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THE COMPOSITIONAL AND MICROBIAL QUALITY OF COMMON AND NEWELY SPICY MISH PRODUCED BY COMMERCIAL DAIRY PLANT IN KHARTOUM, SUDAN

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Abstract: Objective: This study aimed to evaluate and compare the quality of two types of mish using different types of spices. Methods: The common mish flavored with fenugreek (0.7% w/w) and black cumin (0.5% w/w) while the newly spicy mish flavored with green fennel (0.5% w/w) and black pepper (0.15% w/w). The chemical and microbiological properties were examined for both types of mish. The tests were done on the first day of manufacturing and then weekly for 3 weeks for eight samples from four batches for both types of mish. All mish samples were subjected to chemical analysis (protein, fat, total solids and ash) and measure the acidity and enumeration of total bacteria, coliform, yeast and mold and psychrotrophic bacteria. Results: The chemical results showed that there were highly significant differences in the level of fat and acidity ($P<0.001$) between mish samples and also significant differences in total solids during the storage period ($P<0.05$). Microbiological results showed non-significant differences for all groups of microorganisms tested. Conclusions: Both types of mish did well in microbiological quality compared to the other fermented dairy products. However, the newly spicy mish showed slightly lower content of chemical composition compared to the common mish. This study recommended that more work on mish should be done for adding new spices with different ratios in order to maintain the quality of the products.

Keywords: Mish, Spices, Chemical composition, Microbiological quality, Storage period, Health benefit

INTRODUCTION

Recently, developing a variety of fermented milk products for some other beneficial purposes has gotten much more attention, particularly for health purposes and prevention of toxins produced by foodborne pathogens and spoilage bacteria[1]. Although under spontaneous fermentations, the growth of LAB (lactic acid bacteria) cannot be predicted or controlled, which let this procedure practice and carry out traditionally for years [2]. Yoghurt, mish, sour cream and buttermilk are examples of fermented milk products which were made by various micro-organisms addition being added to concentrated milk, cream and skimmed milk [3]. Mish differs from yoghurt in having higher total solids content, higher degree of acidity and the added spices, including black cumin, fenugreek and garlic [4]. Mish, which is a semisolid fermented milk product, is manufactured in Sudan, Egypt and many other Mediterranean countries [5]. Mish, a popular sour product, is a concentrated traditional dairy fermented product, currently being

manufactured by modern dairy industry in Sudan[6].

The methods of mish processing were described previously [4-8]. The shelf life of mish can be extended, but this is not practiced on a large scale. The heat treatment of milk before mish processing improved the quality of this product [9].

Chemical analysis of mish samples revealed 11.83±2.96% total solids, in which 5.09±2.9% for protein, 2.83±0.93% for fat, 2.34±0.98% for lactose and 1.24±0.41% for lactic acid [8]. Mish can be kept in a good microbiological quality for up to 21 days [4,10-11]. However, being judged by acidity, the visual observations of products that the mish produced by modernized methods could be kept for longer period up to 30 days [4]. To this term, the added spices may play a good role in extending this period [9].

Black pepper is regarded as important for its medicinal value [12-16] while fennel has antibacterial activity [17-19]. The object of this study was evaluating the effect of adding green fennel and black pepper into the products and comparing the chemical and microbiological properties of newly spicy mish with common mish during storage.

METHODS

Source of samples

Sixty-four commercially produced mish samples were obtained from retailers in Khartoum State from September 1 to December 31 in 2015.

The samples represent two different type of mish. The common mish flavored with fenugreek (0.7% w/w) and black cumin (0.5% w/w) while the newly spicy mish flavored with green fennel (0.5% w/w) and black pepper (0.15% w/w). Four batches of samples from each types of mish were obtained from the same company (Best Dairy plant); each batch contains 8 samples. All samples were transported to the laboratory in an ice box held whose temperature under 4°C and analyzed for chemical and microbiological characteristics. Guided by date coding, the samples were analyzed in every seven days from the processing date during the shelf life (21 days).

Samples preparation

The fresh milk was obtained from the milk collection center in Khartoum North that belongs to the producing company. After it was received, the Quality Control Unit of plant examined it to assure that it was of good quality. Whole and skim milk powder (New Zealand) were also analyzed first. The spices used were bought from a local market.

Manufacturing procedure for mish samples

Mish was processed by using starter culture consisted of Streptococcus thermopiles and Lactobacillus delbruckii sub sp Bulgaricus (YO-mix 505, Dansco, Denmark). The starter culture was prepared by adding 4.2 grams from dry culture to the milk base (100 liters) and was allowed to be fermented until the desired degree of acidity was reached (0.20-0.25%). The inoculation of starter culture to mish base was added at a rate of 1-2%.

