CHAPTER 4

THE QUALITY OF CORN MILK YOGURT AS AFFECTED BY MILK PROTEIN

4.1 INTRODUCTION

In this research, corn milk was used as a principle ingredient for yogurt making. Making yogurt from corn milk is not only increasing the diversity of fermented products, but also it would be a novel product that combines the good sensory characteristic as well as a high nutritional value of the corn milk with the desirable yogurt flavor. However, to maintain the other physical characteristics of yogurt, for example body and texture, quality and the consumer acceptance, appearance and mouthfeel, the milk based may need to be fortified. An addition of milk based with dried dairy ingredients is a common practice in yogurt manufactory. In general, dried dairy ingredients can be divided into two groups. The first group consists of casein based such as SMP, sodium caseinate and calcium caseinate. Another group is whey protein based; including whey protein isolate (WPI) and whey protein concentrate (Amatayakul et al., 2006b). The addition of dried dairy ingredients causes the increase in density of protein matrix in microstructure, and reduction of syneresis in yogurt (Amatayakul et al., 2006b; Everett and McLeod, 2005; Remeuf et al., 2003; Saint-Eve et al., 2006). For soymilk yogurt, sodium caseinate stimulates acid development and coagulates the soy protein (Granata and Morr, 1996; Karleskind et al., 1991). For a novel corn milk yogurt, it is importance to investigate the properties of the yogurt as affected by the added proteins.

Since the dried dairy ingredients exhibit diverse functional and structural characteristics, so it is appropriate to consider their application in the form of blends. However, the blends to be used should be carefully studied prior to recommending their application. This study aims to examine the effect of sodium caseinate and WPI that incorporated individually or blended in different proportions upon the pH value, lactic acid content, and amount of yogurt starter cultures, physical properties, texture

and microstructure of the corn milk yogurt. The 0-4% sodium caseinate and 0-1% WPI were employed in this study because cow's milk generally contained about 3-4% protein, which consisted of 80% casein and 20% whey protein (Fox and McSweeney, 1998; Walstra *et al.*, 1999).

4.2 MATERIALS AND METHODS

4.2.1 Preparation of the sweet corn milk

The sweet corn used in this study was an ATS-5 that harvested on the 23nd 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.

4.2.2 Starter cultures preparation

S. thermophilus and L. delbrueckii subsp. bulgaricus were prepared using the methods described in section 3.2.2 and 3.2.3, respectively.

4.2.3 Corn milk yogurt preparation

The corn milk was added with distilled water at a ratio of 1:2 for corn milk to distilled water. The ratio used was the best ratio predetermined based upon. The milk was then heated at 90°C prior to fortification with milk protein at 9 different levels using blends of 0, 2 or 4% (w/v) sodium caseinate (BBA, France) and 0, 0.5 or 1.0% (w/v) WPI (Arla, Denmark). The milk mixture was subsequently stirred for 5 min. After stirring, the milks were pasteurized by heating at 95°C for 5 min (Raphaelides and Gioldasi, 2005) and cooled to 40°C. The milk was then inoculated with 2% (v/v) yogurt starter cultures that were composed of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* at a ratio of 1:1. The inoculated milk was poured into 100 ml sterilized plastic cup and incubated at 40°C for 4 h until the pH values reduced to 4.4-4.6. All treatments were carried out in triplicate.

4.2.4 Chemical analysis

Samples of corn milk yogurt were analyzed for total solid, protein content, total acidity and pH value. The total solid, protein content and total acidity were measured according to AOAC methods no. 990.20, no. 991.20 and no. 947.05, respectively (AOAC, 2000). The result of the total acidity was expressed as % lactic

acid. For the pH measurement, a pH meter Consort C830 (CE, Belgium) was employed.

4.2.5 Microbiological analysis

The corn milk yogurts were subjected to microbiological analysis for the viable numbers of yogurt cultures. *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* were enumerated by the methods previously described in section 3.2.7.

