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Effect of different coagulants on yield and quality of tofu from soymilk

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Abstract Soymilk tofu coagulated with four indigenous coagulants was compared in terms of chemical, textural, colour, and sensory attributes with calcium sulphate (CS) coagulated tofu. Coagulants used are Epsom salt (ES), lemon juice (LJ), alum and top water of fermented maize (TWFM). Protein and magnesium content significantly (P < 0.05) increased from 44.5 to 51.7 g/100 g and 252 to 324.6 mg/100 g, respectively, in soybean grain to soymilk. Calcium and magnesium contents increased and decreased significantly (P < 0.05) in tofu coagulated with CS and ES, respectively. Lightness (L) values were 86.3, 86.2, 77.8, 72.4 and 84.6, redness (a) values are 0.34, 0.21, 0.87, 1.05 and 0.32, and yellowness (b) values were 24.0, 23.9, 27.3, 20.3 and 23.4 for CS, ES, LJ, alum and TWFM. The hardness, chewiness and brittleness of textural properties of tofu were significantly (P < 0.05) affected by different sources of coagulation. Sensory evaluation data shows that LJ impacts a significantly acceptable sensory attribute to tofu. This study has demonstrated that tofu quality is affected by the type of coagulant used in curding the soymilk.

Keywords Coagulants · Quality · Yield · Textural attributes · Tofu · Sensory characteristics

Introduction

Tofu is a highly versatile and nutritious food that is made from soybean curds. Although the word "tofu" is

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Institute of Agricultural Research and Training, Obafemi Awolowo University, P.M.B. 5029 Ibadan, Nigeria e-mail: veromum1@yahoo.co.uk Japanese, the food seems to have originated in ancient China, where the Mandarin term is doufu [1]. While tofu is gaining an increasing popularity in western countries, it remains the most important and popular food product in east and south-eastern Asian countries [2]. For most Nigerians, tofu is receiving attention because it is high in protein and its usage as a substitute for meat. Tofu is often sold as a wet block in rural Nigeria and it is generally made and sold on the same day as it is highly perishable.

Tofu, also known as soybean curd, is a soft, cheeselike food made by curdling fresh hot soymilk with a coagulant. It is produced traditionally by curdling fresh hot soymilk with either salt (CaCl₂ or CaSO₄) or an acid (glucuno-d-lactone) [2]. The coagulant produces a soy protein gel, which traps water, soy lipids and other constituents in the matrix forming curds. The curds are then pressed into solids [3, 4]. While there are only three basic steps in tofu making, it is complex and difficult to make a high-quality product. The quality of the beans, the amount of stirring, the coagulants, and the pressing of the curd can have a high impact on the quality of the final product [1].

Yield and quality of tofu have been reported to be influenced by soybean varieties, soybean quality, processing conditions and coagulants [2, 3]. The coagulation of soymilk relies on the complex interrelationship between type of soybean, soymilk cooking temperature, volume, solid content, pH, coagulant type, amount and time [4]. Coagulation of soymilk is a complex interaction of several variables [5].

At this point, the elimination of electrical repulsion charges allows the proteins to aggregate. Lemon juice (LJ) and vinegar can be used as alternatives to calcium sulphate (CS) as they reduce the pH of the system and allow protein to aggregate. Studies have shown that the amount of soy protein used to make the soymilk is critical for tofu yield and quality because tofu is a soy protein gel [6, 7].

Poysa and Woodrow [6] reported that different coagulants produce tofu with different textural and flavour properties. CS creates a bridge by which the soy proteins in the milk can aggregate. It may also interact with proteins to enhance the cross-linking of polymers [8, 9]. The combined heat- and calcium-induced mechanisms work to produce the tofu. The resulting tofu product is affected by such things as pH, concentration of coagulate, and the rate at which the product is stirred [8].