Mish's preparational steps were illustrated previously [4]. The mix was prepared by using high solids non-fat (22%-23%) from skim milk powder plus whole milk powder and fresh milk. The specific amount of the spices that was added to the common mish include salt (1% w/w), garlic (0.05% w/w), black cumin (0.5% w/w) and fenugreek (0.7% w/w). However for the newly spicy mish salt (1% w/w), garlic (0.05% w/w), green fennel (0.5% w/w) and black pepper (0.15% w/w) were used. These were mixed thoroughly to give the final desired product. After filling the mish in the retail plastic containers (250 and 150 grams), the containers were coded with batch number and date. Then the products were left for 48 hours for ripening before their distribution to the markets.

Chemical analysis

The fat content was determined by Gerber method and the ash was determined by gravimetric method [20]. The protein content was determined by Kjeldahl method and the total solids content were determined by the modified method of AOAC [21]. The acidity of the mish samples was also determined by the titration described by the AOAC [21].

Microbiological analysis

All media were obtained in dehydrated forms and prepared according to the manufactures' instructions. Plate count agar (Biomark, B 298) was used to determine the total bacterial count that was incubated at 32 °C for 48 hours [22] and psychotropic bacteria count was incubated at 7º C for 10 days [23]. Yeast extracts glucose chloramphenicol agar (YGC agar, Merck) was used to ration the total yeast and mold. Violet red bile agar (Merck) was used to ration the coliform count according to the manufacturers' instructions.

Sterilization of glass wares such as test tubes, pipettes, flasks and bottles were heated in hot oven at 180 °C for one hour. Ringer solution and tips were sterilized by autoclaving for 15 minutes at 121 °C [24].

Culturing methods

1ml of the mish sample was transferred aseptically by sterile pipette to 9ml sterile ringer's solutions. This procedure was repeated to make serial dilution [25]. 1ml from each selected dilution was cultured at 32 °C for 48 hours using pour plate technique for the total bacterial count while the coliform count was incubated at 37°C for 24 hours. Yeasts and molds count were estimated after incubation at 28°C for 5 days. The plates' content was described in Marshall [24].

Statistical analysis

The data of the present study was analyzed statistically using randomized complete block design. ANOVA test was used to determine the significant levels. The least significant difference was used for mean separation at P<0.05. The analysis was conducted by using SPSS 10.0 package program.

RESULTS AND DISCUSSION

Chemical composition of mish

Table.1 and Table.3 showed that the mean of total solids content was 24.97±0.15% for common mish, of which the minimum was 24% and the maximum was 26%. The newly spicy mish content of total solids was 23.02±0.36%, of which the minimum was 19% and the maximum was 25%. A significant difference was found for the total solids during storage in both common and newly spicy mish (P<0.001). The values reported for the total solids revealed that the higher value was 30.93%±0.187% and the lowest value was 18.59±0.187% [8].

Table.1 The chemical composition of ordinary mish and newly spicy mish

Chemica compo-sition / Types of mish	Total solids		Fat		Protein		Ash		Titratable acidity	
	24.97		3.47		6.42		2.61		2.7	
Common mish	Max	Min	Max	Min	Max	Min	Max	Min	Max	Min
	26	24	3.9	3	6	8	3.4	2.3	2.9	1.8
	23.02		2.75		5.63		2.76		2.3	
Newly spicy mish	Max	Min	Max	Min	Max	Min	Max	Min	Max	Min
	25	19	3.7	2	6	4.6	3.5	2.3	2	1.8
Overall mean	23.99		3.11		6.03		2.69		2.51	
S E	0. 187		0.05		0.078		0.043		0.04	
L S	N S		***		N S		N S		***	

Table.2 The microbiological quality of ordinary mish and newly spicy mish

Micro-bial loads / Types of mish	TBC (log)		Coliform (log)		Yeast and mold (log)		Psychotropic (log)	
	9.66		0		0		0.4	
Com-mon mish	Max	Min	Max	Min	Max	Min	Max	Min
	10	9.1	0	0	0	0	1	0
	9.49		0		0.35		0.5	
Newly spicy mish	Max	Min	Max	Min	Max	Min	Max	Min
	9.1	9	0	0	2	0	2	0
Overall mea	9.58		0		0.18		0.65	

n				
S E	0.036	0	0.06	0.08
L S	N S	N S	N S	N S

Table.3 The effect of storage period on the chemical and microbiological properties of mish samples

