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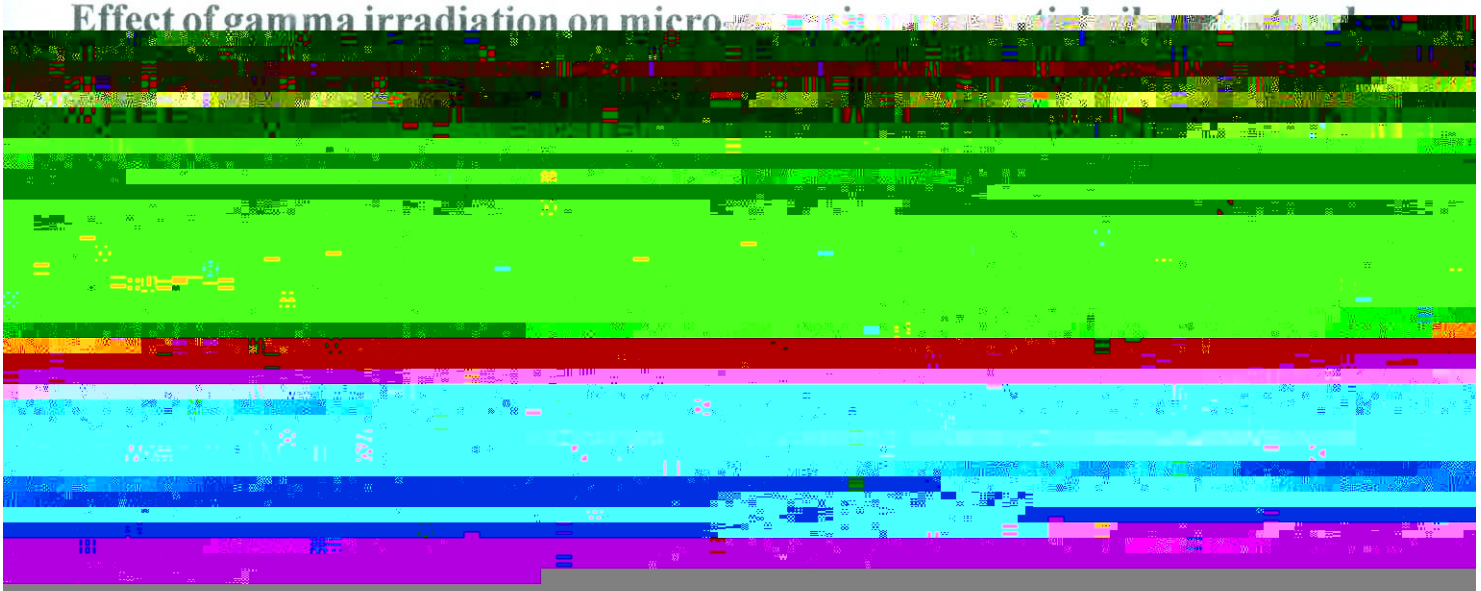
Effect of gamma irradiation on micro



Lanka at 3 different doses of 5 kGy, 7.5 kGy and 10 kGy respectively. Fricke dosimetry was used to measure the absorbed dose and dose calibration.

Un-irradiated (10g) and irradiated spice samples (10g) taken 24 hours after irradiation were each mixed with 99ml phosphate buffer solution. A serial dilution range was prepared by using phosphate buffer after allowing 5 min. for sedimentation.

Spread plate method was adopted to obtain total microbial counts. Potato dextrose agar medium was used for fungal cultures. Plate count agar medium was used to obtain aerobic mesophilic bacterial count, while violet red bile lactose agar was used for detection and enumeration of coliforms or enterobacteriaceae. Bacterial cultures were incubated at 37°C for 24 - 48 h, while fungal cultures were incubated at 30°C for 5 days. Identification of fungi and bacteria were carried out by studying their vegetative and asexual structures and *via* biochemical tests respectively.

All samples meant for storage studies were held at ambient temperature in the laboratory for 3 months and 6 months respectively. At a fixed time interval, designated samples were analyzed for total viable bacterial and fungal counts using spread plate method.