The texture of tofu should be smooth, firm and coherent but not hard and rubbery. Different cultures often prefer different textures of tofu and as such, coagulants used for tofu coagulation defer from one nation to the other. The most often used being small amount of either CS or magnesium chloride. The Chinese have used the calcium salt mined from mountain quarries for 2000 years [10]. The salt is the pure form of gypsum. The Japanese traditionally used sea salt in form of magnesium chloride to coagulate soymilk. Recently, CS and/or glucono-d-lactone (GDL) have been mostly used as coagulant for the production of tofu in Japan [11].

These salts are, however, expensive and not readily available for home use in Nigeria. For this reason, households in Nigeria have sorted out other means of curding soybean milk in tofu processing. Available coagulants used in Nigeria for tofu processing are, epsom salt (magnesium sulphate), LJ, alum and top water on fermented maize grains. The present study assesses the yield and quality of tofu from soybean coagulated with these coagulants using calcium sulphate as a control.

Materials and methods

Materials

Soybeans were obtained from the Institute of Agricultural Research and Training, Obafemi Awolowo University, Ibadan, Nigeria seed store.

Soymilk and tofu processing

Soymilk and tofu were prepared according to Prabhakaran [12] with slight modifications. Soybean grains (400 g) were soaked in 2,500 g (6:1) tap water and stored in the refrigerator $(4-5^{\circ}C)$ for 8 h. The soaked bean which had doubled its original weight was ground in batches with 3,000 g water for 5 min at high speed in LBC10 laboratory blender (Model 2610T, Waring Products, Inc (Waring Laboratory & Science), Calhoun, GA, USA). After grinding, the slurry was filtered through a muslin cloth and squeezed by hand to obtain soymilk. The residue obtained was mixed in 1,000 g of water to make slurry and then filtered again to recover more soymilk solids. The soymilk was heated to boiling point, maintained for 5 min while stirring and cooled to 75°C.

Tofu was prepared by coagulating the soymilk using any one of the coagulants, CS, Epsom salt (ES), LJ alum and top water of fermented maize (TWFM). The coagulant concentrations used for tofu preparation were 0.3% calcium sulphate, 2.5% alum and 0.5% each of other coagulants based on the amount of soymilk used. While 20 mL of LJ and 235 mL of TWFM was used directly for the coagulation, 10 g of ES and alum was dissolved in 235 mL of warm water 5 min before use. The titratable acidities were 0.189, 0.021, 0.014 and 0.182, respectively, for LJ, TWFM, ES and alum. The prepared coagulants were added to the milk and mixture was vigorously stirred with a stainless steel paddle for 15 s. The mixture was allowed to stand for 10 min when the curd has separated from the whey and transferred to a wooden mould for pressing. The bean curd was pressed at 20.5 g/cm² for 60 min. After pressing, tofu samples were weighed and stored in water at 4–5°C before analyses [4].

Chemical analysis

Chemical composition of the soybean, soymilk and tofu was done. The crude moisture, protein and fat content were determined by vacuum oven method, Kjeldahl method using a protein conversion factor of 6.25 and Soxhlet extraction method, respectively [13]. The ash contents were determined using the method of AOAC [14]. Mineral analysis was determined by the digestion of the sample on a mixture of 4 mL of 60% perchloric acid and 25 mL of concentrated nitric acid and mineral constituent read by atomic absorption spectrophotometry using a Perkin-Elmer 560 instrument ((Perkin-Elmer Corp.). Results were expressed on dry-matter basis.

Texture profile analysis (TPA) of curd

Texture of curd was determined by TPA using a Stable Micro System, model TA.-XT2 (Stable Micro System, Texture Technologies Corp., White Plains, NY, USA). Three curd samples of cylindrical shape were cut vertically from a block of curd using a cylindrical cutter (25 mm diameter). Samples were compressed twice to 25% of its original height with a metal disc (60 mm diameter). The TPA curve was recorded and used to calculate the hardness, brittleness and chewiness using the software provided with the Texture Analyzer [15]. Determined parameters (hardness, brittleness and chewiness) using the instrument software were calculated from the penetrometer curve as described by Bourne [16].

