Effect of Low-dosage Electron Beam Irradiation on Antioxidant Activities of Navel Oranges during Storage at a Low Temperature of 3°C

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Abstract Effects of low-dosage electron beam irradiation on antioxidant activities of Navel oranges during storage at a low temperature of 3°C were studied. Oranges were irradiated at dosages of 0.2, 0.4, 0.6, 0.8, and 1.0 kGy and changes in antioxidant compounds and antioxidant activities were investigated. No changes in total phenolic contents or flavonoid contents were observed with an increase in radiation dosage. Also, no differences between non-irradiated and irradiated oranges in DPPH radical scavenging and ABTS radical scavenging activities, FRAP values, and reducing powers were observed. Electron beam irradiation at dosages less than 1 kGy does not affect levels of antioxidant compounds and antioxidant activities of Navel oranges.

Keywords: electron-beam, irradiation, antioxidant, orange

Introduction

Oranges are popular fruits with strong antioxidant properties from high levels of phytochemicals, such as vitamin C, flavonoids, and carotenoids. However, high moisture and sugar contents make oranges vulnerable to insect attack. Thus, removal of insects is necessary to extend product shelf life for higher marketability (1). Fumigation with organic chemicals has been used for post-harvest control of insects, but there is concern about residues and negative effects on human health (2). Moreover, use of methyl bromide has been recommended for reduction because of environmental issues (3).

Irradiation at dosages below 0.5 kGy is effective for insect and parasite disinfection against different species (4). Several studies reported that low dosage irradiation can inhibit fecundity and egg hatchability of fruit insects, and dosage levels of 0.5 to 1.0 kGy are sufficient to eliminate most insects on products. Low-dosage irradiation at less than 400 Gy has been authorized as a feasible method to achieve insect disinfestation by the international organizations: United Nations Food and Agriculture Organization (FAO), International Atomic Energy Agency (IAEA) and World Health Organization (WHO) with no creation of harmful substances (5,6). Generally, gamma rays are used in food irradiation, but there is a possible limit to acceptance by consumers due to perceptions of radioactivity. On the other hand, electron beams use only energy from electrons and do not create any residues. Moreover, electron beam irradiation uses a conveyor system that can shorten processing

times, so research into electron beam irradiation of foods as a practical food irradiation technology has increased (7). However, much research has focused on quality and microbiological properties and there are only a few studies related to biological and phytochemical levels, even with increasing interest in the effects of such treatments in the food industry. Treatment of foods can change food textures and also constituent compounds in either positive or negative ways, which is an important issue for customers in choosing food products. Hence, this study investigated effects of low-dosage electron beam irradiation on biological functionality based on determination of antioxidant compounds and antioxidant activities during storage at a low temperature of 3°C.

Materials and Methods

Oranges and irradiation treatments Fresh oranges (cultivar: Navel, *Citrus sinensis*) of 7 to 8.5 cm diameter harvested in California were obtained from a retail supermarket in Daejeon, Korea in February of 2014. Fruits were packed in polyethylen film (Homeplus Polyethylene, Hwaseong, Korea), and kept in a refrigerator at 3°C for 1 day before irradiation.

Packed oranges at 6 per pack were placed in molded pulp packaging fruit trays. Oranges were then irradiated at room temperature (20±5°C) at 10 MeV×0.5 mA using an electron beam accelerator (EB-Tech. Co., Daejeon, Korea) located at the Korea Atomic Energy

Research Institute, Jeongeup, Korea. Type electron beam irradiation is a single round-trip along the length and conveyor speed was 24 m/min.

Oranges were irradiated using a double-beam that ensured penetration of beam energy to the fruit center. Five mm alanine dosimeters (Bruker Instruments, Rheinstetten, Germany) were placed with oranges and the free radical signal was measured using a Bruker EMS 104 EPR Analyzer for determination of the applied dosage. Dosimeters were placed on the top, bottom, and center of orange surface. Three oranges were used for measurement of radiation dosages with measured dosages of 0.2, 0.4, 0.6, 0.8, and 1 kGy, respectively. Non-irradiated oranges served as controls. Differences in dosimeters readings were small and the dosage absorbed was similar on both the fruit top and bottom. Fruits were exposed to a double upper and lower beam, so a high-dosage region was located at the middle-side of oranges. In the fruit middle, absorbed dosages were higher due to overlapping high dosage areas of the 2 single beams. However, the uniformity ratio of maximum to minimum absorbed dosages in a process ($D_{\max}/D_{\min}$) was acceptable at 1.6 in this study as the acceptance range of uniformity ratio values for an electron beam is between 1.5 and 2, or even higher (8).

