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Quality profile of palm sugar concentrate produced in Songkhla province, Thailand

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Abstract: Quality of palm sugar concentrate produced in Songkhla province in terms of physical, chemical and microbiological aspects was determined. Palm sugar concentrates possessed wide range of turbidity and colour shades. Transmittance values at 650 nm were found to vary between 1.34-50.45 % and L* (Lightness) varied between 1.78-53.93, a* (red colour intensity) varied between 9.87-34.75 and b* (yellow colour intensity) varied between 3.09-78.94. Total soluble solid contents of the samples varied from 59.01 to 73.05°Brix and were likely related to a variation of total microbial count (1.20×10^3 - 4.80×10^6 CFU/ml), yeast and mold count (1.30×10^2 - 5.30×10^4 CFU/ml) and osmophillic yeast count (2.00×10^2 - 1.46×10^5 CFU/ml). Total microbial counts of the product moderately positively correlated with L*, b* and transmittance value, whereas slightly negative correlated with total acidity (as lactic acid), reducing sugar content and osmophillic yeast count. The microbiological quality of all samples did not comply with Thai legislation standard for palm sugar concentrate and the content of total soluble solids of 7 in 30 samples were not in line with the requirements either. Different quality of palm sugar concentrate might be due to the differences in personal hygiene, sanitary facilities, heating processes and storage conditions.

Keywords: Palm sugar concentrate, palm sap, palm syrup, microbial count, total soluble solids

Introduction

In Thailand, palm sugar concentrate is a natural product made from sap of either coconut tree (*Cocos nucifera* L.) or Palmyra palm tree (*Borassus flabellifer* Linn.). However, the product made from Palmyra palm sap is more popular and well-known of. Palmyra palm trees are widely grown in Africa, South Asia, South America, Australia and in other tropical countries (Morton, 1988). In southern Thailand, the palm is widely grown in Petchaburi and Songkhla provinces. Nowadays, Palmyra palms are the most populated in Songkhla province, approximately 1,262,771 millions plants. There are approximately 300 palm sugar concentrate producers in Songkla province (Department of agricultural extension Thailand, 2006). Palmyra palm trees generally have been planted on the dykes of rice fields for shading the rice and tapping palm sap for cooking (Panyakul, 1995; Department of Agricultural Extension Thailand, 2001). Interestingly, one of the tree products, palm sap, is nutritious and indigenous sweet product. When palm sap is heated, it turns to be a palm sugar concentrate and can be kept longer. The unique flavour of palm sugar concentrate has made its popularity as a flavoring reagent in confectionery and baking products. In addition, emphasis on the consumption of natural foods has resulted in the use of palm sugar concentrate as an alternative sweetener (Panyakul, 1995).

Traditionally, palm sap is manually collected from each inflorescence of the Palmyra palm tree. Palm sugar concentrate is produced by evaporating the palm sap in a large opened pan (approximately 60-80 liters/pan) and is heated using the wood fired stove until it becomes concentrated. Producers then determine to stop the process and get finished product by observing intensity of brown color thickness and viscosity of the on-going product. The product would be taken out from a pan to cool down and kept in containers for sale. Overheating process would alter its unique flavour and colour. Once the sap is produced to a palm sugar concentrate, their total soluble solids which are mainly sugar should be at least 65°Brix or above for food safety purpose (Thai Industrial Standards Institute, Ministry of Industry, 2003). However, the other product specific quality is still undefined. It still lacks of standard procedures in controlling critical factors influencing the final product quality during its traditional production.

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The quality of palm sugar concentrate product varies across producers depending on individual production techniques. The differences are based on personal hygiene, sanitary facilities, harvesting condition, heating temperature, heating time and storage conditions. The major chemical reactions occurred during the heating process of sugar riched products such as palm sugar concentrate are Inversion, Maillard and Caramelisation reactions (Fennema, 1996). Micro-organisms in palm sap also play important role in producing reducing sugars (glucose and fructose) from sucrose via hydrolysis reaction. The sugars are responsible for dark colour and the intense caramel flavour that arise during evaporation, masking the flavour character of palm sugar concentrate. This palm sugar concentrate product is similar to the commercial maple syrup in terms of production method. The United States Department of Agriculture Standards (1980) specify the highest grade of maple syrup as a pure syrup which is free of any material other than pure, clear, clean liquid maple syrup in sanitary condition. The characteristics of the highest maple syrup grade are transparency for light transmittance not less than 75.0%, and delicately sweet and original maple flavours. The total soluble solids of finished maple syrup should not be less than 66°Brix. Total soluble solids of maple syrups can be ranged from 62 to 74°Brix with a mean value of 67°Brix (Stuckel and Low, 1996). Maple syrup shall be free of sugar crystals and shall not be damaged in any way. This specification compliments to a direction of improving quality of palm sap and palm sugar concentrate. Palm sugar concentrate producers can also apply the good sanitary practices in collecting and storing sap as well as heating process of the maple syrup production for palm sugar concentrate improvement. Therefore, the purpose of this research was to determine the qualities of 30 palm sugar concentrate samples.

