

QUALITY CHANGES IN FISH BURGER FROM COMMON CARP (*CYPRINUS CARPIO*) DURING REFRIGERATED STORAGE

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Abstract: Fish burgers from Common carp (*Cyprinus carpio*) were assessed for proximate, chemical, microbiological and sensory quality changes over a period of 19 days at refrigerated storage at (4⁰ C). These burgers were separated into three different Experimental Samples (Sample A: Control Sample-No ginger; Sample B: 5gm ginger/kg burger mix; Sample C: 10gm ginger/ kg burger mix) and the changes in the moisture, protein, fat and ash content were significant at P<0.01. TVBN, PV, FFA, TBA content, Total plate count and Psychrophilic counts increased significantly in all the samples during the entire storage period. *Salmonella* spp, *Vibrio* spp, *Staphylococcus aureus*, *Faecal streptococci*, *Escherichia coli* and molds were found to be absent. The color changes of the burgers were also studied during the entire storage period. According to sensory scores, the three Experimental Samples of fish burgers had a shelf life of 15, 17 and 17 days, respectively. A negative correlation was observed between storage period and overall acceptability scores.

Keywords: Common carp, Fish burger, Quality, Colour, Refrigerated Storage.

Introduction

Fish production increased from a level of 0.75 million tons in 1950-51 to 9.58 million tons in 2013-14 comprising 3.44 million tons of marine and 6.14 million tons of inland fish (FAO, 2014). In India, carps are the most important species that supports Indian freshwater fisheries. Among the freshwater fish produced in India, the major carps of India (*Catla catla*, *Labeo rohita* and *Cirrhinus mrigala*) and Chinese carps (*Cyprinus carpio*, *Ctenopharyngodon idella* and *Hypophthalmichthys molitrix*) form a major component of Indian aquaculture, with a market share of over 90%. They can be easily cultured in low – input technology systems due to their herbivorous feeding habit (Sehgal and Sehgal, 2003). Freezing and frozen storage of fish burgers are commonly used because of the consistent, reliable quality, ease of transportation and the fact that they are very close to fresh equivalents (Tokur *et al.*, 2004). The demand for ready to eat and/or ready to cook products

are gradually growing because of their convenience (Yerlikaya *et al.*, 2005). Burgers are simple and a cost effective means of converting carps into value added convenience products. Battered and breaded or coated systems have the benefits of versatility and familiarity because they enhance the flavour and texture of processed food products (Biswas *et al.*, 2004). The main parameters which affect the shelf life of the stored fish products are the production form, features of packing material, storage temperature, packing process and machines that are used (Baygar *et al.*, 2008).

The aim of present study is to develop ready to eat fish products (fish burger) from Common carp (*Cyprinus carpio*) and to determine the rate and the type of a deterioration process that occurs during the refrigerated storage using sensory, bacteriological, physical and chemical assessments.

Materials and Methods

Common carp (*Cyprinus carpio*) harvested from freshwater culture ponds was iced in the ratio of 1:1 and transported to the laboratory within 20 minutes. The fish were washed with chilled water and dressed to remove head and viscera. Meat was separated from dressed fish using rotary type deboner (Safe World, Malaysia). The deboned fish meat was later minced using a mincer (Sirman, Italy) to obtain uniform size meat particles. The fresh minced meat obtained from common carp fish was used for the preparation of fish burger.

Each fish burger was a mean 25g weight and composed of 65% fish mince, 15% potatoes, 1% sugar, 4% corn flour, 0.2% pepper powder, 5% onion, 1.5% green chili, 2.2% bread crumbs, 1.5% salt, 0.1% polyphosphate, 3.5% shrimp essence and 1% egg white. After proper mixing, it was separated into three different Experimental Samples: Sample A: Control Sample, No ginger; Sample B: 5gm ginger/kg burger mix was added; Sample C: 10gm ginger/ kg burger mix was added in order to explore the anti-oxidative property of ginger when added to the value added fishery products. The fish burgers prepared using a standard recipe were flash fried at 180°C for 30 seconds in refined sunflower oil until the color turned slightly brown. The products were later cooled, packed in HDPE pouches (200 g/pouch), sealed, labeled and stored at 4°C for quality evaluation. The analyses were done on the every alternate day up to 19 days. The samples were drawn randomly for analyses and were performed in triplicate for biochemical and duplicate for microbiological parameters.

