss and handling. Therefore, cherries must be carefully handled and stored at ideal temperatures in order to maintain glossiness.

3.7 Consumer Testing

The initial scores for blueberries and sweet cherries were between 5 and 7 for all attributes (Tables 1-2) corresponding to “neither like nor dislike” to “like moderately.” There were no significant ($P>0.05$) effects of treatment on consumer liking of any of the attributes, but

fumigated cherries exhibited a high degree of decay by day 45 such that they were not suitable for consumer testing.

Miller and McDonald [27] found that blueberry texture and flavor were not affected by gamma irradiation up to 1.0 kGy, while Moreno et al. [32] did not observe changes in blueberries treated at 1.6 kGy. A trained panel found no differences in bruising of irradiated and control Bing cherries irradiated at 0.6-0.8 kGy immediately after harvest and stored for 2 days in ambient temperature. [36]

3.8 Gas Concentrations

Gas concentrations in the cherry samples are shown in Fig 4, which stabilized at 12-14% O_2 and 6-8% CO_2. The recommended gas concentrations for cherries packaged in modified atmosphere bags is 3 - 10% O_2 and 10 - 15% CO_2. [13] Irradiated samples had higher CO_2 and lower O_2 than control and fumigated cherries initially, suggesting that irradiation may have caused an increase in respiration rate. However, O_2 levels increased and CO_2 levels decreased in the irradiated samples to achieve concentrations similar to the control and fumigated samples. Irradiation has been known to induce a temporary increase in respiration rate in fresh produce including cut iceberg lettuce, mangoes and plums, citrus, avocadoes. [37, 31] The transient increase in respiration rate of the irradiated cherries does not seem to have affected any of these quality factors as compared to the control.

3.9 Weight Loss

There was no impact of treatment on weight loss of either fruit. Weight loss ranged from 1.2-1.5% for blueberries and cherries (data not shown), most likely due to loss of moisture.

3.10 Survival of *Salmonella spp.* and *Listeria monocytogenes*

Our results show that the *Salmonella* cocktail declined by less than a log initially then remained constant on the inoculated blueberries (Fig. 5A). Irradiation at 400 Gy reduced *Salmonella* counts by under a log immediately and this differences was maintained during storage. *Salmonella* counts on inoculated sweet cherries remained constant for two weeks and then started to decline (Fig. 5B). Upon irradiation, counts declined by 1 log CFU/g and continued to decline gradually until day 14 and rapidly thereafter. D_{10} values for *Salmonella* on diced tomatoes were reported to be in the range of 0.26 kGy to 0.39 kGy, [38] and in the range of 0.26 kGy to 0.32 kGy on green onions. [39] In a recent study, Palekar et al. [40] determined the D value for *Salmonella* Poona on cut cantaloupe to be 0.21 kGy. Thus the ~1 log reduction of *Salmonella* observed in our study is not unexpected.

L. monocytogenes counts remained unchanged on control blueberries during the four week storage study, but increased on cherries (Fig. 5B). On blueberries, irradiation resulted in an initial 1.12 $\log_{10}$ CFU/g reduction in *Listeria* counts ($P<0.05$), but the difference decreased to 0.66 $\log_{10}$ CFU/g by the end of the testing period ($P>0.05$). On irradiated cherries, *L. monocytogenes* counts decreased by 0.9 $\log_{10}$CFU/g and this difference in counts was maintained during storage. Todoriki et al. [41] have reported D_{10} values for *Listeria monocytogenes* on inoculated cherry tomatoes to be 0.20 - 0.22 kGy. In our study, only a log reduction was observed following treatment at 0.4 kGy, in contrast to Mohacsi-Farkas et al. [42] who reported a 2 log reduction on fresh cut cantaloupe after 1 kGy irradiation treatment.

