



Production and Quality Evaluation of Soy Cheese (Tofu) Using Selected Coagulants

*** J. Ndife¹, ²I. Imade and ¹S. Ubbor**

¹Department of Food Science and Technology, Michael Okpara University of Agric., Umudike.

²Dept. of Food Technology, Kaduna Polytechnic, Kaduna Nigeria.

*Corresponding author. E-mail: jothel2000@gmail.com.

Abstract

Food formulations that incorporate soy ingredients are being exploited as veritable alternatives. Cheese analogues are healthy alternatives to benefit from consuming functional foods. There is the need to enhance the production and quality of soy cheese. In this work, soy cheeses (*Tofu*) were prepared from soybean using four selected coagulants – *Moringa oleifera* seeds extract (MC), Tamarind fruit pulp extract (TC), Lime juice (LC) and Alum (AC). The effects of the coagulants on the physico-chemical, microbial and sensory qualities of the soy cheese products were evaluated. The results of the physical properties of the samples showed that MC gave higher yield (12.84 %) and acidity (0.37 %), while AC had the highest values in hardness (84.55 %) and dry matter (36.72 %) contents. The proximate composition showed that MC had the lowest moisture (60.52%) but highest protein (22.51%) contents. The ash contents of the soy cheese from the plant coagulants MC (3.00 %), TC (2.80 %) and LC (3.5%) were lower than that of the Alum coagulant AC (3.89%). The cheese from Alum coagulant (AC) had more minerals than those from plant coagulants. TC had the highest Vitamin C (2.69 mg/100g) while MC had the highest vitamin B₁ (6.07 mg/100g) and B₂ (29.21 mg/100g) contents. There was no detection of coliform in the soy cheese samples. There were no significant difference ($p > 0.05$) in the appearance and overall acceptability of sample MC and sample LC. Sample MC from *Moringa oleifera* seed cake extract coagulant had the highest sensory scores for Flavour (8.35), texture (8.40) and overall acceptability (8.63). The results of quality properties of the cheeses differed with the coagulants used. *Moringa oleifera* coagulated cheese had the most improved quality of all the samples.

Keywords: Cheese, coagulants, *moringa oleifera*, tamarind, lemon juice

Introduction

Cheese is the curd formed by the coagulation of milk by rennet or enzymes in the presence of lactic acid produced by added micro-organisms (Johnson *et al.*, 2001; Achachlouei *et al.*, 2013).

They are produced with a wide range of flavors, textures, and forms by coagulation of the milk protein casein. Usually the milk of buffalo, goat, or sheep is used. During production the milk is usually acidified, by adding the enzyme rennet which causes coagulation. The solids produced are separated and pressed into final form (Achachlouei *et al.*, 2013).

Cheese is very nutritious because of the high protein quality (Butler *et al.*, 2010; Ndife, 2016), and is also a veritable source of dietary calcium. Cheese has fewer digestive problems compared to other dairy products (Oboh and Omotsho, 2005; Onwuka, 2014). However, the high saturated fats content detracts from the nutritional quality and poses a health challenge to consumer perception (Onwuka, 2014). Cheese analogues or imitation cheese seems to be a good alternative to incorporate the health benefits derivable from consuming cheese (Moller *et al.*, 2008; Soybean, a highly nutritious plant legume contain well balanced amino acids and desirable fatty acids. It serves as an important protein source for

many people around the world (Amadou *et al.*, 2009, Barnes, 2010; Butler *et al.*, 2010) Food formulations that incorporate soy proteins into different products are being exploited by food experts and manufacturers as functional foods (Nanri, 2010; Ndife, 2016). The inclined acceptance of soy foods by the general population is due to increase recognition of the health benefits of soy foods, especially by skeptics, who want to reduce their consumption of animal products.

Tofu is the Japanese name for bean curd products. *Tofu* is low in calories, rich in essential amino acids, contains beneficial amount of iron, is low in saturated fat, high in vitamins and contain antioxidants (Achachlouei *et al.*, 2013). *Tofu* is popularly consumed in Nigeria because of the various nutritional and medicinal attributes associated with soybeans products such as reduction of cardiovascular disease, osteoporosis and cancer risks (Butler *et al.*, 2010; Nanri, 2010). Furthermore, the cost of producing cheese analogues is less than products obtained from animal sources (Oboh and Omotsho, 2005; Velasquez and Bhatena, 2007; Rinaldoni *et al.*, 2014).

