

## Effect of Black Pepper (*Piper Nigrum* L.) on the Keeping Quality of Spiced Cottage Cheese

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### Research Article

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#### ABSTRACT

Cottage cheese is a soft, unripened, pleasant mild acidic, meat-like texture with discrete curd particles and diacetyl flavour. Nutritionally, Cottage cheese is designated as slim cheese with a low calorific value of 96 K cal/100 g, low lactose consequently with the reduced cholesterol and saturated fatty acids. Cottage cheese has limited shelf life of 8-10 days, due to the high moisture content (~75%) and high pH (~5.0). This effects wide spread of marketing and distribution. Hence, this study was investigated to improve the shelf life and sensory properties of cottage cheese by using black pepper (*Piper nigrum* L.), a natural antioxidant substance and contain organic compounds having antagonistic activity against microorganisms. The effect of three different levels of Black pepper powder i.e., 0.5, 1.0 and 1.5% on the physico-chemical, microbiological and sensory properties were studied at 3 days interval under refrigeration (4-5°C) temperature. Best results were obtained by using 1 percent black pepper powder by inhibiting psychrotrophs, yeast and molds as well as retard the acidity development and lowering the proteolysis in the cottage cheese. Finally, that addition of black pepper powder at the level of 1.0% improved the properties as well as the aesthetic quality of the spiced cottage cheese and extended the shelf life from 8 to 14 days without any quality deterioration of the cottage cheese.

#### INTRODUCTION

Cottage cheese is a highly regarded dairy product. It is being acclaimed as the best source for high protein, with low fat. In view of characteristic delicate flavour, cottage cheese could be consumed with almost any food. It combines well with salads, vegetables, all types of biscuits and fruits. Nutritionally, cottage cheese is a wholesome low calorie food. Cottage cheese could be a healthy part of a weight loss plan. Moreover, high protein content found in cottage cheese (about 10 per cent by weight) also makes it a great protein source for vegetarians and those who do not like meat products<sup>[1]</sup>. It is body building without leading to obesity as it is easily digestible and assimilable than other concentrated hard cheeses.

Shelf life of cottage cheese hardly exceeds 7 days, due to high moisture content (~75%) and relatively high pH (~5.0). Cottage cheese provides an excellent environment for the growth of many microorganisms especially Gram-negative Psychrotrophic bacteria (e.g. *Alcaligenes* spp, *Pseudomonas* spp, *Achromobacter* spp, *Escherichia* spp, and *Micrococci* spp). Other important spoilage psychrotrophic bacteria found on the surface of cottage cheese are yeasts (*Rhodotorula* spp. and *Torulopsis* spp.) and moulds (*Geotrichum* spp, *Mucor* spp. and *Penicillium* spp). Together with a high water activity and protein level, the pH of cottage cheese (pH=4.7-5.3) is favourable for some acid tolerant spoilage causing microorganisms. Moreover, the salt content in Cottage cheese (1-2%) is too low to retard the growth of these spoilage causing microorganisms<sup>[2,3]</sup>. There has been increased interest in specialty cheese including cheese with additives like herbs, spices and vegetables. It has short shelf life, moreover since it is rich in milk protein and a very costly product. Hence it is out of economy to reach the majority of Indian people. Due to the ever growing demand of cheese by varied health conscious consumers, it is necessary to develop the cottage cheese with high nutritive value by extending the shelf life.

Black pepper (*Piper nigrum* L.) popularly known as “The King of Spices”. By its nature it is spicy, aromatic and carminative due to the presence of piperamides which are the pungent bioactive alkaloids. It is a natural antioxidant. It acts as anti-inflammatory, anticancer, antiperiodic and antipyretic. It contains mainly vitamins A, C, E, K, niacin and  $\beta$ -carotene and traces of minerals such as iron, calcium, phosphorous. It can also help to lowers body cholesterol levels. The amino acids contained by black pepper, works for enhancing the bioavailability of nutrients. Ascorbic-acid (0-10 ppm), Beta-carotene (0.114-0.128 ppm), Lauric-acid (400-447 ppm), myristic-acid (700-782 ppm), palmitic-acid (12,200-13,633 ppm), piperine (17,000-90,000 ppm) <sup>[4]</sup>.

