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Freshly harvested onions were exposed to different doses of gamma irradiation at 0.15 KGy, 0.5 KGy and 1.5 KGy. After exposure, samples were stored at ambient temperature up to 90 days and observed for fungal incidence and determined nutritional content at 30 days of intervals. The data revealed that, fungal incidence on onion samples subjected to 0.15 KGy dosage reduced from 31.8 to 26.1% during storage period without affecting nutritional qualities. In control samples, however, per cent incidence increased from 41.8% to 66.8% at the end of storage period (90 days). Thus, application of gamma irradiation can be considered as a safe and effective post-harvest method for controlling fungal contamination causing deterioration of onions.

Key words: Fungal incidence, gamma irradiation, mycotoxin, nutritional content, onion

INTRODUCTION

Onion (*Allium cepa*), (Family: Liliaceae) is a widely consumed vegetable across the world throughout the year. In India onion is mainly cultivated in three seasons namely *kharif*, late *kharif*, and *rabi* seasons. *Kharif* produce is available in the market from October to December, whereas the late *kharif* produce which comes in the market from January to March followed by the *rabi* crop which accounts for 60% of onion production, hits the markets from April to June. The same *rabi* crop continues to meet the consumer demand till October-November every year before the *kharif* crop is harvested and brought to the market. Therefore, storage of *rabi* crop becomes pivotal to maintain its continuous supply in the markets (Shivakumar and Chandrashekar, 2014). Onion may be exposed to a wide range of fungal contamination during pre-harvest, post-harvest handling and storage. Sprouting, weight loss, rotting and vulnerability to fungal infection are the key factors responsible for reduction in quality, productivity, and shelf life of onion.

Black mould disease caused by *Aspergillus niger* is one of the major post-harvest diseases of onion recorded in India. Fungal contamination results in physical, chemical, nutritional changes and mycotoxin contamination which causes serious health risks among individuals. (Adebayo-Tayo *et al*.2012). Thus, detection and elimination of fungal species is of vital importance to preserve the food commodity from microbial spoilage. Gamma irradiation is an ecofriendly mycotoxin reduction approach and can complement existing technologies to ensure food security and safety. Irradiation could be used for anti-infestation of food grains and pulses: inhibition of sprouting in onions, potatoes, garlic, yam, and ginger, preventing microbial contamination of spices, extending shelf-life under recommended conditions of storage, and overcoming quarantine barriers in international trade. The irradiation process has been approved by the Food and Agriculture Organization (FAO), the World Health Organization (WHO), the International Atomic Energy Agency (IAEA) and the Codex Alimentarius Commission (CAC). Irradiation dose up to 10 KGy is considered as safe and does not impart any nutritional problems and toxicological hazards. Irradiation can be used for the direct purpose of eliminating or reducing the presence of molds and mold spores in foods and in feeds, extending shelf-life under recommended conditions of storage and overcoming quarantine barriers in

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international trade (Fapohunda *et al.* 2012; Calado *et al*. 2014).

MATERIALS AND METHODS

Collection of onion samples

Freshly harvested onion samples were collected from agricultural field (after curing) during harvesting season and was subjected to gamma irradiation. Approximately 500g of onion samples in triplicates were packed in polythene bags (0.1mm thickness), sealed and exposed to different doses of gamma radiation.

Sample exposure for irradiation

Samples were irradiated at Gamma irradiation chamber (Model GC-1200) located at Department of Studies in Physics, University of Mysore with C60 source at the dose rate of 5.46 Kilo Gray/hour (Fig.1). Onions were exposed at 0.15KGy, 0.5KGy and 1.5KGy doses and stored at ambient temperature (26±2°C) conditions. Irradiated samples were observed for physical parameters (colour, texture, sprouting), fungal incidence and nutritional content at regular intervals (0, 30, 60, 90 days). Gamma irradiation process was evaluated by performing nutritional analysis of the samples and compared with control samples. All the analysis were carried out in triplicates (Al-Kuraieef, 2014; Abdullah *et al.*2018).

