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Preliminary Investigation of the Production and Characterization of Peanut Milk Based Stirred Yoghurt

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Abstract: The possibility of producing yoghurt based on peanut milk was studied. Stirred yoghurt was prepared from a mixture of 70% peanut milk and 30% cow milk. The final product was subjected to physiochemical analysis and sensory evaluation. Whole milk yoghurt was used as a control throughout the investigation. Investigations revealed that the peanut milk based yoghurt had 3.47% protein content, 81.02% water holding capacity and 34.43% susceptibility to syneresis compared to 2.76, 65.03 and 47.40% for whole milk yoghurt, respectively. Peanut milk based yoghurt also had higher apparent viscosity compared to whole milk yoghurt. Sensory evaluation of peanut milk based yoghurt using the nine points hedonic scale with the help of sixteen panelists showed that the product had a good appearance, texture and acceptable flavor. The titratable acidity of the peanut milk based yoghurt was 80°T and pH was 4.57. The investigation confirmed that it is possible to produce acceptable peanut milk based yoghurt. Therefore, it is one of the interesting alternative options to yoghurt manufacture in regions with high peanut production.

Key words: Peanut milk based yoghurt, stirred yoghurt, susceptibility to syneresis, titratable acidity, water holding capacity

INTRODUCTION

Peanut (*Arachis hypogaea* L.) is a major source of edible oil and protein meal and is therefore considered to be highly valuable in human and animal nutrition (Nwokolo, 1996). Peanuts may be consumed raw, roasted, pureed, or in a variety of other processed forms and constitute as a multimillion-dollar crop world wide (Yu *et al*., 2005) with numerous potential dietary benefits like high protein content and health promoting oils. It is therefore necessary to adequately research into the possibility of peanut processing and utilization in other edible products.

Over the last 50 years, many ways of producing peanut milk have been developed by various researchers (Beuchat and Nail, 1978; Chan and Beuchat, 1992; Chandrasekhara, 1971; Maltz, 1981; Rubico *et al*., 1988; Salunkhe and Kadam, 1989; Van, 1992) but all these methods are modifications of the Illinois process for preparing soy milk (Chan and Beuchat, 1992). The peanut milk composition depends on the desire of the producer, but in all cases, this low cost milk has high protein content (Kouane *et al*., 2005).

Peanut milk may be produced by soaking and grinding full-fat raw peanuts with water to get a slurry, subject to filtration (Chan and Beuchat, 1992). Alternatively, it may also be produced by grinding unsoaked roasted peanuts, raw full-fat, or partially defatted peanuts to form flour to which

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water may later be added to make an emulsion. Another way is to use the peanut milk or peanut protein isolates to supplement animal milk (toned milk). In all cases, the peanut or peanut flour to water ratio varies greatly from one producer to another. The milk-like product produced is then homogenized and pasteurized in much the same way as fresh milk and also supplemented with vitamins and minerals and is sometimes flavored (Chan and Beuchat, 1992).

For ages it has been well known that peanut milk and peanut milk products have nutritional benefits for young and old people because of their extreme richness in protein, minerals and essential fatty acids such as linoleic and oleic acids, which are considered to be highly valuable in human nutrition. It is extensively used in India and other developing countries by vegetarians and more recently by children allergic to cow milk proteins (Kouane *et al*., 2005). The current interest in peanut milk and peanut milk products is motivated by the fact that dairy and dairy products are always priced too high for the low income earners. Another factor, no less important, is the growing awareness of the nutritional benefits of vegetable proteins in low cholesterol diets by health conscious people (Kouane *et al*., 2005).

