CHAPTER 6
ADDITION OF GELATIN ENHANCED GELATION OF CORN MILK YOGURT

6.1 INTRODUCTION

Appearance and physical characteristics are important quality parameters of yogurt. Good quality yogurt should be thick and smooth with low syneresis. (Amatayakul *et al.*, 2006a, Lucey, 2002; Shaker *et al.*, 2000). The spontaneous syneresis is related to instability of the gel network (i.e. large-scale rearrangements) resulting in the loss of the ability to entrap all the serum phase (Lucey, 2002). The natural corn milk yogurt still has high syneresis. Then, this chapter was aimed to improve the physicochemical properties of the corn milk yogurt.

Texture is a prime characteristic of yogurt quality and addition of a stabilizer, functioning as a gelling agent or thickener, such as gelatin or other hydrocolloids has been shown to provide good stability and desirable texture (Duboc and Mollet, 2001; Kumar and Mishra, 2004; Sodini *et al.*, 2004), since they impart good resistance to syneresis and a smooth sensation in the mouth (Amatayakul *et al.*, 2006a; El-Sayed *et al.*, 2002; Fiszman *et al.*, 1999; Lal *et al.*, 2006), by binding with water to reduce water flow in the matrix space. Some may interact with protein in the food matrix and as a result further increase hydration behavior (Duboc and Mollet, 2001; Tamime and Robinson, 1999).

Gelatin contains 18 amino acids linked together in a partially ordered fashion. The electrically charged groups and their ionization constant are useful for elucidating the stabilization of the structure and the nature of the reaction of gelation with other substances. Gelatin contains about 1% of sugars, which are galactose, glucose, mannose, lactose and xylose (Djagny *et al.*, 2001; Ledward, 2000; Lopes da Silva and Rao, 2006; Poppe, 1997).

Gelatin has long been used in the food industry because of its unique physical properties rather than its nutritional value as a protein. Gelatin is used to improve the texture of yogurt, without affecting the characteristic tasted of the product. The hardness of the finished product depends mainly on the amount of gelatin added. Gelatin is used mainly to reduce syneresis (Fiszman and Salvador, 1999; Poppe, 1997). Modler and Kalab (1983) reported that at a 0.5% concentration, gelatin efficiently promoted immobilization of the aqueous phase in the yogurt network, and significantly reducing susceptibility to syneresis. Kumar and Mishra (2004) studied 3 types of stabilizers, which were gelatin, pectin and sodium alginate at 3 different levels of 0.2, 0.4 and 0.6% (w/w) together with a sample without any addition of stabilizer as a control. The results of the study indicated that all types of the stabilizers affected the syneresis of the mango soy fortified set yogurt (MSFY) samples. The syneresis decreased as the levels of stabilizers increased. However, among different types of stabilizers, the MSFY samples added with gelatin showed the lowest amount of syneresis. The hardness, cohesiveness and adhesiveness of the yogurt increased up to 0.4% of gelatin. The uses of 0.1 and 0.5% (w/w) gelatin as a stabilizer were also reported for corn milk yogurt (Prasertcheeva, 2003) and for yogurt-like soybean product (Cheng *et al.*, 1990), respectively.

Other hydrocolloids including low methoxyl pectin and sodium alginate are extensive used in stabilization of low pH food products such as acidified milk drinks and yogurt. The stabilization effects of low methoxyl pectin and sodium alginate are related to the interaction with calcium in the egg-box model of gelation (Draget *et al.*, 2006; Lopes da Silva and Rao, 2006; May, 2000; Nussinovitch, 1997; Onsøyen, 1997). Although low methoxyl pectin and sodium alginate are quite popular in food processing, they required calcium for gelling. The corn milk naturally contained only 5.78$\pm$0.01 mg/100 g (Appendix B-1), which is not enough for gelling. For this reason, gelatin that is not required calcium for the gel setting was used. The objectives of this study were to examine concentrations of gelatin on starter culture counts, acidity, syneresis, textural characteristics, rheological properties and microstructure of corn milk yogurt.