Storage period / Parameters	Total solids	Fat	Protein	Ash	TBC (log)	Coliform (log)	Yeast And Mold (log)	Psychotropic (log)
Day 1	24.78^{a}	2.82^{b}	5.78^{a}	2.79^{a}	9.55	0	0	0
Day 7	23.44^{ab}	3.19^{a}	5.90^{a}	2.68^{a}	9.49	0	0	0
Day 14	23.35^{ab}	3.24^{a}	6.22^{a}	2.76^{a}	9.63	0	0	0
Day 21	24.38^{b}	3.22^{a}	6.21^{a}	2.53^{a}	9.64	0	0	0
Overall means	23.99	3.12	6.03	2.69	9.58	0	0	0
S E	0.187	0.052	0.078	0.041	0.036	0	0	0
L S	S *	S *	N S	N S	N S	N S	N S	N S

The slightly higher content of total solids found for the common mish compared with newly spicy mish could be due to high fenugreek seeds protein content that revealed 20%-30% [26]. An analysis of fennel showed 9.5% protein [27]. It might also be due to the antimicrobial properties of spices used in mish as black cumin which was used on common mish was found to inhibit the growth of some pathogenic bacteria [9]. Moreover, it was recommended that the processing of mish using spices like black cumin, fenugreek, garlic and other known spices, since those spices were proved to have significant effect as preservative [8].

Table.1 and Table.3 illustrated that the mean fat content of common mish was 3.48±0.04%, the minimum was 3.0% and the maximum was 3.9%, which were higher than the value (2.83±0.93%) found for fat content of mish [8]. The mean fat content of newly spicy mish was lower as it revealed 2.75±0.11%, the minimum was 2.0% and the maximum was 3.0%. The values were less than the result reported previously for mish [8]. They found the maximum fat content of mish was 6.82±0.103% and the minimum value of fat content was 5.27±0.103%. The obtained values were also lower than those reported in a previous report [4]. It was reported that the mean fat content was 6.98±0.151% for mish produced by modern method. The maximum was 7.2% and the minimum was 6.5%. The mean fat content was 5.89±0.285% for mish produced by traditional method while the maximum was 6.5% and minimum was 5.2% [4].

Table.2 showed significant difference ($P<0.05$) for fat content on the interaction between time and treatment. The higher fat content of common mish compared to the newly spicy mish might be due to black cumin seeds, it is rich in unsaturated fatty acids especially polyunsaturated fatty acids (48-70%), monounsaturated (18-29%) and saturated fatty acids (12 -25%) [28].

Table.1 and Table.3 showed that the mean protein value was 6.42±0.045%, the minimum was 6.0% and the maximum was 6.9% for common mish, which were made by adding fenugreek and black cumin. The mean value was 5.6±0.16%, the minimum was 4.6% and the maximum was 6.9% for newly spicy mish, which were made by replacing fenugreek and black cumin by green fennel and black pepper. These results were agreed with El Zubeir et al. who had reported that the mean value of mish protein was 5.09±2.80% [8]. However, the values were lower than those reported by Abdel-Gader et al. [4]. They reported that protein content in mish produced by modern method was 10.260±0.342% for mean value, 9.2% and 10.7% for the minimum and maximum while 7.9±0.372% for mean value, 7.6% and 9% for the minimum and maximum values for mish produced by traditional method.

The higher protein value of common mish might be due to the addition of fenugreek used for common mish. It was reported that fenugreek contain 20-30% protein [26].

However, the addition of fennel used in the newly spicy mish resulted in lowering the protein content. This might be because fennel contains 9.5% protein [27].

Table.1 and Table.3 showed that mean of ash content was 2.60±0.043% for common mish, the minimum was 2.3% and the maximum was 3.4%. The mean ash value was 2.78±0.078% for newly spicy mish, the minimum was 2.3% and the maximum was 3.5%, which were more than the result which reported that the ash content of mish was 1.26±0.6% [8]. Higher value (1.43%±0.161%) also was reported for ash content of mish produced by modern processing method [4]. The slightly higher content of ash appeared for newly spicy mish compared with common mish could be due to fennel seeds, which contain 13.4% mineral [27].

Table.1 and Table.3 showed highly significant differences ($P<0.001$) between treatments on titratable acidity. The mean of titratable acidity was 2.72±0.073% for common mish, the minimum was 1.8% and the maximum was 2.8%. However, the titratable acidity of newly spicy mish was lower than the ordinary mish. The mean was 2.3±0.05%, the minimum was 1.8% and the maximum was 2.9%. These results were much higher than that reported for the titratable acidity values (1.25±0.35% and 1.243±0.41% for roub and mish respectively) [8]. Similarly lower values were also reported for the titratable acidity (3.96±0.035%, 2.48±0.035% and 3.23±0.035%) for mish form three different manufacturing dairy plants [11]. However, the obtained values agreed with those which showed that the titratable acidity was 2.33% (2.28-2.35%) for mish produced by modern method and was more or less similar to 2.35±0.018% obtained for traditional method that ranging between 2.31% to 2.37 % [4]. Higher values were found titratable acidity (1.243±0.41%) for mish samples produced by Butana dairy factory [8].

