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Improving the Properties of Prepared Pastry Margarine by Using Nanotechnology

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ABSRTACT

In recent years nanotechnology-based products have been developed for use in many applications including food. Nano-emulsions are one of the most important applications because they have positive effects on the physical and chemical properties of various fatty products .In this study, pastry margarine was prepared using palm oil, palm stearin in addition of soybean and sunflower oils at the ratio of (5% soybean oil: 5% sunflower oil : 90% palm oil) and (5%soybean oil : 5%sunflower oil : 90% palm stearin) . Nanotechnology was used to modify the emulsifying properties of pastry margarine and to study the effect of these properties on its stability during storage period up to 11 weeks under cooling conditions at (4°C). Chemical, physical and emulsion characteristics of pastry margarine blends processed by nano-emulsion technique were determined and compared with the control samples. Nano-partecils of water phase which used in preparing of table margarine by nano-emulsion technique were examined using Transmission Electron Microscope and Zeta Potential Analyzer. Results showed that the margarine blends processed using this technique led to an improvement in emulsification properties and increased storage stability under refrigeration conditions. obtained results of chemical properties namely acid and peroxide values for margarine blends contained (5% soybean oil: 5% sunflower oil : 90% palm oil) and (5%soybean oil : 5%sunflower oil : 90% palm stearin) were determined and being 0.688 and 0.698mg KOH/gm oil and 3.198 and 3.290 ml.eqv/kg oil at the end of storage period in compared with the control samples which were 0.731 and 0.745 mg KOH/gm oil and 5.181 and 5.233 ml.eqv/kg oil. Results of physical properties namely colour and refractive index were also determined and indicated that all prepared margarine blends using nano-emulsion technique were more stable during storage than the others, while the results of emulsion stability indicated that the consistency of the creaming stability were nearly similar to those of control samples. Finally, it could concluded that using of nano-emulsion technique in preparing of pastry margarine could be prolonged stability and extend storage period.

Keywords: Nano-emulsion-pastry margarine- Transmission Electron Microscope- Zeta Potential Analyzer- Emulsion stability

INTRODUCTION

As a challenge towards the food crises, the world is seeking for a food product that can fulfill all health, safety, low price and satisfaction demands of consumers. fats and oils are one of the most important food constituents as it is the main source of energy and at the same time it has an important role in improving the palatability of foods especially the highly spread fast foods(Badawy *et al*., 2015).

Margarine is water in fat emulsion in which water droplets are kept separated by the fat crystals. It was categorized according to demand by different consumers and based on melting points and hardness Dian and Sahri (2011).

Pastry margarine is used in a wide range of savoury and fruit products. Fats for the pastry are supposed to have a hard consistency so that when they are being blended into the dough, it retains enough formation under shear conditions to be spread as protective thin films and droplets all through the dough (Vereecken, 2010).

In recent years, nanotechnology-based products have been developed to employ in a lot of applications that includes pharmaceuticals and foods. The emulsions with the micro-droplets, occasionally called conventional emulsions, and nano-dispersions, or thermodynamically stable emulsions (surprisingly called micro-emulsions), can be simply manufactured on an manufacturing scale up (Nidhi ,2015).

MATERIALS AND METHODS

Vegetable oils and fats

Refined, bleached and deodorized Palm oil fractions (palm kernel oil, palm stearin and palm oil), soybean oil and sunflower oil were obtained from Arma For Food Industries Company, 10th Ramadan city, Egypt.

Emulsifiers

Monoglycerides and Lecithins were obtained from Sigma Chemical Company, Cairo, Egypt.

Coloring agent

Beta carotene as colouring matter was obtained from Shehab Company for Dairy Pro. Talkha City, El-Dakahlia Governorate, Egypt..

Flavouring agent

Ghee commercial essence used as flavoring agent was obtained from Misr for Oil and Soap Company, El Mansoura city, El-Dakahlia Governorate, Egypt.

Other ingredients

Potassium sorbate, citric acid and sodium chloride were obtained from El-Gomhoria Pharmaceutical and Chemicals Company, El Mansoura city, El-Dakahlia Governorate, Egypt and skim milk was secured from Shehab Company for Dairy products, Talkha City, El-Dakahlia Governorate, Egypt. While commercial essential oil (Thyme extract)) was obtained from local Fathalla market, El-Mansoura city, El-Dakahlia Governorate, Egypt.

Commercial margarine:

Pastry commercial margarine were obtained from supermarket Hayber One, Cairo,Egypt.

Methods

Preparation of fat phase blends

Soybean oil "SBO" and sunflower oil "SFO" were added to palm stearin oil "PSO"and palm oil "PO" with different ratios as mentioned in Table(1) .

Table 1. Vegetable oils and fats blends used in fat phase preparation:

Blends	Vegetable oils and fats			
	SBO	SFO	PO	PS
Blend(3)	5	5		90
Blend(4)	5	5	90	

All blends were melted at 50ºC for 20 min then cooled quickly at 4±1ºC and stored in domestic refrigerator at 4±1 ºC until further chemical and physical analysis and solid fat contents were carried out according to the method described by Mirhosseini *et al*., (2008,a).

Preparation of water phase

Water phase was presented as 12% of emulsion contained adequate amount of water and other ingredients were mentioned in Table (2). Water phase was prepared by two methods as follow:

First method for regular type, all ingredients were dissolved in distillated water and then mixed gradually by

MPW-120 homogenizer,1500 rpm for 5 min as stated by (ESS,2007) at Food Industry Dept. Research Lap. Fac. of Agric. Mansoura University. Second method all ingredients were also dissolved in water but mixed using Omni International 17106 Varriable- Speed Homogenizer, 18,000rpm; 220V at Dep. of Pharmacology and Toxicology, Fac. of Pharm. Mansoura University to obtain nano-emulsion particles.

Table 2. Fat and water phases used in margarine processing:

Ingredients	Function	%
a)Fat phase:(88%)		
1-Fat blends		87%
2-Lecithin	Emulsifier	0.3%
3-Monoglycerides	Emulsifier	0.3%
4-Beta carotene	Coloring agent	0.1%
5-Essence(butter essence)	Flavoring matter	0.3%
b) Water phase (12%)		
1-Water		11%
2-Potassium sorbate	Preservative	0.2%
3-Citric acid	Preservative	0.2%
4-Sodium chloride	Preservative	0.1%
5-Thyme extract	antioxidants	0.02%
6-Skim milk	stabilizer	0.5%

Margarine processing

Pastry margarine with different types of emulsions (traditional and nano) were formulated by dissolving emulsifiers (lecithin 0.3% and monoglycerides 0.3%) and other soluble fat ingredients(Beta carotene 0.1% and essence 0.3%) in fat blends at 70ºC then added to water phase using MPW-120 homogenizer,1500rpm for 5 min at Food Industry Dept. Research Lap. Fac. of Agric. Mansoura university. The prepared margarine was finally cooled to 4±1ºC and stored at domestic refrigerator until further analysis was carried out as stated by (ESS,2007).

