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EFFECT OF BACTERIAL CULTURE, FERMENTATION TIME AND INOCULUM SIZE ON QUALITY OF *EMASI* PRODUCED IN THE KINGDOM OF ESWATINI.

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Abstract

Emasi is a dairy product produced by fermenting milk at room temperature. Traditionally fermented milk may have low hygienic quality hence it is important to produce emasi that resembles the traditionally produced product with improved hygienic qualities. The aim of the study was to determine the effect of bacterial culture, fermentation time and inoculum size on sensory attributes and physiochemical properties of emasi. Commercial mesophilic lactic acid culture, isolated Leuconostoc mesenteroides ssp. mesenteroides/ dextranicum and Lactococcus lactis ssp. lactis, mixed culture from emasi sorghum meal and mixed culture from traditional emasi were used for fermentation for 12, 18 and 24 hours; at 2.5%, 5% and 10% (v/v) inoculum size. The samples were examined for physiochemical properties and sensory attributes. The effect of type of culture on physiochemical properties was significantly different (P < 0.05) between the sources. Titratable acid ranged from 0.96% to 1.22%, whilst viscosity ranged from 3551cP to 2222cP. The isolated bacterial strains had the lowest syneresis percentage whilst mixed culture from emasi sorghum meal had the highest. It ranged from 48.6% to 56.5%. The pH ranged from 4.26 to 3.87. The overall acceptability by the panellists showed that they preferred the commercial mesophilic lactic acid bacteria and isolated bacterial strains compared to the mixed culture of traditional emasi and emasi from sorghum meal produced emasi. Fermentation time and inoculum size did not have any significant effect on the physiochemical properties and sensory attributes of emasi. More studies need to be done on isolating bacterial strains that will produce characteristics that resemble the traditional product emasi.

Keywords: emasi, bacterial culture, inoculum size, fermentation

INTRODUCTION

Milk is a nutritious and essential food for human beings. It also serves as a good medium for the growth of many microorganisms (Robinson and Tamime, 1981, Dlamini *et al.*, 2015). Common micro-organisms that may grow in milk are *Lactobacillus*, *Streptococcus*, *Staphylococcus* and *Micrococcus spp*. (Srujana *et al.*, 2011). Milk from cows is consumed as fresh milk or fermented milk as part of a balanced diet for Eswatini households. Fermentation is the oldest means of preserving food (Robinson and Tamime, 1981). Traditionally fermented milk products are abundant in sub-Sahara Africa and have been widely reported (Mutukumira *et al.*, 1996; Gadaga *et al.*, 1999).

In Eswatini, fermented foods have been produced since time immemorial and traditionally fermented milk is known as *emasi* (Masarirambi *et al.*, 2009). Due to the location of Eswatini (31°S), the country enjoys a sub-tropical climate (Thompson, 2003), which is favourable for the production of traditionally fermented products like *emasi* (Masarirambi *et al.*, 2009). *Emasi* is a fermented dairy milk product obtained from fermentation with mesophilic lactic-acid-producing microorganisms. It may be drunk as a refreshing nutritional drink or used as a relish with the staple food (lipalishi and

liphuthu) in Eswatini. Traditionally, *emasi* is prepared by adding fresh raw milk in the earthenware pots or any other suitable containers to ferment spontaneously at ambient temperatures (Gran *et al.*, 2003).

The increase in human population size and development of modern technology has led to isolation of specific lactic acid bacteria from the traditional product for use in production of commercial *emasi* from pasteurized milk under controlled processing conditions. Commercial production of the product has led to products of better nutritional, physical, chemical and sanitary qualities compared to traditional production (Nsibande and Dlamini, 2000). According to Mutukumira *et al.* (2008), the main difference between the two types of fermented milk could be associated with the culture responsible for milk fermentation. However continued improvement of the quality of commercial *emasi* remains sought after.

Although the commercial sector has made efforts to mimic the production of traditional *emasi* in Eswatini, studies done on similar products have shown that indigenous fermented milk is more superior compared to the commercial product (Feresu and Muzondo, 1989). Previous studies have shown that traditionally fermented milk may have low hygienic quality (Ashmaig *et al.*, 2009). Attempts should be made to produce *emasi* that resembles the traditionally produced product with improved hygienic qualities.

