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Development and Assessment of Shelf-Stable Spicy Snacks Prepared from Buffalo Tripe by Incorporating Sichuan Pepper (*Timur*)

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A study was conducted for the preparation of the shelf stable spicy tripe snack from buffalo tripe with two different spices mixture. The main difference in the spice mixture is that one mix did not contain the Sichuan Pepper (timur) which is called as control batch and another contained it which is called as treated batch. Trial of product development was performed for three times where the combinations of the spices were altered till best combination was obtained. The prepared products were compared to sensory, microbiological and chemical analysis. The timur treated sample was found significantly superior in terms of taste and overall acceptability. The results of microbiological and physiochemical analysis showed that the timur-added snack more shelf-stable than un-added snack. Proximate composition of the best formulation moisture, crude protein, crude fat, crude fiber, total ash, salt and carbohydrates by difference are 43.915±1.5, 10.447±0.5, 15.203±0.5, 1.263±0.7, 2.7±0.7, 5.554±0.6 and 20.919±2.3, respectively.

Key words: Buffalo rumen meat, Buffalo tripe, Shelf-stable Snack, Tripe snack

Introduction

In Nepal, Rumen meat or tripe is one of the most under-utilized meat by-products. Development of products as shelf-stable spicy tripe snack will help to add the value to the by-product and helps to give the buffalo tripe more importance and more economic value.

Buffaloes are slaughtered mainly for meat, the by-products from slaughtered animals are also of good value. Buffalo tripe is one of the important edible offal and weighs about 4.36 to 5.45 kg per animal. Commercial exploitation of tripe for development of processed product manufacture is very limited because of its poor functional properties and inherent toughness due to high collagen content. It is essential to develop technologies for utilization of tripe into processed product manufacture by reducing its toughness. Proteolytic enzymes such as papain, bromelin and ficin have been used for many years to tenderize tough cuts of meat. The best tenderization effects are achieved with papain(Anandh, 2013a).

Among the varied and diverse value added meat products, shelf stable ready-to-eat meat snacks are one of the fastest growing categories in US, Europe and China due to changing lifestyle and favourable image perception of meat snacks. Sales of meat snacks grew 147% between 1997 and 2003 in the US (Roberts, 2003). About 1.5 million tonnes of meat snacks were consumed in Europe in 2005 (Anonymous, 2006). The growing trend in snack meats is mainly due to the consumer convenience, taste, and availability of more variety than in the past. These are looked as potential replacements for junk foods due to low fat and low carbohydrate appeal. Most importantly, consumers view meat snacks as a convenient on-the-go snack food due to time constraints. They can be easily handled and distributed without compromising on safety. Ready-to-serve products such as snack foods, snack bars, and frozen foods are popular with double income households and consumers who are generally short on time(*U.S. Department of Commerce Industry Report*, 2008)

Snacking of perishable foods items like "Tripe" with salt, spices and condiments provide ready to eat products with good shelf stability at ambient temperature. Low moisture and reduced pH are two factors contributing to shelf stability of Snacks. Besides its preservative effect, Snacking also helps to improve desirable characteristics like taste and flavour to the food. Flavour greatly influences acceptability of Snacked products. Snacking controls microbial growth by creating unfavourable condition.

Materials and methods

The materials used in formulation of snack recipe make from tripe is presented in table 1

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Table 1 Snack Recipe

Particulars	Amount (g)
Tripe	1612.9
Oil	20
Cinnamon	1.7
Black Pepper	10
Cardamom	1.7
Cloves	1.1
Schwan Pepper	20
Garlic	50
Cumin	50
Coriander	30
Onion	280
Salt	100
Salt	100
Chilli	50
Turmeric	10
Asafoetida	0.5
Fenugreek	5
Ginger	50
Total	2292.9
Cooking Loss	612.9
Total Product	1679

Product Development

Procurement of fresh, sound buffalo tripe from market was done. Washing and preliminary preparation of the tripe was done. The clean tripe was cooked as whole by braising or stewing. The tripe was cut in desired shape and fried in little measured quantity of oil to brown color and the oil was drained out The spices were mixed in the dried form and heated for a while to develop proper flavor. This was mixed with fried tripe to obtain the formulation as is presented in table 1. The snack was divided in two batches. *Timur* (grinded powder) was added and mixed well in the first batch while the second batch did not contain *timur* and used as control in this study. A preliminary sensory test was conducted to determine the maximum quantity of *timur* that can be added to the snack without any tone of over-spicing in odor and taste. The ready to eat tripe snack was stuffed in small glass jars up to the neck taking care to make it free of air and capped properly.