Over the years, peanut milk has been successfully converted into low cost edible products with high nutritional value. In this area, researchers have focused on products resulting from fermentation, such as yoghurt, buttermilk and ripened cheese analogs (Beuchat and Nail, 1978; Chan and Beuchat, 1991). In this regard, advances in fermented products manufacturing revealed that hexanal, which is one of the compounds responsible for the unwanted beany flavor in peanut milk, completely disappeared as a result of fermentation (Chan and Beuchat, 1991; Hao and Brackett, 1988). Preparation and fermentation of peanut milk may serve as one such effort that can increase the consumption of this valuable crop and hence improve protein availability and consumption (Sunny-Roberts *et al*., 2004).

As early as 1967, a yoghurt-like product (Dahi) was successfully prepared from miltone using lactic cultures, at the Central Food Technological Research Institute (CFTRI) in India (Salunkhe and Kadam, 1989). Miltone is got by supplementing animal milk with spray-dried peanut protein isolates. In 1978, a beverage comparable to flavored butter milk was prepared by fermenting pure peanut milk with *Lactobacillus bulgaricus* and *Lactobacillus acidophilus* and adding sugar plus fruit flavorings (Beuchat and Nail, 1978). However though an acceptable custard-like texture developed in peanut milk fermented with *L. bulgaricus* NRRL B-1909 and *L. acidophilus* NRRL B-1910, excessive whey made the products somewhat undesirable for yoghurt substitutes (Salunkhe and Kadam, 1989).

Although some study has been done on the utilization of peanut milk in yoghurt production, most has been either on the supplementation of animal milk with peanut protein isolates or fermentation of pure peanut milk with lactic acid bacteria. However, excessive whey and the nutty flavor make the product from fermented pure peanut milk somewhat undesirable for use as a yoghurt substitute (Salunkhe and Kadam, 1989).

The main aim of present investigation was to explore the possibility of preparing an acceptable yoghurt product based on peanut milk (70%) supplemented with a small proportion of cow milk (30%). The product was also subjected to physiochemical analysis and sensory evaluation.

MATERIALS AND METHODS

This study was conducted between September and December 2006 from the Key Laboratory of Food Science and Safety, Southern Yangtze University, Wuxi, Peoples Republic of China

Materials

The Spanish red-skinned peanut seeds were purchased from a local supermarket in Wuxi, China. Care was taken to ensure that good quality and mould-free seeds were selected. The whole milk powder was purchased from a dairy factory in Donghuang, Qinghai, China. The flavors were obtained from Shanghai H and K Flavors and Fragrances Co. Ltd. The culture pack of *Lactobacillus bulgaricus* and *Streptococcus thermophilus* were obtained from the Key Laboratory of Food Science and Safety of Southern Yangtze University, Wuxi, China.

Milk Preparation

Whole milk powder was reconstituted at 43°C with moderate mixing at 14% w/v total solid concentration and cooled to room temperature then refrigerated at 4°C overnight for hydration of the powder before usage.

Peanut milk was prepared by a method similar to the one reported by Salunkhe and Kadam (1989) with slight modifications. Sorted peanut seeds were roasted at 130°C for 28 min in an oven (ENGZI 101-1-BS, Shanghai Yuejin Machine Factory, China). The seeds were then de-skinned and weighed before being soaked in 0.5% $NaHCO_3$ for at least 14 h. The de-skinned peanut kernels were then washed with clean water. The kernels were then mixed with water in a ratio of 1:5 [peanuts (g): water (mL)] and transferred to a blender (SS680-A, Shanghai Yuyang Electronics Co.) where they were blended for 5 min. The slurry formed was filtered using a double layered cheese cloth to yield peanut milk.

Yoghurt Preparation

Peanut milk based yoghurt (PMBY) was prepared as follows; To a blend of 70% peanut milk (PM) and 30% Reconstituted Whole Milk (RWM) was added 7% (w/v) sucrose as a sweetener. The toned milk was homogenized at 25 MPa then pasteurized at 85°C for 30 min. The pasteurized milk was cooled to 43°C in a water bath then inoculated with 3%(v/v) starter culture (*Lactobacillus bulgaricus* and *Streptococcus thermophilus*;1:1) and incubated at 43°C for about 4 h. Yoghurt was cooled rapidly to 25°C, stirred gently for approximately 2 min then transferred into glass bottles. Stirred yoghurt was then stored in a refrigerator at ~5°C overnight before being analyzed.

