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Evaporation measurement and validation of meteorological models

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Abstract

Identification of the best-fit meteorological model for evaporation measurement in a particular agroecological zone is a pre-requisite for cost-effective land management. A simulation experiment was conducted at Makerere University with the objective of determining the best-fit model for estimating evaporation. Meteorological data was collected from Makerere Meteorological Station and applied to different models to estimate potential evaporation. Actual evaporation from three mini lysimeters was used to validate models. Actual evaporation from the lysimeter varied between 2.6 to 6.73 mm day^{-1}. Results showed that measured actual evaporation was greater in the afternoon than in the mornings and varied significantly ($P<0.05$) over days. After a linear regression analysis of measured actual evaporation against estimates from meteorological models, models were ranked in the order of best fit as Priestley and Taylor, Makkink, Abtew, Hargreaves, Romanenko, Penman, Turc, CROPWAT, and Penman-Monteith.

Key words: Actual evaporation, land management, lysimeter, simulation, Uganda

Introduction

Evaporation is a key element in the water balance (Stroosnijder, 1987). Excessive evaporation contributes significantly to moisture deficit in agricultural production and thus reduces food production. Evaporation measurement in a particular agroecological zone is a pre-requisite for proper land management. Meteorological models provide the fastest and the cheapest method of estimating evaporation because they do not involve soil-plant complexities. Mini lysimeter measurements of evaporation can be used to validate models to be used for field evaporation estimates (Boast and Robertson, 1982; Stroosnijder, 1987). Although the necessary equipment are available, these studies have not been conducted in Uganda. Thus, this study was conducted to avail information for evaporation measurements and validation of meteorological models.

Materials and methods

Experimental site

The experiment was conducted at Makerere University, Kampala. The site is located between latitude 0° 19'E and longitude of 32°34'E and at an elevation of 1200 m a.s.l. Three simple mini lysimeters (Trambouze *et al.*, 1998) each of base 30x30 cm and 50 cm high were constructed. In order to reduce weight, the lysimeter frame was made of plastic sheets reinforced with metallic angle bars on the sides. The base of the lysimeter was perforated to allow free drainage. The top part of the lysimeter was open to air and covered only whenever there was a rainfall incidence. Each lysimeter was filled with 56kg sandy clay loam (sand 54%, clay 22% and silt 24%). The experiment was conducted during the month of July 2001. Data were analysed using two way ANOVA procedures in Genstat statistical package.

Measurements

The soil in lysimeters was slowly brought to saturation (water added until it starts to drain from the bottom) and allowed to drain for 24 hours to field capacity (Klute, 1986). During the experiment, lysimeters were exposed to sunshine and weights taken at 8.00 am, 1.00 pm and 6.00 pm using a weighing balance of 50 g sensitivity. The soil moisture content in the lysimeter was restored to field capacity every evening by adding the amount lost through evaporation. The daily actual evaporation rate was determined by dividing volume water (cm) lost through evaporation and lysimeter surface area. Atmospheric physical parameters (maximum and minimum temperature, wind speed, relative humidity and sunshine hours) for the month of July 2001 were collected from Makerere meteorological station. At Makerere meteorological station, wind speed is measured using a cup-counter anemometer exposed at a standard height of 2 m. Air temperature and humidity observation were measured at height of 2 m using maximum and minimum thermometers and dry – bulb and wet- bulb thermometers, respectively. The campbell-stokes tropical sunshine recorder was used to record sunshine hours. This instrument is exposed in sectors N.E. to S.E. and S.W to N.W. and the main base mounted at 1.22 m. Nine meteorological methods (Abtew, Hargreaves, Makkink, Priestley and Taylor, Turc, Romanenko, Penman, Penman-monteith and Penman-Monteith FAO CROPWAT software of version 5.7, Oct. 1991) were tested.

Best fit model selection criteria and model ranking

Nine meteorological models were tested in this study with the objective of establishing the best-fit model. The models were; Abtew, Hargreaves, Makkink, Priestley and Taylor, Turc, Romanenko and Penman (Xu and Singh, 2000), Penman-Monteith and CROPWAT. These models are elaborately described by Makkink (1957) and FAO (1998).