Consumption of dairy products provides a wide range of benefits to consumers at low cost when compared with the ever increasing meat prices. As people are concerned with the taste of *emasi*, they always seek for the best product with sensory attributes and rheological properties close to the traditionally produced product. Research into the technology of fermented products is important for the production of consistently safe and high quality products, for both the urban and rural communities (Masarirambi et al., 2009). The aim of the study was to determine the effect of bacterial culture, fermentation time and inoculum size on sensory attributes and physiochemical properties of emasi. The results of the research may be useful to improve income of small entrepreneurs and also may improve the organoleptic quality of commercially produced emasi in the Kingdom of Eswatini.

METHODOLOGY

Site description

The research was conducted at UNESWA Luyengo Dairy Laboratory. The UNESWA, Luyengo campus is located in the upper Middle Veld of Eswatini, at latitude 26° 32' South and longitude 31° 14' East, and at an altitude of 738 m. The study was conducted from the November 2014 to March 2015.

Product preparation

The fresh milk was pasteurised at 85° C for 15 minutes before inoculating with culture. The culture was inoculated at 29° C- 32° C and left to ferment at room temperature. After fermentation the *emasi* was cooled at 4° C for 24 hours before carrying out the tests.

Preparation of lactic acid culture Commercial lactic acid culture

The raw milk was sterilized at 121°C for 15 minutes and cooled to 30°C before inoculating. Two granules of the freeze-dried mesophilic aromatic lactic acid culture (CHN-22, CHR-Hansen, South Africa) were added into a 250mL of sterilized milk. The milk was incubated at 30°C for 24 hours and refrigerated at 4°C before using it as a starter culture for research.

Mixed bacterial culture from emasi sorghum meal

The raw milk was sterilized in an autoclave at 121° C for 15 minutes and cooled to 30° C before inoculating. Fifteen mL of traditional sorghum *emasi* was inoculated into 250 mL of sterilised milk. The milk was incubated at 30° C for 24 hours and refrigerated at 4° C before using it as a starter culture for research.

Mixed bacterial culture from traditional emasi

Fifteen mL of traditional *emasi* was inoculated into 250 mL of sterilised milk. The milk was incubated at 30° C for 24 hours and refrigerated at 4° C before using it as a starter culture for research.

Isolated Leuconostoc ssp. and Lactococcus ssp. culture

The pure strains of *Leuconostoc mesenteroides* ssp. *mesenteroides/ dextranicum* and *Lactococcus lactis* ssp. *lactis* were isolated at the department of Environmental Health Sciences, Faculty of Health Sciences, Mbabane Campus, University of Eswatini. Pure strains of *Leuconostoc mesenteroides* ssp. *mesenteroides/ dextranicum* and *Lactococcus lactis* ssp. *lactis* were obtained separately by growing in MRS broth. The purity of the lactic acid culture was determined by inoculating a loop of each strain on to the MRS agar (Merck Laboratories, Darmstadt, Germany). They were incubated for 48 hrs at 30°C.

Lactic bacteria broth (LBB) was prepared by adding 1% tryptone, 0.5% yeast extract, 0.2% lactose and 1.0% sodium chloride (NaCl) into 200 mL distilled water. The broth was sterilised at 121°C for 15 minutes and cooled to 30°C before inoculating. A colony of the culture strain growing in the petri dishes were inoculated into the broth (30°C) growing separately. They were incubated for 48 hrs at 30°C

Sterilized skimmed milk was inoculated with 1ml of lactic bacteria broth incubated with pure strains separately. The pure strains were separately grown in skimmed milk for 24 hrs at 30°C and cooled in the refrigerator at 4°C.

Sterilized milk was finally inoculated with a mixed ratio of 50: 50 of the isolated lactic acid bacteria grown in skimmed

milk. The inoculated milk was incubated for 24hrs at 30°C and cooled at 4°C before using as a starter culture. Physical analysis (rheological testing)

Viscosity (cP)

The viscosity was determined using Brookfield DV-11+ Viscometer (model No. M/03-163-A0404 Middleborough, Massachusetts. U.SA). Spindle no. 4 was used at speed of 10rmp and the reading on the viscometer was taken after 1 minute for each sample (Moyane and Jideani, 2013).

Synerisis

The synerisis was determined using a Harmonic Series Centrifuge (model no. PLC-O12). Measured 40g of the sample was put in a centrifuge tube and set the centrifuge at 3500rmp for 10 minutes and took the readings by decanting the supertant after centrifuging in a measuring cylinder and the percentage calculated.

