

Ultra-weak bioluminescent and vigour of irradiated rice

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Abstract: Detection of the irradiation dose and the vigour of irradiated rice based on ultra-weak luminescent analysis is one of the analytical detection methods. Rough rice and head rice flour were used in this research for ultra-weak luminescent analysis. The bioluminescent intensity of rough rice was different at irradiation dose after different storage time, and the trend was consonant with the germination rates of irradiated rough rice. The bioluminescent intensity and germination rates of irradiated rough rice at irradiation dose after different storage time were due to the effect of the rice embryo that affected by irradiation, which had self-repair during storage. As a result, the ultra-weak luminescent analysis with rough rice cannot used to detect the irradiation dose of irradiated rice, but it can be used to detect the vigour of irradiation rice. Experimental results show that the irradiation dose has a highly significant effect on the bioluminescent intensities of rough rice flour when cane sugar was added.

Keywords: ultra-weak luminescent, vigour, gamma irradiation, rice

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1 Introduction

Gamma irradiation was used in agricultural product to sterilize and control pests for prolonging shelf-life and reducing health hazards^[1]. The irradiated foods are confirmed to be wholesome and nutritionally adequate and safe for human consumption within 10 kGy dose^[2-3]. However, labelling is deemed necessary in order to ensure the freedom of choice for consumers^[4]. In the international conference “The Acceptance, Control of and Trade in Irradiated Food”, it was recommended that the compliance with labelling regulations should be checked directly in the food^[5]. And this means that the analytical detection method of irradiation treatment has to be

improved and new methods need to be investigated^[1].

Luminescence analysis is one of the analytical detection method includes thermoluminescence analysis, ultra-weak bioluminescence analysis, chemiluminescence analysis, photoluminescence analysis, and so on^[6]. Ultra-weak bioluminescence is one of the most wonderful and attractive phenomena of life and defined as an enzymatic chemiluminescent reaction with high quantum yield^[7]. The previous investigations^[8-10] showed that gamma irradiation may change the structure of large molecules (e.g. starch, protein and fat) and influence the drying characteristics and nutritional quality of rice. The change in molecules structure can result in enzymatic chemiluminescent reaction^[11]. So, the ultra-weak bioluminescence intensity of head rice (without embryo) may be relative to the irradiation dose. The detection of the irradiation treatment in agricultural products based on ultra-weak bioluminescence may be a one kind of method. However, few studied was reported on detection of irradiated rice by the ultra-weak bioluminescent.

The ultra-weak bioluminescence analysis was also an effective method for detecting freshness degree of

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chicken egg^[12], singlet oxygen generation in germinating soybean^[13], content of bacteria^[7], fungi^[14], and so on. Rough rice has the organization of embryo, and its vigour of life may also be detected with ultra-weak bioluminescence analysis. It was testified that, the gamma irradiation could affect the vigour of agricultural products^[15-16]. However, the few studies on ultra-weak bioluminescent analysis on detecting the vigour of irradiated rice were also found.

The objectives of this paper were to: (1) find the relationship between ultra-weak bioluminescent intensity and vigour of irradiated rough rice; (2) study the relationship between ultra-weak bioluminescent intensity of head rice flour and irradiation dose.

2 Materials and methods

2.1 Rough rice

Rough rice (Zhenong 1, late-cropping season rice) harvested in October, 2005, from the experimental farm of Agronomy, Zhejiang University, was used for this experiment. After irradiated, all the rough rice samples were dried (Oven dry at 50°C) to the safe moisture content (13.5±0.1%, dry basis) and then stored for 0 (non-storage), 1, 1.5, 2 years under a commercial storage condition (10±1°C of temperature and 50%–55% of relative humidity).

2.2 Gamma irradiation

Rice samples were exposed to 60°C source at ambient temperature at the Institute of Nuclear-agriculture Sciences, Zhejiang University. The irradiated doses for of rough rice samples were 0 kGy (non-irradiation), 2 kGy, 5 kGy, 8 kGy, and 10 kGy respectively, with dose rate at 1 kGy/h. Dosimetry was performed using 5 mm diameter alanine dosimeters (Bruker Instruments, Rheinstetten, Germany) and the dosimeters were calibrated as per the International standard set by the International Atomic Energy Agency (Vienna, Austria)^[11].

