Study optimum ratio of major ingredients in Khaomak yogurt ice cream

Suttida Sonthisawate[1] and Walairut Chantarapanont[1,*]

Abstract

The objective of this research was to study optimal ratio of yogurt ice cream mix, blended Khaomak and water in Khaomak yogurt ice cream. Mixture Design was used for formulating mixture of yogurt ice cream mix (60–80%), blended Khaomak (5–15%) and water (0–20%). The viscosity and %overrun of Khaomak yogurt ice cream, 80% yogurt ice cream mix, 5% blended Khaomak and 15% water (trt1) gave the highest values which were 174 cP and 11.35%, respectively. Khaomak yogurt ice cream at ratio of 65:15:20 (trt3) gave the highest melting rate. Total soluble solid from Khaomak yogurt ice cream at ratio of 80:15:5 (trt2) and trt1 gave the highest values which were 26.97ºBrix and 25.83ºBrix, respectively. Treatment with high amount of blended Khaomak caused decreasing in L* and increasing in a* and b* ($p \leq 0.05$). The acidity of Khaomak yogurt ice cream was mainly from yogurt ice cream mix, trt2 gave the highest acidity which was 0.45% total acid and pH 4.85. Total bacteria of all treatments were in range of 7.50–7.90 log cfu g^{-1} and lactic acid bacteria were in range of 7.40–7.80 log cfu g^{-1}, yeast and mold were less than 10 cfu g^{-1}. Microbiological analysis of all treatments met the requirement of fermented milk product standard issued by the Ministry of Public Health, Thailand. The ratio of yogurt ice cream mix, blended Khaomak and water at ratio of 80:5:15 (trt1) obtained the highest liking score in all attributes including overall liking (7.08) with just right higher than 50% in Just about right (JAR) test for all attributes.

Keywords: ice cream, yogurt, Khaomak

1. Introduction

Yogurt is a fermented milk product with high nutrition value (Deeth and Tamime, 1981). It consists of protein from milk which helps regulate the digestive system. In addition, it is an essential source of various vitamin contents which include vitamin A, B1 (thiamin), B2 (riboflavin) and a variety of fat-soluble vitamins. The mineral contents of yogurt consist of calcium, potassium, phosphorous, and sodium (Tamime and Robinson, 1999). Traditionally, there are 2 strains of lactic acid bacteria, *Streptococcus thermophilus* and *Lactobacillus delbruekii* subsp.*bulgaricus*, using as starter cultures in yogurt production (Elizabeth *et al*., 2011). Due to acid production, milk protein was precipitated into a state of gel (curd form) which has white or off-

[1] Department of Product Development, Faculty of Agro-Industry, Kasetsart University, 50 Paholyothin Rd., Chatuchak, Bangkok 10900, Thailand

* Corresponding author, e-mail: fagiwlc@ku.ac.th

white in color, unique scent and sour taste. Yoghurt bacteria are claimed to provide certain nutritional and health benefits (Deeth and Tamime, 1981). For this reason, people who usually consume yogurt find a result of a decrease in abnormality of intestinal function (Lankaputhra and Shah, 1995; Shah, 1999) and a better capability of Lactose digestion (Sittisiri and Katekaw, 1999).

Khaomak (The sweet fermented rice) is a Thai traditional fermented food which has several essential health benefits. To produce Khaomak, starter culture in form of starch cake called lookpang-Khaomak is used to ferment glutinous sticky rice until becoming soft juicy mass with fine aroma, sweet and alcoholic taste. Alcohol content below 3.00% in Khaomak is beneficial to blood circulation for people who have low blood pressure. The source of sweetness in Khaomak was mainly due to glucose from the fermentation (Yodtheon, 2011) and glucose is an important source of lactic acid bacteria (Axelsson, 1998). Even though nowadays Khaomak becomes more common in the market, the consumption rate is still low (Dejsungkranont, 2003). There are several reasons for this such as unattractive physical appearance and vary in taste (Khumweera, 2004). To increase application of traditional Thai food as Khaomak for more consumption and to make more flavors and more health benefit in yogurt ice cream, Khaomak yogurt ice cream concept was developed. However, this experiment was the first part of development process. In this study, the objective of this research was to study optimal ratio of major ingredients which were yogurt ice cream mix, blended Khaomak and water in Khaomak yogurt ice cream.