Characterization of Nano-emulsion particles

Transmission electron Microscope Measurement (TEM)

Size of nano-emulsion particles was characterized using Transmission Electron Microscopy, TEM (JEOL TEM-2100) connected to CCD camera at an accelerating voltage of 200 KV according to the method described by Smith,(2015) . Measurements were recorded at the Central Laboratory, Electron Microscope Unit, Faculty of Agriculture, Mansoura University, Mansoura city, Egypt.

Zeta potential measurements

It was measured by Zeta Potential Analyzer Malvern Zeta -sizer Nano-zs90.Measurements were recorded at the Central Laboratory, Electron Microscope Unit, Faculty of Agriculture, Mansoura University, Mansoura city, Egypt KV according to the method described by Khoshnevisan and Barkhi ,(2015).

Physical characteristics

Refractive index (RI), melting point (MP) and colour were determined as the method described by (A.O.A.C.,2000)

Solid Fat Content (SFC)

Solid fat content was determined for different fats blends by Nuclear Magnetic Resonance (NMR) in temperature ranged between10-40ºC according to the method described by (A.O.A.C.,2000) at Food Quality Central Laboratory of Arma Food Industries, 10 Ramadan city, Egypt .

Microstructure of vegetable fat blends

Fat crystals were determined by Polarized Light Microscope (PLM) at Central Laboratory, Faculty of Agriculture, Mansoura University. Samples were examined at cooling system temperature to avoid melting of samples as the method described by Marangoni and Narine,(2002).

Proximate chemical analysis

Acid value (A.V) and free fatty acids "%" (F.F.A%) ,Peroxide value(P.V) and iodine value (IV) were determined according to the method described in A.O.A.C. (2005) at Food Industry Dept. Research Lap. Fac. of Agric. Mansoura University.

Thiobarbturic acid value (TBA)

Thiobarbituric acid (TBA) value was determined using UV-Vis Auto model UV- 2602 according to the method described by Pearson *et al*., (1983) at Food Industry Dept. Research Lap. Fac. of Agric. Mansoura University.

Fatty acids profile of vegetable oils and fats samples and their blends

Fatty acids methyl esters for vegetable oils and fats samples and their blends were determined by gas liquid chromatography (GLC) according to the method described by (Radwan.,1978) at the Food Tech. Res. Institute, Agric. Res. Center, El-Giza, Egypt.

Determination of total phenolic contents of Thyme extract

The Folin-Ciocalteu method was intended for determining total phenolic contents (as gallic acid equivalent) of thyme extract using standardized spectrophotometric in the Food Tech. Res. Institute, Agric. Res. Center, El-Giza, Egypt according to Ivanova *et al.,(*2010).

Determination of radical scavenging activity (DPPH%) of Thyme extract

2,2 diphenyl 1-picrylhydrazyl(DPPH%) assay was carried out according to the method that shown by Brand-Williams *et al*.,(1995) at Food Tech. Res. Institute, Agric. Res. Center, El-Giza, Egypt.

DPPH scavenging effect % = $A_{blank} - A_{sample} / A_{blank} \times 100$

Where A = Absorbance blank = all reagent except sample

Emulsion stability of prepared table and pastry margarine

Some properties were used to determine emulsion stability of prepared table and pastry margarine namely (lower phase%, separated phase and creaming stability) after preparing and prolonged storage as follow:

Lower phase %

Lower phase % was determined and calculated according to the method that described by Mirhosseini *et al*., (2008,a).

Lower phase% = (Lower phase height / initial emulsion × 100)

Separated layers%

Separated layers % was determined and calculated according to method that illustrated by Mirhosseini *et al*., (2008,a).

Separated layers% =

(The height of the separated layer/the total height of the emulsion) × 100

Creaming stability

Emulsion stability was measured by the change in the height of the bottom serum phase (Hs) within time and was compared with total height of emulsion(HE)

according to the method described by Klinkersorn *et al*., (2004).

Creaming index (CI) = 100× (HE/HS)

Where: HS =The height of the bottom serum phase
HE= Total height of emulsion

Oxidative stability of all prepared types of margarine:

Rancimat method according to A.O.A.C. (2000) at food Technology Research Institute, Agricultural Research Center, El-Giza, Egypt.

RESULTS AND DISCUSSION

Chemical and physical properties of (RBD) vegetable oils and fats used in table and pastry margarine processing:

In this study, there were four types of vegetable oils and fats used in table margarine processing, All oils namely soybean oil (SBO), sunflower oil (SFO), palm stearin (PS) and palm oil (PO) are used were refined, bleached and deodorized (RBD). Chemical and physical properties of these vegetable oils and fats were determined and the results were tabulated in Table (3).

It noticeable from chemical properties that acid value (AV) and free fatty acids (FFA %) value were used as a measure of the formation of acidic compounds and secondary products that were formed during oxidation Khalaf, (2015).

From data presented in Table (3) it could be observed that AV and FFA% of (RBD)soybean and sunflower oils were 0.351,0.266 mg KOH/gm oil and 0.228,0.208%) respectively while palm stearin and palm oil being 0.471, 0.373 mg KOH/gm oil and 0.354,0.291%) respectively. These results may be due to the formation of fatty acids composition of vegetable oils and fats.

These results were nearly accordance with those found by (El-Ghonamy *et al*.,2015) whose reported that AV of soybean and sunflower oils were (0.290 and 0.170 mg KOH/gm oil) respectively, while (Oladiji *et al*., 2010) whose reported that FFA% of soybean was ranged from (0.3:1.01%).

Peroxide value(PV) used as an index for the early stage of oxidation process and degree of lipids oxidative rancidity and the formation of hydro-peroxides compounds (Karoui *et al*.,2011). Also in Table (3), results indicated that values of PV were2.630, 1.872, 3.310 and 2.923 ml.eqv/kg oil for soybean, sunflower, palm stearin and palm oil respectively.

Our obtained results were nearly in accordance with those of El-Ghonamy *et al*.,(2015), they found that PV of soybean oil and sunflower oil were 1.57 and 1.39 ml.eqv\kg oil respectively, while El-Gammal (2006) reported that values of PV for palm stearin and palm oil were 2.650 and 3.310 ml.eqv/kg oilrespectively.

Iodine value (IV) is a quantitative measure of unsaturation in lipids. The principle of the assessment is based on the fact that halogens add to the double bonds of unsaturated fatty acids. It can be seen from Table (3) that iodine value of (RBD) soybean, sunflower, palm stearin and palm oil were 122.32, 103.11, 45.77 and 57.98gm/100gm oil respectively.

These results were nearly accordance with those found by Zaliha *et al*., (2014) they reported that values of IV for soybean oil,palm stearin and palm oil were 134.20, 41.90 and 52.69 gm/100gm oil, while Hashem and El-Wasief, (2016) found that IV of sunflower oil was 131.60 gm/100gm oil.

