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Chemical composition and microbiological quality of Metata Ayib: a traditional Ethiopian fermented cottage cheese

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Abstract

The chemical composition and microbiological quality of Metata Ayib, a traditional Ethiopian fermented cottage cheese, were investigated in this study for the first time. A total of nineteen Metata Ayib samples procured from a local market in Merawi town in Northwest Ethiopia were used for the analysis. The Metata Ayib samples analyzed had average moisture, fat, protein and ash contents of 42.3 ± 5.1, 28.7 ± 8.4, 43 ± 6.9 and 3.2 ± 0.65 g/100 g, respectively. The average titratable acidity and pH values of the Metata Ayib samples were 0.43 ± 0.07% lactic acid and 4.0 ± 0.1, respectively. The Metata Ayib samples had a total bacterial count ranging from 2.5 x 10^5 – 6.9 x 10^7 cfu/g and yeast and mould counts ranging from 1.2 x 10^2 – 6.1 x 10^3 cfu/g. However, coliforms were not detected in the Metata Ayib samples analyzed. Based on these results, further investigation is needed into the identification and functional properties of the microflora of Metata Ayib towards possible development of starter cultures.

Introduction

Dairy products constitute an important component of the diets of many Ethiopians. Milk is consumed either in fresh or fermented (sour) form in the country. Although cows' milk is the most widely produced and processed milk, milk from goats, sheep and camels is also consumed mainly in the pastoral lowland areas of the country. The major dairy products produced and consumed in different parts of the country include fresh whole milk, sour milk (Ergo), butter, Arera (defatted sour milk) and a traditional cottage cheese called Ayib (O'Mahony, 1988). Ayib is a traditional cottage cheese made from defatted sour milk by heating the milk on a slow fire to precipitate the casein and some of the remaining fat. Ayib has a shelf life of only a few days and is either consumed at home or marketed.

However, in addition to the above-mentioned dairy products, some other traditional dairy products are expected to be present both in the mixed and pastoral production systems in the country. Metata Ayib of West Gojam Zone of the Amhara National Regional State is an example of such traditional dairy products which has recently been reported by Tassew (2007). Metata Ayib is a traditional fermented cottage cheese produced in Northwest Ethiopia. It differs from the traditional cottage cheese Aiyb in that its production involves the use of different spices and spontaneous fermentation for 15 days. The property of Metata Ayib has not been fully understood and characterized.

Understanding the properties of traditional dairy products and proper scrutiny of the indigenous processing steps are important in order to develop appropriate manufacturing procedure and to standardize the processing parameters. Therefore, this study was initiated to assess the chemical composition and microbiological quality of the traditional fermented cottage cheese Metata Ayib produced in Northwest Ethiopia.

Materials and Methods

Cheese samples

A total of 19 Metata Ayib samples (≈ 300 g each) were procured (each from a different vendor) from a local market in Merawi town located in West Gojam Zone of the Amhara Regional State in Northwest Ethiopia. The samples were placed in sterile bottles and delivered to Haramaya University Dairy Technology Laboratory for analysis. A portion of each cheese sample (≈ 150 g) was aseptically transferred into separate sterile containers and used

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for microbiological analysis. The age of the Metata Ayib samples procured ranged from 4-6 weeks. The cheese samples were analyzed within three days of collection.

Manufacturing protocol of Metata Ayib

The manufacturing steps and ingredients used during preparation of Metata Ayib were identified by interviewing the nineteen vendors from whom cheese samples were purchased using semi-structured questionnaire. Secondary sources were also referred to verify the processing steps and ingredients used (Tassew, 2007). The raw material used for the production of Metata Ayib is a traditional cottage cheese called Ayib (which in turn is manufactured from defatted sour milk). The manufacturing of Metata Ayib involves production of different batches of Ayib by heating defatted sour milk at 40-50°C for 30 min and drainage of whey for three days. The different batches of Ayib are mixed together and the spices *Brassica nigra* and *Coriandrum sativum* are added into the mixture after which whey is drained for three more days. Then the curd is mixed with ten additional spices listed in Table 1. The mixture is allowed to ferment naturally at ambient temperature in a tightly closed container for 15 days. Finally, the vessel is opened and Metata Ayib is ready for consumption.

