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### Shelf Life Extension of Mosambi (Citrus sinensis) by gamma irradiation

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#### ABSTRACT

Changes in quality characteristics of mosambi (Citrus sinensis) irradiated by doses of 0.5 and 1.0 KGy and stored at ambient (20-25<sup>0</sup>C, 65-75% RH) and refrigerated (8-10<sup>0</sup>C, 80-85%RH) condition were studied for a period of one month. Irradiation at both the dosage levels in combination with ambient storage resulted in increased ripening during storage. However, storing at refrigerated condition helped in maximum retention of fruit quality characteristics such as, physiological loss in weight, total soluble solids, titrable acidity, vitamin-C and microbial quality. The data revealed that gamma irradiation at dose level of 0.5 KGy in combination with refrigerated storage proved to be effective for extending the shelf life of fresh mosambi fruit than 1.0KGy dose level.

**Key words:** mosambi, gamma irradiation, physiological loss in weight, total soluble solids, titrable acidity, shelf life.

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## INTRODUCTION

Citrus occupies a place of considerable importance in tropical and subtropical fruits grown all over the world. Saathgudi variety of mosambi is cultivated commercially to a greater extent in India. In the developing country like India, post harvest losses of citrus fruits are in the range of 25-30 percentage as against 5-10 percentage in developed citrus growing countries like Brazil, USA and Australia [1]. Proper storage of citrus fruits for extended period is very essential for the utilization of fruit in the glut season.

With the ready availability of isotopic sources, research of unprecedented magnitude on food irradiation has been conducted in many countries. Radiation energy can prove to be the most effective technique to improve the keeping quality of food in comparison with many traditional processing techniques, as they cause undesirable and harmful effects on the physical, chemical and sensory qualities of food.

The extent to which irradiation effects on the quality characteristics of foods varies widely and depends on dosage used, chemical composition and physical structure of food [2].

Citrus fruits are very sensitive to gamma irradiation and to optimize the effectiveness of ionizing treatments of citrus fruits, the control of some parameters has been reported to be very important namely dosage level, storage temperature and the original quality of fruits [3]. Hence, the present investigation was undertaken to identify the gamma irradiation dose level which helps in minimizing loss in quality of mosambi fruit. In addition, the best combination treatment with respect to enhanced fruit quality at ambient and low storage temperature was also investigated.

## MATERIALS AND METHODS

The fresh well graded saathgudi variety of mosambi (*Citrus sinensis*) fruit having uniform size, shape, firmness and colour were selected in one lot to minimize variations among treatments. Samples were further divided into two groups based on the dosage levels. They were placed in 8cm×11cm ziplock bags. The packages containing the fruits were randomly assigned to each treatment. Non irradiated set of packets which served as control was also placed in ziplock bags. The samples were coded as MT<sub>1</sub>- 0.5KGy treated and stored under ambient conditions; MT<sub>2</sub> – 0.5kGY treated and stored under refrigerated conditions, MT<sub>3</sub> -1KGy treated and stored under ambient conditions, MT<sub>4</sub>-1KGy treated and stored under refrigerated conditions, MU<sub>1</sub>-untreated and stored under ambient conditions, MU<sub>2</sub>-untreated and stored under refrigerated conditions.

Gamma chamber 5000 supplied by BRIT, Department of Atomic energy, Mumbai, was used for giving the irradiation treatment with cobalt 60 as a source . The packed fruits were subjected to radiation at dosage levels of 0.5KGy and 1KGy. The fruits were irradiated at a mean dose rate of 4.04Kg/hr. The time taken to irradiate fruits at 0.5KGy and 1.0KGy was 7.26

min and 14.51 minutes and the samples were maintained at temperature  $18 \pm 2^{\circ}\text{C}$ . Upon irradiation, the zero day packs were analyzed and the remaining packs were transferred for storage at ambient ( $20 \pm 2^{\circ}\text{C}$ ) and refrigerated conditions ( $8 \pm 2^{\circ}\text{C}$ ). Periodic evaluations were done on day 1, 7, 14, 21 and 28 (end of storage). All runs in the experiment were prepared in triplicates.

Observations were made regarding physiological loss of weight (PLW), fruit distribution percentage, total soluble solids (TSS), pH, acidity, pectin, vitamin C and microbial quality.