Essential oil content was obtained from finely milled irradiated and unirradiated spice samples weighing 50g, by steam distillation in a Deryng's apparatus. Gas chromatography (carbo wax 20m) was carried out using a Shimadzu GCL - 8 A FIDC chromatograph, and using a carbo wax 20 m column to study the composition of the volatile components of the essential oils of the irradiated and unirradiated spice samples. For gas chromatography the initial temperature was 70°C and the final temperature was 230°C for pepper and nutmeg. In the case of nutmeg the initial temperature was 100°C and the final temperature was 230°C. The program rate was 5°C/min. The temperature of the detector and the injector was 250°C.

A CRD design with 10 replicates was adopted in this study. The means were analyzed using Duncan Multiple Range Test at 5% level of significance.

RESULTS AND DISCUSSION

The effect gamma irradiation on bacterial and fungal populations in pepper, cardamom and nutmeg are given in Table : 1.

A progressive increase in the dose showed a concomitant decrease in the total viable fungal and bacterial counts in all the spices. The results obtained indicate that 5 kGy gamma irradiation dose was sufficient to reduce the total viable fungal count of pepper from 4.52×10^3 cfu/g to 2.9×10^2 cfu/g. The total viable bacterial count of pepper was reduced from 3.21×10^6 cfu/g to 2.00×10^4 cfu/g. The 7.5 kGy gamma irradiation dose reduced the total viable bacterial count of pepper 3.21×10^6 cfu/g to 1.00 x

Table 1. Effect of gamma irradiation on total viable bacterial count and total viable fungal counts of pepper, cardamom and nutmeg after 24 h, 3 months and 6 months storage

Spice Commodity	Irradiation treatment kGy	24 h. Storage		3 months storage		6 months storage	
		TVBC† cfu/g‡	TVFC† Cfu/g	TVBC cfu/g	TVFC cfu/g	TVBC cfu/g	TVFC cfu/g
Pepper	0	3.21×10^6	4.52×10^3	4.02×10^6	1.00×10^3	4.25×10^7	1.20×10^7
	5	2.20×10^4	2.90×10^2	2.91×10^2	0	2.89×10^2	0
	7.5	1.0×10^2	0	0	0	0	0
	10	0	0	0	0	0	0
Cardamom	0	1.20×10^3	2.00×10^2	1.10×10^3	1.90×10^2	1.00×10^3	1.70×10^2
	5	30	0	28	0	26	0
	7.5	0	0	0	0	0	0
	10	0	0	0	0	0	0
Nutmeg	0	1.02×10^4	4.30×10^2	1.00×10^4	4.10×10^2	9.00×10^3	3.90×10^2
	5	3.00×10^2	0	2.9×10^2	0	2.7×10^2	0
	7.5	0	0	0	0	0	0
	10 kGy	0	0	0	0	0	0

†TVBC - Total Viable Bacterial Count
‡Cfu/g - colony forming unit per gram
†TVFC - Total Viable Fungal Count

Table 2. Effect of Gamma Irradiation on major essential oil v/w content of pepper, Cardamom and Nutmeg ml/100 g.

Spice Commodity	Irradiation dose kGy	Essential oil content mg $100g^{-1}$
Paper	0 (control)	3.1_a
	5	3.1_a
	7.5	3.2
Cardamom	0 (control)	6.3_b
	5	6.2_b
	7.5	6.4_b
	10	6.3
Nutmeg	0 (control)	4.9_c
	5	4.9_c
	7.5	5.0_c
	10	5.5

Values within the column followed by the same letters are not significant according to Duncan 's Multiple Range Test at 5% probability.

Table 3. Effect of gamma irradiation on major volatile oil components or pepper, after 24h. Respective volatile oil components expressed as a percentage of the total essential oil content recorded on GLC.

Irradiation Dose (kGy)	Pinene L (%)	Pinene B (%)	Sabinene (%)	Limonene (%)	Caryophyllene (%)
0 (control)	15.82_a	23.04_b	13.12_c	18.73_d	18.16_c
5	16.0_a	21.09_b	12.85_c	18.76_d	17.96_c
7.5	15.91_a	20.90_b	12.89_c	18.75_d	17.81
10	16.03	21.88	13.10	19.10	17.81

Values within the column followed by the same letters are not significant according to Duncan 's Multiple Range Test at 5% probability.