Results of physical properties were also presented in Table (3). Refractive index values were 1.022, 1.218, 1.441 and 1.428 for SBO, SFO, PS and PO respectively.

Colour is one of the main factors for determining quality of oils. Vegetable oils have minimum values of colour index are more suitable for edible purposes Askar,(2017). From data presented in Table (3) it could be observed that the light yellow colour was in palm stearin and palm oil, it is primarily due to B-carotene content, it was higher in palm oil (1.367 at 440 mm) compared with palm stearin (1.095at 440 mm), this may be due to the effect of temperature which was used in different steps of processing steps for palm oil (El-Gammal,2007).

Also, results in Table (3) indicated that colour were (1.222 and1.412) respectively for soybean oil and sunflower oil. These values are nearly close to Saafan,(2014).

Melting point of fats is a parameter used to characterize oils and fats quality and is related to their physical properties, such as hardness and thermal behavior. Results in Table (3) indicated that melting point in palm stearin and palm oil were 45 and 26 at 40ºC respectively. This due to their content of palmitic acid.

These results were nearly in accordance with those found by El-Waseif and Hashem, (2017) whose reported that melting point of palm stearin and palm oil was 38.5 and 27.5 respectively.

Table 3. Chemical and physical properties of (RBD) vegetable oils and fats used in table and pastry margarine processing:

Vegetable oils and fats samples(RBD) properties	SBO	SFO	PS	PO
Acid value(AV)mg KOH/gm oil	0.351	0.266	0.471	0.373
Free fatty acid (FFA%)(as oleic acid%)	0.228	0.208	0.354	0.291
Peroxide value(PV) (ml.eqv\kg oil)	2.630	1.872	3.310	2.923
Iodine value(IV) (gm\100gm oil)	122.32	103.11	45.77	57.98
Refractive index(RI) (at40ºC)	1.022	1.218	1.441	1.428
Colour(at 440 mm)	1.222	1.412	1.095	1.167
Melting point at40ºC(MP)	NM	NM	45	26

NM: Not measured SBO:soybean oil, SFO:sunflower oil, PS:Palm stearin, PO:Palm oil
RBD: Refined, Bleached and Deodorized

Fatty acids profile of (RBD) vegetable oils and fats used in table and pastry margarine processing:

Most studies on lipids for health and nutrition have focused on their fatty acids composition. However, the distribution of these fatty acids in the triacylglycerol molecule is specific for the native fats and oils (Ract *et al*., 2015).

From data mentioned in Table (4), it is noticeable that saturated and unsaturated fatty acids could be specified in all samples. The amount of saturated fatty acids were increased with nearly four time in palm oil fractions namely palm stearin and palm oil in compare with those of soybean and sunflower oils. Also, it could be noticed that from the same table the amount of saturated and unsaturated fatty acids were approximately equal in palm oil. Palmatic acid (C16:0) was considered as the predominate saturated fatty acid in both of palm stearin and palm oil with the percentage of 55.02 and 44.93%. On the other hand the epidemic unsaturated fatty acid in all oils

and fats samples was linoleic acid (C18:2) which ranged between (17.24%) to (55.12%). Data showed that levels of the values of linoleic acid (C18:2"ώ6") were (55.12, 50.54, 17.24 and 30.54%) for soybean oil, sunflower oil, palm stearin and palm oil respectively.From these mentioned data, it could be concluded that palm stearin and palm oil were convenient to hard stock fat processing (margarine and shortening) according to their fatty acids profile in the processing of diverse fat spreads and restrain stable against oxidation (Miskandar *et al*., 2002).

Table 4. Fatty acids profile of (RBD) vegetable oils and fats used in table and pastry margarine preparing:

Fatty acids	Vegetable oils and fats			
Saturated fatty acid (SFA)	**SFO**	**SBO**	**PS**	**PO**
Caproic acid (C6:0)	0.04	0.02	ND	ND
Caprylic acid (C8:0)	0.34	0.06	ND	ND
Capric acid (C10:0)	ND	ND	ND	ND
Lauric acid (C12:0)	0.23	0.10	0.28	0.19
Myristic acid (C14:0)	0.07	0.07	1.16	1.32
Palmatic acid (C16:0)	7.56	10.30	55.02	44.93
Margaric acid (C17:0)	0.04	0.09	0.16	0.10
Stearic acid (C18:0)	3.40	3.66	11.27	5.61
Arachidic acid (C20:0)	0.33	0.37	0.36	0.32
Behenic acid (C22:0)	0.50	0.45	ND	ND
TSFA	12.51	15.12	68.25	51.97
Unsaturated fatty acids(USFA)				
Palmitoleic acid (C16:1)	0.11	0.19	0.20	0.06
Heptadecenoic acid (C17:1)	0.02	0.04	0.02	0.21
Oleic acid (C18:1)	28.02	27.62	13.09	15.47
Linoleic acid (C18:2 "ώ6")	55.12	50.54	17.24	30.54
Linolenic acid (C18:3 "ώ 3")	4.07	6.03	1.17	1.31
Eicosenoic acid (C20:1)	0.15	0.46	0.03	0.12
TUSFA	87.49	84.88	31.75	47.71
TFA	100	100	100	100

ND: Not detected TSFA: Total Saturated Fatty Acids
TUSFA: Total Unsaturated Fatty Acids TFA: Total Fatty Acids

Total phenolic content (TPC)"mg/ g" and antioxidant activity (DPPH%) of Thyme extract:

Phenolic compounds are considered the major group of acting compounds as primary antioxidants and the terminator of free radicals. TPC was determined as Gallic acid equivalents (Zeyada *et al*., 2007). Results in Table (8) showed the percentage of total phenolic content in Thyme extraction which was 44.90±1.3 mg/ g as Gallic acid, while the antioxidant activity which was evaluated using the DPPH method reached to 58.80±4.6 %. Eghdami *et al*., (2013) reported that the total phenolic content of the extract of Thyme extract was 32.34±2.63 mg/ g as Gallic acid, while the antioxidant activity was 64.34±0.32 %.

Table 5. Total phenolic content (TPC)"mg/ g" and antioxidant activity (DPPH %) of Thyme extract

Antioxidant extract	TPC (mg/g) as Gallic acid	Antioxidant activity by DPPH%
Thyme extract	44.90± 1.3	58.80±4.6

Chemical and physical properties of fat blends used in fat phase preparation:

The composition of vegetable fats blends and chemical and physical properties have relationship and it has effect on fat phase formulation. Chemical and physical properties could be important to concurrence all blends for monitoring during processing and storage (El-Waseif and Hashem, 2017). The degree of lipids hydrolysis was contemplated by acid value and amounts of free fatty acids in all prepared blends. Results in Table (6) showed that AV and FFA% for blends 3 and 4 were 0.644 and 0.576 mg KOH/gm oil and 0.498 and 0.395% respectively.Oxidation stability of vegetable fat blends were explored by determination of peroxide and thiobarbituric acid values. Peroxide value is an indicator of oxidative rancidity stages of lipids and determined as milliequivalent of peroxide oxygen combined with one kg of oil/ fat Sanghi and Tiwle, (2015).Peroxide values of these blends 3 and 4 being 3.756 and 3.887 ml.eq√ kg oil respectively. These obtained results indicated that these blends were suitable for different types of margarine processing and containing a considerable amount of antioxidants (Warner and Lazlo, 2005).

