

## Induction of $\gamma$ irradiation for decontamination and to increase the storage stability of black teas

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### Abstract

The handling and storage of black tea at high humidity increases its moisture content, which encourages microbial growth.  $\gamma$  irradiation is one of the most efficient techniques for the reduction of microorganisms in food. The present study aimed to evaluate the effect of  $\gamma$  irradiation on black tea quality and microbial load, during storage. Irradiation had a significant effect on reducing the microbial load, while untreated samples had a high microbial load. Untreated samples absorbed moisture up to 8.65% in 12 months time, whereas it was 6.0% in the case of irradiated samples. The quality of the irradiated teas exhibited stability in major parameters like theaflavin (TF), thearubigin (TR), highly polymerized substances (HPS), and total liquor colour (TLC). There was no significant difference among the quality characteristics of 7 kGy and 10 kGy irradiated samples during the entire period of storage when compared with 0 day control. Results indicated a deterioration in quality of well packed, but untreated control samples during storage. Irradiation resulted in complete removal of microbes and extended the shelf life of black tea.

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*Keywords:*  $\gamma$  irradiation; Black tea; Microbial load; Moisture content; Quality parameters

### 1. Introduction

The greatest strength of the Indian tea industry is that it produces wide varieties of tea. Biochemical constituents in relation to the environmental and edaphic factors contribute to its uniqueness. Black teas are preferred by the drinkers due to the refreshing taste and unique aroma. The black tea aroma is composed of more than 600 compounds, which include various group of compounds, viz., aldehydes, alcohols, terpenes, nitro compounds, hydrocarbons, etc. Major compounds of black tea aroma are linalool and its oxides, methyl salicylate, geraniol, *trans*-2-hexenal and *cis*-3-hexenol.

Manufactured tea is stored for some time before it reaches the consumer. Storage results in the microbial contamination of the processed tea (Bouakline, Lacroix, Roux,

Gangneux, & Derouin, 2000; Wilson, Dettenkofer, Jonas, & Daschner, 2004). If tea leaves are contaminated by pathogenic microorganisms, the tea infusion may pose a potential health risk (Hauer, Jonas, Dettkekofer, & Daschner, 1999). Presence of fungal strains and aflatoxin are also reported in tea (Elshafie, Allwatia, & Albahy, 1999; Hasah & Abdelsater, 1993; Yde, Rillaer, & Maeyer-Cleempoel, 1981).

Irradiation has become an effective means of processing and preserving food products (Fan, Niemira, & Sokorai, 2003; Molins, 2001; WHO, 1988).  $\gamma$  irradiation has been employed for the decontamination and/or sterilization of dehydrated vegetables, fruits, seasonings and animal foods, and to prolong the storage period (Chwla et al., 2003; Fu, Shen, Bao, & Chen, 2000; Mahrou, Caillet, Nketsa-Tabiri, & Lacroix, 2003). Recently, codex general standard for irradiated foods (CAC, 2003) and the food and drug administration (FDA), amended the food additive regulation to provide control of food borne pathogens in foods (FDA, 2005).  $\gamma$  irradiation of food, up to an overall dose of 10 kGy, is acceptable in several countries for commercial

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food processing (Lacroix & Quattara, 2000).  $\gamma$  irradiation can extend the shelf life of treated foods without inducing the formation of any radionuclide in food products. World Health Organization (WHO) expert committee on the wholesomeness of irradiation of food, has ruled that foods subjected to low dosages (10 kGy) of irradiation are safe and do not require toxicological testing (WHO, 1981). Although studies have been made in many food products on the effect of  $\gamma$  irradiation, no such study has been made in tea, to the best of our knowledge. The present study investigated the effect of  $\gamma$  irradiation on microbial growth in black tea and stability of quality parameters during long-term storage.

