CHAPTER III
MATERIALS AND METHODS

4.1 Materials

4.1.1 Fresh corn silk

Corn silk from sweet corn (*Zea mays*) was obtained from the corn milk processing facility of the National Corn and Sorghum Research Center in Nakhon Ratchasima Province. The fresh corn silk sample was packed in polyethylene bags and kept at 4°C until further experiment

4.1.2 Dried corn silk

Dried corn silk was prepared by drying the fresh corn silk at 50°C in a hot air oven (Memmert, Germany) over night. The dried corn silk was then milled in a high speed mill (Moulinex Optiblend Duo, KRUPS, French and Kenwood CG100, USA.), packed in polyethylene bags and kept at 4°C until further experiment

4.2 Extraction of DF from fresh and dried corn silk

4.2.1 Extraction of DF from fresh corn silk (FDF)

The preparation of dietary fiber from fresh corn silk (FDF) was conducted by the modified alcoholic extraction process of Thumthanaruk (1996) (93). The fresh corn silk sample was boiled in water about 3 h with changing water every hour. The prepared sample was extracted with 95% ethanol (1:5 kg/l). The mixture was occasionally agitated overnight then filtered through a nylon bag using a hydraulic press. The same alcoholic extraction process was repeated twice. Finally, the ethanol-insoluble solid fraction was collected and dried in a hot-air oven at 60°C for 6 h. After that dietary fiber was milled in a high speed mill (Moulinex Optiblend Duo, KRUPS,

French and Kenwood CG100, USA) then kept in polyethylene bags at room temperature.

4.2.2 Extraction of DF from dried corn silk (DDF)

The preparation of dietary fiber from dried corn silk (DDF) was conducted using the same method as described above for fiber extraction from fresh corn silk except the boiling step was omitted.

4.2.3 Yield of DF from dried and fresh corn silk (DDF and FDF)

The yield of corn silk dietary fiber, DDF and FDF was calculated by the following equation based on the weight of corn silk before extraction and the weight of dried dietary fiber after the extraction.

$$\text{Yield (\%)} = \frac{\text{Weight of dietary fiber (g)}}{\text{Weight of corn silk (g)}} \times 100$$

4.3 Determination of properties of DF from dried and fresh corn silk (DDF and FDF)

4.3.1 Physical properties

4.3.1.1 Determination of particle size

The particle size of DDF and FDF was determined by shaking 50 g of the samples on Retsch test sieves (Retsch, type Vibro, GmbH & Co., Germany) size 35, 40, 60, 80, 100, 120 and 140 mesh stacked in an order of decreasing opening size according to Wongmethinee *et al*. (33) The weight of sample remaining on each sieve was recorded and calculated as percentage of total weight. Triplicate analyses of the samples were carried out.

4.3.1.2 Water holding capacity

According to the modified method of Elkhalifa *et al.* (94), 2 g of sample were mixed with 30 ml distilled water contained in a centrifuge tube by a Vortex mixer. The slurry was allowed to stand for 30 min and then centrifuged at 4000 x g for 25 min. After centrifugation the supernatant was drained off. The remaining wet precipitate was then weighed. The result was expressed as g of water retained per g of sample.

4.3.1.4 Emulsifying activity

Emulsifying activity of DDF and FDF were determined following the simple system of emulsifying activity measurement reported by Elkhalifa *et al.* (94). Approximately 2 g of sample were suspended in 50 ml of water and then 50 ml of soybean salad oil was added. The mixture was emulsified with a homogenizer (IKA ULTRA TURAX-T25 with designation of dispersing tool, S25N-25F) at 135,000 rpm for 1 min. The emulsion obtained was divided evenly into 50-ml centrifuge tubes and centrifuged at 4,000xg for 10 min. The emulsifying activity was calculated following the formula:

$$\text{Emulsifying activity (\%)} = \frac{\text{Height of emulsified layer (cm) x 100}}{\text{Height of whole layer in the centrifuge tube}}$$

$$\text{Emulsion stability (\%)} = \frac{\text{Height of emulsion layer after heating (cm)} \times 100}{\text{Height of whole layer in the centrifuge tube}}$$

4.3.1.5 Water activity

The water activity of dietary fiber samples, DDF and FDF was measured using a water activity meter (Novasina IC-500 Aw-Lab, Axair Ltd., Pfäffikon, Switzerland) at 25±1°C in triplicates. The sample was weighed into a plastic sample container. The container was then placed into the equilibrium chamber and held until a constant value was detected.