Also, results in the same table revealed that the values of TBA in all blends are not exceeded 3mg malonald/kg oil, this data could be resulted the blends were more stable in oils to word formation of secondary oxidation products (El-Gammal,2006). Iodine values for blends 3 and 4 were 108.532 and 108.330 gm/ 100gm oil which considered these blends are suitable in both margarine types processing. Melting point (MP) values in Table (6) were increased with the increment of palm oil and palm stearin content and this due to a high content of saturated fatty acids in these vegetable fats. These data were in agreement with those given by Widermann, (2008) who reported that these degrees of melting points were suitable for the fat phase used in fat blends prepared.

Results in Table (6) indicated that colour of blends 3 and 4 were 1.335 and 1.333 respectively. These results were in line with results for Chowdhury *et al*., (2007). From above mentioned data, it could be concluded that all fat blends are suitable for preparation of both margarine types and healthy in general where that all parameters in under safe limit and the melting point for all blends were below the human body temperature which can be digested easy.

Table 6. Chemical and physical properties of selected fat blends used in fat phase preparation:

Properties	Blend (3)	Blend (4)
Acid value (AV) (KOH / gm oil)	0.644	0.576
Free fatty acid (FFA%) (as oleic acid%)	0.498	0.395
Peroxide value (PV) (ml.eqv / kg oil)	3.756	3.887
Thiobarbituric acid value (TBA) (Mg mal. / kg oil)	0.410	0.403
Iodine value (gm /100gm oil)	108.532	108.330
Refractive index (RI) (at 40ºC)	1.041	1.027
Colour (at 440 mm)	1.335	1.333
Melting point (ºC)	44.1	41.8

Solid fat content (SFC) of fat blends:

The Solid Fat Content (SFC) is a measure (in percentage) of the amount of solid fat presented in samples at temperature (Zaliha *et al*., 2014).

Solid Fat Content is responsible for many product characteristics including general appearance, ease of spreading and oil exudation and also an important factor to select the suitable blends used in all fat products. It gives

also an indication of how fat performs at various temperature (Zeitoun, 1999).

Sahri and Dian, (2011) mentioned that quality of margarine for various categories of usage depends on their SFC, crystallizing and melting behavior.

Revealed results showed that addition of palm oil fractions namely (palm oil and palm stearin) >70% of the prepared blends increase the amount of SFC to be suitable for plastic fats processing.

Results in Table (7) showed that the value of SFC being 80.56 and 89.25% which presented in blends 3 and 4 which were suitable for pastry margarine processing which have a plastic fat properties and allow the dough to be folded several time (Wan *et al*.,2015).

Table 7. Solid fat content (SFC) of blends used in pastry margarine preparing

Vegetable Fat Blends Temperature	Blend (3)	Blend (4)
10ºC	80.56	89.25
20ºC	78.31	79.85
25ºC	70.71	70.15
30ºC	60.01	67.06
35ºC	55.27	59.17
40ºC	50.35	49.92

Blend (3) = SBO: SFO: PS Blend (4) = SBO: SFO: PO

Microstructure of fat blends used fat phase preparation:

Polymorphism determines the structural stability of fat phase, influenced by the properties of crystal lattice and the actual amount of fat in the structure of various crystal forms Marangoni, (2016). The studies in crystal size and transformation of polymorphic in fat phase of emulsions indicated that triacylglycerol have ability to crystallize in different polymorphic forms. Rousseau *et al*., (1996) reported that there were three forms of crystal morphology α the form of solid structure and changed quickly to β' by temperature and β the most stabile structure but less than surface area than β'. Differing from crystal to crystal in its melting point and crystal structure (Saadi *et al*., 2011). Crystal size properties are fundamental for final product and acceptability as small crystal lead to steady lipids products, whereas large crystals will produce sandy feel in mouth and for the food with high fat content, the strength of crystal determines the 60–80% hardness. (Zaliha *et al*., 2016). Figure 1 (c and d) shows the microscope images of fat phase of blends. It was indicated that the images of blends 3 and 4 showed different crystal morphology of blends 3 and 4 which contained (5% SBO:5% SFO: 90% PS) and (5% SBO: 5% SFO: 90%PO) at the same temperature, It was formed different shapes of crystals.

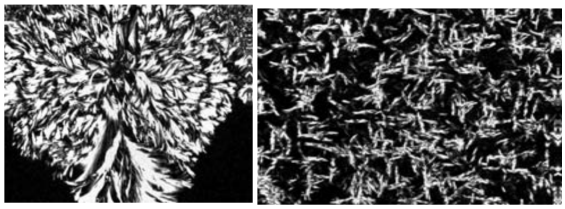

Fig.1.(c)(Blend3) **Fig. 1.(d)(Blend4)**

Fig. 1. c and d. Microscopic images of selected vegetable fats and oils blends (1, 2, 3 and 4) used in fat phase preparation

Characterization of Nano-emulsion particles:

a) Transmission Electron Microscopy (TEM) of water phase used in prepared table and pastry margarine by nano-emulsion:

Morphology and structure of nano-emulsion particles were studied using TEM. Combination of bright field imaging at increasing magnification and of diffraction modes was used to reveal the forms and sizes of nano-emulsion droplets (Savardekar and Bajaj, 2016 and Williams and Carter, 2009). Figure 2 (a and b) showed that regular water phase of emulsion size being 165.40: 220.41 nm while our obtained results from TEM indicated that particles size of water phase which processed by nano-emulsion technique were smaller than those of the origin type and ranged from 7.83 to 49.33 nm as presented in Figure 2 (a and b). Also, the particles in Figure 2 (a and b) seemed to be a smooth, spherical and tetragonal shapes which resulted to the stability and consistency of the fat products (margarine).

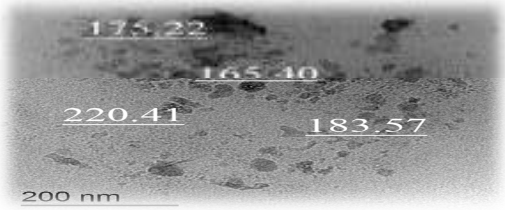

Fig. 2. a. Regular water phase

Fig. 2. b. Water phase by nanoemulsion particles

Fig. 2. a and b. TEM micrographs characterization illustrating the size and morphology of water phases used in prepared pastry margarine at 200nm and which by nanotechnology at 100nm

b) Zeta potential measurements of water phase used in prepared pastry margarine by nano-emulsion:

Zeta potential (ZP) is a key indicator of the stability of the solution (Horie and Fujita, 2011 and El-Gammal, 2016).Zeta potential (ZP) is a part of standard procedure frequently employed for thorough characterization of developed nano-emulsion systems (Isailović and Savić, 2017 and Khoshnevisan and Barkhi, 2015).

