CHAPTER 5
EFFECT OF LACTOSE ADDITION ON ACID PRODUCTION AND QUALITY OF CORN MILK PRODUCTS

5.1 INTRODUCTION

The worldwide production and consumption of yogurt increased dramatically during the last quarter of the past century (Vinderola *et al.*, 2002). Yogurt is principally produced by the symbiotic action of two types of LAB including *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* (Adam *et al.*, 2004; Afonso and Maia, 1999; De Brabandere and De Baerdemaeker, 1999; Lourens-Hattingh and Viljoen, 2001; Tamime and Robinson, 1999; Vaningelgem *et al.*, 2004; Walstra *et al.*, 1999; Xanthopoulos *et al.*, 2001). Cow's milk is the most common raw material for yogurt making. LAB converts lactose, which cow's milk contains 4.8-4.9% (Fox and McSweeney, 1998; Walstra *et al.*, 1999), into lactic acid. Lactic acid helps to destabilize the casein micelles by progressively converting the colloidal calcium/phosphate complex to the soluble calcium phosphate fraction which diffuses into the aqueous phase of the milk. As a result, micelles are gradually depleted of calcium, leading to coagulation of the casein. The coagulation starts at pH 5.3 and completes at pH 4.4-4.6. Lactic acid also gives distinctive flavor and taste (Duboc and Mollet, 2001; Lucey, 2004; Tamime and Robinson, 1999; Vinderola *et al.*, 2002).

Yogurt have been made from a variety of food resources, such as soymilk (Granata and Morr, 1996), coconut milk (Siripanporn *et al.*, 2000), grape juice (Öztürk and Öner, 1999), a combination of mango pulp-soymilk and buffalo milk (Kumar and Mishra, 2004), and a solution of SMP, soymilk and saccharified-rice (Park *et al.*, 2005). Cheng *et al.* (1990) reported addition of lactose to improve the acid production in soymilk yogurt.

In this research, corn milk was used as a material for yogurt making. Corn milk yogurt would offer several distinct nutritional advantages over milk yogurt to the consumer such as reduced levels of cholesterol and saturated fatty acids (USDA, 2004). However, sugar in corn milk is sucrose which cannot provide lactic acid.

(Azanza *et al.*, 1996; Olsen *et al.*, 1990; Pomeranz, 1987). Beside, zein is the main protein in sweet corn, which is an alcohol-soluble protein and display high hydrophobic property hence cannot coagulate by acid (Lásztity, 1996; Pomeranz, 1987). This study determined the effects of lactose on acid production, structure, and survivals of starter cultures of fermented corn milk products.

5.2 MATERIALS AND METHODS

5.2.1 Fermented corn milk for acid production study

5.2.1.1 Sweet corn milk

The sweet corn used in this study was an ATS-5 that 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.

5.2.1.2 Starter cultures preparation

S. thermophilus and *L. delbrueckii* subsp. *bulgaricus* were prepared according to the procedures in section 3.2.2 and 3.2.3.

5.2.1.3 Fermented corn milk

The corn milk was added with distilled water in a ratio 1:2 of corn milk to distilled water. Then, the milk was fortified with 0, 2 or 4% (w/v) lactose (Fonterra, New Zealand). The fermentation of corn milk was prepared following the method in section 4.2.3.

5.2.2 Corn milk yogurt for quality study

From 5.2.1, it was chosen the best concentration of lactose that gave the highest acid production and added with 4% (w/v) sodium caseinate (BBA, France). The fermentation of corn milk yogurt was prepared following the method in section 4.2.3. The effect of lactose addition on sodium caseinate fortified yogurt was determined for its structure. The sodium caseinate fortified yogurt without lactose addition was used as a control treatment.

5.2.3 Microbiological analysis

The corn milk samples were collected for the viable numbers of yogurt starter cultures when a pH value of the samples reached 4.4-4.6. The methods for

enumeration of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* were followed the section 3.2.7.

