

Communication

Temperature determines post-harvest quality of spinach leaves after gamma irradiation

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ABSTRACT: The relative importance of radiation dose, storage time, and temperature in radiation processing of spinach (*Spinacia oleracea* L.), was evaluated in terms of the postharvest quality through a model study using leaf disks. Physiological activity and chlorophyll and carotenoid contents were measured to represent the postharvest quality (in terms of external appearance) of spinach, a leafy green vegetable. At 22°C and 30°C, the higher gamma-radiation dose caused a greater decrease in the physiological activity, depending on the storage time of 4 d. However, this decrease was not significant at 4°C and 15°C. Total chlorophyll and carotenoid contents were substantially decreased by 3 kGy at 15°C, and dose-dependently by 0.5–3 kGy at 22°C. In contrast, the proportion of lutein in total carotenoid was significantly increased in the 2–3-kGy samples only at 22°C, while that of β -carotene was slightly decreased, indicating progression of leaf aging. These data suggest that the unfavorable effect of gamma irradiation on the postharvest quality of spinach could be avoided or controlled by the storage time or temperature rather than the radiation dose. The current study could be available to improve radiation processing of other leafy green vegetables.

Ionizing radiation such as X-rays, gamma rays, and electron beams can be used to improve postharvest quality of fresh fruits and vegetables.¹ Currently, a major use of irradiation to agricultural commodities is as a phytosanitary treatment to prevent the spread of invasive species and/or to overcome quarantine barrier to trade.^{2,3} In phytosanitary or postharvest irradiation treatments, researches have been focused on various factors affecting irradiation efficacy, such as low oxygen, dose rate, host, and temperature.^{2,4,5} Irradiation

of various fresh-cut vegetables was effective to eliminate bacteria or human pathogens such as *E.coli* O157:H7, but it could cause a substantial loss of vitamin C, depending on the vegetable, radiation dose, and storage time.^{4,6} This unfavorable effect of irradiation deteriorates the postharvest quality of vegetables. In case of leafy green vegetables such as spinach and lettuce, a change in chlorophyll and carotenoid contents affects the postharvest quality, associating with physiological activity or external appearance. Therefore, the object of this study is to reveal the relative importance of the radiation dose, storage time, and temperature in terms of the postharvest quality for radiation processing of spinach. We investigated postharvest physiological activities and photosynthetic pigment contents of spinach leaves for 4-d storage at different temperatures in darkness after gamma irradiation of 0.5, 1, 2, and 3 kGy. The physiological activities and photosynthetic pigment contents were determined by chlorophyll fluorescence and HPLC analyses through a model study using leaf disks, respectively.^{7,8}

In leafy green vegetables such as Romaine lettuce and spinach, the population of internalized *Escherichia coli* O157:H7 cells is reduced with a D10-value of 0.39 kGy or 0.27 kGy, respectively.⁹ Moreover, irradiation of electron beam with a dose above 1.07 kGy eliminates *E. coli* O157:H7 and *Salmonella* from spinach leaves, but the respiration rate of spinach is not affected by irradiation up to 2.1 kGy, implying no product damage.¹⁰ To reveal the unfavorable effect of gamma irradiation on the postharvest quality of spinach, physiological activity of leaf disks was investigated during 4-d storage in darkness at 4, 15, 22, or 30°C after irradiation with various doses from 0.5 to 3 kGy. Since spinach is a leafy green vegetable species, the physiological activity was estimated by a chlorophyll fluorescence parameter for

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photosynthetic activity (Fv/Fm) rather than the respiration rate previously used by Neal et al.¹⁰

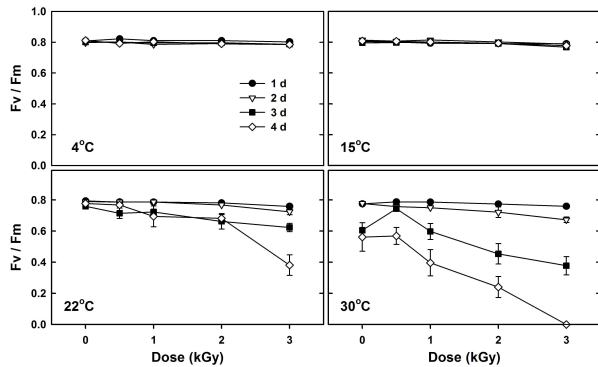


Figure 1. Change in physiological activity of the control and irradiated spinach leaves during the post-irradiation storage at different temperatures. Physiological activity was estimated by a chlorophyll fluorescence parameter for photosynthetic activity, the maximum photochemical efficiency of photosystem II or Fv/Fm.¹¹ The optimal Fv/Fm values for higher plants such as spinach are about 0.8. Bars represent means \pm S.E. (n = 8).