From presented data in Figure 3 (a) it could be summarized that the distribution the average size, density and the formation of aggregates of particles were more stable in water phase which prepared using nano-emulsion technique than those other particles prepared by usual aggregation in regular water phase as presented in Figure 3 (b).So, it could be summarized from results in Figure 2 (a and b) and Figure 3 (a and b) that prepared particles with nano-emulsion technique increase stability of particles size and this property is extremely important for fat products processing.

Changes of some chemical properties of prepared pastry margarine blends during cooling storage at 4±1ºC for 11 weeks:

One of the most important problem for oil preservation, is storage condition especially in long time storage, because the oils highly reasonable to oxidation and if storage condition it wasn't suitable very quickly oxidized and will be rancid Khalaf, (2016). pastry margarine was prepared and formulated using blends (3 and 4) by different techniques (regular and nano-emulsion) and some chemical properties namely (acid, peroxide, TBA and iodine values) were determined and all obtained results were illustrated in Tables (7, 8, 9and 10)

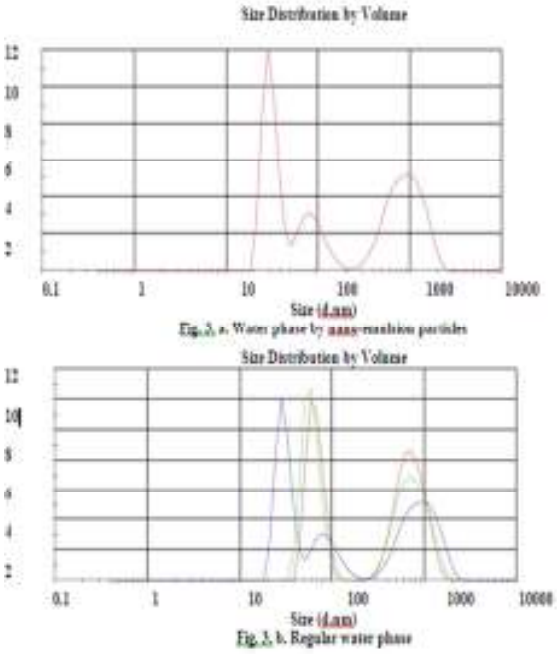

Fig. 3. a and b. Zeta potential measurements of water phases used in prepared pastry margarine which by regular method and nano technique

Changes of Acid value (A.V):

Results of Tables (8) illustrated that acid value of stored prepared pastry margarine blends by nano-emulsion to evaluate the stability during cold storage at (4±1ºC) for 11 weeks.

The results of acid value of prepared pastry margarine at the storage period were detected in Table (8) .Acid values of prepared pastry margarine blends (3 and 4) by nano-emulsion were (0.501and 0.510 mg KOH/kg oil) respectively in compared with those of (0.531 and 0.528 mg KOH/kg oil) for control prepared pastry margarine blends 1 and 2 respectively at zero time ,while commercial sample was 0.511 mg KOH / kg oil.

Data also in Table (8) showed there was slightly increasing in acid values prolonged storage period. For prepared pastry margarine blends for (3 and 4) by nano-emulsion, acid value reached to (6.888 and 0.698 mg KOH/kg oil), while control blends (3 and 4) reached to (0.980 and 0.982 mg KOH/kg oil) and the commercial sample was 0.851 mg KOH / kg oil after 11 weeks at 4±1ºC.

From results in Table (8) it was observed that using of nano-emulsion technique for preparing pastry margarine improve blends more firmer and more stabile to prolonged storage period. Also, all obtained values of AV were below (1%) and these values indicated that these prepared table and pastry margarine blends is safety for consumable as mentioned in (E.O.S., 2009).

Also, it was a remarkable that acid values of prepared pastry margarine by nano-emulsion technique were slightly increased in compare of values for both of commercial and control pastry margarine.

Table 8. Changes of Acid value of pastry margarine blends during storage at 4±1ºC

Storage period (weeks)	Commercial pastry margarine	Prepared pastry margarine blends (Control)		Prepared pastry margarine blends by nano-emulsion	
		Blend (3)	Blend (4)	Blend (3)	Blend (4)
Zero time	0.511	0.531	0.528	0.501	0.510
(1)	0.540	0.577	0.542	0.512	0.527
(2)	0.555	0.590	0.565	0.520	0.527
(3)	0.598	0.633	0.622	0.536	0.545
(4)	0.622	0.675	0.685	0.540	0.559
(5)	0.674	0.780	0.753	0.557	0.564
(6)	0.699	0.801	0.790	0.561	0.577
(7)	0.714	0.851	0.832	0.590	0.598
(8)	o.738	0.888	0.875	0.609	0.633
(9)	0.780	0.910	0.911	0.621	0.654
(10)	0.822	0.944	0.952	0.633	0.668
(11)	0.851	0.980	0.982	0.688	0.698

Blend (3) = SBO: SFO: PS Blend (4) = SBO: SFO: PO)

Oxidation and rancidity parameters:

Peroxide value (P.V):

Early stage of lipids oxidation could be expressed by peroxide value Ali and El-Anany(2012). Peroxide value used as a tool the determine the primary oxidation products namely hydro-peroxide Pereira *et al*., (2016). As mentioned before the smallest particle size can play a vital role in retarding oxidation steps. Results in Table (9) and Figure (5) confirmed that prepared pastry margarine by nano-emulsion in both of blends (3 and 4) were superior than the other prepared control blends.Results of PVs indicated that all blends with these small and dense particles were more stable and prevent oxidation steps up to 8 weeks under cold storage. Also, it was noted that the increment in both of blends (3 and 4) were slightly increased and reached to 3.198 and 3.290 ml. eqv/ Kg oil.

Table 9. Changes in peroxide value of prepared pastry margarine blends during storage at 4±1ºC for 11 weeks:

Storage period (Weeks)	Commercial pastry margarine	prepared pastry margarine blends (Control)		Prepared pastry margarine blends by nano-emulsion	
		Blend (3)	Blend (4)	Blend (3)	Blend (4)
Zero time	2.444	2.476	2.468	2.425	2.431
(1)	2.452	2.528	2.513	2.433	2.435
(2)	2.502	2.579	2.584	2.433	2.437
(3)	2.537	2.628	2.632	2.450	2.565
(4)	2.578	2.699	3.811	2.521	2.572
(5)	2.658	3.113	3.144	2.544	2.605
(6)	2.688	3.322	3.354	2.692	2.640
(7)	2.777	3.898	3.900	2.781	2.790
(8)	3.110	4.143	4.212	2.915	3.945
(9)	3.343	4.590	4.775	3.023	3.112
(10)	3.776	4.671	4.998	3.135	3.238
(11)	4.058	5.181	5.233	3.198	3.290

Blend (3) = SBO: SFO: PS Blend (4) = SBO: SFO: PO

These obtained results from previous tables may be due to that addition of palm oil and palm stearin which contained high amount of palmitic acid increase resistance of processed table margarine blends to oxidation steps. The results also showed that the stability of these blends which processed with nano-emulsion technology were also more stable than commercial sample.

