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Irradiation Detection in Korean Traditional Soybean-Based Fermented Powdered Sauces: Data for Establishing a Database for Regulation of Irradiated Foods

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Abstract

To facilitate establishing regulations for irradiated foods, Korean traditional soybean-based fermented powdered *doenjang* (PD), *kanjang* (PK), *kochujang* (PKC) and *chungkukjang* (PC) were irradiated at 1, 3, 5 and 7 kGy, and subjected to irradiation detection analyses as part of establishing a database for detecting irradiated foods. Photostimulated luminescence (PSL) and electron spin resonance (ESR) were applied as the detection methods. Using PSL analysis, the irradiated PD, PK and PKC could be easily distinguished from the non-irradiated ones, while irradiation of the PC at 5 kGy or higher was detectable. The ESR spectra of the irradiated PD, PK and PKC exhibited symmetrical multiplet lines, which might be induced from the crystalline sugar, whereas, the PC showed a single signal at the paramagnetic centers. The signal intensity increased with incremental increases of irradiation doses distinguishing the irradiated samples from the control. In addition, the peak height also revealed that irradiation induced an increment in the intensity of single and/or multiplet lines of the ESR signals, resulting in clear confirmation of irradiation. Thus, the data from this study could be used as references for detecting irradiated soybean-based fermented powdered sauces.

Key words: soybean, powdery sauces, irradiation, detection

INTRODUCTION

Doenjang (soybean paste), *kanjang* (soy sauce), *kochujang* and *chungkukjang*, traditional Korean soybean-based fermented foods, make up one of the common food groups which are consumed in every single household in Korea. They have characteristic flavors, taste, color, nutrients and functional properties, which are generated by the microflora produced from microorganisms such as mold, *Bacillus* spp., lactic acid bacteria and yeast during fermentation (1). However, due to the formation of hydrolytic enzymes, the decomposition of some nutrients, acids, alcohol and flavor components continues, causing a short shelf life, although the decomposition process imparts taste and flavor to the products (2). In general, heat sterilization and/or the addition of synthetic preservatives have been used to improve shelf life. However, heat sterilization causes some problems with changes in, color, texture and functional properties. Additionally, consumers are not willing to purchase the traditional sauces when preservatives have been added to prolong the shelf life (3,4). As an alternative, irradiation techniques have been recognized as an effective method for microbial control with a minimal influence on food texture and functional properties.

Soybean-based powdered *doenjang* (PD), *kanjang* (PK), *kochujang* (PKC) and *chungkukjang* (PC) are one category of food items which have been recently approved for irradiation treatment. Besides those powdery sauces, other food items are also newly approved for irradiation treatment. Furthermore, it is expected that more foods will be approved in the future. Upon approving more food items, regulatory standards for those irradiated foods are need to be issued and implemented to assure safe and effective application of irradiation techniques and to facilitate full disclosure of the irradiation treatment to consumers, who can then make informed choices about choosing an irradiated product over an non-irradiated one. The detection methods developed by the European Committee for Standardization (CEN) to provide reliable

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information to consumers have been successfully applied to identify irradiation treatment. Thus, the aim of this study was to identify irradiation treatment on the Korean traditional soybean-based fermented PD, PK, PKC and PC by using photostimulated luminescence (PSL) and electron spin resonance (ESR) analysis.

MATERIALS AND METHODS

Materials

PD, PKC and PC were purchased from a local market in Daejeon, Korea, and PK was provided by a local food company for the experiment. Each sample was further dried in a laboratory dry oven (OF-22, JO10014, JEIO Tech., Kimpo, Korea) at 40°C overnight to reach a moisture content of approximately 10~11%.

