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Research Article/Araştırma Makalesi

Impact of low-dose gamma irradiation treatment on microbial and chemical properties of raw milk white cheese

Düşük doz gama ışınlama işleminin çiğ sütten üretilen beyaz peynirin mikrobiyal ve kimyasal özellikleri üzerine etkisi

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ABSTRACT

Using raw milk in cheese-making has microbiological risks, however a considerable number of artisan cheese makers around the world prefer raw milk, as heat treatment damages natural microbiota and alters the characteristic taste and flavor of the cheese. The potential of the gamma irradiation process to make the raw milk cheese microbiologically safe was investigated in this study. A total of 5 groups of cheese were produced, including control groups (control pasteurized milk cheese and control non-irradiated raw milk cheese). After the white cheese samples were kept in brine for 1 night, they were vacuum-packaged and each group was subjected to gamma irradiation at doses of 1, 2, and 3 kGy, respectively. Chemical composition, pH, acidity, Urea-PAGE images, total aerobic mesophilic bacteria (TAMB), coliform group bacteria, *E. coli*, yeasts and molds, *Lactobacillus* and *Lactococcus* counts of cheese samples were determined during 60 days of storage at 4°C. All analyzes were performed on days 1, 30, and 60 of production. We found that irradiation reduced the microbiological load of the cheeses and slowed down the degradation of α_{s1}-CN and β-CN especially at 3 kGy. Irradiation at 3 kGy successfully inhibited the coliforms and *E. coli*, however yeast and mold counts were still high due to initial high levels. Our study showed that irradiation of the raw milk cheese is a promising method to reduce the microbial counts to acceptable limits which could reduce the required ripening periods for the microbial safety of raw milk cheeses.

Key Words: Food irradiation, irradiation dose, white cheese, low-fat

ÖZ

Peynir yapımında çiğ sütün kullanılması mikrobiyolojik riskler taşımasına rağmen, ısıl işlemin doğal mikrobiyotayı bozması ve ürünün tipik tat ve aromasını değiştirmesi nedeniyle dünya çapında önemli sayıda artizan peynir üreticisi çiğ süt kullanımını tercih etmektedir. Bu çalışmada çiğ süt peynirinin ışınlama işlemi ile mikrobiyolojik açıdan güvenli hale getirilme potansiyeli araştırıldı. Kontrol grupları (kontrol pastörize süt peyniri ve kontrol ışınlanmamış çiğ süt peyniri) dahil toplam 5 grup peynir üretildi. Beyaz peynir örnekleri salamurada 1 gece bekletildikten sonra vakumla paketlenerek 3 gruba ayrıldı ve her gruba sırasıyla 1, 2, ve 3 kGy dozlarında gama ışınlaması uygulandı. Peynir örneklerinin kimyasal kompozisyonu, pH, asitlik, Urea-PAGE görüntüleri, total aerobik mezofilik bakteri (TAMB), koliform grup bakteri, *E. coli*, maya - küf, Laktobasil ve Laktokok sayıları 4°C'de 60 günlük depolama süresince analiz edildi. Analizler üretimin 1, 30, ve 60. günlerinde yapıldı. Işınlamanın peynirlerin mikrobiyolojik yükünü azalttığını ve özellikle 3 kGy'de α_{s1}-CN ve β-CN'nin bozunmasını yavaşlattığını bulduk. 3 kGy'deki ışınlama, koliformları ve *E. coli*'yi başarılı bir şekilde inhibe etti, ancak maya ve küf sayıları, başlangıçtaki yüksek seviyeler nedeniyle hala yüksekti. Çalışmamız, çiğ süt peynirinin ışınlanmasının, mikrobiyal yükü kabul edilebilir sınırlara düşürmek ve çiğ süt peynirleri için gerekli olgunlaşma sürelerini kısaltabilecek umut verici bir yöntem olduğunu gösterdi.