Various types of coagulants are used for the coagulation of milk. These coagulants are reported to impart both physical and chemical as well as sensory effects on the

quality of the curds (Achachlouei *et al.*, 2013; Gopalakrishnan *et al.*, 2016). Thus, the objective of this study was to study the potentials of selected coagulants that include *Moringa oleifera* seed cake extract, Tamarind pulp, Alum salt (Aluminum sulphate) and lime juice for the production of soy cheeses and to evaluate the physicochemical properties, microbial qualities and sensory acceptability of the products. This will help determine which coagulant is most suitable for tofu production and development

Materials and Methods

Raw material procurement

The soy beans and coagulant materials (*Moringa oleifera* seeds, lime fruit, tamarind seeds, and Alum (Potassium aluminum sulphate) used for this study were purchased from Kaduna Central market in Kaduna State of Northern Nigeria. The chemicals and reagents used were of analytical standard. The equipment and utensils used were standardized for the purpose of this research.

Preparation of coagulants

Alum: Alum salt (Potassium aluminum sulphate) was milled into powder and 200 g of the salt was dissolved in 250 ml water,

filtered and the filtrate was used as Alum coagulant.

Lime: Lime fruits were cut into equal halves and their juice squeezed out; the extracts from the juice (200 ml) was sieved to get rid of seeds. The juice obtained served as the lime coagulant.

Tamarind: Tamarind fruit pod were sorted to remove extraneous materials and unhealthy ones. The pods were removed from the fruits which were then soaked in water for 2h to allow for softening of the fruit pulp. This was pulverized with equal volume of water in a warring blender, then filtered by squeezing through a muslin cloth. The filtrate was used as Tamarind coagulant.

Moringa seed extract:

Moringa seed were sorted to obtain healthy. The seeds (300 g) were soaked in 300 ml of water and then pulverized in a warring blender (Bin atone) for 2 min to extract the water-soluble components. The extract was recovered by squeezing the pulverized slurry through a folded muslin cloth. This fine extract was used as *Moringa* coagulant.

Production of cheese samples

The soy beans were sorted to remove extraneous materials, washed to remove dirt before blanching the seeds for 25 minutes

in boiling water. The soy bean seeds were then steeped in water twice the volume of the soya beans for 6 h before dehulling manually. The soy beans were drained out of water and milled into paste with an attrition mill. The soy paste was spread and folded muslin cloth by squeezing out as much filtrate as possible. The soy milk obtained was divided into four portions (10 L each) and mixed with salt and spices. The different liquid coagulants comprising of Alum water, Tamarind seeds extracts, *Moringa oleifera* seeds extracts

and Lime juice (200 ml each) were added to the separate soy milk portions and allowed to coagulate for 4 h before cooking for 30 minutes until curd formation. The curds were drained, and pressed to expel the whey before cutting them into uniform sizes. The soy cheeses were labeled according to their coagulants as: AC (Alum), TC (Tamarind seeds extracts), MC (*Moringa oleifera* seeds extracts), and LC (Lime juice) respectively (Table 1), and kept in the refrigerator at 5 °C for subsequent analysis.

Table 1: Formulation of Soy Cheeses

Ingredients	Samples			
	AC	MC	TC	LC
Coagulants (ml)	200	200	200	200
Soy milk (L)	10	10	10	10
Boulioun (g)	25	25	25	25
Onion (g)	100	100	100	100
Pepper (g)	100	100	100	100
Salt (g)	10	10	10	10

AC (Alum), TC (Tamarind seeds extract), MC (*Moringa oleifera* seeds extract); LC (Lime juice)

Methods of Analysis

Some physico-chemical, microbial and sensory parameters were analyzed to ascertain the quality of the soy cheeses using standard procedures viz:

Determination of physical properties

The physical properties of % yield, % compression and acidity were determined by using procedures described by Onwuka (2018).

Determination of chemical properties

The determination of the chemical composition of the samples for dry matter content, moisture, ash, protein, fat, fiber and carbohydrate contents were determined by methods described by AOAC (2005). The vitamin and mineral contents were also determined by procedures described by AOAC (2005).

Mineral contents determination

Each sample was digested by the wet ashing method prior to mineral content determination using atomic absorption spectrophotometer for Ca, Mg and Fe and Corning 400 flame photometer for K and Na (AOAC, 2005). The phosphorus content was determined colorimetrically with Jenway 6100 spectrophotometer using the method described by Nielsen (2003).

Vitamin contents determination

The spectrophotometric method described by Jacobs, (1999).was used; the absorbance of the sample and the standard solutions were measured with a UV-Spectrophotometer at their respective wavelengths. The concentrations of the vitamins in the prepared samples were run against known standards calibrated curve.

Microbial contents determination

The determination of the microbial quality (mesophilic aerobic bacteria, coliforms and fungi counts) of the cheese products were performed by the method outlined in compendium of methods for the microbiological examination of foods (APHA, 1992) with some modifications.

