Effect of coagulant type and concentration on the yield and quality of soy-lupin tofu

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Abstract

Soy-lupin tofu samples were prepared by replacing 30% soybean with lupin flour. Four different coagulants i.e. calcium sulphate, calcium lactate, magnesium sulphate and magnesium chloride were used at three different concentrations (0.3, 0.4 and 0.5% w/v of the "milk") to study their effect on yield and quality improvement. The results revealed that the tofu samples prepared using magnesium sulphate had higher moisture content and fresh yield than those prepared from other coagulants. The L^*, $a^*$and b^* colour coordinates showed no significant differences among the samples. Fat content was affected by the type and concentration of the coagulants. Magnesium sulphate and magnesium chloride at 0.5% level produced tofu with lower fat contents. Protein contents, however, were not affected by type or concentration of coagulant. Texture profile analysis revealed that the hardness and chewiness of samples changed with the type and concentration of the coagulant whereas cohesiveness and springiness were not affected significantly. Sensory evaluation for appearance, colour, flavour, mouthfeel and overall acceptance of the selected samples showed no significant differences. Based on the higher fresh yield magnesium sulphate was found to be a better coagulant for soy-lupin tofu preparation.

Keywords

legumes; lupin; tofu; physicochemical; sensory; texture.

Introduction

Tofu, a very popular food in the Orient particularly Far Eastern countries, is traditionally made by curding soybean milk using different coagulants. It is used as a meat substitute due to its high protein contents with good balance of amino acids and better digestibility (Liu, 1999; Read, 2002). Due to its recognised nutritional benefits, there is an increase of tofu consumption among the Western countries in recent years (Oboh, 2006).

Comparable in protein content (32% compared with 37% in soybean) and functional properties yet lower in price, Australian Sweet Lupin (*Lupinus angustifolius* L.) is proved to be a valuable alternative to soybean in many foods (Jayasena & Quail, 2004; Jayasena *et al.*, 2004). In addition, lupin is lower in fat (6% compared with 18% in soybean) and higher in dietary fibre contents (30% compared with 9% in soybean) that provides a healthier choice to be used as ingredient in many foods (Hall & Johnson, 2004; Jayasena *et al.*, 2009). Compared to soybean, pea and faba bean, lupin has lower antinutritional factors such as trypsin inhibitors that can interfere with digestion, phytic acid which binds to minerals such as calcium and zinc thus reducing the bioavailability, saponins and lectins that can act as gastric irritants (Hudson, 1994; Petterson & Fairbrother, 1996). Research evidence suggests that lupin could be incorporated up to 40% in the raw material of tofu making without any significant influence on physicochemical and sensory qualities but the major concern was lower yield (Jayasena *et al.*, 2010). Protein is the main functional component that determines the quantity and quality of tofu and studies have shown that lupin protein and soy protein have similar functional properties (Jayasena *et al.*, 2004). Coagulation of the protein is the most important step in the tofu-making process as yield and quality of tofu mostly depends on this step. Coagulation occurs due to cross linking of protein molecules in the bean extract with the divalent cations.

Calcium sulphate and 'nigari' (sea water extract) are the most common coagulants used in tofu manufacturing process (Shih *et al.*, 1997). However, recent studies have shown that calcium lactate, magnesium sulphate and magnesium chloride are very effective coagulants for soy tofu preparation (Prabhakaran *et al.*, 2006). The coagulation of soymilk depends on many factors including variety of soybean, soymilk heating temperature and time, pH and coagulant type and concentration (Hou & Chang, 2004). In addition, yield and quality of tofu have been reported to be influenced by coagulants (Cai *et al.*, 1997; Poysa & Woodrow, 2004; Oboh, 2006). As with type of coagulant, the concentration of the coagulant also affects the nature of tofu. The amount of coagulant added in tofu manufacturing is one of the critical points as it determines the product texture, flavour and yield (Wilson, 1995).