CONCLUSIONS

Irradiated blueberries and cherries were not different than control fruit for any quality attribute other than firmness, and irradiation did not improve or reduce shelf-life. Fumigation did not impact quality attributes initially but shelf-life was compromised due to development of sliminess and mold. In our study, the fruit was allowed to warm up to 21.1°C, a process that took about 12 hours. Fumigation at that temperature took 2 hours and aeration required another 4 hours resulting in an 18 hour break from the cold chain. In contrast, the irradiated fruit were exposed to ambient temperatures for approximately two hours. The extended exposure to higher temperatures for fumigation is most likely the cause of the greater damage, decay, and shorter shelf-life observed in the fumigated berries compared to the control. At 0.4 kGy the modest reduction in *Salmonella* and *Listeria* counts will not contribute significantly to improving safety. Our results show that irradiation at a target dose of 0.4 kGy does not adversely or positively impact blueberry or sweet cherry quality or shelf-life and can serve as a good alternative to methyl bromide fumigation.

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Table 1 – Effect of irradiation and fumigation on quality attributes and sensory scores (1-9 hedonic scale) of blueberries during storage.

	Day 11			Day 22			Day 31		
	Control	**IRR**	**MB**	**Control**	**IRR**	**MB**	**Control**	**IRR**	**MB**
Soluble Solids Content (SSC)	12.0	12.6	12.0	11.8	12.3	12.0	11.7a	12.0b	12.0b
Titratable Acidity (%)	0.38	0.3	0.36	0.37	0.32	0.37	0.36	0.33	0.37
SSC/TA Ratio	31.6	42	33.3	31.9	38.4	32.4	32.5	36.4	32.4
Consumer testing									
Appearance	6.7	6.9	7.0	6.6	6.6	6.8	6.5	6.4	6.7
Flavor	5.6	6.2	6.0	5.5	5.5	6.2	5.5	5.2	6.3
Texture	5.6	6.0	6.2	5.9	5.9	6.0	6.0	5.9	6.0
Overall Liking	5.6	6.0	6.1	5.7	5.8	6.0	5.8	5.7	5.9

Values in the same row/column that are followed by the same letter are not significantly different. Letters a-c in a row represents differences between treatments on any given day. Letters x-y in a row represents differences between days for a given treatment.

Table 2 – Effect of irradiation (IRR) and methyl bromide fumigation (MB) and on quality attributes and sensory scores (1-9 hedonic scale) of sweet cherries during storage.

	Day 6			Day 32			Day 45		
	Control	**IRR**	**MB**	**Control**	**IRR**	**MB**	**Control**	**IRR**	**MB**
Soluble Solids Content (SSC)	19.6	19.6	18.7	18.6	21.1	14.3.0	19.2	17.1	18.4
Titratable Acidity (%)	0.67	0.68	0.64	0.43	0.53	0.35	0.5	0.42	0.47
SSC/TA Ratio	29.3	28.8	29.2	43.3	39.8	40.9	38.4	40.7	39.2
Color									
L	21.89	21.97	21.62	21.51	21.62	21.42	22.04	21.89	22.46
a	13.37y	14.5y	13.54	12.64x	13.1x	12.77	12.52x	12.0x	14.03
b	16.07y	15.99x	15.64x	14.95ax	18.32cy	16.42by	16.1ay	16.57ax	18.34bz
Gloss	13by	16cy	9a	8x	10x	10	12by	10ax	9a
Consumer testing									
Appearance	6.8	6.9	7.2	6.1	7.5	6.7	6.2	7.3	NA
Flavor	7.1b	6.5a	7.1b	7.0	7.3	7.0	7.1	6.8	NA
Texture	6.8	6.8	6.9	7.0	7.5	6.4	6.5	6.6	NA
Overall Liking	6.7	6.7	6.9	6.7	7.6	6.5	6.6	6.3	NA

Letters a-c in a row represents differences between treatments on any given day. Letters x-y in a row represents differences between days for a given treatment.