Physiochemical analysis

pН

The pH of the samples was determined using pH meter, (EUTECH instruments pH 700, pH/mV/OC/OF meter, model no. ECPH 700425, Auckland, New Zealand). The pH and temperature probe were immersed in the sample. The instrument was calibrated using standard buffer solutions at pH 4 and 7 before testing the samples.

Acidity (titration for lactic acid concentration)

Acidity of the samples was determined by measuring 10mL of emasi sample in a beaker, adding 3-5 drops of phenolphthalein indicator into the sample and titrating using 0.1N of sodium hydroxide (NaOH) until the sample changed to a light pink colour. The volume of the NaOH used to titrate each sample was recorded. The percentage of lactic acid was calculated using the equation (NZIFST, 2010):

Acid (%) =

Weight of sample

Sensory evaluation

The sensory evaluation analysis was conducted using the acceptance test at UNESWA Luyengo campus. Assessors were randomly selected so as to take part in the study. Respondents were informed of the study before answering questions. To evaluate sensory parameters a structured questionnaire was used to capture information and people's reaction towards the different products based on a 7-point hedonic scale. The attributes evaluated include taste, appearance, texture, aroma and overall acceptability. The rating scores used were as follows:

Like very much=7, Like moderately=6, Slightly like=5, Neither like nor dislike=4, Slightly dislike=3, Moderately dislike=2 and Very much dislike=1.

Statistical analysis

The data collected from the study was subjected to three-way analysis of variance (ANOVA) using statistical software Statistix© (Statistix, 2006). Where means were significantly (P < 0.05) different, least significant difference (LSD) was used to separate means at 95% confidence level.

RESULTS

Physiochemical properties of emasi

Effect of bacterial culture, fermentation time and inoculum size on physiochemical properties of emasi Titratable acidity (%)

The effect of commercial lactic culture, isolated Leuconostoc ssp. and Lactococcus ssp., mixed culture from traditional emasi and mixed culture from emasi sorghum meal on titratable acidity of *emasi* were significantly (P < 0.05) different. Mixed culture from emasi sorghum meal had the highest titratable acid content of 1.22% whilst the commercial lactic culture had the lowest content of 0.96% (Table 1). The effect of fermentation time at 12, 18 and 24 hours on titratable acid were all significantly different (P<0.05). At 24 hours the highest titratable acid percentage of 1.18% was recorded whilst at 18 hours it was 1.10%. The lowest titratable acid was recorded at 12 hours 1.01% (Table 1). The effect of inoculum size on titratable acidity was not significantly (P<0.05) different at 2.5% and 5% (Table 1). However, at 10% inoculum size the mean was significantly (P<0.05) different from the others.

pН

The effect of commercial lactic culture, isolated Leuconostoc ssp. and Lactococcus ssp., mixed culture from traditional emasi and mixed culture from emasi sorghum meal on pH of emasi were significantly (P<0.05) different among the treatments. The commercial lactic culture had the highest pH of 4.26 whilst the isolated Leuconostoc ssp. and Lactococcus ssp. culture (4.18) and mixed culture from traditional emasi (ml of NaOH used) (concNaOH) (0.090 milli equivalent weight of lactic acid) (1999) had lower readings (Table 1). The mixed culture from emasi sorghum meal recorded the lowest pH reading of 3.87. The effect of fermentation time on pH was not significantly (P>0.05) different at 18 hours (4.07) and 24 hours (3.99) (Table 1). However at 12 hours (4.15) there was a significant difference (P<0.05) with 24 hours (3.99). There was no significant (P>0.05) difference in the effect of inoculum size on pH of emasi (Table 1).

Viscosity

The effect of commercial lactic culture, isolated Leuconostoc ssp. and Lactococcus ssp., mixed culture from traditional emasi and mixed culture from emasi sorghum meal on viscosity of *emasi* were significantly (P<0.05) different. Mixed culture from emasi sorghum meal had the lowest viscosity of 2222 cP whilst the mixed culture from traditional emasi (2798 cP) and the isolated Leuconostoc ssp. and

Lactococcus ssp culture (3144 cP) had higher readings (Table 1). The commercial lactic acid culture produced the highest viscosity reading of 3551 cP. The effect of fermentation time on viscosity of *emasi* was significantly (P<0.05) different at 12 hours (3199cP) and 24 hours (2681cP). There was no significant (P>0.05) difference at 24 hours and 18 hours (Table 1). There was no significant (P>0.05) difference in the effect of inoculum size on viscosity of *emasi* (Table 1).