In the present study, coagulants such as calcium sulphate, calcium lactate, magnesium sulphate and magnesium chloride have been utilised at different concentrations to find a combination of coagulant type and concentration that could provide a better yield and quality of soy-lupin tofu.

Materials and methods

Materials

Soybean was purchased from the local market (Perth). Australian Sweet Lupin flour was obtained from Irwin Valley Pty Ltd, Australia. Calcium sulphate ($CaSO_4$. $2H_2O$) and magnesium chloride ($MgCl_2$. $6H_2O$) were obtained from Ajax Finechem Pty Ltd Australia. Magnesium sulphate ($MgSO_4$ anhydrous) was obtained from Mallinckrodt Chemicals, USA and calcium lactate ($C_6H_{10}CaO_6$. $5H_2O$) was obtained from VWR International.

Preparation of soy-lupin tofu samples

Soy-lupin tofu samples were prepared by replacing 30% soybean with lupin flour. The ratio of 70:30 soybean: lupin flour was chosen based on the studies by Jayasena *et al.* (2010) which revealed that tofu prepared by replacing up to 40% of soybean with lupin bean (equal to around 30% lupin flour) had a quality equivalent to traditional tofu prepared solely from soybean. Soy-lupin tofu samples were prepared according to the method developed by Jayasena *et al.* (2010).

A sample of 210 g soybean was washed and soaked in 630 ml water (bean: water, 1:3 w/v) overnight for 16 hours. Soaked soybean and 90 g lupin flour was then blended with 3 L water (1:10, dry bean + flour: water) using speed 2 of Breville 5 speed blender (Model BLR 50) for 2 min. The slurry was filtered through cheese cloth squeezed by hand pressure to obtain the extract hereafter called "milk". The milk was boiled for 5 minutes with occasional stirring followed by cooling to 78 °C. The coagulant solution/suspension made by dissolving/mixing 9.0, 12.0 and 15.0 g (0.3, 0.4 and 0.5% w/v of the milk) calcium sulphate, calcium lactate, magnesium sulphate or magnesium chloride in 20 ml distilled water was added and stirred properly. The coagulants were soluble in 20 ml water at room temperature except calcium sulphate. To increase the solubility of calcium sulphate, the suspension was prepared in 20 ml hot water. The mixture was let stand for 30 minutes for coagulation. The coagulated curd was transferred to a mould (22 cm x 15 cm x 12.5 cm), drained off the whey fraction gravimetrically for about 5 minutes and pressed for 45 minutes using a weight of 6 kg. The soy-lupin tofu was removed from the cheese cloth and weighed immediately. Samples were taken for physicochemical analysis before storing in distilled water at 5±1 °C. Samples were prepared in triplicate for each treatment. As the main objective of the experiment was to study the effect of type and concentration of different coagulants on the yield of soy-lupin tofu, the sample prepared by using 0.3% (w/v of milk) calcium sulphate, the most commonly used coagulant type and concentration, was taken as a control.

Physicochemical analysis

Yield

The tofu yield was calculated on the basis of the weight of pressed tofu obtained from 70:30 (w/w) soybean: lupin flour and expressed as g/100g raw material (bean+ flour). Dry tofu yield was calculated by excluding the moisture content.

Moisture, protein and fat contents

AOAC (2000) methods were used to determine moisture (method 925.10), protein (method 950.36) and fat (method 963.15) contents of the tofu samples.

Instrumental colour measurement

Tofu colour was measured using Minolta spectrophotometer CM-508i (Minolta Co. Ltd. Japan) and expressed as L^* (lightness), a^* ($+a^*$ = redness, $-a^*$ = greenness) and b^* ($+b^*$ = yellowness, $-b^*$ = blueness) colour coordinates according to the methods specified by the equipment manual. The instrument was equipped with a pulsed xenon arc lamp as light source, a silicon photodiode array detector and has the illumination/measurement area of Ø11mm. The instrument was calibrated using the white-coloured disc (L* = +98.82, a* = –0.07 and b* = –0.45) supplied with the instrument prior to the analysis. Three readings were recorded for each sample.