Materials and Methods

Sample collection

Palm sugar concentrate 30 samples were randomly collected from primary producers in Songkhla Province, Thailand. The palm sugar concentrate samples were kept in a sterile gas jar under room temperature (28°C) and transported to Department of Food Technology, Prince of Songkla University, Hat-Yai, Songkhla, Thailand. Physical, chemical and microbiological qualities of 30 palm sugar concentrates were analyzed in three replications.

Colour measurement

Colour intensity was measured by a Hunter Lab Colourflex colorimeter providing a result in CIE Lab system (L* (lightness-darkness), a* (redness-greenness) and b* (yellowness-blueness)). All samples were measured in three replications (Palou *et al*., 1999).

Transmittance measurement

The transmittance value or the turbidity of palm sugar concentrate was determined by measuring percentage of light transmission at a wavelength of 650 nm with a Hunter Lab Colourflex (Tiapaiboon, 2004). For a liquid sample with high solids content, like coatings, the effects of high scattering can be accommodated by measurement using a thin path length cell (typically 2 mm or less) on a sphere instrument in the total transmittance mode to collect the highly scattered light (Hunter and Harold, 1987). All samples were analyzed in three replications.

pH measurement

The pH of palm sugar concentrate was measured by a pH meter. Calibration was standardized using pH 7.0 and 4.0 buffers (A.O.A.C, 2000). Each sample was measured in three replications.

Total acidity determination

Total acidity was determined by titration method with 0.1 N sodium hydroxide, which was standardized using potassium hydrogenpthalate (3.2% w/v) prior each titration. A few drops of 1% phenolphthalein were used as an indicator.

A sample of palm sugar concentrate (15 ml) was titrated with 0.1 N sodium hydroxide until reached end point and persisted for 15-20 seconds, and the colour was changed to pink colour at pH 8.1. Three titrations were performed for each sample. The percentage of total acidity obtained was equivalent to lactic acid content (A.O.A.C, 2000).

$$\text{Total acidity (\%w/v as lactic acid)} = \frac{V1 \times N \times 90 \times 100}{1000 \times V2}$$

when N = normality of NaOH solution
V1 = volume of 0.1 N NaOH used for titration (ml)
V2 = volume of sample (ml)
90 = molecular weight of lactic acid (g/mol)

Total soluble solids measurement

Total soluble solids of palm sugar concentrate was measured using Atago hand-held refractometer. All samples were measured in three replications and reported in degree brix (°Brix) (A.O.A.C, 2000).

Sugar content analysis

The determination of total sugars and reducing sugars by Lane and Eynon volumetric method was employed. This method consists of the chemical preparation, sample preparation and titration method (A.O.A.C, 2000). The chemicals used were (1) Fehling's solution (methylene blue and standard sugars) and (2) clearing agent solution (10% neutral lead acetate and 10% potassium oxalate).

Samples of palm sugar concentrate were prepared before analysis. 25 ml of 10% neutral lead acetate solution was added to 60 g (W) of a palm sugar concentrate sample. The mixture was diluted to 250 ml with distilled water and filtrated through filter paper. After filtration, 10 ml of 10% potassium oxalate was added to 100 ml (V1) of the sample and filtrated through filter paper. The clarified sample was separated into two portions; the first one was taken to determine reducing sugars and the second one was taken to determine total sugars by the titration method. Before titration, 30 ml (V2) of the second portion was hydrolyzed by adding 5 ml of conc. hydrochloric acid. The mixture was heated at 70°C for 15 min and cooled down to room temperature. The clarified sample was neutralized by adding 10% sodium hydroxide and then adjusted to 250 ml with distilled water, and was then ready to determine the total sugars.