4.3.1.6 Color

Color of samples was measured by a chromameter (Color Techno System Corporation, Tokyo, Japan). The color value was expressed as L*, a* and b* values. The L* value shows the depth of lightness, which 100 means lightness and 0 means darkness. The a* value means redness (+) and greenness (-) and the b* value means yellowness (+) and blueness (-) of the product (13).

4.3.1.7 pH

The pH of the samples was measured using a pH meter (Mettler Delta 340, Toledo Ltd., England). Sample with a known weight in g was mixed with 50 ml distilled water and allowed to stand for 30 min at room temperature before measuring.

4.3.2 Chemical properties

4.3.2.1 Moisture content

The moisture content was determined using the AOAC method (1990) (95) as the procedure given in Appendix B.

4.3.2.2 Total dietary fiber

The total dietary fiber content (TDF) of corn silk fiber was analyzed using an enzymatic gravimetric method according to AOAC (1990) (95). The procedure is given as Appendix A.

4.5 Application of dietary fiber from dried and fresh corn silk in food models

4.5.1. Preparation of corn silk fiber-added cake

Cakes were prepared to test the effect of fiber ingredients in a baked product. DDF and FDF were used to substitute 15-20% of wheat flour in the regular cake recipe while other ingredients were kept constant. The control formula for cake preparation was based on the one reported by Prakongpan (5) as shown in Appendix D. Cake batter was prepared using an electric mixer (Verasu, Homemate HOM-12KP81, Thailand). All ingredients were mixed and blended at a low speed for 0.5 minutes in a mixing bowl. The speed was increased to high then continued mixing for 3 min and pour to a square pan size 0.5 pound, baked at 180°C for 30-40 min until a wooden pick inserted in the center came out clean. The finished cake was cooled to room temperature. The diagram of golden layer cake preparation is shown in Figure 4.1

The fiber-enriched cakes were prepared by substituting 10-15% of the wheat flour (by weight) in the control formula with either DDF or FDF.

The procedure for cake making was the same as described above for the control cake.

