

## PHYSICOCHEMICAL EVALUATION OF COOKING BUTTER AND HYDROGENATED OILS

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### ABSTRACT

This research aimed to evaluate the physical and chemical properties of cooking butter and hydrogenated oils sold in Assiut Governorate, in which 70 samples (35 of each type) were collected from different localities in Assiut Governorate in the period from November 2021 to April 2022, and a number of tests were conducted to find out the physicochemical evaluation, which included pH, moisture%, free fatty acids%, acid value, peroxide value, *p*-anisidine value and total oxidation, then the obtained results were compared with the permissible limits of the Egyptian Standard specifications. The average results for cooking butter and hydrogenated oil samples were 6.02 and 5.45 for pH, respectively, 24.16 and 0.33% for moisture%, respectively, 1.86 and 0.4% for free fatty acids%, respectively, 2.63 and 0.57 for acid value, respectively, 2.18 and 1.83 for peroxide value, respectively, 2.23 and 1.98 for *p*-anisidine value, respectively, and 6.59 and 5.61 for total oxidation respectively; and when compared with the Egyptian Standards, it was found that 97.14 and 34.29% of the cooking butter and hydrogenated oil samples, respectively, were above the permissible limits for moisture%. Also, 97.14% of the cooking butter samples were above the permissible limits for peroxide value, as well as, all the cooking butter samples exceeded the permissible limit for free fatty acids%, but 28.57% of the hydrogenated oil samples exceeded the permissible limit for acid value.

**Keywords:** Physicochemical evaluation, cooking butter, hydrogenated oils

### INTRODUCTION

Edible fats & oils are important nutritional components with a variety of functions in our bodies (Endo, 2018). Edible fats & oils are food substances of plant, animal or microbial origin that are manufactured for human consumption. Fats & oils are edible as they consist of

carboxylic acid with long hydrocarbon chains. The carboxylic group provides the site for enzymes accelerating the metabolism of food substances & ultimately absorption of diet (Arunima and Rajamohan, 2013).

Several physical & chemical parameters as peroxide value, moisture content & acid value are parameters of interest to determine the shelf-life quality and consequently the economic value of fats & oils (Endo, 2018). Moreover, free fatty acids (FFAs) formation might be an important measure of the rancidity of food. FFAs are formed due to the hydrolysis of triglycerides and may get promoted by the reaction of oil with moisture (Freja *et al.*, 1999).

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Peroxide value (PV) is a widely used measure of primary lipid oxidation indicating the amount of peroxides formed in fats & oils during oxidation (Ozkan *et al.*, 2007). Peroxide is the 1<sup>st</sup> compound that is produced after the oxidation of fats & oils. It had negative impacts on human health and may contribute to different diseases (Pizzino *et al.*, 2017). Rancidity of vegetable oils may pose health risks including cancer and inflammation because of the formation of toxic and reactive oxidation products (Mukherjee and Mitra, 2009).

Microorganisms may cause chemical changes in edible fats & oils leading to lowering the quality of edible fats & oils (Okpokwasili and Molokwu, 1996). The lipolytic fungal activity on fats & oils triglycerides used in baking may cause rancidity, acidity, bitterness, soapiness & other off flavors. These activities may occur in seeds or other plant parts in which oils are taken (Larry, 1987).

The Egyptian Standards put specifications in order to judge the fatty products like cooking butter and hydrogenated oils. The researchers compare their results to be compatible or incompatible with the permissible limits of the Egyptian Standards.

This study aimed to evaluate the quality of cooking butter and hydrogenated oils sold in Assiut governorate. The evaluation was done according to the physical & chemical properties of such products.

## MATERIALS AND METHODS

### Samples collection:

A total of 70 random samples of cooking butter and commercial hydrogenated oils (35 each). The samples were collected from different localities in Assiut governorate, in their packages as sold to the consumer, in the period from November 2021 to April 2022. The collected samples were transferred to the laboratory as soon as possible to be examined.

### Physicochemical examination:

#### 1) pH measurement:

pH value of the samples was measured according to Tadesse *et al.* (2017). Accurately 50 g of the sample was warmed to 40° C in order to be melted. pH meter (Adwa ad11 waterproof pH-temp pocket tester) was calibrated with 2 standard buffer solutions having pH of 4 and 7, then the electrode was inserted into the melted sample, and between each measurement, the electrode was rinsed with warm distilled water.