The titration method requires 3 steps: (1) standardization of the Fehling's solutions, (2) preliminary titration of the sample and (3) accurate titration of the sample (F= Titre x weight (g) of dextrose in 10 ml). Three titrations were performed for each sample. The sugar contents in samples were calculated using two equations below;

$$\text{Reducing sugars (\%w/w)} = \frac{F \times 250 \times 250 \times 100}{W \times V1 \times V3}$$

$$\text{Total sugars (\%w/w)} = \frac{F \times 250 \times 250 \times 250 \times 100}{W \times V1 \times V2 \times V3}$$

when F = factor of the Fehling's solutions

W = weight of sample (g)

V1 = volume of sample after added10% neutral lead acetate solution and filtrated (ml)

V2 = volume of sample used for hydrolysis (ml)

V3 = volume of sample used for titration (ml)

Microbiological analysis

Samples were diluted in sterile 0.1% peptone water and poured on plate count agar to determine total microbial count, and on acidified potato dextrose agar to determine yeast and mold count. Osmophillic yeast count was also analysed using spread plate technique on osmophillic potato dextrose agar. The plates were incubated for 48 hours at 37°C for total microbial count, for 5 days at 37°C for yeast and mold count, and for 3 days at 37°C for osmophillic yeast count. Microbial count was expressed in Colony Forming Units per millilitre (CFU/ml) (Kiss, 1984).

Experimental design and statistical analysis

Completely Randomized Design (CRD) was designed for this experiment, with three replications. To test whether palm sugar concentrate qualities varied significantly among samples, statistical parameters such as F-test and Analysis of Variance (ANOVA) was employed. Duncan's New Multiple Range Test (DMRT) was used to test significant difference between each pairs of mean. Statistical software, SPSS for Window V 11.0 was used for testing mean difference (Steel and Torrie, 1980). A 95% confident interval ($P \leq 0.05$) was set throughout the data analysis to identify significant differences. Principal Component Analysis (PCA) was applied to observe relationship among all quality indicators from 30 palm sugar concentrate samples by XLSTAT software (www.XLSTAT.com).

Results and Discussion

The physical, chemical and microbiological qualities of 30 product samples varied in a wide range. Table 1 summaries of the product quality by means, standard deviations and ranges. The product overall presented large differences in appearance (colour and turbidity), extremely varied in tastes (sugar contents and acidity) and introduced concerns on product safety (microbial counts and critical pH level).

Colour

The colour measured in CIE Lab system is described by colour theory, that the L* value indicates level of lightness (L*=100) to darkness (L*=0), a* positive value indicates red shade whereas negative value indicates green shade colour, and b* positive value indicates yellow shade while negative values indicates blue shade colour (Hunter and Harold, 1987).

The CIE L*, a* and b* values of palm sugar concentrate samples are determined. The results show that lightness (L* value) ranges from 1.78 to 53.93, redness (a* value) ranges from 9.87 to 34.75

Table 1. Physical, chemical and microbiological properties of 30 palm sugar concentrate

Qualities		Range
Colour	L*	1.78-53.93
	a*	9.87-34.75
	b*	3.09-78.94
Transmittance (%) at 650 nm		1.34-50.45
pH		4.50-5.37
Total acidity (%w/v as lactic acid)		0.24-0.86
Total soluble solid (^{o}Brix)		59.01-73.05
Total sugar (%w/w)		23.77-71.89
Reducing sugar (%w/w)		3.54-23.94
Total microbial count (CFU/ml)		$1.20x10^3$– $4.80x10^6$
Yeast and mold count (CFU/ml)		$1.30x10^2$– $5.30x10^4$
Osmophillic yeast (CFU/ml)		$2.00x10^2$–$1.46x10^5$

Note: Each value is the mean of 30 samples with triplicate determinations

and yellowness ranges (b* value) 3.09 to 78.94 (Table 1). Most of samples appeared in red-brown to brown colour shades. The lowest L* value was found in sample No. 10. From observation, visible quality (colour and turbidity) in sample No. 10 was the poorest as it presented darkest turbid brown while sample No. 24 presented the lightest yellow. The decreasing of L* value is responsible for darker colour, and it may also contribute to the non-enzymatic browning reaction during heating process (Maillard and Caramelisation reactions) (Fennema, 1996; Apriyantono *et al*., 2002; Burdurlu *et al*., 2003). Overall, L*, a* and b* values of 30 palm sugar concentrate samples were significant different ($P \leq 0.05$). The heating process of palm sugar concentrate samples could be a main factor affecting the variation of L* values. The samples which were heated at 100°C or above for a long time can cause increasing of the product colour from brown to dark.