**Physiological loss in weight:** PLW of fruits were weighed periodically on a physical balance and the loss in weight was calculated as physiological loss in weight.

$$\text{PLW}\% = (W_i - W_s / W_i) \times 100$$
. Where  $W_i$  = initial weight,  $W_s$  = weight at sampling period.

**Fruit distribution percentage:** Distribution of various components in fruits has been determined by the method given by Kudachikar et al., (2003) [5].

**Total soluble solids :** TSS content was determined by using Erma hand refractometer having a range of 0-32%. Two-three drops of the filtered juice were put on the refractometer lens for TSS measurement and expressed as  $^{\circ}\text{Brix}$  making temperature adjustment at  $20^{\circ}\text{C}$ .

**pH :** Ten ml of juice was weighed. Mixture was allowed to stand for 30 minutes. The supernatant was decanted and collected. pH meter was calibrated with standard buffer solution. Electrode was dipped into the supernatant and reading was noted.

**Titration acidity:** Titration acidity as percent malic acid was determined by the titration method as described by Ranganna [4].

**Pectin:** Pectin was measured by Carna and Haynis method as described by Ranganna [4].

**Vitamin C:** Vitamin C was estimated as described by Ranganna [4]. The 2, 6 Dichlorophenolindophenol method was used to assay vitamin C content in juice by colorimeter. A wavelength of 520 nm was used to determine total vitamin C content [4].

**Microbial count:** Yeast and mold of irradiated and non irradiated fruits was determined by the method of serial dilution using potato dextrose agar (PDA) media [10]. One gram of juice was taken and dissolved in previously sterilized 9 ml of distilled water. One ml of this solution was further diluted by dissolving in 9 ml of distilled water. This way a dilution of  $10^{-4}$  was obtained. One ml of aliquot each of  $10^{-4}$  was placed on three petri dishes containing the PDA medium and incubated for 48 hrs at  $30 \pm 2^{\circ}\text{C}$ . The colonies so formed were counted and expressed as log cfu/g of sample.

**Statistical analysis:** All the determinations were made in 3 different batches. Statistical analysis of data was carried out by Duncan’s multiple range method using Sys-Cat software package and the differences between treatments were considered significant at  $p < 0.05$ .

## RESULTS AND DISCUSSION

### Physiological loss in weight (PLW)

PLW results are presented in Fig.1. Least PLW was found to be registered by 0.5KGy irradiated sample (MT<sub>2</sub>) stored at 15°C. There was a significant difference ( $p < 0.05$ ) among the treatments and days of storage at ambient and refrigerated conditions. On the other hand, maximum PLW was noticed in treated and untreated samples stored under ambient conditions during 28 days storage. In fruits stored under ambient conditions the increase was obvious from 7<sup>th</sup> day where as, for fruits stored under refrigerated conditions, increase in PLW was negligible till the end of the storage period.

The weight loss can be mainly said to occur due to transpiration. Transpiration not only caused desiccation, shriveling, accelerated softening and loss of attractive appearance of fruit but the resultant water stress also accelerated the senescence [1]. However, the minimum reduction in weight loss of refrigerated samples is attributed to the synergistic effect of gamma irradiation and refrigerated storage on rate of respiration and senescence, as irradiation at low doses is known to delay the process of senescence in fruits and vegetables [6].

### Fruit distribution % (Pulp, Peel and juice distribution)

Water loss and softening are the major deteriorative changes that take place during storage of sweet oranges [8].