Reconstituted whole milk yoghurt RWMY (as a control of the experiment) was prepared from RWM alone but following the same procedure described above for PMBY. The experiment was repeated three times.

Physical properties

Water Holding Capacity

Water Holding Capacity (WHC) of the yoghurt was determined by the method of (Harte *et al*., 2003) with some modifications. The yoghurt was subjected to 15 min centrifugation at 10,000 rpm using TGL-16C centrifuge (Shanghai Anke Scientific Machine Co., China). The following formula was used to calculate WHC:

$$\text{WHC (\%)} = \left(1 - \frac{W_1}{W_2}\right) \times 100$$

W_1 = Weight of whey after centrifugation
W_2 = Yoghurt weight
All measurements were carried out in triplicate.

Susceptibility to Syneresis

The yoghourt Susceptibility to Syneresis (STS) was determined using the drainage method (Hassan *et al*., 1996) with some modifications. Syneresis (released whey) was measured by placing a

100 g yoghurt sample on a cheese cloth placed on top of a funnel. After 2 h of drainage, the weight of the whey collected in a beaker was measured and used as an index of syneresis. The following formula was used to calculate STS:

$$\text{STS}(\%) = \frac{W_1}{W_2} \times 100$$

W_1 = Weight of whey collected after drainage
W_2 = Yoghurt weight

Apparent Viscosity

Apparent viscosity and the shear rate of the yoghurt were measured at 10°C using a rheometer (AR 1000 rheometer, England). The data were analyzed with supporting rheometer software (Rheology advantage). The flow curves of the yoghurt were obtained by varying the shear rate from 0.00 to 100 sec^{-1} and the corresponding viscosity values measured.

Chemical Analyses

Titratable acidity and pH

The pH of yoghurt samples was measured with a pH meter (ZD-2, Shanghai Analytical Instruments Factory, China). Estimation of Titratable Acidity (TA) was by a previously reported procedure of Thorner (°T) (Frazier *et al*., 1968), in which 10 mL of sample was titrated against 0.1 M NaOH using phenolphthalein as an indicator.

Determination of Moisture, Ash, Crude Protein and Total Solids

Moisture, ash, fat, protein and total solids were determined according to AOAC (1995) (Ceirwyn, 1995). Fat content was determined by the Rose-Gottlieb method. Crude protein was determined by the Kjeldahl method using a conversion factor of 6.25(0.7) + 6.38(0.3) = 6.289 for the toned milk, 6.25 for pure peanut milk and 6.38 for the control. All measurements were carried out in triplicate.

Sensory Evaluation

Yoghurt samples were analyzed for appearance and color, body and texture/mouth feel and flavor after overnight storage at 4-5°C. Sixteen panelists who had knowledge of food science and sensory quality of food were used to rate the samples on the basis of the nine points hedonic scale (Stone and Sidel, 1993). Three samples were presented to panelists in three digit random number cups containing approximately 25 mL of sample per cup. Sample 1 was prepared from a blend of 70% PM and 30% RWM, sample 2 was similar to 1 but had a yoghurt flavor (0.02%v/v) added to it and sample 3 was the control prepared from pure RWM. The sensory scores included; Like extremely = 9, Like very much = 8, Like moderately = 7, Like slightly = 6, Neither like nor dislike = 5, Dislike slightly = 4, Dislike moderately = 3, Dislike very much = 2, Dislike extremely = 1.

Statistical Analysis

Unless otherwise stated, results were analyzed statistically using a computer program SAS system for windows (SAS, 2002) for analysis of variance (ANOVA) by one way and comparison of means by Duncan's multiple comparison test where $p<0.05$ was considered for significant difference.