## 2. Materials and methods

### 2.1. Sample preparation and storage

Commercial black tea of CTC and orthodox types (both BOP grades) were obtained from tea manufacturing unit, Coonoor, The Nilgris, Tamil Nadu, India. Tea samples were packed separately in metallized polyester pouches. Twenty four such packets for each type were made, each containing 250 g of tea. Half of these packets were used for  $\gamma$  irradiation and the remaining packets served as untreated control. Each 250 g packet of tea was stored at room temperature (25–30 °C) in a dark, cool, dry place and withdrawn at monthly interval for the respective analyses. All analyses were carried out at one month interval during the period between November 2004 and December 2005 at UPASI Tea Research Institute, Valparai, Coimbatore District.

### 2.2. $\gamma$ irradiation treatment

$\gamma$  irradiation treatment was carried out at the spices irradiation unit of board of radiation and isotope technology (BRIT), Babha atomic research centre (BARC), Mumbai using  $^{60}\text{Co}$  gamma cell (Model: Gamma Chamber 5000, BARC, Mumbai). Packed samples were irradiated at 7.0 kGy and 10.0 kGy at room temperature (25–30 °C). The irradiation stand was rotated to ensure that the samples received a uniform radiation dose.

### 2.3. Moisture content

Moisture content of the black tea samples was monitored adopting (ISO, 1980). Samples were dried at  $103 \pm 2$  °C for a period of 16 h and moisture content was expressed in percentage.

### 2.4. Enumeration of microbes

Yeast and mould colony forming units (CFU) were determined by Indian standard procedure (IS, 1999) using spread plate methodology on yeast extract dextrose chloramphenicol agar (YDA) medium (pH 6.6) using serial dilu-

tion technique. Plate count of bacteria, was analyzed on tryptone yeast extract dextrose agar (TYA) medium (pH 7.0) using the standard procedure (IS, 1995). YDA plates were incubated at  $25 \pm 1$  °C for three days, whereas TYA plates were incubated at  $37 \pm 1$  °C for two days. The microbial content of the samples was measured by observing the average number of viable cells expressed as CFU/ml in the samples. The presented data is the average counts observed in three petri dishes for each sample.

### 2.5. Biochemical analyses of quality parameters

Major quality parameters of the black tea such as theaflavin (TF), thearubigin (TR), highly polymerized substances (HPS) and total liquor colour (TLC) were analysed according to the procedure of Thanaraj and Seshadri (1990).

### 2.6. Statistical analysis

Data obtained were analyzed statistically (ANOVA) wherever possible and percent loss against control was computed. Differences among the results obtained by different treatments were analyzed statistically and means were separated by least significant difference (LSD) at 5% probability level.

## 3. Results and discussion

### 3.1. Enumeration of microbes

Through out the experimental period, no colony forming unit of either bacteria or yeast and mould was detected in  $\gamma$  irradiated (at 7 kGy and 10 kGy) samples of both CTC and orthodox tea, whereas, in non-irradiated control samples, CFU was detected in both types of tea in all the months (Table 1). In general, a higher number of bacterial CFU were noticed than the fungal CFU. Between the two types of teas, CTC had a higher number of CFU ( $5 \times 10^3$ /g of bacteria and  $4 \times 10^1$ /g of fungus) than that in orthodox tea ( $4 \times 10^2$ /g of bacterial and  $2 \times 10^1$ /g of fungal CFU) at the end of experimental period. A gradual increase in the number of CFU was noticed in the control samples with the increase in storage period. Several studies indicated that irradiation was an effective way of preserving food products (Chen, Xu, Chen, Chen, & Dong, 1993; Yang, Perng, Liou, & Wu, 1993; Yu, Zhang, Cheng, & Zheng, 1993; Zhang, Liu, Li, Yang, & Tian, 1993; Zhou, Jin, Wei, Fu, & Xiong, 1996). Our result show that, like spices,  $\gamma$  irradiation could be successfully employed in black tea manufacturing process to reduce the natural contaminants of bacterial and fungal species, which are responsible for the degradation of quality parameters (Ramasamy & Raju, 1993). Both 7 kGy and 10 kGy resulted in no microbial activity during the entire period of storage of one year in both CTC and orthodox teas, which indicate that irradiation