**Table 1. Effect of Gamma irradiation on Distribution of various components (%) of mosambi fruit.**

TREATMENTS	STORAGE PERIOD, DAYS				
	0	7	14	21	28
MT <sub>1</sub>	Pu – 41 <sup>a</sup> ± 1.63 Pe- 42 <sup>a</sup> ± 1.69 J – 15 <sup>a</sup> ± 2.05	Pu – 41 <sup>a</sup> ± 1.24 Pe - 32 <sup>a</sup> ± 1.24 J – 24 <sup>a</sup> ± 2.05	Pu – 36 <sup>a</sup> ± 1.24 Pe – 29 <sup>a</sup> ± 2.01 J – 33 <sup>a</sup> ± 2.05	Pu – 41 <sup>a</sup> ± 1.63 Pe- 26 <sup>a</sup> ± 2.05 J – 31 <sup>a</sup> ± 1.24	Pu – 25 <sup>a</sup> ± 1.65 Pe – 27 <sup>a</sup> ± 1.63 J – 50 <sup>a</sup> ± 2.44
MT <sub>2</sub>	Pu – 50 <sup>b</sup> ± 2.05 Pe- 24 <sup>a</sup> ± 2.05 J – 23 <sup>a</sup> ± 2.05	Pu – 41 <sup>a</sup> ± 3.26 Pe-34 <sup>b</sup> ± 2.86 J – 24 <sup>b</sup> ± 2.87	Pu – 42 <sup>b</sup> ± 2.87 Pe-33 <sup>b</sup> ± 2.05 J – 28 <sup>a</sup> ± 2.05	Pu – 41 <sup>a</sup> ± 2.86 Pe-28 <sup>a</sup> ± 2.05 J – 28 <sup>a</sup> ± 2.05	Pu – 38 <sup>a</sup> ± 2.05 Pe-27 <sup>b</sup> ± 1.63 J – 35 <sup>a</sup> ± 2.05
MT <sub>3</sub>	Pu – 45 <sup>c</sup> ± 2.44 Pe – 39 <sup>a</sup> ± 2.05 J – 15 <sup>b</sup> ± 1.69	Pu – 39 <sup>b</sup> ± 2.05 Pe-32 <sup>c</sup> ± 2.05 J – 29 <sup>c</sup> ± 2.82	Pu – 42 <sup>c</sup> ± 2.05 Pe-26 <sup>c</sup> ± 2.86 J – 30 <sup>b</sup> ± 2.05	Pu – 45 <sup>b</sup> ± 2.05 Pe-27 <sup>b</sup> ± 2.86 J – 31 <sup>c</sup> ± 1.63	Pu – 33 <sup>b</sup> ± 1.63 Pe-26 <sup>c</sup> ± 2.44 J – 24 <sup>a</sup> ± 2.44
MT <sub>4</sub>	Pu – 47 <sup>c</sup> ± 2.44 Pe - 37 <sup>a</sup> ± 1.63 J – 17 <sup>c</sup> ± 1.28	Pu – 46 <sup>b</sup> ± 2.05 Pe-29 <sup>c</sup> ± 2.05 J – 24 <sup>c</sup> ± 0.82	Pu – 43 <sup>c</sup> ± 2.44 Pe-34 <sup>c</sup> ± 2.05 J – 25 <sup>c</sup> ± 1.63	Pu – 43 <sup>b</sup> ± 2.05 Pe-26 <sup>b</sup> ± 2.05 J – 29 <sup>c</sup> ± 2.86	Pu – 39 <sup>b</sup> ± 2.05 Pe-35 <sup>c</sup> ± 2.49 J – 26 <sup>a</sup> ± 2.49
MU <sub>1</sub>	Pu – 41 <sup>d</sup> ± 2.86 Pe – 39 <sup>b</sup> ± 1.63	Pu – 39 <sup>b</sup> ± 2.44 Pe-34 <sup>d</sup> ± 2.05	Pu – 35 <sup>d</sup> ± 2.05 Pe-31 <sup>c</sup> ± 2.05	Pu – 38 <sup>c</sup> ± 1.63 Pe-31 <sup>c</sup> ± 2.86	Pu – 38 <sup>c</sup> ± 2.05 Pe-27 <sup>d</sup> ± 2.05

	J-20 <sup>d</sup> ± 1.63	J-24 <sup>d</sup> ± 1.63	J-33 <sup>d</sup> ± 2.05	J-33 <sup>d</sup> ± 2.44	J-39 <sup>a</sup> ± 2.86
MU <sub>2</sub>	Pu-45 <sup>d</sup> ± 1.63	Pu-43 <sup>b</sup> ± 2.05	Pu-42 <sup>d</sup> ± 2.44	Pu-47 <sup>d</sup> ± 2.05	Pu-45 <sup>d</sup> ± 2.05
	Pe-38 <sup>c</sup> ± 2.44	Pe-37 <sup>d</sup> ± 2.05	Pe-39 <sup>c</sup> ± 2.44	Pe-31 <sup>d</sup> ± 2.86	Pe-32 <sup>d</sup> ± 2.40
	J-18 <sup>e</sup> ± 1.63	J-20 <sup>e</sup> ± 2.05	J-14 <sup>e</sup> ± 2.05	J-22 <sup>e</sup> ± 2.05	J-25 <sup>a</sup> ± 2.05

Values are mean ± SD of tropical samples, means with in columns for given parameter bearing different superscript are significantly different at p < 0.05.