Yield and colour analysis

The yield of tofu was calculated as the weight (g) of fresh tofu obtained from a specified amount of the soymilk used for its preparation [12]. The colour of tofu, expressed in L, a, and b values, according to the method described by Obatolu et al. [17].

Sensory evaluation

Sensory evaluation was done on freshly made tofu. Ten semi-trained panellists evaluated the sensory attribute of the fresh tofu. Panellists were familiar with product sensory evaluation, most having participated in previous related projects. Panellists were given no time limit for the evaluation, though most panellists took 10-15 min to complete the rating of all the samples. Tofu was cut into cubic samples and placed on a plastic plate with a random number [4]. The attributes evaluated were colour, flavour, mouth feel and overall acceptability. For each sample, panellists scored their liking of these characteristics using the nine-point hedonic scale (1 = dislike extremely, 2 = dislike very much,3 = dislike moderately, 4 = dislike slightly, 5 = neither like nor dislike, 6 = like slightly, 7 = like moderately, 8 = like very much, and 9 = like extremely).

Statistical analyses

Data were analysed in triplicate and subjected to the analysis of variance (ANOVA) using the Statistical Analysis Systems software package [18]. Significant differences (P < 0.05) among group means were calculated with the Duncan's multiple range test.

Results and discussion

Table 1 shows the chemical composition of the soybean, the soymilk and tofu coagulated with different coagulants. The tofu was processed from the same soybean and the same batch of soymilk. A significant (P < 0.05) increase and decrease of protein and fat, respectively, was observed when soybean was processed into soymilk. Similar trend of significant increase and decrease in the content of protein and fat, respectively, was observed when the soymilk was coagulated into tofu irrespective of the source of coagulant. It could be assumed that the coagulants allow the release of fats during processing, probably suggesting that the processing method considerably decreases the fat-binding capacity of protein. After the addition of the various coagulants into the soymilk, a quick formation of curd was observed with separation of whey.

The moisture content of tofu samples varied from 70.6% in alum coagulated tofu to 79.9% in CS coagulated tofu (control). The variation in the moisture content of tofu prepared with different coagulants is probably due to the differences in gel network within the tofu particles that is influenced by different anions and its ionic strengths towards the water holding capacity of soy protein gels. Alum was observed to be rapid in its action of coagulating the soy proteins.

Table 1 Effect of coagulants on chemical composition and yield of tofu samples

Nutrients (g/100 g)	CS	ES	LJ	Alum	TWFM	Soybean	Soymilk
Moisture [*]	79.9 ± 1.1^{b}	$75.7 \pm 1.0^{\circ}$	73.3 ± 0.7^{cd}	70.6 ± 1.3^{d}	78.1 ± 1.3^{b}	8.2 ± 0.7^{e}	88.9 ± 0.9^{a}
Protein ^{**}	$58.2 \pm 1.5^{\rm a}$	$54.2 \pm 1.3^{\circ}$	56.2 ± 1.2^{bc}	$54.8 \pm 1.3^{\circ}$	57.4 ± 1.6^{ab}	$44.5 \pm 0.9^{\rm e}$	51.7 ± 1.2^{d}
Fat **	$13.7 \pm 0.8^{\circ}$	$13.3 \pm 0.4^{\circ}$	$12.9 \pm 0.6^{\circ}$	$12.3 \pm 0.2^{\circ}$	$13.1 \pm 0.7^{\circ}$	21.8 ± 0.8^{a}	16.3 ± 0.6^{b}
Ash ^{**}	7.9 ± 0.03^{a}	7.2 ± 0.05^{a}	$5.8 \pm 0.03^{\circ}$	6.2 ± 0.02^{b}	$5.2 \pm 0.03^{\circ}$	5.1 ± 0.01^{a}	4.6 ± 0.04^{b}
Calcium ^{**} (mg)	312.7 ± 15.5^{a}	$208.2 \pm 18.8^{\circ}$	$210.9 \pm 10.8^{\circ}$	$223.1 \pm 19.6^{\circ}$	222.3 ± 7.0 ^c	254.8 ± 10.5^{b}	$204.3 \pm 12.6^{\circ}$
Magnesium**(mg)	$237 \pm 3.3^{\circ}$	307 ± 6.4^{a}	238 ± 2.5^{c}	$231 \pm 4.3^{\circ}$	$233 \pm 5.3^{\circ}$	252 ± 6.7^{b}	324.6 ± 8.0^{bc}
Yield (gm)	$565.7 \pm 11.3^{\rm a}$	518.3 ± 15.0^{b}	477.1 ± 9.9 ^c	442.4 ± 6.7^{d}	532.8 ± 8.9^{ab}	-	_