2.3 Head rice flour

After gamma irradiation and drying, half rough rice samples were dehulled in a Satake dehuller and milled in a Satake TM-05 Grain-testing mill (Satake Co., Japan). The resulting milled rice was then ground in an Udy

cyclone mill (Satake Co., Japan) with a 100-mesh sieve for study the relationship between ultra-weak bioluminescent intensity of head rice flour and irradiation dose. The head rice flour samples were also stored for 0 (non storage), 1, 2, 3 years under the same conditions as rough rice samples.

2.4 Ultra-weak bioluminescent detection

Preliminary experiments showed that sample weight (including grain and flour) of 10 g was enough to detect the ultra-weak bioluminescence^[15,16]. In this study, the ultra-weak bioluminescence of grain and flour samples were measured with a BPCL ultra-weak luminescence analyzer (Institute of Biophysics, Chinese Academy of Sciences, China) at room temperature (30±1°C). In each experiment, each sample was put in a special experimental vessel (appurtenance of BPCL ultra-weak luminescence analyzer) for measuring the ultra-weak luminescence. The ultra-weak bioluminescence of sample was calculated with subtracting the ultra-weak luminescence of vessel from the total ultra-weak luminescence of sample and the vessel^[17]. The experiment was conducted by five replicates.

2.5 Vigour of rice and Germination test

One hundred seeds were symmetrically placed on two filter papers, which had dipped in water for one hour, in a 15-cm-diameter Petri dish. Seeds were incubated under 12 h light and 12 h darkness for 14 days in growth chambers at constant 30°C (GB/T 3543.4-1995, National Standard of China). Seed germination rate was detected at the 14th day. The germination tests were carried out after storage for 0 (non-stored), 1, 1.5 and 2 years. Seed sample was considered as germinated seed when the root was 5 mm long.

Experiment for each sample had three replications. The standard deviation and ANOVA analysis was calculated using SAS software (SAS Institute Inc. 1999).

3 Results and discussion

3.1 Effect of dose on ultra-weak bioluminescent intensity for rough rice

The effect of dose on ultra-weak bioluminescent intensity for rough rice after different storage time was shown in Figure 1. It was obvious that the bioluminescent intensity of rough rice was different at

irradiation dose after different storage time.

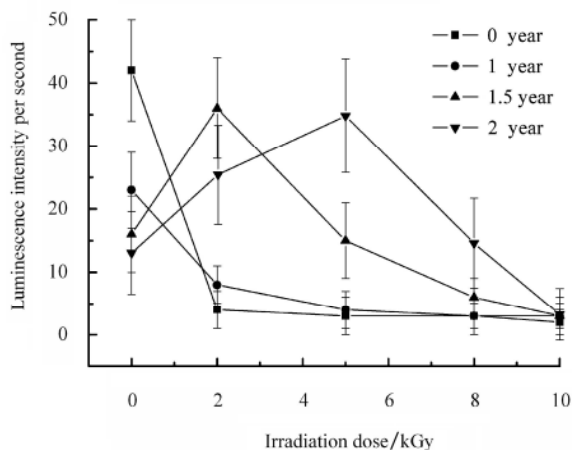


Figure 1 Effect of dose on the ultra-weak bioluminescent intensity of irradiated rough rice after different storage

With the increasing storage time, the bioluminescent intensity of non-irradiated rough rice decreased, but the bioluminescent intensities of rough rice irradiated with 5 kGy and 8 kGy increased. The bioluminescent intensity of 2 kGy irradiated rough rice had a higher value after 1.5 year storage. It may be because that the 2 kGy irradiated rough rice had a higher vigour after 1.5 year storage. The bioluminescent intensity of 10 kGy irradiated rough rice had not a significant difference among four year storages.

3.2 Relation between ultra-weak bioluminescent intensity and vigour for irradiated rough rice

It was known that the rough rice had the embryo, which had different life activity for germination. The life activity of rough rice was testified to be different with the diverse irradiation dose^[15]. The changing of ultra-weak bioluminescent intensity of rough rice (Figure 1) may be affected by the different life activity of rough rice.

The effect of irradiation dose on germination rate of irradiated rough rice after different storage time was shown in Figure 2. The trend of germination rate for irradiated rough rice at different dose and after different storage time was similar with that of bioluminescent intensity for rough rice sample at different dose after different storage time.