Table 1  
Effect of  $\gamma$  irradiation on microbial load in black tea (CTC and orthodox) samples during one year storage

Treatment	Colony forming units (CFU/g) black tea sample							
	Quarter I		Quarter II		Quarter III		Quarter IV	
	Bacteria	Fungi	Bacteria	Fungi	Bacteria	Fungi	Bacteria	Fungi
CTC (untreated control) (g)	$4.1 \times 10^3$	$3.7 \times 10^1$	$4.5 \times 10^3$	$3.7 \times 10^1$	$4.8 \times 10^3$	$3.9 \times 10^1$	$5 \times 10^3$	$4 \times 10^1$
Orthodox (untreated control) (g)	$3.3 \times 10^2$	$1.9 \times 10^1$	$3.7 \times 10^2$	$2 \times 10^1$	$3.9 \times 10^2$	$2 \times 10^1$	$4 \times 10^2$	$2 \times 10^1$
CTC 7 kGy	ND	ND	ND	ND	ND	ND	ND	ND
CTC 10 kGy	ND	ND	ND	ND	ND	ND	ND	ND
Orthodox 7 kGy	ND	ND	ND	ND	ND	ND	ND	ND
Orthodox 10 kGy	ND	ND	ND	ND	ND	ND	ND	ND

ND: not detectable.

tion at 7 kGy or above was effective in securing the microbial safety of the black teas.

### 3.2. Moisture content

Data on moisture content during storage in CTC and orthodox teas are presented in Table 2. Untreated control samples accumulated a moisture percentage of about 8.65 in 12 months time, whereas the irradiated samples retained the moisture level in both CTC and orthodox tea at around 6.0%. No significant difference was found between 7 kGy and 10 kGy treatments on moisture content. Das, Gogoi, Gogoi, and Goswami (1994) also found an increase in the moisture content of tea from 3.45% to 6.18% when stored at ambient temperature for six months. It is assumed that the metabolic activities of the microbial population in the non-irradiated samples have accelerated the moisture content. Increase in the moisture content over a period of time resulted in the deterioration of quality constituents of tea during storage. Tea, being hygroscopic, is more susceptible to lipid hydrolysis, auto oxidative reactions and enzymatic browning during storage. Deterioration of black tea is characterized by a loss of aroma and astringency and some-

Table 2  
Changes in moisture content (%) during one year storage of  $\gamma$  irradiated and untreated control samples of black tea (CTC and orthodox)

Months	CTC			Orthodox		
	Control	7 kGy	10 kGy	Control	7 kGy	10 kGy
1	3.78	3.78	3.78	3.61	3.61	3.61
2	4.36	3.85	3.82	3.95	3.66	3.65
3	4.61	3.97	3.89	4.37	3.80	3.89
4	5.00	4.41	4.45	4.99	4.04	4.08
5	5.35	4.64	4.55	5.23	4.25	4.19
6	5.79	4.72	4.70	5.45	4.38	4.32
7	5.94	5.23	5.02	5.58	4.58	4.57
8	6.31	5.36	5.22	5.83	4.89	4.74
9	6.74	5.54	5.24	6.37	5.06	4.96
10	7.27	5.75	5.47	7.48	5.17	5.21
11	7.96	5.83	5.69	7.96	5.63	5.61
12	8.65	6.13	6.09	8.36	5.96	5.85
SE	1.25	0.31	0.21	1.01	0.25	0.17
CD	2.45	0.61	0.41	1.98	0.49	0.33

times by the acquisition of undesirable taints that cause loss of quality constituents. Reactions were found to be accelerated by moisture (Hazarika & Mahanta, 1983).