6.2 MATERIAL AND METHODS

6.2.1 Sweet corn milk

The sweet corn used in this study was an ATS-5. It was harvested on the 23rd day after silking of the corn plant. The sweet corn was purchased from the same place and duration time as section 3.2.1. The preparation of corn milk solution and storage condition were followed the method in section 2.2.1.

6.2.2 Starter cultures preparation

S. thermophilus and *L. delbrueckii* subsp. *bulgaricus* were prepared using the same method as previous experiment (section 3.2.2 and 3.2.3).

6.2.3 Corn milk yogurt preparation

Distilled water was added to the extracted corn milk in a ratio of 1:2, corn milk to distilled water. The milk was then heated at 90°C prior to fortification with 2% (w/v) lactose (Fonterra, New Zealand) and 4% (w/v) sodium caseinate (BBA, France). The used sodium caseinate contained 95.14$\pm$0.28 mg calcium per 100 g (Appendix B-2). Subsequently, gelatin (Bloom 246 g, Gelita, New Zealand) was added to the mixture at concentrations of 0, 0.2, 0.4 and 0.6% (w/v). The mixture was stirred for 5 min, following by heating at 95°C for 5 min (Raphaelides and Gioldasi, 2005) then cooled down to 40°C. Consequently 2% (v/v) of yogurt starter culture composed of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* at a ratio of 1:1 was inoculated. The inoculum was poured into 100 ml UV-sterilized plastic cups and incubated at 40°C for 4 h until a pH 4.4-4.6 was reached.

6.2.4 Microbiological determination

The corn milk yogurt samples were subjected to microbiological analysis mainly for the viable numbers of yogurt starter cultures in the samples. The methods for enumeration of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* were followed the section 3.2.7.

6.2.5 Analysis of acidity and syneresis

Total acidity of the corn milk yogurt was measured according to AOAC methods no. 947.05 (AOAC, 2000). The determination of syneresis was using the methods as described in section 4.2.7.2.

6.2.6 TPA

The textural properties of corn milk yogurt were measured following the same method as described in section 4.2.8.

6.2.7 Microstructure

Microstructure of corn milk yogurt was determined using the same method as section 4.2.9.

6.2.8 Rheological measurement

Oscillatory testing were performed using a control stress Rheometer AR 2000 (TA Instruments-Waters, Inc., New Castle, DE USA), using a 60 mm 2° steel cones and plate system (truncation 54 μm, TA Instruments Ltd.) with a sample gap of 900 to 1,000 μm. (Afonso and Maia, 1999). Excess sample was trimmed off prior to the analysis. Initially, a strain sweep, at a constant frequency of 1 Hz, with a displacement ranging from 0.0001 to 0.1 rad, was carried out, so that the linear viscoelastic region of the samples could be determined. The measuring system was thermostatted at 10°C, all experiments were done in triplicate.

6.2.9 Statistical analysis

Statistical analysis was performed using a SPSS program version 10.0.1. All data were determined for an Analysis of Variance using a Completely Randomized Design. If the F value from the Analysis of Variance was significant, LSD was then used to determine differences among treatment means.

6.3 RESULTS AND DISCUSSION

6.3.1 Starter culture counts and total acidity

At the various levels of added gelatin, the amounts of starter culture grown in the corn milk yogurt were in the range 11.70-11.88 log CFU/ml and 9.13-9.50 log CFU/ml for *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus*, respectively. In Figure 6.1 it could be seen that an increase in concentration of gelatin, significantly increased product acidity ($P<0.05$) although at gelatin concentrations of 0.4% and 0.6% (w/v), the acidities were similar. These results agreed with those Kumar and Mishra (2004) who found that the acidity of a mango soymilk set yogurt increased with increasing levels of added gelatin.