With the increasing storage time, the germination rate and the bioluminescent intensity for non-irradiated rough rice decreased (Figure 1). The bioluminescent intensity

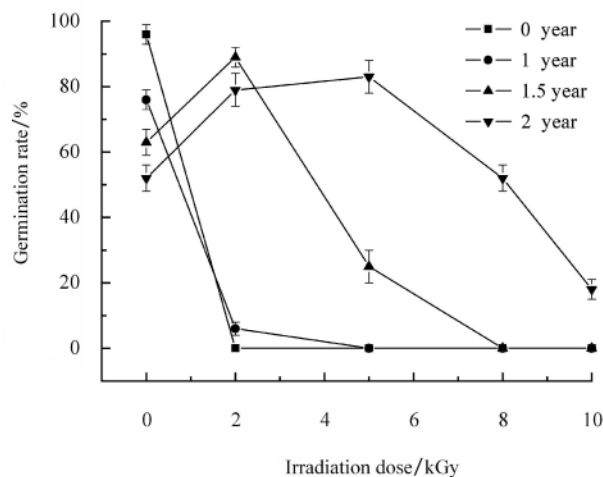


Figure 2 Effect of dose on the germination rate of irradiated rough rice after different storage

had a significant positive correlation with the life activity of life-form^[7,13]. For example, the germination rate of 5 kGy and 8 kGy irradiated rough rice sample increased with the increasing storage time because of the increasing life activity. The change of life activity for irradiated rough rice at different storage may be due to the interaction between self-repair (increasing the life activity) and the respiration (decreasing the life activity) of irradiated organism^[18,19]. The life activity of rough rice decreased evidently after irradiation and increased and had a high degree after 1.5 year and 2 year storage respectively due to the self-repair during storage. With the changing of life activity and germination rate of irradiated rough rice, the bioluminescent intensities of irradiated rough rice samples were changed similarly (Figure 1). The germination rates of 10 kGy irradiated rough rice sample were all very low during storage, which is the reason for no significant difference in bioluminescent intensity of 10 kGy irradiated rough rice between different storage time.

After 0 and 1 year storage time, the high germination rates (>75%) appeared for non-irradiated rough rice because of its higher life activity; but the germination rate was very low when the dose was more than 2 kGy due to the low life activity.

This is because of the effect of irradiation on rice embryo and embryo is damaged by γ -ray^[7,15]. After one year storage, the self-repair of irradiated rough rice had not represented. After 1.5 and 2 year storage, the higher

germination rates of rough rice appeared at 2 kGy and 5 kGy, respectively. It was because that the self-repair of rough rice sample occurred with the increasing storage time and had a higher degree at 2 kGy and 5 kGy after 1.5 and 2 year storage respectively. These testified the changing of bioluminescent intensities of irradiated rough rice sample at irradiation dose after different storage time (Figure 1).

3.3 Effect of dose on ultra-weak bioluminescent intensity of head rice flour

The embryo was taken out and the head rice flour was used to detect the effect of dose on the ultra-weak bioluminescent intensity. The effect of dose on ultra-weak bioluminescent intensity of head rice flour after different storage time was shown in Figure 3. The bioluminescent intensities of head rice flour increased with the increasing dose. It was assumable due to the changing of molecules structure caused by irradiation^[7,13].

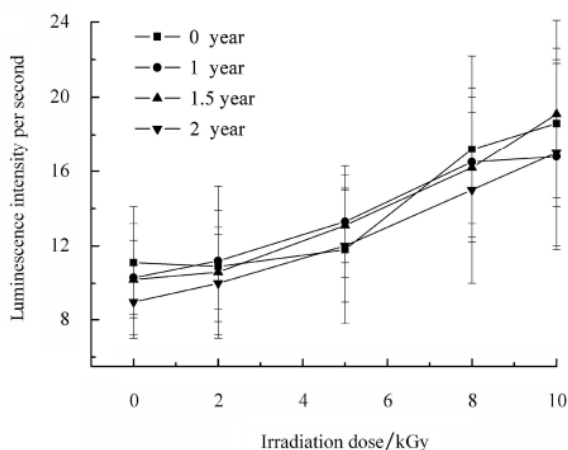


Figure 3 Effect of dose on the ultra-weak bioluminescent intensity of irradiated head rice flour after different storage

The bioluminescent intensities were not significant affected by storage time and the irradiation dose have not a significant effect on the bioluminescent intensities of head rice flour ($p > 0.1$, Table 1).