Oranges were stored at $3\pm 2^{\circ}\text{C}$ and all orange samples were evaluated at day 0 (irradiation day), and days 15, 30, 37, and 45 after irradiation. Oranges was squeezed using a household electric juice squeezer (MSJ-100M; Tongyang/Magic, Seoul, Korea), immediately chilled to -70°C , then freeze-dried in a lyophilizer (SFDSM12-60Hz; Samwon, Seoul, Korea) after (No.2, diameter 8 μm , WhatmanTM, Maidstone, UK). Freeze-dried orange powder was diluted in each way with distilled water and used for each measurement.

Total phenolic contents (TPC) Total phenolic contents of samples were determined following the method of Folin-Ciocalteu (9). An orange juice sample of 0.2 mL was mixed with 0.2 mL of Folin-Ciocalteu phenol reagent (Sigma-Aldrich, St. Louis, MO, USA) and left at room temperature for 3 min. After 3 mL of 10% sodium carbonate (Samchun Pure Chemical Co., Ltd., Anyang, Korea) was added, the mixture was left to stand for 1 h in the dark. The absorbance was measured at 765 nm using a UV spectrophotometer (UV-1800; Shimadzu Corp., Kyoto, Japan). Total phenolic contents were calculated on the basis of a calibration curve using a gallic acid (Sigma-Aldrich) standard.

Flavonoid contents Total flavonoid contents of samples were determined following a partly modified method of Zhishen *et al.* (10). A 250 μL of sample was diluted in 1 mL of distilled water, then 75 μL of 5% sodium nitrite (Samchun Pure Chemical Co., Ltd.) was added. After 5 min at room temperature, 150 μL of 10% aluminium chloride (Junsei Chemical Co., Ltd., Tokyo, Japan) was added, followed by standing for 6 min. Then, 500 μL of sodium hydrate (Samchun Pure Chemical Co., Ltd.) was added and left for 11 min, then the absorbance was measured at 517 nm using a UV

spectrophotometer (UV-1800; Shimadzu Corp.). Total flavonoid contents were calculated on the basis of a calibration curve for the standard naringin (Sigma-Aldrich).

DPPH radical scavenging activity The DPPH radical scavenging activity was determined following the method described by Blois (11). One mL of 0.2 mM DPPH (Sigma-Aldrich) in methanol (Samchun Pure Chemical Co., Ltd.) was added to 1 mL of different concentrations of samples, followed by mixing for 10 s. Mixtures were left to stand for 30 min in the dark, then the absorbance was measured at 517 nm using a UV spectrophotometer (UV-1800; Shimadzu Corp.). EC_{50} values based on mg of fresh weight (FW)/mL were effective concentrations at which DPPH radicals were scavenged by 50%.

ABTS radical scavenging activity ABTS radical scavenging activities were determined following the method of Fellegrini *et al.* (12). Five mL of 7 mM ABTS (Sigma-Aldrich) and 88 μL of 140 mM potassium peroxodisulfate (Samchun Pure Chemical Co., Ltd.) were mixed and left in the dark to stand for 14 to 16 h. Then, the solution was mixed with ethanol (Samchun Pure Chemical Co., Ltd., 1:88 v/v in water) and the absorbance was adjusted to 0.70 ± 0.02 at 734 nm determined using a UV spectrophotometer (UV-1800; Shimadzu Corp.). One mL of ABTS solution was added to 50 μL of different concentrations of samples and mixed for 30 s. The mixture was left for 2 min and 30 s in the dark and the absorbance was measured at 734 nm using a UV spectrophotometer (UV-1800; Shimadzu Corp.). EC_{50} values as mg of raw weight/mL were effective concentrations at which ABTS radicals were scavenged by 50%.

Ferric reducing antioxidant power (FRAP assay) Ferric-reducing antioxidant potential values were determined following the method of Benzie and Strain (13). FRAP reagent was prepared using a 300 mM acetate buffer (Samchun Pure Chemical Co., Ltd.) at pH 3.6, 10 mM 2, 4, 6-tripyridyl-s-triazine (Sigma-Aldrich) in 40 mM HCl, (Samchun Pure Chemical Co., Ltd.) and 20 mM ferric chloride hexahydrate (Samchun Pure Chemical Co., Ltd.) at volume ratios of 10:1:1, then 30 μL of a sample was mixed with 900 μL of FRAP reagent and 90 μL of water, followed by incubation (IB-E; Jeio Tech, Daejeon, Korea) at 37°C for 10 min. Absorbance values were then measured at 593 nm using a UV spectrophotometer (UV-1800; Shimadzu Corp.) FRAP values were calculated on the basis of an FeSO_4 (Samchun Pure Chemical Co., Ltd.) calibration curve at different concentrations.