Texture profile analysis (TPA)

Texture profile of the tofu samples was determined using TA-XT2*i* texture analyser (Stable Micro System, Godalming, UK). A sample was obtained from the central portion of raw tofu and cut into cubes (1.5 cm x 1.5 cm x 1.5 cm). A test speed of 1.0 mm/s and a 15 mm diameter cylindrical probe was used for the analysis. The probe compressed the sample twice to 25% of its original height by using a load cell of 5 kg. The TPA settings used were:

Pre-test speed	: 2.0mm/s
Test speed	: 1.0mm/s
Post-test speed	: 1.0mm/s
Distance	: 25%
Time between bite	: 3s

Three replicate tests were carried out for each tofu sample. The TPA curves were recorded and used to calculate hardness, cohesiveness, springiness and chewiness (Bourne, 2002).

Sensory evaluation

A total of 53 semi trained panellists from the Curtin University's staff and students participated in the sensory evaluation. Four soy-lupin tofu samples that included control sample and three other high yielding samples (one out of each coagulant type) were selected for sensory evaluation studies. Samples were cut into rectangular pieces (4 cm x 2 cm x 2 cm) and deep fried for 4 min in commercially available vegetable oil. Tofu samples were placed in plastic cups which were labelled with 3 digits random numbers. Panellists were also served with crackers and water for the purpose to cleanse the palate between evaluations. A separate set of raw tofu samples (without deep-frying) having different code numbers was used for colour evaluation. The sensory attributes evaluated were appearance, colour, flavour, mouthfeel (oral texture) and overall acceptance. 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, 9 = like extremely) was used for sensory evaluation.

Statistical analysis

The data collected for each of the physicochemical properties of different tofu samples were analysed by analysis of variance (ANOVA) using SPSS for Window version 17.0. The differences of means between groups were compared by using Tukey's honestly significant difference (HSD) test. Since the results obtained from sensory evaluation were non-parametric, the statistical analysis was conducted using Kruskal-Wallis test. Statistical significance was established at $p \leq 0.05$.

Results and discussion

Moisture content and yield (wet and dry)

The moisture content remained same for different coagulant types and concentrations except for 0.5% magnesium sulphate that had a significantly higher value than the control (Table 1). Coagulant concentration had shown no effect on moisture contents within a coagulant. However, when the data were analysed on the basis of coagulant type (accumulating the effect of all three concentrations) the samples prepared by using magnesium sulphate had significantly ($p \leq 0.05$) higher moisture contents than the other coagulants (Table 2).

The fresh tofu yield ranged from 168-198 g/100g raw material for different coagulant type and concentration combinations which was lower than the values reported in the previous studies (Jayasena *et al.*, 2010). The difference could be due to the use of different soybean samples/varieties. Soybean was purchased from the local retail market at about one year interval and there was no information available on the variety or age of the beans. Whereas soybean variety is one of the major factors that affects substantially on the tofu yield (Hou & Chang, 2004). Tofu yield was within the range of 150-200 g/100g soybean using Korean and Canadian soybean cultivars as reported by Abd Karim *et al.* (1999), No and Meyers (2004), Noh *et al.* (2005), Yoon and Kim (2007). In contrast Mujoo *et al.* (2003) reported tofu yield that ranged from 269 to 343 g/100g bean for seven American soybean varieties.

Concentration of coagulants ranging from 0.3 to 0.5% (w/v of milk) had no significant effect on fresh tofu yield. Similar results have been reported by earlier researchers (Prabhakaran *et al.*, 2006) that there was no effect on soy tofu yield when 0.4 or 0.5% (w/w of soy milk) of calcium sulphate, calcium chloride, magnesium sulphate, calcium lactate or calcium acetate were used.