Irradiation

The PD, PK, PKC and PC were packed in polyethylene bags and irradiated at 1, 3, 5 and 7 kGy with a ^{60}Co irradiator (point source, AECL, IR-79, MDS Nordion International Co., Ltd., Ottawa, Ontario, Canada) at the Korea Atomic Energy Research Institute (KAERI). The strength was approximately 100 kCi at a dose rate of 70 Gy/min at 15±0.5°C and the actual doses were within the range of ±2% of the target dose. Dosimetry was performed using 5 mm diameter alanine dosimeters (Bruker Instruments Inc., Ettlingen, Germany), and the free radical signal was measured using a Bruker EMS 104 EPR Analyzer (Bruker Instruments Inc., Ettlingen, Germany).

PSL measurement

PSL analysis was conducted in accordance with the CEN 13751 (5). The materials were dispensed onto a disposable petri-dish (50 mm in diameter) spread into a thin layer, and subjected to a SURRC PSL Irradiated Food Screening System (PPSL 0021, Scottish Universities Research and Reactor Center, Glasgow, UK). The petri-dish with the sample was loaded into sample chamber and measured for 60 s, while the photons, which were detected every second, were accumulated. When the accumulated photon count (APC) reached a threshold at or below 700 counts/60 s, the food was classified as non-irradiated, whereas at an upper threshold of 5,000 counts/60 s or more, they were classified as irradiated foods. Signal levels between the two thresholds were classified as an intermediate for which further investigation is needed to make a reliable determination. All samples were measured in triplicate.

ESR measurement

ESR analysis was performed in accordance with the CEN 1787 (6). The sample (200 mg) was loaded into an ESR tube (4.0 mm in diameter) to produce the same density of sample and fill the cavity of the ESR spectrometer. ESR measurement was performed with an ESR X-band spectrometer (JES-TE 300, Jeol Co., Tokyo, Japan). All the spectra were recorded at ambient temperature and in the same fixed position in the ESR cavity to ensure a uniform and reproducible measurement. The spectrometer was set with a magnetic center field of 328 mT, microwave frequency of 9.21 GHz, microwave power of 0.4 mW, modulation amplitude of 0.4 mT, modulation frequency of 100 kHz and a sweep time of 10.486 s. Each sample was measured in triplicate.

Statistical analysis

The ESR data was analyzed using a Bruker Win-EPR (version 2.11) software, and plotted using an OriginPro 7.0 (OriginLab Corporation, Northampton, MA, USA) software program.

RESULTS AND DISCUSSION

PSL detection

The photon counts from PSL measurements for PD, PK, PKC and PC are shown in Table 1. The photon counts from non-irradiated samples gave the photon counts below 500 counts/60 s, which was less than the threshold value for non-irradiated foods (700 counts/60 s); whereas the photon counts of the irradiated PD, PK and PKC at above 1 kGy were higher than 5,000 counts/60 s, which was the threshold value for irradiation treatment. The clear positive results of PD, PK, and PKC successfully differentiated them from their non-irradiated counterparts. However, only the PC irradiated at 5 kGy or higher yielded photon counts above 5,000 counts/60 s. The PC at 1 and 3 kGy yielded intermediate PSL signals of 3044.8±987.2 and 3564.8±1275.1, respectively. In addition, upon comparing the photon counts at 5 and 10 kGy, the PC yielded much lower photon counts than did the PD, PK, and PKC (Table 1). Due to the low PSL signals from the PC, it was assumed that the amount of inorganic minerals in the sample might be very small compared to the PD, PK, and PKC. Thus, PSL analysis can be a reliable method for detecting irradiation treatment of PD, PK, and PKC, but not PC.