Anahtar Kelimeler: Gıda ışınlaması, ışınlama dozu, beyaz peynir, az yağlı

Introduction

Raw milk had been used for cheese making over centuries. Artisan and farmstead raw milk cheese makers still exist in many countries around the world. Today about 10% of the total cheese production in most European countries is made from raw milk (Beuvier et al., 2004). Some traditional raw milk cheeses in Europe have a protected designation of origin (PDO). Food and Drug Administration (FDA) legally permitted using raw milk for about 30 of the 72 defined cheeses (FDA, 2012). Artisan cheese makers prefer to use raw milk for its native microbiota and enzymes that help develop their signature aroma and texture (Beuvier et al., 2004). Especially in Turkey, lack of the starter cultures for most traditional cheese types drives local dairy farms and family-owned dairies to use raw milk for cheese making. They do not prefer pasteurization as it impairs natural microbiota and alters the original taste and flavor of the cheese. However, raw milk cheeses carry microbiological safety risks, as they might be contaminated with pathogens and have a high microbial load (Beuvier et al., 2004). The Centers for Disease Control (CDC, 2012) advises that risk groups, such as children below 5 years old, pregnants, and people with chronic diseases should not consume raw milk products. There are legislations made for raw milk cheeses in many countries. It is prohibited to sell raw milk cheese before 4 months of ripening in Turkey (Turkish Food Codex Cheese Bulletin, 2015). FDA stated that, raw milk cheese must be aged before consumption for at least 60 days (FDA, 2012). Irradiation of the raw milk cheese could reduce microbial counts to acceptable levels eliminating the costly ripening period for microbial safety. Gamma irradiation comprises the exposure of the food to ionizing radiations obtained by gamma rays from radioactive isotopes of Co-60 or Cs-137. This exposure excites the atoms in the treated food. While the energy is not sufficient to create radioactivity, it is detrimental for microorganisms in the food at certain doses. Irradiation inactivates microorganisms by damaging the cell, mainly by damaging the chromosomal DNA and the damage either suppresses multiplication or cause death by terminating most cell functions (Odueke et al., 2016). Food irradiation has been used for extending the shelf life of the certain food products by many countries and it has the potential to preserve and extent the cheese shelf life without impairing nutritional and sensory characteristics. If applied properly, food irradiation is safe and doesn't cause unwanted organoleptic changes. However, it has been demonstrated by several researchers that higher doses impair organoleptic and nutritional properties of the food. Irradiation of the dairy products is not as widely accepted as other products such as meat and poultry, fruits, vegetables, and spices. The main reason for that is the sensitivity of the lipids to irradiation even at low doses. Polyunsaturated fatty acids are prone to oxidation by free radicals produced by ionizing energy. Irradiation can oxidize the casein and form methyl radicals. Unwanted off-flavors can occur through oxidation, polymerization, de carboxylation, and dehydration reactions caused by irradiation (Arvanitoyannis and Tserkezou, 2010). However, use of irradiation for reducing the microbial load of the cheese is still promising with a careful adjustment of the irradiation dose. In previous studies, irradiation have been applied successfully at doses under 5 kGy to improve the safety and shelf-life of various cheese types including Queso Blanco (Ham et al., 2009), Mozzarella (Huo et al., 2013), Karish (Aly et al., 2012), Ras (Shalaby et al., 2016), Cheddar (Seisa et al., 2004) and Feta (Konteles et al., 2009). There is no previous study on irradiation of raw-milk Turkish White cheese to the authors knowledge. Effectiveness of irradiation on different microorganisms found to be different between studies on different types of cheeses (Arvanitoyannis and Tserkezou, 2010). Irradiation of Artisan Hard cheese at 5 kGy effectively reduced *E. coli*, coliforms, ACC and Listeria spp. monocytogenes counts below the recommended limit (Nyamakwere et al., 2022). Irradiation of post-process contaminated feta cheese at 2.5 and 4.7 kGy was effective to inhibit Listeria

monocytogenes, while pre-process contaminated cheeses still had high load (Konteles et al., 2009). Irradiation resistance among microorganisms varies depending on several factors such as population size, properties of the substrate and microorganisms (Blank et al., 1992). Proteins and lipids in cheese can provide irradiation protection for microorganisms. Therefore, effectiveness of irradiation on reducing the microbial load below acceptable limits is dependent on not only the irradiation dose, but also the type of the cheese and its initial microbial load. In order to determine the effective irradiation doses and conditions, each cheese type should be investigated individually. In this study, we aimed to examine the impact of irradiation at doses up to 3 kGy on chemical and microbiological properties of raw milk cheese samples during 60 days of storage.