Moisture, fat, protein and ash were analyzed as per AOAC (2000). pH value of the homogenate was recorded using a digital pH meter (Oakton, Eutech instruments, Malaysia). Total volatile base nitrogen (TVBN) was determined by the micro diffusion method of

Conway (1962). Thiobarbituric acid value (TBA) was assessed by the method of Tarladgis *et al.* (1960), peroxide value (PV) and free fatty acid (FFA) were estimated according to Jacobs (1958) and Olley and Lovorn (1960) respectively. The samples were evaluated for Commission Internationale de L'Eclairage lightness (L*). Redness (a*), yellowness (b*) and hue angle (arctan, b*/a*), which describes hue or color of the ground meat and saturation index $(a^{*2}+b^{*2})^{0.5}$ which describes the brightness or vividness of color. All values were determined from the mean of eight measurements of each fillet. The Spectrocolorimeter was standardized using white (L*=0) standard tiles and working standards before being used in instruments.

Total plate count (TPC), *Staphylococcus aureus*, *Escherichia coli* and *Faecal streptococci* were determined by spread plate method using Soyabean Casein Agar medium, Baird Parker Agar, Tergitol 7 Agar and Kenner Faecal streptococcus Agar, respectively and were incubated at 37 °C for 24 to 48 h. Psychrophilic bacteria was determined using Nutrient Agar and plates were incubated at 4±1°C for 1 week. *Salmonella* and *Vibrio* were analyzed by streaking method using Brilliant green Agar and Thiosulphate Citrate Bile salt Sucrose mediums. For molds, Potato Dextrose Agar Medium was used as per APHA (1992).

Sensory characteristics of the fish burgers were evaluated by selected panel members of the institute who have experience in evaluation of similar products, on a ten-point scale (IS: 6273[II] 1971; Vijayan 1984). Scores were assigned with '1' being the least and '10' being the highest for attributes. The characteristics covered under the taste panel were appearance, colour, flavor, taste, texture and overall acceptability. The products were deep fried in refined sunflower oil until they were cooked before being presented to the panelists. Scores 10, 9, 8, 7, 6, 5, 4, 3, 2, 1 were taken for excellent, like extremely, like very much, like moderately, like slightly, neither like nor dislike, dislike slightly, dislike moderately, dislike very much and dislike extremely respectively for each of the sensory characteristics. The observations were converted to equivalent numerical scores and a sensory score of 4 was taken as the borderline of overall acceptability.

The SPSS (Statistical Package for Social Sciences) 19 (IBM 2010) statistical package was used for analysis of the experimental results. Sufficient numbers of samples were carried out for each analysis. The results were expressed as mean ± standard deviation (SD). The correlation coefficients between the parameters were carried out using the same software. One way ANOVA was performed by the Duncan test to find the significance difference between storage days.

Result and Discussion

The yield of mince was 55.57% from whole Common carp and the initial moisture, protein, fat and ash content of Common carp mince were found to be $77.84 \pm 0.31\%$, $16.95 \pm 0.10\%$, $3.15 \pm 0.07\%$ and $1.17 \pm 0.03\%$ respectively.

Proximate Composition

The proximate composition of the product is given in Tables 1, 2, 3 and 4. During refrigerated storage, the moisture content of fish burger decreased from 55.25 % to 50.84 %, 55.26% to 51.72 % and 55.66 % to 52.24 % in Sample A, Sample B and Sample C respectively. A decrease in the moisture content was noticed in fish burgers by Vanitha *et al.* (2015), the moisture content in fish burger decreased from an initial value of 55.08 % to 52.78% in fish burgers. Moisture content reduction in fish burger might be due to release of water from fish burger during cooking (Haq *et al.*, 2013). During storage, protein decreased significantly ($p < 0.01$). The decrease may be attributed to the leaching out of the water soluble nitrogenous components, during storage along with moisture. An increase in the crude fat content has been observed throughout the storage period. The increase in fat can be attributed to the decrease in moisture content as they are inversely proportional. Ejaz *et al.* (2013) found the lipid content of Pangus (*Pangasius sutchi*) fish mince was 4.89% and fish burger was 6.82%. The ash content in the product was higher than the fresh fish mince, due to the addition of the ingredients during the preparation of fish burgers. The remaining percentage of the proximate composition is thought to be due to carbohydrates (Tokur *et al.*, 2006).