Other studies have reported that variations in the yields of PSL photon counts largely depend on the amount of inorganic minerals contained in a food sample and/or dispersed accidentally on the surface of sample at the time of measurement. Sanderson et al. (7) used PSL to detect irradiation in herbs, spices, shrimps, fruits and vegetables. They indicated that a low PSL sensitivity of the accompanying minerals of very clean samples or

Table 1. Photostimulated luminescence (PSL) measurements at various irradiation doses (Unit: photon counts)

	Doses (kGy)				
	0	1.0	3.0	5.0	10.0
Powdery *doenjang*	374.6±35.8[1)]	499370.7±36469.8[2)]	188602.4±93271.7	698726.8±10707.7	919420.5±73783.1
Powdery *kanjang*	492.0±91.5	214154.5±10393.1	404361.1±39120.7	534842.7±4351.5	715716.8±119805.2
Powdery *kochujang*	297.2±134.7	172510.9±4323.9	364798.6±18168.4	412803.1±21509.5	529056.2±30052.8
Powdery *chungkukjang*	330.1±16.3	3044.8±987.2[3)]	3564.8±1275.1	15380.6±8354.5	15827.1±9280.1

[1)]Less than 700 counts/sec (negative).
[2)]More than 5,000 counts/sec (positive).
[3)]Between 700 and 5,000 counts/sec (intermediate).

blends that emit only a low amount of photons from irradiated materials might hinder the successful application of PSL analysis. Also, Bayram and Delincée (8) found weak PSL signals of some of Turkish export foodstuffs such as spices, tea, dried fruits and nuts resulting in a detection problem when using the PSL system. They reported that those food samples were confirmed to be irradiated by Thermoluminescence (TL) analysis. In addition, CEN 1787 (6) suggested that for irradiated samples with insufficient PSL sensitivities, negative results might occur. In contrast, non-irradiated samples with high PSL sensitivity may sometimes test positive for irradiation. In principle, the PSL method can be applied as a screening method to detect irradiation treatment of any food containing inorganic mineral debris. And the PSL sensitivity of a sample depends on the quantities and types of minerals within the individual sample.

ESR detection

To confirm the irradiation treatment of samples by another detection method, ESR analysis was applied to the samples. The ESR spectra of PD, PK, PKC and PC are shown in Fig. 1~4, respectively. As shown in the figures, the PD, PK and PKC showed the multiplet ESR spectra structure with a symmetrical line shape. The symmetrical multiplet lines are associated with free radicals where the unpaired electrons are localized on carbon atom (9). Whereas, a pair of single signals occurred from the PC. According to the CEN 13708 (10), irradiated food containing crystalline sugar shows typical multi-component ESR spectra, and due to different mono- and di-saccharides, various ESR specta types can also be due to changes in the saccharide composition. On the other hand, according to the CEN 1787 (6), a single signal is observed in the ESR spectra of a food con-

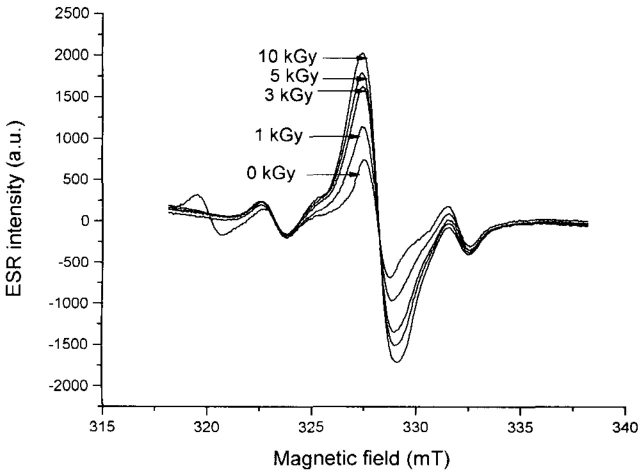

Fig. 2. ESR spectra of the powdered *kanjang* at the various irradiation doses (kGy).

