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Analysis of Fats and Fatty Acids in Several Bakery Cookies of Bangladesh

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Abstract

Cookie is one of the most common baked goods consumed by people throughout the globe including in Bangladesh. Cookies have a considerable amount of fats, despite not being considered as fatty foods by consumers. Since cookies are eaten by individuals of all ages and socioeconomic groups, an experimental investigation was conducted to evaluate the quality and quantity of the fat fraction of bakery cookies available in Dhaka. Ten different bakery cookies (5 savory and 5 sweet) from five different manufacturers were collected from several local supermarkets of Dhaka and analyzed to study the total fat contents, relative fatty acids compositions, moisture and ash contents. Extraction of fat from cookie samples was done using n-hexane. The fatty acid compositions of the samples were determined by gas-liquid chromatograph equipped with flame ionization detector. The results showed the total fat content to be ranged from 16.13-26.84 g/100g of cookies. The fatty acid composition exhibited that the relative percentage of saturated fatty acids (SFA), monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA) were in the range of 7.76-78.20, 9.66-79.56 and 0.38-9.64%, respectively. The major FAs were found to be palmitic acid (C16:0), palmitoleic acid (C16:1), stearic acid (C18:0), oleic acid (C18:1) and linoleic acid (C18:2). The moisture and ash contents in the samples were found in the range of 0.79-4.80 and 0.73-4.23%, respectively.

Keywords: bakery cookies; fatty acids compositions; gas-liquid chromatography.

I. Introduction

Cookies are a popular ready-to-eat, inexpensive, and easy food item appreciated by people of all ages around the world. Cookies have high energy content but little nutritional benefit. This is owing to their high fat and carbohydrate content, as well as the fact that they contain several vitamins, macro- and microelements, proteins, dietary fibers, and other nutrients. Cookies come in many different shapes, dimensions, and components but the principal constituents in cookies are always wheat, fat (butter or vegetable shortenings) and sugar.[1] Cookies have relatively low moisture content (1–5%).[2] Fats are probably the most essential ingredients utilized in cookie production, according to Manley.[3] Fats have a crucial function in the preparation and baking of dough.[4-6] Inclusion of fats influences the overall aspect of the product while also enhancing the quality, palatability, and texture of the cookies by interaction with certain other components. In expansion, fats decrease the abrasive effect of flour and sugar and upgrade air circulation resulting in an increased volume and open surface to make a milder mouthfeel.[7,8] Lowering the fat content or substituting solid oil for liquid oil led to a substantial reduction in dough rigidity, demonstrating that fat is an important structural component.[9]

Since the earliest stages in the use of fatty acid (FA) measurements, gas chromatography (GC) has been an essential analytical technology. Despite the development of GC-MS, GC analysis of FAs equipped with a flame ionization detector (FID) is still the most widely used method.[10-13]

After isolating the fats from the food materials preceding to GC examination, the analytes are converted into more volatile and non-polar derivatives.[12] Fat extraction and afterwards transformation of the FAs into corresponding methyl esters (FAMEs) is the most prevalent method for assessing FAs.[14] Generally, the procedures consist in the first step to heat the sample in methanol with sodium hydroxide, and then the second step is the esterification of free faty acids (FFAs) using methanolic BF_3[15] or methanolic KOH.[16] The direct transesterification of fats by base catalyzed procedure has been found to be more appropriate for nutrition analysis since it is easier to use and involves fewer aggressive chemicals.[16-18] Within the frame of its coordination complex with methanol, boron trifluoride has been utilized as a catalyst for the esterification of fatty acids. Because boron trifluoride alcoholates act like strong acids, they can facilitate fat methanolysis in the same way as HCl or H_2SO_4 added to methanol does, with the extra benefit of the boron trifluoride's high electropolarity.

Fats have aroused interest in the media because their prevalence in daily diet is linked to a variety of diseases. Primitive trials on human have demonstrated that consuming substantial amount of fatty acids can alter one's lipid profile by raising the quantity of low density lipoprotein cholesterol (LDL-c) and lowering high density lipoprotein cholesterol (HDL-c) at the sametime.[19, 20] As a result, these may lead to an enhanced incidence of heart disease, non-insulin dependent diabetes, and malignancy.[21-23]

On the quality of fat used in Bangladeshi bakery cookies, there's exceptionally small data accessible. The selection of a better type of fat in the production of cookies in Bangladesh is typically based on technological and

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economic factors, without regard for nutritional and health consequences. Therefore, it's necessary that the fat composition of bakery cookies should be analyzed. Taking into account the aforementioned, the present work was embraced to assess the attribute and quantity of fat contents as well as the relative fatty acid compositions in various local bakery cookies of Dhaka city.