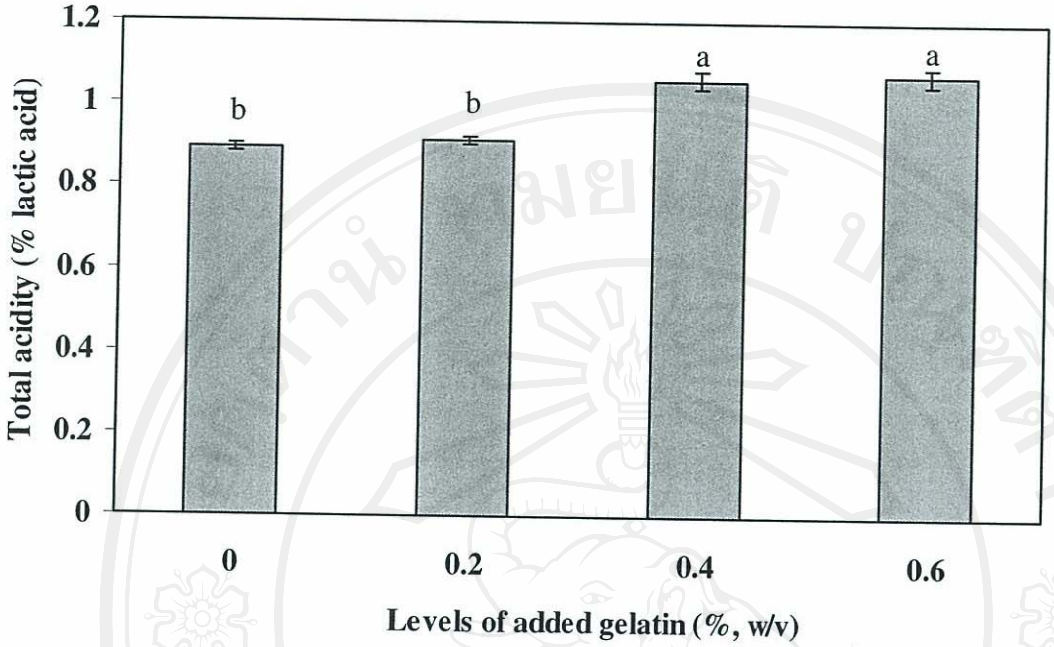

Figure 6.1 Total acidity of corn milk yogurt with different concentrations of gelatin addition. Bars with different superscript were significantly different (P<0.05).

6.3.2 Syneresis

Syneresis of the corn milk yogurt was affected by the addition of gelatin as shown in Figure 6.2. Increased levels of gelatin significantly reduced the extent of syneresis (P<0.05). This might be due to effective immobilization of the aqueous phase by the gelatin in the yogurt network which thus significantly reduced the susceptibility to syneresis (Fiszman *et al*., 1999; Molder and Kalab, 1983; Keogh and O'Kennedy, 1998). It was worth noting that large amounts of gelatin would induce the formation of a gel network with the sodium caseinate in the yogurt mixture (Lal *et al*., 2006).