Changes of Thiobarbituric acid value (TBA):

Thiobarbituric acid (TBA) test is considered the most common method of measurement of oxidative changes in food products and biological samples (Saafan, 2014). Determination of TBA value is more dependable than PV in examination of lipids deterioration as it is considered the secondary stages of oxidation or assemblage of secondary products (Ramadan *et al*.,2010). Results in Table (10) mentioned that TBA values were gradually increased in all prepared margarine and the commercial one under investigation. Values of TBA of control prepared table margarine were slightly increased during first three weeks of storage, while progress increasing was started after four weeks of storage. For pastry margarine, results in Table (10) mentioned that TBA values were gradually increased in all prepared margarine and the commercial one under investigation. Results in the same Table (10) showed that prepared pastry margarine blends by nano-emulsion were more stable than control blends and commercial sample over the period of storage. TBA values for prepared pastry margarine blends by nano-emulsion were 0.601 and 0.604 mg mal.kg oil for blends 3 and 4 respectively and reached to 0.821 and 0.820 mg malonald./ kg oil for blends 3 and 4 respectively after 11 weeks of storage while TBA values of control blends were 0.647 and 0.646 mg malonald./ kg oil for blends 3 and 4 respectively and reached to 0.971 and 0.977 mg malonald./ kg oil for blends 3 and 4 respectively, whilst TBA values of commercial sample was 0.635 mg malonald./ kg oil at zero time and reach to 0.898 mg malonald./ kg oil after 11 weeks of storage under the same circumstances. From data presented in Table (10) concerning PV and TBA values it could be recommended the prepared pastry margarine blends by nano-emulsion could be commercially exploited and exhibited good technique properties having a stable manner in the product during processing and storage period.

Table 10. Changes in thiobarbituric acid of prepared pastry margarine blends during storage at 4±1ºC for 11 weeks

Storage period (weeks)	Commercial pastry margarine	prepared pastry margarine blends (Control)		prepared pastry margarine blends by nano-emulsion	
		Blend (3)	Blend (4)	Blend (3)	Blend(4)
Zero time	0.635	0.647	0.646	0.601	0.604
(1)	0.643	0.666	0.668	0.601	0.610
(2)	0.677	0.678	0.680	0.631	0.611
(3)	0.695	0.690	0.692	0.644	0.627
(4)	0.711	0.738	0.743	0.644	0.640
(5)	0.750	0.764	0.765	0.661	0.663
(6)	0.772	0.798	0.802	0.677	0.680
(7)	0.790	0.824	0.840	0.710	0.712
(8)	0.793	0.870	0.883	0.761	0.754
(9)	0.793	0.898	0.977	0.784	0.777
(10)	0.857	0.925	0.939	0.807	0.804
(11)	0.898	0.971	0.977	0.821	0.820

Blend (3) = SBO: SFO: PS Blend (4) = SBO: SFO: PO

So, it could be concluded that blending of different vegetable fats namely PO and PS increasing of the stability of prepared margarine blends due to the increasing of saturated fatty acids and convert into stable product Basuny (2014). All obtained values of TBA were below (1mg malonald. /kg oil) and these data indicated that these prepared pastry margarine blends were safety for human consumption as stated by (E.O.S., 2009).

According to above presented data of AV, PV and TBA values of different prepared pastry margarine blends, it could be accomplished that prepared pastry margarine blends by nano-emulsion are more stable than commercial pastry margarine and control prepared pastry margarine in which these could be stored at 4ºC for 11 weeks. This mainly appropriate to the addition of palm oil and palm stearin which were considered had more established fatty acids compositions and the it's small particles size which had high stability of processing and storage.

Changes of Iodine value (IV):

Iodine value (IV) is a measurement of degree of unsaturation and is used to characterize fats and oils quality (Hazirah *et al.*, 2014).

For pastry margarine, data in Table (11) and Figure (6) revealed that IV of prepared pastry margarine by nano-emulsion were 104.350 and 104.342 gmI /100gm oil for blends (3 and 4) respectively at zero time and reached to 100.908 and 100.837 gmI /100gm oil for blends (3 and 4) respectively after 11 weeks of cooling storage while IV of commercial sample was 104.242 gmI /100gm oil and reached to 99.211 gm /100gm oil at the end of storage period under the same circumstances.

Also, data were detected in the same Table (11) observed that nano-emulsion technique more efficient and effective for preparing pastry margarine in compared with regular method and this is illustrated by comparing between results of control blends and prepared Pastry margarine blends by nano-emulsion .Iodine values of all pastry margarine blends were changed during storage and were in the recommended range which it wasn't less than 90 gmI /100gm oil (E.O.S., 2009). It could be concluded from data tabulated in Tables (10 and 11) that nano-emulsion technique can be used as advisable method to formulate new and stable blends for pastry margarine processing.

Table 11. Changes in Iodine value of pastry margarine blends during storage at 4±1ºC for 11 weeks

Storage period (weeks)	Commercial pastry margarine	Processed pastry margarine blends (Control)		Processed pastry margarine blends by nano-emulsion	
		Blend (3)	Blend (4)	Blend (3)	Blend(4)
Zero time	104.242	102.330	102.330	104.350	104.342
(1)	104.202	102.202	102.209	104.302	104.233
(2)	104.120	101.723	101.808	104.201	104.222
(3)	104.001	101.553	101.522	104.002	103.802
(4)	103.875	100.010	100.143	103.660	103.604
(5)	103.721	99.601	99.710	103.122	103.196
(6)	102.903	99.043	99.122	102.984	102.122
(7)	101.335	98.880	98.821	102.577	101.412
(8)	101.227	97.020	97.025	102.037	101.363
(9)	100.564	96.980	96.881	101.844	101.123
(10)	99.854	96.393	96.688	101.620	100.993
(11)	99.211	95.952	95.980	100.908	100.837

Blend (3) = SBO: SFO: PS Blend (4) = SBO: SFO: PO

Changes of physical properties of prepared pastry margarine blends during storage at 4±1ºC for 11 weeks:

Changes of Refractive index (RI)

Refractive index is physical criteria used to estimate the degree of unsaturation and identifies the molecules present in lipids (El-Gammal, 2007).