4.2.6 Color

Color of corn milk yogurt was measured by a Minolta Data Processor DP-301 colorimeter (Minolta, Japan).

4.2.7 Physical properties

4.2.7.1 Whey drainage

Whey drainage was removed from the corn milk yogurt using a syringe within 24 h after the yogurt fermentation was completed. The relative amount of whey drained off (in ml per 100 ml of initial sample) was calculated as the whey drainage. This method was modified from the method of Fiszman *et al.* (1999).

4.2.7.2 Syneresis

A method of Wu et al. (2001) was used to measure the syneresis of corn milk yogurt. Within 24 h after the yogurt fermentation completed, the analysis had to be carried out. The analysis was done using a Whatman filter paper number 1 to cover a Buchner funnel. After that, an amount of 20 g of the yogurt was spread in a thin layer to cover the surface of the filter paper. The funnel was then arranged on top of an Erlenmeyer flask and the flask was connected to a vacuum pump. When all the system was set up, the yogurt was filtered in a vacuum condition for 10 min. The liquid that passed the filter paper was collected and recorded. The percentage of syneresis was calculated as the weight of the liquid divided with the weight of the initial sample multiple by 100.

4.2.7.3 Water holding capacity

The water holding capacity was measured by a centrifuge method according to a modified method of Parnell-Cluies *et al.* (1986). Within 12 h of the production of corn milk yogurt, an amount of 10 g sample was centrifuged at 3,000 rpm for 60 min at 10°C. The supernatant was then removed within 10 min and the pellet weight was

recorded. The water holding capacity was expressed as percentage of pellet weight relative to the original weight of corn milk yogurt.

4.2.7.4 Consistency

Yogurt samples were stirred within 24 h after the yogurt fermentation completed. Yogurts were stirred for 1 min at 1,500 rpm at a temperature of 10°C then poured the samples into the slotted compartment of a Bostwick consistometer box until it was full of the sample. Consistency was determined by measuring the distance that was expressed in cm unit, over which the samples flowed in a Bostwick consistometer at 10°C after the door of the equipment was removed for 30 s. The measurement was applied following a method of González-Martínez *et al.* (2002).

4.2.8 Texture Profile Analysis (TPA)

TPA was carried out within 24 h after the yogurt fermentation completed by a modified method of Kumar and Mishra (2004), TPA tests were measured using a TA-XT Plus (Stable Micro Systems, UK) with a 5 kg load cell. Experiments were evaluated by compression tests which generated plot of force (g) vs. time (s). A 35 mm diameter cylinder aluminum probe was used to measure textural profile of a set yogurt sample prepared in a 100 ml cup at a temperature of 10±0.5°C. In the first stage, the corn milk yogurts were compressed by 30% of their original depth. The speed of the probe was fixed at 0.5 mm/s during the pre-test, compression and the relaxation of the samples. The data presented were averages of three replications.

4.2.9 Microstructure

Within 24 h after the yogurt fermentation completed, 0.3 g of samples were taken at about 1 cm below the surface and mixed with 0.3 g of 3% aqueous agar solution (Oxoid code L13, England) at 45°C, and the mixtures were solidified by cooling at 20°C. The gelled samples were cut into approximately 1 mm cubes and had a primary fixative for 20 min at room temperature in a 2.5% glutaraldehyde solution in phosphate buffer (0.1 M, pH 7.3). The primary fixative was conducted 2 times to crosslink proteins. The fixed samples were then washed with phosphate buffer, and postfixed in a 1% osmium tetraoxide solution in phosphate buffer for 1 h for stabilizing unsaturated lipids. The postfixed samples were washed with phosphate buffer and dehydrated in a graded ethanol series (15, 30, 50, 70, 80, 95 and 100%, 30 min in each). Dehydrated samples were then dried with a Pelco CPD-2-critical point

drier (Ted Palla Co., Redding, CA, USA). Dry sections were fractured with a blade and fragments were mounted on aluminum SEM stubs, and vacuum gold coated with a Fine Coat Jeol-JFC-1100 (Jeol Ltd., Akishima, Japan). Microstructure of the corn milk yogurts was examined with a scanning electron microscope model Jeol JSM-5910LV (Jeol Ltd., Tokyo, Japan), using a magnification of 5500X. The measurement was applied following a method of Sandoval-Castilla *et al.* (2004).