Sensory Analysis: Control and treated sample were subjected to the 9-point hedonic rating method for sensory analysis.

Proximate analysis: Moisture, protein, fat, ash and salt contents of treated samples were determined by procedures prescribed by (AOAC, 2005) using hot air oven, Kjeldhal apparatus, Soxhlet apparatus, Muffle furnace and titration arrangements respectively. Carbohydrates were estimated by difference of these proximate.

Microbiological analysis: All the microbiological parameters were determined by following standard methods of (APHA, 1984).

Physiochemical analysis like water activity and peroxide value was calculated by procedure described by AOAC (2005).

Statistical analysis: All the parameters were statistically analyzed by t-test between the control and sample except sensory which was done by chi-square test.

Results and Discussion

a. Proximate Composition

The proximate composition of the treated sample is presented in table 2.

Table 2 Proximate composition of the treated sample.

Proximates	Value per 100gm treated sample
Moisture	43.915±1.05
Crude Protein	10.447±0.517
Crude Fat	15.203±0.445
Crude Fiber	1.263±0.087
Total Ash	2.7±0.097
Salt	5.554±0.196
Carbohydrates by difference	20.919±2.303.

Values are Mean ± Standard error of mean for triplicate data.

b. Crude fiber content of the sample was determined because the spices used to prepare the product contains significant amount of fiber in it. The fiber is included in the carbohydrate. Physiochemical Parameters

i. Water Activity

Water activity of any product is very important to determine. It indicates the level of moisture present for the microbiological activity. Food designers use water activity to formulate shelf-stable food. If a product is kept below a certain water activity, then mold growth is inhibited. This results in a longer shelf life. Table 3 shows water activity of control and the treated samples.

Table 3 Water activity of control and treated sample.

S. No.	Control Sample	Treated Sample
1	0.73	0.67
2	0.74	0.67
Mean	0.735	0.67

The water activity of both control and treated sample is below 0.75 which means that there is a hurdle for bacteria to grow and increase their population in snacks. It helps to make the spicy snacks shelf-stable on the basis of low water activity. Since the water activity of the treated sample was found to be 0.67 the product can be considered safe also in terms of yeast and mold development.

ii. Peroxide Value

Peroxide values of the samples were determined on 0, 15, 30, 45 and 60 days. The value of the experiment is given in figure 1.

The peroxide value of the samples was surely at low level, it was due to the use of very less amount of cooking oil and also presence of higher amount of different spices. Spices are known to have very significant antioxidant activity. It was found that PV of both control and treated sample increased significantly within the storage period but was found under safe level. The PV of the snacks stored for 60 days was acceptable under Nepal standard (<10 mEqO_2/Kg). This might be due to oxidation of fat/oil catalyzed by light, oxygen and metal.