Murlidhar and Goswami <sup>[4]</sup> reported that the black pepper is an antioxidant phenolic compound and contain main flavour ingredient is piperine. Piperine, the active ingredient in pepper, exerts substantial analgesic and antipyretic effects. Addition of pepper to foods increases their keeping characteristics and prevents their spoilage, due to the antimicrobial properties of pepper. The essential oil of pepper is found to be inhibitory to *Vibrio cholerae*, *Staphylococcus albus*, *Clostridium diphtheriae*, *Shigella dysenteriae*, *Streptomyces faecalis*, *Bacillus* spp, *Pseudomonas* spp, etc. Pepper leaf oil also exhibits antifungal activity.

The spices are used as seasonings in foods and beverages, because it contains Antimicrobial activities which mainly improves the shelf life of food. Spices contain phenolic compounds, one of the most important groups of natural antioxidants, which can reduce oxidative cell damage. Antimicrobial activity of spice against different types of microbes, including some foodborne pathogens have been investigated. Makhil et al. <sup>[5]</sup> reported that promising results were obtained by using spices in dairy products like cheese and paneer, contains phytochemicals mainly to inhibit psychrotrophic bacteria, yeast and molds.

Spices are used on surface of some chesses to impart flavour and to improve the shelf life by arresting the growth of spoilage causing organisms <sup>[6]</sup>. Black pepper possesses more antimicrobial properties than ginger used in soy- milk and kunun- zaki on 0.4 -1% and used as a natural antimicrobial preservative to extend the shelf life of food <sup>[7]</sup>. The antimicrobial action of phenolic compound which effect on cellular membrane as it causes structural and functional damage to plasma membrane, it arrests the growth of microorganisms. Therefore, the objective of this study, were made to enhance the shelf life of the cottage cheese without any quality deterioration of the product by incorporating black pepper at different levels of i.e., 0.5, 1.0 and 1.5% on the physico chemical, microbiological and organoleptic properties was investigated on the spiced cottage cheese.

## MATERIALS AND METHODS

Fresh skim milk (<0.5%) procured from Students Experimental Dairy Plant (SEDP), Dairy Science College, Karnataka Veterinary Animal and Fishery Sciences University, Bengaluru, pasteurized skim milk was transferred to a cheese vat and was manufactured by the following procedure <sup>[8]</sup>.

Black pepper (*Piper nigrum* L.) powder by SNAPIN™, DUVAL Enterprises Pvt Ltd., Mumbai was used for the dressing of cottage cheese. Stater cultures: Cultures such as *Lactococcuslactis* ssp. *Lactis*, *L lactis* ssp. *cremoris* and *L. lactis* ssp. *lactisbv. diacetylactis* and in the form of freeze dried direct Vat set (FD-DVS) was obtained from Chr. Hansen Laboratories, Denmark.

The three levels of black pepper powder was achieved based on the results of sensory evaluation of the fresh product. Calculated amount of black pepper powder were mixed with the curd dressing to attain the desired concentrations in the final product.