Material and Methods

Cheese manufacture

Cheese samples were produced at the pilot dairy plant of the Harran University Department of Food Engineering. A total of 200 L milk was used for cheese production provided by Harran University animal farm. After preheating to 35-40 °C, fat was removed using separator (Alfa Laval, Sweden). Milk used for cheese making had 9.45% total solids, 1.7% fat and its pH was 6.72. One part of the milk was separated for control cheese and low pasteurized at 63 °C for 30 minutes, while rest of the raw milk was used for cheese making without any heat treatment. No starter culture was used in cheese manufacture. When the milk temperature reached to 30±1 °C, 0.02% CaCl2 and rennet (CHY-MAXTM M, 600 international milk-clotting units (IMCU) per mL; Chr-Hansen A/S, Hoersholm, Denmark) that is sufficient to curdle the milk in 1 h, were added. Curd was cut into 1 cm3 cubes and allowed to sit for 15 min before collecting into cheese cloth. After all cheese curd was transferred into cheese cloth, whey was drained for 30 min on its own weight and then pressed under added weight for about 3.5 h. Cheese was then cut into 7x7x7 cm^3 blocks, brine salted at 16% salt (NaCl) concentration for 12 h. After draining the brine, cheese blocks were vacuum-sealed in 20x30 cm 80 µ vacuum-sealer bags. Cheese making was replicated twice, and all analysis were performed at least in duplicate.

Irradiation treatment

Vacuum packed cheese samples were irradiated on the first day of the production in a cobalt-60 gamma irradiator cell (Izotop, Ob-Servo Sanguis Co-60 Research Irradiator) at the Turkey Atomic Energy Research Institute, Ankara. Irradiation was applied only to raw milk cheese samples and the applied doses were 1, 2, and 3 kGy with a dose rate of 1.5 kGy/h. No irradiation was applied to control pasteurized milk cheese (Cp) and control raw milk cheese (Cr) samples. Cheese samples stored at 4°C for 60 days and all analysis was done at day 1, day 30, and day 60.

Chemical analysis

Prior to chemical analysis, each cheese sample (whole package) was ground for homogeneity after sterile sampling for the microbiological analysis. Moisture content was determined by the standard drying method at 105±2 °C (IDF, 1982), fat content was determined according to the method of Van Gulik (ISO 3433 | IDF 222, 2008), nitrogen content by the Kjeldahl method (AOAC, 1998) and NaCl content by the Mohr titration method (IDF, 1988). The pH was measured by a pH meter (Testo 230, Germany).

Moisture in non-fat solids (MNFS) were calculated based on these parameters and expressed as mass fraction in %. All measurements were carried out in duplicates and results were expressed as mean value ± standard error (SE).

Urea-page gel electrophoresis was conducted to assess proteolysis (Andrews and Alichanidis, 1983). A separating gel with 12.5% total acrylamide, 4% cross-linking agent at pH 8.9 was used. Final gels were monitored with Bio-imaging systems (mini BIS PRO, Israel) and photographed.

Microbiological analysis

Enumeration of total aerobic mesophilic

bacteria (TAMB), coliforms, *E. coli*, yeasts and moulds, *Lactobacillus* and *Lactococcus* were done during storage on day 1, day 30 and day 60, as colony-forming units (cfu)/mL. Cheese samples (10 g) were weighed inside a sterile cabin into a sterile stomacher bag containing 90 mL of sterile peptone solution (0.1 % m/V) and homogenized for 2 min in the stomacher. Prepared homogenate was then immediately used for preparing the successive dilutions. TMAB counts were determined on Plate Count Agar (PCA, Merck) at 30 °C for 24-48 h (Messer et al., 1985); yeast and molds counts were determined on Potato Dextrose Agar (PDA, Merck) at 25 °C for 5 days (Frank et al., 1985). Coliform group bacteria were determined on Violet Red Bile Agar (VRBA, Merck) at 37 °C for 24 h; *Lactobacillus* count was determined on MRS agar under anaerobic conditions at 30±1 °C for 72 h (Harrigan., 1998). *Lactococcus* counts were determined on M17 agar at 30±1 °C for 48 h (Cabezas et al., 2007). *E. coli* count was determined using two layers of agar, first incubated for 2 h with Triptic Soy Agar (TSA) and after adding a layer of VRBA, incubated 24 h at 44±1 °C (Harrigan., 1998).

Statistical analysis

SPSS version 18 (SPSS Inc., Chicago, IL, USA) was used for analyzing the results. All the experiments were carried out in duplicate. One way ANOVA was used to find the statistical differences between the chemical and microbial properties of the samples. Differences between means were determined by Duncan's multiple range test. Mean values and the standard deviations (SD) were reported.