RESULTS AND DISCUSSION

Physical Properties

The water holding capacity of PMBY was significantly ($P<0.05$) higher than that of RWMY as shown in Table 1. On the other hand, the Susceptibility To Syneresis (STS) of PMBY was

Table 1: Water holding capacity and susceptibility to syneresis values of different yoghurt preparations

Type of yoghurt	WHC (%)	STS [%(w/w)]
PMBY	81.02±1.59[a]	34.43±0.63[b]
RWMY	65.03±2.65[b]	47.40±0.78[a]

Results are reported as means±Standard deviation. Means bearing different letter(s) in a column are significant at level of p<0.05. RWMY: Reconstituted Whole Milk Yoghurt, PMBY: Peanut Milk Based Yoghurt

Table 2: Mean pH and TA values of different yoghurt preparations

Type of yoghurt	pH	Titratable acidity (T °)
PMBY	4.57±0.52[a]	80.00±1.79[b]
RWMY	4.27±0.52[b]	104.00±0.89

[a]Results are reported as means±Standard deviation. Means bearing different letter(s) in a column are significant at level of p<0.05. RWMY: Reconstituted Whole Milk Yoghurt, PMBY: Peanut Milk Based Yoghurt

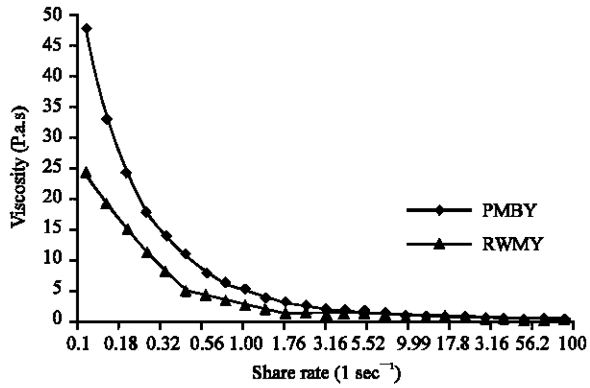

Fig. 1: Viscosity versus shear rate of two different yogurt preparations. RWMY-reconstituted whole milk yoghurt, PMBY-peanut milk based yoghurt

significantly ($p<0.05$) lower compared to that of RWMY. This may be due to a complex synergistic interaction between peanut proteins and cow milk proteins in the PMBY. This observation also suggests that peanut milk based yoghurt was superior to whole milk yoghurt in terms of high WHC and reduced STS.

Apparent Viscosity

As shown in Fig. 1, the apparent viscosity of PMBY was much higher than that of RWMY at all shear rates. At the minimum shear rate of 0.10 sec^{-1}, the viscosity of PMBY was 47.4 Pa.s while that of RWMY was 24.58 Pa.s. At the maximum shear rate of 99.96 sec^{-1}, the viscosity of PMBY was 0.162 Pa.s while that of RWMY was 0.124 Pa.s. However in both cases, the apparent viscosity decreased as the shear rate increased, so both yoghurts exhibited a shear thinning behavior. The fall in viscosity with shear rate might be due to the destruction of the interactions within the yoghurt network structures (Mohameed *et al*., 2004). These interactions include electrostatic and hydrophobic forces, which are considered as weak physical bonds (Mohameed *et al*., 2004). The shear rate range applied in this study was enough to destroy these physical bonds.

Chemical Properties

As shown in Table 2, the pH of PMBY was significantly ($p<0.05$) higher than that of RWMY. On the other hand, the TA of PMBY was significantly ($p<0.05$) lower than that of RWMY. However, both are used to measure level of acidity. The pH depends on the acid concentration and the amount of dissociated hydrogen ions in solution while the TA measures all hydrogen ions (Tamime and Robinson, 1999).