Reducing power Reducing powers were determined following the method of Oyaizu (14) using 250 μL of a 200 mM sodium phosphate buffer (Sigma-Aldrich) at pH 6.6 and 250 μL of 1% potassium ferric cyanide (Samchun Pure Chemical Co., Ltd.) added to 250 μL of different concentrations of samples. Then, mixtures were incubated (IB-E; Jeio Tech) at 50°C for 20 min. After cooling of incubated mixtures on ice, trichloroacetic acid (10% w/v, Samchun Pure Chemical Co.,

Ltd.) was added and mixtures were centrifuged (Supra 30K; Hanil Science Industrial, Incheon, Korea) at $8,293\times g$ for 8 min. Upper layers (0.5 mL) of mixtures were mixed with 0.5 mL of water and 50 μL of 0.1% ferric chloride (Samchun Pure Chemical Co., Ltd.), and the absorbance was measured at 700 nm using a UV spectrophotometer (UV-1800; Shimadzu Corp.). EC_{50} values as mg of FW/mL represented effective concentrations at which the absorbance was 0.5 measured using a UV spectrophotometer (UV-1800; Shimadzu Corp.) as an indicator of reducing power.

Statistical analysis All experiments were conducted at least in triplicate for which mean values and standard deviations were determined. Also, experimental data were analyzed using the Statistical Package of Social Science (SPSS) Software (SPSS 21.0 for Windows, SPSS Inc., Chicago, IL, USA). Data were also analyzed using a one-way analysis of variance (ANOVA) followed by Duncan multiple range test and expressed as mean values $\pm$ standard deviations (SD). Significance was defined at $p < 0.05$.

Results and Discussion

Total phenolic contents Plants contain many phenolic compounds and contained phenolic hydroxyl groups exhibit different biological

properties, such as antioxidant, anti-obesity, and anti-inflammatory activities (15). The Folin-Ciocalteu method uses colorimetry regarding oxidation-reduction reactions of polyphenols from plants, allowing easy and rapid quantification of total levels of phenolic compounds based on visible-light spectrophotometry (16). TPC levels of non-irradiated and irradiated oranges significantly ($p < 0.05$) increased with the storage period, compared to irradiation day 0, regardless of the radiation dosage (Table 1).

Polyphenols from irradiated plants can be degraded due to elimination of free radicals and reactive oxygen species produced by irradiation, or polyphenol levels can be increased due to breakage of polyphenol chemical bonds to produce low molecular weight fragments (17,18). There was no significant ($p > 0.05$) difference between TPC levels and irradiation dosage, compared with controls. Therefore, low-dosage electron beam irradiation did not affect TPC levels. Kim *et al.* (19) reported that TPC levels of flowering cherry were not altered, and Bae *et al.* (20) reported that rice bran and barley bran were not changed by gamma irradiation treatment.

Flavonoid contents Several plant flavonoids exert protective antioxidants effects against external stress or disease (21). *Citrus* fruits contain more than 60 kinds of flavonoids and over 90% are hesperidin and naringin (22,23). In this study, there were no significant ($p < 0.05$) relationships between irradiation dosage and

Table 1. Changes in total phenolic contents of oranges during storage at $3 \pm 2^\circ\text{C}$ for 45 days after electron beam irradiation
(Unit: mg of GAE¹⁾/100 g of FW)