5.2.4 Chemical analysis

At pH value of samples below 4.6, samples of fermented corn milk and corn milk yogurt were analyzed for total solid, total acidity, pH value, and amount of reducing sugar, non reducing sugar as well as invert sugar. The total solid and total acidity were measured according to AOAC methods no. 990.20 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. The amount of reducing sugar, non reducing sugar and invert sugar were measured according to Lane and Eynon method (James, 1995).

5.2.5 Color

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

5.2.6 Physical properties

Physical properties of corn milk yogurt including whey drainage, syneresis, water holding capacity and consistency were evaluated according to the method as described in section 4.2.7.

5.2.7 TPA

TPA was carried out using a method as described in section 4.2.8.

5.2.8 Statistical analysis

The data collected from the acid production experiment were analyzed statistically by an Analysis of Variance using a CRD. If the F value is significant, the Least Significant Difference (LSD) was used to determine the differences between the treatment means. The statistical calculation was performed using a SPSS statistical software version 10.0.1.

The collected data from the quality experiment were analyzed by an analysis of various using a T-test. The statistical calculation was performed using a SPSS statistical software version 10.0.1.

5.3 RESULTS AND DISCUSSION

5.3.1 Effect of lactose on the quality of fermented corn milk

5.3.1.1 Effect of lactose on the numbers of S. thermophilus and L. delbrueckii subsp. *bulgaricus*

As shown in Figure 5.1, lactose of 2 or 4% (w/v) significantly promoted the growth of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* ($P<0.05$). This result agreed with the studies of Husson-Kao *et al.* (2000), Vaningelgem *et al.* (2004) and Vasiljevic and Jelen (2002). However, the comparable results were obtained from the additions of 2 or 4% (w/v) lactose. Hence, using 2% (w/v) of lactose was enough for the growth of yogurt cultures in fermented corn milk.

S. thermophilus grew faster than *L. delbrueckii* subsp. *bulgaricus* in all samples because *S. thermophilus* could utilize both lactose and sucrose for its growth, but *L. delbrueckii* subsp. *bulgaricus* could use only lactose. This was in agreement with the results of Amoroso and Manca de Nadra (1992), Thomas and Crow (1983); van den Bogaard *et al.* (2004), Wang *et al.* (2002) and Wang *et al.* (2003). There was also a reported on a synergistic between *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* that *L. delbrueckii* subsp. *bulgaricus* produced growth factors for *S. thermophilus* (Amoroso and Manca de Nadra, 1992). The sweet corn milk of this study also contained about 5.78±0.01 mg/100 g (Appendix B-1) that could support the growth of S. *thermophilus* (Makhlouf *et al.*, 1995; Tamime and Robinson, 1999; Warman and Havard, 1998).

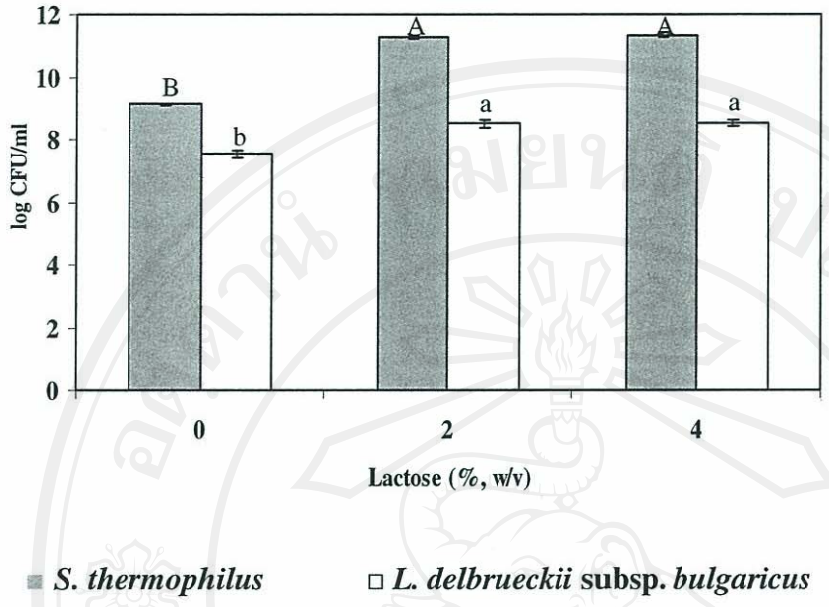

Figure 5.1 The viable numbers of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* in fermented corn milk supplemented with different concentrations of lactose. Bars with different superscript were significantly different ($P<0.05$).