2. Materials and Methods

2.1 Cultures preparation

Pure freeze dried culture of *Streptococcus thermophilus* TISTR 458 and *Lactobacillus delbruekii* subsp. *bulgaricus* TISTR 895 were obtained from Thailand Institute of Scientific and Technological Research (TISTR), Thailand. Both *S. thermophilus* and *L. bulgaricus* were maintained on MRS agar (Difco, USA) slants at 4ºC. Before inoculation into sample, each species was streaked for single colony isolation and transferred into 10 ml of MRS broth (Difco, USA). After inoculation at 42±1ºC for 24 h, 1 loop of inoculum was transferred to 10 ml of MRS broth and incubated at 42±1ºC for 24 h. Then, 1 ml of each culture was pipetted and grew in 25 ml in MRS broth at 42±1ºC for 18 h to achieve a population of 10^9 cfu g^{-1} (from growth study – data not shown) for using in yogurt preparation (Kaewthai, 1999). Number of lactic acid bacteria was confirmed by using the dilution plate count method in duplicate at each dilution (Dave and Shah, 1996).

2.2 Yogurt preparation

Pasteurized milk (100% cow's milk, Meiji, Thailand) was heated to 80ºC for 2 min and dramatically cooled down to 40±2ºC and then 5% (v/v) of mixed starter cultures between *S. thermophiles* TISTR 458 and *L. bulgaricus*TISTR 895 at ratio of 2:1, respectively, was added and incubated at 40±2ºC for 5–12 h. The pH was recorded every hour until reaching pH 4.50 by using a pH meter (CONSORT, Model C830P, Belgium). After that, yogurt was stored at 4ºC until using for yogurt ice cream mix preparation (no more than 3 days storage) (Jitjumroen, 2004).

2.3 Yogurt ice cream mix preparation

Pasteurized milk (cow's milk, Meiji, Thailand) 27.25% and whipping cream (Anchor, New Zealand) 22.65% were mixed together and heated until temperature reaching 50ºC, then sugar (MitrPhol, Thailand) 20.95% and gelatin (McGarett, Thailand) 0.50% were added to the solution and hold at that temperature for 30 min. After that, it was homogenized by using blender (Juice extractor MR-1258, MARA®, Thailand) for 1 min, pasteurized at 80ºC for 2 min, cooled down to 4ºC within 30 min and ripening at 4ºC for 24 h (Surapat, 2001; Tamime and Robinson, 1999). After ripening at 4ºC, ice cream mix was mixed with yogurt 28.65%, which prepared as above, at ratio of 3:1, respectively. Yogurt ice cream mix was used in next experiment without storing.

2.4 Khaomak preparation

Five hundred grams of glutinous sticky rice were washed and soaked with water for 5–6 h, steamed for 20 min allowed to cool to room temperature, then washed with water twice and drained. The steamed glutinous sticky rice (1000 g) was mixed with 10 g of Lookpang-Khaomak powder (Kewkajee, 2013) in 1000 ml beaker, covered with aluminum foil, incubated at 30±2ºC for up to 72 h (Yodtheon, 2011). After that, Khaomak was blended by blender for 1min, pasteurized at 80ºC for 2 min and stored at 4ºC for using in next experiment within 3 days (Jitjumroen, 2004).

2.5 Study of optimum ratio of yogurt ice cream mix, blended Khaomak and water

The experiment was designed by using Mixture Design for formulating mixture of yogurt ice cream mix (60–80%), blended Khaomak (5–15%) and boiled water (0–20%). Five formulations were obtained as shown in Table 1.