From the results in Table 2, PMBY generally had a lower level of acidity compared to RWMY. Since lactose is the main precursor for lactic acid production in yoghurt, then the relatively lower acidity of PMBY may be attributed to the small amounts of lactose in the toned milk from which the yoghurt was prepared. RWMY had a much higher level of acidity due to relatively higher levels of lactose present in whole milk from which it was prepared. The TA 80°T (0.72%) for PMBY is in agreement with the recommendation of International Dairy Federation (IDF) which suggested a minimum of 0.7% lactic acid for commercial yoghurts (Tamime and Robinson, 1999). The production of lactic acid beyond the point of coagulation is monitored principally in relation to consumer preference and hence the selected end point varies not only from country to country, but also with the type of yoghurt. For example: Bulgarian yoghurt has an acidity of up to 1.48% ('165°T) while for Netherland Standards a maximum of 1.17% (130 °T) is allowed (Tamime and Robinson, 1999).

The moisture content of peanut milk was significantly different ($p<0.05$) from that of RWM and toned milk as indicated in the Table 3. Though the moisture content of toned milk was higher than that of RWM, it was not significantly different ($p<0.05$). The moisture contents of PMBY and RWMY as shown in Table 4 are in agreement with 81.9% as reported earlier (Buttriss, 1997).

The total solids content of toned milk and RWM were not significantly different ($p<0.05$) from each other but differed ($p<0.05$) from PM. The level of solids in milk (including the fat content) for the manufacture of yoghurt ranges from as low as 9% in low fat yoghurt to as high as 30% in other types of yoghurt (Tamime and Robinson, 1999). Therefore, the total solids in PM, RWM and toned milk as indicated in Table 3 are within the recommended range for milk to be used in yoghurt production. It is recommended that the composition of total solids in milk used for production of commercial yoghurts should fall within the range of 14-15% (Tamime and Robinson, 1999).

The ash contents of PM, RWM and toned milk differed ($p<0.05$) from each other as shown in the Table 3. The ash content 0.27% for P.M was not very different from 0.2% (Kouane *et al*., 2005). The ash content 0.76% for RWM is in agreement with 0.75% reported earlier (Scott, 1986). Peanut milk has much lower ash content compared to cow milk.

The crude protein content for the different milk types was in the order PM>Toned milk>RWM. The crude protein contents of PM and toned were not significantly different ($p<0.05$) from each other but were significantly ($p<0.05$) different from RWM. The protein content 3.01% for RWM is not far from 3.2% reported earlier (Buttriss, 1997) for cow milk. The crude protein content of 3.76% for PM is close to 3.7% reported earlier on (Rubico *et al*., 1987) for peanut milk.

The amounts of protein in PMBY (3.47%) and RWMY (2.76%) were lower than those of the milks from which they were produced. This shows that some degree of proteolysis must

Table 3: Chemical composition of different milk preparations

Milk type	Composition (%) Moisture	Ash	Fat	Crude protein	Total solids
PM	86.71±0.17^{a}	0.27±0.02^{c}	6.86±0.09^{a}	3.76±0.53^{a}	13.29±0.17^{b}
RWM	85.97±0.05^{b}	0.76±0.03^{a}	5.15±0.07^{c}	3.01±0.09^{b}	14.03±0.05^{a}
Toned milk	86.21±0.2^{b}	0.39±0.02^{b}	6.60±0.01^{b}	3.72±0.16^{a}	13.72±0.16^{a}

Results are reported as means±Standard deviation. Means bearing different letter(s) in a column are significant at level of $p<0.05$. RWM: Reconstituted Whole Milk, PM: Peanut Milk

Table 4: Mean chemical composition of different yoghurt preparations

Yoghurt preparation	Composition (%) Moisture	Ash	Fat	Crude protein
PMBY	82.14±0.09^{a}	0.43±0.05^{b}	5.33±0.03^{a}	3.47±0.05^{a}
RWMY	82.02±0.06^{a}	0.84±0.08^{a}	2.92±0.06^{b}	2.76±0.11^{b}

