

EFFECT OF GAMMA RADIATION ON MICROBIOLOGICAL LOAD OF BEEFBURGER DURING COLD STORAGE ,  
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SUMMARY: Freshly prepared beefburger samples were irradiated on the same day of manufacture with 0.0 and 5.0 KGy. The unirradiated and irradiated samples were stored at  $2^{\circ}\text{C} \pm 1$  . The samples were analysed microbiologically during storage for total viable counts of bacteria, sporeformers, molds, yeasts , *Strept. faecalis*, *Staph. aureus* and enteropathogenic *E. coli*. The detection of *B. cereus*, *Cl. perfringens* and *Salmonella spp.* was also carried out.

INTRODUCTION: Beefburger consists of chopped fresh and/or frozen beef, with or without the addition of beef fat as such and/or seasoning. It does not contain more than a total of 30% fat. Some procedures added vegetables (minced onion , ground garlic, celery flakes and parsley ) in amount not more than 5% (Henrickson, 1978) .

Microbiological studies were carried out on raw hamburgers (beef or beef/pork + minor ingredients) samples from food service establishments by Tamminga *et al.* (1982). Total aerobic counts (/g) were  $10^6 - 10^8$  . Enterobacteriaceae counts in the range  $10^3 - 10^6$ /g. Salmonellae were present in 32.4% of raw samples. Studies were also conducted by Rossi Junior *et al.* (1985) on hamburger. They found that in some samples , the overall, hygienic quality was relatively poor, and could present a health hazard .

Studies were conducted by Niemand *et al.* (1980) on bacteriological characteristics of ground beef-irradiated at 3 KGy , and of vacuum packaged beef cuts irradiated at 2 KGy, and on changes in bacteriological characteristics during storage at  $4^{\circ}\text{C}$ . Counts of all spp. were considerably reduced by irradiation; counts of pseudomonas and enterobacteria remained low, whereas counts of total aerobes, total anaerobes, lactic acid bacteria and micrococci subsequently increased. Staphylococci and Salmonella were eliminated by these irradiation doses. Taste and odour of irradiated samples were inferior to those of control samples ; irradiation at low temperature may minimize this problem . Beefburger samples were found to be contaminated with *Strept. faecalis*, *Staph. aureus*, enteropathogenic *E. coli* , *B. cereus* *Cl. perfringens*. (Youssef, 1987). Therefore, it was thought beneficial to study the effect of gamma radiation, with suitable dose, on eliminating such harmful organisms from chilled beefburger as well as its effect on extending their shelf-life in cold condition .

MATERIAL AND METHODS: Every two portions of fresh prepared beefburger samples were kept in polyethylene bags. The bags of beefburgers were irradiated on the same day of manufacture with 5.0 KGy. The unirradiated and irradiated samples were stored at  $2^{\circ}\text{C} \pm 1$  . The irradiation process was done at the National

Center for Radiation Research and Technology, Nasr City Cairo, using the Egypt's Industrial Mega-Gamma Irradiator. The source was cobalt 60. The dose rate at the time of experiments was 1 KGy/2.8 minute. 10 grams of the samples were mixed well with 90 ml of saline solution (8.5 g NaCl + 1 g peptone/L). Serial dilution method was used for the microbiological tests .

METHOD: Total bacterial counts were determined as recommended by the American Public Health Association (APHA),(1960). Aerobic sporeformers count was determined according to the method described by Chalmers (1955). Yeasts and molds count was determined according to the Oxoid Manual (1982).

*Streptococcus faecalis* was counted on Kanamycin aesculin azide agar medium as recommended by Mossel et al. (1973) .

*Staphylococcus aureus* was numerated on laboratory prepared Baird-Parker medium as recorded by IAEA (1970). DN -ase test was used as a confirmation test (Oxoid Manual, 1982) .

Enteropathogenic *E. coli* (EEC) was counted using the MPN method as reported by IAEA, (1970) .

*Bacillus cereus* was detected as described by Mossel et al. (1967) . Confirmation test of suspected colonies were made biochemically by testing acid from different sugars. Colonies which ferment glucose but not manitol, xylose or arabinose were considered to be *B. cereus* .

*Clostridium perfringens* was detected as mentioned by Stephen et al. (1975). Positive colonies are characterized by the ability to liquify gelatin after 24 -44 hrs. (Hauschild and Hilsheimer, 1974).

*Salmonella spp.* were detected using the most probable number technique according to ISO (1978) .