The best model was selected on the basis of unbiased and high precision. The bias was assessed by examining the value of the slope (a) and intercept (b), of the regression between observed and estimated values. Precision was assessed using standard error (S.E) and determination of correlation coefficient, r for each model. For the ideal model, a= 1, b =0 S.E = 0 and r = 1 (Majaliwa, 1998). The models were ranked after a comparison of mean values of the above four parameters for each model in terms of closeness to ideal values. The best model was given a ranking value of 1.

Results and discussion

The results indicated that evaporation significantly ($P<0.05$) varied with time of the day, and over the days. Evaporation in the afternoons (1.00 p.m-6.00 p.m.) was generally higher than that in the mornings (8.00 a.m-1.00 p.m.). This could be explained by the diurnal variation of temperature since there is a strong agreement between daily air temperature and evaporation (Xu and Singh, 1998). At mid-day (12.00 pm), the sun is at its maximum elevation and the maximum amount of solar radiation is being received. Due to the atmosphere being heated from below by the slow process of conduction and convection, a delay of between two or three hours after mid-day generally occurs before the maximum air temperature and maximum evaporation are reached. The maximum temperature therefore occurs in the afternoons (Mwebesa, 1970) and hence maximum evaporation. For two groups of the pairwise (1, 4, 5 and 2, 3, 4), a significant difference ($P<0.05$) was observed for daily evaporation (Fig. 1).

The models were significantly different ($P<0.05$) with respect to the slope (Table 1). The best-fit models are; Priestley and Taylor, Makkink, Hargreaves, Abtew, Romanenko, Penman, Turc, CROPWAT, and Penman- Monteith respectively. The models can also be classified in two groups of

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Effect of selected preservation methods on the shelf life and sensory quality of 'Obushera'

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Abstract

'Obushera' is a predominantly lactic acid bacteria fermented gruel produced from sorghum and or millet flour, whose shelf life is limited to 1 - 2 days. A study aimed at exploring possibilities of extending the shelf life of 'obushera' with least effect on its quality characteristics was conducted. Sorghum grains were germinated, dried and ground into flour, which was used to produce porridge. This porridge was then fermented, into 'obushera', for 48 h with previously made 'obushera' as a starter. The 'obushera' was divided into four portions each of which was subjected to one of the following treatments: pasteurisation, refrigeration, pasteurisation combined with refrigeration and a control. The treated 'obushera' was analysed for pH, alcohol content, total acidity, lactic acid bacteria counts and yeast counts at two day intervals during storage. The acceptability of the 'obushera' subjected to the different treatments was evaluated using a 25 member panel. Pasteurisation stopped the fermentation process by reducing the number of lactic acid bacteria and yeasts to almost undetectable levels. It also increased the shelf life of 'obushera' from 2 days to over one month. Refrigeration only slowed down the rate of fermentation and subsequent deterioration of the 'obushera'. The refrigerated porridge became unacceptable by the eighth day. There was no significant difference in quality of 'obushera' treated with pasteurisation alone and that subjected to pasteurisation combined with refrigeration. The sensory evaluation results showed that the pasteurised 'obushera' was acceptable. The results show that it is possible to increase the shelf life of 'obushera' beyond the traditional two days with little adverse effects on its sensory properties using pasteurisation at 78°C for 10 minutes.

Key words: Cereal beverage, fermented, sensory evaluation, Uganda

Introduction

Lactic acid fermentation of foods is widely practised in Uganda as a household-level technology to process and preserve a number of products. One such products is 'obushera'. 'Obushera' is a sweet and sour non-alcoholic beverage popular among the people of south, west and central Uganda. It is consumed as a refreshing drink and a weaning food. 'Obushera' is prepared by mixing pre-germinated sorghum or millet flour in warm water to make a slurry of 8 to 10% solids (w/w) to which previous 'obushera' or pre-germinated flour is added to initiate the lactic fermentation. The traditional processing and properties of 'obushera' have been reviewed by Muyanja (2001). Today, there are a number of small enterprises that engage in the production and sale of 'obushera'. With increasing urbanisation and industrialisation, it has become necessary to improve the production process and increase the shelf life of 'obushera'.