Results and Discussion

Chemical analysis results

Chemical composition, pH and acidity of the cheese samples are presented in Table 1. Pasteurized control cheese (Cp) had higher dry matter than other cheese samples. Irradiation had no impact on moisture, fat, MNFS and salt levels. Most other studies also showed that macro components such as fat were not influenced by irradiation doses below 10 kGy (Nyamakwere et al., 2022, Lalaguna, 2003, Olson, 1998). There was a decrease in the protein content of 3 kGy cheese samples. Some previous studies have also found a reduction in protein content due to gamma irradiation (Nyamakwere et al., 2022, Shalaby et al., 2016, Omer and Elshirbiny, 2005) and they attributed this to the destruction of natural microbiota and milk enzymes by irradiation resulting in a decrease in total nitrogen content. It was stated that gamma-irradiation could destroy biogenic amine molecules (Kim et al., 2004) that can reduce the total nitrogen content of the cheese. Although biogenic amines appear at low concentrations in cheese, raw milk cheese might have higher amounts of biogenic amines due to microbial decarboxylase activity on free amino acids (Rabie et al., 2011). Cp had higher pH and lower acidity than other cheese samples which could be due to the limited microbial growth and acid development in pasteurized milk during the cheese making steps. Irradiation did not influence the pH of the cheese samples significantly ($p>0.05$) in accordance with the previous studies (Nyamakwere et al., 2022, Lalaguna, 2003). However, acidity of the 3 kGy cheese on day 1 was lower than the Cr and 1 kGy. During the storage, pH of the cheese samples was stable except for the Cr. There was an increase in the acidity of Cp and 3 kGy cheese samples during the 60 days of storage. We did not see any significant difference at composition throughout the storage ($p>0.05$).

Table 1. Composition and pH of White cheeses (Cp: Control pasteurized cheese, Cr: Control raw milk cheese) at day 1, day 30 and day 60

Parameters	Day	Irradiation dose (kGy)				
		0 (Cp)	0 (Cr)	1	2	3
w(Moisture)/%	1	57.97±0.27 ^{aA}	59.54±0.71 ^{bA}	60.71±0.35 ^{bA}	61.91±0.00 ^{bA}	60.33±0.00 ^{bA}
	30	56.44±0.00 ^{aA}	58.95±0.00 ^{bA}	61.17±0.00 ^{bA}	60.74±0.00 ^{bA}	61.34±0.00 ^{bA}
	60	56.69±0.09 ^{aA}	59.09±0.35 ^{bA}	58.83±0.35 ^{bA}	60.01±0.35 ^{bA}	59.43±0.00 ^{bA}
w(Fat)/%	1	7.8±0.2 ^{aA}	7.5±0.7	8.7±0.3 ^{aA}	7.0±0.0 ^{aA}	8.0±0.0 ^{aA}
	30	8.0±0.0 ^{aA}	8.0±0.0 ^{aA}	8.0±0.0 ^{aA}	8.5±0.0 ^{aA}	7.5±0.0 ^{aA}
	60	8.5±0.1 ^{aA}	8.2±0.3 ^{aA}	8.2±0.3 ^{aA}	8.7±0.3 ^{aA}	9.0±0.0 ^{aA}
w(Protein)/%	1	27.72±1.35^{a}	27.35±2.30^{a}	27.91±3.47^{a}	25.12±1.62^{a}	23.45±0.78^{b}
	30	27.37±0.14^{a}	26.46±1.62^{a}	26.46±1.21^{a}	24.56±0.23^{a}	22.33±4.39^{b}
	60	27.36±3.75^{a}	26.24±2.76^{a}	26.94±1.28^{a}	25.58±0.97^{a}	22.33±0.56^{b}
w(Salt)/%	1	6.20±0.25 ^{aA}	5.87±0.02 ^{aA}	5.40±1.66 ^{aA}	5.70±1.17 ^{aA}	5.45±0.24 ^{aA}
	30	6.01±0.54 ^{aA}	5.75±0.91 ^{aA}	5.02±0.01 ^{aA}	5.40±0.07 ^{aA}	5.21±0.01 ^{aA}
	60	5.34±0.00 ^{aA}	5.20±0.23 ^{aA}	5.03±0.46 ^{aA}	6.03±1.44 ^{aA}	6.09±0.23 ^{aA}
w(MNFS)/%	1	62.88±1.02 ^{aA}	64.36±2.54 ^{aA}	66.54±3.22 ^{aA}	66.57±1.69 ^{aA}	66.57±0.52 ^{aA}
	30	61.35±0.58 ^{aA}	64.08±1.54 ^{aA}	66.49±1.35 ^{aA}	66.38±0.07 ^{aA}	66.31±4.70 ^{aA}
	60	62.00±4.15 ^{aA}	64.41±2.90 ^{aA}	64.11±1.52 ^{aA}	65.77±0.79 ^{aA}	65.31±0.33 ^{aA}
pH	1	6.38±0.04^{aA}	5.20±0.01^{bA}	5.19±0.07^{bA}	5.25±0.01^{bA}	5.23±0.08^{bA}
	30	6.43±0.00^{aA}	5.36±0.00^{bB}	5.28±0.01^{bA}	5.27±0.00^{bA}	5.30±0.06^{bA}
	60	6.53±0.01^{aA}	5.37±0.01^{bB}	5.33±0.01^{bA}	5.35±0.02^{bA}	5.35±0.01^{bA}
w(Acidity)/% lactic acid	1	0.05±0.00^{aA}	0.24±0.01^{cA}	0.23±0.01^{cA}	0.21±0.01^{bcA}	0.18±0.00^{bA}
	30	0.07±0.01^{aB}	0.23±0.01^{bA}	0.23±0.00^{bA}	0.22±0.00^{bA}	0.22±0.00^{bB}
	60	0.08±0.00^{aB}	0.23±0.00^{bA}	0.24±0.01^{bA}	0.23±0.01^{bA}	0.26±0.00^{bC}