Chemical analysis

The pH of fish burger decreased from 6.65 to 6.06 in Sample A, whereas in Sample B and Sample C the pH decreased from 6.63 to 6.04 and 6.59 to 6.00, respectively. Metin *et al.* (2002) reported that the pH of fish burgers decreased from 6.5 to 5.6 which may be due to fermentation of potato and bread ingredients of burger. At the initial stage of storage period, PV of Sample A, Sample B and Sample C fish burgers was 4.84, 4.80 and 4.72 meqO₂/kg of fat and it increased to 7.27, 7.26 and 7.18 meqO₂/kg of fat at the end of 5th day of storage and subsequently decreased to 5.23, 5.17 and 5.12 meqO₂/kg of fat respectively, at the end of 19 days of storage period. The decrease of the PV at the end of the storage may occur owing to decomposition of hydro peroxides into secondary oxidation products and similar trend was also observed by the Yerlikaya *et al.* (2005) during the refrigerated studies of fish patties from anchovy. The FFA increased significantly ($P < 0.01$) from 1.21 to 1.77 % of Oleic acid

for Sample A, 1.19 to 1.72 % of Oleic acid for Sample B and 1.18 to 1.69 % of Oleic acid for Sample C fish burgers, respectively. The increase in FFA may be due to cooking process of mince prior to preparation of product (Ninan *et al.*, 2011) and flash frying after preparation of the product might have deactivated the lipolytic enzymes.

TBA content showed an increasing trend in Sample A, Sample B and Sample C fish burgers from the initial value of 0.49, 0.47 and 0.45 mg MA/kg of sample to 0.89, 0.87 and 0.83 mg MA/kg of sample, respectively by the end of storage period. TBA is a secondary breakdown product of lipid oxidation and widely used as an indicator of degree of lipid oxidation (Aubourg, 1999). The TVBN content increased in Sample A, Sample B and Sample C fish burger from an initial value of 5.41 to 8.23 mg /100g sample, 5.39 to 7.99 mg/100g sample and 5.37 to 7.93 mg /100g sample, respectively, during 19 days storage period at refrigerated temperature. According to Adebona (1978) the increase in TVBN with length of storage is mainly attributed to the production of ammonia.

Table 1: Changes in the moisture content of fish burgers during refrigerated storage.

Storage period (Days)	Moisture* (%)		
	Fish burger		
	Sample A	Sample B	Sample C
0	55.25±0.08 ^j	55.26±0.03 ^j	55.66±0.32 ^g
1	55.02±0.22 ⁱ	55.08±0.08 ⁱ	55.36±0.29 ^{fg}
3	54.54±0.39 ^h	54.68±0.34 ^h	54.94±0.04 ^f
5	54.13±0.35 ^g	54.24±0.10 ^h	54.83±0.03 ^{ef}
7	53.67±0.19 ^f	53.88±0.15 ^g	54.36±0.24 ^{de}
9	53.52±0.37 ^f	53.87±0.19 ^f	54.03±0.03 ^{cd}
11	53.01±0.08 ^e	53.37±0.37 ^e	53.69±0.20 ^{bcd}
13	52.44±0.36 ^d	52.80±0.01 ^d	53.22±0.02 ^{ab}
15	51.94±0.19 ^c	52.77±0.01 ^c	53.07±0.04 ^a
17	51.38±0.21 ^b	52.26±0.11 ^b	52.68±0.10 ^a
19	50.84±0.12 ^a	51.72±0.09 ^a	52.24±0.07 ^a

*Each value is represented as the mean±SD of n= 3.

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Table 2: Changes in the crude protein content of fish burgers during refrigerated storage.