The magnesium sulphate delivered the highest fresh (wet) tofu yield among coagulants (Table 2). It might be due to the higher moisture content of the samples prepared by using magnesium sulphate since tofu yield and moisture contents are highly correlated (Cai *et al.*, 1997). This is probably due to the difference in gel network within the particles which is influenced by different anions and their ionic strengths towards the water holding capacity of protein gels (Obatolu, 2008).

The dry yield, on the other hand, demonstrated no significant difference for different coagulants and their concentration (Table 1). This confirmed that the higher fresh yield delivered by magnesium sulphate was due to higher moisture holding capacity of those samples.

Colour

Table 3 shows the values of $L*$, $a*$ and $b*$ of soy-lupin tofu prepared by using different coagulant types at different concentrations. The coagulant type and concentration had no significant effect on $L*$, $a*$ or $b*$ values of the samples.

Having a greater L^* value is a favourable characteristics as consumer prefer lighter or whiter tofu (Hou & Chang, 2004) and good coagulant should produce a tofu with higher L^* value (Tay *et al.*, 2006). The different coagulating agents used at different concentrations in this study were found to have no significant effect ($p \leq 0.05$) on the colour of tofu. All of the samples had a creamy white colour which is a desirable characteristic for good quality tofu. The results are in agreement to those of Prabhakaran *et al.* (2006) who found no difference in the colour of soybean tofu samples prepared by using 0.4-0.5% (w/v of soy milk) of calcium sulphate, magnesium sulphate, magnesium chloride, calcium acetate and calcium lactate. However the results are contrary to the study of Obatolu (2008) which revealed a significant difference in L^*, a^* and b^* colour coordinates of tofu samples prepared from Epsom salt, alum and lemon juice. The difference in tofu colour in case of Obatolu (2008) might be due to the use of such coagulants that had their own colour such as lemon juice that imparted a change in the tofu colour.

Fat and protein contents

Coagulant concentration had an effect on the fat content of tofu but it depended upon the type of coagulant (Table 4). There was no effect of concentration on the fat contents of tofu samples in case of calcium salts (calcium sulphate and calcium lactate) whereas a significant decrease in fat content with increase in coagulant concentration was determined in case of magnesium salts (magnesium sulphate and magnesium chloride) (Table 4). The results are in agreement to those of Cai and Chang (1998) who demonstrated a decrease in tofu fat content with an increase in coagulant concentration. Although lower fat content may be preferable considering health benefits, higher quantity of coagulant (0.5% w/v magnesium sulphate or magnesium chloride of milk) is required to achieve the lower fat value.

Analysing the cumulative effect, samples prepared by using calcium lactate had the highest and those prepared by using magnesium sulphate had the lowest fat contents (Table 5). This was most probably be due to the release of fats during the tofu making process as some coagulants can considerably decrease the fat binding capacity of the protein network formed during curding (Obatolu, 2008).

The coagulant concentration had no significant effect on protein contents (Table 4). The results are in agreement to previous studies (Cai & Chang, 1998). The protein content of lupin containing tofu might be able to improve by optimizing the processing conditions especially protein coagulation conditions. Zee *et al*. (1988) showed that faba bean having 24% less protein contents than soybean produced a tofu with 50% more protein content than soy tofu. This was mainly due to the reduction in protein loss during faba bean tofu preparation.

Textural properties

The textural properties of tofu play a critical role in tofu consumer acceptability (Sun & Breene, 1991; Hou & Chang, 2004). The textural properties of tofu samples prepared using different types of coagulant at different concentrations demonstrated a variation in their textural properties (Table 6). The hardness and chewiness of tofu samples demonstrated substantial changes with different coagulants and their concentrations. The lowest hardness and chewiness was demonstrated by the sample prepared by using 0.5% magnesium sulphate. There was no significant difference in hardness among the samples prepared from calcium sulphate and calcium lactate. Similarly, both cohesiveness and springiness were not significantly affected ($p \leq 0.05$) by the type and the concentration of coagulants.