#### RESULTS AND DISCUSSION:

##### Spoilage flora of beefburger:

##### Total bacterial count :

Data in Table (1) showed that the total bacterial count(TBC) were sharply reduced by irradiation treatments . The initial TBC in unirradiated sample was  $1.1 \times 10^6$ , was reduced to  $2.5 \times 10^2$  cells/g as a results of irradiation with 5.0 KGy. These results are in agreement with those obtained for sausage(Hassan, 1976 and Emam, 1987) and Shams El-Din (1978) in case of camel meat .

During cold storage at  $2^{\circ}\text{C} \pm 1$  progressive increase in the TBC of unirradiated and irradiated beefburger with almost the same rate specially after the first period of storage However, the total bacterial counts remained lower in irradiated samples allover the storage period (8 weeks) . At the end of 8<sup>th</sup> week of storage the TBC reached  $9.5 \times 10^8$  cells/g and  $2.0 \times 10^6$  cells /g in the control and irradiated samples. At this time the control and irradiated samples were rejected. The control samples were rejected due to high levels of bacteria while the irradiated samples were rejected due to the appearance of fungi spots on the surface of beefburger. Daelman and Hoof (1975) found that a temperature of  $2^{\circ}\text{C}$  gave a sausage of excellent microbial quality even after storage for 21 days .

### Sporeforming bacteria :

Data in Table (1) showed that sporeforming bacteria were more resistant to gamma radiation than total bacteria since about 4 log cycles were reduced in total counts by 5 KGy, while only one log cycle was occurred by the same dose in sporeformers. Similar results were reported by Shams El-Din ( 1978 ) ; and Vankooji (1981) . The main reason that spores are radiation resistant is probably due to their low water content , which reduces the efficiency of ionizing event (Tallentire, 1970) .

During cold storage, relatively slow rates of increasing in sporeformers counts either unirradiated or irradiated samples were observed . These results may be due to that refrigerated temperature was unsuitable for the growth of these microorganisms. The mean counts increased from  $9.0 \times 10^2$  to  $1.7 \times 10^4$  cells/g. and from  $4.0 \times 10^2$  to  $1.0 \times 10^3$  cells/g after 8 weeks in unirradiated and irradiated beefburger samples respectively.

### Yeast and mold counts :

Data in Table (1) showed that gamma irradiation with dose of 5.0 KGy inhibited either yeasts or molds in beefburger samples. However, the yeast were started to multiply after one week of storage whereas few colonies of molds were appeared after 2 weeks in the irradiated samples . During storage the yeast counts of unirradiated and irradiated samples increased by almost the same rate after the first period of storage reaching  $1.2 \times 10^5$ ,  $2.7 \times 10^5$  CFU/g at the end of the 8<sup>th</sup> week for the irradiated and unirradiated samples respectively. On the other hand the counts of mold increased sharply in irradiated samples after the 2<sup>nd</sup> week of storage reaching  $7.4 \times 10^5$  CFU/g at the end of the 8<sup>th</sup> week, whereas, their counts in the control were only  $1.9 \times 10^4$  CFU/g at the same week. As previously mentioned the irradiated samples were rejected due to the appearance of fungus spots on the surface of beefburger. However, it is notable that the irradiated samples did not reach the borderline of acceptability as regards to the total bacterial counts till the 8<sup>th</sup> week of storage (Table 1) .

These results are in accordance with those of Emerson *et al.* (1964) who reported that the spoilage of unirradiated fish, fruits and vegetables samples was due primarily to the bacterial growth, the yeast and molds were the principal cause of spoilage in irradiated samples. The same findings were also obtained by Corelett *et al.* (1965) and Youssef (1981) for irradiated fish and Bolti fish fillet respectively .

### Pathogenic bacteria :

Data in Table (2) showed that the initial counts of *Strept. faecalis*, enteropathogenic *E. coli* and *Staph. aureus* were  $1.2 \times 10^5$ ,  $1.6 \times 10^4$  and  $1.0 \times 10^4$  cells/g respectively in unirradiated beefburger samples. Application of gamma radiation at 5.0 KGy dose level completely suppressed these pathogens in beefburger samples. On the other hand , the counts of these organisms in the uni-radiated beefburger reduce as storage period advanced at  $2^\circ\text{C} \pm 1$  , reaching  $6.5 \times 10^2$  cells/g in case of *Strept. faecalis* . Meanwhile, no growth of either enteropathogenic *E. coli* or *Staph. aureus* was observed after 6 weeks storage. The decrease in number of pathogens at low temperature may be due to the unsuitable temperature for growth



of these pathogens as reported by El-Mongi (1983) .