**Table 2. Effect of gamma irradiation on Total soluble solids (°Brix) of mosambi Fruit**

TREATMENTS	STORAGE PERIOD, DAYS				
	0	7	14	21	28
MT <sub>1</sub>	6.60 <sup>a</sup> ± 0.08	7.45 <sup>b</sup> ± 0.04	8.25 <sup>a</sup> ± 0.04	10.1 <sup>a</sup> ± 0.04	11.4 <sup>a</sup> ± 0.06
MT <sub>2</sub>	6.55 <sup>a</sup> ± 0.04	6.85 <sup>b</sup> ± 0.04	7.05 <sup>b</sup> ± 0.04	7.25 <sup>b</sup> ± 0.04	7.45 <sup>b</sup> ± 0.04
MT <sub>3</sub>	6.45 <sup>a</sup> ± 0.04	7.25 <sup>c</sup> ± 0.04	8.01 <sup>c</sup> ± 0.08	9.45 <sup>c</sup> ± 0.04	12.1 <sup>c</sup> ± 0.04
MT <sub>4</sub>	6.55 <sup>a</sup> ± 0.04	6.75 <sup>c</sup> ± 0.04	6.95 <sup>c</sup> ± 0.04	7.45 <sup>c</sup> ± 0.04	7.75 <sup>c</sup> ± 0.04
MU <sub>1</sub>	6.50 <sup>b</sup> ± 0.08	6.85 <sup>d</sup> ± 0.04	7.40 <sup>d</sup> ± 0.08	7.95 <sup>c</sup> ± 0.05	8.55 <sup>c</sup> ± 0.04
MU <sub>2</sub>	6.55 <sup>c</sup> ± 0.04	6.75 <sup>d</sup> ± 0.04	7.05 <sup>d</sup> ± 0.04	7/25 <sup>d</sup> ± 0.04	7.35 <sup>c</sup> ± 0.04

Values are mean ± SD of tropical samples, means with in columns for given parameter bearing different superscript are significantly different at p < 0.05.

The results on percentage distribution of components of fruits such as pulp, peel and juice revealed that there was a significant difference (p < 0.05) between the samples with regard to the percentage distribution of components and also a significant interaction effect between the samples and the storage period (Table 1).

During the initial stages, percent of juice (15-20%) was less for treated and untreated samples, when compared to pulp and peel, which ranged from 41-50% and 24-42% respectively.

At the end of storage period, highest juice yield (50%) was observed for 0.5KGy irradiated sample (MT<sub>1</sub>) stored under ambient conditions and least percent (25%) was observed for 0.5KGy treated (MT<sub>3</sub>) sample stored under refrigerated conditions.

This increasing trend in juice yield for samples stored under ambient conditions may be attributed to maturation of fruit as juice content in citrus fruits shows a positive correlation with the harvest maturity [7]. Lesser juice yield in refrigerated samples can be due to inhibitory effect of low temperature on fruit respiration and maturity.

At ambient storage, a decreasing trend in peel weight was observed for treated and untreated samples where as much reduction was not seen in refrigerated samples. 1KGy had a slightly more pronounced effect on peel than lower dose (0.5KGy). Throughout the entire period of study, peel thickness reduced to a greater extent especially in samples irradiated at 0.5 and 1KGy stored under ambient conditions. This phenomenon can be attributed to the profound sample moisture loss in the samples.

### **Total Soluble solids (TSS)**

An increased trend was observed in TSS content for all the samples and significant interaction effect was observed between treatments and storage period. At the beginning of storage, increased TSS was observed in all the samples maximum being in 1KGy irradiated sample (MT<sub>3</sub>) which was on par with 0.5KGY irradiated sample stored under ambient conditions (MT<sub>1</sub>). Increase in TSS was much pronounced in the later stages of storage with 1KGy irradiated sample (MT<sub>3</sub>) recording the highest percentage increase in TSS followed by 0.5KGY irradiated sample stored under ambient condition (MT<sub>1</sub>). Least percentage increase was found in control samples stored under refrigerated conditions. This increasing trend in TSS can be due to hydrolysis of acid and polysaccharides in to simple sugars and due to the loss of moisture which occurs during the normal ripening of the fruits [9]. (Table 2). Higher TSS values in 1KGy and control samples can be attributed to the solubilization of pectin substances and rapid ripening [10].