Mycological analysis of gamma irradiated samples

Approximately100 g (two bulbs) of onion and from each treatment were subjected to mycological analysis by blotter method. Outer scales of onion were discarded, then washed, air dried and cut into small bits of 1 cm with sterile knife. 1 g of the respective samples were then surface sterilized with 1% sodium hypochlorite solution, air dried and placed on to moistened blotter paper at the rate of 15-20 bits per Petriplate and incubated at 26±2°C for 7 days. Samples were observed for fungal growth during incubation period. Per cent incidence of fungal isolates was calculated according to (Ghiasian *et al*. 2004).

$$\text{Incidence (\%)} = \frac{\text{Number of samples infected by genus/species}}{\text{Total number of samples}} \times 100$$

Nutritional analysis

Nutritional content (pyruvic acid, ascorbic acid and carbohydrates) of irradiated samples was determined and compared with control samples (unirradiated) to know the influence of gamma irradiation treatment on onion quality.

Estimation of pyruvic acid content

Pyruvic acid was determined by modified method of Schwimmer and Weston (1961). Onion (50 g) was blended in mixer with 50 ml of water and kept undisturbed for 10 min. The homogenate was filtered through two layers of cheese cloth. Onion filtrate (1 ml) was diluted with water and 0.0125% DNPH in 2N HCl (1:1) and incubated at a 37^0C in water bath for 10 min. To the resultant solution, 2.5% 0.6N sodium hydroxide was added and absorbance was measured at 420 nm (UV-1800, Shimadzu. USA). Pyruvic acid content was determined against standard calibration curve (25–200 µl) with 1 mM sodium pyruvate. Pyruvic acid content was expressed in µmol /g.

Estimation of ascorbic acid content

Ascorbic acid content was determined by spectrophotometric method as given by Kapur *etal.* (2012). Standard ascorbic acid solution (500 µg/ml) was prepared and diluted to 5, 10, 15, 20, 25 µg/ml. Onion sample extract was prepared by blending 5 g of sample in the blender. Then sample was mixed with 25 ml of 5% metaphosphoric acid-acetic acid solution in a 50mL volumetric flask and diluted up to the mark with the same solution. Then it was filtered and centrifuged at 4000 rpm for 15 min (R-24, Remi centrifuge, India). Supernatant solution obtained was mixed with 4-5 drops of bromine water until colour appears, to this 10 % of thiourea solution was added slowly to remove excess bromine and the clear solution was obtained. 2,4- dinitrophenyl hydrazine solution (1 ml) was added to the standard and sample preparations and kept at water bath (37°C) for 3 h. After incubation, solutions were cooled on ice bath for 30 min and 5 ml of chilled 85% H_2SO_4 was added. The absorbance of the resulting-coloured solutions was recorded at 521 nm using spectrophotometer (UV-1800, Shimadzu. USA). Standard calibration curve was used to determine the concentration of ascorbic acid in the investigated samples.

Estimation of carbohydrate content

The total carbohydrate content of irradiated and unirradiated onion samples was estimated by phenol sulphuric acid method (Dubios *et al.* 1956). Briefly, 1 g of crushed onion sample was transferred into a boiling tube and kept in water bath for 3 h with 5 ml of 2.5N HCl and allowed to cool at room temperature. The volume was made upto 100 ml after neutralization with sodium carbonate. Sample (0.2 ml) in triplicates was transferred into test tube and made upto 1 ml with water. For blank sample 1 ml water was taken. Phenol solution (1 ml) and 96% sulphuric acid (5 ml) was added to each tube, mixed well, and kept in water bath at 30°C for 20min. The absorbance was recorded at 490 nm in a Spectrophotometer (UV-1800, Shimadzu. USA) and the total amount of carbohydrate present in the sample was calculated using the standard graph prepared from standard glucose (100 mg/ ml) calibration curve.