Synerisis

The effect of commercial lactic culture, isolated *Leuconostoc* ssp. and *Lactococcus* ssp., mixed culture from traditional *emasi* and mixed culture from *emasi* sorghum meal on synerisis of *emasi* were significantly different (P<0.05). The mixed culture from *emasi* sorghum meal had the highest synerisis percentage of 56.5% whilst the mixed culture from traditional *emasi* (55.5%) and commercial lactic acid culture (50.7%) had lower percentages (Table 1). The isolated *Leuconostoc* ssp. and *Lactococcus* ssp had the lowest synerisis percentage of 48.6%.

Table1: Effect of bacterial culture, fermentation time and inoculum size on physiochemical properties of *emasi*

Sensory attributes of emasi

Effect of culture type on sensory attributes of emasi

There was no significant (P>0.05) difference in the effect of bacterial culture on appearance of *emasi* (Figure 1). There was however a significant (P<0.05) difference in the effect of bacterial culture on aroma of *emasi*. The mixed culture from traditional *emasi* (MCTE) had the lowest degree of liking of 4.49 for aroma, whilst the isolated bacterial culture (ILLC) had the highest score (5.34) of them all.

There was a significant (P<0.05) difference in the effect of bacterial culture on taste of *emasi*. The isolated *Leuconostoc* ssp. and *Lactococcus* ssp. Culture (ILLC) scored the highest degree of liking (5.36) whilst the mixed culture from traditional *emasi* (MCTE) scored the lowest (4.38) score as the panellist "slightly disliked" the taste (Figure 1).

The effect of bacterial culture on texture of *emasi* had a significant (P<0.05) difference, higher scores were recorded for commercial lactic culture (COMM) (5.14) and isolated *Leuconostoc* ssp. and *Lactococcus* ssp. The panellists "liked" Culture (ILLC) (5.39) for texture as compared to mixed culture from traditional *emasi* (MCTE) (4.54) and sorghum meal (MCES) (4.60) which the panellists "neither liked or dislike".

	Bacterial Culture				Fermentation time (hrs)			Inoculum size (%)		
Physiochemical qualities	СОММ	ILLC	MCTE	MCES	12	18	24	2.5	5	10
Titratable acidity (%)	0.96 ^a	1.04 ^b	1.17 ^c	1.22 ^d	1.01 ^a	1.10 ^b	1.18 ^c	1.06 ^a	1.08 ^a	1.15 ^b
pH	4.26 ^a	4.18 ^b	3.99°	3.87 ^d	4.15 ^a	4.07 ^{ab}	3.99 ^b	4.12 ^a	4.08 ^a	4.02 ^a
Viscosity (cP)	3551 ^a	3144 ^b	2978°	2222 ^d	3199 ^a	2905 ^{ab}	2681 ^b	2994 ^a	2874 ^a	2917 ^a
Synerisis (%)	50.7 ^a	48.6 ^b	55.5°	56.5 ^d	50.3ª	52.8 ^b	54.0 ^b	51.5 ^a	51.6 ^a	53.9 ^b

Means with different letter in each constituent are significantly (P<0.05) different.

Key: **COMM** is commercial lactic acid culture, **ILLC** is isolated *Leuconostoc ssp.* and *Lactococcus ssp.* culture, **MCTE** is mixed culture from traditional *emasi* and **MCES** is mixed culture from *emasi* sorghum meal.

The effect of fermentation time on synerisis was not significantly (P>0.05) different at 18 hours (52.8%) and 24 hours (54.0%) (Table 1). However, at 12 hours (50.3%) there was a significant (P < 0.05) difference from 18 and 24 hours as the lowest synerisis percentage was recorded of 50.3%.

There was no significant (P>0.05) difference in the effect of inoculum size on synerisis of *emasi* at 2.5% and 5% (Table 1). However, there was significant (P<0.05) difference at 10% with the highest synerisis of 53.9%.

In terms of overall acceptability of *emasi*, there was a significant difference (P<0.05) with the commercial lactic culture (5.43) and isolated *Leuconostoc* ssp. and *Lactococcus* ssp. culture (5.58) scoring higher as the panellists "liked" better than the mixed culture from traditional *emasi* (4.90) and sorghum meal (5.02).