Sensory evaluation

The protocol described by Iwe (2002) was used. The organoleptic properties of soy cheese were evaluated by 20-member semi-trained panelists, randomly selected from the staff and students of the university. Quality attributes of the cheese, including appearance, aroma, texture, taste and general acceptability, were evaluated using 9-point hedonic scale, with 1 as dislike extremely, 5 as neither like nor dislike and 9 as like extremely.

Experimental design and statistical analysis

The experimental set-up was of a completely randomized design. Results are presented as mean \pm standard deviations. The data obtained from the various analysis were subjected to analysis of variances using the statistical package for social sciences (SPSS), version 16.0. One way analysis of

variance (ANOVA) was used for comparison of the means. Differences between means were considered to be significant at $p < 0.05$ using the Duncan multiple range test.

Results and Discussion

Physical properties of soy cheese samples.

Table 2 shows the results of the physical properties of the soy cheese samples.

Coagulation of the protein and oil (emulsion) suspended in the boiled soy milk is the most important step in the production of high yield of tofu (Moller *et al.*, 2008). The soy cheese produced from *Moringa oleifera* seed extract (MC) had highest yield (12.84%) and acidity (0.37%), while the Alum-salt coagulated cheese (AC) had the highest values in hardness (84.55%) and dry matter (36.72%) contents, in comparison to the soy-cheese from other plant coagulants.

Table 2: Physical Properties of Soy Cheeses

Para meter s(%)	AC	MC	TC	LC
Yield	11.3 5 ^b ±0 .55	12.8 4 ^a ±0 .48	10.4 2 ^c ±0 .40	11.2 0 ^b ±0 .63
Hardn ess	84.5 5 ^a ±0	80.4 6 ^b ±0	66.2 8 ^d ±0	75.9 2 ^c ±0

	.31	.35	.35	.40
Acidit y	0.28 b±0.	0.37 ^a ±0.0	0.23 b±0.	0.32 ^a ±0.0
	03	6	05	4
Dry Matter	38.7 2 ^a ±0	36.8 1 ^a ±0	28.5 6 ^c ±0	34.4 0 ^b ±0
	.01	.04	.06	.08

*Means within a row with different letters are significantly different at $p < 0.05$: AC (Alum-potassium aluminum sulphate), TC (Tamarind fruit pulp extracts), MC (*Moringa oleifera* seed extracts), and LC (Lime juice).

There were significant differences ($p < 0.05$) in some of the physical properties of cheese samples. This is an indication that the selected coagulant under consideration differ substantially in their coagulating ability, the difference could be as a result of their chemical components (Johnson *et al.*, 2001; Ghebremichael *et al.*, 2005). Firm *Tofu* contains low amount of moisture and has firmness comparable to cooked meat and a somewhat rubbery feel similar to that of pannier (Prestamo *et al.*, 2000). Salt coagulants are mostly used in the production of Chinese-type brittle tofu. Acid coagulants have been reported to produce higher and firmer yields of curds than salt and enzyme precipitated curds (Gopalakrishnan *et al.*, 2016). The production techniques employed help to create tofus with unique physical and chemical characteristics (Butler *et al.*, 2010).

Chemical compositions of soy cheese samples.

The results of the proximate composition of the different soy cheese samples as indicated in Table 3, shows the moisture content to be lowest in sample MC (60.52 %) and highest in sample LC (65.23 %). The protein and fat contents of the soy curds produced ranged from 17.36 - 22.51% and 5.53-7.56% respectively. There was significant difference ($p < 0.05$) in the protein content of the soy cheeses. The high protein content of the *Moringa* coagulated sample MC (22.51%) could possibly be attributed to the likelihood that the protein in the *Moringa* seeds and the other plant extracts (TC and LC) might have been transferred into the soy curd unlike the Alum salt coagulant AC (17.36%). The reduction of the moisture contents

of the cheese samples could lead to increased protein concentration. Furthermore, the protein content of the soy cheeses produced, was higher than those of the commercial soy cheese (12%) reported by Rinaldoni *et al.*, (2014). This variation could be as a result of the difference in the variety of soybean, coagulants used and the condition under which the coagulation was carried out (Moller *et al.*, 2008; Amadou *et al.*, 2009).

The fat contents of samples MC (7.56%) and TC (6.80%) were higher than that of AC (5.53%) and LC (6.12%). The fat contents of these soy cheese samples (5.53 - 7.56%) were however lower than the average value (9.0%) reported by Amadou *et al.* (2009) for some commercial soy cheeses.