Fat content

The Soxhlet fat extraction method was used to determine the fat content of Metata Ayib samples as described by AOAC (1995) method 933.05. Three grams of cheese sample was fed into a Soxhlet apparatus fitted with a 1-L round-bottomed flask and a condenser. The extraction was carried out for 8 h using n-hexane (boiling point 68-70°C). The solvent was removed by heating at 70°C in a hot dry oven (Model IV 50C, England). The recovered oil was then weighed and expressed as percent fat.

Moisture content

The moisture content of the cheese samples was determined by drying three grams of Metata Ayib samples in a forced draft oven (Model IV 50C, England) at 102 ± 2°C for 3 h (Bradley *et al*., 1993). The moisture content of the cheese samples were calculated according to Bradley *et al*. (1993).

Ash content

The ash content was determined by igniting the pre-dried cheese samples (≈ 2.0 g) (that were used for moisture determination) in a Muffle Furnace (Isotemp® Programmable Muffle Furnace 650-750 Series, Fisher Scientific) at 550°C. The samples were ignited until constant mass was achieved (≈ 4 h). The ash content was calculated according to Bradley *et al*. (1993).

Table 1. Spices used in the preparation of Metata Ayib

Vernacular name (Amharic)	Common name	Scientific name
Senafitch	Mustard	*Brassica nigra*
Dimbillael	Coriander	*Coriandrum sativum*
Zingebile	Ginger	*Zingiber officinale*
Korerima	Korerima	*Aframomum korerima*
Netchshinkurt	Garlic	*Allium sativum*
Zekakibe (Basobila)	Basil	*Ocimum basilium*
Tosign	Thyme	*Thymus serrulatus*
Tenadam	Rue	*Ruta graveolence*
Netch-azmud	Bishop's weed	*Trachyspermum ammi*
Tikur-azmud	Black cumin	*Nigella sativa*
Keyshinkurt	Shallot	*Allium cepa*
Abish	Fenugreek	*Trigonella foenum-graecum*

Protein content

The total nitrogen content of the cheese samples (1 g) was determined by the Kjeldahl method using Gerhardt apparatus (Kjeldatherm KB/KBL, Vapodest 40) as described by the International Dairy Federation (IDF, 1993). The crude protein content of the cheese samples was determined by multiplying the nitrogen content by the factor 6.38.

pH determination

The pH of the cheese samples was measured using a digital pH meter (Jenway 3320 model pH Meter, Jenway Limited).

Titratable acidity

The titratable acidity of the cheese samples was determined by measuring 10 g of finely ground cheese and adding water (40°C) to a volume of 105 mL. After vigorous shaking, the mixture was filtered and an aliquot of the filtrate (25 mL) was titrated with 0.1 N sodium hydroxide solution using phenolphthalein as indicator. The titratable acidity was expressed as % lactic acid and calculated according to the procedure of AOAC (1995) method 920.124. Each sample was analyzed in duplicate for each of the variables indicated above.

Microbiological analysis

The cheese samples intended for microbiological analysis were ground into a fine paste using a sterile pestle and mortar before weighing.

Standard plate count

The total viable bacteria count was determined by adding 1 g of cheese sample into test tubes having

9 mL of sterile quarter strength Ringer's solution. After thorough mixing using a vortex mixer (K MS1 Minishaker IKA, Fisher Scientific), serial dilutions were prepared under a horizontal laminar flow cabinet (Model HLE4B, ENVAIR Limited, USA) and 1 mL of appropriate dilutions were pour plated in duplicate using standard plate count agar (CDH, India) that was sterilized in a bench-top autoclave (Astell Scientific Ltd., UK). The plated samples were allowed to solidify and then incubated at 32ºC for 48 h (6). Colony counts were made using colony counter (Model SC5, Stuart Scientific, UK). The estimated number of total viable bacteria count per g of sample was calculated according to IDF (1991a).