### Procedure for Manufacture of Cottage Cheese with Black Pepper

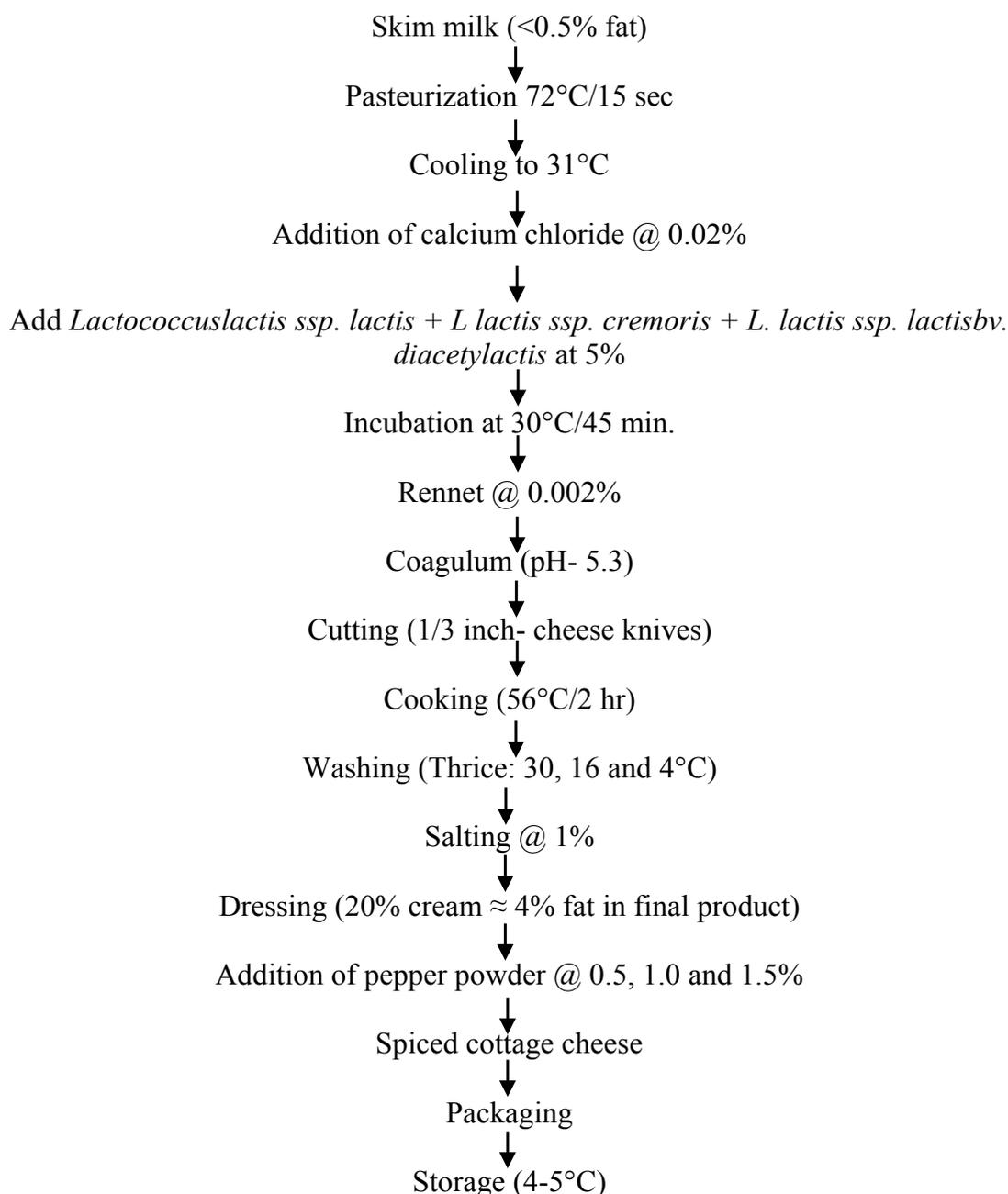
Cottage cheese was prepared by using skim milk <0.5% fat (**Figure 1**). The spiced cottage cheese samples were analyzed for sensory, biochemical as well as microbiological quality for 3 days interval during the storage at refrigeration temperature.

### Physico-Chemical Analysis of Cottage Cheese

The following physico chemical parameters were analyzed in the cottage cheese dressed with black pepper powder.

#### Determination of moisture

The moisture content was determined as per the procedure in IS: SP 18 Part (XI) <sup>[9]</sup>.



**Figure 1.** Procedure for manufacture of cottage cheese with black pepper.

#### **Determination of fat**

Fat percentage was determined by Mojonnier fat extraction method as per IS: SP 18 Part (XI) <sup>[9]</sup>.

#### **Determination of protein**

The protein content was determined by Kjeldahl method described in in IS: SP 18 Part (XI) <sup>[9]</sup>.

#### **Determination of water activity**

The water activity was determined by using Rotronic water activity meter.

#### **Statistical Analysis**

All treatments were evaluated in triplicate. The results obtained were statistically analyzed by using R Programme, R-Version 3.1.3 for ANOVA.