4.2.10 Statistical analysis

The collected data from chemical and microbiological analysis, color, and TPA were analyzed statistically by an Analysis of Variance using a Factorial Experiment in CRD with 2 factors. The first factor was concentrations of sodium caseinate, which were 3 concentrations, 0, 2 and 4% (w/v). The second factor was concentrations of WPI, which were 3 concentrations, 0, 0.5 and 1% (w/v). CRD was used to evaluate the results of physical properties. If the F value from the Analysis of Variance was significant, a Duncan's New Multiple Range test was utilized to determine differences between treatment means (Montgomery, 2001). The statistical calculation was performed using a SPSS statistical software version 10.0.1.

4.3 RESULTS AND DISCUSSION

4.3.1 Total solid and protein content

Figure 4.1 revealed that fortification of milk protein in corn milk yogurt affected the solid content of the product. The total solid was significantly increased with higher level of sodium caseinate and/or WPI concentration (*P*<0.05). The gel structure of yogurt was not observed in corn milk yogurt without supplementation of milk protein. The fortification of 4-5% protein to obtain the enough solid content for yogurt making was stated by Remeuf *et al.* (2003). Lee *et al.* (1990) reported that soymilk exhibited the good gel at 8.12% protein contents, while Yazici *et al.* (1997) showed that supplemented the soymilk with calcium required only 5.4% protein to form gel.

The corn milk yogurts added with WPI alone at any tested concentrations could not form good gel. The sodium caseinate was a better fortified protein, and it produced enough solid to form a good gel of yogurt at the concentration of 4% (w/v). Tamine and Robinson (1999) reported that the minimum solids concentration of

cow's milk yogurt was 8.2-8.65%, but the consistency of yogurt would be greatly improved as the solids increased from 12 to 20%. The optimum total solids to produce yogurt from soymilk (Lee *et al.*, 1990) and a combination of mango pulp, soymilk and buffalo milk (Kumar and Mishra, 2004) were 11.49 and 12.26%, respectively.

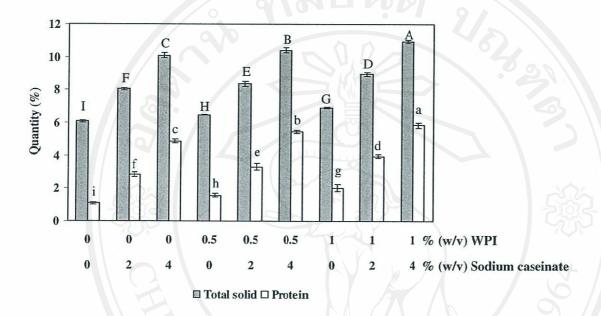


Figure 4.1 Total solid and protein contents of corn milk yogurts supplemented with different concentrations of sodium caseinate and WPI. Bars with different superscript were significantly different (*P*<0.05).

4.3.2 pH value and total acidity of corn milk yogurt

Figure 4.2 showed pH value and total acidity of corn milk yogurt which were added with different ratio of sodium caseinate and WPI. The acidity of yogurt fortified with sodium caseinate and/or WPI significantly increased as proportion of sodium caseinate in the yogurt increased while the increase in pH were found in yogurts that fortified with high proportions of sodium caseinate and WPI. This result suggested that sodium caseinate noticeably influenced the growth of cultures, which produced acidity, while the effect of WPI was much smaller than that of sodium caseinate.