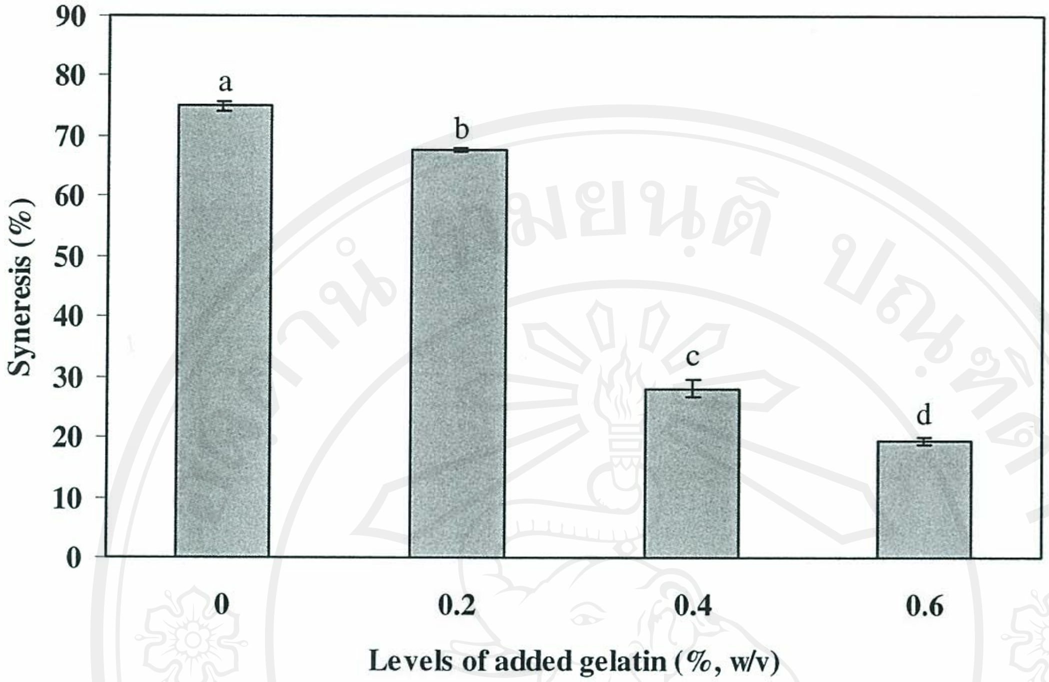

Figure 6.2 Syneresis of corn milk yogurt with different concentrations of gelatin addition. Bars with different superscript were significantly different (P<0.05).

6.3.3 Textural characteristics

Figure 6.3 showed the effect of gelatin addition on hardness, adhesiveness and springiness, nearly all of these parameters significantly increased with increasing gelatin concentration (P<0.05). However to high a concentration of a stabilizer, such as gelatin (0.6%, w/v) could impair the palatability of a natural yogurt gel (Lucey, 2004). Therefore, a medium concentration 0.4% (w/v) of gelatin could be appropriate to ensure good textural quality.