Physiological stability of Shelf-life

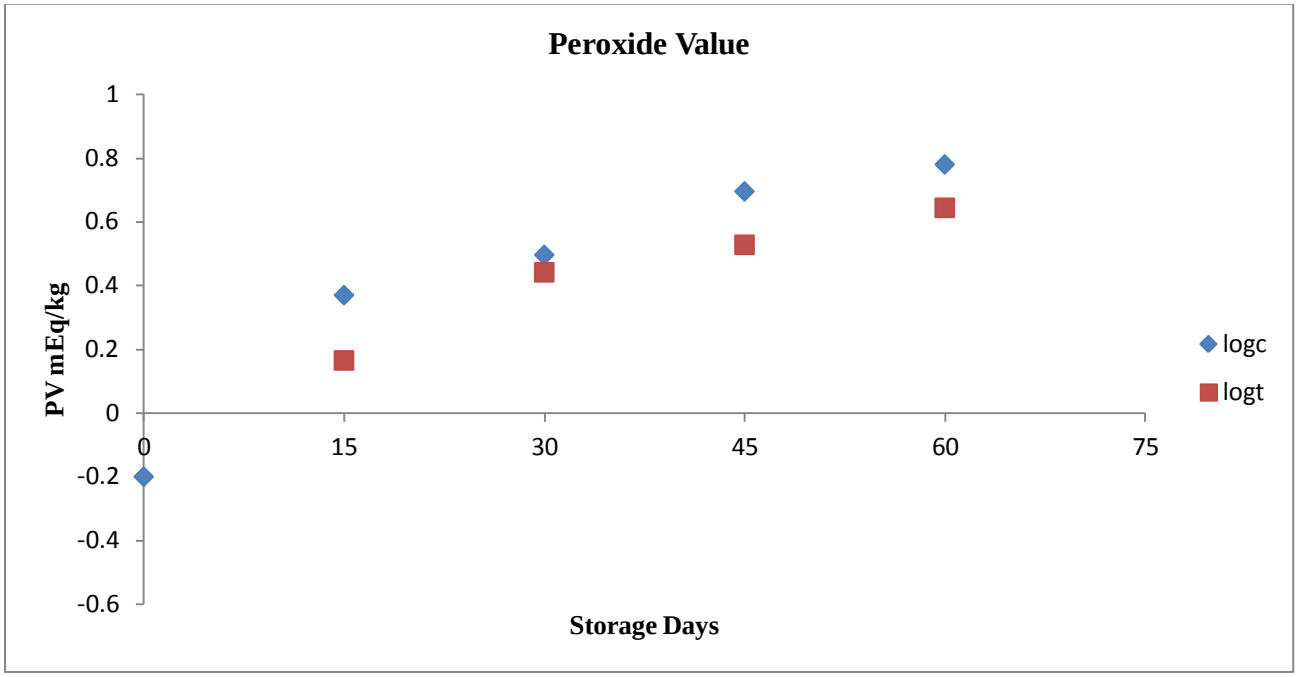

Fig: 1 Peroxide Value Analysis of Snacks within different storage days

The PV of the control sample was higher than that of treated sample within the 60 days of storage. Any difference in the PV shown between the two samples was due to the timur, which was present in the treated sample and not in the control sample. This shows that the antioxidant activity of the timur Microbiological Analysis

The TPC of the control and treated sample were calculated and in both cases the TPC was found below 6 log cycle which is at safe level for shelf-stable meat products. It was also found that in both cases that within the storage days up to 60 days, there was slight increase in the log cycle of aerobic count. It might be due to the breakdown of the fat and protein in smaller form, which might be utilized by the microorganisms.

Microbiological stability of Shelf-life

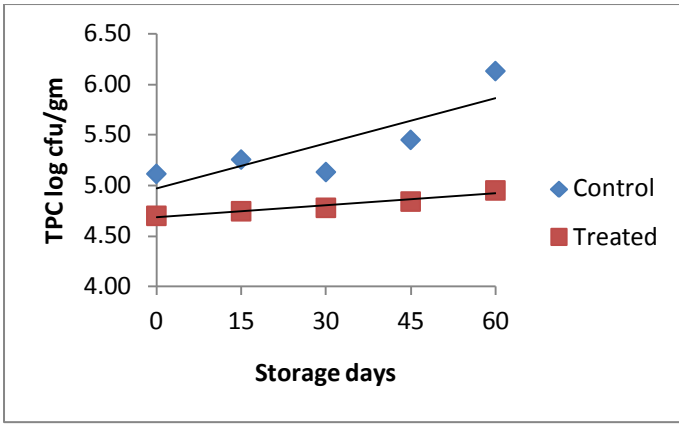

Fig: 2 Microbiological Stability of Snacks within different storage days. The straint line show the linear regression line.

The regression line shows the slope of treated and control sample to be 0.004 and 0.0148 respectively. The y-intercept of the lines are 4.6827 and 4.9709 respectively. The Y-intercept values shows that initial microbial count of the treated sample was lesser than that of control sample. This shows the effectiveness of timur having anti-microbial activityThe lower value of slope, similarly indicate the inhibitory effect timur towards the growth of microorganisms Hence, timur shows the antimicrobial activity on the snacks and helps to inhibit the growth of aerobic organisms and enhance the stability of the product.