II. Materials and Methods

Materials

A total of ten (five sweet and five savory) cookie samples of the locally promoted brands with a strong local appeal were acquired from several bakery shops found in Dhaka city. The brands were chosen based on the ones with the largest consumption among those sold in bakeries. The sweet cookie samples were coded with letters A1, A2, A3, A4, A5 and the savory ones were assigned B1, B2, B3, B4, B5. All samples were collected and analyzed before the recommended expiry date.

Chemicals

Solvents of analytical grade, such as ultra pure n-hexane, anhydrous Na_2SO_4 were bought from Merck, Germany. BF_3-CH_3OH complex (Merck, Germany) and NaOH (BDH, UK) were also utilized in this study.

Sample preparation and fat extraction

Each finely grounded cookie sample was placed in an oven at 50^{o}C until complete dryness. Total fat extraction from the samples was carried out by using n-hexane as the extraction solvent. Approximately 10g of homogenized cookie sample was weighed in a round bottom flask then extracted with 30 mL of n-hexane under reflux for 1 hour. The whole process was repeated another two times. The solvents were then removed by evaporation and was dried in an air-oven at 40-60ºC for 1 hour and then cooled. The extracted fat was then weighed and the total fat content (%) was calculated.

Preparation of fatty acid methyl esters (FAMEs)

The extracted fats were methylated and converted into corresponding FAMEs by transesterification with alcoholic sodium hydroxide (NaOH). Approximately 0.5g of each fat extract was taken to which 10 mL of methanolic NaOH was added. The resulting solution was shaken well and refluxed for 1 hour. The mixture was then evaporated and drops of dilute hydrochloric acid were added to the flask to neutralize NaOH. A little water was added to the mixture followed by the neutralization and was transferred to a separatory funnel. Extraction with 15 mL of n-hexane separates the aqueous and hexane layer. The aqueous layer was discarded and the hexane layer was collected. In order to make it free from water, the hexane portion was filtered using anhydrous sodium sulfate (Na_2SO_4). The filtrate was dried and after evaporation, the content was treated with 1 mL boron trifluoride methanol (BF_3-MeOH) complex and was refluxed for 15 minutes. The mixture was then evaporated and transferred to a separatory funnel containing 5-10 mL of n-hexane. The mixture was shaken vigorously and the methyl esters of fatty acid were made free from water by adding anhydrous Na_2SO_4 and filtered. The filtrate was collected in a GC vial for analysis.

GC analysis of FAMEs

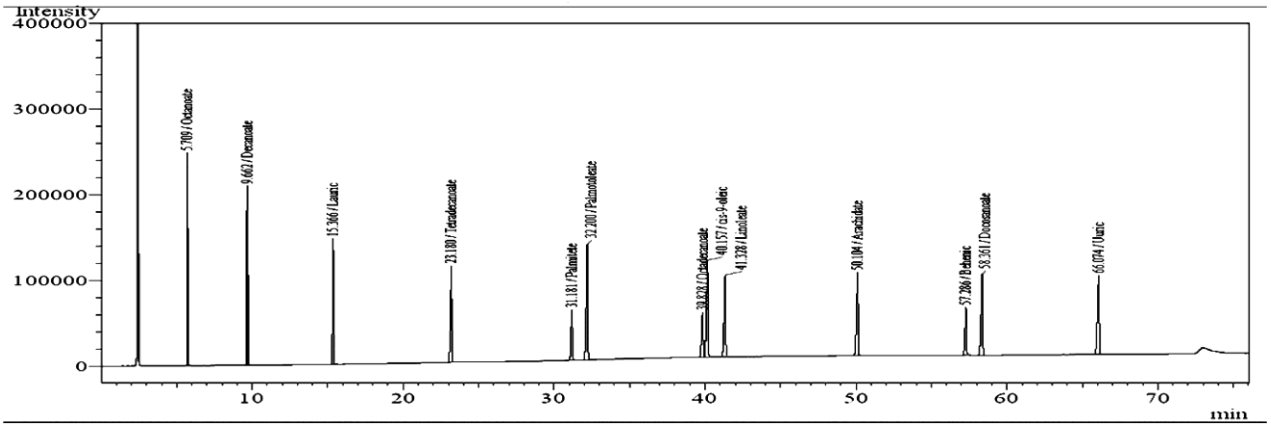
0.5 µL volume of each sample was injected into GC-FID for separation and quantification of the FAMEs. The analysis was carried out using an HP-5 capillary WCOT quartz column (30 m long, 0.25 mm inner diameter, 0.25 µm film thickness). The temperature program was employed as follows: initial temperature of 105^{o}C; held for 1 min, increased at a rate of 5^{o}C min^{-1} to 150^{o}C; held for 1 min, temperature increased at a rate of 2.25^{o}C min^{-1} to 280^{o}C; held for 7 minutes; total run time was 75.78 minutes. The detector and injector temperatures were 290 and 285^{o}C, respectively. Nitrogen was used as carrier and makeup gas. The flow rate was 1 mL min^{-1} with a split ratio of 80:20. Hydrogen and air were used for flame. Each sample produced a chromatogram with distinct peaks at various retention times. The retention time of each fatty acid was compared using a mixture of standard methyl esters of fatty acids run in the GC-FID under the same conditions as the samples.