Irradiation dosage (kGy)	Storage period (days)				
	0	15	30	37	45
0	114.58 ^c $\pm$ 0.54 ²⁾³⁾	133.23 ^{aB} $\pm$ 0.50	136.16 ^{bB} $\pm$ 4.18	134.65 ^{cB} $\pm$ 1.53	142.85 ^{bA} $\pm$ 0.17
0.2	132.04 ^{dD} $\pm$ 0.08	131.63 ^{aD} $\pm$ 0.25	135.54 ^{bC} $\pm$ 0.99	138.35 ^{bB} $\pm$ 0.71	141.40 ^{bA} $\pm$ 0.19
0.4	115.60 ^{cD} $\pm$ 1.51	124.69 ^{bB} $\pm$ 0.64	121.27 ^{dC} $\pm$ 0.38	143.47 ^{aA} $\pm$ 0.84	144.90 ^{bA} $\pm$ 4.46
0.6	125.28 ^{bC} $\pm$ 0.62	119.39 ^{cD} $\pm$ 3.14	127.30 ^{cC} $\pm$ 1.74	130.91 ^{dB} $\pm$ 0.63	149.73 ^{aA} $\pm$ 1.02
0.8	116.72 ^{cD} $\pm$ 3.21	130.86 ^{aC} $\pm$ 1.73	133.36 ^{bC} $\pm$ 0.79	140.17 ^{bB} $\pm$ 0.89	144.50 ^{bA} $\pm$ 1.65
1.0	114.90 ^{cD} $\pm$ 0.35	129.74 ^{aC} $\pm$ 2.73	140.64 ^{aA} $\pm$ 3.23	130.93 ^{dC} $\pm$ 1.31	136.29 ^{cB} $\pm$ 0.69

¹⁾GAE, gallic acid equivalents

^{2)a-d}Values with different letters within a column differ significantly ($p < 0.05$).

^{A-D}Values with different letters within a row differ significantly ($p < 0.05$).

³⁾Means $\pm$ SD ($n=3$).

Table 2. Changes in flavonoid contents of oranges during storage at $3 \pm 2^\circ\text{C}$ for 45 days after electron beam irradiation
(Unit: mg of naringin/100 g of FW)

Irradiation dosage (kGy)	Storage period (days)				
	0	15	30	37	45
0	9.87 ^{cD} $\pm$ 0.17 ¹⁾²⁾	12.07 ^{aC} $\pm$ 0.28	12.07 ^{bC} $\pm$ 0.21	12.63 ^{bB} $\pm$ 0.10	13.09 ^{cA} $\pm$ 0.36
0.2	12.14 ^{aB} $\pm$ 0.24	11.87 ^{aC} $\pm$ 0.01	11.69 ^{cC} $\pm$ 0.05	11.91 ^{cB} $\pm$ 0.14	13.36 ^{cA} $\pm$ 0.10
0.4	11.18 ^{bC} $\pm$ 0.17	12.56 ^{aB} $\pm$ 1.56	12.58 ^{aB} $\pm$ 0.03	13.25 ^{aB} $\pm$ 0.06	14.61 ^{aA} $\pm$ 0.23
0.6	11.87 ^{aC} $\pm$ 0.13	11.79 ^{aC} $\pm$ 0.17	11.72 ^{cC} $\pm$ 0.14	12.36 ^{bB} $\pm$ 0.38	13.73 ^{bA} $\pm$ 0.13
0.8	11.90 ^{aE} $\pm$ 0.18	12.28 ^{aD} $\pm$ 0.20	12.61 ^{aC} $\pm$ 0.08	13.04 ^{aB} $\pm$ 0.21	13.94 ^{bA} $\pm$ 0.11
1.0	11.41 ^{bC} $\pm$ 0.13	12.56 ^{aA} $\pm$ 0.33	12.73 ^{aA} $\pm$ 0.15	11.99 ^{cB} $\pm$ 0.15	12.60 ^{dA} $\pm$ 0.09

^{1)a-d}Values with different letters within a column differ significantly ($p < 0.05$).

^{A-E}Values with different letters within a row differ significantly ($p < 0.05$).

²⁾Means $\pm$ SD ($n=3$)

flavonoid contents, compared with controls, throughout the storage period (Table 2). Some studies have suggested that more 1 kGy of ionizing radiation can induce degradation of phenolic compounds (24). However, Yun *et al.* (25) reported that irradiated apples did not show significant differences in flavonoid contents, compared with non-irradiated apples. Kwon *et al.* (26) also found that there was no significant effect of irradiation on flavonoid contents of fresh vegetable juice. Thus, an increase in flavonoid contents was more affected by storage time than by the radiation dosage.

DPPH and ABTS radical scavenging activities DPPH scavenging and ABTS scavenging activity changes in orange juice are shown in Table 3 and 4. The DPPH radical scavenging activity showed no trend over the storage period. There was a slight increase in activity at day 45, compared to irradiation day 0 ($p < 0.05$). The ABTS radical scavenging activity decreased with the storage period. On the other hand, the radiation dosage did not affect either DPPH or ABTS radical scavenging activities during storage with irregular IC_{50} levels during storage, in agreement with Kim *et al.* (27) who reported that irradiated kiwi fruits exposed to low-dosage electron beam irradiation showed no significant changes in radical scavenging activities. Jo *et al.* (28) also found that grape by-products irradiated with up to 3 kGy did not show any increase or decrease in scavenging activities, and Jeong (29) reported no difference in DPPH radical scavenging activities of pepper powder with irradiation.