Milk casein was a source of amino acid and nitrogen for growth of LAB (Oliveira *et al.*, 2001; Shihata and Shah, 2000). Previous work discovered that both sodium caseinate and whey protein hydrolysate could enhance the lactic acid

production in soymilk yogurt (Granata and Morr, 1996). The result of this study that WPI yogurt produced relatively lower acidity than sodium caseinate yogurt was consistent with the study of Karleskind *et al.* (1991). Karleskind *et al.* (1991) explained that WPI and cheese whey did not contribute adequate amounts of potential essential nutrients for the bacteria culture in soymilk yogurt whereas 4.0% (w/v) sodium caseinate could effectively stimulate the growth of bacteria. Granata and Morr (1996) also confirmed that whey protein hydrolysate was less effective than sodium caseinate for stimulation acid production in soymilk yogurt. In case of cow's milk yogurt, however, whey protein could stimulate the growth of yogurt starter cultures (Dave and Shah, 1998; Granata and Morr, 1996; Karleskind *et al.*, 1991).

Most of pH values and total acidity of the corn milk yogurts would be decreased as lower concentration of sodium caseinate, WPI or sodium caseinate to WPI ratios. The reason might be due to the difference in the buffering capacity of milk blend. Whey protein generally has lower buffering capacity than casein (Walstra and Jenness, 1984). Therefore, lowering the concentration of sodium caseinate would result in a reduction in buffering capacity. However, the reduction in buffering capacity might not be the only factor contributing to the lower pH values of yogurt with low ratio of sodium caseinate to WPI.

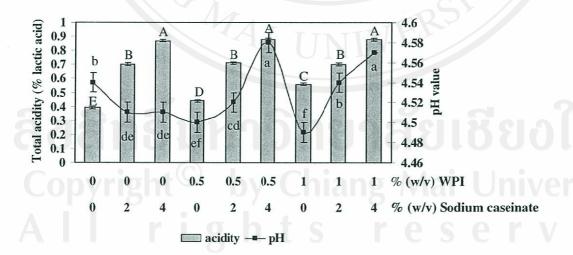


Figure 4.2 Total acidity (% lactic acid) and pH value of corn milk yogurts supplemented with different concentrations of sodium caseinate and WPI. Means with different letters were significantly different (*P*<0.05).

4.3.3 Viable cell counts of starter cultures

The effects of type and concentration of milk proteins on growths of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* showed a similar trend as observed in the case of acidity (Figure 4.3). Sodium caseinate was more effective than WPI for stimulation growths of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus*. The result suggested that addition of 2% (w/v) sodium caseinate with or without addition of WPI provided nutrients that were enough to obtained >9.26 log CFU/ml of *S. thermophilus* and >8.27 log CFU/ml of *L. delbrueckii* subsp. *bulgaricus* (Figure 4.2). Although, the acidity content of the corn milk yogurt would be increased with an increasing of the concentration of sodium caseinate and/or WPI, but the counts of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* also increased. It might be due to the buffering capacity of corn milk yogurt that also increased with higher level of milk proteins (Amatayakul *et al.*, 2006a; Gastaldi *et al.*, 1997).

The better growth of *S. thermophilus* in corn milk yogurt with or without fortification of sodium caseinate and/or WPI would be mainly because *L. delbrueckii* subsp. *bulgaricus* could not utilize sucrose in corn milk (Amoroso and Manca de Nadra, 1992; Thomas and Crow, 1983; Wang *et al.*, 2002).

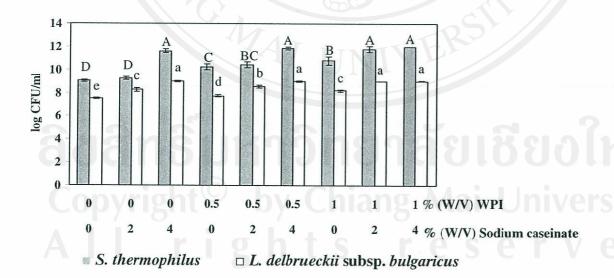


Figure 4.3 The viable numbers of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* in corn milk yogurts supplemented with different concentrations of sodium caseinate and WPI. Bars with different superscript were significantly different (*P*<0.05).