Coliform count

One g of cheese sample was added into a test tube having 9 mL of sterile quarter strength Ringer's solution. After mixing the sample using a vortex mixer, serial dilutions were prepared and 1 mL of appropriate dilutions were pour plated in duplicate using violet red bile agar (CDH, India) that has been boiled according to the manufacturer's instructions. The plated samples were then allowed to solidify and incubated at 30ºC for 24 hours (Harrigan, 1998). Colonies were counted using colony counter. Colonies surrounded by dark red colour were considered as coliforms. Estimated number of coliforms per g of sample was calculated by the formula described by IDF (1991a).

Yeast and mould count

Yeast and mould count was made according to IDF (1991b) using chloramphenicol yeast extract agar. Plating was done as stated above for standard plate count and colonies were counted after incubating the plates at 25°C for 5 days. All analysis was done in duplicate.

Statistical analysis

The proximate composition and microbial count data were analyzed by descriptive statistics using SPSS program, version 12.0.

Results and Discussion

Spices used in Metata Ayib preparation

Table 1 shows the spices used during preparation of Metata Ayib. In Ethiopia, different spices are traditionally used to enhance flavour and keeping quality of milk and milk products (Beyene, 1994; Gonfa *et al*., 2001). Use of spices in the preparation of the traditional cottage cheese Ayib has been reported from different parts of the country (Fita, 2004; Kassa, 2008). The spices used in the preparation of Metata Ayib in the present study (Table 1) are in line with the types of spices used in the processing and storage of Ayib in Shashemene area in Southern Ethiopia (Kassa, 2008). Spices possess natural antimicrobial compounds and can be used for preservation of foods (de Souza *et al*., 2005).

Table 2. Proximate composition and acidity of Metata Ayib samples (n = 19)

Constituents	Mean ± SD
Moisture (g 100/g)	42.3 ± 5.1
Fat (g 100/g)	28.7 ± 8.4
Crude protein (g 100/g)	43.0 ± 6.9
Ash (g 100/g)	3.2 ± 0.65
Titratable acidity (%lactic acid)	0.43 ± 0.07
pH	4.00 ± 0.1

n = number of samples; SD = standard deviation

Chemical composition

Table 2 presents the gross chemical composition of Metata Ayib samples. The moisture content of the Metata Ayib samples ranged from 37.6-49.3%. The value of the moisture content of Metata Ayib observed in this study suggests that it is an intermediate moisture food and is much lower than the corresponding value (79%) reported for the traditional cottage cheese Ayib (Ashenafi, 1992). The lower moisture content of Metata Ayib may partly contribute to its reported long shelf life.

The fat content of the Metata Ayib samples ranged from 20.1-36.9%. The average fat content observed for the Metata Ayib samples analyzed is much higher than the fat contents 2 g/100 g and 7% of Ayib reported by Ashenafi (1992) and Gonfa *et al*. (2001), respectively. Similarly, the Metata Ayib samples analyzed have higher crude protein content than the traditional cottage cheese Ayib which was reported to be 15 g/100 g protein (O'Mahony, 1988; Ashenafi, 1992). The ash content of the Metata Ayib samples analyzed in the present study is also higher than the ash content (1.16%) of Ayib reported by Kassa (2008). The higher crude protein and ash contents of Metata Ayib suggest that it could serve as an important source of amino acids and minerals for human beings. Analysis of the mineral and amino acid composition of Metata Ayib could give an insight about the major types of amino acids and minerals that it contains and thus its actual nutritional value. Hence the amino acid and mineral profiles of Metata Ayib deserve investigation.

The pH of Metata Ayib samples analyzed ranged from 3.99-4.18 which shows that it is an acidic dairy product. The traditional cottage cheese Ayib was reported to have a pH value ranging from 3.7-4.6 (Ashenafi, 1992). Kassa (2008) reported a pH

value of 4.34 for Ayib samples collected from a local market in Shashemene area in Southern Ethiopia. Among other factors, the lower pH of Metata Ayib may partly contribute to its reported long shelf life. Tassew (2007) reported that Metata Ayib can be kept at ambient temperature for about three years without being spoiled.