### 3.3. Biochemical analyses of quality parameters

Theaflavin (TF), a major quality constituent, which contributes to the brightness and briskness of the tea liquor, showed deterioration in all the samples during storage (Table 3). However, compared to untreated control, irradiated samples had very little change. There was a loss of 22.4% TF in the untreated control sample of CTC tea compared to orthodox at 37.3%, whereas, irradiation at 7 kGy and 10 kGy resulted in reducing loss significantly (Fig. 1). A significant deterioration of TF content was found in case of untreated control between 0 month and 12th month. The loss of TF during storage is a major factor responsible for the deterioration of black tea quality.

Thearubigin (TR) also showed a decline in both types of tea during storage. In case of untreated control CTC tea, 35% loss in TR was recorded after 12 months of storage, whereas in orthodox tea it was 24%.  $\gamma$  irradiation could reduce this deterioration to a great extent. CTC tea treated at 7 kGy and 10 kGy resulted in 17.5% and 13.2% loss of TR respectively, when compared to zero day samples, during 12 months storage. In case of orthodox tea, these losses were 6.7% and 4.6%, respectively (Fig. 2). TR content of

Table 3  
Changes in theaflavin (TF) content (%) during one-year storage of  $\gamma$  irradiated and untreated control samples of black tea (CTC and orthodox)

Treatment	TF content (%)			
	Quarter I	Quarter II	Quarter III	Quarter IV
<i>CTC</i>				
Control	$0.98 \pm 0.05a$	$1.07 \pm 0.03a$	$1.06 \pm 0.03a$	$0.84 \pm 0.03a$
7 kGy	$1.08 \pm 0.07b$	$1.32 \pm 0.04c$	$1.07 \pm 0.04a$	$0.89 \pm 0.04b$
10 kGy	$1.01 \pm 0.06a$	$1.22 \pm 0.04b$	$1.02 \pm 0.04a$	$1.00 \pm 0.03c$
<i>Orthodox</i>				
Control	$0.71 \pm 0.03a$	$0.86 \pm 0.04a$	$0.60 \pm 0.03a$	$0.47 \pm 0.02a$
7 kGy	$0.76 \pm 0.05b$	$1.05 \pm 0.03c$	$0.77 \pm 0.02b$	$0.69 \pm 0.04c$
10 kGy	$0.71 \pm 0.05a$	$1.00 \pm 0.03b$	$0.77 \pm 0.03b$	$0.64 \pm 0.03b$

Values are mean of three months  $\pm$  S.E. Means in each column followed by same letters are not significantly different at  $P = 0.05$  according to Fisher's LSD test.

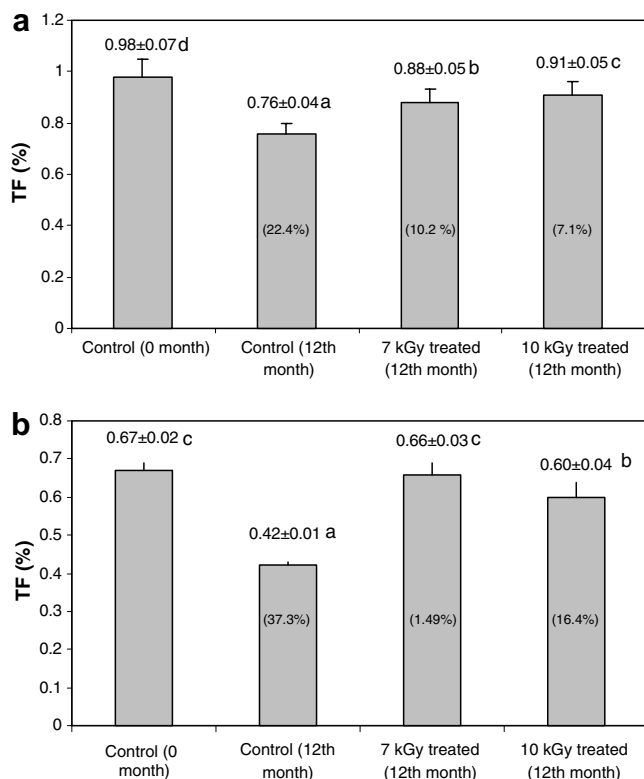


Fig. 1. Variations in theaflavin content (TF%) of black tea after 12 months of storage due to  $\gamma$  irradiation. (a) CTC tea and (b) orthodox tea. Values are mean  $\pm$  S.E. Means followed by same letters are not significantly different at  $P = 0.05$  according to Fisher's LSD test. Values in parenthesis are percent loss.

untreated control samples showed significant deterioration between 0 and 12th month while in all the other samples it was non-significant.