Means with different superscripts within the same column are significantly (P < 0.05) different

CS calcium sulphate coagulated tofu, ES epsom salt coagulated tofu, LJ lemon juice coagulated tofu, Alum coagulated tofu, TWFM top water of fermented maize coagulated tofu

* On wet weight basis; **On dry weight basis

The mineral composition of the tofu is not significantly different except CS- and ES-coagulated tofu that was significantly (P < 0.05) higher in calcium and magnesium, respectively. The amount of these minerals is significantly higher than that observed value in soymilk (Table 1). This is expected as the salts are composed of calcium and magnesium, respectively.

Visualized by naked eye as the separation of whey occurred, the coagulating speeds of TWFM and LJ were found to be very slow. A time period of more than 10 min was required for the curd formation to occur with these coagulants. The slower the coagulating action of the coagulants, the better yield of tofu produced than rapid acting ones. Yield range lowest (442 g) in alum coagulated tofu and highest (565.7) in CS coagulated tofu (Table 1). A similar observation of better yield for slow acting coagulants was observed by previous researchers [12, 19] who used different concentration levels of calcium and magnesium in coagulating soymilk. Although lemon has a slow coagulating action in the present study, the yield was significantly lower than ES that coagulated at a faster rate than LJ.

Cai et al. [3] earlier correlated high moisture content with higher tofu yield. Tofu with high moisture content appeared smooth while tofu with low moisture content had a coarse texture by visual examination. Yield and quality are affected by several factors, such as variety or cultivar, soybean growth environment [20, 21] and tofu-processing methods [8, 21, 22]. However, because all other factors are the same in the present study, the difference in yield and quality could be attributed to the coagulant which is the only different variable. Sun and Breene [23] reported coagulant as an important factor in the quality of tofu.

The texture of tofu has been reported to be an equally important attribute as nutritional quality that affects the product acceptability [5, 23]. The textural characteristics are the important determinants for consumption. Hardness, brittleness and chewiness of tofu are some textural parameters that ultimately

determine the quality of tofu. Texture profile analysis was therefore carried out to study the effects of different coagulants on the firmness of tofu matrix. Hardness value is defined as the maximum force at the first cycle of compression, or the force necessary to attain a given product deformation (25% of original height) while brittleness is the force required to attain a significant break or fracture [24, 25].

The current study shows a significant difference in the textural characteristics of tofu produced with different coagulants. The hardness, chewiness and brittleness force ranged from 525.6 in LJ to 1,008(g)^c, 1.4 to 4.5 and 1,035 to 3,678(g)^c, respectively, in LJ-coagulated tofu to alum-coagulated tofu (Table 2). Sun and Breene [23] had earlier reported similar differences in textural properties of soymilk tofu coagulated with various protein coagulants. The higher the force required in breaking the tofu, the less brittle the tofu is [4]. This relationship between hardness and brittleness is observed in the present study as alum-coagulated tofu that had the highest hardness force is the least brittle and chewable. The low values observed for LJ coagulated tofu is an indication that the tofu coagulated with LJ is significantly (P < 0.05) softer and more fragile than other tofu coagulated with CS, ES and TWFM. Most Nigerians would have a preference for soft tofu as this is often compared to cow milk curd known as "wara". A typical soft tofu is characterized by a bland taste and fine texture with 84–90% moisture content [10, 24]. Next to alum in hardness, less chewable and less brittle is tofu coagulated with ES. The hardness probably is due to denseness and compact structure of the tofu, an implication that alum has the ability to create a more dense structure by making protein molecules closer due to loss of water during coagulation step. Low holding water capacity has been associated with higher hardness in tofu [4, 25].