In general soy-lupin tofu samples prepared by using calcium salts had higher hardness than those prepared from magnesium salts (Table 7). It may probably be caused by the ability of these salts to create a network structure with the protein molecules coming closer due to the loss of water during coagulation (Obatolu, 2008).

Among the coagulants, tofu samples made by using magnesium salts (magnesium sulphate and magnesium chloride) had lower hardness and chewiness than the samples prepared by using calcium salts *i.e.* calcium sulphate and calcium acetate (Table 7). Previous researchers also found that the tofu made by using 0.4-0.5% (w/v of soymilk) magnesium sulphate had lower hardness than those prepared with the same concentrations of calcium sulphate, calcium acetate and calcium lactate (Prabhakaran *et al.*, 2006). This may have link with the higher moisture content of the samples prepared by using magnesium sulphate (Table 1). According to Wang and Hesseltine (1982), cross-linking between protein molecules along with the presence of calcium ions are required for soy protein coagulation. Magnesium ions can also be used instead of calcium ions, since this divalent cation can form cross-linking between protein molecules. However, the sites of cross-linking in the protein molecules may be different for both calcium and magnesium causing the latter to form a loose network encompassing many air gaps within the network. This might be a reason why magnesium sulfate is rarely used alone as a coagulant for firm tofu preparation. It is commonly used along with other coagulants such as magnesium chloride and calcium chloride. ‘‘Modified nigari’’ is a popular name used for such type of coagulant mix (Hou *et al.*, 1997).

Sensory evaluation

The results for sensory evaluation are presented in Table 8. The scores for colour and appearance, which are the first deciding factors that determine the acceptance or rejection of a product, reflected that the tofu samples prepared by using different coagulants had a good acceptability. Acceptance by colour and appearance were not affected by the use of different coagulants. All samples had similar creamy white colour which is the acceptable colour for tofu (Hou & Chang, 2004).

Flavour, a combination of both taste and odour, was a concern in soy-lupin tofu as lupin has a natural beany flavour. However, the results indicated that all of the samples had acceptable flavour scores (Table 8). Besides lupin itself, the concentration and type of coagulant used could determine the tofu flavour (Kao *et al.*, 2003). However in our study different coagulants had no significant effect on the flavour of soy-lupin tofu. It is possible that deep frying could have masked the beany flavour of the product.

Sensory acceptability of texture, which is perceived by touching and/or mouth feel, is an important determinant of consumer acceptability of tofu (Obatolu, 2008). The results showed that calcium salts had a greater coagulating power than magnesium salts causing them to produce a better texture profile in terms of hardness and chewiness than that produced by magnesium salts (Table 7). However sensory score for mouthfeel (oral texture) showed no significant difference among the samples prepared by using different coagulants.

The evaluation of overall acceptability is important in determining how well a product is accepted by consumers. Although a non-specific indication of the reasons, it is a good indication of the potential consumer demand of the product. The overall acceptability of soy-lupin tofu samples prepared by using different coagulants was not significantly different ($p \leq 0.05$) to the control tofu sample. All samples had similar scores for overall acceptability. All samples received ≥ 6 out of 9 scores for overall acceptability.

Conclusions

This study contributes to a wider variation of coagulant options such as calcium lactate, magnesium sulphate and magnesium chloride to be used in the production of lupin incorporated tofu. However, use of magnesium sulphate may be preferred as it produced a higher fresh (wet) yield of lupin incorporated tofu. The market value of tofu depends on the yield and quality of fresh produce. Since fresh soy-lupin tofu prepared by using magnesium sulphate produced a softer product a combination of coagulants may be applied to get a better texture. However as tofu is mostly consumed after deep frying which changes the texture of the product and masks minor differences, a little softness of soy-lupin tofu prepared by using magnesium sulphate may not effect on its acceptability and marketing.