Most of the work done on 'obushera' and related fermented gruels has concentrated on the microbiology and nutritional aspects of the products (Muyanja, 2001; Lorri, 1993). Halm *et al.*, (1993), Lorri and Svanberg (1993) and Mbugua (1984) reported that these products are predominantly lactic acid fermented although yeasts are involved as well. Earlier findings indicate that lactic acid fermented cereals have improved safety due to the production lactic acid and other antimicrobial agents

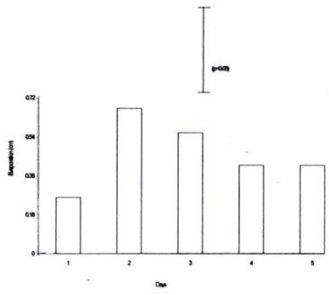

Figure 1. Mean evaporation for five days.

Table 1. A summary of means for slope (a), intercept (b), Standard Error (S.E), and correlation coefficient (r) for regression of 5 days of evaporation from 3 mini lysimeters with potential evaporation from meteorological models with.

Model	Intercept (b)	Standard error (s.e)	Correlation coefficient (r)	Slope (a)	1-a	Best fit model
Makkink	0	0.353	0.563	1	0	1
Priestley & Taylar	0.01	0.193	0.663	1	0	1
Hargreaves	-0.02	0.197	0.567	1.02	0.02	3
Abtew	0.05	0.197	0.563	0.95	0.05	4
Romanenko	0.17	0.277	0.283	0.82	0.18	5
Penman	0.16	0.35	-0.073	0.82	0.18	5
Turc	-0.04	0.227	0.443	0.74	0.26	7
CROPWAT	0.02	0.227	0.453	2.78	1.78	8
Penman-Monteith	-0.04	0.207	0.547	3.2	2.2	9
LSD(0.05)	ns	ns	ns	1.67	ns	

ns = Nonsignificant

no pairwise significant difference as; group one (Makkink, Priestley and Taylar, Hargreaves, Abtew, Romanenko, Penman, and Turc), and group two (CROPWAT, and Penman- Monteith).

Conclusion

Evaporation in the afternoon was greater than in morning. Nine models were evaluated in this study using meteorological data from Makerere university weather station. Priestley and Taylor and Makkink models have been found to be the best models.

Acknowledgement

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(Byaruhanga *et al.*, 1999; Kingamukono *et al.*,1994; Olsen *et al.*, 1995; Svanberg *et al.*,1992). Some other bacterial species e.g., *Bacillus* species and coliforms have been implicated in the spoilage of fermented cereal gruels (Lorri, 1993; Holzapfel, 1984). Little work, however, has been done on the product shelf-life and manufacturing practices yet these are important to the small enterprises that deal in these products. A major problem facing the producers and consumers of 'obushera' is its short shelf life. Traditionally, a batch of 'obushera' product has to be consumed within 1-2 days of its production. This is because beyond this period, the fermentation process continues into the alcoholic stage and the product becomes too sour, which is undesirable. To extend the shelf life of 'obushera', the processing method (s) should stop the fermentation and also preserve the product without adversely affecting its quality.

Pasteurisation and chemical preservation have been used to preserve food singly or in combination. Efiuvwevwere and Akoma (1997) used pasteurisation and chemical preservation to extend the shelf-life of kunun-zaki, a Nigerian fermented cereal gruel. Gimbi *et al.* (1997) used pasteurisation alone to preserve a fermented millet-based weaning gruel. Chemical preservation alone however, did not yield good results mainly because it does not stop the fermentation process. Producers of 'obushera' in urban areas in Uganda use refrigerators for storage, however, this does not stop the fermentation process. Low temperatures just slow down the fermentation process. Thus, with time the product becomes too sour and alcoholic. Very little information exists in literature on the improved production and preservation of 'obushera'. This work was undertaken to provide recommendations with regard to extension of the shelf life of 'obushera' using pasteurisation while optimising the product quality.