Table 3: Proximate Content of Soy Cheeses

Parameters (%)	AC	MC	TC	LC
Moisture	63.40 ^b ±1.23	60.52 ^b ±0.53	68.41 ^a ±0.20	65.23 ^a ±0.25
Protein	17.36 ^c ±1.20	22.51 ^a ±1.34	19.60 ^b ±1.10	20.16 ^b ±1.25
Fat	5.53 ^c ±0.58	7.56 ^a ±0.80	6.80 ^b ±0.53	6.12 ^b ±0.47
Ash	3.89 ^a ±0.10	3.00 ^b ±0.53	2.80 ^b ±0.70	3.5 ^a ±0.20
Carbohydrate	6.34 ^b ±1.24	7.46 ^a ±1.40	4.82 ^c ±0.98	6.28 ^b ±1.15

*Means within a row with different letters are significantly different at $p < 0.05$: AC (Alum-potassium aluminium sulphate), TC (Tamarind fruit pulp extracts), MC (*Moringa oleifera* seeds extracts), and LC (Lime juice)

The ash contents of the soy cheeses were AC (3.89%), MC (3.00 %), TC (2.80 %) and LC (3.5%) respectively. The highest ash content was obtained in AC (3.89%) with Alum coagulant, followed by the Moringa sample MC (3.00 %). The inorganic nature of the Alum coagulant must have contributed to the increase in the mineral and ash contents of sample AC. The increase in mineral content of *Tofu* may be attributed to the use of inorganic coagulants as demonstrated in previous studies (Amadou *et al.*, 2009; Achachlouei *et al.*, 2013).

Mineral contents of soy cheese samples.

The mineral content of the soy cheese samples is presented in Table 4. The mineral contents ranged from 0.11 to 41.03ppm. Sample LC had the highest calcium content (41.03 ppm) while sample AC had the lowest phosphorus content (9.02ppm). The highest iron and manganese values were recorded in sample MC (3.66 ppm) and sample AC (0.64 ppm) respectively.

Table 4: Mineral Contents of Soy Cheeses

Parameters(ppm)	AC	MC	TC	LC
Calcium	41.03 ^b ±0.65	29.00 ^c ±0.74	31.02 ^c ±0.59	58.01 ^a ±0.60
Phosphorus	9.02 ^c ±0.01	14.06 ^a ±0.01	12.06 ^b ±0.02	10.07 ^c ±0.02
Iron	1.13 ^b ±0.20	3.66 ^a ±0.51	1.73 ^b ±0.18	4.29 ^a ±0.32
Manganese	0.6 ^a ±0.03	0.11 ^b ±0.04	0.26 ^b ±0.01	0.34 ^b ±0.01

*Means within a row with different letters are significantly different at $p < 0.05$: AC (Alum-potassium aluminium sulphate), TC (Tamarind fruit pulp extracts), MC (*Moringa oleifera* seeds extracts), and LC (Lime juice)

The percentage mineral content is considered as a quality criterion in nutritional adequacy of foods for wellbeing. The differences in mineral content would majorly depend on the type of substrate and the nature of the coagulants (Achachlouei *et al.*, 2013). The use of salt coagulants has been reported to make tofu that is rich in minerals with tender but slightly

brittle texture (Amadou *et al.*, 2009).

Vitamin contents of soy cheese samples.

Table 5 shows the results of the vitamin contents of the soy cheese samples. The vitamin contents were generally low. The vitamins

analyzed were essentially water soluble vitamins which are known to be heat labile. The samples were subjected to cooking to hasten the coagulation process during the production.

Sample TC had the highest Vitamin C (2.69 mg/100g) content while sample MC had the highest vitamin B2 (29.21 mg/100g) and B1 (6.07 mg/100g) contents. The

soy cheeses must have derived some of the vitamins from the different coagulants especially the plants based coagulants. It has been reported by several researchers that Moringa is very rich in these water soluble vitamins. The alum salt coagulant (AC) produced the least vitamin content in the cheeses, as there was no contribution of vitamin from the salt coagulant

Table 5: Vitamin Contents of Soy Cheeses

Parameters(mg/100g)	AC	MC	TC	LC
Vitamin C	1.07 ^c ±0.65	2.43 ^a ±0.74	2.69 ^a ±0.59	1.88 ^b ±0.60
Vitamin B ₂	2.31 ^c ±0.01	29.21 ^a ±0.01	22.34 ^b ±0.02	19.62 ^b ±0.02
Vitamin B ₁	1.10 ^c ±0.20	6.07 ^a ±0.51	4.82 ^b ±0.18	3.70 ^b ±0.32

*Means within a row with different letters are significantly different at $p < 0.05$: AC (Alum-potassium aluminium sulphate), TC (Tamarind fruit pulp extracts), MC (*Moringa oleifera* seeds extracts), and LC (Lime juice).