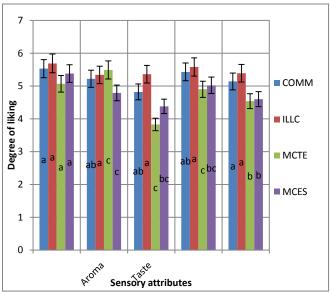


Figure 1. Effect of bacterial culture type on sensory attributes of *emasi*.

Note. Error bars are based on standard error of estimate. Means with different letter in each constituent are significantly (P<0.05) different .

Key: **COMM** is commercial lactic acid culture, **ILLC** is isolated *Leuconostoc* ssp. and *Lactococcus* ssp. culture, **MCTE** is mixed culture from traditional *emasi* and **MCES** is mixed culture from *emasi* sorghum meal

Effect of fermentation time on sensory attributes of *emasi* There were no significant (P>0.05) differences in the effect of fermentation time on appearance, aroma, taste and overall acceptability of *emasi*. However, there was a significant (P>0.05) difference in the effect of fermentation time on texture of *emasi*. Higher degree of liking scores were recorded at 18 hours (5.05) and 24 hours (5.00) compared to 12 hours (4.17) with the lowest scores.

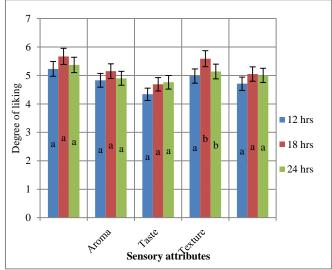


Figure 2: Effect of fermentation time on sensory attributes of *emasi*.

Note. Error bars are based on standard error of estimate. Means with different letter in each constituent are significantly (P<0.05) different.

Effect of inoculum size on sensory attributes of emasi

There was no significant (P>0.05) difference in the effect of inoculum size on appearance, taste, texture and overall acceptability of *emasi*. However, there was a significant (P<0.05) difference in the effect of inoculum size on aroma of *emasi*. Higher degree of liking scores were recorded at 2.5% (5.18) and 5% (5.17) compared to 10% (4.53) with the lowest scores.

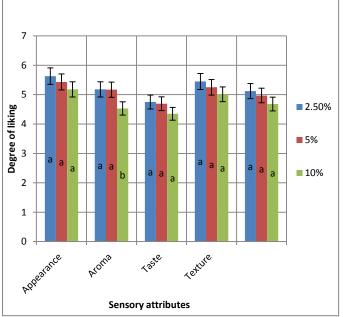


Figure 3: Effect of inoculum size on sensory attributes of *emasi.*

Note. Error bars are based on standard error of estimate. Means with different letter in each constituent are significantly different (P<0.05).

DISCUSSION

The effect of culture on sensory attributes and physiochemical properties of *emasi*.

The effect of culture on physiochemical properties of *emasi* such as titratable acidity, viscosity, synerisis and pH showed significant differences between the bacterial culture species. The commercial lactic culture had the lowest titratable acid of 0.96% and the highest pH reading of 4.26. The pH value of the commercial culture was within the range of 4.22 and 4.30 recorded by Moyane and Jideani (2013), but the titratable value was a bit higher compared to what they recorded. The organic acid (predominantly lactic acid) causes the pH of sour milk to drop (Gadaga *et al.*, 2004).

The lactic acid bacteria (LAB) that produce exopolysaccharides are often used to increase the viscosity of

stirred fermented milks, such as yoghurt and to decrease the susceptibility to synerisis (Dlamini et al., 2009). The commercial lactic culture had the highest viscosity reading of 3551 cP compared to other culture species and a low synerisis percentage of 50.7%. The value of viscosity of the culture is higher than that reported by Moyane and Jideani (2013), of 3330 to 1240 cP of commercial emasi products. Viscosity is influenced by the size of the particles and whey proteins from denaturation caused by pH (Masson et al., 2010). The synerisis of the sample was very high compared to that of 24.7% to 25% recorded by Nsibande and Dlamini (2000). Synerisis can be minimized by using stabilizers to the fermentation media or by increasing the total solids using milk powder (Schellhass and Moris, 1985). The sample was prepared without a stabilizer hence that is why the synerisis was high. Synerisis is also increased if the milk is not homogenised prior to fermentation (Schellhass and Moris, 1985). The sample was not homogenized hence the high synerisis percentage.