#### 2) Moisture %:

The moisture% of the samples was done according to AOAC (1990). A porcelain dish was firstly washed and then dried by heating for at least 1 h in the drying oven set at 102° C, then cooled in a desiccator to the temp of the weighing room and the dish was weighed with an analytical balance. Approximately, 5 g of the samples were weighed into the porcelain dish. The test portion and the dish were heated for 2 hrs in the drying oven set at 102° C. The test portion and the dish were cooled in the desiccator after drying to the temp of the weighing room and the dish and its content were weighed. The drying, cooling and weighing procedures were repeated for periods of half an hour until the difference in mass between 2 consecutive weightings of the dish was not exceed 1 mg or until the mass was increased. The moisture % was calculated using the formula below:

$$\text{Moisture\%} = [(M_1 - M_2) / M] \times [100]$$

where:

M<sub>1</sub> = mass (g) of the test portion and the dish before drying

M<sub>2</sub> = mass (g) of the test portion and the dish after drying

M = mass (g) of the sample

#### 3) Free fatty acids% (FFAs%) (AOAC, 2000):

About 5 - 10 g of each sample were weighed into a 50 ml conical flask and 50 ml hot ethanol (99%) and 1 ml phenolphthalein indicator (1 g phenolphthalein in 100 ml ethanol) were added. The mixture was boiled for 5 min and then directly titrated against

0.1N NaOH (0.4 g NaOH in 100 ml distilled water) until the faint pink color persisted for 15 sec. The FFAs% was calculated using the formula below:

$$\text{FFAs\%}^* = (V \times N \times 28.2) / W$$

where:

\* FFA as oleic acid

V = volume of 0.1N NaOH used in titration

N = normality of NaOH (0.1)

W = sample weight (g)

28.2 is the normality of oleic acid

#### 4) Acid value (AV) (AOAC, 2000):

The acid value of the samples was measured upon the titration done in the previous FFAs%. The acid value was calculated as the equation below:

$$\text{AV} = (V \times N \times M) / W$$

where:

V = volume of 0.1N NaOH used in titration

N = normality of NaOH (0.1)

M = molecular weight of NaOH (40)

W = sample weight (g)

#### 5) Peroxide value (PV) (IDF, 2006):

##### Calibration curve determination:

**Standard stock solution of Fe (III)** was prepared by dissolving 0.5 g iron powder into 50 ml 10N HCl, then 2 ml hydrogen peroxide (H<sub>2</sub>O<sub>2</sub> 30%) was added (the excess of H<sub>2</sub>O<sub>2</sub> was removed by boiling for 5 min). The solution was cooled to room temp and was diluted with distilled water into the mark of 500 ml of the volumetric flask, in which, the concentration of the standard stock solution was 1000 ug/ml.

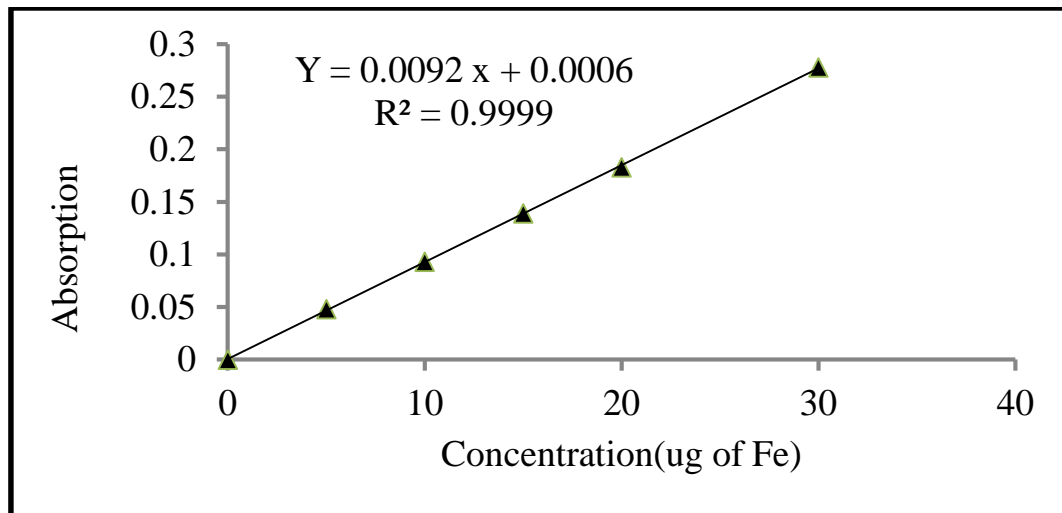
**Standard working solution of Fe (III)** was prepared by dissolving 5 ml of the standard stock solution in 50 ml of chloroform:methanol (70:30), in which, the concentration of the standard working solution was 100 ug/ml.