Materials and methods

Samples and preparation

The production of 'obushera' used in this study was based on the traditional process. Red sorghum grains were purchased from a market in Kampala. The grain was screened to remove dirt and chuff. The sorghum grain was then kept on a damp cotton cloth covered with banana leaves for three days. This allowed germination of the seeds. The germinated grain was sun dried on stainless steel trays. The dry germinated grain was then gently pounded in mortar, with a pestle, to separate the grain from the germinated roots. The pounded grain was winnowed to retain clean grain. The clean grain was then milled into a flour using a hammer mill.

Preparation of 'obushera' base

Sorghum flour was mixed with hot water in the ratio of 1:4 (flour:water) in the following order: the flour was first mixed with a small amount of cooled water to form a slurry to which the rest of boiling water was added to make 12 l of the flour-water mixture. The mixture was left to stand for about 5 minutes, this was then referred to as 'obushera' base. Two batches of the 'obushera' base were prepared.

Preparation of starter culture

To start the fermentation, pre-germinated sorghum flour was added to each of the two batches of 'obushera' base at a rate of 5% (w/w) of the sorghum flour. The mixture was thoroughly stirred and fermented at ambient temperature (25 - 28°C) for 48 h to yield duplicate batches of 'obushera' culture. The preliminary 'obushera' cultures were recycled in 'obushera' base at an inoculation rate of 5% (v/v) followed by incubation at ambient temperature for 48 h. This procedure was repeated four times leading to duplicate batches of obushera culture hereafter referred to as starter culture.

Preparation of obushera

The 12 l of 'obushera' base were left to cool to a temperature below 35°C. The cool 'obushera' base was then inoculated with 5% (v/v) starter culture and left to ferment for 48 h at ambient temperature (25 - 28°C) to yield 'obushera'. The resultant 'obushera' was subjected to the treatments described below.

Treatments

All batches of 'obushera' were packed and sealed in clean and sterile 250 ml glass bottles. The first was kept on the shelf at ambient temperatures (25 – 28° C). This served as the control. The second batch was kept in a refrigerator (7°C). The third batch was pasteurised by the following procedure. The filled bottles were put in a hot water gyrating bath. The internal temperature of 'obushera' was monitored with a mercury thermometer. When the internal temperature of 'obushera' in the bottles reached 78°C, the bottles were sealed and held at that temperature for 10 min. This was to allow for pasteurisation of this batch of 'obushera'. The 'obushera' was cooled, immediately after pasteurisation, by putting the jars in a gyrating cold water bath. This batch was then kept on a shelf at ambient temperature (25 - 28°C). The pasteurisation at 78°C for 10 min was established in preliminary work (results not shown) which was aimed at optimising pasteurisation conditions. The fourth batch was pasteurised as described above and then kept in a refrigerator at a temperature of 7°C.

Drawing of samples for analysis

At intervals of two days, 2 bottles were taken from each of the treatments for chemical and microbiological analyses.

Chemical analysis

Determination of pH and titratable acid

The pH of 'obushera' was determined using a glass electrode connected to a standard PW9420 pH meter (Philips). Titratable acidity was determined by titrating 10 ml of sample diluted with 10 ml of distilled water against 0.1M sodium hydroxide solution and phenolphthalein as indicator. The titratable acidity was calculated as percent lactic acid (w/w) (Ayebo and Mutasa, 1987).

Determination of alcohol content

The per cent alcohol content (m/m) of 'obushera' was determined using the method described by Kirk and Sawyer (1991) in which results obtained in specific gravity were converted to percent alcohol.

Microbial analysis

The viable colonies of yeasts were enumerated by spread plating 0.1 ml of serial dilutions of the samples onto sterile Potato Dextrose Agar (Oxoid CM139) and incubating at 25°C for 5 days. The number of colony forming units (c. f. u.) per ml of sample were counted and calculated after incubation (Anon., 1987).