5.3.1.2 Effect of lactose on acid production of fermented corn milk

Mean pH values and total acidity of fermented corn milk samples added with different lactose concentrations are exhibited in Figure 5.2. The pH values of all samples including control were below 4.6 after 4 h of fermentation time. The drop in pH or rise in total acidity in yogurt added with lactose was more than that of the control. This was because S. *thermophilus* and *L. delbrueckii* subsp. *bulgaricus* could utilize lactose and convert to lactic acid. The similar result was discovered in soymilk yogurt by Cheng *et al.* (1990). This study showed that adding lactose at the levels of 2 or 4% (w/v) was not significantly ($P\geq0.05$) affected the pH values of the fermented corn milk. In addition, the drop in pH or increase in total acidity did not seem to be factors that affected the viability of starters during fermentation (Figure 5.1). Adding 2% (w/v) of lactose to corn milk would be the optimum amount as Olsen *et al.* (1990)

found that the growth of S. *thermophilus* and *L. delbrueckii* subsp. *bulgaricus* in corn milk fermentation required only 1.21-1.27% lactose.

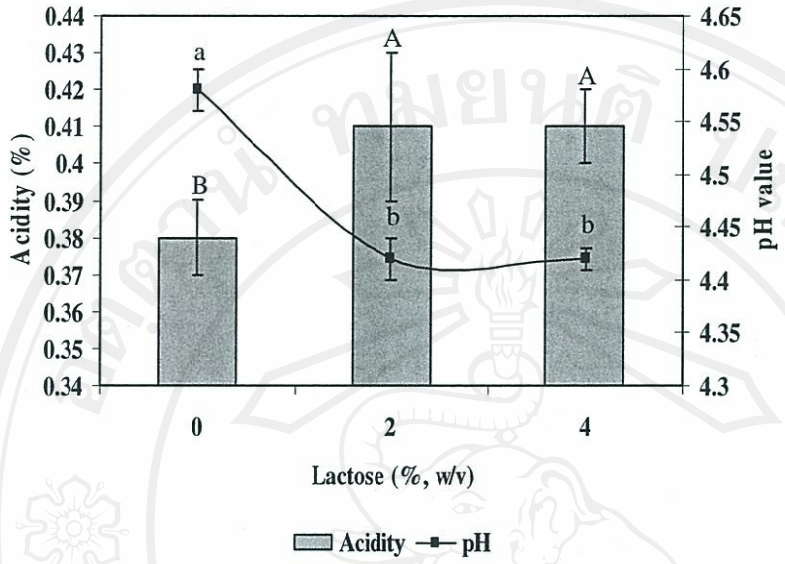

Figure 5.2. Total acidity (% lactic acid) and pH value of fermented corn milk samples supplemented with different concentrations of lactose. Means with different letters were significantly different ($P<0.05$).

5.3.1.3 Effect of lactose on total solid and sugar of fermented corn milk

Figure 5.3 showed amounts of total solid, reducing sugar, invert sugar and non reducing sugar in fermented corn milk added with different levels of lactose. The concentrations of lactose significantly ($P<0.05$) influenced on the amounts of total solid, reducing sugar and invert sugar in fermented corn milk after the pH value of these fermented milk samples were below 4.6. The amounts of total solid, reducing sugar and invert sugar increased in the samples added with high concentrations of lactose. The reason was because lactose is a reducing sugar (Walstra *et al.*, 1999). The hydrolysis of lactose also liberated reducing sugar (James, 1995). Olsen *et al.* (1990) also observed that sweet corn contained reducing sugar such as fructose and glucose.