The photosynthetic activity or physiological activity was kept constant in all the irradiated samples at 4°C and 15°C, while it was significantly decreased depending on the radiation dose (>1 kGy) and storage time (>2 d) at 22°C and 30°C (Figure 1). This trend was clearly manifested as correlative visible damages as well as the lower physiological activity (Figure 2). These results suggest that postharvest irradiation of spinach with a dose of gamma rays above 1 kGy should require stricter cold-chain storage.

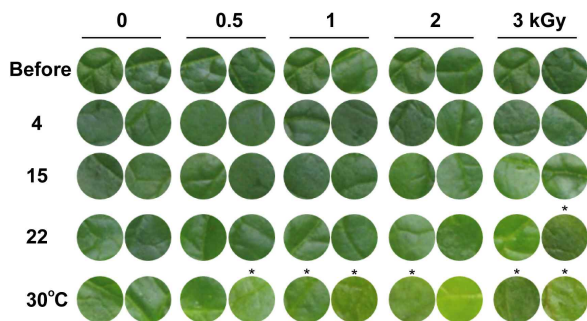


Figure 2. Dark-induced senescence of the control and irradiated leaves at different temperatures. Leaf disks were incubated in darkness for 4 d at 4, 15, 22, or 30°C, respectively. Asterisks represent decomposition of leaf tissues in spite of the relatively greenish leaf color.

Contents of chlorophylls and carotenoids are essentially associated with postharvest quality of leafy green vegetables after irradiation. Lester and Hallman reported that gamma irradiation at 2.0 kGy significantly reduced carotenoids such as lutein/zeaxanthin, neoxanthin, violaxanthin, and β -carotene as well as vitamin C in baby-leaf spinach.⁵ In the present study, total chlorophyll content in all the irradiated samples was maintained almost constant at 4°C during the storage time of 4 d, while it was significantly decreased by about 40% in the 3-kGy sample at 15°C and by 18 to 52% in the 0.5-3-kGy samples at 22°C. This trend was also observed in the total carotenoid content, but less conspicuous. The proportion of lutein in total carotenoid or the ratio of lutein to total chlorophyll was significantly increased in the 2-3-kGy samples during the storage at 22°C, but not at 4°C, while that of β -carotene was slightly decreased at both temperatures (Table 1). The increased lutein and decreased β -carotene in total carotenoid indicates progression of leaf senescence or aging.¹² The dependence on the radiation dose and storage time has been shown for vitamin C loss of fresh-cut vegetables during the storage after gamma irradiation.^{4,6} Similarly, the present data demonstrate that both the radiation dose and storage time are main factors to affect the content and composition of chlorophylls and carotenoids in spinach after gamma irradiation, which are associated with the postharvest quality. Moreover, it is suggested that the unfavorable effect of gamma irradiation on the postharvest quality of spinach could be enhanced by the storage temperature.

Table 1. Change in proportion of β -carotene or lutein in total carotenoid during the storage at different temperatures after gamma irradiation

Dose (kGy)	β -carotene (%)		Lutein (%)	
	4°C	22°C	4°C	22°C
0	19.0 \pm 0.7 ^a	16.8 \pm 1.2 ^a	41.4 \pm 0.1 ^a	44.4 \pm 0.8 ^a
0.5	18.5 \pm 0.6 ^a	15.2 \pm 1.1 ^a	41.5 \pm 0.3 ^a	45.4 \pm 0.4 ^a
1	18.3 \pm 0.6 ^a	16.0 \pm 1.0 ^a	41.5 \pm 0.4 ^a	46.7 \pm 0.9 ^{ab}
2	18.4 \pm 0.6 ^a	14.4 \pm 0.7 ^a	41.5 \pm 0.3 ^a	47.7 \pm 0.6 ^b
3	17.8 \pm 0.5 ^a	13.8 \pm 0.6 ^a	41.3 \pm 0.1 ^a	50.0 \pm 0.8 ^{bc}
Pared t-test	L	* J	L	* J

Means in the same column followed by different letters are significantly different by a Student's t-test (P < 0.05). Asterisks indicate significant differences between two groups (P < 0.01).

In conclusion, by the model study using spinach leaf disks, it was shown that gamma irradiation could affect the postharvest quality or physiological activity by reducing the contents of chlorophylls and carotenoids, or by modulating the composition of carotenoids. Despite any explicit limitations of this model study, the storage time, radiation dose, and temperature were found to be essential factors synergistically contributing to the unfavorable effect of gamma irradiation on the postharvest quality of spinach. The overall considerations about these factors will help to optimize the phytosanitary or postharvest irradiation treatments for leafy green vegetables.

KEYWORDS: Ionizing radiation; postharvest quality; storage temperature; carotenoid; chlorophyll; photosynthesis

Received March 14, 2014; Accepted March 30, 2014

ACKNOWLEDGEMENT

This research was supported by the Nuclear R & D Program of the Ministry of Science, ICT and Future Planning (MSIP), Republic of Korea.

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