With regards to *B. cereus* the results presented in Table(2) indicated that this organism was present in either unirradiated or irradiated beefburger samples. These results revealed that irradiation with 5.0 KGy dose level was not sufficient to eliminate *B. cereus* from beefburger product. Ingram and Roberts (1980) reported that some spores of *B. cereus* are among the most resistant *B. spp.* These finding confirmed the previous results of sporeforming bacteria (Table 1) . Meanwhile , neither *Cl. perfringens* nor *Salmonella spp.* were detected in beefburger samples .

Table 1. Effect of gamma radiation on spoilage flora counts of beefburger during storage at  $2^{\circ}\text{C} \pm 1$ , (cells/g).

Storage period (weeks)	Total bacterial counts		Sporeformers count	
	0.0 KGy	5.0 KGy	0.0 KGy	5.0 KGy
0	$1.1 \times 10^6$	$2.5 \times 10^2$	$9.0 \times 10^2$	$4.0 \times 10$
1	$6.5 \times 10^6$	$2.7 \times 10^2$	$1.4 \times 10^3$	$8.0 \times 10$
2	$8.5 \times 10^6$	$3.3 \times 10^3$	$2.7 \times 10^3$	$1.1 \times 10^2$
4	$4.3 \times 10^7$	$1.1 \times 10^4$	$4.1 \times 10^3$	$2.3 \times 10^2$
5	$1.2 \times 10^8$	$1.1 \times 10^5$	$6.0 \times 10^3$	$5.5 \times 10^2$
8	$9.5 \times 10^8$	$2.0 \times 10^6$	$1.7 \times 10^4$	$1.0 \times 10^3$

  

Storage period (weeks)	Yeast		Mold	
	0.0 KGy	5.0 KGy	0.0 KGy	5.0 KGy
0	$1.0 \times 10^3$	NG	$7.0 \times 10$	NG
1	$2.3 \times 10^3$	$3.5 \times 10^2$	$8.5 \times 10$	NG
2	$2.9 \times 10^3$	$2.3 \times 10^3$	$5.5 \times 10^2$	$1.0 \times 10$
4	$4.6 \times 10^3$	$5.0 \times 10^3$	$8.5 \times 10^2$	$2.6 \times 10^2$
6	$1.5 \times 10^4$	$2.4 \times 10^4$	$3.0 \times 10^3$	$1.5 \times 10^4$
8	$1.2 \times 10^5$	$2.7 \times 10^5$	$1.9 \times 10^4$	$7.4 \times 10^5$

NG = No growth was observed .

Table 2. Effect of gamma radiation on some pathogenic bacteria and incidence of *B. cereus* in beefburger stored at  $2^{\circ}\text{C} \pm 1$  .

Storage period (weeks)	<i>Strept. faecalis</i>		<i>Staph. aureus</i>		<i>E. coli</i>	
	0.0 KGy	5.0 KGy	0.0 KGy	5.0 KGy	0.0 KGy	5.0 KGy
0	$1.2 \times 10^5$	NG	$1.0 \times 10^4$	NG	$1.6 \times 10^4$	NG
1	$2.1 \times 10^4$	NG	$5.5 \times 10^3$	NG	$7.4 \times 10^2$	NG
2	$7.1 \times 10^3$	NG	$3.0 \times 10^2$	NG	$3.1 \times 10^2$	NG
4	$4.7 \times 10^3$	NG	$1.0 \times 10^2$	NG	$3.6 \times 10$	NG
6	$1.5 \times 10^3$	NG	NG	NG	NG	NG
8	$6.5 \times 10^2$	NG	NG	NG	NG	NG

NG = No growth was observed .

Table (2) Cont.

Incidence of *B. cereus*

Storage period (weeks)	0.0 KGy	5.0 KGy
0	+	+
1	+	+
2	+	+
4	+	+
6	+	+
8	+	+

+ Present .

CONCLUSION: Total bacterial counts and sporeformers count of beefburger samples were greatly reduced by treatment with gamma radiation with 5 KGy. The same dose was sufficient to eliminate yeasts, molds, *Strep. faecalis*, *Staph. aureus* and enteropathogenic *E. coli* but did not affect the presence of *B. cereus*. *Cl. perfringens* and *Salmonella spp.* were not detected in all samples. After 8 weeks storage, the unirradiated samples were spoiled bacteriologically while the irradiated ones were rejected due to the appearance of fungal spots on the surface of beefburgers inspite of its lower bacterial counts (  $2 \times 10^6$  cells/g ) .

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