The isolated Leuconostoc mesenteroides ssp. mesenteroides/ dextranicum and Lactococcus lactis ssp. lactis had higher titratable acid of 1.04% (v/v) as compared to the commercial lactic culture and a lower pH reading of 4.18. The isolated Leuconostoc ssp. and Lactococcus ssp. culture had the lowest synerisis percentage compared to other cultures (commercial lactic culture, mixed culture from traditional emasi and emasi sorghum meal). This can be due to their ability to produce exopolysaccharides thus increasing viscosity and reducing synerisis (Dlamini et al., 2009). Synerisis or whey separation is not desirable in cultured dairy products because it indicates an inferior product (Nsibande and Dlamini, 2000). Isolated yoghurt starter can be used in yoghurt manufacturing in an industrial large scale (Bashiti, 2010). The isolated Leuconostoc ssp. and Lactococcus ssp. culture was as good as the commercial lactic culture. The use of pure cultures enables different acid flavours to be developed leading to a range of fermented (cultured) products (Kurmann et al., 1992).

The mixed culture produced *emasi* had a titratible acid of 1.17% and a very low pH value of 3.99 as compared to the emasi produced using the commercial culture and the isolated bacterial strains culture. The low pH in fermented milk offers a selective environment for yeast growth, but is unfavourable for some bacteria (Fleet, 1990) and spoilage may become evident when the yeast population reaches 10^5 to 10^6 cells/g (Fleet, 1990). Synerisis is high when acidity is high (Nsibande and Dlamini, 2000). A low viscosity reading of 2796 cP was recorded in this emasi and this though, was within the range of Moyane and Jideani (2013) but lower than that of the emasi from the isolated *Leuconostoc* ssp/*Lactococcus* ssp. culture and commercial lactic culture. However, the synerisis was higher as compared to that of the commercial and isolated bacterial strains culture.

The mixed culture from *emasi* sorghum meal had the highest titratable acidity of 1.22% and also the lowest pH value of 3.87. According to Sanet-Bali *et al.* (2012), traditional fermented milks are considered safe because of the low pH and the production of antimicrobial substances by fermenting organisms. The synerisis percentage of the mixed culture from sorghum meal was the highest of them all which was 56.5% and the viscosity was also low at 2222cP. High synerisis results in low viscosity due to the high whey content of the product. This could be attributed to the fact that the culture is mixed and undefined.

Sensory evaluation

The sensory scores indicated that the consumers "liked" the appearance of all bacterial culture species presented as *emasi*.. In terms of aroma the panellists neither liked nor disliked the mixed culture from traditional *emasi* and sorghum meal *emasi*, however the commercial lactic culture and the isolated bacterial strains were ranked higher with panellists "liking" the culture. The aroma, in general, of dairy products is complex due to the heterogeneous nature of milk (Moyane and Jideani, 2013).

In terms of taste of the culture, the isolated bacterial strains and commercial lactic culture species were both ranged higher showing that the consumers "liked" them whilst the two mixed culture from traditional *emasi* and sorghum meal where ranked lower as the panellists "slightly disliked" the culture species. The two mixed culture from traditional emasi and sorghum meal's taste scores might have been influenced by the high acidic content of the samples.

In a study done by Moyane and Jideani, (2013), samples which had low viscosity also recorded lowest consumer acceptance on texture and smoothness sensory attributes. The texture of isolated Leuconostoc ssp. and Lactococcus ssp. culture had the highest scores where the panellists "liked" the product; this can be due to the high viscosity reading and very low synerisis percentage. The commercial lactic culture also had high sensory scores. The two mixed culture from traditional emasi and sorghum meal had low texture scores from the panellists, this can be due to the high synerisis percentages and low viscosity readings. The overall acceptability of both the commercial lactic culture and isolated bacterial strains had high scores as the panellists "liked" the culture species compared to the two mixed culture from traditional emasi and sorghum meal which had low score as the panellists "slightly disliked" them.