Table 4. Effect of gamma irradiation on volatile oil components of cardamom.

Does	Pinene (%)	Sabinene (%)	Limonene (%)	Terpineol (%)	Linalyl Acetate(%)	Terpenyl Acetate (%)
0 kGy (control)	2.29^a	1.08^b	46.54^c	2.10^d	3.00^c	32.90^f
5 kGy	2.60^a	1.18^b	48.95^c	2.00^d	3.16^c	30.98^f
7.5 kGy	2.62^a	1.166^b	48.82^c	2.03^d	3.00^c	30.71^f
10 kGy	2.56^a	1.168^b	50.05^c	2.00^d	3.01^c	30.02^f

Values within the column followed by the same letters are not significant according to Duncan 's Multiple Range Test at 5% probability.

Table 5. Effect of gamma irradiation on volatile oil components of nutmeg.

Doses	Sabinene (%)	Phellandrene (%)	Cineole (%)	Terpineol (%)	Elemicene (%)
5 kGy	49.20^a	5.47^b	6.50^c	5.91^a	2.72^c
5 kGy	43.41^a	5.12^b	6.40^c	7.00^c	3.20^c
7.5 kGy	47.95^a	4.91^b	5.50^c	6.18^d	3.30^c
10 kGy	50.06^a	4.41^b	5.91^c	6.02^d	2.92^c

Values within the column followed by the same letters are not significant according to Duncan 's Multiple Range Test at 5% probability.

10^2 cfu/g, and it completely eliminated the fungi from the samples. A dose of 10 kGy values within the column followed by the same letters are not significant according to Duncan's Multiple Range Test at 5% Probability.

Effectively eliminated all bacetria and fungi from pepper. These observations are in agreement with the observation of Munasiri *et al*(1987).

The 5 kGy gamma irradiation reduced the total viable bacterial count of cardamom from 1.20×10^3 cfu/g to 30 cfu/g. It was evident from this study that gamma irradiation was effective in the elimination of fungi from these samples.

The results obtained in this study indicate that the total viable bacterial count of nutmeg was reduced from 1.02×10^4 cfu/g to 3.00×10^2 cfu/g at 5 kGy gamma irradiation. This dose completely eliminated the fungi from nutmeg samples. A dose of 7.5 kGy was effective in eliminating all bacteria and fungi from cardamom and nutmeg resulting in zero counts(Table 1).

It is evident from Table 1 that there was only little variation in total viable bacterial counts and fungal counts of irradiated spice samples following 3 months and 6 months storage. However in unirradiated samples fungal and bacterial counts were significantly lower after 3 months and 6 months storage. It is possible that the anaerobic environment created by CO_2 build up within the packs due to respiratory activity of the surviving micro organisms was responsible for the slight decline in total viable microbial counts in these instances. Observation thus indicate that gamma irradiation reduced the total viable microbial load of the spice samples considered in this study, and enabled quality maintenance of spices, during storage periods extending to 6 months with no microbial decontamination.

It was observed that in unirradiated pepper samples, the total viable bacterial and fungal count increased with storage time. The total viable bacterial and fungal counts were high in unirradiated pepper due to the high relative humidity and temperature (28°C - 30°C) in the ambient storage environment.

The fungi associated with pepper, cardamom and nutmeg were identified as *Aspergillus niger, Aspergillus flavus, Pencillium ittalicum*, and *Rhizopus* sp., while bacterial contamination was attributed to *Bacillus* sp, *Pseudomonas* sp, *E. coli* and *Streptococcus sp.*

Results indicate that there was no significant difference between the essential oil content and volatile oil components of irradiated and unirradiated pepper, cardamom and nutmeg samples as presented in Table: 2, 3, 4 and 5. This study indicates that gamma irradiation technology could be beneficial to the spice industry in reducing microbial contamination and in storage quality maintenance.

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