RESULTS AND DISCUSSION

Mycological analysis of irradiated and control (unirradiated) onion samples were carried out to know the per cent incidence of fungal pathogens associated with them. The results (Table-1) revealed that the per cent incidence recorded for the onion samples subjected to 0.15KGy irradiation was comparatively lesser (31.8%) than control samples (41.8%) and the incidence was reduced in the following storage period (30,60 and 90 days) from 31.8% to 26.1%. Similar results were observed by Abdullah *et al.* (2018) where irradiation dose of 0.15KGy reduced fungal and bacterial load significantly in onion bulbs with retention of organoleptic properties. In control samples, per cent incidence was increased from 41.8% to 66.8%at the end of storage period (90 days). Gamma irradiation of onion samples at 0.15KGy reduced fungal incidence but it was not effective in complete elimination of *Aspergillus* species (Fig 2). The results are in acceptance with Tripathi *et al.* (2011) where they observed that, gamma irradiation of onion doses below 1KGy was unable to decontaminate *A. niger* causing back mould rot disease of onion after 60 days of storage. Calado *et al*. (2014) concluded that the fungal load and their mycotoxin contamination may be substantially reduced with 5-10KGy doses of gamma irradiation. Many factors affect the radiolysis process such as doses absorbed, fungal load, initial concentration of mycotoxin, position in the irradiated system, moisture content, and/or the presence of other matrix components (Calado *et al.* 2014). However, per cent incidence of 0.5 and 1.5KGy irradiated samples was not controlled, rather it got increased and this may be due to the extent of physical damage caused by gamma irradiation at higher doses and thus samples got spoiled within 45days of storage period. Abdullah *et al*. (2018) noted that 0.15 KGy has been approved as the maximum dose for onions by the Codex Alimentarius Commission.

Effect of gamma irradiation was evaluated by performing nutritional analysis of irradiated and unirradiated onion samples. Samples were observed for colour and sprouting during storage period. Pyruvic acid content, ascorbic acid content and carbohydrates content were estimated, and the results were tabulated (Table1). In the present study, it was found that sprouting was effectively controlled in all the irradiated samples irrespective of dosage treated. Gamma irradiation was found effective in preventing sprouting losses in onion bulbs even after 120 and 270 days of storage (Tripathi *et al*. 2011). Samples treated with 0.5 and 1.5KGy were spoiled within 60 days of storage. Pyruvic acid content was increased from 6.89 to 9.90 µmol/g in control samples and 6.26 to 12.9 µmol/g in irradiated samples (0.15KGy) till storage period. Pyruvic acid level, as an index of onion pungency, has become a routine procedure to ensure the quality of onions for both shippers and consumers (Abrameto *et al.*2010). The estimation of pungency in bulbs has become necessary as the popularity of high pungent Indian onion cultivars in India has increased. Assessment of bulb pungency level in different Indian cultivars of onion has been reported. Ascorbic acid content was decreased in both radiated and irradiated samples. The reduction in ascorbic acid after irradiation treatment at 0.5 to 1.5KGy seems to indicate that radiolysis could accelerate the conversion of ascorbic acid to dehydroascorbic acid (Hussain and Gooneratne, 2017).

Carbohydrate content was decreased with time of storage in control samples whereas in irradiated samples much variation was observed between the different dosage treatments. Irradiation induced changes in nutritional value depend on several factors such as irradiation dosage, the temperature and atmosphere in which irradiation

Fig.1 : Exposure of sample to gamma irradiation. A- Gamma irradiation unit; B- Exposure of nion samples to gamma irradiation

Table.1 : Mycological and nutritional analysis of gamma irradiated onion sample.

Sl.No.	Storage period	Dosage (KGy)	Sprouting (%)	Ascorbic acid (mg/100g)	Carbohydrates (mg/100g)	Pyruvic acid (µ mol/g)	Incidence (%)
		Control	Nil	11.2±0.16	88.5±0.18	6.89±0.12	41.3±1.6
		0.15	Nil	11.1±0.20	88.3±0.35	6.26±0.16	31.8±1.1
1	0 day	0.5	Nil	11.0±0.13	87.9±0.16	5.13±0.11	32.6±2.3
		1.5	Nil	11.2±0.21	88.6±0.09	6.12±0.13	34.9±2.4
		Control	6.9	10.3±0.25	86.7±0.11	7.86±0.04	43.9±1.9
		0.15	Nil	9.6±0.12	91.1±0.23	6.69±0.13	29.9±1.7
2	30days	0.5	Nil	9.8±0.15	93.3±0.36	4.12±0.15	41.6±2.0
		1.5	Nil	9.8±0.15	96.4±0.22	5.69±0.11	48.6±2.6
		Control	14.2	9.7±0.08	81.3±0.16	9.6±0.25	49.3±1.5
		0.15	Nil	8.8±0.11	93.1±0.08	11.6±0.38	26.7±1.4
3	60days	0.5	Spoiled	NA	NA	NA	NA
		1.5	Spoiled	NA	NA	NA	NA
		Control	29.8	6.9±0.16	78.5±0.19	9.9	66.8±2.8
		0.15	Nil	8.4±0.09	82.7±0.06	12.9	26.1±0.9
4	90days	0.5	Spoiled	NA	NA	NA	NA
		1.5	Spoiled	NA	NA	NA	NA