The effect of fermentation time on sensory attributes and physiochemical properties of *emasi*

The effect of fermentation time at 12hrs, 18hrs and 24hrs on titratable acid were all significantly (P<0.05) different. At 12hrs the titratable acid percentage was low at 1.01% and gradually increasing at 18hrs and 24hrs being the highest at 1.18%. The pH values are also noticed to be decreasing as

the fermentation time increase. pH and titratible acid percentage influence synerisis and viscosity. As the pH decrease and T.A increase the synerisis increased but at 18hrs and 24hrs there was no significant (P<0.05) difference. The viscosity decreased due to the increase in synerisis from 12hrs, 18hrs and 24hrs with readings as follows: 3199 cP, 2805 cP and 2681 cP. Eissa *et al.* (2010) stated that prolonged metabolic activity of micro-flora in yoghurt causes changes in the micro-structure of the media and hence affecting viscosity.

Sensory evaluation

The sensory score indicated that fermentation time did not have an effect on the appearance, aroma, taste and overall acceptability of *emasi*. The panellists liked the *emasi* regardless of the fermentation time. However, fermentation time had an effect on the texture of the product. Low scores were given at 12 hrs with panellists slightly disliking it whilst at 18 hrs and 24 hrs the panellists liked the texture.

The effect of inoculum size on sensory attributes and physiochemical properties of *emasi*

The effect of inoculum size (2.5%, 5% and 10% v/v) on bacterial culture species had no significant difference on viscosity and pH. However there was a significant difference on T.A and synerisis of *emasi*. The titratable acid at 2.5% and 5% were not significantly (P<0.05) different but at 10% the percentage was high at 1.15%, this affected the synerisis also with 10% inoculum size having the highest synerisis percentage.

The inoculum size had no effect on appearance, taste, texture and overall acceptability as the panellists liked all the inoculum sizes. However, on aroma there was a significant difference with 10% recording low sensory scores as the panellists "slightly disliked". According to Moyane and Jideani, (2013), aroma of dairy products is complex due to the heterogeneous nature of milk. The significant difference in aroma does not affect the overall acceptability of the product.

CONCLUSION

The outcome of the study showed that the type of culture used to ferment milk influenced consumer acceptance of *emasi* compared to inoculum size and fermentation time. The commercial lactic culture and isolated *Leuconostoc* ssp. and *Lactococcus* ssp culture had higher viscosity readings and low synerisis percentages whilst the two mixed culture species from traditional *emasi* and sorghum meal had low viscosity readings and high synerisis percentages. The sensory evaluation indicated that consumers prefer the commercial lactic culture and isolated *Leuconostoc* ssp. and *Lactococcus* ssp culture species as compared to the two mixed culture from traditional *emasi* and sorghum meal. The inoculum size and fermentation time did not have any significant effect on the physiochemical properties and consumer preference of *emasi*. The best culture species from the study were the commercial lactic culture and the isolated *Leuconostoc* ssp. and *Lactococcus* ssp culture. The inoculum size best for fermenting is the 2.5%, there was no significant difference between the levels inoculum size, I would advise the lowest size as it is economic when fermenting *emasi* at commercial level. 18-24 hours would be the best time for fermenting *emasi* as during that time the mesophilic bacteria fermenting the milk have coagulated the media to a desired texture and taste preferred by the consumers.

REFERENCES

- Ashmaig, A., Hasan, A. and Gaali, E.E. (2009). Identification of lactic acid bacteria isolatedfrom traditional Sudanese fermented camel's milk (Gariss). *African Journal of Microbiology Research* 3:451-457
- Bashiti, T.A.I. (2010). Production of yoghurt by locally isolated starters: *Streptococcus thermophilus* and *Lactobacillus bulgaricus*. *Journal of Al Azhar University* 12:56-58
- Dlamini, A.M., Peiris, P.S., Bavor, J.H. and Kasipathy, K. (2009). Rheological characteristics of an exopolysaccharide produced by a strain of *Klebsiella* oxytoca. Journal of Bioscience and Bioengineering 107:272-274
- Dlamini, A. M., Amusan, R., Magongo, B.A. (2015). Survival of indicator organisms (coliforms) and index bacteria (*Escherichia coli*) in Swaziland traditionally fermented emasi. *Global Journal of Biology, Agriculture and Health Sciences* 4:74-78
- Eissa, E. A., Mohamed Ahmed, I.A., Yagoub, A.E.A. and Babiker, E.E. (2010). Physicochemical, microbiological and sensory characteristics of yoghurt produced from goat milk. *Livestock Research for Rural Development* 22:137-139.
- Feresu, S., and Muzondo M. I. (1989). Factors affecting the development of two fermented milk products in Zimbabwe. MIRCEN *Journal of Applied Microbiology and Biotechnology* 5:349-355.
- Fleet, G.H. (1990).Yeasts in dairy products. *Journal of Applied Bacteriology* 68: 11.
- Gadaga, T.H., Mutukumira, A.N., Narvhus, J.A. and Feresu, S.B. (1999). A review oftraditional fermented foods and beverages of Zimbabwe. *International Journal of Food Microbiology* 53:1-11
- Gadaga, T.H., Nyanga, L.K. and Mutukumira, A.N. (2004). The occurrence, growth and control of pathogens in African fermented foods. *African Journal of Food and Agriculture Nutrition Development* 4:20-23.
- Gran, H.M., Gadaga, H.T. and Narvhus, J.A. (2003). Utilization of various starter cultures in the production of *amasi*, a Zimbabwean naturally fermented raw milk product. *International Journal of Food Microbiology* 88:19–28