The viable colonies of lactic acid bacteria were enumerated by using the pour plate method (Harrigan and McCance, 1976) using 1 ml of serial dilutions of sample on MRS Agar (Oxoid CM361) and incubated at 28°C for 48 h. Typical colonies of lactic acid bacteria were counted. The number of c.f.u. per ml of sample were calculated after incubation (Anon., 1987).

Sensory evaluation

A panel of 25 untrained persons was used to evaluate acceptability of 'obushera' subjected to different treatments using a nine point hedonic scale where scores 1 = Like extremely and 9 = Dislike extremely (Amerine *et al.*, 1965). This was done every two days during the storage period. On the 14th day fresh control and refrigerated 'obushera' were prepared. The porridge was presented to the panellists in cups coded with 3 digit random numbers.

Shelf-life

After monitoring the changes in the stored 'obushera' for 14 days, the shelf-life of the pasteurised 'obushera' was estimated using the equation for the slope of the titratable acid graph $y = mx + c$ where y is the % titratable acid at a given time, m is the slope of the graph, x is the time in days and c is the intercept at the y axis. Since a titratable acid content of 0.95%, often attained in spontaneous fermentations after 96 h, obushera becomes too sour and undesirable, the time taken for the pasteurised 'obushera' to attain 0.95% titratable acid was taken to be the estimated shelf-life of the product.

Data analysis

The microbial counts were converted to Log_{10} c. f. u. /ml. This, together with the chemical and sensory data were subjected to ANOVA using Statistical Package for Social Science (SPSS). Correlations, standard deviations and the least significance difference between means were determined at a probability level of 5%.

Results

There was a general increase in the number of lactic acid bacteria and yeasts with subsequent increase in the amount of titratable acid and alcohol in the 'obushera' during storage (Figs. 1, 2, 3 and 5). Figures 1 and 2 show a reduction in the number of yeasts and lactic acid bacteria upon pasteurisation as compared to the control. There was a concomitant decrease in pH with the increased titratable acid in 'obushera' during storage (Figs. 3 and 4). Alcohol production also increased with the control and refrigerated 'obushera' showing the highest increase (Fig. 5). Acid production and lactic acid bacteria

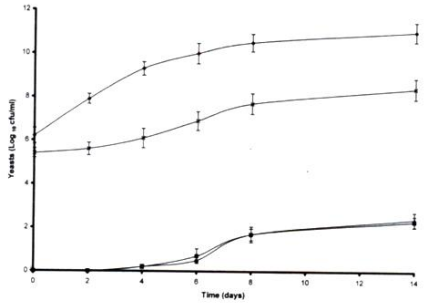

Figure 1. Changes in yeast population of pasteurised (■), pasteurised and refrigerated (▲), refrigerated (X) and the control (◆) 'obushera' during storage. (Error bars are standard deviation).

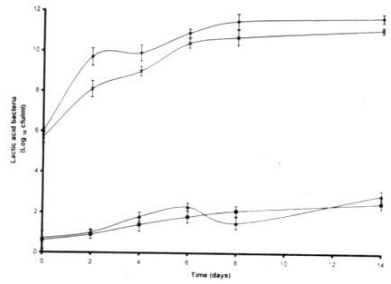

Figure 2. Changes in lactic acid bacteria population of pasteurised (■), pasteurised and refrigerated (▲), refrigerated (X) and the control (◆) 'obushera' during storage. (Error bars are standard deviation).

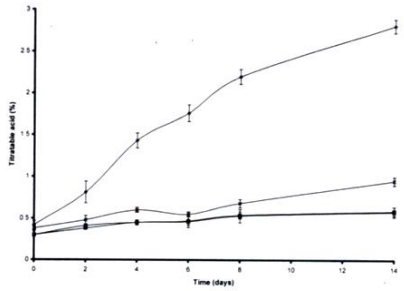

Figure 3. Changes in the % titratable acid of pasteurised (■), pasteurised and refrigerated (▲), refrigerated (X) and the control (◆) 'obushera' during storage. (Error bars are standard deviation).