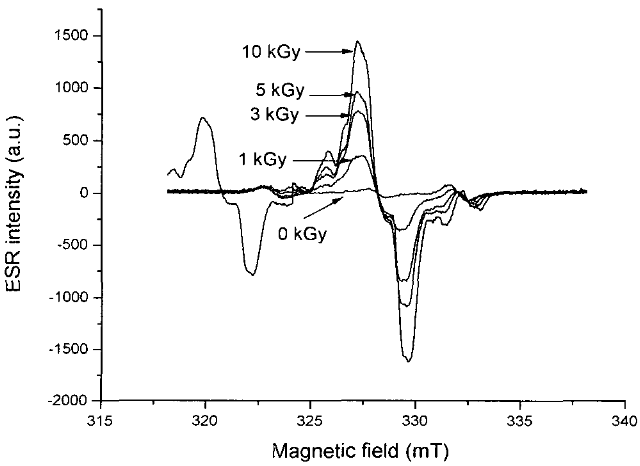

Fig. 1. ESR spectra of the powdered *deonjang* at the various irradiation doses (kGy).

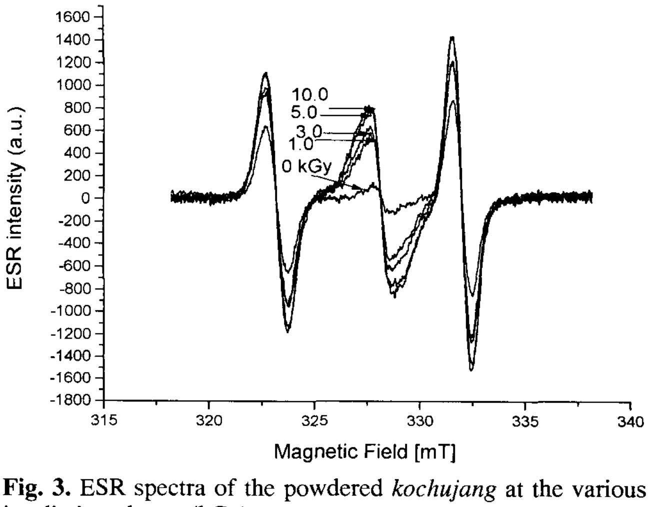

Fig. 3. ESR spectra of the powdered *kochujang* at the various irradiation doses (kGy).

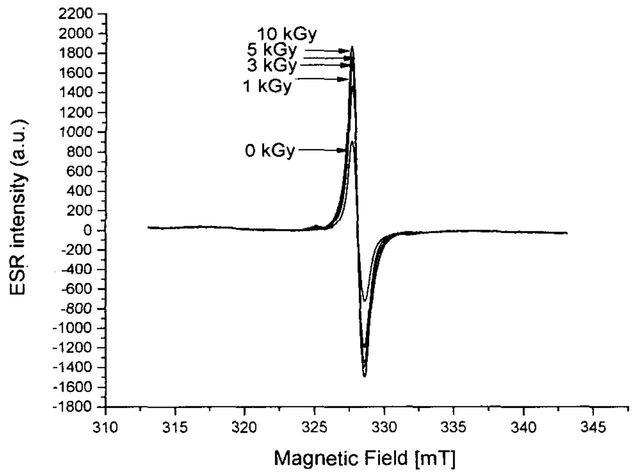

Fig. 4. ESR spectra of the powdered *chungkukjang* at the various irradiation doses (kGy).

taining cellulose, due to the cellulose radicals formed by gamma irradiation.

The signal intensity of irradiated foods is usually much greater than that of non-irradiated foods, which can differentiate them from the non-irradiated food. In principles, ESR spectroscopy detects paramagnetic centers (e.g. radicals), which are either due to irradiation or due to other compounds present (10). Thus, the intensity of the signal varies according to the concentration of paramagnetic compounds and with the applied dose. As shown in the spectra, the multiplet resonance lines of which a central signal of about 5~6 mT occurred at the paramagnetic center could determine irradiation treatment. Also, an irradiation induced increment in the intensity of single resonance and a radiation specific multiplet line was observed. In addition, it was found that there was a slightly positive correlation between the ESR intensity and irradiation doses showing that the main central peak of samples increased with each incremental increase in irradiation dose.