Transmittance value (turbidity)

The transmittance value is determined by measuring the percentage of light transmittance at 650 nm. The transmittance values of 30 palm sugar concentrate samples were found to vary from 1.34 to 50.45 as shown in Table 1. The turbidity of palm sugar concentrate depends greatly on its protein content and the polyphenolic compounds, which is dissolved from Kiam wood (*Cotylelobium lanceotatum* craih.) during collecting stages of the palm sap (Taipaiboon, 2004; Loetkitsomboon, 2004). Balange and Benjakul (2009) reported that the total phenolic content in Kiam wood (intact form) as extracted by water at room temperature was 29.33 mg tannin per g of dry Kiam wood.

Generally after harvesting, fresh palm sap is treated with plant preservatives namely *Cotylelobium lanceotatum* craih*., Hopea odorata.* D*., Shorea floribunda.* D., *Saccoglottis gabonensis, Vernonia amygdalina, Euphobia* sp*., Nauclea* sp. or *Rubiacae* sp. (Faparusi and Bassir, 1972; Ogbulie *et al*., 2007). In Thailand, the most popular one is called Kiam wood (*Cotylelobium lanceotatum* craih.). Kiam wood has been traditionally submerged in palm sap to prevent or retard microbial fermentation (Chanthachum and Beuchat, 1997). However, the interaction of protein and polyphenol can form soluble complexes and grow to a large colloid size or haze (Kermasha *et al*., 1995; Siebert *et al*., 1996).

pH and total acidity

Main organic acids present in palm sugar concentrate are lactic and tartaric acids (Stuckel and Low, 1996). Micro-organisms, mainly lactic acid bacteria, have produced organic acids (lactic acid), then increases total acidity and decreased pH. Thus, total acidity in this study is calculated based on lactic acid equivalence. The pH of palm sugar concentrate samples varied from 4.50 to 5.37, while total acidity varied from 0.24 to 0.86 %w/v. The pH value and total acidity content were significantly different among samples. High percentage of total acidity indicates the initial spoilage or fermentation of fresh palm sap as a raw material used for palm sugar concentrate production. A consequence of high acidity means low pH condition, it will accelerate Maillard reaction and then the brown colour of the palm sugar concentrate

sample will be promoted during heating step. In addition, darkness intensity in the product can be increased during storage.

Total soluble solids

Total soluble solids of 30 palm sugar concentrate samples varied from 59.01 to 73.05°Brix as shown in Table 1. According to the Thai Industrial Standards Institute, Ministry of Industry (2003) states that the total soluble solids in palm sugar concentrate shall not be less than 65°Brix. From this result, only 23 samples have met the product standard. The Thai palm sugar concentrate standard is similar to maple syrup standard stated by the United States Department of Agriculture Standards (1980) as the total soluble solids of the finished maple syrup shall not be less than 66°Brix. This criterion is set to prevent growth of micro-organisms in finished product during storage under room temperature.

Total sugars and reducing sugars

Total sugars and reducing sugars were determined by titration method and reported its results in percentage (%w/w) (A.O.A.C, 2000). Total sugars of samples varied from 23.77 to 71.89 %w/w, while reducing sugars varied from 3.54 to 23.94 %w/w. Total sugars and reducing sugars were significantly different among all samples. This might be due to the effect of contamination from micro-organisms in samples. The micro-organisms can convert sucrose to glucose and fructose (invert sugar) and finally to organic acids or alcohols in palm sap (Willits and Hills, 1976). In general, total sugars and reducing sugars are primary substances in Caramelisation reaction during heating step (Martins *et al*., 2001). Heat from the process, especially at high temperature and long heating time, will accelerate hydrolysis reaction of sucrose to be reducing sugars. Then, reducing sugars can interact with amino acids and form dark color via Maillard reaction (Aider *et al*., 2007).