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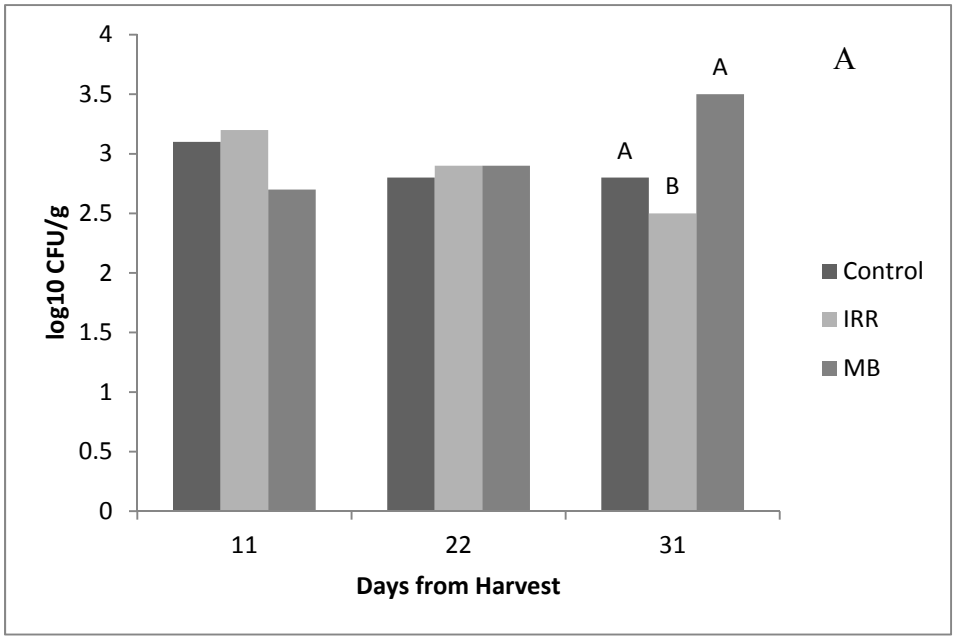


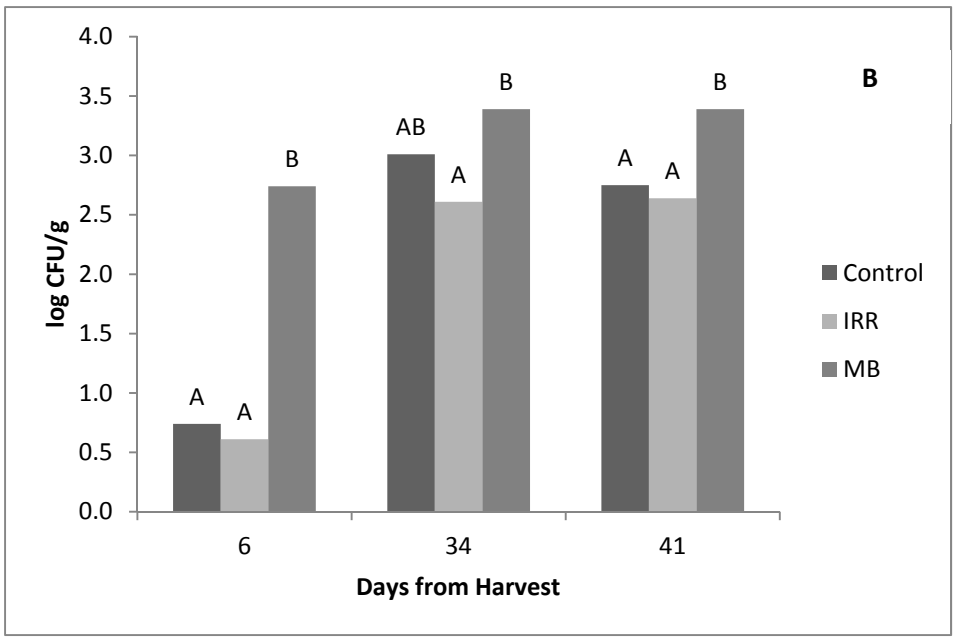


Figure 1 – Effect of irradiation at 0.4 kGy on yeast and mold counts on A. blueberries until day 31 after harvest and B. sweet cherries until day 41 after harvest. Values on the same day that are followed by the same letter are not significantly different.