4.3.4 Color

Figure 4.4 showed color coordinate values of L* (lightness), C* (chroma or purity of color) and h (hue angle). Sodium caseinate and/or WPI significantly affected color of corn milk yogurt (*P*<0.05). Increase of lightness and decrease of color purity of corn milk yogurts were observed at higher concentration of sodium caseinate and/or WPI. The results were agreed with the observation of González-Martínez *et al.* (2002) that studied the change of color values as affected by casein. The higher milk protein content would increase the total acid content in corn milk yogurt, and then caused the higher coagulation of protein. The coagulation of protein then affected the structure and surface properties of yogurt. Mor-Mur and Yuste (2003) reported that the increasing in the protein coagulation would enhance the light absorption that resulted in the lighter tones.

According to the h values, 97.00-100.53, the color of all yogurt samples were yellow (Minolta, 1994). The h value of corn milk yogurt was decreased as the proportion of sodium caseinate and WPI concentration in the yogurt increased (Figure 4.5).

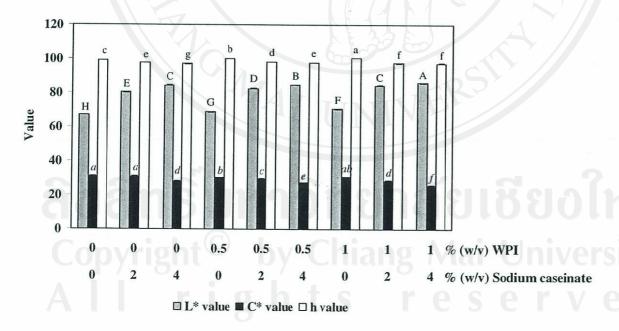


Figure 4.4 Color of corn milk yogurts fortified with different concentrations of sodium caseinate and WPI. Bars with different superscript were significantly different (P<0.05).

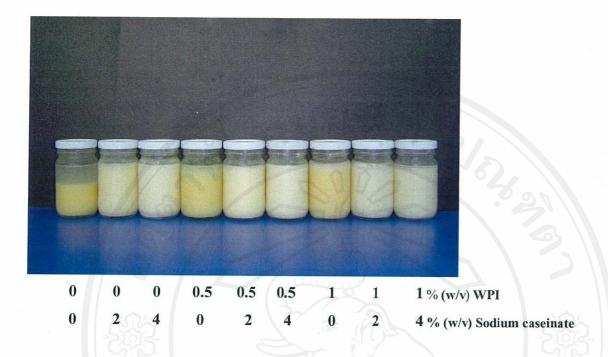


Figure 4.5 Corn milk yogurts fortified with different concentrations and types of milk protein.

4.3.5 Physical properties

The results (Table 4.1) revealed that the substrates in corn milk yogurt could not make a curd structure of yogurt. Thus, the physical properties of corn milk yogurt without addition of milk protein were not presented. Although corn milk could support the growth of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* as appeared in the reduction of pH (Figure 4.2). It might be due to the effects of total solid, protein content and type of protein in corn milk. The total solid and protein content in the corn milk yogurt were 6.07±0.08% and 1.09±0.07%, respectively (Figure 4.1), which was less than those in cow's milk yogurt. Important proteins in corn milk were zein (prolamins) and corn glutelin (Lásztity, 1996; Pomeranz, 1987) that could not make a gel characteristic in acid condition of yogurt. Protein in cow's milk contained whey protein and casein micelles (Amatayakul *et al.*, 2006b). Remeuf *et al.* (2003) and Lucey (2004) cited that heat treatment and acid production by LAB resulted in denaturation of whey protein and casein. The denatured whey proteins became mostly associated with casein micelles during heat treatment or acidification

process. Aggregation of casein particles occurred as the pH approached the isoelectric point of 4.6 (Lucey, 2004; Shaker *et al.*, 2000).