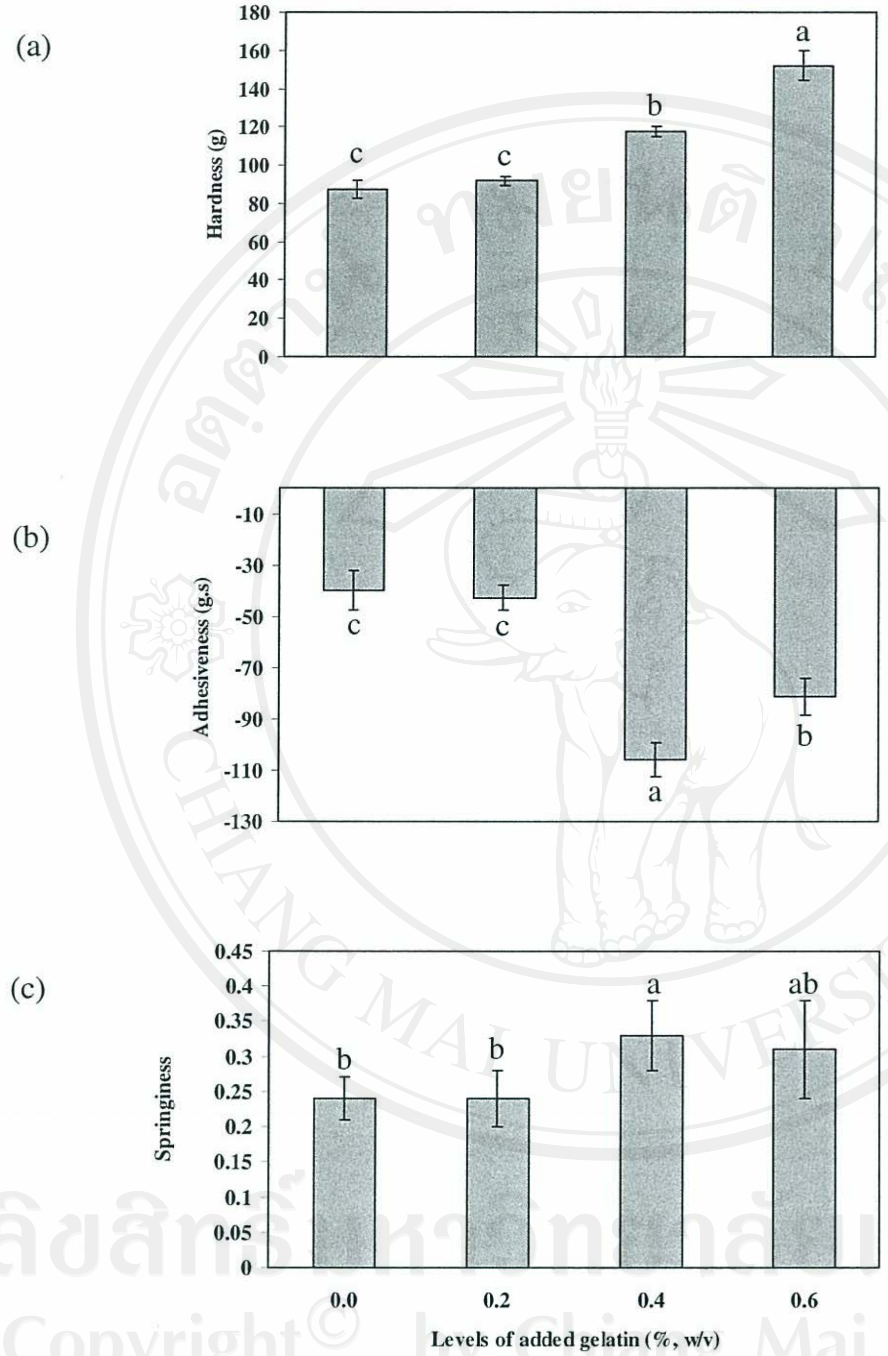

Figure 6.3 TPA of corn milk yogurt with different concentrations of gelatin addition. Bars with different superscript were significantly different (P<0.05).

6.3.4 Microstructure

Figure 6.4 (a)-(c) showed the microstructures of corn milk yogurts as affected by gelatin concentration. At higher concentrations of gelatin (Figure 6.4 (c)) the micrographs showed a dense, highly branched-structure and homogeneous spongy-like interior with few air cells. At the intermediate concerns of gelatin (Figure 6.4 (b)) the structure was less dense and less branched and the spongy-like interior contains more air cell. The control sample (no added gelatin) showed the most open structure (Figure 6.4 (a)). These results agree with those of Fiszman *et al.* (1999) who investigated the effect of gelatin addition on the microstructure of acidic milk gels and yogurt. They found that the smooth bridge of gelatin with a double network structure seemed to be located inside the casein micelles, which could retain the aqueous phase more efficiently, thus reducing syneresis.

(a)

(b)

(c)