**Calibration curve points** were determined according to the spectrophotometer absorption reading of the prepared following solutions:

Point*	Solutions (10 ml)				Fe (III) content in µg	Spectrophotometer absorption reading**
	Standard working solution (ml)	Ammonium thiocyanate (15 g in 50 ml dist. water) (ml)	10N HCl 2% (ml)	Chloroform: methanol (70:30) (ml)		
1	0	0.05	0.05	9.9	0	0
2	0.5	0.05	0.05	9.4	5	0.048
3	1	0.05	0.05	8.9	10	0.093
4	1.5	0.05	0.05	8.4	15	0.139
5	2	0.05	0.05	7.9	20	0.183
6	3	0.05	0.05	6.9	30	0.278

\* Each point in the calibration curve was obtained after 3 replicates measurement

\*\* Spectrophotometer absorption reading was at 500 nm against blank reagent solution



Y= absorption

X = concentration

0.0092 = slope of calibration graph

0.0006 = Y- intercept

### Sample preparation:

Accurately, 5 g of each sample was weighed into a centrifuge tube, in addition, 1 g of anhydrous sodium sulfate was added. The centrifuge tube was placed in the oven at 40° C for fat melting. The fat layer was separated by centrifugation (at 5000 rpm for 5 min) and filtered with dry filter paper in the oven at 40° C, in which the filtered fat was used for analysis.

### Sample analysis:

Firstly, iron (II) solution was prepared by dissolving 0.2 g barium chloride dihydrate in 25 ml distilled water as 1<sup>st</sup> solution, then 0.25 g iron (II) sulfate heptahydrate was dissolved in another 25 ml distilled water as 2<sup>nd</sup> solution which was added slowly with stir to the 1<sup>st</sup> solution, then 2 ml 10N HCl was added, and finally, the obtained iron (II) solution was filtrated to obtain a clear solution.

About 0.05 - 0.3 g of the prepared sample was weighed in a test tube, then 9.6 ml chloroform:methanol (70:30) was added with mixing for 4 sec, after that, 0.05 ml ammonium thiocyanate (15 g in 50 ml distilled water) was added with mixing for another 4 sec, and final addition of 0.05 ml iron (II) solution with mixing for 4 sec, then the test tube was incubated at room

temperature for 5 min. Moreover, another test tube with the same contents and procedures omitting the sample was prepared as a blank.

The absorption was read at 500 nm against blank by spectrophotometer (721, VIS spectrophotometer, Prolab, China). The peroxide value (expressed as mequiv) was calculated according to the formula below:

$$PV = [A_s - A_b] / [55.84 \times m \times W_s \times 2]$$

where:

$A_s$  = absorbance of the sample

$A_b$  = absorbance of the blank

$m$  = slope of the calibration curve (0.0092)

$W_s$  = sample weight (g)

55.84 is the atomic weight of iron (III)

2 is the factor to convert mequiv (ml equivalent O<sub>2</sub>/Kg fat) of Fe to mequiv of peroxide

### 6) *p*-ansidine value (*p*-AV) (AOCS, 1993):

#### Sample preparation:

Accurately, 5 g of each sample was weighed into a centrifuge tube, in addition, 1 g of anhydrous sodium sulfate was added. The centrifuge tube was placed in the oven at 40° C for fat melting. The fat layer was separated by centrifugation (at 5000 rpm for 5 min) and filtered with dry filter paper in the oven at 40°

C, in which the filtered fat was used for analysis.

### Sample analysis:

About 0.5 – 2 g of the prepared sample were dissolved in 25 ml isooctane (2,2,4, trimethyl pentane) using a 25 ml volumetric flask. The blank was only 25 ml isooctane without the sample. The measurement of the absorbance (Ab) was run at 350 nm against blank using spectrophotometer (721, VIS spectrophotometer, Prolab, China).

After that, 5 ml of the previous fat solution (the sample plus 25 ml isooctane) was pipetted into a test tube and 1 ml *p*-ansidine 0.25% reagent (0.25 g *p*-ansidine in 100 ml glacial acetic acid) was added with mixing. Also, the blank was prepared as 5 ml isooctane plus 1 ml *p*-ansidine 0.25%. After 10 min incubation at room temperature in a dark place, the absorbance (As) was read at

350 nm against blank using spectrophotometer (721, VIS spectrophotometer, Prolab, China). The *p*-AV was calculated according to the formula below:

$$p\text{-AV} = [ 25 \times (1.2 A_s - A_b)] / m$$

where:

$A_s$  = absorbance of fat solution after reaction with *p*-ansidine

$A_b$  = absorbance of fat solution before reaction with *p*-ansidine

$m$  = sample mass (g)

25 is the volume of isooctane (ml) in which the test sample was dissolved

1.2 is the correction factor of the fat solution with 1 ml *p*-ansidine

### 7) Total oxidation value (TOTOX) (De Abreu *et al.*, 2010):

$$\text{TOTOX} = 2 \text{ PV} + p\text{-AV}$$

## RESULTS

**Table 1:** Physicochemical properties of the examined samples.

Parameter	Cooking butter samples			Hydrogenated oils samples		
	Min.	Max.	Average	Min.	Max.	Average
pH	5	7.3	6.02	4	7.2	5.45
Moisture%	13.67	41.73	24.16	0.1	1.2	0.33
Free fatty acids%	0.62	5.36	1.86	0.17	2.76	0.4
Acid value	1	7.6	2.63	0.24	3.92	0.57
Peroxide value	0.16	3.96	2.18	0.32	3.35	1.83
<i>p</i> -ansidine value	0.62	13.54	2.23	0.86	5.57	1.98
Total oxidation	1.05	16.26	6.59	2.75	11.21	5.61

**Table 2:** Physicochemical evaluation of the examined cooking butter samples according to the Egyptian Standards (2005a)

Parameter	Egyptian Standards (2005a)	Samples number			
		Below*		Above**	
		No./35	%	No./35	%
Moisture%	Not more than 16%	1	2.86	34	97.14
Free fatty acids%	Not more than 0.4%	0	0.00	35	100.00
Peroxide value	Not more than 0.6 mequiv	1	2.86	34	97.14

\* Below Egyptian Standards (2005a)

\*\* Above Egyptian Standards (2005a)

**Table 3:** Physicochemical evaluation of the examined hydrogenated oil samples according to the Egyptian Standards (2005b).

Parameter	Egyptian Standards (2005b)	Samples number			
		Below*		Above**	
		No./35	%	No./35	%
Moisture%	Not more than 0.2%	23	65.71	12	34.29
Acid value	Not more than 0.6	25	71.43	10	28.57
Peroxide value	Not more than 10 mequiv	35	100.00	0	0.00

\* Below Egyptian Standards (2005b)

\*\* Above Egyptian Standards (2005b)

## DISCUSSION

The physicochemical properties of the examined samples were shown in Table 1. The pH values of the examined cooking butter and hydrogenated oil samples ranged from 5 to 7.3 and 4 to 7.2, respectively, with averages of 6.02 and 5.45, respectively. The obtained pH values of the cooking butter samples were higher than those reported by Şenel *et al.* (2011), Erkaya *et al.* (2015), Akgül *et al.* (2021) but lower than those obtained by Mourad and Bettache (2018).

The moisture content of the examined cooking butter and hydrogenated oil samples ranged from 13.67 to 41.73% and 0.1 to 1.2%, respectively, with averages of 24.16 and 0.33%, respectively. The obtained moisture content of the cooking butter samples (Table 1) exceeded the values reported by Hanaa *et al.* (2014), Lina *et al.* (2018), Akgül *et al.* (2021). Also, the obtained results of the hydrogenated oil samples for moisture content were higher than those obtained by Kandhro *et al.* (2013).

Free fatty acids (FFAs) formation is due to hydrolysis, cleavage and oxidation of lipids' double bonds. FFAs value of fresh butter which is between 0.14 to 0.39% is generally assumed to be passable for butter fat and a higher content of FFAs is related to poor storage condition (Nadeem *et al.*, 2014; Erkaya and Sengul, 2015). In the present study, the FFAs% of the examined cooking butter and hydrogenated oil samples varied between 0.62 to 5.36% and 0.17 to 2.76%

with averages of 1.86 and 0.4%, respectively. These obtained results of the cooking butter samples for FFAs% (Table 1) were lower than those obtained by Lina *et al.* (2018), while, the obtained results of the hydrogenated oil samples were higher than those obtained by Tahir *et al.* (2013), Tripathi and Yadav (2021). The acid value results (Table 1) of the examined cooking butter and hydrogenated oil samples ranged from 1 to 7.6 and 0.24 to 3.92 with averages of 2.63 and 0.57, respectively. It was observed that the obtained acid value of the examined cooking butter samples was lower than the value of Saba *et al.* (2018).