## **RESULTS AND DISCUSSION**

The sensory characteristics of cottage cheese by incorporation of black pepper at different levels are presented in **Table 1**. Cottage cheese containing 1 per cent black pepper awarded highest sensory scores. **Table 1** reveals that colour and appearance

scores for all treated and control samples were statistically significant ( $P \leq 0.05$ ). Increase in the incorporation of black pepper at above 1 per cent levels leads to decrease the sensory scores for spiced cottage cheese. Judges opined that the cottage cheese prepared by incorporating the black pepper above 1 per cent makes product different from that of control because presence of fine black specks in treated samples due to incorporated whole pepper powder.

**Table 1.** Effect of different levels of black pepper on sensory characteristics of spiced cottage cheese.

Pepper (%)	Colour and Appearance	Body and Texture	Flavour	Overall acceptability
Control	8.11 <sup>a</sup>	8.17 <sup>a</sup>	8.20 <sup>a</sup>	8.10 <sup>a</sup>
0.50	8.11 <sup>a</sup>	8.17 <sup>a</sup>	8.20 <sup>a</sup>	8.21 <sup>a</sup>
1.00	8.30 <sup>b</sup>	8.23 <sup>b</sup>	8.50 <sup>b</sup>	8.76 <sup>b</sup>
1.50	7.50 <sup>c</sup>	7.69 <sup>c</sup>	7.20 <sup>c</sup>	7.43 <sup>c</sup>
CD ( $P \leq 0.05$ )	0.11	0.05	0.28	0.28

The colour and appearance for control and 0.5 per cent level of black pepper were found to be non - significant. The highest score with respect to body and texture of 8.23 was recorded for cottage cheese containing 1.0 per cent black pepper whereas, significantly lowest score of 7.69 were awarded to product with 1.5 per cent. Statistical analysis revealed that different levels of black pepper has significant effect on body and texture of cottage cheese. This could be due to the large number of fine black pepper particles and slightly hard or dry body.

There was decrease in the sensory scores with increase in black pepper levels. The highest score with respect to flavour of 8.50 was recorded for cottage cheese containing 1.0 per cent black pepper whereas, lowest score of 7.20 was awarded to product with 1.5 per cent. Cottage cheese prepared by incorporation of black pepper powder at 1 per cent was found to be highly acceptable compared to other samples. Wendoref and Wee [6] reported that coating of cheese with mixed spice oil was organoleptically highly acceptable than the uncoated cheese. Essential oils or spices have been used extensively as flavour ingredients in wide variety of beverages and confectionery products.

The cottage cheese with 1.0 per cent level of pepper secured highest average sensory scores of 8.76 compared to 0.5 and 1.5 per cent levels of pepper, while the lowest score of 7.43 was secured by cottage cheese with 1.5 per cent of pepper. The control, 0.5, 1.0 and 1.5 per cent levels of pepper were found to have significant difference. Incorporation 1 per cent black pepper powder and it was significantly superior to that of control. Further incorporation of black pepper powder had significantly decreased the sensory scores. This may be due to presence of pepper particles as indicated by judges. Similarly Arunkumar [10] incorporated of black pepper powder at different levels 0.5, 1.0, 1.5 and 2.0 per cent in paneer spreads and reported that incorporation of 1 per cent black pepper powder was found to be highly acceptable compared to the other paneer spreads.

Incorporation of black pepper powder in cottage cheese showed decrease in the moisture and water activity. The moisture content of spiced cottage cheese samples were declined slowly and steadily during storage, the fresh samples varied from 73.45 to 73.00%. At the end of storage, it ranges from 71.20 to 71.00% depending upon the storage period.

The pH increases and acidity decreases by incorporation of black pepper. The pH of the fresh samples of cottage cheese ranges from 4.73 to 4.80 afterwards, it steadily decreased upto a certain period of storage depending upon the level of black pepper added to the cottage cheese.

The acidity in control and the spiced cottage cheese were 0.5, 1.0 and 1.5. Initial acidity of the fresh spiced cottage cheese samples ranges from 0.61 and 0.62% slow and gradual increase in the acidity during storage. Acidity of the control sample increased after 7 days of storage, in comparison to the spiced cottage cheese. Addition of black pepper powder in cottage cheese delayed the acid development, because of its antimicrobial activity against the spoilage causing microorganisms, which arrest the growth and improve the biological value of spiced cottage cheese. **Figure 2** illustrates the cottage cheese samples with addition of black pepper powder exhibits an increase in acidity to a certain period followed by subsequent rise in acidity after 12 days of storage.