Microbial contents of soy cheese samples.

The mean bacteria and fungi (yeasts and mould) counts of the soy cheese samples that were

studied, as shown in Table 6, ranged from 7.8×10^2 to 6.0×10^4 cfu/g and 1.0×10^1 to 5.5×10^1 cfu/g. There were significant differences ($p < 0.05$) in the bacteria and fungi counts of the soy cheeses.

Table 6: Microbial Contents of Soy Cheeses

Parameters(cfu/g)	AC	MC	TC	LC
Total bacteria counts	$5.5^c \times 10^3$	$1.1^b \times 10^4$	$7.8^d \times 10^2$	$6.0^a \times 10^4$
Total fungi counts	$1.6^c \times 10^1$	$1.0^c \times 10^1$	$4.0^b \times 10^1$	$5.5^a \times 10^1$
Total coliform counts	Nil	Nil	Nil	Nil

*Means within a row with different letters are significantly different at $p < 0.05$: AC (Alum-potassium aluminium sulphate), TC (Tamarind fruit pulp extracts), MC (*Moringa oleifera* seeds extracts), and LC (Lime juice)

Sample LC had the highest bacteria (6.0×10^4 cfu/g) and fungi counts (4.0×10^1 cfu/g). while TC and MC had the lowest bacteria (1.1×10^4 cfu/g) and fungi (1.0×10^1) loads respectively. There were no coliform bacteria detected in the soy cheese samples. The microbiological quality of the samples is an indication of the hygienic conditions under which the product was handled and processed and points to the type of fermentation and subsequent spoilage envisaged when stored

(Oboh and Omotsho, 2005; Onwuka, 2014).

Sensory evaluation of soy cheese samples

The mean sensory scores on the organoleptic preference for different soy cheese samples are shown in Table 7. There were significant differences ($p < 0.05$) in the sensory attributes analyzed between the different types of soy cheeses.

Table 7: Mean Sensory Scores of Soy Cheeses

Parameters	AC	MC	TC	LC
Appearance	7.31 ^b ±0.63	7.76 ^a ±0.45	5.54 ^c ±0.70	7.56 ^a ±0.50
Flavour	8.04 ^b ±0.71	8.35 ^a ±0.82	7.16 ^c ±0.65	8.22 ^b ±0.75
Texture	8.31 ^a ±0.55	8.40 ^a ±0.60	7.33 ^b ±0.74	7.50 ^b ±0.63
Overall acceptability	8.17 ^b ±0.48	8.63 ^a ±0.53	7.40 ^c ±0.61	7.53 ^c ±0.48

*Means within a row with different letters are significantly different at $p < 0.05$; AC (Alum-potassium aluminium sulphate), TC (Tamarind fruit pulp extracts), MC (*Moringa oleifera* seeds extracts), and LC (Lime juice).

Samples MC and AC had the highest scores of 7.76 and 7.31 respectively, for appearance attribute. Similar trends were obtained in the panelists' scores for attributes of flavour and texture. The difference in scores for texture among the soy cheeses implies that the coagulants had noticeable effect on the procedures. Soy cheese samples MC and AC had the best overall acceptance scores of 8.63 and 8.17 respectively. The low

acceptability of TC (7.40) and LC (7.53) coagulated with tamarind and lime respectively, could be attributed to the impact these coagulants had on the colour and texture of the soy curd (Barnes, 2010; Butler *et al.*, 2010). Cai and Chang (1999) reported that acid coagulants tend to affect the taste and texture of *tofu* more than *tofu* coagulated by alkaline coagulants. The alum coagulant had no strong perceivable smell and taste in the

soy cheese (AC). Salt coagulants have been reported to produce tofu that is tender but slightly brittle in texture (Gopalakrishnan *et al.*, 2016).

Conclusion and Recommendations

The result of the study showed that *Moringa oleifera* seed cake extract was a better coagulant when compared with other coagulants used in the production of soy cheeses. *Moringa oleifera* coagulated soy cheese (MC) had superior sensorial quality and higher nutrient contents compared

to the other soy cheeses. The alum salt coagulated soy cheese (AC) though lower in nutrient values was organoleptically better than the Tamarind (TC) and lime (LC) coagulated soy cheeses. Because of the high nutritional and health benefits of *Moringa oleifera*, the soy cheese produced could serve as functional food.

Further research should be made to identify the contribution of each coagulant type on the nutritional quality of soy *tofu*. The production of soy *tofu* with probiotic advantages will be a welcome development.

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