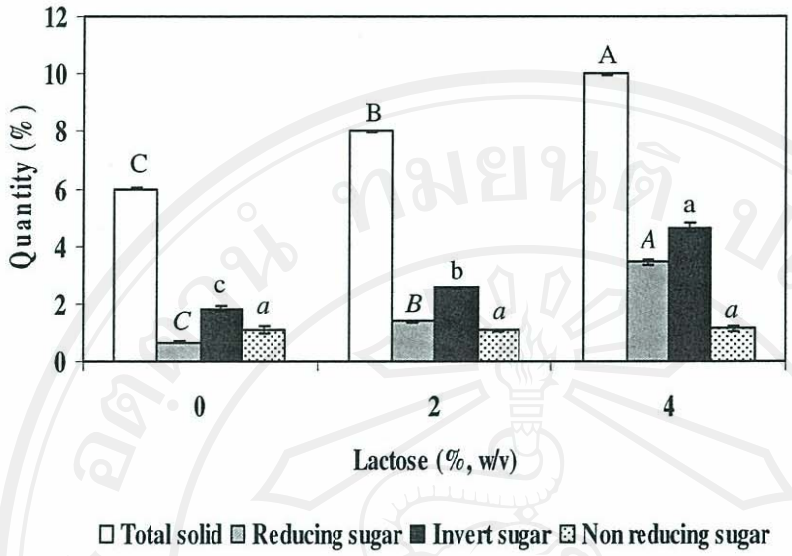

Figure 5.3 Total solid and sugars content of fermented corn milk samples supplemented with different concentrations of lactose. Bars with different superscript were significantly different (P<0.05).

5.3.1.4 Effect of lactose on color of fermented corn milk

Figure 5.4 showed that the concentration of added lactose was not influenced on L* (lightness) and h values but affected on C* value. González-Martínez (2002) explained that lightness of cow's milk yogurt increased with more coagulation of protein. Accordingly, no change in lightness of the yogurt samples could be partly because coagulation of the zein in corn milk was not occurred.

The result of C* value revealed that adding 2 or 4% (w/v) lactose had a little affect on purity of color of fermented corn milk. The h values of 98.70-99.37 of all fermented corn milk samples indicated that the color of these samples were yellow (Minolta, 1994).

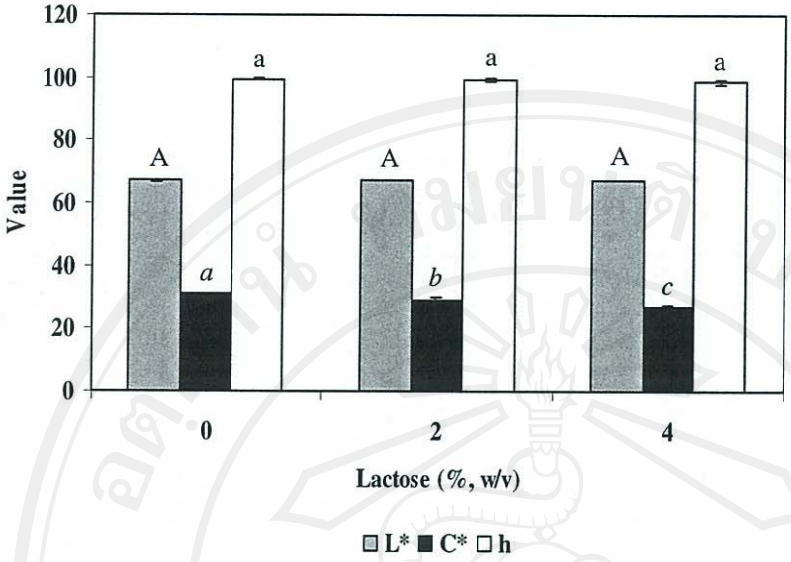

Figure 5.4 Color of fermented corn milk samples supplemented with different concentrations of lactose. Bars with different superscript were significantly different (P<0.05).