Increase in concentration of black pepper powder, slowly changes in pH because of fact that black pepper at higher concentration which exert a strong antimicrobial activity against the spoilage causing and also lactose fermenting organisms. With the elevation in concentration of pepper powder, occurrence of yeasts and molds was delayed, because it has an antifungal action (**Table 2**).

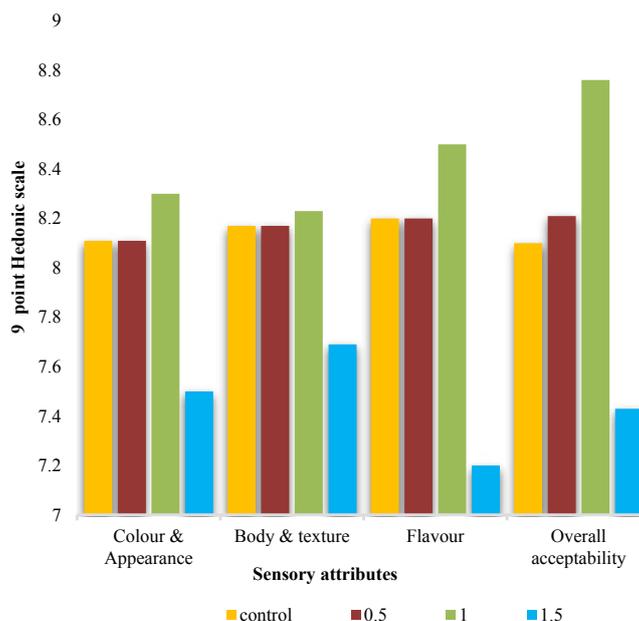


Figure 2. Sensory evaluation using 9 point hedonic scale.

Table 2. Chemical characteristics of spiced cottage cheese with different levels of black pepper.

Pepper (%)	Moisture (%)	Acidity (%)	pH (%)	Water activity (a <sub>w</sub> )
Control	73.45 <sup>a</sup>	0.61 <sup>a</sup>	4.73 <sup>a</sup>	0.982 <sup>a</sup>
0.5	73.30 <sup>a</sup>	0.60 <sup>a</sup>	4.70 <sup>a</sup>	0.978 <sup>a</sup>
1.0	73.00 <sup>b</sup>	0.61 <sup>a</sup>	4.83 <sup>b</sup>	0.979 <sup>a</sup>
1.5	73.00 <sup>b</sup>	0.60 <sup>a</sup>	4.80 <sup>b</sup>	0.979 <sup>a</sup>
CD(P ≤ 0.05)	0.32	NS	0.08	NS

Use of black pepper was effective in controlling the growth of microorganisms in cottage cheese. The fresh control sample showed yeast and molds counts of 0 to 0.30103 log cfu/g while spiced cottage cheese exhibited no occurrence of yeast and molds up to 8 days. The yeast and molds in the spiced cottage cheese samples was noticed during 12 to 18 days of storage. Therefore with progressing storage period, yeast and molds count was gradually increased, which was followed by a gradual rise towards the end of storage period regardless of treatment. Yeast and molds counts of the fresh cottage cheese should not exceed 1 log cfu/g [11]. Further breakdown of lactose into lactic acid that happened during the end of storage period intensified the proliferation of yeast and molds. Fresh samples of spiced cottage cheese, yeast and molds count was below 1 log cfu/g.

Coliforms count of fresh spiced cottage cheese samples during storage were below 1 log cfu/g. The count remained within 1 log cfu/g in T<sub>2</sub> and T<sub>3</sub> stored upto 4 days. The corresponding count in the control and T<sub>1</sub> samples reached to 1.875061 and 1.30103 log cfu/g respectively. On the 16<sup>th</sup> day the control sample showed higher coliforms, when compared with the spiced cottage cheese samples. Each sample showed a gradual increase in coliforms counts by a rapid growth the end of the storage. While on 18<sup>th</sup> and 20<sup>th</sup> day T<sub>1</sub> and T<sub>2</sub> samples showed similar observations when compared with T<sub>3</sub> sample (Figure 3).