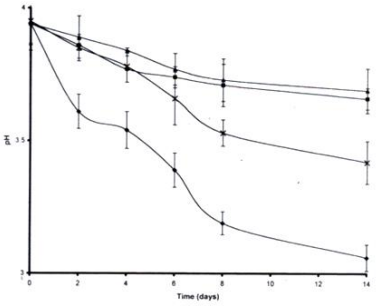

Figure 4. Changes in the pH of pasteurised (■), pasteurised and refrigerated (▲), refrigerated (X) and the control (◆) 'obushera' during storage. (Error bars are standard deviation).

growth were positively correlated ($\alpha = 0.05$) with $r = 0.99$ for pasteurised, $r = 0.81$ for pasteurised and refrigerated, $r = 0.81$ for the refrigerated and $r = 0.87$ for the control obushera. Alcohol production and yeasts growth were positively correlated ($\alpha = 0.05$) with $r = 0.89$ for pasteurised, $r = 0.92$ for pasteurised and refrigerated, $r = 0.95$ for the refrigerated and $r = 0.98$ for the control obushera. The sensory evaluation results show that the pasteurised 'obushera' was acceptable (Table 1). The refrigerated and control treatments exhibited an offensive odour, thus were not acceptable, after six and 2 days of strorage, respectively.

Discussion

Pasteurisation

Pasteurisation at 78°C for 10 minutes reduced the population of lactic acid bacteria from 5.8 $\log_{10}$ c. f. u./ml to 0.6 $\log_{10}$ c. f. u./ml and that of the yeasts from 6.2 $\log_{10}$ c.f.u./ml to undetectable levels. Efiuvwevwere and Akoma (1997) reported a similar reduction of lactic acid bacteria in "Kunun-zaki",

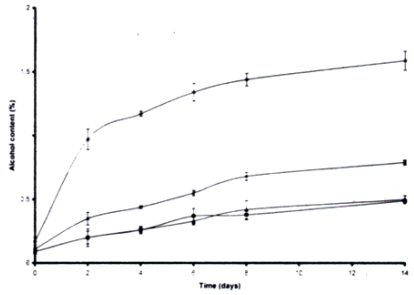

Figure 5. Changes in the % alcohol content of pasteurised (■), pasteurised and refrigerated (▲), refrigerated (X) and the control (◆) 'obushera' during storage. (Error bars are standard deviation).

Table1. Mean sensory scores for 'obushera' subjected to different treatments.

Treatment	Mean score for acceptability[c] at							
	0 days	2 days	4 days	6 days	8 days	10 days	12 days	14 days
Pasteurised	3.00 (0.25)[a]	2.65 (0.29)	3.05 (0.56)	3.04 (0.49)	2.80 (0.37)	2.96 (0.61)	2.98 (0.39)	2.75 (0.11)
Pasteurised and refrigerated	2.50 (0.43)	2.75 (0.44)	3.21 (0.41)	2.69 (0.30)	2.95 (0.35)	2.72 (0.43)	3.30 (0.51)	3.04 (0.17)
Refrigerated	2.77 (0.33)	2.86 (0.32)	3.10 (0.28)	4.35 (0.63)	nd[b] (0.30)	nd	nd	4.04
Control	2.52 (0.36)	nd	nd	nd	nd	nd	nd	2.58 (0.48)

a. Figures in parentheses are standard deviation
b. Sensory score not determined because product had offensive odour
c. 1 = Like extremely and 9 =Dislike extremely.

a fermented millet beverage. Yeasts in Oyokpo, a Nigerian millet beer were reduced after pasteurisation (Iwuagwu and Izuagbe, 1985). Lactic acid bacteria counts in the pasteurised obushera that was stored at room temperature showed a slight increase of 2 $\log_{10}$ cycles through the storage period. Despite a reduction to an undetectable level after pasteurisation, yeast growth was detected after 2 days of storing the 'obushera' at ambient temperature.