### **Pectin**

A common characteristic underlying tissue softening during fruit ripening is partial degradation of pectic polysaccharides, with pectin solubilization and liberation of uronic acids, galactose and arabinose [13].

Fig. 1. shows the effect of irradiation on pectin content of mosambi fruit. All the samples in the study exhibited increasing trend throughout the storage period. It has been statistically proved that there is a significant interaction ( $p < 0.05$ ) between treatments and storage period. Treated fruits (0.5 KGy and 1.0KGY) stored under ambient conditions (MT<sub>1</sub> and MT<sub>3</sub>) had the maximum increase in pectin content i.e, 84.7% and 86.1% respectively. The level of increase in treated samples stored under refrigerated conditions was less than the fruits stored under ambient conditions. It is also evident that there was more significant increase in pectin content for all the treatments up to 15 days, whereas from 15 days till the end of storage, percentage increase in pectin content was less. The changes observed in the present study are in line with the hypothesis that the insoluble proto pectin and pectate fractions are broken down to soluble form by radiation [13]. Decrease in fruit firmness and softening in oranges are due to enzymatic degradation of insoluble pectin to the more soluble pectic acid and pectinates.

### **pH**

An increasing trend in pH was observed throughout the storage period, though it was not very obvious. Most of the samples exhibited a slight increase in pH up to 15 days of storage, after which a negligible increase was observed in refrigerated samples.

Increase in pH can be correlated to the decreased acidity and increased sugar content which occur due to utilization of organic acids as reducing substrates.

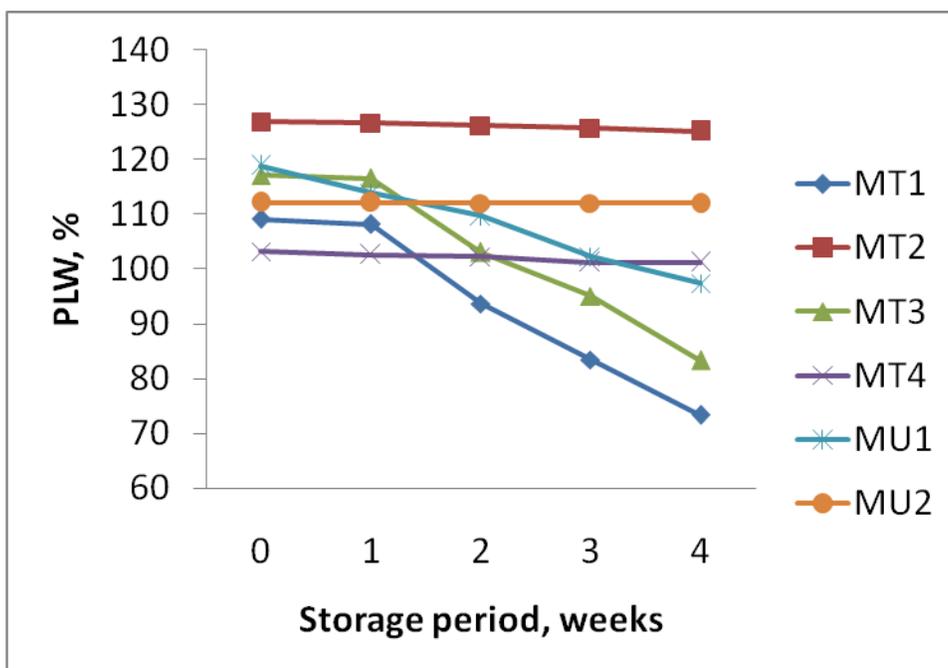


Figure 1. Effect of gamma irradiation on physiological loss in weight (PLW) of mosambi fruit during storage