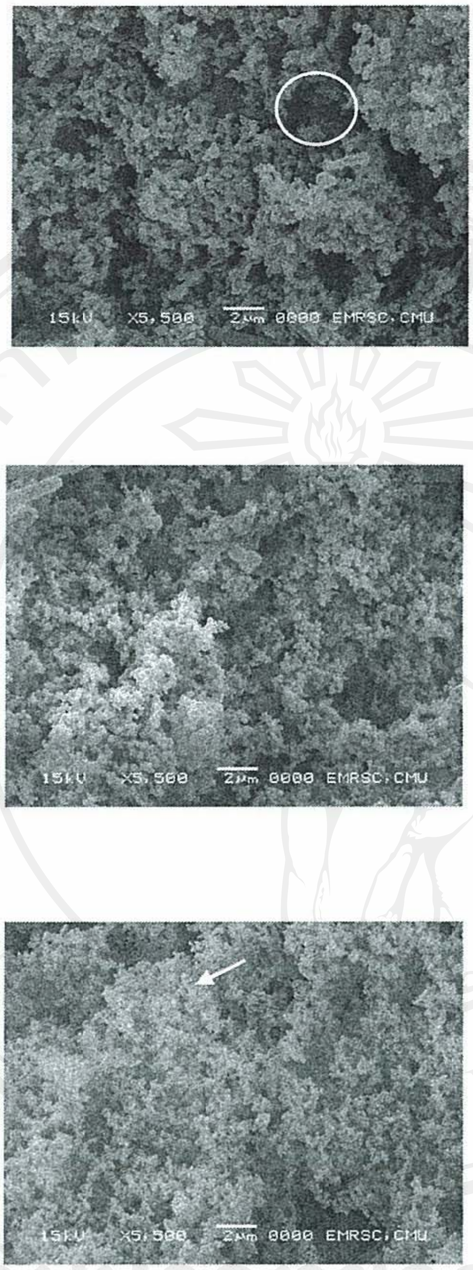

Figure 6.4 Scanning electron micrographs of corn milk yogurt with different levels of gelatin addition 0% (a), 0.4% (b), 0.6% (c) (light circle is air cells, and the light arrow indicates a dense and highly branched-structure).

6.3.5 Rheological properties

A constant displacement of 0.002 rad was selected (Figure 6.5) for the frequency sweeps which were performed at 0.01-1 Hz.

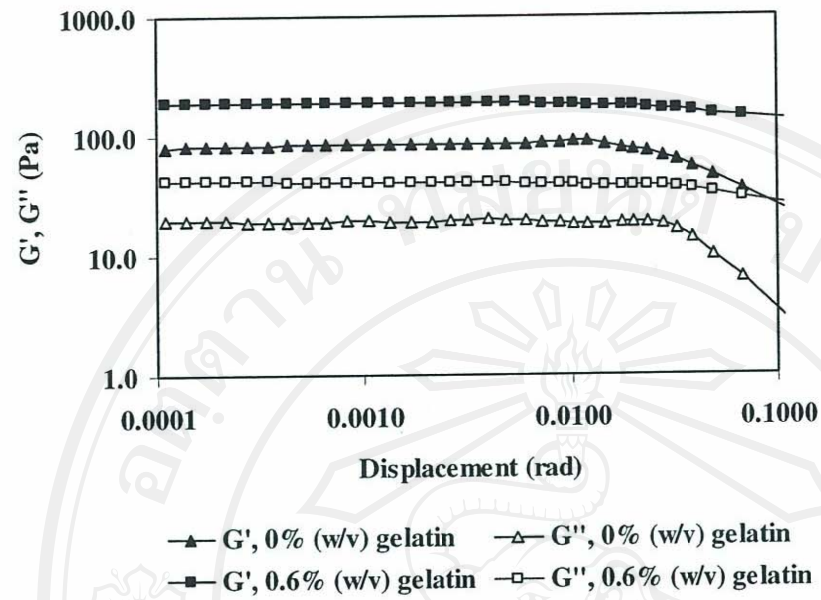

Figure 6.5 Strain sweep at constant frequency of 1 Hz of corn milk yogurt with 0.6% (w/v) added gelatin compared with the control (no added gelatin).

Table 6.1 and Figure 6.6 showed the viscoelastic behavior of the corn milk yogurts, at the lowest concentration of added gelatin (0.2%, w/v) both the storage (G′) and loss (G″) moduli exhibited a similar pattern to that of the untreated (no added gelatin) sample whereas in the samples having larger amounts of gelatin, both moduli increase significantly ($P<0.05$). Probably due to a higher cross-link density induced by stronger gelation of the system (Apichartsrangkoon, 2002; Apichartsrangkoon 2003; Apichartsrangkoon *et al.*, 1999; Apichartsrangkoon and Ledward, 2002). It also worth noting that the overall G′ plots were less frequency dependent than the G″ plots and their loss tangent (ratio of G″/G′) were as low as 0.2 which was an indication of true gel behavior with solid-like structures (Ross-Murphy, 1984). These results were in accord with the TPA (Figure 6.3) and microstructure (Figure 6.4) results.