Although pasteurisation destroys most vegetative forms of microbes in food, some species of yeast and lactic acid bacteria as well as spores may survive the pasteurisation process (Jay, 1992). The survival of thermo-resistant species of lactic acid bacteria and yeast was probably responsible for growth of lactic acid bacteria and yeasts during storage of the pasteurised 'obushera'. The detection of yeast cells after 2 days of storage suggests the presence and survival of ascospores which could have germinated after that time of storage at ambient temperature.

During the storage of pasteurised 'obushera', the titratable acid and alcohol content increased from 0.3 - 0.58% and 0.09 - 0.49%, respectively. The pH of the pasteurised 'obushera' was reduced from 3.94 to 3.66. The strong correlation between lactic acid bacteria and titratable acid and that between yeast growth and alcohol content of pasteurised 'obushera' indicate that the surviving micro-organisms were responsible for the subsequent changes in these parameters during storage.

Pasteurisation combined with refrigeration

According to Gould (1996) spores of psychrotrophic bacteria, are easily destroyed by pasteurisation while the spores of mesophiles and thermophiles, which might survive pasteurisation, cannot grow at the refrigeration temperatures. Thus, a combination of pasteurisation and refrigeration would result in an enhanced shelf life. However, there was no significant difference, for all the parameters measured, between the pasteurised and the pasteurised and refrigerated 'obushera'. This implies that pasteurisation alone is enough to achieve increased shelf life of 'obushera' without additional refrigeration. Refrigeration can, however, be used to chill 'obushera'.

Refrigeration

The use of refrigeration alone only slowed down the growth of yeasts and lactic acid bacteria in 'obushera'. Lactic acid bacteria and yeast counts in the refrigerated 'obushera' increased tremendously during the storage period. The increase in the control treatment was, however, more than that observed in the refrigerated 'obushera'. The pH and titratable acid of refrigerated 'obushera' ranged between 3.42 - 3.95 and 0.38 - 0.95 %, respectively during the storage period. The alcohol content increased from 0.11 to 0.79%. The refrigerated 'obushera' was not acceptable to the panellists after 8 days. This observation lends support to the fact that refrigeration alone only slows down the growth of micro-organisms but does not stop the growth and activity of the micro-organisms (Jay, 1992). This also explains the lower counts of yeasts and lactic acid bacteria observed in the refrigerated 'obushera' as compared to the control.

Sensory evaluation and shelf-life

The control 'obushera' became unacceptable and was rejected by the panellists after 2 days. The refrigerated sample remained acceptable for 8 days after which the panellists rejected it. The 'obushera' samples subjected to pasteurisation alone and the combined treatment of pasteurisation and refrigeration remained acceptable through the monitoring storage period of 14 days. At the end of 14 days of storage, the 'obushera' subjected to pasteurisation and pasteurisation combined with refrigeration were most acceptable, with their mean scores for acceptability index not being different from those of freshly prepared 'obushera'. This shows that 'obushera' can be preserved by pasteurisation, refrigeration and combined pasteurisation and refrigeration with no adverse effects on its sensory properties. Similarly,

Efiuvwevwere and Akoma (1997) successfully used pasteurisation of kunun-zaki at 70°C for 30 minutes with out adverse effects on the sensory properties of the product. The shelf-life projections based on the changes in the titratable acid and pH indicated that the pasteurised 'obushera' could be stored at ambient temperature for 32 days without decreased sensory quality.

Conclusions

The results show that it is possible to increase the shelf life of 'obushera' beyond the traditional two days with out adverse effects on its sensory properties using simple preservation methods. Although refrigeration increased the shelf life, this was to a limited extent. Pasteurisation at 78°C for 10 minutes alone was adequate to increase the shelf life of the product with no added advantage resulting from combined pasteurisation and refrigeration. Refrigeration, however, may be used to chill 'obushera'.

Although pasteurisation reduced the microbial population in 'obushera', some heat resistant forms of microbes survived and these were responsible for some changes in 'obushera' during storage at ambient and refrigeration temperature. Such surviving microbes do not cause spoilage of 'obushera' during the normal storage time.

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