FRAP value and reducing power The FRAP assay is based upon reduction of a ferric tripyridyltriazine (Fe^{3+} -TPTZ) complex to the ferrous tripyridyltriazine (Fe^{2+} -TPTZ) state by a reductant at low pH (22). Oyaizu's method (14) used in this study is also based on potassium ferricyanide (Fe^{3+}) reactive substances to form potassium ferrocyanide (Fe^{2+}), which then reacts with ferric chloride to form a ferric-ferrous complex. Both methods are indicators of the reductive ability of antioxidants based on a principle different from the radical scavenging activity.

FRAP values of oranges decreased gradually with storage, but there was no trend for irradiation dosage (Table 5). Low-dosage irradiation did not affect the FRAP reducing power (Table 6). The reductive ability of irradiated oranges initially decreased slightly during storage, but a gap in FRAP values between non-irradiated and irradiated oranges decreased by the end of storage ($p < 0.05$). Generally, irradiation leads to an increase in reducing powers of food products (19,30,31). However, Kim *et al.* (32) and Lee *et al.* (33) showed that irradiation treatments did not produce changes in the reducing power of *Eriobotrya japonica* extracts and tamarind juice. Lee *et al.* (34) suggested that the reducing power of the disaccharide sucrose was produced by gamma irradiation due to generation of radiolytic end-products. Thus, the reducing power of fruits might be represented by the composition of sugar contents. Castell-Perez *et al.* (35) suggested that low-dosage electron beam irradiation caused no significant changes in fruit sugar contents. Therefore, low-dosage

Table 3. Changes in DPPH radical scavenging activities of oranges during storage at $3 \pm 2^\circ\text{C}$ for 45 days after electron beam irradiation

(Unit: mg/mL)

Irradiation dosage (kGy)	Storage period (days)				
	0	15	30	37	45
0	$2.42^{cC} \pm 0.09^{1)2)}$	$2.92^{aB} \pm 0.05$	$2.56^{dC} \pm 0.05$	$3.38^{cA} \pm 0.16$	$2.87^{dB} \pm 0.11$
0.2	$2.47^{cC} \pm 0.21$	$2.99^{aB} \pm 0.07$	$2.48^{eC} \pm 0.01$	$2.88^{dB} \pm 0.05$	$3.39^{bA} \pm 0.03$
0.4	$2.81^{bB} \pm 0.19$	$2.53^{bC} \pm 0.09$	$3.16^{aA} \pm 0.04$	$3.30^{cA} \pm 0.22$	$3.31^{bA} \pm 0.07$
0.6	$3.06^{bBC} \pm 0.09$	$2.99^{aC} \pm 0.04$	$2.76^{dD} \pm 0.03$	$3.93^{aA} \pm 0.04$	$3.14^{cB} \pm 0.08$
0.8	$3.42^{aB} \pm 0.27$	$2.96^{aC} \pm 0.08$	$2.86^{bC} \pm 0.02$	$3.94^{aA} \pm 0.02$	$3.38^{bB} \pm 0.03$
1.0	$2.88^{bC} \pm 0.11$	$2.42^{dD} \pm 0.04$	$2.85^{bC} \pm 0.06$	$3.60^{bB} \pm 0.05$	$4.07^{aA} \pm 0.12$

^{1) a-e}Values with different letters within a column differ significantly ($p < 0.05$).

^{A-D}Values with different letters within a row differ significantly ($p < 0.05$).

²⁾ Means $\pm$ SD ($n=3$)

Table 4. Changes in ABTS radical scavenging activities of oranges during storage at $3 \pm 2^\circ\text{C}$ for 45 days after electron beam irradiation

(Unit: mg/mL)