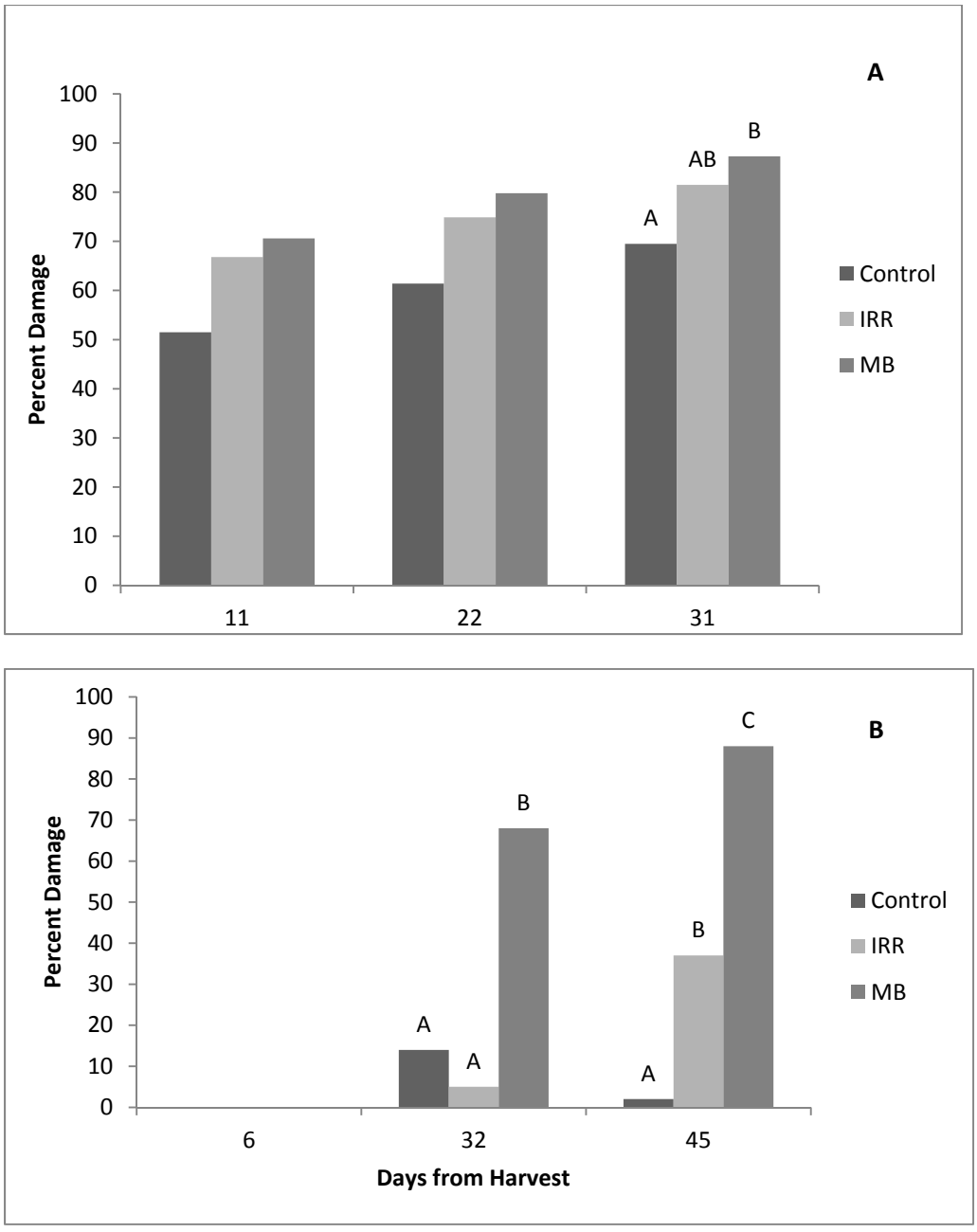


Figure 2 – Effect of irradiation (IRR) and fumigation (MB) on the extent of damage and decay in A. blueberries and B. sweet cherries during storage. Values on the same day that are followed by the same letter are not significantly different.