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Table 1 Moisture content and yield of soy-lupin tofu samples prepared by using different coagulant types and concentrations

Coagulant		Moisture (%)	Tofu Yield (g/100g raw material)	
Type	Concentration (% w/v of milk)		Fresh	Dry
Calcium sulphate	0.3 (control)	75.4 ± 1.5^{c}	$^{A}172 \pm 6^{c}$	42.0 ± 1.1^{a}
	0.4	75.6 ± 0.4^{bc}	$^{A}173 \pm 6^{c}$	42.1 ± 0.8^{a}
	0.5	76.6 ± 0.6^{bc}	$^{A}178 \pm 7^{bc}$	41.4 ± 1.5^{a}
Calcium lactate	0.3	77.1 ± 1.1^{abc}	$^{A}180 \pm 5^{abc}$	41.0 ± 0.8^{a}
	0.4	76.4 ± 0.5^{bc}	$^{A}175 \pm 5^{bc}$	41.3 ± 0.5^{a}
	0.5	75.3 ± 0.7^{c}	$^{A}168 \pm 4^{c}$	41.5 ± 1.0^{a}
Magnesium sulphate	0.3	78.0 ± 0.6^{ab}	$^{A}186 \pm 8^{abc}$	40.8 ± 1.5^{a}
	0.4	77.7 ± 0.2^{abc}	$^{A}192 \pm 3^{ab}$	42.8 ± 0.4^{a}
	0.5	79.5 ± 0.7^{a}	$^{A}198 \pm 5^{a}$	40.6 ± 1.6^{a}
Magnesium chloride	0.3	76.4 ± 0.3^{bc}	$^{A}179 \pm 3^{bc}$	42.0 ± 0.2^{a}
	0.4	77.4 ± 0.4^{abc}	$^{A}182 \pm 7^{abc}$	41.0 ± 0.8^{a}
	0.5	76.8 ± 0.8^{bc}	$^{A}180 \pm 6^{abc}$	41.8 ± 0.6^{a}

Means (± SEM; n = 3) with different superscripts (in lower case) within the same column are significantly different ($p \leq 0.05$). Means with similar superscripts (in upper case) in the fresh yield column show a non significant difference ($p \leq 0.05$) within a coagulant type.

Table 2 Effect of different coagulants (cumulative effect) on the moisture content and yield of soy-lupin tofu samples

Coagulant	Moisture (%)	Wet yield (g/100g raw material)	Dry yield (g/100g raw material)
Calcium sulphate	75.9 ± 1.5^{b}	173.8 ± 9.7^{b}	41.8 ± 1.1^{a}
Calcium lactate	76.3 ± 1.5^{b}	174.5 ± 8.9^{b}	41.3 ± 1.2^{a}
Magnesium sulphate	78.4 ± 1.1^{a}	191.9 ± 9.2^{a}	41.4 ± 2.2^{a}
Magnesium chloride	76.8 ± 0.9^{b}	180.4 ± 8.4^{b}	41.6 ± 0.9^{a}

Means (± SEM; n = 3) with different superscripts (in lower case) within the same column are significantly different ($p \leq 0.05$).

Table 3 Colour analysis of soy-lupin tofu samples prepared from different coagulant types and concentrations

Coagulant		Colour coordinates		
Type	Concentration (% w/v of milk)	L^*	a^*	b^*
Calcium sulphate	0.3 (control)	85.8 ± 0.3 [a]	0.24 ± 0.12 [a]	16.7 ± 0.1 [a]
	0.4	86.5 ± 0.1 [a]	0.26 ± 0.11 [a]	16.4 ± 0.4 [a]
	0.5	86.6 ± 0.1 [a]	0.13 ± 0.05 [a]	16.9 ± 0.3 [a]
Calcium lactate	0.3	87.0 ± 0.4 [a]	0.16 ± 0.02 [a]	17.1 ± 0.3 [a]
	0.4	86.7 ± 0.1 [a]	0.14 ± 0.03 [a]	16.8 ± 0.3 [a]
	0.5	86.3 ± 0.4 [a]	0.11 ± 0.04 [a]	17.0 ± 0.3 [a]
Magnesium sulphate	0.3	87.1 ± 0.1 [a]	0.20 ± 0.08 [a]	17.0 ± 0.2 [a]
	0.4	87.3 ± 0.1 [a]	0.19 ± 0.03 [a]	16.7 ± 0.1 [a]
	0.5	87.5 ± 0.2 [a]	0.21 ± 0.09 [a]	16.7 ± 0.2 [a]
Magnesium chloride	0.3	86.4 ± 0.5 [a]	0.21 ± 0.08 [a]	17.1 ± 0.5 [a]
	0.4	87.1 ± 0.2 [a]	0.22 ± 0.10 [a]	17.2 ± 0.4 [a]
	0.5	87.0 ± 0.2 [a]	0.19 ± 0.08 [a]	17.1 ± 0.3 [a]