Table 1 Percentage of yogurt ice cream mix, blended Khaomak and water in Khaomak yogurt ice cream

Treatment (trt)	Yogurt ice cream mix	Blended Khaomak	Water
1	80	5	15
2	80	15	5
3	65	15	20
4	75	5	20
5	75	10	15

Khaomak yogurt ice cream from each formulation was produced by mixing all ingredients with blender for 1 min and freezing by home style ice cream maker (HOMEMATE®, Model HOM-4002, Malta) for 30 min. After this step, each treatments was sampled for analyzing viscosity using a digital Brookfield (Brookfield Viscosity, Model DV II, USA) (Ibanoglu and Esra, 1999), color by using Spectrophotometer (Minolta, Model CM–3500d, USA) (Akin *et al*., 2007), overrun (Arbuckle, 1986), total soluble solid by using refractometer (ATAGO, Model PAL–α, Japan) (AOAC, 2005), pH by using a pH-meter (CONSORT, Model C830P, Belgium) and titratable acidity as %lactic acid (Akin *et al*., 2007). After that, the process continued with placing 50 g of ice cream in plastic cup, covering with a plastic lid, hardening at -18±2ºC for 24 h and storing at -18±2ºC until analyzing (Jitjumreon, 2004). All treatments were analyzed for melting rate (Garcia *et al*., 1995), total plate count (BAM, 2002), yeast and mold (BAM, 2002) and lactic acid bacteria (Dave and Shah, 1996). All treatments were analyzed in 2 replications.

Sensory evaluation of Khaomak yogurt ice cream was evaluated by 50 untrained panelists using 9-Point Hedonic scale (1 = dislike extremely to 9 = like extremely) and Just about right (1 = too little, 2 = just right, 3 = too much) in color, odor, sweetness, flavor, smoothness and overall liking. Each panelist received 5 samples for tasting, and was asked to gargle with water after tasting each sample. Samples were presented in a random order and assigned in three–digit sample number. All treatments were analyzed in 2 replications. The data were analyzed statistically using SPSS statistical software program version 16 (SPSS Inc., Chicago, IL, USA) Analysis of variance (ANOVA) and Duncan's Multiple Range Test was used to determined significant difference among results.

3. Results and Discussion

From mixture design, 5 formulations were obtained as shown in Table 1. All treatments after freezing by home style ice cream maker before hardening at -18±2ºC were sampled for analyzing physical and chemical properties. The physical properties of Khaomak yogurt ice cream before hardening were presented in Table 2 trt 1 gave the highest viscosity (174 cP) and %overrun (11.35%) with significant difference ($p \leq 0.05$) among other treatments except in viscosity for trt2 (166 cP). Results also showed that trt2 and trt1 gave the highest total soluble solid which were 26.97ºBrix and 25.83°Brix, respectively. The viscosity and overrun of the samples increased when the total soluble solid increased. The addition of high amount of yogurt ice cream mix and blended Khaomak caused an increase in the viscosity and %overrun as shown in trt1 and trt2. Guven and Karaca (2002) reported that the increase of viscosity and overrun were observed in the fruit frozen yogurt, depending on the increase of fruit concentration and sugar content.

Color of the ice cream samples were significant different ($p \leq 0.05$). Whiteness (L*) of the ice cream samples decreased as blended Khaomak content increased. The color of Khaomak yogurt ice cream was from blended Khaomak which had effect in increasing greenness (-a*) and yellowness (b*) color. The results showed that increasing of blended Khaomak increased greenness (-a*) from -0.38 to -0.20 and yellowness (b*) from 15.52 to 17.59 in Khaomak yogurt ice cream.

Table 2 Physical properties of Khaomak yogurt ice cream with different ratio of yogurt ice cream mix, blended Khaomak and water.

Treatment (trt)	Viscosity (cP)	L*	a*	b*	Overrun (%)	Total Soluble solid (ºBrix)
1	174.00±13.90^{a}	87.36^{a}	-0.20^{a}	15.78^{c}	11.35^{a}	25.83±0.30^{b}
2	166.00±4.90^{a}	86.94^{c}	-0.68^{d}	17.59^{a}	9.77^{b}	26.97±0.90^{a}
3	120.00±7.10^{c}	86.20^{d}	-0.40^{c}	17.40^{a}	7.71^{c}	22.90±0.30^{d}
4	143.00±4.70^{b}	87.44^{a}	-0.22^{a}	15.52^{d}	9.12^{b}	22.50±0.30^{d}
5	145.00±11.40^{b}	87.18^{b}	-0.38^{c}	16.47^{b}	9.60^{b}	24.43±0.30^{c}

Note: Mean in the same column with the same superscript were not significantly different ($p>0.05$).

Mean ±SD in each row, if not indicated, SD ≤ 0.50.