Results are reported as means±Standard deviation. Means bearing different letter(s) in a column are significant at level of $p<0.05$. RWMY: Reconstituted Whole Milk Yoghurt, PMBY: Peanut Milk Based Yoghurt have occurred during fermentation of milk to yoghurt. Although the yoghurt starter cultures are considered to be only weakly proteolytic, *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* may, during the fermentation, cause a significant degree of proteolysis (Tamime and Robinson, 1999). This activity is important because the enzymatic hydrolysis of milk proteins results in the liberation of peptides of varying sizes and free amino acids and these possible changes may be involved during the formation of the gel and can affect the physical structure of yoghurt (Tamime and Robinson, 1999).

The difference in fat contents of PM, RWM and toned milk was significant ($p<0.05$). The fat content was in the order PM>toned milk> RWM. The fat content 5.15% for RWM is in agreement with 5.14% for Jersey cow milk (Scott, 1986; Tamime and Robinson, 1999). The fat contents of PMBY (5.33%) and RWMY (2.92%) were lower than those of the respective milks from which they were produced (Table 4). This may be attributed to lipid metabolism by the Starter culture micro-organisms (*S. thermophilus* and *L. delbrueckii* subsp. *Bulgaricus*) and the process of homogenization. The extent of lipolysis in homogenized milk is much greater than in non-homogenized milk, due, in large measure, to the destruction of the protective layer of the fat globule membrane (Tamime and Robinson, 1999).

The toned milk which was used to produce PMBY had a relatively high fat content (6.60%) compared to most commercial milk products. However since this toned milk was composed of 70% P.M and only 30% RWM, then most of its fat was predominantly constituted by the health promoting, low cholesterol peanut oils (Ory *et al*., 1992; Savage and Keenan, 1994). The contributions of each of these milk types to the fat content was estimated by preparing toned milk in the same way but using skimmed milk in place of whole milk, then measured the fat content of that toned milk. In that case the fat content was all contributed by PM. The difference between the fat content of toned milk prepared using whole milk and that prepared using skimmed milk equals to the fat contribution of whole milk to the toned milk as summarized in the expression below:

$$F_{cwm} = F_{(PM+RWM)} - F_{(PM+SKM)}$$

Where:

F_{cwn} = Percentage fat contributed by whole milk

$F_{(PM+RWN)}$ = Percentage fat content of toned milk prepared from PM and whole milk

$F_{(PM+SKM)}$ = Percentage fat content of toned milk prepared from PM and skimmed milk

In the experiment $F_{(PM+SKM)}$ was 4.99% while $F_{(PM+RWN)}$ was 6.60% giving F_{cwn} = 1.61%. Therefore, 4.99% was the fat contribution by peanut milk. When this same approach was applied to PMBY, $F_{(PM+SKM)}$ was 3.68% while $F_{(PM+RWN)}$ was 5.33% giving F_{cwn} 1.65. Hence 3.68 was the fat contribution by peanut milk in PMBY. The total fat content of PMBY was 5.33% and was significantly different from that of RWMY.

Animal milk fat has a major impact on the mouth feel of yoghurt, approximately 1% being regarded as the minimum to produce the desired response from the consumer (Tamime and Robinson, 1999). Different yoghurts may have different fat levels; for instance full fat natural yoghurt (3.0-3.5%), luxury' fruit yoghurts (>4%) and Greek-style yoghurts (>8.0-10%) (Tamime and Robinson, 1999). It was vital to use whole milk instead of skimmed milk to extend PM because much as the peanut oils alone are good, health promoting and would be sufficient for yoghurt production, they are inferior in terms of sensory quality of yoghurt just like other plant oils (Barrantes *et al*., 1996). It was also reported that the sensory properties of yoghurt made by substituting milk fat with plant oils were characterized as being inferior when compared with equivalent products made with animal fat (Barrantes *et al*., 1996). Therefore the small proportion of 1.65% contributed by whole milk in our product may serve to improve the sensory properties of PMBY.