Microbiological quality

The contamination and the growth of micro-organisms in palm sap after harvesting is the main factor influencing on the quality of finished product (palm sugar concentrate). Total microbial count of all samples ranged from 1.20×10^3 to 4.80×10^6 CFU/ml. The yeast and mold count presents ranged from 1.30×10^2 to 5.30×10^4 CFU/ml. The osmophillic yeast count also ranged from 2.00×10^2 to 1.40×10^6 CFU/ml (Table 1). As Thai Industrial Standards Institute, Ministry of Industry (2003) indicates that total microbial and the yeast and mold counts in palm sugar concentrate samples shall not be more than 500 CFU/ml and 100 CFU/ml, respectively. According to these criteria, all samples in this study did not meet the product standard. The micro-organisms can naturally contaminate in the raw material and the finished product, especially during harvesting and storage. Since typical harvesting process is conducted in an open-condition and the bamboo tube is reused to collect the palm sap. Therefore, contaminations of micro-organisms in the raw material are promoted. The bamboo tubes sometimes are not considered to be cleaned before and after usages. Moreover, most of the producers do not have good practice in sanitary during palm sap harvesting process. In fact, when palm sugar concentrate is produced under high temperature for evaporation to convert palm sap to palm sugar concentrate, substantial amount of micro-organisms are destroyed. However, a few micro-organisms, especially osmophillic yeasts are found to survive, and grow after processed. The details of palm sugar concentrate production could be used for discussion in this study. For example, in case of types of container at the site of the producers (such as traditional earth jar (E), plastic bottle (B), Tin can size 20 liters (T1) and Tin can size 100 liters (T2)), samples (No. 3, 4, 5 and 6), which were stored in traditional earth jars were found to contain high load of osmophillic yeast count. In addition, samples No. 12(T2), 13(E), 15(T1) and 21(T1) were found much more osmophillic yeast count as they were stored in opened containers for longer times (2-5 months) in any containers. Hence, the quality of palm sugar concentrate does not only vary across producers but also varies within processing and storage steps as well, it depends on individual production factors such as personal hygiene, sanitary facilities, heating temperature, heating time and storage conditions (Willits and Hills, 1976; Morselli and Whalen, 1991; Ogbulie *et al*., 2007).

Product profiling

The overall quality of palm sugar concentrate samples

The chemical, physical and microbiological qualities of the palm sugar concentrate samples are summarized as can be seen in Table 1. Large variation of the quality indicators introduces food safety concerns as it might be due to effects of sugar fermenting process, which is based on micro-organism activities. The micro-organisms were found in a large number in all samples, more than 100 times specified in the product standard.

Table 2. Correlation coefficient matrix from quality of palm sugar concentrate samples

Variables	L*	a*	b*	%T	pH	Total acidity	Total soluble solid	Reducing sugar	Total sugar	Total microbial	Yeast and mold	Osmophillic yeast
L*	1	0.435	0.996	0.965	0.406	-0.477	0.302	-0.478	0.375	0.564	-0.100	-0.075
a*	0.435	1	0.475	0.428	0.439	-0.178	0.374	-0.475	0.213	0.014	0.066	0.268
b*	0.996	0.475	1	0.946	0.417	-0.495	0.332	-0.495	0.387	0.496	-0.092	-0.057
%T	0.965	0.428	0.946	1	0.358	-0.376	0.268	-0.413	0.328	0.692	-0.099	-0.030
pH	0.406	0.439	0.417	0.358	1	0.137	0.454	-0.638	0.055	0.186	0.363	0.186
Total acidity	-0.477	-0.178	-0.495	-0.376	0.137	1	-0.105	0.086	-0.578	-0.173	0.069	0.220
Total soluble solid	0.302	0.374	0.332	0.268	0.454	-0.105	1	-0.223	0.274	-0.033	0.055	0.224
Reducing sugar	-0.478	-0.475	-0.495	-0.413	-0.638	0.086	-0.223	1	0.074	-0.175	-0.263	-0.229
Total sugar	0.375	0.213	0.387	0.328	0.055	-0.578	0.274	0.074	1	0.127	0.014	-0.027
Total microbial	0.564	0.014	0.496	0.692	0.186	-0.173	-0.033	-0.175	0.127	1	0.116	-0.115
Yeast and mold	-0.100	0.066	-0.092	-0.099	0.363	0.069	0.055	-0.263	0.014	0.116	1	0.184
Osmophillic yeast	-0.075	0.268	-0.057	-0.030	0.186	0.220	0.224	-0.229	-0.027	-0.115	0.184	1