Storage period (Days)	Crude Protein* (%)		
	Fish burger		
	Sample A	Sample B	Sample C
0	18.12±0.01 ^j	18.26±0.03 ^k	18.25±0.02 ^g
1	18.05±0.02 ^j	18.22±0.08 ^j	18.17±0.04 ^g
3	17.86±0.04 ⁱ	18.05±0.04 ⁱ	18.05±0.04 ^f
5	17.65±0.02 ^h	17.80±0.03 ^h	17.77±0.04 ^e
7	17.37±0.04 ^g	17.48±0.04 ^g	17.58±0.03 ^d
9	17.07±0.04 ^f	17.29±0.06 ^f	17.26±0.04 ^c
11	16.71±0.03 ^e	17.08±0.06 ^e	17.00±0.01 ^b
13	16.51±0.06 ^d	16.91±0.05 ^d	16.89±0.03 ^a
15	16.31±0.03 ^c	16.71±0.04 ^c	16.80±0.03 ^a
17	16.16±0.02 ^b	16.62±0.05 ^b	16.70±0.02 ^a
19	16.07±0.04 ^a	16.41±0.02 ^a	16.58±0.01 ^a

*Each value is represented as the mean±SD of n=3.

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Table 3: Changes in the fat** content of fish burgers during refrigerated storage.

Storage period (Days)	Fat* (%)		
	Fish burger		
	Sample A	Sample B	Sample C
0	13.24±0.06 ^a	13.20±0.01 ^a	13.16±0.21 ^a
1	13.59±0.01 ^b	13.55±0.03 ^{ab}	13.40±0.28 ^b
3	14.11±0.14 ^c	13.93±0.01 ^b	13.88±0.20 ^c
5	14.56±0.02 ^d	14.32±0.01 ^c	14.21±0.27 ^d
7	14.99±0.02 ^e	14.90±0.00 ^d	14.82±0.10 ^e
9	15.65±0.01 ^f	15.56±0.01 ^e	15.42±0.16 ^f
11	16.09±0.09 ^g	16.01±0.01 ^f	15.81±0.14 ^g
13	16.76±0.14 ^h	16.41±0.02 ^g	16.19±0.14 ^h
15	17.05±0.19 ⁱ	16.88±0.01 ^h	16.71±0.24 ⁱ
17	17.53±0.12 ^j	17.12±0.02 ⁱ	17.08±0.12 ^j
19	17.89±0.09 ^k	17.52±0.01 ⁱ	17.38±0.17 ^k

** On dry weight basis

*Each value is represented as mean±SD of n=3.

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Table 4: Changes in the Ash content of fish burgers during refrigerated storage.

Storage period (Days)	Ash* (%)		
	Fish burger		
	Sample A	Sample B	Sample C
0	2.69±0.01 ^a	2.70±0.01 ^a	2.72±0.01 ^a
1	2.72±0.00 ^{ab}	2.73±0.01 ^a	2.78±0.01 ^a
3	2.77±0.01 ^{bc}	2.78±0.04 ^a	2.89±0.06 ^b
5	2.79±0.00 ^{cd}	2.85±0.09 ^b	2.92±0.05 ^c
7	2.82±0.12 ^{cde}	2.89±0.07 ^{bc}	3.09±0.01 ^d
9	2.83±0.10 ^{de}	2.90±0.01 ^{cd}	3.15±0.01 ^e
11	2.85±0.05 ^e	2.97±0.01 ^d	3.17±0.06 ^{ef}
13	2.97±0.01 ^f	3.04±0.02 ^e	3.19±0.07 ^{ef}
15	3.04±0.03 ^g	3.11±0.01 ^e	3.20±0.03 ^f
17	3.09±0.02 ^{gh}	3.17±0.01 ^f	3.24±0.07 ^{fg}
19	3.15±0.01 ^h	3.23±0.02 ^f	3.29±0.02 ^g

* Each value is represented as the mean±SD of n=3.

^{abcdefghijklm}Means followed by the same superscript within a column are not significantly different (P > 0.01).

In Sample A, Sample B and Sample C fish burgers, the L* values increased from 24.73, 27.69 and 29.51 to 28.96, 30.25 and 32.68, respectively. The hue angle of Sample A, Sample B and Sample C increased from 0.98, 1.33 and 1.39 to 1.35, 1.39 and 1.45, respectively. The a* values of Sample A, Sample B and Sample C decreased from 18.59, 13.89 and 16.78 to 11.75, 10.46 and 14.01 respectively, b* values of Sample A, Sample B and Sample C decreased from 18.23, 18.42 and 23.35 to 14.11, 14.56 and 19.42 and saturation indexes of Sample A, Sample B and Sample C were decreased from 26.03, 23.07 and 28.75 to 18.78, 18.02 and 24.09 respectively, by the end of refrigerated storage period. The color alteration in burgers occurs in meat when the heme protein becomes denatured, iron is oxidized into ferric, and the hemepigment remains intact (Ganhao *et al.*, 2010).