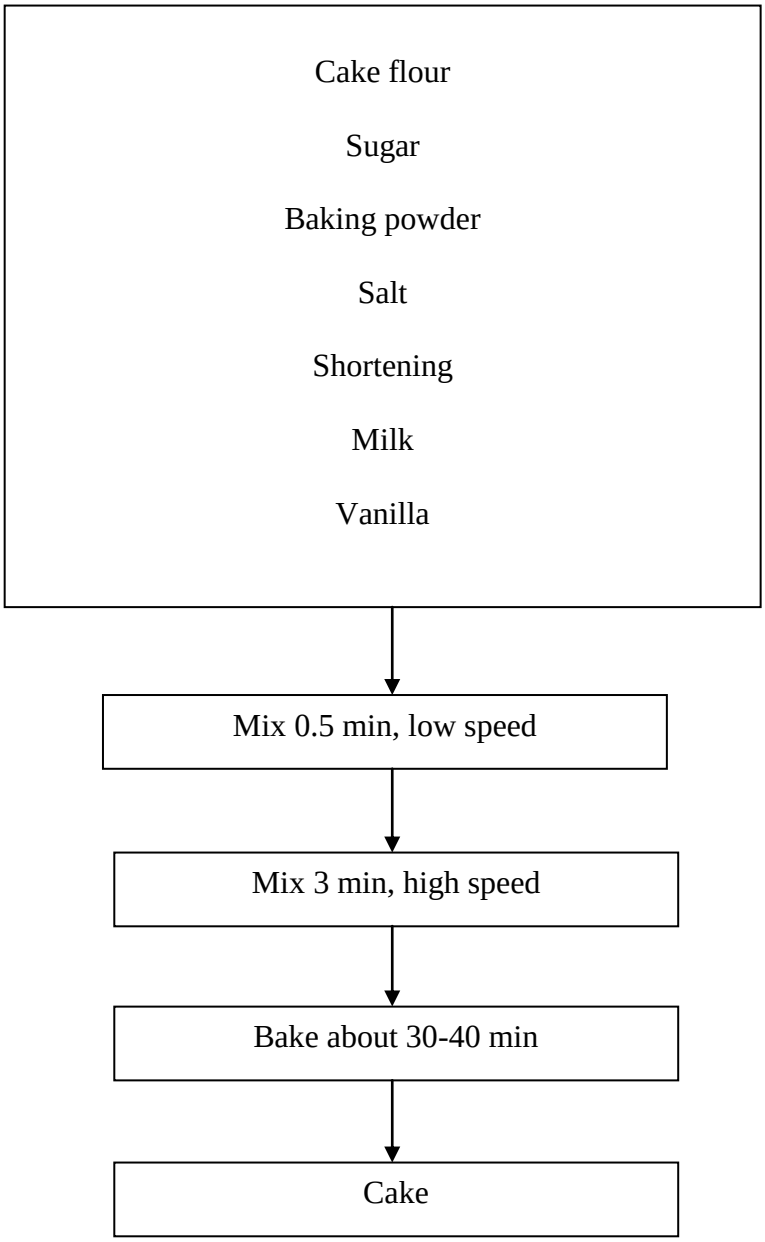

Figure 4.1 Diagram of golden layer cake preparation

4.5.2 Quality determination of corn silk fiber-added cake

a. Volume (V, ml) of cakes was measured using the sesame seed displacement method.

b. Color values of crust and crumb of cake were measured by using a chromameter.

c. Water activity of cakes was measured by a water activity analyzer

4.5.3 Sensory screening test

Sensory screening test of products were performed The food samples were evaluated by thirty untrained panelists recruited from staff and graduate students of the Institute of Nutrition, Mahidol University. Cake samples were prepared on the day before the evaluation, packed in polypropylene bags and stored at 4°C. All samples were coded with a three-digit random number and randomly served to each panelist, one at a time. Panelists were asked to rinse his/her mouth before testing the next sample. The test was performed in air-conditioned testing booths under a daylight fluorescent lamp at the Sensory Science Laboratory of the Institute of Nutrition. Nine-point hedonic scales were used for evaluating color, flavor, general appearance, texture and overall acceptability of the product. The scale ranged from 1 to 9 where 1 = “dislike extremely”, 5 = “neither like nor dislike”, and 9 = “like extremely”.

4.5.4 Determination of TDF content of corn silk fiber-added cakes

Corn silk fiber-added cakes were determined for TDF content using the enzymatic-gravimetric method of AOAC. (95)

4.5.5 Preparation of fried batter coated chicken

DDF and FDF corn silk fiber were added to batter suspension for coating deep-fried chicken pieces at 3% by weight while the other ingredients were kept constant. The fiber ingredient was allowed to hydrate for about 5 min before frying. The formula of batter coated suspension appears in Appendix E. (96)

The batter mix was used to prepare deep-fried chicken pieces. Skinned, boneless chicken breast was cut into 3x3 cm pieces then dipped in a batter suspension to obtain good coating. Then the batter coated chicken pieces were deep-fried in palm oil in a deep-fryer at 180°C for 3 min. The fried battered coated products were cooled on a paper towel before storage. The diagram of fried batter coated chicken preparation is shown in Figure 4.2