It could be observed from presented data in Table (12) that refractive index values were slightly decreased during storage for all prepared pastry margarine blends and commercial sample. From the same table the RI values of prepared pastry margarine blends attributed to substantially regular combination of saturated fatty acids from palm oil and palm stearin which used in processing of blends. Results in Table (12) showed that values of refractive index at the beginning of storage of prepared pastry margarine by nano-emulsion were 1.024 and 1.023 for blend (3 and 4) respectively and reached to 1.017 and 1.016 for blends (3 and 4) after 11 weeks of storage in compared with control prepared pastry margarine blends which being 1.021 and 1.020 for blends (3and 4) respectively and reached to.012 and 1.010 for blends (3 and 4) at the end of storage. From data in Table (12) it could be renowned that it is relevance between RI and IV which two parameters of fats and oils effect with amount of saturated and unsaturated fatty acids.

Table 12. Changes in refractive index of prepared pastry margarine blends during storage at 4±1ºC

Storage period (weeks)	Commercial pastry margarine	Prepared pastry margarine blends (Control)		Prepared pastry margarine blends by nano-emulsion	
		Blend (3)	Blend (4)	Blend(3)	Blend(4)
Zero time	1.023	1.021	1.020	1.024	1.023
(1)	1.022	1.020	1.019	1.024	1.023
(2)	1.020	1.019	1.017	1.024	1.023
(3)	1.019	1.018	1.017	1.023	1.023
(4)	1.019	1.018	1.016	1.023	1.022
(5)	1.019	1.017	1.016	1.023	1.022
(6)	1.017	1.016	1.016	1.022	1.022
(7)	1.017	1.016	1.014	1.022	1.020
(8)	1.015	1.015	1.014	1.018	1.017
(9)	1.015	1.015	1.012	1.017	1.017
(10)	1.015	1.014	1.010	1.017	1.017
(11)	1.014	1.012	1.010	1.017	1.016

Blend (3) = SBO: SFO: PS Blend (4) = SBO: SFO: PO

Changes of Colour:

Colour is one of the criteria which responsible for fat spreads stability during storage periods Janaki, (2013).Obtained results tabulated in Table (13) and Figure (8) showed that margarine blends prepared with nano-emulsion technique were more stable in colour prolonged storage periods under cooling conditions in compared with commercial sample and other prepared blends. Data in Table (11) revealed that limited changes in colour were observed in all prepared pastry margarine during storage period. Adding of satisfactory quantity of β- carotene gave a inconsistent fortification adjacent to light induced oxidation and to the low temperature of refrigerated storage.This may be due to small size of nano-emulsion particles, which increase the surface area of distribution of colour.Table (13) showed the results of colour index of pastry margarine. Results showed that prepared pastry margarine blends by nano-emulsion were more stable than control blends and commercial sample over the period of cooling storage which were 1.312 and 1.314 for blends 3 and 4 respectively for prepared pastry margarine by nano-emulsion at zero time and reached to 1.101 and 1.103 for blends 3 and 4 respectively for prepared pastry margarine by nano-emulsion after 11 weeks of cooling storage,while for control blends there were 1.311 and 1.300 for blends 3 and 4 respectively at zero time and reached to 1.014 and 1.015 for blends 3 and 4 respectively after 11 weeks of cooling storage, while for commercial sample there was 1.320 at zero time and reached to 1.019 at the end of cooling storage.

Table 13. Changes in color of pastry margarine blends during storage at 4±1ºC

Storage period (weeks)	Commercial pastry margarine	Prepared pastry margarine blends (Control)		Prepared pastry margarine blends by nano-emulsion	
		Blend(3)	Blend (4)	Blend(3)	Blend(4)
Zero time	1.320	1.311	1.300	1.312	1.314
(1)	1.320	1.300	1.300	1.308	1.313
(2)	1.320	1.243	1.265	1.308	1.310
(3)	1.272	1.221	1.222	1.308	1.310
(4)	1.270	1.221	1.201	1.300	1.305
(5)	1.270	1.195	1.198	1.300	1.290
(6)	1.251	1.133	1.198	1.262	1.264
(7)	1.210	1.111	1.133	1.262	1.264
(8)	1.110	1.031	1.101	1.126	1.128
(9)	1.110	1.030	1.044	1.121	1.120
(10)	1.102	1.022	1.023	1.119	1.112
(11)	1.019	1.014	1.015	1.101	1.103

Blend (3) = SBO: SFO: PS Blend (4) = SBO: SFO: PO

In conclusion, it could be recommended that pastry margarine blends should not have hydrogenated oils or using interesterification method and have got to be free trans fatty acids and which performed as suitable substitutes for saturated fatty acids.

Emulsion stability of fresh and stored prepared pastry margarine blends:

Lower phase %:

Data in Table (14) illustrated that the lower phase% increased in prepared pastry margarine by nano-emulsion more than those of control blends which increases the emulsion stability. Results in Table (14) mentioned that lower phase% for blends of prepared pastry margarine by nano-emulsion(0.7% for blend 3 and 0.8% for blend 4)were near to lower phase% of commercial sample which was 0.9%, while control blends were 0.5% for both blends 3 and 4. It could be due to using of the suitable emulsifiers (lecithin and mono di-glyceride) and small size and diffusion of water phase molecules.

Table 14. Lower phase % of fresh pastry margarine before storage

	Commercial pastry margarine	prepared pastry margarine blends (Control)		prepared pastry margarine blends by nano-emulsion	
		Blend (3)	Blend (4)	Blend (3)	Blend (4)
Lower phase %	0.9	0.5	0.5	0.7	0.8

Our obtained results are in accordance with Singh, (2015) who mentioned that the small droplet size gives nano-emulsion unique rheological and textural properties, which render them transparent and pleasant to the touch; both of these unique features can be desirable in preparing of margarine. These could be agreed with Jahniaval *et al.,*

(2003) Whose illustrated that the fat crystals were also dense and stick together and this make the emulsion more stable.

Separated layer %:

For emulsions of pastry margarine, results in Table (15) showed that emulsions in prepared pastry margarine by nano-emulsion in both blends and the commercial sample were more stable more than the emulsion of control prepared blends during storage period. Where separated layer % of the emulsions of commercial sample and prepared pastry margarine by nano-emulsion were 1.6% of both blends 3 and 4 while were 1.9 and 1.8 % for control blends 3 and 4 respectively after 11 weeks of cooling storage at 4±1°C.