The peroxide value (PV) is related to hydroperoxides which are unstable & readily decompose forming mixtures of volatile aldehyde compounds. The oxidative degradation compounds are generally termed "secondary oxidative products" which are determined in oils and fats by methods such as *p*-anisidine (*p*-AV) (Ramadan and Mörsel, 2004). The obtained results in Table 1 showed the peroxide value (PV) of the cooking butter and hydrogenated oil samples ranged from 0.16 to 3.96 and 0.32 to 3.35 with averages of 2.18 and 1.83, respectively. The obtained results of the examined cooking butter samples for PV were higher than those of Asdagh and Pirsá (2020), Hassan *et al.* (2022), also, the obtained results of the hydrogenated oil samples were higher than the results of Kandhro *et al.* (2013), Tripathi and Yadav (2021).

The *p*-anisidine results tabulated in Table 1 of the examined cooking butter and

hydrogenated oil samples varied between 0.62 to 13.54 and 0.86 to 5.57 with averages of 2.23 and 1.98, respectively. The obtained results for *p*-anisidine were higher than those of Tadesse *et al.* (2017) for the cooking butter samples and also higher than those of Tripathi and Yadav (2021) for the hydrogenated oil samples.

The resultant total oxidation (TOTOX) from peroxide and *p*-anisidine values of the examined cooking butter and hydrogenated oil samples ranged from 1.05 to 16.26 and 2.75 to 11.21 with averages of 6.59 and 5.61, respectively.

In order to judge the examined cooking butter samples, their obtained results in Table 1 were compared with the Egyptian Standards (2005a) tabulated in Table 2, in which the majority of the cooking butter samples were incompatible with the Egyptian Standards, as 97.14%, 100% and 97.14% of the cooking butter samples were above the permissible limits for moisture%, FFAs% and peroxide value, respectively.

For the judgment of the examined hydrogenated oil samples, their obtained results in Table 1 were compared with the Egyptian Standards (2005b) tabulated in Table 3, in which 34.29% and 28.57% of the hydrogenated oil samples were incompatible with the Egyptian Standards for moisture% and acid value, respectively. But all the hydrogenated oil samples were compatible with the Egyptian Standards for peroxide value (Table 3).

In conclusion, it was found that the majority of the examined cooking butter samples were unacceptable for peroxide value and free fatty acids % according to the Egyptian Standard; while, about a third of the examined hydrogenated oil samples were unacceptable for acid value according to the Egyptian Standard.

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### التقييم الفيزيائي الكيميائي للزبد الفلاحي والزيوت المهدرجة

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هدف هذا البحث إلى تقييم الخواص الفيزيائية والكيميائية للزبد الفلاحي والزيوت المهدرجة المباعة بمحافظة أسيوط، حيث تم جمع ٧٠ عينة (٣٥ من كل نوع) من مناطق مختلفة بمحافظة أسيوط في الفترة من نوفمبر ٢٠٢١ إلى أبريل ٢٠٢٢، وقد تم إجراء عدد من الإختبارات لمعرفة التقييم الفيزيائي الكيميائي والذي تضمن كل من الأس الهيدروجيني ونسبة الرطوبة ونسبة الأحماض الدهنية الحرة وقيمة الحمض وقيمة البيروكسيد وقيمة بارا أنسيدين والأكسدة الكلية، ثم مقارنة النتائج التي تم الحصول عليها بالحدود المسموح بها في المواصفات القياسية المصرية. وكانت متوسطات النتائج لعينات الزبد الفلاحي والزيوت المهدرجة بالنسبة للأس الهيدروجيني 6.02 و 5.45 على التوالي، ونسبة الرطوبة 24.16 و 0.33% على التوالي، ونسبة الأحماض الدهنية الحرة 1.86 و 0.4% على التوالي، وقيمة الحمض 2.63 و 0.57 على التوالي، وقيمة البيروكسيد 2.18 و 1.83 على التوالي، وقيمة بارا أنسيدين 2.23 و 1.98 على التوالي، والأكسدة الكلية 6.59 و 5.61، على التوالي، وبالمقارنة مع المواصفات القياسية المصرية فقد كان 97.14 و 34.29% من عينات الزبد الفلاحي والزيوت المهدرجة على التوالي، أعلى من الحدود المسموح بها لنسبة الرطوبة، وأيضا 97.14% من عينات الزبد الفلاحي أعلى من الحدود المسموح بها لقيمة البيروكسيد، وكذلك كل عينات الزبد الفلاحي تجاوزت الحد المسموح به للأحماض الدهنية الحرة، ولكن 28.57% من عينات الزيوت المهدرجة تجاوزت الحد المسموح به لقيمة الحمض.

**الكلمات المفتاحية:** التقييم الفيزيائي الكيميائي، الزبد الفلاحي، الزيوت المهدرجة