According to the similar results, obtained from using 2 and 4% (w/v) lactose, it was concluded that adding 2% (w/v) of lactose to corn milk would be the optimum amount. The 2% (w/v) of lactose was chosen for making yogurt in the experiment 5.2.2.

5.3.2 The effect of lactose on quality of corn milk yogurt

Yogurt added with lactose showed similar results of acid production and the amount of starter cultures (section 5.3.1.1 and 5.3.1.2). The viable numbers of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* in corn milk yogurt added with 2% (w/v) lactose were compared to the control yogurt (Figure 5.5). The amount of the starter cultures in corn milk yogurt added with lactose was significantly (P<0.05) higher than that of the control yogurt. The pH value was lower, and acidity content was higher in the yogurt added with lactose (Figure 5.6). These results revealed that lactose promoted the growths of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus*, and enhanced the acid production.

The influence of lactose on color of the corn milk yogurt was similar to those on the fermented corn milk (Figure 5.7). Hypothesis that protein in the corn milk yogurt was not coagulated was confirmed by the similar results ($P \geq 0.05$) of physical and texture properties of control and tested yogurts (Tables 5.1-5.2). Adding 2% (w/v) lactose increased the total solid of yogurt (Figure 5.6), but it could not cause the change of physical and texture properties to a greater extent. Conversely, for cow's milk yogurt, addition of SMP or milk protein affected the physical properties and textural profile parameters of the yogurt (Amatayakul *et al.*, 2006b; Gastaldi *et al.*, 1997; Oliveira *et al.*, 2001).

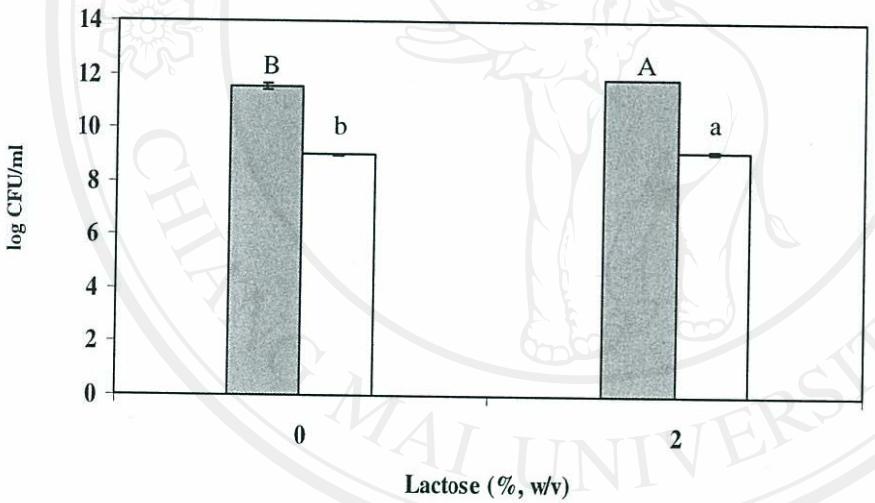

Figure 5.5 The viable numbers of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* in corn milk yogurts added with 0 and 2% (w/v) lactose. Bars with different superscript were significantly different ($P<0.05$).