Table 15. Separated layer % of pastry margarine blends during storage at 4±1°C

Storage period (weeks)	Commercial pastry margarine	prepared pastry margarine blends (Control)		Prepared pastry margarine blends by nano-emulsion	
		Blend (3)	Blend (4)	Blend (3)	Blend (4)
Zero time	1.1	1.2	1.2	1.1	1.1
(1)	1.1	1.2	1.2	1.1	1.1
(2)	1.1	1.2	1.2	1.1	1.1
(3)	1.1	1.3	1.3	1.2	1.2
(4)	1.2	1.3	1.4	1.2	1.2
(5)	1.3	1.4	1.4	1.2	1.2
(6)	1.3	1.4	1.4	1.3	1.3
(7)	1.4	1.5	1.4	1.3	1.3
(8)	1.5	1.6	1.6	1.5	1.4
(9)	1.6	1.6	1.6	1.5	1.6
(10)	1.6	1.7	1.8	1.5	1.6
(11)	1.6	1.9	1.8	1.6	1.6

Blend (3) = SBO: SFO: PS Blend (4) = SBO: SFO: PO

Creaming stability (creaming index):

Creaming index can be appears due to gravity separation. Upward movement of droplets on the fact that their density is less than the density of their environment is called creaming index (Peng *et al.,* 2010). Flocculation is result of the aggregating of two or more droplet at conditions that each droplet has maintained its entirety (Esmaeelian, 2016). The results in Table (16) revealed that creaming index was increased at a lower rate for blends (3 and 4) of prepared pastry margarine by nano- emulsion compared with control blends of both types of margarines. Using nano-emulsion technique in preparing pastry margarines protect the emulsions from oiling off and watering off and increased it's stability during storage period.

Table 16. Changes of creaming stability of the pastry margarine blends during storage at 4±1°C

Storage period (weeks)	Commercial pastry margarine	prepared pastry margarine blends (Control)		Prepared pastry margarine blends by nano-emulsion	
		Blend (3)	Blend (4)	Blend (3)	Blend (4)
Zero time	1.2	1.4	1.4	1.3	1.3
(1)	1.2	1.4	1.4	1.3	1.3
(2)	1.2	1.4	1.4	1.3	1.3
(3)	1.3	1.4	1.5	1.3	1.3
(4)	1.3	1.5	1.6	1.3	1.4
(5)	1.4	1.6	1.6	1.3	1.4
(6)	1.4	1.6	1.6	1.5	1.5
(7)	1.4	1.6	1.8	1.5	1.6
(8)	1.4	1.7	1.8	1.5	1.6
(9)	1.5	1.8	1.8	1.6	1.6
(10)	1.6	1.8	1.9	1.7	1.7
(11)	1.6	1.9	1.9	1.7	1.7

Blend (3) = SBO: SFO: PS Blend (4) = SBO: SFO: PO

It could be concluded that emulsion stability of prepared pastry margarine by nano-emulsion and with an addition of lecithin and monoglyceride can be maximized by using vegetable fats and oils (88%) this recommended blends have stable and homogenized emulsion.

Oxidative stability of fresh prepared table and pastry margarine with commercial margarine:

Rancimat index is a fast oxidation measurement of the degree of its resistance to oxidation; it indicates the expiry of the inherent anti-oxidative protection of the lipids and the potential of shelf life Dachtler *et al.,* (2013).

From obtained results in Table (17) it was showed that all blends of prepared pastry margarine blends by nano-emulsion have a long induction period .The results in the same Table (17) also revealed that blends of prepared pastry margarine by nano-emulsion had the same induction period (resistance hours) of commercial margarine (28hous at 110°C) which reflects that this blends were more stable toward oxidation. The effectiveness of nano-emulsion technique of preparing table and pastry margarine has been attributed mainly to its ability to remain the margarine stable at high temperature compared with regular method of preparing control margarine blends and the commercial margarine.

Table 17. Oxidation stability of fresh prepared table and pastry margarine compared with commercial and control margarine at 110°C

Margarine blends	Commercial pastry margarine	(Control)		Prepared pastry margarine by nano-emulsion	
		Blend (3)	Blend (4)	Blend (3)	Blend (4)
Induction period(hr)	28	19	19	28	28

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تحسين خصائص مارجرين المخبوزات المصنعة باستخدام تكنولوجيا النانو

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في السنوات الأخيرة تم تطوير المنتجات القائمة على تكنولوجيا النانو لاستخدامها في العديد من التطبيقات بما في ذلك الغذاء و تعتبر تكنولوجيا المستحلبات النانوية من أهم التطبيقات لما لها من تأثيرات إيجابية على الخواص الفيزيائية والكيميائية لمختلف المنتجات الدهنية. في هذه الدراسة تم تصنيع مارجرين المخبوزات باستخدام خليط زيت النخيل وإستيارين النخيل وزيت فول الصويا وزيت عباد الشمس بنسبة (٥ زيت فول الصويا: ٥٪ زيت عباد الشمس: ٩٠٪ زيت النخيل) و (٥٪ زيت فول الصويا: ٥٪ زيت عباد الشمس: ٩٠٪ ستيارين النخيل) .ثم استخدام تقنية النانو لتعديل خصائص الاستحلاب لمارجرين المخبوزات ودراسة تأثير ذلك على ثباتها خلال فترة التخزين حتى ١١ أسبوعًا تحت ظروف التبريد عند درجة حرارة (٤ درجة مئوية)وتم تحديد الخواص الكيميائية والفيزيائية لمستحلبات خلطات مارجرين المخبوزات التي تم تصنيعها بواسطة تقنية النانو ومقارنتها بعينات الكنترول . تم فحص جزيئات الطور المائي المستخدم في معالجة مارجرين المخبوزات بطريقة تقنية النانو باستخدام جهاز Transmission Electron Microscope و Zeta Potential Analyzer.أظهرت النتائج أن خلطات المارجرين المجهزة باستخدام هذه التقنية أدى إلى تحسن في خصائص الاستحلاب وزيادة ثباته أثناء التخزين في ظل ظروف التبريد .و كانت نتائج الإختبارات الكيميائية (رقم الحموضة و البيروكسيد) للخلطات المصنعة من (٥٪ زيت فول الصويا: ٥٪ زيت عباد الشمس: ٩٠٪ زيت النخيل) و (٥٪ زيت فول الصويا: ٥٪ زيت عباد الشمس: ٩٠٪ نخيل استيارين) ٠.٦٨٨ و ٠.٦٩٨ مجم أيدروكسيد بوتاسيوم/جم زيت و ٣.١٩٨ و ٣.٢٩٠مللمكافئ/كجم زيت في نهاية فترة التخزين(١١ أسبوعًا) مقارنة بعينات الكنترول التي كانت ٠.٩٨٠ و٠.٩٨٠ مجم أيدروكسيد بوتاسيوم/جم زيت و ٥.١٨١ و ٥.٢٣٣ مللمكافئ/كجم زيت . تم تقدير بعض الخواص الفيزيائية وهي اللون و معامل الانكسار وأشارت النتائج إلى أن جميع خلطات المارجرين التي تمت تصنيعها باستخدام تقنية المستحلبات النانونية كانت أكثر ثباتًا أثناء التخزين مقارنةً بالخلطات الأخري. وتشير الدراسة إلي أهمية استخدام تقنية المستحلبات النانونية لإطالة فترة الثبات والتخزين في مختلف المنتجات الدهنية مثل المارجرين