Microbiological analysis

The changes in the total plate count (TPC) and psychrophilic count during storage were enumerated and the results are presented in Table 5 and 6 respectively. TPC analyses of fish products did not exceed the maximum levels (7 Log. cfu/g of meat) of microbiological criteria for fresh and frozen fish given by the ICMSF (1978). A steady increase in TPC was observed during refrigerated storage. The initial TPC of three samples of fish burgers were 5.0×10^2 cfu/g of sample, 2.84×10^2 cfu/g of sample and 1.56×10^2 cfu/g of sample, respectively. At the end of 19 days of storage, the counts increased to 7.0×10^4 cfu/g of sample, 5.80×10^4 cfu/g and 3.71×10^4 cfu/g, respectively. According to Vanitha *et al.* (2015), the total plate count increased from an initial number of 4.06×10^2 cfu/g of meat to 5.52×10^4

cfu/g of meat for fish burger at the end of 15 days of storage. The psychrophilic bacteria count of Sample A, Sample B and Sample C fish burgers were initially 2.63×10^2 cfu/g of sample, 1.62×10^2 cfu/g of sample and 1.07×10^2 cfu/g of sample, respectively. At the end of the storage period these values reached 3.71×10^4 cfu/g of sample, 3.09×10^4 cfu/g of sample, 2.23×10^4 cfu/g of sample, respectively. *Salmonella* spp, *Vibrio* spp, *Staphylococcus aureus*, *Faecal streptococci*, *Escherichia coli*, yeast and mould were not detected in Sample A, Sample B and Sample C, fish burgers during the entire storage period. This was also found in many studies (Baygar *et al.*, 2008; Kilinc and Cakli 2005; and Vanitha *et al.*, 2015).

Table 5: Changes in the total plate count (TPC) of fish burgers during refrigerated storage.

Storage period (Days)	Total plate count* (cfu/gram of sample)		
	Fish burger		
	Sample A	Sample B	Sample C
0	5.00×10^2 (2.69)	2.84×10^2 (2.45)	1.56×10^2 (2.19)
1	7.16×10^2 (2.85)	4.24×10^2 (2.62)	2.39×10^2 (2.38)
3	1.07×10^3 (3.03)	9.50×10^2 (2.97)	4.30×10^2 (2.63)
5	1.90×10^3 (3.27)	1.80×10^3 (3.25)	8.40×10^2 (2.92)
7	4.78×10^3 (3.67)	3.10×10^3 (3.49)	1.30×10^3 (3.11)
9	9.50×10^3 (3.97)	5.10×10^3 (3.70)	2.36×10^3 (3.37)
11	1.03×10^4 (4.01)	9.90×10^3 (3.99)	4.70×10^3 (3.67)
13	2.91×10^4 (4.46)	2.10×10^4 (4.32)	9.70×10^3 (3.98)
15	3.71×10^4 (4.57)	2.91×10^4 (4.46)	2.44×10^4 (4.38)
17	5.80×10^4 (4.76)	3.71×10^4 (4.57)	2.91×10^4 (4.46)
19	7.00×10^4 (4.84)	5.80×10^4 (4.76)	3.71×10^4 (4.57)

*Each value is represented as the mean of two estimates

Figures in the parenthesis indicates Log. Bacterial count

cfu = colony forming units

Table 6: Changes in the Psychrophilic bacterial count of fish burgers during refrigerated storage

Storage period (Days)	Psychrophilic count* (cfu/gram of sample)		
	Fish burger		
	Sample A	Sample B	Sample C
0	2.63×10^2 (2.42)	1.62×10^2 (2.21)	1.07×10^2 (2.03)
1	4.67×10^2 (2.67)	3.89×10^2 (2.59)	2.95×10^2 (2.47)
3	6.76×10^2 (2.83)	6.45×10^2 (2.81)	5.75×10^2 (2.76)
5	1.04×10^3 (3.09)	1.04×10^3 (3.02)	9.77×10^2 (2.99)
7	3.38×10^3 (3.53)	2.95×10^3 (3.47)	2.08×10^3 (3.32)
9	6.30×10^3 (3.80)	5.75×10^3 (3.76)	4.07×10^3 (3.61)
11	1.02×10^4 (4.01)	9.54×10^3 (3.98)	7.76×10^3 (3.89)
13	1.62×10^4 (4.21)	1.02×10^4 (4.01)	1.02×10^4 (4.01)
15	2.44×10^4 (4.38)	1.47×10^4 (4.17)	1.23×10^4 (4.09)
17	2.91×10^4 (4.46)	2.10×10^4 (4.32)	1.47×10^4 (4.17)
19	3.71×10^4 (4.57)	3.09×10^4 (4.49)	2.23×10^4 (4.35)