Whey drainage could be defined as the appearance of whey (serum) on the yogurt surface (Farnsworth *et al.*, 2006). Table 4.1 showed that the whey drainage of corn milk yogurts decreased with the increase in proportion of sodium caseinate or WPI in yogurt substrate. The increase of total solids and protein content might enhance the interactions between particles and, as a result, the ability of yogurt to hold water increased (Amatayakukl *et al.*, 2006b; Gastaldi *et al.*, 1997). Fortification of sodium caseinate and WPI gave the variation of whey drainage. For example, at 0.5% (w/v) WPI, the whey drainage decreased with increase in concentrations of sodium caseinate. On the other hand, proportion of the sodium caseinate was not significant on the whey drainage at 1.0% (w/v) WPI ($P \ge 0.05$). It might be because the excessive whey protein to casein ratio induced the shrinkage of the gel, which then led to increase whey drainage or whey separation (Farnsworth *et al.*, 2006; Lucey, 2004)

The effect of milk protein on syneresis and water holding capacity of corn milk yogurt could be seen in Table 4.1. The syneresis reduced with increase of proportions of sodium caseinate and/or WPI. In contrast, water holding capacity of the corn milk yogurt increased with increase in proportions of the fortified proteins while the ratios of the combination of sodium caseinate and WPI were not significant difference ($P \ge 0.05$). Generally, the increase in total solid of liquid milk caused the increase in density, but it reduced pore size in protein matrix of yogurt gel. Then, reduction of syneresis and improvement of the water holding capacity of the gel exhibited (Amatayakul et al., 2006b; Lorenzen et al., 2002; Sodini et al., 2004). The similar effects were observed by Keogh and O'Kennedy (1998). Keogh and O'Kennedy (1998) explained that casein and \(\beta-lactoglobulin interact chemical on heating. This effectively increased the concentration of gel-forming protein in the yogurt matrix and reduced syneresis through increased entrapment of serum within the interstices of the whey protein molecules attached to the surface of the casein. Augustin et al. (1999) noted that the denatured whey protein content had the major influence on resistance of yogurts to syneresis.

Table 4.1 revealed that the addition of sodium caseinate and/or WPI supported the increase in consistency of corn milk yogurt. The corn milk yogurts

fortified with <4.0% (w/v) sodium caseinate which had <10.08% total solid (Figure 4.1), had a soft gel. This characteristic of soft gel could be due to the decreasing of fraction of solid. Therefore, the increase in total solid through additions of sodium caseinate and/or WPI caused the gel formation. The less open gel structure formed with high protein content would be made the aggregate network with high consistency. This was in agreement with results found by Gozález-Martínez *et al.* (2002) who studied the influence of substituting milk powder for whey powder on yogurt quality. However, in this study, corn milk fortified with low concentration of milk protein generally produced yogurt with very weak gel. The hard gel was obtained at 4 and 1% (w/v) sodium caseinate and WPI, of which the amounts of solid and protein content were 10.94 and 5.86%, respectively (Figure 4.1).

Table 4.1 Physical properties of corn milk yogurts supplemented with different levels of sodium caseinate and WPI.

Sodium caseinate (%)	WPI (%)	Whey drainage (%)	Syneresis (%)	Water holding capacity (%)	Bostwick distance (cm)
0	0.0	ND	ND	ND	> 24
2	0.0	13.22 <u>+</u> 0.20 ^b	77.72 <u>+</u> 0.71 ^b	32.61 <u>+</u> 1.06 ^c	> 24
4	0.0	0.58 <u>+</u> 0.13 ^f	74.93 <u>+</u> 0.88 ^{cd}	40.63 ± 1.22^{b}	10.03 <u>+</u> 0.50 ^c
0	0.5	23.82±0.18 ^a	81.76 <u>+</u> 0.75 ^a	32.80 <u>+</u> 1.07 ^c	> 24
2	0.5	2.35 <u>+</u> 0.40 ^d	77.21 <u>+</u> 1.33 ^{bc}	40.79±1.69 ^b	11.46 <u>+</u> 0.13 ^b
4	0.5	1.50 <u>+</u> 0.28 ^e	72.55 <u>+</u> 2.32 ^{de}	42.34 <u>+</u> 1.47 ^{ab}	6.15 <u>+</u> 0.84 ^e
0	1.0	0.62 <u>+</u> 0.18 ^f	71.43 <u>+</u> 1.24 ^e	42.17±0.42 ^{ab}	19.54 <u>+</u> 0.85 ^a
2	1.0	6.43 <u>+</u> 0.71 ^c	75.52 <u>+</u> 1.90 ^{bc}	42.43 <u>+</u> 2.20 ^{ab}	7.54 <u>+</u> 0.27 ^d
4	1.0	6.27 <u>+</u> 0.91 ^c	67.84 <u>+</u> 1.61 ^f	44.52 <u>+</u> 2.62 ^a	5.72 <u>+</u> 0.18 ^e