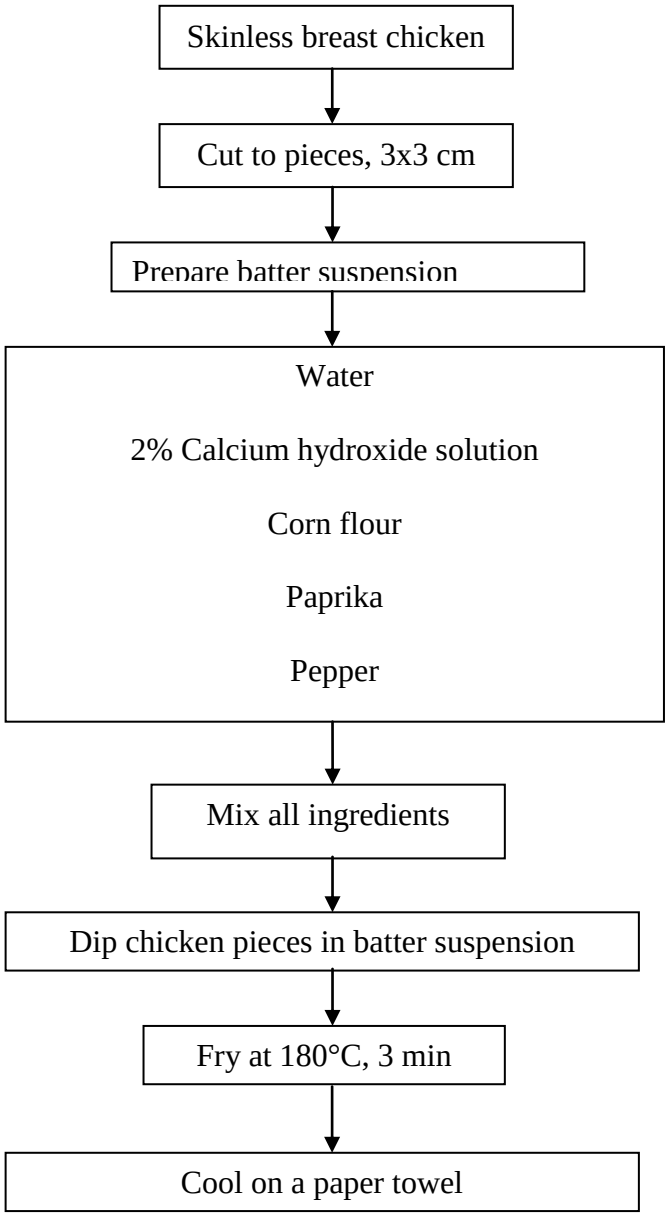

Figure 4.2 Diagram of deep-fried chicken preparation

4.5.6 Quality determination of fried batter coated chicken

a. Yield

Yield of cooked product was measured as a percentage of weight retained after frying. Yield was calculated as

$$\text{Yield (\%)} = \frac{\text{Cook weight}}{\text{Raw weight}} \times 100$$

b. Batter pick-up

The amount of batter adhering to the samples during coating before frying was considered as the batter pick-up. The batter pick-up of DDF and FDF fiber added-batter was measured as a percentage ratio of the weight of batter coating and the weight of batter-coated food. (33) The batter pick-up was calculated using the formula shown below.

$$\text{Batter pick-up (\%)} = \frac{\text{Weight of batter-coated sample - weight before coating}}{\text{Weight of batter-coated sample}} \times 100$$

c. Oil uptake of frying batter

Batter suspension before frying and the batter crust removing from deep-fried chicken pieces were analyzed for moisture content, fat content and oil uptake in triplicate analyses. Moisture and fat contents were analyzed using the hot-air oven method and Soxhlet extraction method as the procedure given in Appendix A. and C., respectively. The criterion "U_R", expressed the oil uptake ratio between weight of oil uptake and the weight of water removing. The value was used to evaluate the effectiveness of dietary fiber ingredient to reduce oil absorption during deep-fat frying. The difference between moisture content before and after frying was calculated as water removing and the difference between initial and final fat content of frying batter was used as oil uptake. (33) The value of U_R was calculated as follows.

$$U_R = \frac{\text{Oil uptake (g)}}{\text{Water removed (g)}}$$

4.5.7 Sensory acceptability test

Sensory acceptability test of products were performed The food samples were evaluated by thirty untrained panelists recruited from staff and graduate students of the Institute of Nutrition, Mahidol University. The fried batter coated chicken

samples were prepared on the same day of the evaluation. All samples were coded with a three-digit random number and randomly served to each panelist, one at a time. Panelists were asked to rinse his/her mouth before testing the next sample. The test was performed in air-conditioned testing booths under a daylight fluorescent lamp at the Sensory Science Laboratory of the Institute of Nutrition. Nine-point hedonic scales were used for evaluating color, flavor, general appearance, texture and overall acceptability of the product. The scale ranged from 1 to 9 where 1 = "dislike extremely", 5 = "neither like nor dislike", and 9 = "like extremely".

4.6 Statistical analysis

The means of scores from chemical analysis and physical analysis were analyzed for significant difference at $p \leq 0.05$ by using an independent sample t-test. For sensory acceptability screening test used One-way ANOVA and Duncan's Multiple Range Test. All statistical analyses were performed using SPSS/PC^{+TM} Program version 13.0.