The chemical properties of Khaomak yogurt ice cream before hardening were presented in Table 3 The acidity of Khaomak yogurt ice cream was mainly from yogurt ice cream mix. Treatments with high percentage of yogurt ice cream mix gave lower pH in final product. Schmidt *et al*., (1997) reported that the addition of yogurt in yogurt ice cream had an effect on pH value. The pH values have a correlation with titratable acidity with decreasing in pH value resulting in increasing titratable acidity.

Table 3 Chemical properties of Khaomak yogurt ice cream with different ratio of yogurt ice cream mix, blended Khaomak and water.

Treatment (trt)	Total acidity (%)	pH
1	0.43^{b}	4.93^{c}
2	0.45^{a}	4.85^{d}
3	0.35^{e}	5.38^{a}
4	0.38^{d}	5.17^{b}
5	0.42^{c}	4.94^{c}

Note: Mean in the same column with the same superscript were not significantly different (p>0.05).

All treatments after hardening at -18±2°C for 24 h were sampled for analyzing melting rate, microbiological test and sensory evaluation. Melting rate of all treatments correlated with their %overrun by increasing %overrun resulting in decreasing melting rate (Figure 1). Similar results were found in Pelan *et al.*, (1997). Trt3 gave the highest melting rate due to the lowest percentage of yogurt ice cream mix.

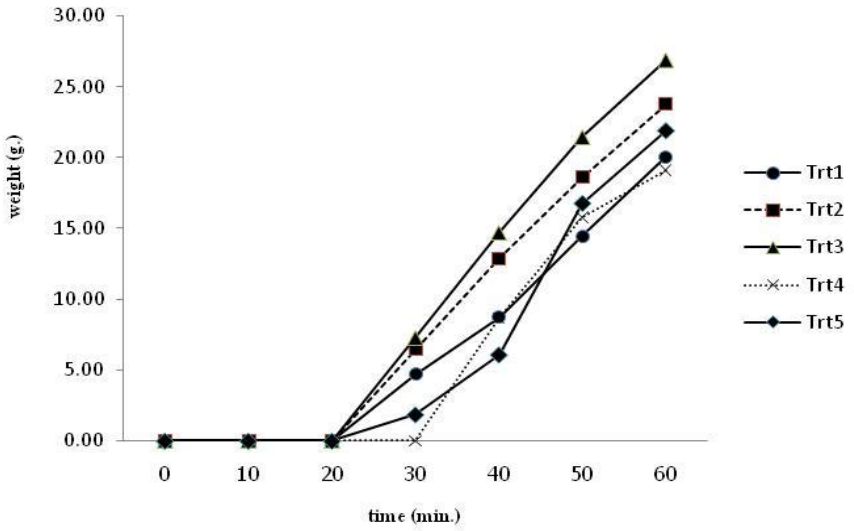

Figure 1 Melting rate of Khaomak yogurt ice cream with different ratio of yogurt ice cream mix, blended Khaomak and water.

The microbiological analyzes of all treatments were presented in Table 4. Total bacteria of all treatments were in range of 7.50–7.90 log cfu g^{-1} and lactic acid bacteria of all treatment were in range of 7.40–7.80 log cfu g^{-1}, yeast and mold were less than 10 cfu g^{-1}. The results showed that there were no significant difference ($p>0.05$) in total number of bacteria in all treatments except trt4 ($p\leq0.05$). However, there were significant difference in number of lactic acid bacteria in trt3 and trt4 ($p\leq0.05$). These might be from the effect of ratio of yogurt ice cream mix, blended khaomak and water because trt3 and trt4 had the least total soluble solid (22.90ºBrix and 22.50ºBrix, respectively) which had effect in cryogenic protection for lactic acid bacteria during freezing. Standard of fermented milk product issued by the Ministry of Public Health required minimum of 10^{7} cfu g^{-1} of bacteria with maximum of 100 colony yeast and mold (FDA, 2005). All treatments met the requirement of Thai FDA.

Table 4 Total bacteria, Yeast/Mold and Lactic acid bacteria of Khaomak yogurt ice cream with different ratio of yogurt ice cream mix, blended Khaomak and water.