Higher titratable acidity degree was found for the common mish compared with newly spicy mish (Fig.1). The variation could be attributed the difference of lactic acid when comparing mixture and mish to fermentation process and the addition of spices [8]. Similarly, significantly ($P<0.05$) higher acidity was found for Mudaffara cheese flavored with sesame compared to that flavored with black cumin [29]. Moreover, the medical and aromatic plants can show functional properties such as antioxidant, anti-inflammatory, antiallergic, anti-depressive and antimicrobial effects, depending on the active substances in their content. With these properties, they can also be used as spices, food supplementary and additives [30].

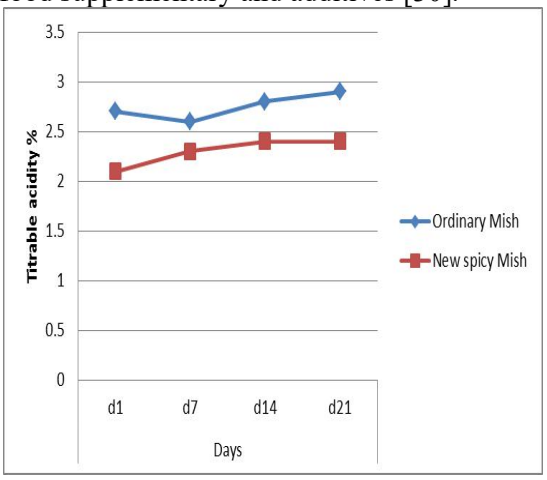

Fig.1 Titratable acidity measurement of common and newly spicy mish during storage

Microbiological composing of mish samples

The present data showed no significant differences were obtained in all microbiological means (total bacterial count, coliform count, yeast and molds count and psychotropic bacterial count). The mean for total bacterial count obtained was log 9.67 for common mish, the maximum was log 10.00 and the minimum was log 9.00 (Table.2 and Table.3). However, the total bacterial count mean was log 9.49 for newly spicy mish, the maximum was log 10.10 and the minimum was log 9.00 The obtained bacterial count might indicate the lower standards of hygiene practice in studied dairy plant [8-9,31]. Coliform, psychotropic bacteria, yeast and mold' s loads were log 9.11, log 0.96, log 6.24 and log 8.88 respectively for total bacterial count, yeast and mold, coliform and psychotropic bacteria [9]. The obtained values were also lower than those reported by Abdel-

Gader et al. [4]. They reported that the means of microbiological loads for mish produced by companies who used modern method were 5.2×10^4, 1.4×10 and 6.75 ± 0.3 respectively for total bacterial count, yeast and molds and coliform count. The means of microbiological loads for mish from the company used modern method were 2.27×10^5, 4.96×10^4 and 9.48×10^3 respectively for total bacterial count, yeast and molds and coliform count. Lower average of bacterial counts in mish samples produced after pasteurization of milk was reported for mish [9]. Hence, in order to eliminate the hazard of disease transmission, fermented dairy products should be prepared only from high-grade milk that has been adequately pasteurized, followed by cooling to ensure that the product is safe and free from post pasteurization contaminants and pathogenic microorganisms [9].

Total bacterial count of newly spicy mish showed slightly less values than common mish might be due to fennel seeds (Table.2 and Table.3). Dusko et al. showed that fennel was used to treat many bacterial, fungal, viral and mycobacterial infectious diseases. The antibacterial activity of fennel was shown [18-19]. Commonly fennel was added as one of the important spices to roub; A Sudanese sour milk; in most of prepared dishes in rural area of Sudan. The fennel decoction can be used as a natural conservation enhancer in cottage cheese, while bringing antioxidant properties to the final product [32]. Black pepper, which also was introduced in the newly introduced mish, was proved to have medicinal values [12-16].

The lower values of psychotropic bacterial count, yeast and mold count and the absent of coliform bacteria in the two types of mish (Table.2 and Table.3) might be due to spices used; as black cumin which was used on common mish inhibits the growth of some pathogenic bacteria [9]. Fennel was also used in newly spicy mish showed bacterial activity against Enterococcus facecalis, Staphylococcus aureus, Escherichia coli, pseudomonas aeruginosa, Salmonella typhi, Salmonella typhimurium and Shigella flexneri [19]. On the other hand, black pepper (Piper nigrum) and garlic (Allium sativum) are aromatic plants commonly used in dairy products as natural preservative additives due to their antimicrobial and antioxidant activity [30].

CONCLUSIONS

The newly spicy mish examined in this study showed good microbiological and chemical quality than the common mish. Hence, it is recommended that the use of spices in dairy products for their preservation and functional properties should be encourage. However, further studies are needed on how to use the different types, concentrations and mixtures of spices in order to get acceptable and good quality dairy products.

COMPETING INTERESTS

Nothing to be declared.

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