Irradiation dosage (kGy)	Storage period (days)				
	0	15	30	37	45
0	$8.16^{bCB} \pm 0.14^{1)2)}$	$8.66^{aA} \pm 0.22$	$8.58^{aAB} \pm 0.31$	$8.70^{aA} \pm 0.33$	$7.92^{aC} \pm 0.21$
0.2	$9.02^{aA} \pm 0.22$	$8.18^{bB} \pm 0.14$	$7.52^{dC} \pm 0.19$	$8.32^{aB} \pm 0.33$	$7.67^{aC} \pm 0.10$
0.4	$8.44^{bA} \pm 0.13$	$7.52^{dC} \pm 0.09$	$7.60^{dBC} \pm 0.09$	$7.71^{bB} \pm 0.06$	$6.99^{bD} \pm 0.03$
0.6	$7.83^{cB} \pm 0.18$	$7.68^{dCB} \pm 0.15$	$8.00^{bCB} \pm 0.33$	$8.61^{aA} \pm 0.44$	$6.53^{cC} \pm 0.03$
0.8	$9.11^{aA} \pm 0.30$	$7.93^{bBC} \pm 0.19$	$8.35^{aBB} \pm 0.25$	$7.52^{bC} \pm 0.05$	$6.57^{dD} \pm 0.43$
1.0	$8.83^{aA} \pm 0.22$	$7.83^{cC} \pm 0.13$	$8.36^{aBB} \pm 0.21$	$7.46^{bD} \pm 0.11$	$7.15^{bD} \pm 0.16$

^{1) a-d}Values with different letters within a column differ significantly ($p < 0.05$).

^{A-D}Values with different letters within a row differ significantly ($p < 0.05$).

²⁾ Means $\pm$ SD ($n=3$)

Table 5. Changes in ferric reducing antioxidant potential (FRAP) values of oranges during storage at 3±2°C for 45 days after electron beam irradiation (Unit: mM)

Irradiation dosage (kGy)	Storage period (days)				
	0	15	30	37	45
0	7.22 ^{cC} ±0.34 ¹⁾²⁾	8.37 ^{aA} ±0.13	7.75 ^{abB} ±0.04	6.84 ^{cD} ±0.05	7.65 ^{aB} ±0.06
0.2	8.74 ^{aA} ±0.06	8.23 ^{abB} ±0.01	8.24 ^{abB} ±0.16	7.62 ^{aC} ±0.06	6.91 ^{bD} ±0.05
0.4	7.42 ^{cAB} ±0.13	8.01 ^{bA} ±0.09	6.75 ^{cC} ±0.70	7.33 ^{bBC} ±0.12	6.92 ^{bBC} ±0.11
0.6	8.24 ^{bA} ±0.10	7.53 ^{cB} ±0.09	7.07 ^{bcC} ±0.22	6.56 ^{ed} ±0.02	6.44 ^{dD} ±0.02
0.8	7.14 ^{cAB} ±0.09	7.39 ^{dA} ±0.04	7.51 ^{bA} ±0.50	6.68 ^{dC} ±0.03	6.85 ^{bcBC} ±0.03
1.0	8.36 ^{abA} ±0.36	7.94 ^{bbB} ±0.04	7.71 ^{abB} ±0.21	6.35 ^{fd} ±0.06	6.78 ^{cc} ±0.06

^{1)a-f}Values with different letters within a column differ significantly ($p<0.05$).

^{A-D}Values with different letters within a row differ significantly ($p<0.05$).

²⁾Means±SD ($n=3$)

Table 6. Changes in reducing power of oranges during storage at 3±2°C for 45 days after electron beam irradiation (Unit: mg/mL)

Irradiation dosage (kGy)	Storage period (days)				
	0	15	30	37	45
0	6.01 ^{dA} ±0.06 ¹⁾²⁾	5.09 ^{dC} ±0.05	5.94 ^{bA} ±0.09	5.23 ^{cB} ±0.03	5.03 ^{bC} ±0.09
0.2	5.91 ^{dB} ±0.15	6.43 ^{bA} ±0.05	5.78 ^{bbB} ±0.20	5.29 ^{cC} ±0.20	5.22 ^{bC} ±0.18
0.4	7.02 ^{cA} ±0.11	5.46 ^{cc} ±0.11	5.86 ^{bbB} ±0.04	4.82 ^{dD} ±0.10	4.97 ^{bD} ±0.23
0.6	7.65 ^{aA} ±0.14	6.47 ^{bbB} ±0.11	5.74 ^{bcC} ±0.17	5.67 ^{bc} ±0.14	4.25 ^{cd} ±0.13
0.8	7.63 ^{aA} ±0.11	6.66 ^{abB} ±0.14	6.50 ^{abB} ±0.19	4.94 ^{dC} ±0.15	5.02 ^{bC} ±0.08
1.0	7.26 ^{bA} ±0.09	6.59 ^{abB} ±0.07	5.34 ^{cd} ±0.17	6.14 ^{aC} ±0.15	5.54 ^{ad} ±0.03

^{1)a-d}Values with different letters within a column differ significantly ($p<0.05$).