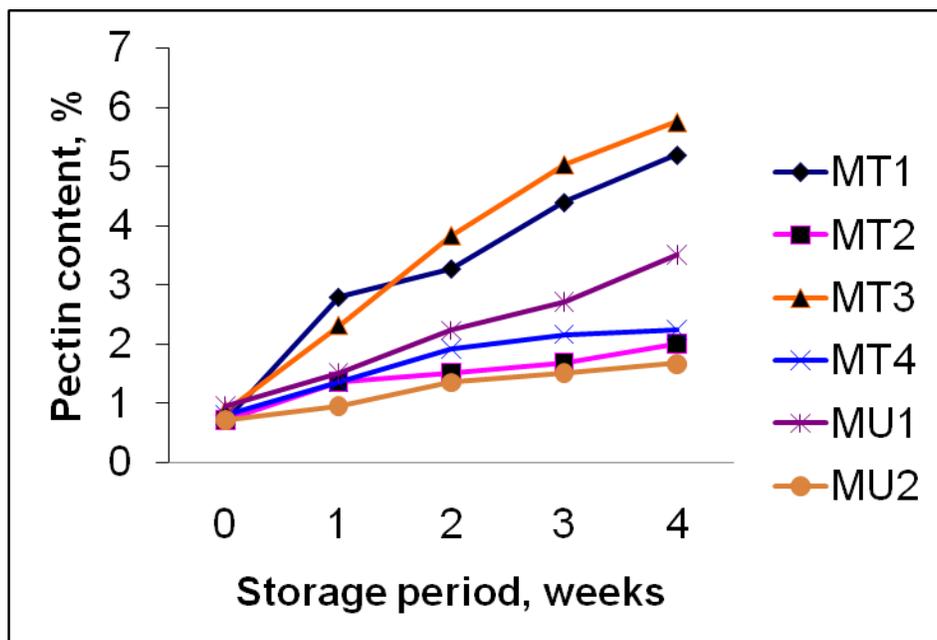


Figure 2. Effect of gamma irradiation on pectin content of mosambi fruit during storage

**Titration acidity**

The data on titrable acidity of the treated and untreated mosambi fruits shows a significant difference ( $p < 0.05$ ) between the treatments. Maximum percentage decrease (14.8%) was recorded by control samples ( $MU_1$ ) followed by 1KGy irradiated sample (13.9%) stored under ambient conditions ( $MT_3$ ). Least decrease (6.0%) in titrable acidity was observed in 0.5KGy irradiated and control samples stored under refrigerated conditions, by the end of storage period.

The rate of decrease in acidity was consistent throughout the storage period which might be either due to dilution effect (or) hydrolysis of acid in to sugar [11].

Studies have shown that titrable acidity was lower in citrus fruits stored at  $6 \pm 1^\circ \text{C}$  and  $8 \pm 1^\circ \text{C}$  than in fruits stored at  $20\text{-}30^\circ \text{C}$  [8]. Loss in acidity may be largely due to the utilization of organic acids as respiration substrates and as carbon skeletons for synthesis of new compounds during ripening [12].

### **Vitamin C**

A significant difference ( $p < 0.05$ ) was found among the samples with regard to the Vitamin C content. It is clear from the fig.1 that the samples irradiated and stored under refrigerated conditions had a comparatively less decrease in Vitamin C content over the entire 30 days. However, treated and untreated samples stored and ambient conditions have significant loss in Vitamin C content.

So, it can be concluded that irradiating and storing at low temperature had a beneficial effect in minimizing Vitamin C loss. This behaviour is in agreement with previous works that have reported that irradiation of ranges at doses ranging between 0.3 and 2KGy does not affect content of Vitamin C throughout the storage period at various conditions like 7 weeks at  $7^\circ \text{C}$  [3].

It is also possible that high levels of antioxidants present in the fruit may quench free radicals generated by irradiation, avoiding the reaction of free radicals with Vitamin C and thereby preventing its degradation [14].