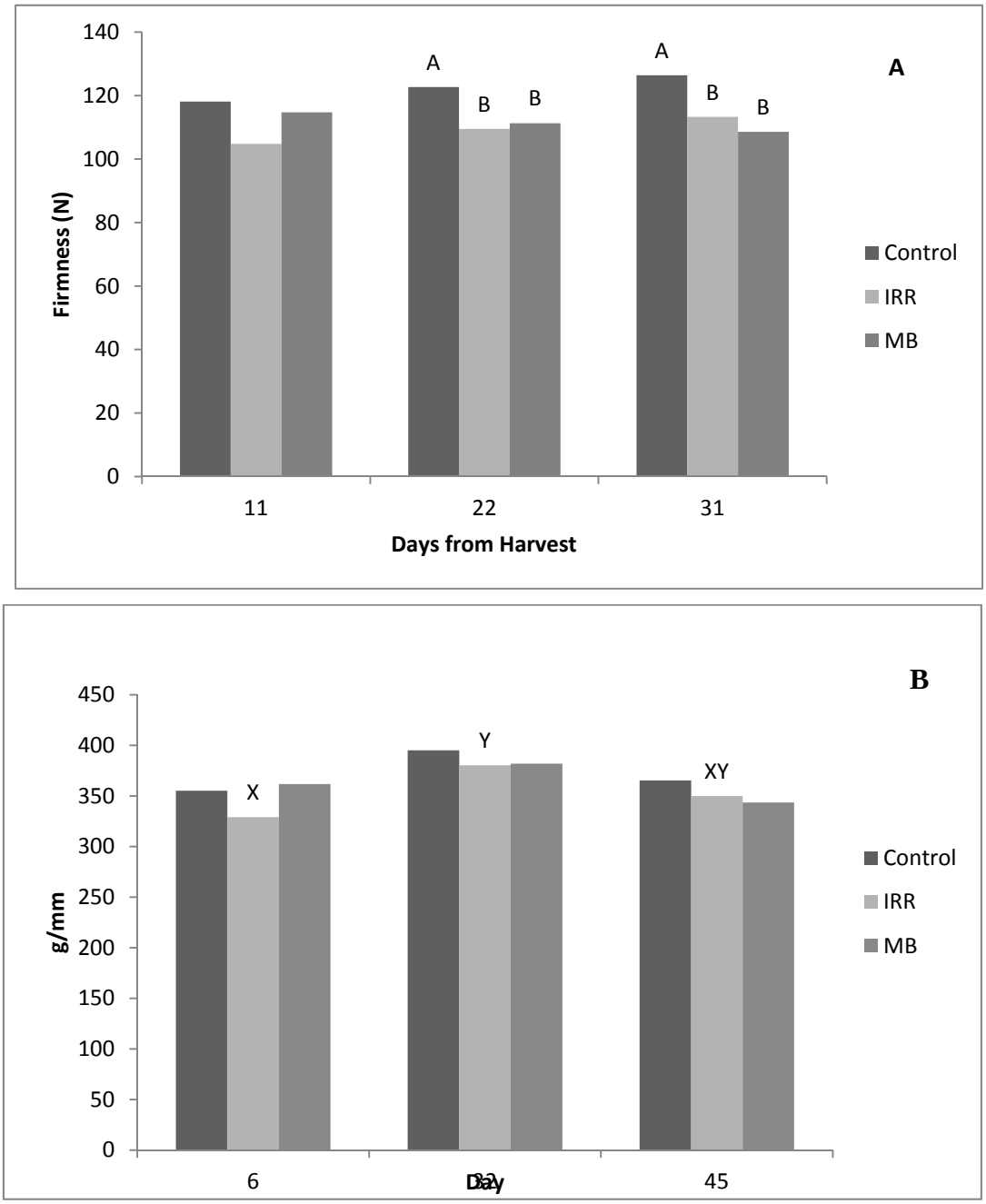


Figure 3 – Effect of irradiation (IRR) and fumigation (MB) on the firmness of A. blueberries and B. sweet cherries during storage. Values on the same day that are followed by the same letter are not significantly different.