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

ND = Not determined

4.3.6 Texture profile

The addition of milk proteins positively affected the textural properties of corn milk yogurts. Hardness of the corn milk yogurts was increased as higher levels of sodium caseinate and/or WPI were added (Figure 4.6). The similar trend was found by Amatayakul *et al.* (2006b); Gastaldi *et al.* (1997); Oliveira *et al.* (2001). The increase in solid and casein contents led to the increase in protein network and number of bond between particles in the yogurt matrix. (Gastaldi *et al.*, 1997; Keogh and O'Kennedy, 1998). According to Amatayakul *et al.* (2006b), the denatured whey protein on the surface of casein micelles was increased with the reduction in sodium caseinate to WPI ratio. This would prevent the coalescence of casein micelles and network formation, and resulted in decreasing of hardness of yogurt. The same trend was shown for the studies of adhesiveness. The springiness was not significantly affected by the additions of sodium caseinate or WPI.



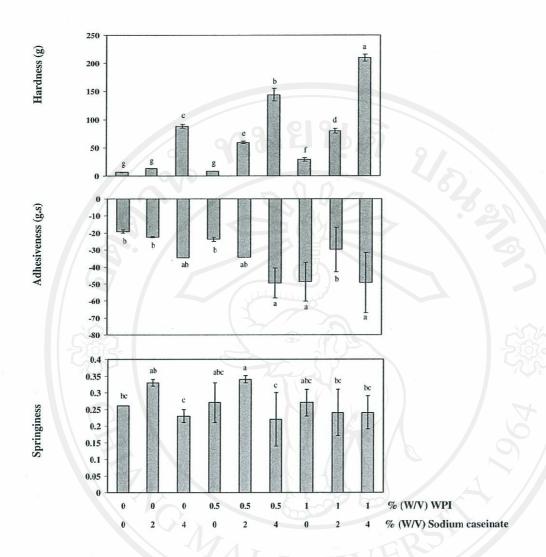


Figure 4.6 Texture profiles of corn milk yogurts supplemented with different concentrations of sodium caseinate and WPI. Bars with different superscript were significantly different (*P*<0.05).

4.3.7 Microstructure

Scanning electron micrographs in Figure 4.7 revealed that the microstructure of gel of corn milk yogurt was influenced by both types and concentrations of milk proteins, i.e. casein and whey protein. A thick compact structure and big holes were observed in the control corn milk yogurt. It might be because the control corn milk mixture contained less quantities of solid and protein content. Moreover, zein in corn milk could not form a gel at pH value of 4.6. Therefore the control corn milk yogurt could not exhibit a gel structure of yogurt. Adding enough amount of milk protein, for

example 4.0% (w/v) sodium caseinate, 1.0% (w/v) WPI and a combination of 4% sodium caseinate and 1.0% (w/v) WPI, could enhance the interactions between particles and higher the density of the matrix. This phenomenon induced the increase in the immobilization of free water in the yogurt gel (Amatayakul *et al.*, 2006b; Lorenzen *et al.*, 2002; Sodini *et al.*, 2004). Consequently, the water holding capacity of yogurt was improved, and the syneresis was reduced.

