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Ethanol extraction-based drying enhanced ESR radical detection in oranges irradiated to different ionizing radiations during storage

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ABSTRACT

The detection of cellulose radicals by ESR spectroscopy in oranges irradiated by different radiation sources has been studied during storage. The peel of oranges was found to be more suitable for ESR analysis in comparison with flesh parts because of its higher fiber and lower moisture contents. The oranges were exposed to gamma ray, electron-beam, and X-ray sources, and the paramagnetic centres were investigated. The samples extracted with absolute ethanol followed by oven drying showed a reduced drying time (five-fold) and enhanced signal sensitivity (double) in comparison to freeze dried samples. Further, the ESR spectrum showed that a radiation-induced signal could be detected in samples exposed to a dose above 0.4 kGy regardless of the radiation source. Irradiated oranges were stored at different conditions such as dark at 4 °C, fluorescent light at 20 °C, and indirect natural light, to study ESR spectral behavior. Fading of signal intensity was predominant when the samples were stored under natural and artificial light. The identification of cellulose radicals using ESR technique was improved for irradiated oranges containing a total fiber content of more than 60% (dry basis) after ethanol extraction even after a prolonged storage.

1. Introduction

The import of fruit from other countries is associated with the risk of migration of potentially damaging insect pests to new areas. To overcome this barrier to international trade, known as the quarantine barrier, technological solutions are being developed worldwide. Food irradiation could be one of the potential solutions to this problem. Phytosanitary irradiation is environmentally safe to replace a chemical fumigant (methyl bromide), an ozone-depleting substance. A dose not exceeding 1 kGy is generally permitted for fresh fruit and vegetables (IAEA, 2014). The safety, wholesomeness, and nutritional adequacy of irradiated foods are now well established and accepted by all major health and food authorities (WHO, 1999). Food irradiation is being used commercially in more than 55 countries around the world (IAEA, 2014). Further developments in the design and adaptation of uses of machine radiation sources (e-beam facilities and X-ray machines) (Cleland, 2006; Pillai et al., 2012; Jo et al., 2015) are also being investigated for phytosanitary irradiation of food. However, various national and international regulations and labeling requirements limit the use of this technology. Reliable methods of identification to enforce regulations and traceability are mandatory for the acceptability of irradiated food (Chauhan et al., 2009).

The ability to analytically identify irradiated food complementary to certification helps enhance consumer confidence. Significant progress has been made in this field (Sanyal et al., 2009; Ahn et al., 2012; Sanyal et al., 2012; Kwon et al., 2013). The interactions between biological materials and different forms of energy are very complex and depend on the irradiation and post-irradiation conditions. Electron spin resonance (ESR) spectroscopy is a non-destructive technique for the detection of paramagnetic species that are generated during the process of irradiation. Three European standards for the detection of irradiated food via ESR spectroscopy have been released by the European Committee of Normalization (CEN) and adopted by the Codex Alimentarius Commission as Codex Standards (EN1786, 1996; EN1787, 2000; EN13708, 2001). Upon irradiation of foods of plant origin, unpaired electrons are induced and trapped in cellulose which may be used as probes. The cellulose radicals have been characterized by a pair of lines to the left and right of the central ESR signal. However, the success in ESR spectroscopy to detect irradiated food of plant origin is limited by the stability of the radiation-induced cellulose signals

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which are influenced by the cellulose and the moisture content of the food commodity. Foods containing a high level of moisture show a fast reduction of the radiation-induced ESR signal during storage (Raffi and Stocker, 1996; Yordanov and Gancheva, 2000). Even in the case of dry food samples such as spices, it did not lead to favorable results because the radiation-induced signal showed a rapid reduction in signal intensity with the storage time, leading to complete disappearance before the maximal general commercial storage time (Ahn et al., 2014a). Many researchers have attempted to address this problem by studying various irradiated foods using freeze-drying (Yordanov et al., 2006), oven-drying (Akram et al., 2012), or other techniques (Yordanov et al., 2005; Kikuchi et al., 2010). A procedure to increase ESR sensitivity was proposed by de Jesus et al. (1999), who extracted the fruit pulp of kiwi, papaya and tomato with ethanol, and measured the dried residue to detect the 'cellulosic' signal.

A large quantity of oranges is imported to South Korea and informative labeling and reliable detection methods are needed to identify irradiated fruit. In the present study, effects of various sample extractions on the EPR spectra of oranges were evaluated before and after irradiation. The main objective of this investigation was to assess the efficacy of sample pretreatments to identify irradiated oranges during prolonged storage using ESR spectroscopy and to understand the role of dietary fiber and moisture contents of oranges in ESR signals.

2. Materials and methods

2.1. Materials and irradiation treatment

Navel oranges, imported from the United States, were purchased from a local market in Daegu, Korea. Samples packed in cardboard boxes were distributed in 3 groups and stored at $4 \pm 1\,^{\circ}C$ before irradiation. Gamma ray were carried out at levels ranging from 0 to 2 kGy at a dose rate of 2.1 kGy/h using a Co-60 source (100 kCi, AECL, IR-79, MDS Nordion International Co., Ltd., Ottawa, Canada) at the Korean Atomic Energy Research Institute, Jeongeup, Korea. Electron beam and X-ray irradiation was carried out with an electron accelerator (ELV-4, 10 MeV, Fujifilm, Tokyo, Japan) in EB-Tech, Korea. The absorbed doses were measured using an alanine-electron paramagnetic resonance dosimetry system, with an EMS 104 EPR analyzer (Bruker Biospin, Rheinstetten, Germany). In order to study the ESR signal behavior during storage, samples were stored at 3 different conditions: dark at $4\,^{\circ}C$, fluorescent light at $20\,^{\circ}C$, and indirect natural light.