A big variation in the gross chemical composition was observed among the different Metata Ayib samples for the variables considered. This could be attributed to the difference in the manufacturing procedure especially the spices used by the different producers. About twelve types of spices are used as ingredients during manufacturing of Metata Ayib (Table 1). Each individual producer may not use all the spices listed above and this could have contributed to the difference in chemical composition of Metata Ayib samples obtained from different producers. This calls for a need to standardize the manufacturing procedure with respect to spices used so as to produce Metata Ayib with consistent properties.

Microbiological quality

The Metata Ayib samples had total viable bacteria count ranging from 2.5 x 10^5 to 6.9 x 10^7 cfu/g. The total viable bacteria count of Metata Ayib samples observed in this study is lower than the corresponding value reported for Ayib samples. Ashenafi (1990) reported that the majority (92%) of Ayib samples collected from open air markets in Awassa had aerobic mesophilic counts greater than 10^7 cfu/g. The lower total plate count of the Metata Ayib samples analyzed in the present study as compared to that reported for the traditional cottage cheese, Ayib, might be associated to its low pH and low moisture content. The various spices used during manufacturing of Metata Ayib might also have exerted an inhibitory effect on the general microflora of the Metata Ayib samples. Kassa (2008) reported that treatment of Ayib with garlic juice improved the shelf life of Ayib without significantly affecting its overall acceptability. More research is needed to understand the factors involved in Metata Ayib preservation.

Coliforms were not detected in the Metata Ayib samples analyzed in the present study. However, over 32% of Ayib samples purchased from local markets in Awassa had coliform counts of more than 10^2 cfu/g (Ashenafi, 1990). The absence of coliforms in the Metata Ayib samples could be attributed to the low pH (4.0) of Metata Ayib which might have inhibited the growth of coliforms. The Metata Ayib samples had yeast and mould counts ranging from 1.2 x 10^2 to 6.1 x 10^3 cfu/g. The yeast and mould count observed in the present study is less than that reported for Ayib samples (Ashenafi, 1990). Ayib samples collected from local market in Shashemene area were reported to have mean aerobic mesophilic bacteria, Enterobacteriaceae and yeast and mould counts of 7.11, 3.55 and 6.35 log10 cfu/g, respectively (Kassa, 2008) which indicated a suboptimal microbial quality of market Ayib in the area. Since Metata Ayib is an acidic product, growth of yeasts and moulds on it is expected. It was reported that yeasts and moulds, which can grow at low pH values, could affect the flavour and keeping quality of Ayib (Ashenafi, 1990). Identification of the major yeast and mould species present in Metata Ayib could help design an effective method used to prevent spoilage of Metata Ayib by yeasts and moulds.

Fermentation of Metata Ayib takes place spontaneously without the use of a defined starter culture. Thus, isolation and characterization of the predominant microorganisms (lactic acid bacteria) responsible for the fermentation process would help to develop a starter culture for the production of Metata Ayib with acceptable taste, flavour and shelf-life attributes. The results of the present study suggest that production of Metata Ayib vis-à-vis the traditional cottage cheese Ayib is a novel indigenous practice that results in better microbiological quality and improved shelf-life. This is significant because most of the smallholder farmers who produce Metata Ayib do not have refrigeration facilities to preserve the product, thus, the preservative function of the spices used in production of Metata Ayib needs to be investigated further.

Conclusion

The data generated in this study is the first of its kind on Metata Ayib, a traditional Ethiopian fermented cottage cheese. The results revealed that Metata Ayib is a novel indigenous dairy product and has a better microbiological quality as compared to the traditional cottage cheese Ayib. Thus, further scientific investigations need to be conducted to fully understand the identity and functional properties of Metata Ayib microflora in order to improve and scale-up its production to a commercial level.

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