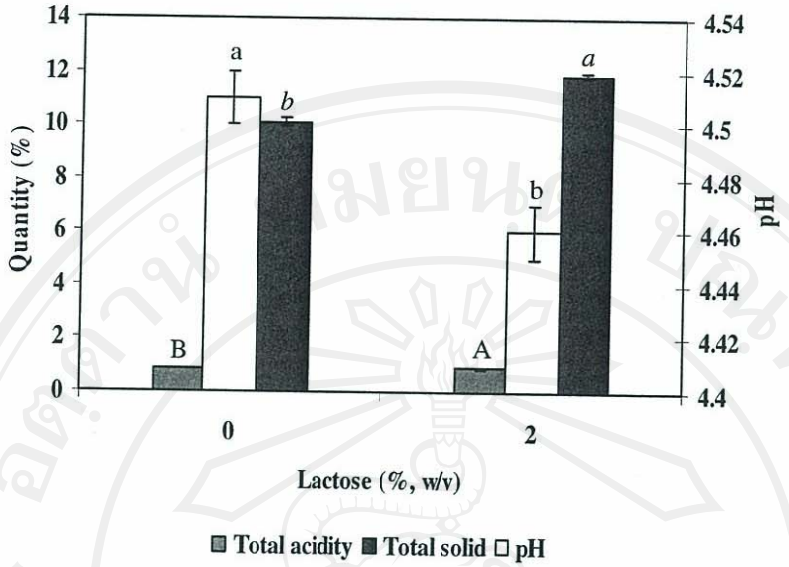

Figure 5.6 Total solid, total acidity (% lactic acid) and pH value of corn milk yogurts added with 0 and 2% (w/v) lactose. Bars with different superscript were significantly different (P<0.05).

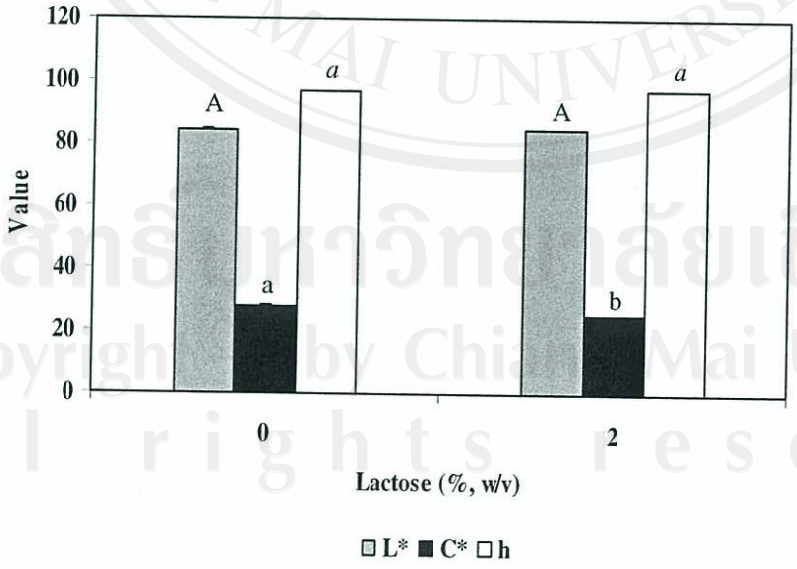

Figure 5.7 Color of corn milk yogurts added with 0 and 2% (w/v) lactose. Bars with different superscript were significantly different (P<0.05).

Table 5.1 Physical properties of corn milk yogurts added with 0 and 2% (w/v) lactose

Lactose (%, w/v)	Whey drainage (%)	Syneresis (%)	Water holding capacity (%)	Bostwick distance (cm)
0	0.55±0.10	74.64±1.17	40.09±0.72	10.05±0.12
2	0.53±0.08	74.11±0.41	40.66±1.45	10.74±0.99

* Values in a column were not significantly different treatments ($P \geq 0.05$)

Table 5.2 Textural profile parameters of corn milk yogurts added with 0 and 2% (w/v) lactose

Lactose (%, w/v)	Hardness (g)	Adhesiveness (g.s)	Springiness
0	87.73±3.49	-36.06±7.86	0.25±0.01
2	88.65±3.78	-43.13±3.19	0.22±0.03

* Values in a column were not significantly different treatments ($P \geq 0.05$)

5.4 CONCLUSIONS

This work demonstrated that lactose promoted the growth of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus*, enhanced acid production in both fermented corn milk and corn milk yogurt. Adding 2% (w/v) of lactose to corn milk was not affected the physical properties and textural profile parameters of corn milk yogurt. The 2% (w/v) lactose was an adequate concentration for the growth of starter cultures and acid production of corn milk products.