The corn milk yogurt added with 4.0% (w/v) sodium caseinate had spongy structure with regular pore size. Its pore size was smaller than that of the control corn milk yogurt (Fig 4.7 (b) and (a), respectively). The corn milk yogurt added with 1.0% (w/v) WPI exhibited a non-spongy network with irregular pore size. Thus, the corn milk yogurt added with 1.0% (w/v) WPI contained less consistency and hardness.

The gel structure of corn milk yogurt added with 4.0% (w/v) sodium caseinate and 1.0% (w/v) WPI exhibited irregular pore size and finer network than that of the corn milk yogurt added only with sodium caseinate. Moreover, its micrograph of the fortified corn milk yogurt displayed a shrinkage structure. This shrinkage structure might be responsible for low value of syneresis, but high values of whey drainage, water holding capacity, consistency, hardness, adhesiveness and gumminess of the yogurt

These observations could be compared with those of several authors. Saint-Eve *et al.* (2006) observed that the protein composition including sodium caseinate, SMP and whey protein concentrate, caused differences in the structure of the gel network. They found that sodium caseinate provided gel with heterogeneous structure with large pores. When yogurts were enriched with whey protein, the protein network presented a more uniform distribution of the gel and smaller pores than sodium caseinate yogurt. In addition, Remeuf *et al.* (2003) stated that gel of whey protein concentrated-enriched yogurts exhibited a different structure with a very fine network, containing numerous very small pores. Puventhiran *et al.* (2002) reported that the addition of whey protein led to a structure where casein micelles appeared in the form of individual entities surrounded by finely flocculated protein and linked to very small whey protein aggregates. This structure might increase the number of bonds between particles so; explain the dense and finely branched network in yogurt from whey protein-enriched milk based. Thus, when yogurts were enriched with caseinate,

interactions between casein micelles were more likely. Yogurts fortified with casein preparations showed fusion of casein micelles. This indicated a lower degree of interaction between whey protein and casein micelles, possibly leading to the coarse network of sodium caseinate yogurt, contrary to whey protein yogurt (Saint-Eve *et al.*, 2006).



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Figure 4.7 Scanning electron micrographs of corn milk yogurt without protein supplementation (a) and corn milk yogurt supplemented with 4.0% (w/v) sodium caseinate (b), 1.0% (w/v) WPI (c), 4.0% (w/v) sodium caseinate and 1.0% (w/v) WPI (d).

4.4. CONCLUSIONS

Sodium caseinate and WPI were significantly affected the production of lactic acid, pH value, counts of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus*, color, physical and textural properties, and the microstructure of corn milk yogurts. Lactic acid production, pH values and amount of starter cultures were enhanced by the addition of milk proteins. The lightness of the corn milk yogurt was higher, but purity of color was lower with increase of sodium caseinate and/or WPI concentration. The yellowness of yogurt was decreased with the increase of WPI concentration. Addition of sodium caseinate enhanced the yellowness of yogurt. Water holding capacity and consistency were increased, whereas syneresis was decreased with increasing of sodium caseinate and/or WPI concentration. Whey drainage was mainly reduced by adding sodium caseinate.

The corn milk alone could not exhibit the gel structure of yogurt. Fortification of corn milk with 4.0% (w/v) sodium caseinate was suitable for corn milk yogurt production, which the protein content of this corn milk yogurt was within the range of cow's milk yogurt. It was because the yogurt contained the highest amounts of starter cultures and the lowest whey drainage. Adding 4.0% (w/v) sodium caseinate or 1.0% (w/v) WPI in the corn milk yogurt produced homogeneous networks with smaller pore size compared to that in the control corn milk yogurt. The micrograph of 4.0% (w/v) sodium caseinate added yogurt displayed more homogeneous and bigger network than that of 1.0% (w/v) WPI added yogurt. Combination of 4.0% (w/v) sodium caseinate and 1.0% (w/v) WPI gave the fine network of shrinkage gel with irregular and heterogeneous pore size.

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