Means (± SEM; n = 3) with similar superscripts in a column are not significantly different ($p \leq 0.05$).

Table 4 Fat and protein contents of soy-lupin tofu samples from various coagulant types and concentrations

Coagulant		Fat	Protein
Type	Concentration (% w/v of milk)	(g/100g db)	(g/100g db)
Calcium sulphate	0.3 (control)	^{A}23.8 ± 0.9^{ab}	55.0 ± 0.5^{a}
	0.4	^{A}23.4 ± 0.7^{ab}	53.8 ± 1.1^{a}
	0.5	^{A}22.9 ± 0.9^{ab}	53.2 ± 0.5^{a}
Calcium lactate	0.3	^{A}25.6 ± 0.4^{a}	55.3 ± 0.4^{a}
	0.4	^{A}24.3 ± 0.7^{a}	54.9 ± 0.3^{a}
	0.5	^{A}24.2 ± 1.0^{a}	56.5 ± 1.2^{a}
Magnesium sulphate	0.3	^{A}22.9 ± 0.4^{ab}	55.4 ± 0.4^{a}
	0.4	^{B}20.1 ± 1.2^{bc}	55.3 ± 0.6^{a}
	0.5	^{C}16.8 ± 0.6^{c}	54.9 ± 0.8^{a}
Magnesium chloride	0.3	^{A}26.0 ± 0.3^{a}	54.8 ± 1.1^{a}
	0.4	^{B}22.4 ± 1.4^{ab}	55.3 ± 0.2^{a}
	0.5	^{C}17.9 ± 0.4^{c}	55.8 ± 0.2^{a}

Means (± SEM; n = 3) with different superscripts (in lower case) within the same column are significantly different ($p \leq 0.05$). Means with different superscripts (in upper case) in the fat content column show a significant difference ($p \leq 0.05$) within a coagulant type.

Table 5 Effect of coagulant type (cumulative effect) on the fat and protein content of soy-lupin tofu samples

Coagulant	Fat (g/100g db)	Protein (g/100g db)
Calcium sulphate	23.4 ± 1.3^{b}	54.0 ± 0.7^{a}
Calcium lactate	24.7 ± 1.2^{a}	55.6 ± 0.9^{a}
Magnesium sulphate	20.0 ± 2.9^{c}	55.2 ± 0.8^{a}
Magnesium chloride	22.1 ± 3.7^{b}	55.3 ± 0.5^{a}

Means (± SEM; n = 3) with different superscripts within the same column are significantly different ($p \leq 0.05$).

Table 6 Textural properties of soy-lupin tofu samples made from different coagulant types and concentrations