Moisture and ash contents

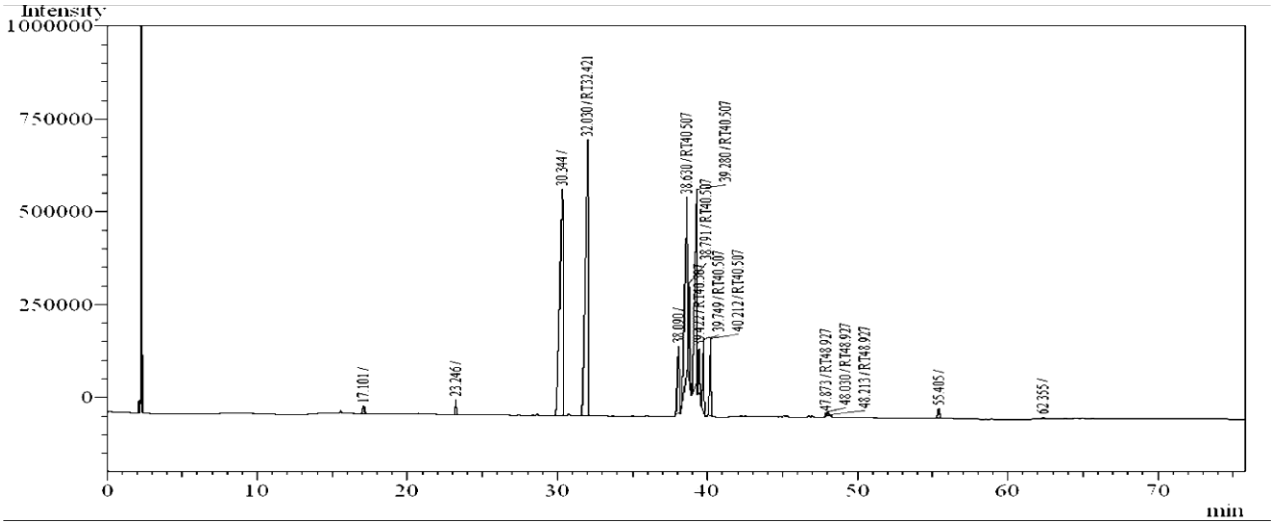
Weight loss techniques using heating crumbs from crushed cookies were used to measure the cookie moisture content. In three previously weighed porcelain crucibles, 2.0g of sample was weighed and placed in each of them. The crucibles were placed in a muffle furnace at 105°C for 4 hours. The residues were cooled and the weights were measured. For each crucible, the percentage of moisture was obtained and then the mean value was calculated. The moisture content residues in the same porcelain crucibles were heated in a muffle furnace for an additional 4 hours at 700^{o}C. The crucibles were then cooled in desiccators and weighed.

Statistical Analyses

Each sample of the triplicate treatments was analyzed and the results were expressed as mean values ± standard deviation (SD).



(a)



(b)

Fig 1. GC Chromatogram of (a) the standard fatty acids and (b) the sample B3

Table 1. Total fat, ash and moisture content of the cookie samples*

Sample	Fat content (g/100g)		Moisture content (%)		Ash content (%)	
	(Mean ± SD)	RSD (%)	(Mean ± SD)	RSD	(Mean ± SD)	RSD
A1	18.16±0.23	1.25	2.11±0.0	13.71	0.73±0.0	5.48
A2	21.51±0.58	2.72	1.73±0.0	12.14	1.17±0.0	11.20
A3	23.83±0.30	1.28	1.43±0.0	18.81	0.83±0.0	11.02
A4	16.13±0.53	3.31	2.22±0.0	8.76	0.94±0.0	4.79
A5	20.62±0.57	2.76	2.85±0.0	7.53	1.21±0.0	9.43
B1	20.94±1.21	5.77	0.79±0.0	8.96	1.82±0.0	0.25
B2	20.33±1.64	8.09	2.26±0.03	2.34	4.23±0.0	1.21
B3	26.84±1.22	4.53	4.82±0.02	8.58	1.29±0.0	10.05
B4	16.31±2.12	13.02	2.16±0.0	18.73	3.60±0.0	3.47
B5	20.56±1.09	5.28	4.15±0.0	5.66	2.80±0.0	4.93

*Each fatty acid value in the table represents the mean ± standard deviation of three replicates.