Values are means ± standard deviations. Means within the same row marked with different lowercase letters are significantly different and means within the same column marked with different uppercase letters are significantly different ($p<0.05$).
MNFS: moisture in non-fat substance

Urea PAGE results

Urea-PAGE results are given at Fig 1. We observed differences between α_{s1} and β-CN bands and their degradation products of irradiated, non-irradiated and control pasteurized cheese samples. The cheese sample Cp had the highest amount of α_{s1}-CN band through the storage which could be due to the fact that pasteurization inactivates native milk proteases and microbiota to some extent. Proteolysis of the α_{s1}-CN occurs first by the coagulants with the cleavage of Phe23-Phe24 bond, forming α_{s1}-CN (f1-23) and α_{s1}-CN I (f24-199). Then residual coagulant, native milk enzymes and other microbial enzymes hydrolyze the α_{s1}-CN I (f24-199) into smaller fragments (Dave et al., 2003). The α_{s1}-CN band was almost invisible in Cr and became more distinct as the irradiation dose increased. This indicates that irradiation could be effective in inactivating some of the proteolytic enzymes and microbiota that would act on α_{s1}-CN. While the intensity of the α_{s1}-CN band of 2 and 3 kGy cheese samples didn't change much during the storage, a decrease observed with the others. The α_{s1}-CN I (f24-199) band of irradiated samples were similar on day 1. However, Cp had a slighter α_{s1}-CN I (f24-199) band, which could be due to other factors that limit rennet action on α_{s1}-CN, such as high pH. The intensity of the α_{s1}-CN I (f24-199) band of Cp and irradiated cheese samples increased during the storage. There was a decrease in the amount of α_{s1}-CN I (f24-199) band in Cr on day 30, while it increased in irradiated cheese samples with increasing irradiation doses. The non-irradiated raw milk cheese appeared to have high proteolytic activity due to native milk enzymes and microbial enzymes that consumed both α_{s1}-CN and α_{s1}-CN I (f24-199) rapidly. Irradiation could have inactivated some of the proteolytic enzymes and microbiota that could further degrade α_{s1}-CN I (f24-199). The increase in the α_{s1}-CN I (f24-199) band intensity also indicates that irradiation didn't inhibit residual rennet