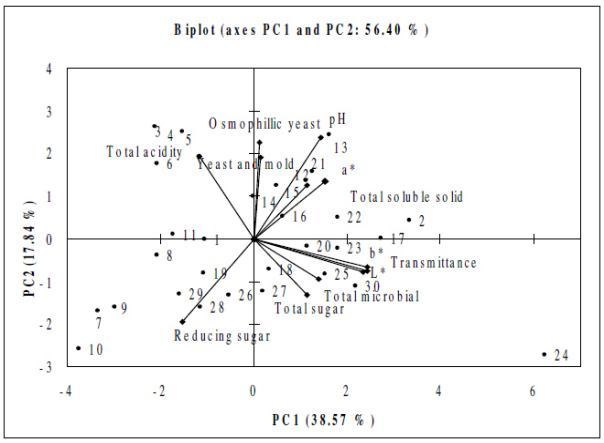

Figure 1. Biplot PC1-PC2 of the quality of palm sugar concentrate samples

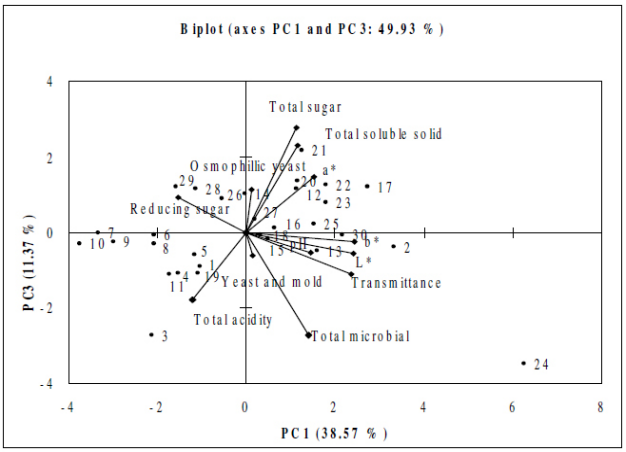

Figure 2. Biplot PC1-PC3 of the quality of palm sugar concentrate samples

Relationship among product qualities

The product samples (n=30) contained large variation of quality indicators. All quality values were significantly difference ($P \leq 0.05$) across samples. The data measured on 12 physical, chemical and microbiological qualities from 30 observations (samples), were analyzed using multivariate technique- Principal Component Analysis (PCA). The data set was analyzed based on correlation matrix (Table 2) from each quality (variable). The most related variables are grouped into the PC1 and then the rest will be grouped into PC2, PC3 and so on. The graphical illustrations from PCA mainly present (1) plots of scores to represent the quality (variable) positions according to each PC and (2) plots of scores to represent the observation (sample) positions according to PCs. Graphical PCAs are shown in Figures 1 and 2, which are composed from PC1-PC2 and PC1-PC3, presenting 67.77% of variance from the original data set. Figure 1 PCA reveals negative relationships between reducing sugars and these parameters (such as pH, total soluble solids and %transmittance), also negative relationship between total acidity and these parameters (including total sugars, %transmittance, L*, b* and total microbial count). Given 67.77% of the variance explained by the 3 PCs, Figure 2 also reveals a group pattern of 7 samples (no. 1, 8, 9, 10, 11, 19 and 27) whose total soluble solid values did not meet the palm sugar concentrate standard. The samples contained low total soluble solids, definitely low total sugars, but also contained high total acidity and low pH, low transmittance value and low a* as well. For example, sample 10 presented highest dark colour and lowest L*, b*, transmittance value and pH, whereas sample 24 contained the highest light colour and highest L*, b*, transmittance value and pH.

Conclusion

The 30 palm sugar concentrate samples produced by traditional evaporating process in an open pan, were randomly sampling from primary producers in Songkhla province. The physical, chemical and microbiological qualities of palm sugar concentrate differed among samples. The samples contained large variation in all qualities measured. Microbiological quality of all 30 samples was not in line with the requirements by Thai legislation for palm sugar concentrate. Total soluble solids of 7 out of 30 samples did not meet the requirements of more than 65°Brix either.

As we found samples with low total soluble solids, contained high total acidity but low pH, low transmittance value and low a*, this finding confirms us of how the qualities changed. The palm sap during harvesting – before processing, if contaminated, its sugar would be used by micro-organisms and resulting in higher acid content. Then the acids synergist with heat during process and reducing sugar also increased. After the process, some of micro-organisms are destroyed but colour, turbidity and acidity still remained. Then osmophillic yeast would grow in the product because of its high sugar content, if the storage condition is poor. In order to improve quality of palm sugar concentrate, the quality of palm sap before processed is one of major concern. Suitable temperature and time during evaporating process are also of importance. Good practices such as hygiene, sanitary facilities and equipment could greatly contribute to extend the product shelf-life during storage. The factors also affect intensities of brown colour, sweet taste, thickness and viscosity of palm sugar concentrate as the qualities are mainly developed during the heating process. Future work could firstly aim at educating the producers to understand effects of personal hygiene sanitary facilities and equipment, on the product quality and price.

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