Table 2. Fatty acid compositions of various bakery cookies consumed in Bangladesh*

Fatty acids (%)		Sample									
		A1	A2	A3	A4	A5	B1	B2	B3	B4	B5
C8:0	Caprylic acid	0.99	-	-	-	-	0.84	-	-	-	-
C10:0	Capric acid	1.45	-	-	-	-	1.06	-	-	-	-
C12:0	Lauric acid	4.62	0.20	-	-	-	-	0.18	0.50	0.20	-
C14:0	Myristic acid	47.06	1.15	1.04	0.94	1.10	-	0.97	0.56	1.10	1.03
C16:0	Palmitic acid	24.08	-	-	-	-	5.08	0.19	24.23	-	-
C18:0	Stearic acid	-	30.44	7.53	8.17	6.27	-	32.62	1.85	29.90	32.60
C20:0	Arachidic acid	-	0.47	0.42	0.31	0.39	0.81	0.40	0.06	0.36	-
C22:0	Behenic acid	-	-	-	-	-	-	0.06	0.08	-	-
	∑SFA[a]	78.20	32.26	8.98	9.42	7.76	7.78	34.42	27.27	31.55	33.62
C16:1n-7	Palmitoleic acid	9.66	42.60	47.20	45.09	52.41	27.03	45.52	25.59	47.61	47.77
C18:1n-9	Oleic acid	-	9.38	31.42	32.85	27.15	0.22	4.14	3.40	3.74	3.59
	∑MUFA[b]	9.66	51.98	78.62	77.93	79.56	27.24	49.66	28.98	51.34	51.35
C18:2n-6	Linoleic acid	-	8.39	7.56	6.43	8.80	0.38	7.81	3.90	6.57	6.66
C18:3n-3	α-Linolenic acid	-	1.24	0.58	1.68	0.37	-	0.08	0.50	2.89	-
	∑PUFA[c]	0.00	9.64	8.14	8.12	9.17	0.38	7.89	4.40	9.46	6.66

*Results are expressed as the percentage of total fatty acids. 'A' represents sweet cookies whereas 'B' representing savory cookies.
[a] Saturated fatty acids, [b]Monounsaturated fatty acids, [c]Polyunsaturated fatty acids
"-" indicates Not Detected

III. Results and Discussion

The average fat contents obtained from the different cookie samples were expressed as relative to 100g of the product. The amount of total fat in the samples varied from 16.13±0.53 to 26.84±1.22 g/100g of the samples (Table 1). In case of sweet cookies, the highest amount of fat was found in sample A3 (23.83%), while sample A4 (16.13%) had the least, and as for savory cookies, B3 (26.84%) had the highest fat content whereas B4 (16.31%) had the least. The fat content of cookie samples differed due to the manufacturers' use of different recipes and depended on the quantity of fat used in cookie preparations.

The fatty acid (FA) compositions of various cookie samples are as shown in Table 2. The profile of fatty acids showed a lot of variation, implying that several different types of fats were used in making of these cookies. Overall, except for A1, monounsaturated fatty acids (MUFA) dominated the fatty acid composition of all analyzed cookies, accounting for 9.66 to 79.56% of total fatty acids. Saturated fatty acids (SFA) in the cookie samples varied considerably, ranging from 7.76 to 78.20 % of the total FAs. In case of sweet cookies, A1 had the highest percentage of SFA (78.20%), while A5 (7.76%) had the lowest, and as for savory cookies, B5 had the highest percentage of SFA whereas B1 (7.78%) had the lowest. Myristic acid (C14:0), stearic acid (C18:0) and a small percentage of arachidic acid (C20:0) were the major SFAs found in all sweet cookie samples; only A1 had a large proportion of palmitic acid (C16:0). The main SFA in the analyzed savory cookie samples were myristic (C14:0), palmitic (C16:0) and stearic acid (C18:0), while a small quantity of arachidic acid (C20:0) was also determined in a few samples. The presence of palmitic acid suggested that palm oil could be present.[24]

In terms of monounsaturated fatty acid (MUFA), the highest level was detected in A5 (79.56%) while A1 (9.66%) had the least amount. Both sweet and savory cookie samples, with the exception of A1, had very high percentages of MUFA. Among the MUFAs, the major contributor was palmitoleic acid (C16:1n-7) which varied in the range of 9.66-52.41 and 25.59-47.77% in sweet and savory cookies, respectively, followed by oleic acid (C18:1n-9) which was found in the range of 9.38-32.85%

in sweet cookies and 0.22-4.14% in case of savory ones. Oleic acid was absent in B1.