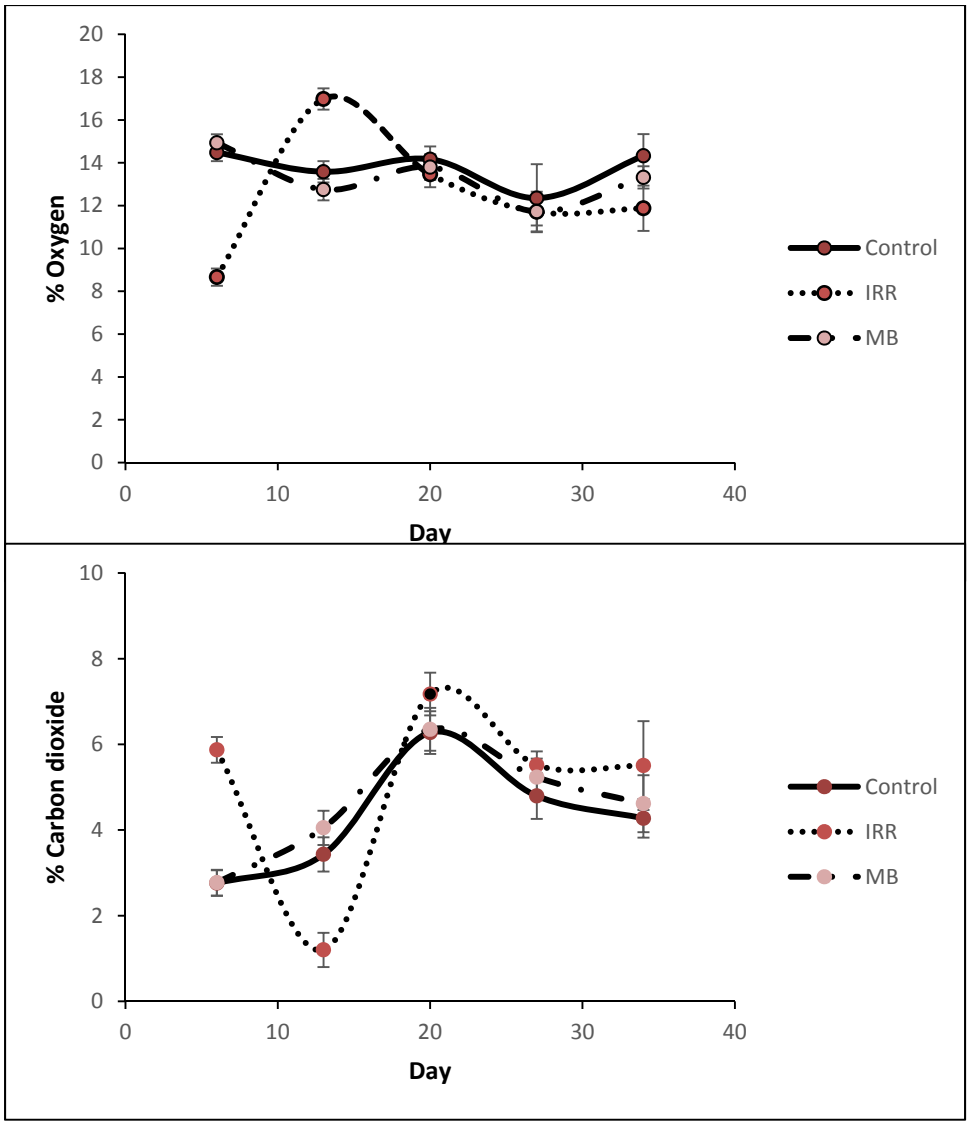
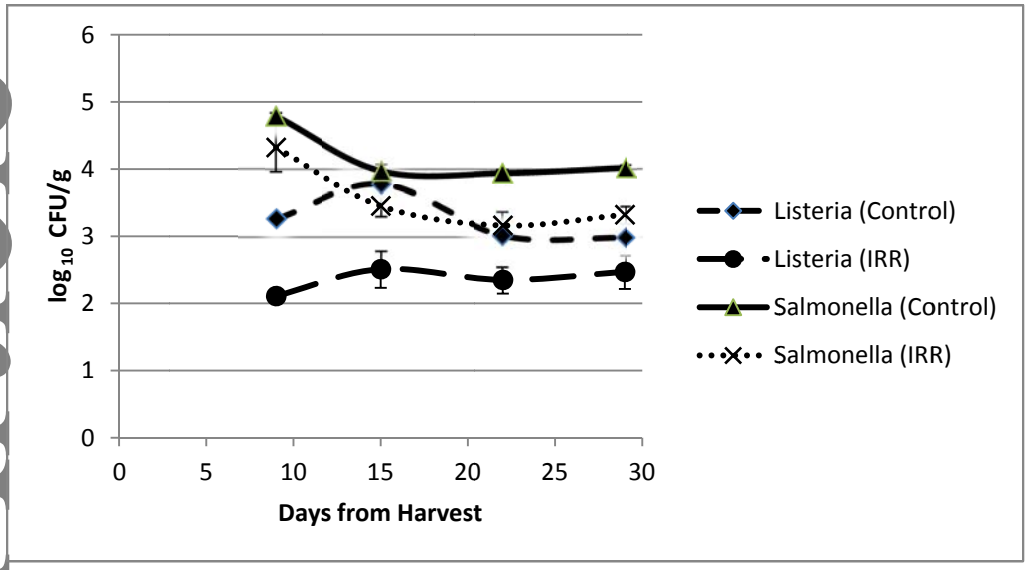