2.2. Sample pretreatments for ESR measurement

Both the non-irradiated and irradiated flesh and peel parts of oranges were cut into small pieces (3×3 cm) and subjected to freeze drying and solvent extraction methods. For freeze drying, 30 g of the samples were frozen at $-35\,^{\circ}C$ for 24 h followed by drying for 30 h using a freeze dryer (Bondiro, Ilsin Bio Base, Yangju, Korea). Three solvent extractions followed by oven drying were

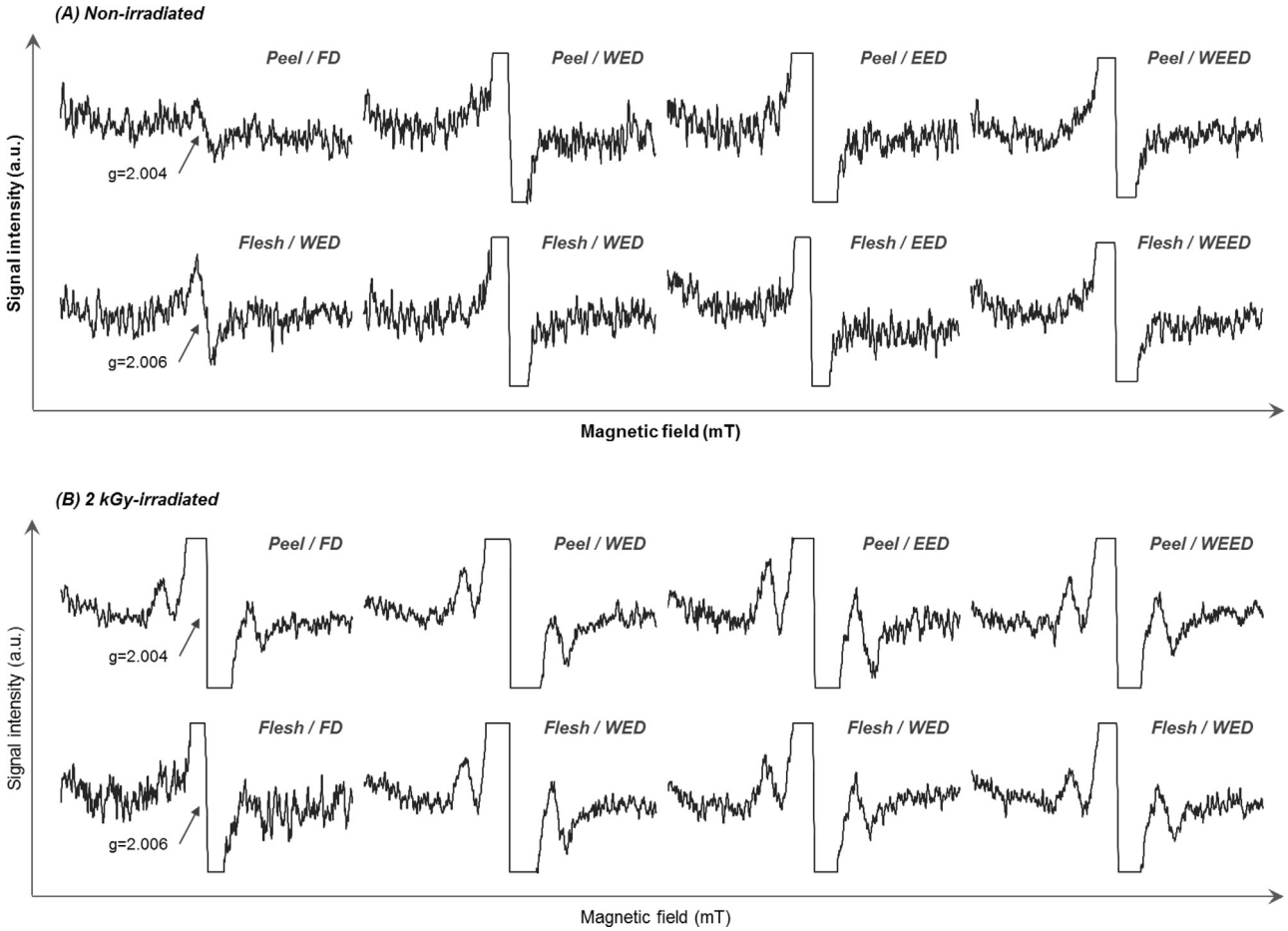

Fig. 1. ESR signals of dried peel and flesh parts of non-irradiated (A) and 2 kGy-irradiated (B) oranges (FD: freeze drying, WED: water-extraction drying, EED: 97% ethanol-extraction drying, WEED: water- and ethanol-extraction drying).