*Each value is represented as mean of two estimates

Figures in the parenthesis indicates Log. psychrophilic count
cfu = colony forming units

Sensory Analysis

The overall mean acceptability scores were found to decrease significantly ($p<0.01$) in all the three samples of fish burgers during the entire period of storage at refrigerated temperature (Table 7). On comparing, the overall acceptability scores with refrigerated storage period, the Sample A (Control, 0% Ginger), Sample B (0.5% Ginger) and Sample C (1% Ginger) fish burgers have been acceptable for 15 days, 17 days and 17 days, respectively. The results of correlation coefficient (r) and regression analysis between refrigerated storage period (days) and the overall acceptability scores for fish burgers are presented in Table 8. Vanitha *et al.* (2015) reported that, the shelf life of fish burgers prepared from Catla (*Catla catla*) was found to be 15 days. Coban (2013) reported that the fish fingers contained 1% ginger oil produced from *Sarda sarda* as an alternate product can be stored for 17 days in 4°C without undesirable changes of sensory and chemical quality. The increase in shelf life of Sample B and Sample C compared to Sample A (Control) may be attributed due to the antibacterial effect of ginger.

Table 7: Changes in the Overall acceptability scores of fish burgers during refrigerated storage

Storage period (Days)	Overall acceptability scores		
	Fish burger		
	Sample A	Sample B	Sample C
0	9.15 \pm 0.10 ^h	9.22 \pm 0.06 ^h	8.98 \pm 0.10 ⁱ
1	8.76 \pm 0.01 ^g	8.80 \pm 0.17 ^g	8.72 \pm 0.14 ^{hi}
3	8.29 \pm 0.12 ^g	8.55 \pm 0.17 ^f	8.36 \pm 0.19 ^h
5	7.72 \pm 0.18 ^f	7.79 \pm 0.19 ^e	7.64 \pm 0.22 ^g
7	7.32 \pm 0.16 ^f	7.35 \pm 0.16 ^e	7.22 \pm 0.18 ^f
9	5.54 \pm 0.15 ^e	6.73 \pm 0.17 ^d	6.64 \pm 0.26 ^e
11	5.47 \pm 0.19 ^e	6.34 \pm 0.27 ^d	6.06 \pm 0.13 ^d
13	4.68 \pm 0.11 ^d	5.31 \pm 0.17 ^c	5.37 \pm 0.24 ^c
15	3.73 \pm 0.18 ^c	4.25 \pm 0.24 ^b	4.45 \pm 0.23 ^b
17	3.51 \pm 0.12 ^b	4.05 \pm 0.17 ^b	4.10 \pm 0.21 ^b
19	3.20 \pm 0.09 ^a	3.65 \pm 0.12 ^a	3.87 \pm 0.19 ^a

*Each value is represented as mean \pm SD of n=7

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Table 8: Correlation Coefficient and Regression equations for mean overall acceptability scores during storage period and shelf life of fish burgers during refrigerated storage.

Fish Burgers	Regression equation	Correlation coefficient	Shelf-life (Days)
Sample A	$y = -0.695x + 10.21$	0.988*	15
Sample B	$y = -0.597x + 10.13$	0.990*	17
Sample C	$y = -0.562x + 9.863$	0.998*	17

Conclusion

Fish burgers prepared from Common carp (*Cyprinus carpio*) were assessed for proximate, chemical, microbiological and sensory quality changes over a period of 19 days at refrigerated storage at (4°C) with different levels of antioxidants. The Ginger incorporated at 0.5% and 1% level shows better sensory scores and shelf life over normal burger prepared from Common carp (*Cyprinus carpio*).

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