Low microbial count in the prepared snacks might be due to heat treatment, spices, salt and low water activity. Because of antioxidant and antimicrobial properties, spices have dual function – in addition to imparting flavor and taste, they play major role in food preservation by delaying the spoilage of food. Salt gives flavor and has a preservative effect. Salt can preserve meat by retarding microbial growth. It retards microbial growth by extracting water from meat and thus lowering water available for the growth of microorganisms. Secondly, it extracts water from microbial cell and cause death by plasmolysis. In the third possible mechanism salt ionizes and the ions diffuse into microbial cells and interfere with the metabolism.

a. Sensory Analysis

The raw formulation of the spices were taken from the (Das, Nath, Hazarika, & Laskar, 2013). The recipe was for the chicken meat pickle and it was not used to make the spicy snacks so further change in the spice recipe was done. The recipe formulation refined by making certain number of formulations were made by heat and trial method and sensory was performed for higher preferences.

For first attempt the some spices were changed as chilli was used about 15% of the weight of the cooked tripe and salt was also used about 15% of weight of the cooked tripe. For treated sample, timur was incorporated about 5% of the weight of the cooked tripe. The expected result was to make a spicy-snack with high sensory acceptance. The product was made and the sensory analysis was allowed to perform. It was found that the mean overall acceptance of first trial was below 7. With the result a second attempt was made to improve the overall acceptance. For the second attempt, chilly was used about 10% of the weight of the cooked tripe and salt was also used about 10% of weight of the cooked tripe. For treated sample, timur was incorporated about 2% of the weight of the cooked tripe. The product was made again and the sensory analysis was allowed to perform again. It was found that the mean overall acceptance of the second trial was around 8 which were more appreciable than first attempt. In the third attempt, many spices were altered with their amount to be contributed. For treated sample, timur was again incorporated about 2% of the weight of the cooked tripe. The product was made again and the sensory analysis was allowed to perform again. Sensory analysis was performed using nine point hedonic scales. The analysis of result with chi-suqare test showed that where the taste and overall acceptability of the treated sample was found significant higher ($P<0.05$) than the control.

Table: 4 Final Sensory Analyses of Tripe Snacks

Parameter	Chi-test ($P<0.05$)
Color	0.068
Odor	0.249
Texture	0.695
Taste	0.022
Overall	0.0002

Shelf-Stability Investigation

Shelf-stability of the samples was investigated under two basis i.e. microbiological and physiological basis.

In microbiological basis, the samples of different storage days were taken for the TPC, Yeast and Mold growth check. It was found that resulted log cycle of the TPC and Yeast and Mold was under the permitted level of the TPC count for shelf-stable cooked meat products. This leads to the conclusion that the product is microbiologically safe up to 60 days. It was also found that the treated sample has significantly lower count of TPC and could be more stable than that of control sample. The results from this study are quite comparable to the study of (Rajbanshi, Adhikari, & Subba, 2014). In the shelf stable intermediate moisture type chicken meat pickle was developed and evaluated to find its quality and storage stability. Pre-cooked and fried lean minced chicken meat was mixed with salt, spices and vinegar and packed air tight in glass jar. The product had appreciable sensory quality and low total microbial, yeast and mould counts. Salmonella, Staphylococci and E. coli were found absent in the product. Microbial count and peroxide value lay within the acceptable levels for sixty days. (Rajbanshi, Adhikari, & Subba, 2014). Similarly, in physiological basis, the PV of the samples of different storage days were taken and it was found that both control and treated sample has PV under the safe level up to 60 days. It was also seen that treated sample has ever lesser PV within the storage days.

Conclusions

From the current study, the inclusion of timur in snack food make from tripe, shows several benefit. Firstly, the nutritional value of the final product increased as is evident by an increase in the fibre content of the product. Secondly, The sensorial properties are enhanced as the final produce scored better in terms of taste and overall acceptance compared to the control sample. The antioxidant and antimicrobial property of timur showed that the treated sample had significantly higher shelf-life and lower aerobic microbial count. In addition to all those added value timur addition is a way of product diversification and utilization of byproduct to convert it to tasty final product, which if explored further might have potential for commercialization.

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