^{A-D}Values with different letters within a row differ significantly ($p<0.05$).

²⁾Means±SD ($n=3$)

electron beam irradiation of oranges might not induce changes in the reducing power.

Exposure to low-dosage electron beam irradiation up to 1.0 kGy might have no effect on orange TCP and flavonoid contents. Also, there were no significant ($p<0.05$) differences between non-irradiated and irradiated oranges in DPPH radical scavenging and ABTS radical scavenging activities, FRAP values, and reducing powers. Thus, electron beam irradiation up to 1 kGy did not affect the level of antioxidant compounds and antioxidant activities of Navel oranges at low temperature.

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References

- McDonald H, Arpaia ML, Caporaso F, Obenland D, Were L, Rakovski C, Prakash A. Effect of gamma irradiation treatment at phytosanitary dose levels on the quality of 'Lane Late' navel oranges. *Postharvest Biol. Tec.* 86: 91-99 (2013)
- Park MG, Sung BK, Cho JY. Residual characteristics of methyl bromide and hydrogen cyanide in banana, orange, and pineapple. *J. Appl. Biol. Chem.* 54: 214-217 (2011)
- MBTOC. Reports of the methyl bromide technical options committee (MBTOC). 2006 Assessment of alternatives to methyl bromide. UNEP, Nairobi (2006)
- Frazier WC, Westhoff DC. Preservation by radiation. pp. 159-170. In: *Food Microbiology*. 4th ed. McGraw-Hill, New York, NY, USA (1988)
- World Health Organization. Wholesomeness of Irradiated Food: Report of a Joint FAO/IAEA/WHO Expert Committee. Technical Report Series, Geneva, Switzerland, pp. 7-34 (1981)
- Kume T, Furuta M, Todoriki S, Uenoyama N, Kobayashi Y. Status of food irradiation in the world. *Radiat. Phys. Chem.* 78: 222-226 (2009)
- Kwon JH. Safety and Understanding of Irradiated Food. Korea Food Safety Research Institute, Seoul, Korea. pp. 9-29 (2010)
- IAEA. Techniques for high dose dosimetry in industry, agriculture and medicine. Proceedings of a Symposium. IAEA, Vienna. p. 233 (1998)
- Folin O, Denis W. On phosphotungstic-phospho-molybdic compounds as color reagents. *J. Biol. Chem.* 12: 239-249 (1912)
- Zhishen J, Mengcheng T, Jianming W. The determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals. *Food Chem.* 64: 555-559 (1999)
- Blois MS. Antioxidant determinations by the use of a stable free radical. *Nature* 181: 1199-1200 (1958)
- Fellegrini N, Roberta R, Min Y, Catherine RE. Screening of dietary carotenoids and carotenoid-rich fruit extract for antioxidant activities applying 2,2'-azino bis (3-ethylenbenzothiazolin-6-sulfonic acid) radical cation decolorization assay. *Method. Enzymol.* 299: 379-389 (1998)
- Benzie IFF, Strain JJ. The ferric reducing ability of plasma (FRAP) as a measure of "Antioxidant power": The FRAP assay. *Anal. Biochem.* 239: 70-76 (1996)
- Oyaizu M. Studies on products of browning reaction--anti-oxidative activities of products of browning reaction prepared from glucosamine. *Jpn. J. Nutr.* 44: 307-315 (1986)
- Lee S, Moon HK, Lee SW, Moon JN, Kim JK. Effects of drying methods on quality characteristics and anti-oxidative effects of Omija (*Schizandra chinensis* bailon). *Korean J. Food Preserv.* 21: 341-349 (2014)
- Lee HR, Jung BR, Park JY, Hwang IW, Kim SK, Choi JU, Lee SH, Chung SK. Antioxidant activity and total phenolic contents of grape juice products in Korean market. *Korean J. Food Preserv.* 15: 445-449 (2008)
- Song EJ, Lee SY, Kim KBWR, Park JK, Kim JH, Lee JW, Byun MW, Ahn DH. Effect of gamma irradiation on the physical properties of alginic acid and λ -carrageenan. *J. Korean Soc. Food Sci. Nutr.* 36: 902-907 (2007)
- Urban, WM. Food irradiation. 1st ed. Academic Press, London, England. pp. 83-117 (1986)
- Kim KH, Jo JE, Lee SA, Yook HS. Anti-oxidative properties of ethanolic extracts