Note: NA- Not analyzed (Onion samples irradiated at 0.5 and 1.5KGy were spoiled within 60 days of storage, hence analysis was not performed)

is performed, packaging and storage period (Hussain and Gooneratne, 2017). Overall change in nutritional aspects of onion samples subjected to irradiation dosage of 0.15KGy was insignificant. At doses below 1KGy, the nutritional losses are considered insignificant (Shea, 2000). Similar result was obtained by (Sharma *et al*. 2020) in their study where the ascorbic acid, total soluble solids and pyruvic acid content was not much altered in onion bulbs with 0.2KGy irradiation treatment along with

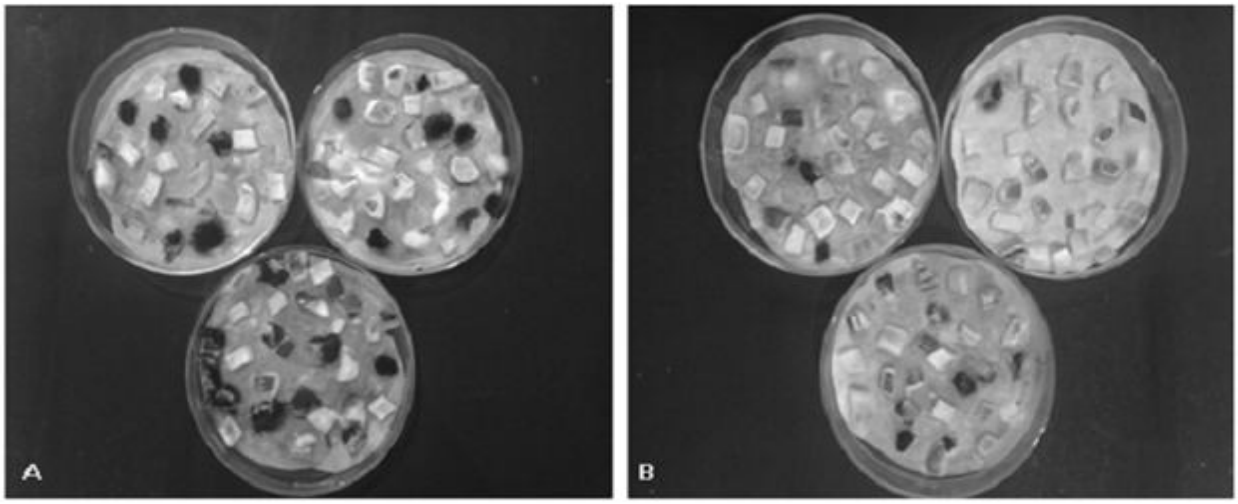

Fig.2 : Percent incidence of onion samples after 90 days of storage. A- Control samples; B- Samples irradiated at 0.15KGy

acceptable texture and colour up to three months. Irradiation dose of 0.13KGy was effective in storage of onions up to 64 days without significant loss of nutrients and pungency level along with retention of color and texture (Kallai *et al*. 2015). Carbohydrates are not significantly affected during irradiation at less than 10KGy (Aziz and Mahrous, 2007). In general macronutrients (protein, lipid, and carbohydrate) quality does not suffer due to irradiation and minerals have also been shown to remain stable.

In comparison to the adverse health and environmental impact of fungicides, application of gamma irradiation can be considered as a safe and effective post-harvest method for controlling fungal contamination causing deterioration of onions. Further Gamma irradiation at 0.15KGy improves the quality of onions by reducing fungal incidence and sprouting without affecting organoleptic properties.

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