- Kurmann, J.A., Rasic, J.L. and Kroger, M. (1992). Encyclopedia of fermented fresh milk products. *An international inventory of fermented milk, cream, buttermilk, whey, and related products* Van Nostrand Reinhold. ISBN 0-442- 00869-4 New York, USA.
- Masson, L.M.P., Calado, V.M.A., Deliza, R. and Rosenthal, A. (2010). Rheological behaviour of fermented dairy beverages obtained from the ultra-high pressure homogenization (UHPH), International Conference on Food Innovation. Food Innovation. Dijon, France
- Masarirambi, M.T., Mhazo, N., Dlamini, A.M. and Mutukumira, A.N. (2009). Common indigenous fermented foods and beverages produced in Swaziland: A review. *Journal of Food Science and Technology* 46:505-508
- Moyane, J.N. and Jideani, A.I.O. (2013). The physicochemical and sensory evaluation of commercial sour milk (*amasi*) products. *African Journal of Food Science* 7: 52-62
- Mutukumira, A.N., Dube, D.M.J., Mupunga, E.G. and Feresu, S.B. (1996). Smallholder milk production, milk handling and utilization: A Case Study from the Nharira/ Lancashire Farming Area, Zimbabwe. *Livestock Research Rural Development* 8:40-50
- Mutukumira, A.N., Narvhus, J.A., Marzin A., Feresu, S.B. and Abrahamsen, R.K. (2008). Characteristics of fermented milk produced by *Lactococcus lactis* subsp. *lactis* biovar. *Diacety lactis* C1, *Lb. plantarum* B1 and *Leuc. mesentoroides* subsp. *Mesenteroides* E9 isolated from traditional fermented milk in Zimbabwe. *Milchwissen* 63:438-442

- Nsibande, R.T. and Dlamini, A.M. (2000). The composition and fermentation quality of Swaziland *emasi*. UNISWA Research Journal Agriculture Science Technology 3:48-53
- NZIFST (2010). New Zealand Institute of Food Science and Technology. Food Science Experiments. URL <u>http://www.nzifst.org.nz/myfiles/Expt 5.</u> <u>Sour_Cream_Chemistry guide.doc</u> Accessed 11 April 2015
- Robinson, R. K. and Tamime, A. Y. (1981). Microbiology of fermented milks. *Dairy Microbiology*. Applied Science Publishers. London, U.K. 2:245-278
- Samet-Bali, O., Ennouri, M., Dhouib, A.And Attia, M. (2012). Characterisation of typical Tunisian fermented milk: Leben. *African Journal of Microbiology Research* 6:2169-175
- Schellhaas, S.M. and Moris, H.A. (1985). Rheological and scanning electron microscopic examination of skim milk gels obtained by fermenting with ropy and non-ropy strains of lactic acid bacteria. *Food Microstructure Journal* 4:279-287
- Srujana, G., Reddy, R.A., Reddy, K.V. and Reddy, R.S. (2011). Microbial quality of raw pasteurized milk samples collected from different places of Warangal District, (A.P.) India. *International Journal of Pharmacy and Biological Sciences* 2:139-143
- Statistix (2006). Statistix for Windows. *Analytical Software* Tallahassee, FL, U.S.A. pp333
- Thompson, C.F. (2003). *Swaziland Business Year Book* 2003. Christina Forsyth Thompson, Mbabane, Swaziland