There was, however, no significant difference in the brittleness of the control (CS) and TWFM coagulated tofu. Contrary to the result of this study, Prabhakaran

Table 2 Textural properties of tofu from the various coagulants

	Hardness (g) ^c	Chewiness (kg)	Brittleness Force (g) ^c	Colour determination		
				L	а	b
CS ES LJ Alum TWFM	548.0 ± 22.8^{c} 764.2 ± 20.4^{b} 525.6 ± 39.9^{c} 1008.5 ± 24.1^{a} 542.9 ± 20.9^{c}	$\begin{array}{l} 2.4 \pm 0.06^{\rm c} \\ 3.6 \pm 0.07^{\rm b} \\ 1.4 \pm 0.06^{\rm d} \\ 4.5 \pm 0.03^{\rm a} \\ 2.3 \pm 0.08^{\rm c} \end{array}$	$1167 \pm 34.0^{\circ} 1482 \pm 52.9^{\circ} 1035 \pm 40.5^{\circ} 3678 \pm 38.6^{\circ} 1166 \pm 26.7^{\circ} $	$\begin{array}{c} 86.3 \pm 2.4^{a} \\ 86.2 \pm 5.2^{a} \\ 77.8 \pm 3.5^{b} \\ 72.4 \pm 3.9^{c} \\ 84.6 \pm 4.0^{a} \end{array}$	$\begin{array}{c} 0.34 \pm 0.02^{\rm c} \\ 0.21 \pm 0.04^{\rm d} \\ 0.87 \pm 0.0^{\rm b} \\ 1.05 \pm 0.01^{\rm a} \\ 0.32 \pm 0.05^{\rm c} \end{array}$	$\begin{array}{c} 24.0 \pm 0.7^{b} \\ 23.9 \pm 0.9^{b} \\ 27.3 \pm 0.6^{a} \\ 20.3 \pm 0.4^{c} \\ 23.4 \pm 0.5^{b} \end{array}$

Means with different superscripts within the same row are significantly (P < 0.05) different

CS calcium sulphate coagulated tofu, ES epsom salt coagulated tofu, LJ lemon juice coagulated tofu, Alum coagulated tofu, TWFM top water of fermented maize coagulated tofu

et al. [12] previously observed that the hardness and chewiness of magnesium sulfate-coagulated tofu was less than the hardness and chewiness of tofu prepared from calcium sulfate. The differences in textural characteristics as a result of coagulant could be attributed to the higher moisture content of this tofu and probably an occurrence of incomplete precipitation of soy proteins which causes a loose network encompassing many air gaps within it instead of having a compact protein network [22, 25]. A firm tofu with a greater hardness means more work to break down the internal bonding. Tofu with high chewiness, is more difficult to eat. It has also been reported that the coagulant concentration and type of anion may affect the hardness of tofu [26, 27]. Tofu coagulated with TWFM had a textural characteristic (brittleness force, hardness and chewiness similar to CS-coagulated tofu while alumcoagulated tofu significantly differs in textural parameters from calcium sulphate.

With regard to the colour, either white or creamy white colour is the desirable tofu characteristic [5]. Hunter colorimetric readings indicate significant differences in L, a and b value of tofu from the different coagulants. The lightness (L) values were 87.3, 86.2, 77.8, 72.4, and 84.6, redness (a) values are 0.34, 0.21, 0.87, 1.05, and 0.32, yellowness (b) values were 24.0, 23.9, 27.3, 20.3, and 23.4, respectively, for tofu-coagulated with CS (control), ES, LJ, alum and TWFM. Coagulant significantly (P < 0.05) affected the colour properties of tofu in the present study. Tofu coagulated with ES and TWFM are, however, significantly similar in colour to the control tofu. The present result is contrary to the study of Prabhakaran et al. [12] who found no difference in the colour of tofu made from different coagulating agents. Alum- and LJ-coagulated tofu are off the L or creamy white of a desirable colour for tofu.