Treatment (trt)	Total bacteria (log cfug^{-1})	Yeast/Mold (cfu g^{-1})	Lactic acid bacteria (log cfug^{-1})
1	7.90^{a}	<10 est.	7.70^{ab}
2	7.80^{a}	<10 est.	7.70^{ab}
3	7.80^{a}	<10 est.	7.60^{b}
4	7.50^{b}	<10 est.	7.40^{c}
5	7.90^{a}	<10 est.	7.80^{a}

Note: Mean in the same column with the same superscript were not significantly different (p>0.05).
est. = estimate plate count which mean that number of colony showing on the plate at the lowest dilution were not in range of 25–250 colony/plate.

Results of sensory evaluation of Khaomak yogurt ice cream were shown in Table 5. Treatment at ratio of 80:5:15 of yogurt ice cream mix, blended Khaomak and water (trt1), obtained the highest liking score in all attributes including overall liking (7.08) with %just right higher than 50.00% in JAR test for all attributes as shown in Table 6. Adding high percentage of blended Khaomak (15%) gave the lowest overall liking score due to high alcohol flavor in treatment. Even though blended Khaomak was pasteurized during preparation which resulted in low alcohol content (1.00–2.00% - data not shown), it still had effect on Khaomak yogurt ice cream flavor when Khaomak was added in high amount.

Table 5 Liking scores (mean value) of Khaomak yogurt ice cream with different ratio of yogurt ice cream mix, blended Khaomak and water.

Sensory attribute	Treatment (trt)				
	1	2	3	4	5
Color	7.5±0.7^{a}	7.4±1.0^{ab}	7.2±1.3^{b}	7.4±07.^{ab}	7.6±0.8^{a}
Odor	6.4±1.5^{a}	6.0±1.7^{a}	5.5±1.8^{b}	6.3±1.4^{a}	6.1±1.4^{a}
Sweetness	7.1±1.3^{a}	6.5±1.6^{bc}	6.1±1.6^{c}	6.9±1.3^{ab}	7.0±1.1^{a}
Flavor	7.0±1.3^{a}	6.1±1.8^{b}	5.6±1.6^{c}	6.7±1.0^{a}	6.9±1.1^{a}
Smoothness	7.1±1.3^{a}	6.9±1.3^{b}	6.1±1.5^{c}	6.2±1.3^{b}	6.8±1.2^{b}
Overall liking	7.1±1.3^{a}	6.3±1.7^{b}	5.3±1.8^{c}	6.3±0.9^{b}	6.5±0.9^{b}

Note: Mean±SD in the same row with the same superscript were not significantly different (p>0.05).

Table 6 Percentage on Just about right scores of Khaomak yogurt ice cream with different ratio of yogurt ice cream mix, blended Khaomak and water.

Trt	Color			Odor			Sweetness			Flavor			Smoothness		
	too little	just right	too much	too little	just right	too much	too little	just right	too much	too little	just right	too much	too little	just right	too much
1	0	98	2	36	58	6	4	78	18	30	66	4	18	82	0
2	2	94	4	26	42	32	8	68	24	28	62	10	16	80	4
3	4	90	6	22	26	52	20	62	18	6	60	34	46	50	4
4	0	98	2	38	58	4	12	76	12	14	64	22	60	36	4
5	2	94	4	44	46	10	6	84	10	18	62	20	30	68	2

Conclusion

Khaomak yogurt ice cream consisted of 80.00% yogurt ice cream mix, 5.00% blended Khaomak and 15.00% water (trt1), was the optimal ratio from this study with the highest overall liking scores (7.08) and %just right higher than 50.00% in JAR test for all attributes. The numbers of viable count of total bacteria, yeast and mold and lactic acid bacteria of all treatments met the Thai standard requirement. This ratio was chosen for study in next experiment of Khaomak yogurt ice cream product development.

Acknowledgement

Thanks to Kasetsart University Research and Development Institute for funding this research.

References

Akin, M.B., Akin, M.S. and Kirmarci, Z. 2007. Effects of inulin and sugar levels on the viability of yogurt and probiotic bacteria and the physical and sensory characteristics in probiotic ice cream. Food Chemistry. 98–99.

Arbuckle, W.S. 1986. Ice cream (4th Eds.). Van Norstrand Reinhold, New York.