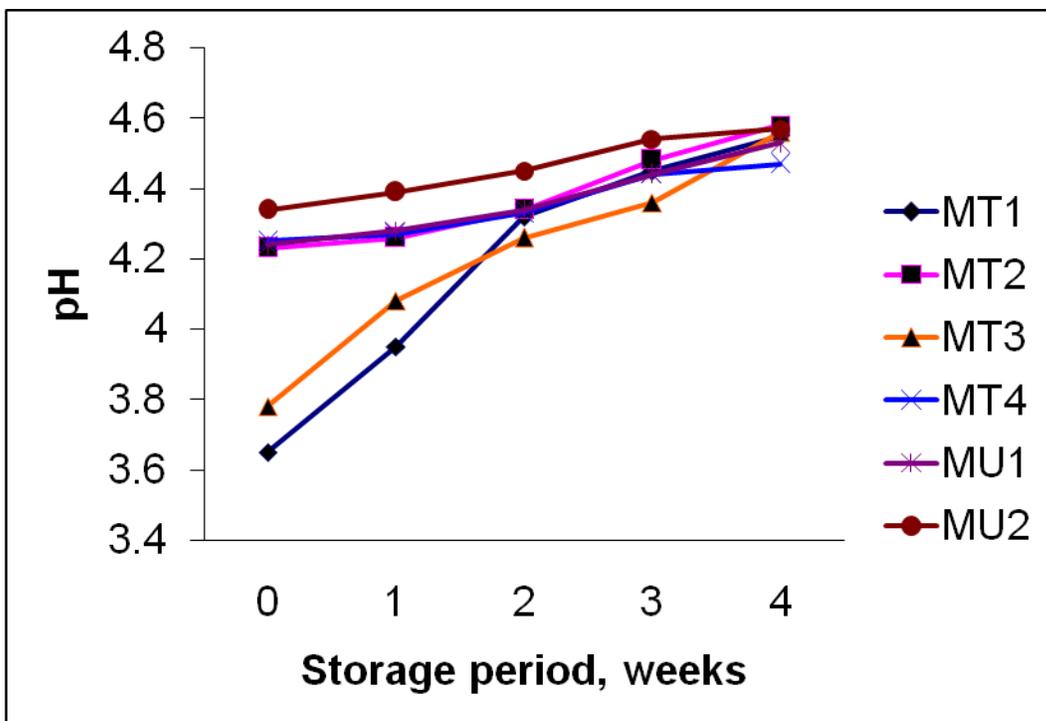


Figure 3. Effect of gamma irradiation on pH of mosambi fruit during storage

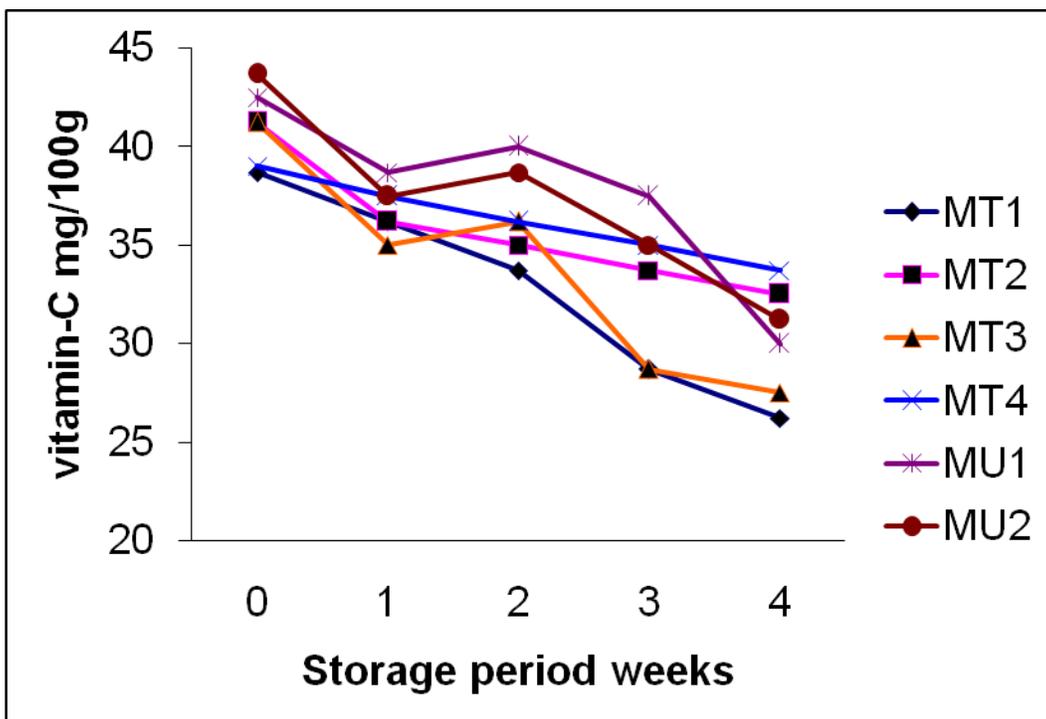


Figure 4. Effect of gamma irradiation on vitamin C content of mosambi fruit during storage