Table 6.1 Viscoelastic behavior of corn milk yogurt with various amounts of added gelatin measured at a frequency of 0.1 Hz

Gelatin (%, w/v)	G′ (Pa)	G″ (Pa)
0.0	87.56 ± 5.28[c]	18.32 ± 1.22[c]
0.2	102.82 ± 5.58[c]	20.16 ± 1.06[c]
0.4	170.33 ± 8.43[b]	36.59 ± 2.05[b]
0.6	206.40 ± 17.43[a]	42.22 ± 6.09[a]

* Values in a column followed by different letters were significantly different treatments (*P*<0.05)

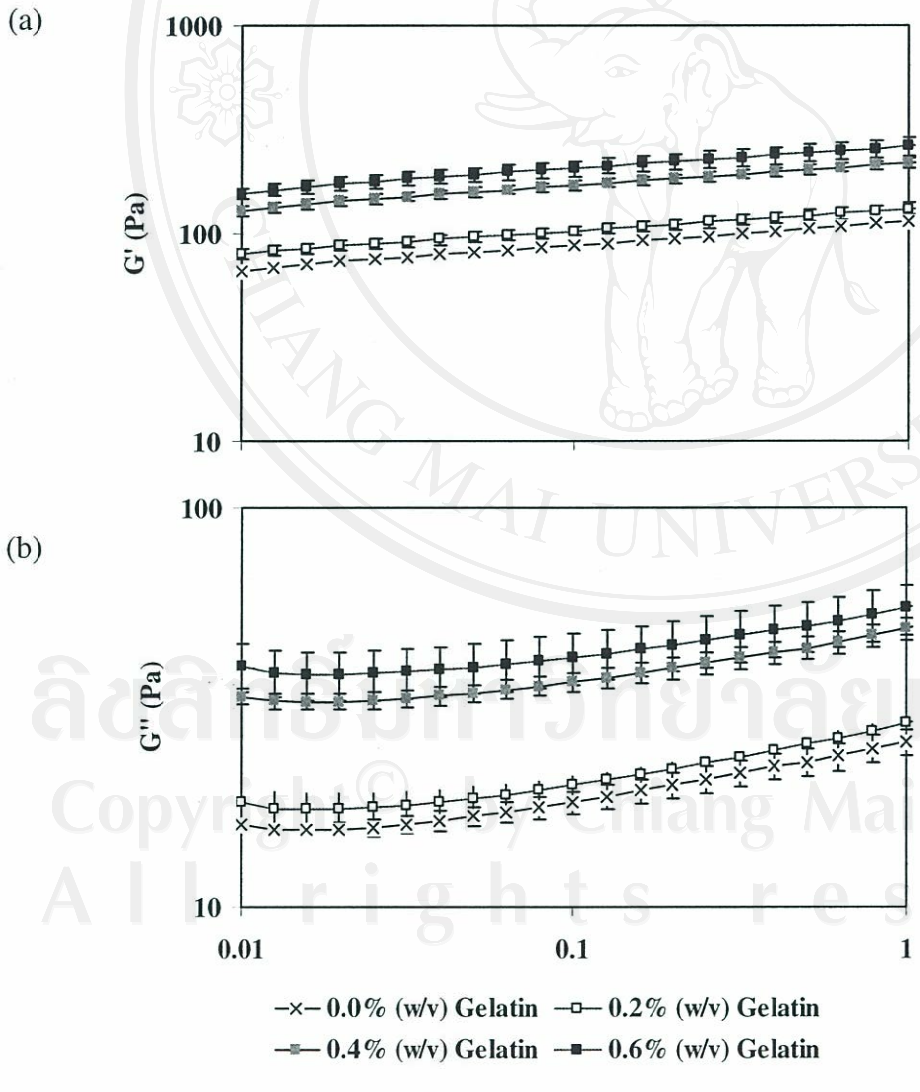

Figure 6.6 Dynamic viscoelastic behavior of corn milk yogurt with different concentrations of gelatin addition.

6.4 CONCLUSIONS

The highest amount of additional gelatin (0.6%, w/v) gave very hard gel structure with the firmest structure. It led to a very dense and compact structure which could impair palatability. Thus, the optimum level of additional gelatin might be 0.4% (w/v).