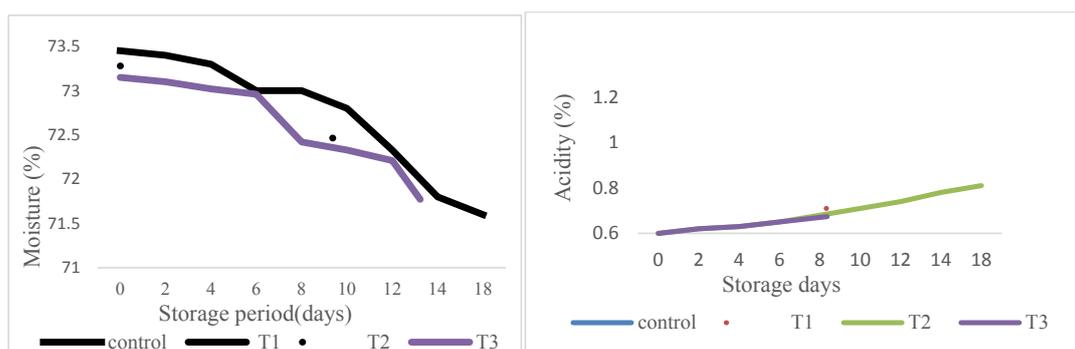


Figure 3. Chemical characteristics of spiced cottage cheese with different levels of black pepper (Moisture, Acidity) stored at 4-5°C of samples T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub> treated with black pepper of 0.5, 1.0 and 1.5% respectively.

Although the antimicrobial activity of phenolic compounds is well established in spices, the inhibitory mechanisms of these compounds are against microorganisms. It is also advocated that the antimicrobial action of essential oils from spices due to impairment of a variety of enzyme systems, including those involved in energy production and structural component synthesis of microorganisms [12]. Similarly Udensi et al. [14] reported the comparative studies of Incorporation of black pepper in soymilk and kunun-zaki.

Final results reported that, as spice concentration increases there was a reduction in microbial load. Black pepper recorded lower microbial load, thus having more antimicrobial activity. Black pepper used as a natural antimicrobial preservative were used to extend the shelf life of food and dairy products (Table 3).

**Table 3.** Different levels of black pepper on (A) Yeast and molds (B) coliforms (log cfu/g) counts of spiced cottage cheese at refrigeration temperature at 4-5°C.

Days	Control	Levels of Black pepper		
		T <sub>1</sub> (0.5)	T <sub>2</sub> (1.0)	T <sub>3</sub> (1.5)
<b>A. Yeast and Moulds count</b>				
0	0.30103	-ve	-ve	-ve
4	0.77815	-ve	-ve	-ve
8	1.977724	0.69897	0.30103	-ve
12	2.824776	1.832509	1.681241	0.7781
16	ND	2.491362	2.133539	1.97724
18	ND	ND	2.352183	2.133539
20	ND	ND	ND	2.824776
<b>B. Coliforms</b>				
0	<1	<1	<1	<1
4	1.875061	1.30103	<1	<1
8	2.096961	1.748188	1.447158	1.20412
12	2.491362	2.09691	1.778151	1.68121
16	3.50515	2.824776	2.3522183	2.120574
18	ND	ND	2.565848	2.447158
20	ND	ND	ND	3.875061

### CONCLUSION

The consumption of cheese products is increasing in recent years, mainly due to excellent nutritional properties. The nutritive value of cottage cheese is comparatively higher than some of the other cheese products. Consumption of this spiced cottage cheese not only improves the nutritional status but also improves the therapeutic properties. In this investigation the different levels of black pepper powder on the cottage cheese was carried out mainly to increase the shelf life. Among three levels i.e., 0.5, 1.0 and 1.5 per 100 g of cheese, it was noticed that with increasing the level of black pepper powder, cottage cheese underwent minimum changes. Addition of 1.0 percent black pepper powder in cottage cheese enhanced the shelf life from 8 to 14 days. Keeping quality of cottage cheese by 5 days against the control sample without any noticeable adverse effect on the typical flavour of cottage cheese. Hence, it was also observed that addition of black pepper powder in cottage cheese at 1.0% level considerably improved the flavour as well as the aesthetic quality of the product as well as extended the shelf life of the product from 8 to 14 days without effecting the sensory and textural properties in comparison to the control sample.

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