activity. The β-CN band of all cheese samples appears to be similar on day 1. However, its degradation products differed between cheeses. Plasmin and neutral proteases degrade β-CN into ɣ-caseins (Fox, 1989). The sample Cp appear to have higher number of ɣ-CN fractions. When we look at the 30th and 60th day results, the intensity of the β-CN was higher at higher irradiation doses. A slight β-CN-I band was visible at 3 kGy sample after 30 days of storage. β-CN-I (f189-192) is correlated with bitterness. Thus, it is possible that irradiation may have inactivated some microorganisms that break down bitter peptides, leading to bitter β-CN-I (f189-192) fraction in 3 kGy cheeses (Öztürk et al., 2013). In the end of the 60 days of storage, irradiated cheese samples exhibited smaller amounts of α_{s1}- and β-CN degradation products as compared to both Cr and Cp. Seisa et al. also reported that irradiation slowed down the enzymatic hydrolysis of the β-CN and α_{s1}-CN during ripening of Cheddar cheese at 16°C (Seisa et al., 2004). They claimed that irradiation can cause protein degradation resulting in differences in the development of the peptide profile of Cheddar cheese. Ham et al. reported a dose dependent decrease in α_{s1}-CN and βA1-CN in both milk and Queso Blanco cheese after irradiation (Ham et al., 2009). They did not see difference at βB-CN levels at 5 kGy and below. They found an increase in αs0-CN and βA3-CN due to irradiation and they postulated that βA3-CN was formed by the breakdown of βA1-CN during irradiation. Nyamakwere et al. did not report any difference in the amount of whey proteins and caseins after irradiation at 5 kGy, except for β-CN which was reduced dramatically (Nyamakwere et al., 2022).