Among the polyunsaturated fatty acids (PUFA), linoleic acid (C18:2n-6) was present in the highest amount ranging from 6.43-8.80% in sweet cookies and 0.38-7.81% in savory cookies. Linolenic acid (C18:3n-3) was found in the range of 0.37-1.68 and 0.08-2.89% in sweet and savory cookies, respectively. PUFA was absent in B1.The low PUFA content may suggest the use of solid fats.[1]

The identification and quantification of trans fatty acids (TFA) were not doable due to the inadeauacy of the standard used for analyzing the fatty acid profiles of the cookie samples, but it is very much possible that some of the cookies may contain considerable amount of TFAs. This is because of the vegetable oils are high in unsaturated fatty acids with only non-conjugated double bonds in the cis configuration. But these unsaturated components can be isomerized to the trans during extraction, heating, processing, oxidation, and partial hydrogenation for the production of a wide range of bakery items including cookies.[25] The structure, physical properties, chemical stability, and physiological effects of trans fatty acids are similar to those of saturated fatty acids, in comparison to cis unsaturated fatty acids.[19]

The highest percentage of saturated fatty acids discovered in this investigation was obtained from A1 and B2. The lowest amount of unsaturated fatty acids was detected in A1. Apart from A1, the cookies produced by the other bakeries had a high percentage of MUFAs. The samples also had a considerable percentage of PUFAs, except for B1. As per World Health Organization, eating foods high in saturated fatty acids and fatty acid trans isomers has detrimental health effects. High amount of saturated and trans fatty acids in the diet can raise levels of the harmful LDL cholesterol in the bloodstream, leading to heart artery blockages and heart attacks.[26, 27] Furthermore, particularly beneficial monounsaturated fatty acids, such as oleic acid, as well as necessary fatty acids, such as linoleic acid were present in low amounts with the exception of A3, A4 and A5 as they had considerable amount of oleic acid.

The results indicate that all the cookie samples that are analyzed contain significant amount of fats, so excessive consumption of these cookies may lead to obesity and have negative health consequences like hypertension and cardiovascular diseases.[28]

Moisture content in cookies is vital since it influences variables such as taste, texture, stability, appearance, shape, and weight, as well as shelf life, texture, freshness and resistivity to microbial contamination.[29] The maximum limit of moisture content is 5.0% recommended by Bangladesh Standards and Testing Institution (BSTI) for cookies (BDS383 2001).[30] The results showed that the average moisture content of the cookies varied between 1.43-2.85 and 0.79-4.82% for sweet and savory cookies, respectively. Among all the samples, the percentage of moisture in B1 was the lowest, while B5 had the highest moisture content. The moisture contents of both the sweet and savory cookies evaluated are relatively low and within 1.0-5.0%. This means that these cookies have longer shelf lives and can be stored for longer periods of time prior to consumption without spoiling.

The mineral elements or inorganic materials in general that are found in cookie samples are related to the ash content of the cookies.[31] The ash content of a food is essential because the quantity of minerals can affect its nutritional value, quality and microbial stability.[32] The results showed that the average ash content of the cookies varied between 0.73-1.21 and 1.29-4.23% for sweet and savory cookies, respectively. The sample with the least ash content was A1, whereas the sample with the most ash content was B2. In comparison to the savory cookies, the sweet cookies had a lower overall ash content. This could be because savory cookies include salt.

IV. Conclusion

This investigation demonstrates that the average fat content of the cookies ranged from 16.13±0.53-26.84±1.22 g/100g of the samples which is alarming. The results obtained showed that, in general, the most abundant fatty acids present in the cookies were palmitoleic and stearic acid followed by oleic and linoleic acid. Either way, these cookies should be consumed in moderate amounts to remain healthy and avoid the risk of being obese. Since cookies, nowadays, have become a regular and popular food item among people of various socioeconomic classes, the results may aid in the development of public health policies that can affect a large population. Moreover, the findings of this study can be used to enrich dietary charts available in Bangladesh.

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