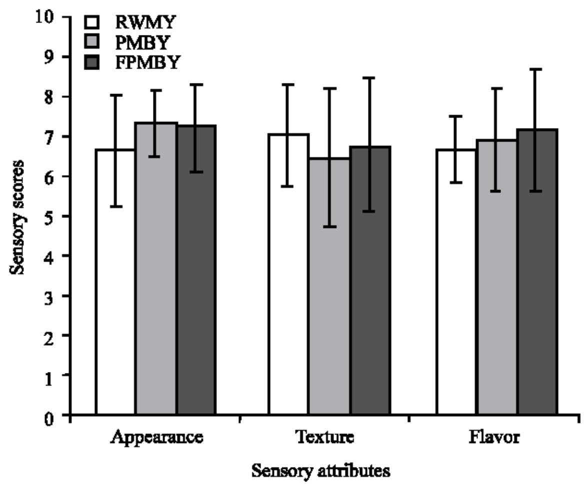

Fig. 2: Mean sensory scores for 3 different yogurt preparations. RWMY-reconstituted whole milk yoghurt, PMBY-peanut milk based yoghurt, FPMBY-Flavored peanut milk based yoghurt

Table 5: Sensory scores of different sensory attributes of three yoghurt preparations

	Scores for different sensory attributes		
Yoghurt type	Appearance	Body and texture	Flavor
RWMY	6.69±1.4[a]	7.06±0.84[b]	6.69±1.12[c]
PMBY	7.38±1.31[a]	6.50±1.76[b]	6.94±1.70[c]
FPMBY	7.25±0.84[a]	6.81±1.31[b]	7.19±1.53[c]

Results are reported as means±Standard deviation, for sixteen replicates of each sensory attribute. Means bearing different letter(s) in a column are significant at level of $p<0.05$. RWMY: Reconstituted Whole Milk Yoghurt, PMBY Peanut Milk Based Yoghurt, FPMBY: Flavored Peanut Milk Based Yoghurt

Sensory Evaluation

The results of sensory evaluation of yoghurt on the basis of appearance, texture and flavor are summarized in the Table 5. All mean scores for the different sensory attributes of the three yoghurt preparations were within the commercially acceptable range (4-9 scores) recommended for yoghurt by the Karl Ruther nine points scheme (Tamime and Robinson, 1999).

PMBY and FPMBY had higher scores than RWMY in terms of appearance and color (Fig. 2). However there was no significant difference ($p<0.05$) in appearance scores among the three yoghurt preparations as shown in Table 5. Therefore the panelists could not clearly distinguish between the appearance of peanut milk based yoghurt and whole milk yoghurt.

As shown in Fig. 2, RWMY had higher body and texture scores than PMBY and FPMBY. However there was no significant difference ($p<0.05$) in body and texture among the three yoghurt preparations (Table 5). Since the panelists could not clearly tell the difference in body and texture, then the mode of yoghurt preparation did not affect ($p<0.05$) the texture scores of yoghurt.

The flavor scores were in the order of FPMBY>PMBY>RWMY as shown in Fig. 2. It is interesting to note that the flavor of peanut milk based yoghurt was more appreciated by the panelists than whole milk yoghurt and the scores were even higher when it was flavored. This shows that the application of flavors improves the acceptability of peanut milk based yoghurt. However, the difference was not significant ($p<0.05$) in flavor scores of the three yoghurt preparations (Table 5). Therefore, the flavors of all the different yoghurts were generally appreciated by the panelists.

CONCLUSION

The investigation revealed that PMBY had higher protein content, WHC, lower STS and higher viscosity compared to RWMY. Peanut milk based yoghurt also had a relatively good sensory texture, appearance, flavor and relatively high quantity of peanut oils. The investigation also confirmed that it is possible to produce acceptable peanut milk based yoghurt. Therefore, it is one of the interesting alternative options for yoghurt manufacturers in regions with high peanut production.

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