For the PD and PKC, Fig. 1 and 3, the intensities of irradiated samples were significantly higher than those of the non-irradiated samples, differentiating clearly the irradiated samples from the control. On the other hand, the PK and PC, (Fig. 2 and 4) ESR spectra at 0 kGy showed a small signal, but were still relatively low compared to the signal at 1 kGy making it possible to distinguish irradiated samples from the control, but due to only small differences in signal intensity of samples irradiated at doses between 0 kGy and 1 kGy, in some cases, a mistake can possibly be made in determining irradiation treatment for the PC sample. These results indicate that ESR analysis can be a reliable detection method for the PD and PKC, but may not be always reliable for the PC. To provide more information on detection, the PK and PC were subjected to TL analysis, but it turned out that the extraction of silicate minerals from the samples was not successful. The major reason may be that silicate minerals are generally present on the surface of raw food materials, and are frequently removed by the processing of prepared foods. Thus, it could be explained that based on the results of this experiment that the ESR signals at 0 kGy can be used as the control signal for comparing some PCK provided from different companies. Irradiation treatment was confirmed by the peak intensity of ESR signals. Table 2 shows the peak heights of PD, PK, PKC and PC. As shown in the figures, the peak heights of the irradiated PD and PKC at 1 kGy were greater than at 0 kGy, whereas for the PK and PC at 1 kGy, the peak heights were about 2 times higher than at 0 kGy.

Upon considering the multiplet spectra, Lee et al. (9) reported that the multiplet lines, which were indicative of free radicals, were formed in the protein portions by irradiation, and this might be due to the different structures of proteins. In addition, Pshezhetskii et al. (11) reported that the ESR spectrum, which consisted of singlet and multiplet lines, was induced by deamination and decarboxylation of amino acids. As shown in the figures, from the multiplet ESR spectra observed in the PD, PK and PKC, we could assume that the multi-component ESR signals might be a result of sugars formed during decomposition of the soybeans during fermentation. Also, since the overall spectrum structure depends on the radical composition and on the crystallinity of the mono- and di-saccharides present in the sample, variations in the spectrum characteristics occur when different sugars are formed (10).

CONCLUSION

As a part of our efforts for establishing a data-base

Table 2. Peak heights[1)] of the ESR spectra of the powdered sauces at various irradiation doses

Materials	Doses (kGy)				
	0	1.0	3.0	5.0	10.0
Powdery *deonjang*	105.0±2.65	709.0±30.45	1599.0±57.24	2022.0±64.86	3151.7±189.74
Powdery *kanjang*	1402.7±71.07	2119.3±348.72	3050.7±250.33	3304.7±101.86	3744.0±71.25
Powdery *kochujang*	288.5±59.10	1143.5±109.60	1280.5±183.14	1579.5±139.30	1705.5±70.0
Powdery *chungkukjang*	1637.5±41.72	2670.5±20.51	3043.0±295.57	3149.0±210.72	3388.0±90.51

[1)]The peak intensity in the paramagnetic center.

for irradiated food detection, Korean traditional soybean-based fermented powdered *deonjang* (PD), *kanjang* (PK), *kochujang* (PKC) and *chungkukjang* (PC) were irradiated (1, 3, 5 and 10 kGy) and subjected to PSL and ESR analyses to identify irradiation treatment. PSL measurement showed that the irradiated PD, PK and PKC yielded photon counts of higher than 5,000 counts/60 s differentiating them from the non-irradiated samples. But, the PC could only be distinguished from control when the irradiation dose was 5 kGy or higher, yielding only intermediate photon counts at 1 and 3 kGy. On the other hand, the ESR spectra of the PD, PK and PKC exhibited symmetrical multiplet lines, which might be induced by the crystalline sugar. In contrast, the ESR spectra of PC showed a single signal at the paramagnetic center. The signal intensity increased with each increased increment of irradiation doses, distinguishing the irradiated samples from the control. Also, it was found that there was a positive correlation between the ESR intensity and the irradiation doses. The peak height also suggested that the irradiation increased the intensity of single and/or multiplet lines of ESR signals.

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