- from flowering cherry (*Prunus serrulata* L. var. *spontanea* Max. wils.) fruit with various doses of gamma radiation. J. Korean Soc. Food Sci. Nutr. 40: 1378-1383 (2011)
20. Bae SM, Kim JH, Cho CW, Jeong TJ, Yook HS, Byun MW, Lee SC. Effect of γ -Irradiation on the antioxidant activity of rice hull, rice bran, and barley bran. J. Korean Soc. Food Sci. Nutr. 31: 246-250 (2002)
 21. Dixon RA, Achnine L, Kota P, Liu CJ, Reddy MS, Wang L. The phenylpropanoid pathway and plant defence-A genomics perspective. Mol. Plant Pathol. 3: 371-390 (2002)
 22. Monforte MT, Trovato A, Kirjavainen S, Forestieri AM, Galati EM, Lo Curto RB. Biological effects of hesperidin, a *Citrus* flavonoid. (note II): Hypolipidemic activity on experimental hypercholesterolemia in rat. Farmaco 50: 595-599 (1995)
 23. Bok SH, Lee SH, Park YB, Bae KH, Son KH, Jeong TS, Choi MS. Plasma and hepatic cholesterol and hepatic activities of 3-hydroxy-3-methyl-glutaryl-CoA reductase and acyl CoA: Cholesterol transferase are lower in rats fed citrus peel extract or a mixture of citrus bioflavonoids. J. Nutr. 129: 1182-1185 (1999)
 24. Ahn HJ, Kim JH, Kim JK, Kim DH, Yook HS, Byun MW. Combined effects of irradiation and modified atmosphere packaging on minimally processed Chinese cabbage (*Brassica rapa* L.). Food Chem. 89: 589-597 (2005)
 25. Yun HJ, Lim SY, Hur JM, Jeong JW, Yang SH, Kim DH. Changes of functional compounds in, and texture characteristics of, apples, during post-irradiation storage at different temperatures. Korean J. Food Preserv. 14: 239-246 (2007)
 26. Kwon SC, Jo C, Lee KH. Gamma irradiation for sanitation of vegetable fresh juice containing non-thermal process materials. J. Korean Soc. Food Sci. Nutr. 38: 964-969 (2009)
 27. Kim KH, Kwon JS, Lee JO, Park SH, Yook HS, Lee BC. Physicochemical changes of electron beam-irradiated Korean kiwifruits at low dose levels. J. Korean Soc. Food Sci. Nutr. 36: 603-608 (2007)
 28. Jo JE, Yook HS, Kim KH, Baek JY, Moon YJ, Park SJ, Jang SA. Effect of drying methods and gamma irradiation on the color changes and antioxidant activity of grape by-products. J. Korean Soc. Food Sci. Nutr. 39: 1826-1831 (2010)
 29. Jeong MS. Microbial reduction and quality stability of electron-beam irradiated red pepper and cinnamon powders. MS Thesis, Kyungpook National University, Daegu, Korea (2013)
 30. Oufedjikh H, Mahrouz M, Amiot MJ, Lacroix M. Effect of γ -irradiation on phenolic compounds and phenylalanine ammonia-lyase activity during storage in relation to peel injury from peel of *Citrus clementina* Hort. Ex. Tanaka. J. Agr. Food Chem. 48: 559-565 (2000)
 31. Song HP, Kim BD, Shin EH, Song DS, Lee HJ, Kim DH. Effects of gamma irradiation on the microbiological and general quality characteristics of fresh yam juice. Korean J. Food Preserv. 17: 494-499 (2010)
 32. Kim HJ, Jo C, Kim TH, Kim DS, Park MY, Byun MW. Biological evaluation of the methanolic extract of *Eriobotrya japonica* and its irradiation effect. Korean J. Food Sci. Technol. 38: 684-690 (2006)
 33. Lee JW, Kim JK, Srinivasan P, Choi JI, Kim JH, Han SB, Kim DJ, Byun MW. Effect of gamma irradiation on microbial analysis, antioxidant activity, sugar content and color of ready-to-use tamarind juice during storage. LWT-Food Sci. Technol. 42: 101-105 (2009)
 34. Lee JW, Oh SH, Kim JH, Byun EH, Kim MR, Kim KS, Lee HJ, Byun MW. The non-enzymatic browning reaction occurred by gamma irradiation in sugar-Lysine aqueous model solution. J. Korean Soc. Food Sci. Nutr. 35: 583-587 (2006)
 35. Castell-Perez E, Moreno M, Rodriguez O, Moreira RG. Electron beam irradiation treatment of cantaloupes: Effect on product quality. Food Sci. Technol. Int. 10: 383-390 (2004)