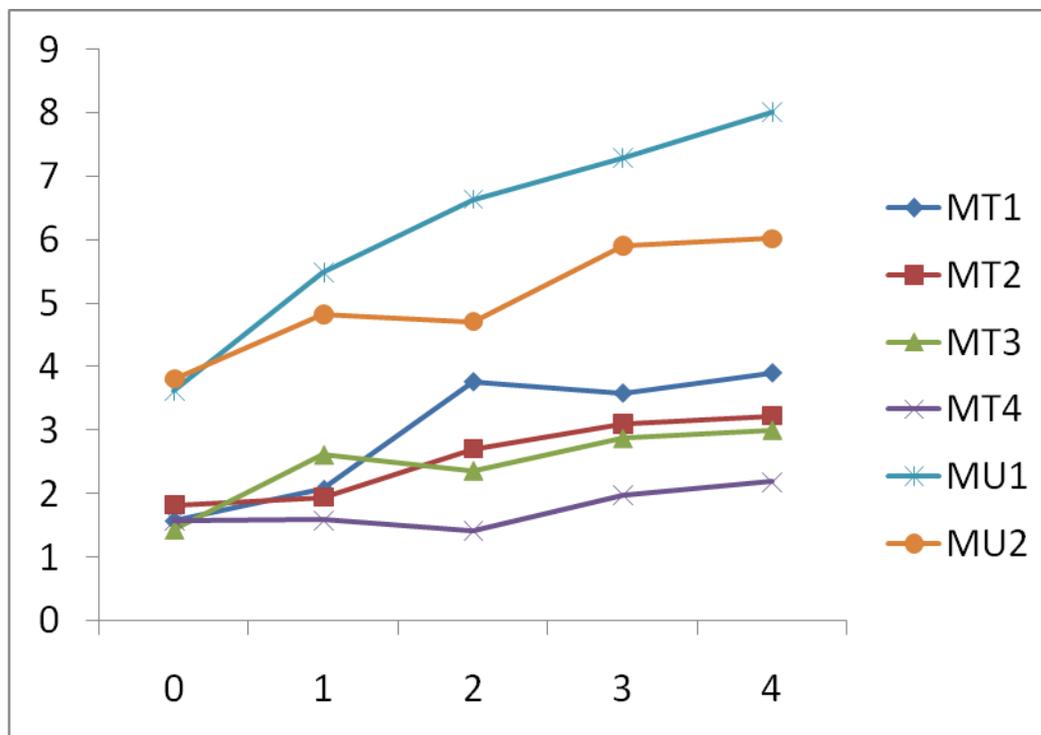


Figure 5. Effect of gamma irradiation on microbial quality (Yeast and mold count) of mosambi fruit during storage

### Yeast and mold count

The yeast and mold counts of mosambi have markedly reduced by the irradiation treatment which further decreased with increase in the irradiation dose. The least increase in yeast and mold count was observed for 1 KGy irradiated samples ( $2.19 \pm 0.34$  log cfu/ml sample) stored under refrigerated conditions whereas maximum increase was observed for control samples ( $8.02 \pm 0.15$  log cfu/ml), stored under ambient conditions.

### CONCLUSION

The physiological state of citrus fruits at the time of irradiation influences the fruits capacity to with stand high doses of irradiation and the present study clearly indicated the necessity of establishing the maximum tolerable dosage for citrus fruits. It was apparent from the data that irradiation of the fruits at 0.5 and 1KGy doses and storage under ambient conditions had a rather negative impact on the shelf life and nutritional properties. However, control samples were comparable to 0.5KGy refrigerated for some of the quality attributes and considerable decrease in some attributes. The results indicated the effectiveness of gamma irradiation for extending the shelf life of mosambi. The radiation treatment in combination with refrigeration maintained acceptable quality storage for 28 days.



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