Figure 4 – Change in O_2 and CO_2 concentrations during storage of sweet cherries bulk packaged in liner bags.



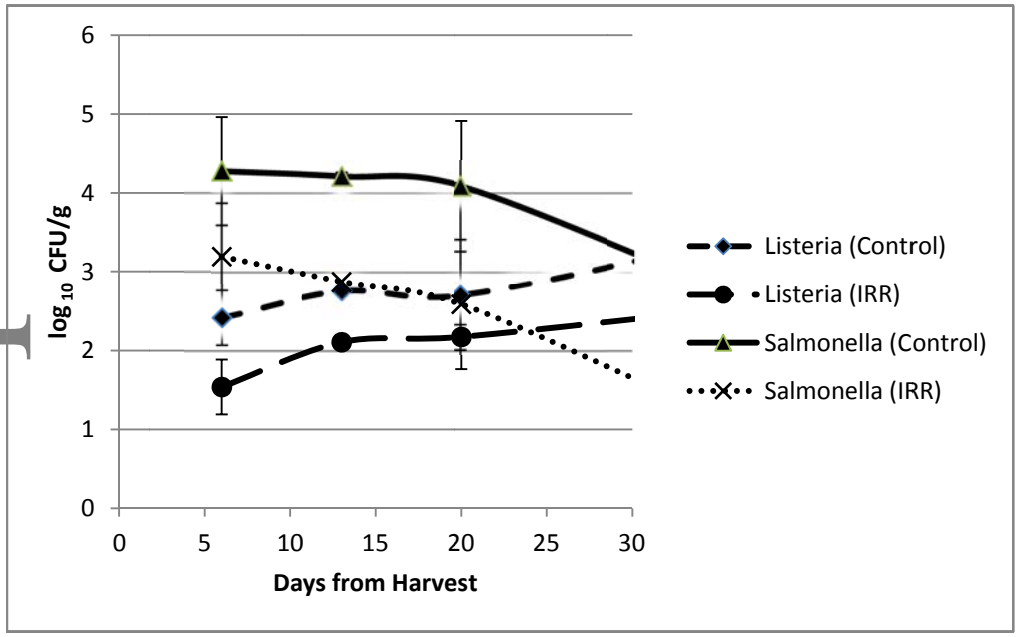


Figure 5 – Effect of irradiation at 0.4 kGy on survival of *L.monocytogenes* and *Salmonella* spp. on A. blueberries and B. sweet cherries during storage.