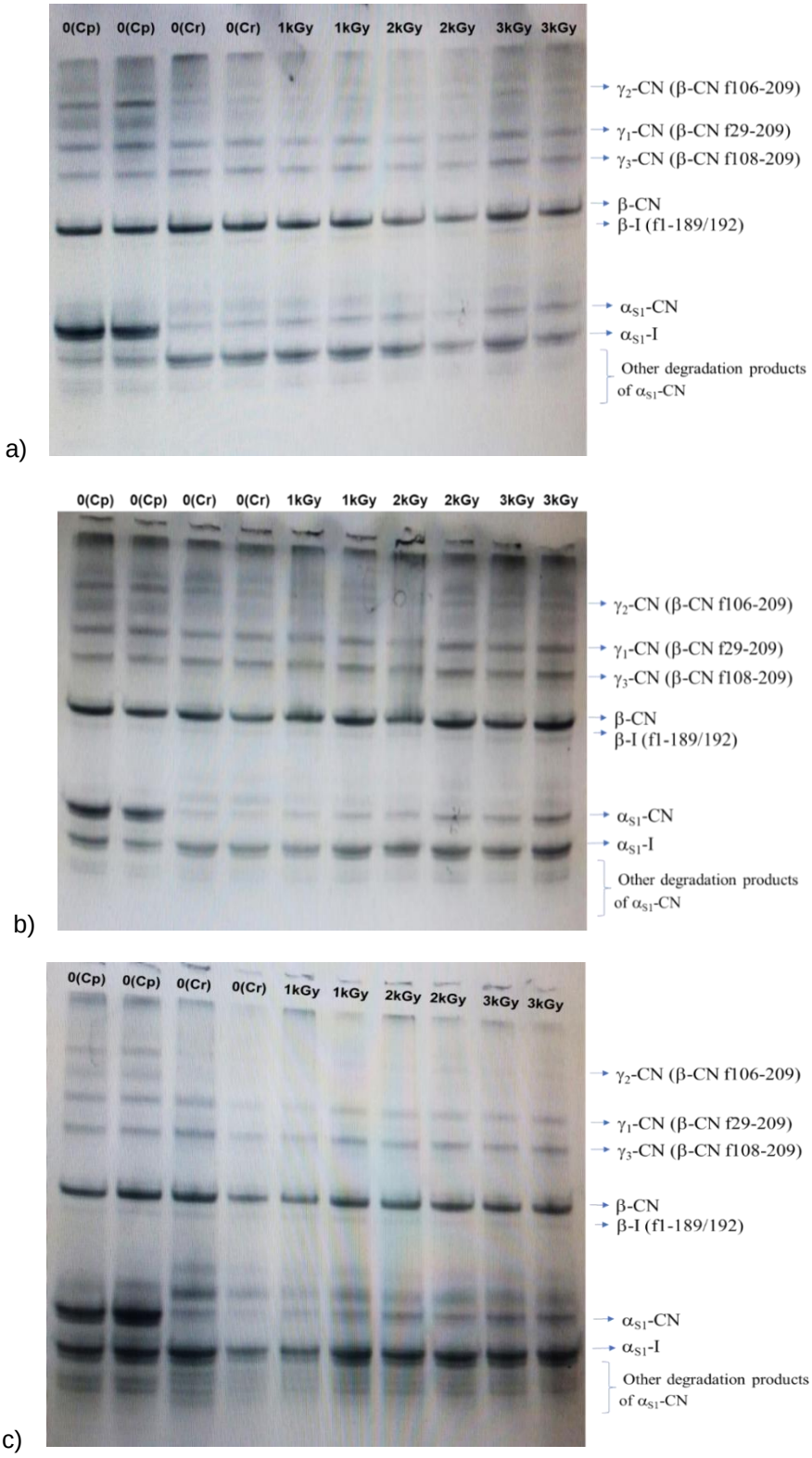

Figure 1. Urea-PAGE results of the non-irradiated (0(Cp): Control pasteurized cheese, (0)Cr: Control raw milk cheese) and irradiated (1 kGy, 2 kGy, 3 kGy) cheese samples a) on day 1, b) on day 30 and c) on day 60.

Microbiological analysis results

Changes of microbial populations of irradiated and non-irradiated White cheeses during storage are given in Fig 2. Impact of the irradiation on microbial counts was dose dependent and 3 kGy irradiation was more effective. Irradiation of the raw milk cheese at 2 and 3 kGy dose reduced the TMAB counts to the levels achieved by pasteurization (Cp). Other studies have also reported a decrease in total aerobic bacteria counts due to irradiation (Nyamakwere et al., 2022, Konteles et al., 2009). There was an increase in TAMB count of the Cp at day 30 with a higher rate than Cr, and a decrease was observed at day