Coagulant		Hardness (g)	Cohesiveness	Springiness (cm)	Chewiness (g.cm)
Type	Concentration (% w/v of milk)				
Calcium sulphate	0.3 (control)	348.1 ± 56.2^{a}	0.81± 0.02^{a}	0.88 ± 0.03^{a}	246.1 ± 37.0^{a}
	0.4	310.9 ± 12.7^{ab}	0.80 ± 0.01^{a}	0.88 ± 0.02^{a}	218.7 ± 8.3^{ab}
	0.5	251.5 ± 14.3^{abc}	0.80 ± 0.01^{a}	0.87 ± 0.04^{a}	173.5 ± 7.7^{bcd}
Calcium lactate	0.3	255.4 ± 10.6^{abc}	0.81 ± 0.03^{a}	0.85 ± 0.04^{a}	175.4 ± 5.5^{abcd}
	0.4	268.3 ± 4.9^{abc}	0.80 ± 0.01^{a}	0.88 ± 0.03^{a}	187.9 ± 2.1^{abcd}
	0.5	307.9 ± 20.9^{ab}	0.80 ± 0.01^{a}	0.87 ± 0.03^{a}	214.1 ± 17.0^{abc}
Magnesium sulphate	0.3	175.9 ± 10.3^{cd}	0.81 ± 0.01^{a}	0.91 ± 0.05^{a}	128.2 ± 3.6^{de}
	0.4	201.5 ± 5.2^{cd}	0.80 ± 0.01^{a}	0.86 ± 0.04^{a}	147.4 ± 6.6^{cde}
	0.5	133.7 ± 16.5^{d}	0.79 ± 0.01^{a}	0.86 ± 0.04^{a}	91.1 ± 11.9^{d}
Magnesium chloride	0.3	194.0 ± 15.4^{cd}	0.80 ± 0.01^{a}	0.90 ± 0.03^{a}	133.3 ± 12.3^{de}
	0.4	235.7 ± 15.0^{bcd}	0.80 ± 0.01^{a}	0.84 ± 0.03^{a}	167.1 ± 11.7^{bcd}
	0.5	207.9 ± 0.4^{bcd}	0.80 ± 0.01^{a}	0.88 ± 0.04^{a}	139.4 ± 6.4^{de}

Means (± SEM; n = 3) with different superscripts within the same column are significantly different ($p \leq 0.05$).

Table 7 Effect of different coagulants (cumulative effect) on the textural properties of soy-lupin tofu samples

Coagulant	Hardness (g)	Cohesiveness	Springiness (s)	Chewiness (s)
Calcium sulphate	303.4±66.4^{a}	0.80±0.01^{a}	0.88±0.03^{a}	278.3±67.3^{a}
Calcium lactate	277.3±31.6^{a}	0.80±0.02^{a}	0.87±0.03^{a}	256.7±30.6^{a}
Magnesium sulphate	188.5±18.8^{b}	0.81±0.01^{a}	0.91±0.04^{a}	167.7±18.0^{b}
Magnesium chloride	192.8±44.2^{b}	0.80±0.01^{a}	0.86±0.04^{a}	177.4±39.8^{b}

Means (± SEM; n = 3) with different superscripts within the same column are significantly different ($p \leq 0.05$).

Table 8 Sensory evaluation of soy-lupin tofu prepared by using different coagulant types and concentrations

Coagulant		Appearance	Colour	Flavour	Mouthfeel	Overall acceptability
Type	Concentration (%w/v of milk)					
Calcium sulphate	0.3% (control)	6.1 ± 0.2[a]	6.4 ± 0.2[a]	5.9 ± 0.2[a]	5.9 ± 0.2[a]	6.2 ± 0.2[a]
Calcium lactate	0.3%	6.2 ± 0.2[a]	6.3 ± 0.2[a]	6.3 ± 0.2[a]	5.8 ± 0.2[a]	6.3 ± 0.2[a]
Magnesium sulphate	0.5%	6.2 ± 0.2[a]	6.3 ± 0.1[a]	5.9 ± 0.2[a]	5.7 ± 0.2[a]	6.1 ± 0.2[a]
Magnesium chloride	0.4%	6.3 ± 0.1[a]	6.3 ± 0.2[a]	5.8 ± 0.2[a]	5.6 ± 0.2[a]	6.0 ± 0.2[a]

Means (± SEM; n = 53) with different superscripts within the same column are significantly different ($p \leq 0.05$).