Association of Official Analytical Chemists (AOAC). 2005. Official methods of analysis of AOAC international (18th Eds.). AOAC International, Gaithersburg, MD.

Axelsson, L. 1998. Lactic acid bacteria: classification and physiology (pp. 1–72). In Salminen, S. and Wright, A. Lactic acid bacteria: Microbiology and Functional aspects. Marcel Dekker, Inc., New York.

Bacteriological Analytical Manual (BAM). 2002. Food and Drug Administration Bacteriological Analytical Manual (8th Eds.). AOAC International, USA.

Dave, R.I. and Shah, N.P. 1996. Evaluation of media for selective enumeration of *Streptococcus thermophilus*, *Lactobacilllusdelbruekii*spp. *bulgaricus*, *Lactobacillus acidophilus* and *Bifidobacterium*spp. J. Dairy Sci.79: 1529–1536.

Deeth, H.C. and Tamime, A.Y. 1981. Yogurt: Nutritive and therapeutic aspects. J. Food Prot. 44(1): 78–86.

Dejsungkranont, M. 2003. Properties of yeast and mold involve in khaomak and satho fermentation. M.S. Thesis, Kasetsart University, Bangkok. (In Thai).

Elizabeth, W. Ng., Yeung, M. and Tong, P.S. 2011. Effect of yogurt starter cultures on the survival of *Lactobacillus acidophilus.* Invest J of Food Microbiology. 145: 169–175.

Food and Drug Administration. 2005. Notification of the Ministry of public health No. 289: fermented milk. Ministry of Public Health, Bangkok, Thailand.

Garcia, R.S., Marshall, R.T. and Heymann, H. 1995. Low fat ice cream from freezed concentrated versus heat-concentrated nonfat milk solid. J. Dairy Sci. 78: 2345–2351.

Gu¨ven, M. and Karaca, O.B. 2002. The effects of varying sugar content and fruit concentration on the physical properties of vanilla and fruit ice-cream-type frozen yogurts. International Journal of Dairy Technology. 55: 27–31.

Ibanoglu, S. and Esra, I. 1999. Rheological properties of cooked tarhana a cereal-based soup. Food Research International. 32: (1999) 29–33.

Jitjumroen, W. 2004. Use of corn germ for substituting skim milk powder in the production of probiotic frozen yogurt. M.S. Thesis, Kasetsart University, Bangkok. (In Thai).

Kaewthai, N. 1999. Screening of lactic acid bacteria from raw milk as yogurt starter. M.S. Thesis, Kasetsart University. Bangkok. (In Thai).

Kewkajee, S. Look-pang Khaomak. URL (www.kawmaktrat.pantown.com) (September 22, 2013).

Khumweera, P. 2004. Properties of sweetened rice (Kaomak). M.S. Thesis, Kasetsart University. Bangkok. (In Thai).

Lankaputhra, W.E.V. and Shah, N.P. 1995. Survival of *L. acidophilus* and *Bifidobacterium*spp. In the presence of acid and bile salts. Cult. Dairy Prod. J. 30: 2–7.

Pelan, B.M.C., Watts, K.M., Cambel, I.J. and Lips, A. 1997. The stability of aerated milk protein emulsion in the presence of small molecule surfactants. J. Dairy Sci. 80: 2631–2638.

Schmidt, K.A., Kimand, J. and Jeon, I.J. 1997. Composition of carbohydrates and concentration of β–galactosidase of commercial frozen yogurt. J. Food. Qual. 20: 349–358.

Shah, N.P. 1999. Probiotic bacteria: antimicrobial and antimutagenic properties. Probiotica. 6: 1–3.

Sittisiri, P. and Katekaw, S. 1999. Eat-Live for health (1st Eds.). Sookjai, Inc. Bangkok. (In Thai).

Surapat, S. 2001. Process of ice cream. Handbook of ice cream and products (pp. 19–23). Kasetsart University, Bangkok. (In Thai).

Tamime, A.Y. and Robinson, R. K. 1999. Yoghurt: Science and Technology (2nd Eds.). Woodhead Publishing, Ltd., Cambridge.

Yodtheon, J. 2011. Development of Khaomak Beverage. M.S. Thesis, Kasetsart University. Bangkok. (In Thai).