Table 1 Effect of inducement on bioluminescent by ANOVA analysis

	No was added in wheat flour	Water was added	Cane sugar was added
Effect of dose	$p > 0.1$	$p < 0.05$	$p < 0.01$
Effect of storage time	$p > 0.1$	$p > 0.1$	$p > 0.1$

Note: Irradiation dose: 0 kGy, 0.6 kGy, 1.5 kGy, 2.4 kGy and 3 kGy. Storage time: 0 year, 1 year, 1.5 year and 2 year.

3.4 Inducement of bioluminescent

The ultra-weak bioluminescent intensity of head rice flour was testified to be a possible way to forecast the dose of irradiated rough rice. But the lower relativity of bioluminescent intensities of head rice flour and the dose indicated that forecasting the dose was not feasible yet.

Water as an evocator was added in head rice flour (15% moisture content) from the injection aperture of the BPCL ultra-weak luminescence analyzer during ultra-weak bioluminescent detection. When water was used as a revulsant of ultra-weak luminescence, the effect of dose on the ultra-weak bioluminescent intensity of head rice flour at different storage time was shown in Figure 4. Compared with Figure 3, a similar result could be found. The irradiation dose has a significant effect on the bioluminescent intensities of head rice flour when water was added ($p < 0.05$, Table 1).

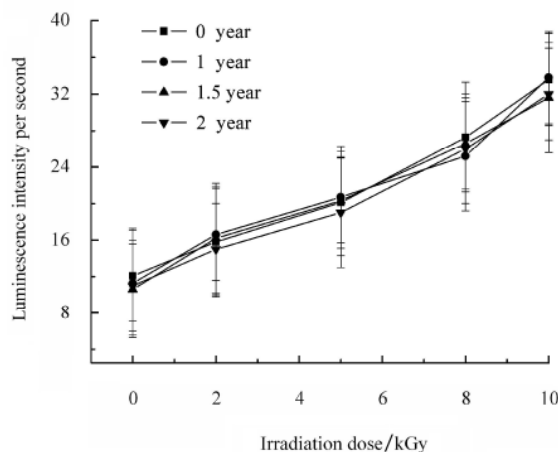


Figure 4 Effect of dose on the ultra-weak bioluminescent intensity of irradiated head rice flour added with water after different storage

Cane sugar solution as an evocator was added in head rice flour from the injection aperture of the BPCL ultra-weak luminescence analyzer during ultra-weak bioluminescent detection. When cane sugar solution was used as a revulsant of ultra-weak luminescence, the effect of dose on the ultra-weak bioluminescent intensity of head rice flour at different storage was shown in Figure 5. Compared with Figure 3, 4 and 5, a most distinct trend could be seen from Figure 5, and the irradiation dose has a highly significant effect on the bioluminescent intensities of head rice flour when cane sugar was added ($p < 0.01$, Table 1).

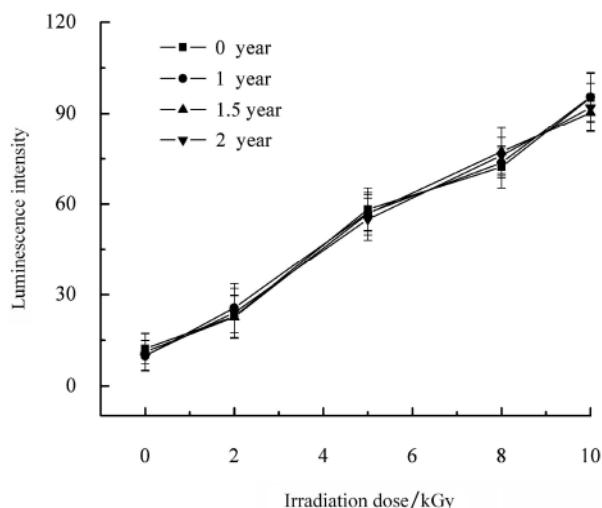


Figure 5 Effect of dose on the ultra-weak bioluminescent intensity of irradiated head rice flour added with cane sugar solution after different storage

4 Conclusions

The bioluminescent intensity and vigour (germination rate) of rough rice sample were different with the increasing storage time and irradiation dose. So, the ultra-weak luminescent analysis could be used to distinguish the different vigour of rough rice seed.

The bioluminescent intensities of head rice flour increased with the increasing dose. The bioluminescent intensities versus irradiation dose were not significantly affected by different storage time. The irradiation doses have a highly significant effect on the bioluminescent intensities of head rice flour when cane sugar was added.

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