60. Irradiated cheese TAMB counts were rather stable during the storage except for 3 kGy, which decreased at day 60.

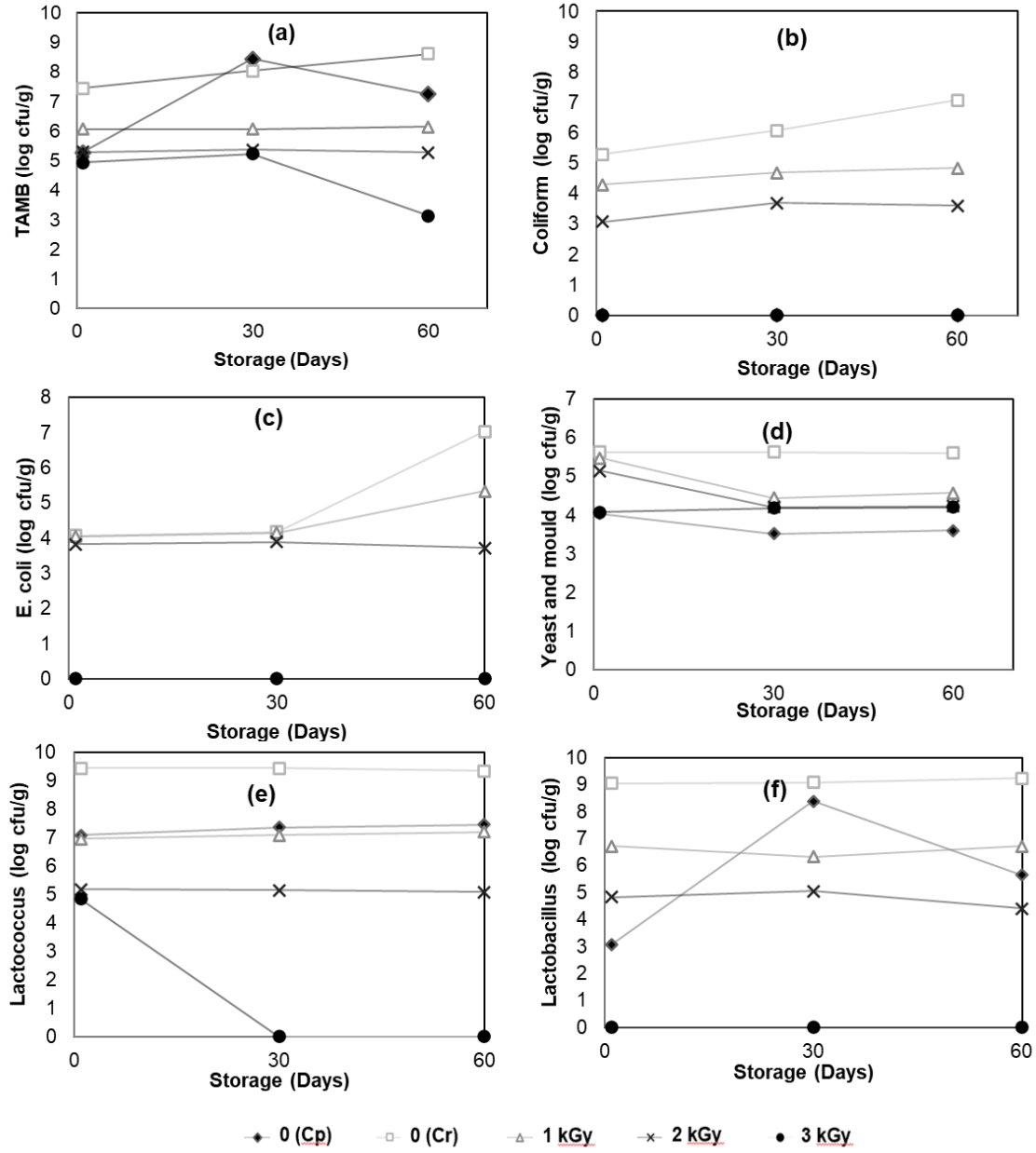

Figure 2. Changes of microbial populations of irradiated (1 kGy, 2 kGy, 3 kGy) and non-irradiated (0(Cp): Control pasteurized cheese, (0)Cr: Control raw milk cheese)White cheeses during storage: (a) TAMB (total aerobic mesophilic bacteria), (b) coliform bacteria, (c) *E. coli*, (d) yeasts and molds, (e) *Lactococcus* and (f) *Lactobacillus*

Coliform group bacteria and *E. coli* were not detected at Cp and 3 kGy cheese samples at any storage point. On the other hand, Cr had about 5 log cfu/g coliforms at day 1, and counts increased about 2 logs by 60 days (Fig 2b). Irradiation reduced the coliforms 1 log at 1 kGy dose, and 2 logs at 2 kGy on day 1. Irradiation doses below 3 kGy was not effective on *E. coli* counts (Fig 2c). While there was an increase in *E. coli* counts of Cr and 1 kGy on day 60, 2 kGy sample counts didn't change. Previous studies have also reported a dose dependent reduction in coliform counts, where a complete inhibition was achieved at high irradiation doses (Nyamakwere et al., 2022, Shalaby et al., 2016, Konteles et al., 2009). It is claimed that irradiation can cause death of the pathogens or sublethal injury preventing their multiplication by damaging their cellular membrane or DNA (Wang et al., 2018). Irradiation at 3 kGy reduced the yeast and mold counts to a similar level with Cp, however that was still high probably due to the initial high load of the raw milk.

The Cr cheese had the highest *Lactococcus*

counts (Fig 2e). Irradiation at 1 kGy was effective to reduce the *Lactococcus* counts to a level achieved by pasteurization, and a higher reduction was observed at 2 and 3 kGy doses on day 1. *Lactococcus* counts were stable during the storage except for the 3 kGy samples, where a complete reduction occurred at day 30. No *Lactobacillus* colonies were detected at 3 kGy sample throughout the storage (Fig 2f). The highest *Lactococcus* counts were observed at Cr and irradiation reduced the counts in a dose dependent manner. Seisa et al. reported that irradiation greatly reduced the number of non-starter lactic acid bacteria of the Cheddar cheese (Seisa et al., 2004).

Conclusion

Irradiation treatment effectively inhibited *E. coli* and coliforms at 3 kGy dose. All microbial group counts were reduced by irradiation application in a dose dependent manner. We determined that microbial safety of the irradiated cheese at 3 kGy dose was similar to pasteurized milk cheese for the analyzed microbial groups. It should be considered that irradiation would influence the ripening and flavor development in cheese since it has a detrimental effect on most microbial groups and slowed down the degradation of α_{s1}-CN and β-CN especially at 3 kGy. Irradiation treatment could be applied after some aging period to prevent any undesirable effects on ripening. Safety of raw milk artisan cheeses are concerned, and irradiation could be a good option to avoid pasteurization which impairs native flora and the desired flavor development during initial cheese making process. Irradiation appears to have an influence on proteins and degradation of caseins. More study is required to investigate the possible radiolytic products that could form after irradiation of the cheese.

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