The result of the sensory evaluation is as presented in Table 3. Basic regular tofu is a white, essentially bland, soft product, closely resembling pressed white milk curd [28]. High sensory scores in Table 3 indicate whitest colour, best flavour, smoothest mouth feel and is most overally accepted. The colour, flavour, mouth feel and overall acceptability ranged, respectively, from 4.0 alum-coagulated tofu to 6.8 CS-coagulated tofu (control), 3.8 in alum-coagulated tofu to 7.6 in LJcoagulated tofu, 4.4 in alum-coagulated tofu to 7.2 in LJ-coagulated tofu and 4.5 in alum-coagulated tofu to 7.0 in LJ-coagulated tofu. Except for the colour, the LJ-coagulated tofu was most preferred while alumcoagulated tofu was the least preferred in all the sensory characteristics. The reason for the low score in colour and flavour, respectively, of LJ-coagulated tofu

 Table 3 Coagulant effect on sensory characteristics of tofu

	Colour	Flavour	Mouth feel	Overall acceptability
CS ES LJ Alum TWFM	$\begin{array}{l} 6.8 \pm 0.06^{a} \\ 6.3 \pm 0.07^{ab} \\ 5.5 \pm 0.03^{c} \\ 4.0 \pm 0.3^{d} \\ 5.8 \pm 0.6^{bc} \end{array}$	$\begin{array}{c} 6.2 \pm 0.03^{b} \\ 4.3 \pm 1.0^{c} \\ 7.6 \pm 0.02^{a} \\ 3.8 \pm 0.05^{d} \\ 6.1 \pm 0.01^{b} \end{array}$	$\begin{array}{l} 6.8 \pm 0.04^{ab} \\ 5.0 \pm 0.06^c \\ 7.2 \pm 0.04^a \\ 4.4 \pm 0.03^d \\ 6.2 \pm 0.08^{ab} \end{array}$	$\begin{array}{l} 6.8 \pm 0.02^{a} \\ 5.7 \pm 0.02^{b} \\ 7.0 \pm 0.04^{a} \\ 4.5 \pm 0.03^{c} \\ 6.7 \pm 0.02^{a} \end{array}$

Means with different superscript within columns are significantly different (P < 0.05)

CS calcium sulphate coagulated tofu, *ES* epsom salt coagulated tofu, *LJ* lemon juice coagulated tofu, *Alum* coagulated tofu, *TWFM* top water of fermented maize coagulated tofu

is probably due to the greenish-yellowish colour and lemon flavour, respectively, impacted in the tofu. Despite a long history of tofu consumption in some parts of the world, the characteristic beany taste of soy products is still being discriminated against by the consumer. In the present study, LJ could have effectively caused the elimination or reduction of the L2 and L3 lipoxygenase isozymes from soybean tofu, thereby making LJ-coagulated tofu more preferred in terms of flavour, mouth feel and overall acceptability. Previous taste tests conducted in Japan showed that soymilk prepared from soybean free L2 and L3 lipoxygenase was preferred by consumers over soymilk prepared from standard varieties [11, 29]. There was however no significant difference (P < 0.05) in the overall acceptability of the control tofu (calcium sulphate), LJ and TWFM tofu. The low sensory score in colour of LJ- and alum-coagulated tofu agree with the result of the instrumental colour analysis in this study. ES-coagulated tofu was preferred in terms of colour to LJ and TWFM tofu.

Conclusion

All the different coagulants had significant effects (P < 0.05) on the quality of the tofu, with the exception of tofu coagulated with CS and TWFM that were not significantly different from one another. Fat was reduced when soybean was processed to milk and finally to tofu. CS- and TWFM-coagulated tofu gave the highest yield while LJ-coagulated tofu has the best textural and sensory characteristics. This study has demonstrated that tofu quality is affected by the type of coagulant used in curding the soymilk. Based on the result of the present study, and considering the availability of the coagulants, TWFC and LJ are recommended in coagulating soymilk for tofu making in the Nigerian community.

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