Highly polymerized substances (HPS) and total liquor colour (TLC), which contribute to the colour and body of the liquor and cuppage of black tea, also deteriorated during storage. HPS showed 44.0% and 34.4% deterioration in the untreated control samples of CTC and orthodox, whereas the reduction was 21.7% and 30.6% in the case of TLC. These changes were significantly low in the case of irradiated samples (Fig. 2).

It was observed that the control samples were stable only for a period of six months followed by deterioration of all of the quality parameters. The study reveals stability of major quality parameters like TF, TR, TLC and HPS in irradiated tea during one year storage. Principal changes undergone during storage are due to the formation of compounds related to TF and TR (Roberts & Fernando, 1981).

#### 4. Conclusion

The results clearly indicated that quality deterioration occurred even in well packed untreated control samples. Various doses of  $\gamma$  irradiation resulted in complete absence of microbial growth and contained the quality deterioration during storage in both CTC as well as orthodox tea.

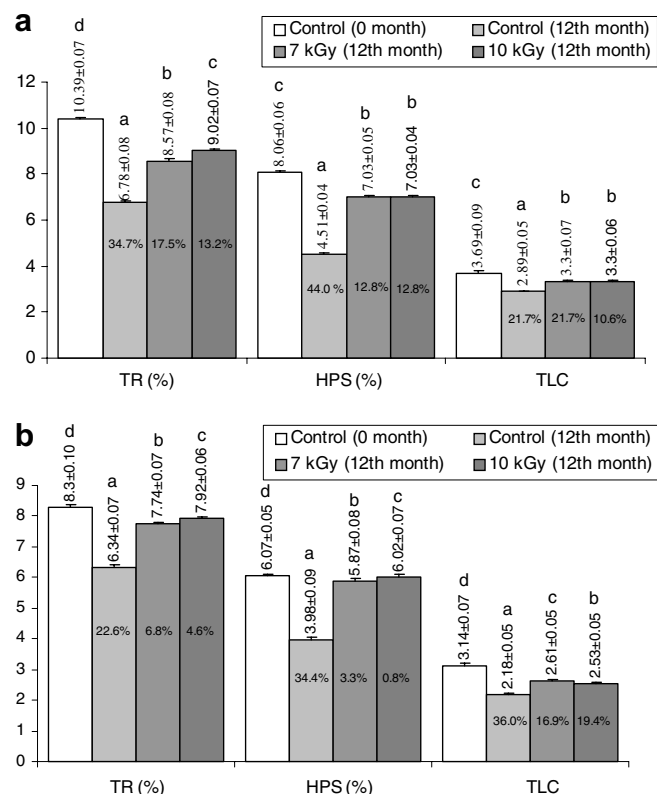


Fig. 2. Variations in thearubigins (TR%), highly polymerized substances (HPS%) and total liquor colour (TLC) of black tea after 12 months storage due to  $\gamma$  irradiation. (a) CTC tea and (b) orthodox tea. Values are mean  $\pm$  S.E. Means followed by same letters are not significantly different at  $P = 0.05$  according to Fisher's LSD test. Values in parenthesis are percent loss.

In conclusion, irradiation dose of 7 kGy can be effective to control microbial growth in black tea and in extending their shelf life without any significant deterioration of quality constituents. This technology will enable food processors to deliver larger amounts of high quality tea with extended shelf life.

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