Extraction of antioxidant phenolic compounds from spent coffee ground by oil infusion method and production of scrub coffee oil glycerin soap bar

By
Mr. Vaivit Kositarat
ID 5614892

A special project submitted to the faculty of Biotechnology, Assumption University in part of fulfilment of the requirements for the degree of Bachelor of Science in Biotechnology
2016

Senior Project

Extraction of antioxidant phenolic compounds from spent coffee ground by oil infusion method and production of scrub coffee oil glycerin soap bar

By

Mr. Vaivit Kositarat

ID 5614892

A special project submitted to the faculty of Biotechnology, Assumption University in part of fulfilment of the requirements for the degree of Bachelor of Science in Biotechnology

Title	: Extraction of antioxidant phenolic compounds from spent coffee ground by oil infusion method and production of scrub coffee oil glycerin soap bar
By	: Vaivit Kositarat
Advisor	: A. Sireerat Laodheerasiri
Level of study	: Bachelor of Science
Department	: Agro Industry
Faculty	: Biotechnology
Academic year	: 2016

Advisor

(A. Sireerat Laodheerasiri)

ABSTRACT

The spent coffee grounds (SCG) contain large amount of phenolic compounds which are investigated as a potential source of antioxidant. Melanoidins is one of phenolic compounds formed during a roasting process of coffee bean and it is brown-colored compound. To increase the sustainability of SCG, *Coffea arabica* was extracted by oil infusion method. Five oils which were canola oil, corn oil, coconut oil, sunflower oil and mineral oil were used as the oil carrier for extracting antioxidant compounds from SCG, the 10%, 20% and 30% w/w of SCG were infused in each oil carriers at room temperature for 24, 48, 72, 96 and 120 hrs. The result showed that, the increasing of concentration of infusion ratio and the time period of infusion effected the increasing of the amount of total phenolic compounds (TPC) in all oil carriers. The TPC of 30% (w/w) SCG at 120 hrs of canola oil, corn oil, coconut oil, sunflower oil and mineral oil were 31.99±0.06, 35.54±0.87, 29.05±0.74, 30.37±0.5 and 22.04±0.72 mg/ml, respectively. The highest TPC was found in 30% w/w SCG infused in canola oil at 72 hrs. which presented 36.10 ± 0.93 mg/ml however 30% w/w SCG infused in corn oil at 120 hrs. showed the highest percentage increase of TPC comparing with carrier coconut oil (59.4 % ± 3.89). All five SCG infused oils contain scavenging activity to DPPH; canola oil, corn oil, coconut oil, sunflower oil and mineral oil contains 92.31%±1.1, 91.25%±1.9, 48.94%±0.4, 94.71%±0.3 and 10.51%±0.4, respectively. The 30% w/w infused in sunflower oil at 120 hrs. exhibited the highest antioxidant activity of 94.71% ± 0.3 and IC50 was 29.07 mg/ml whereas coconut oil showed the percentage increasing of antioxidant activity comparing with oil carrier was 265.23% ± 5.8. For the brown-colored development, the infusion ratio and period of infusion influenced the increasing of color in all oil carriers significantly. Peroxide value were increased when SCG was infused in canola oil, corn oil, and sunflower oil, the value were 16 meg/kg, 8 meq/kg and 36 meq/kg, respectively. however peroxide value was unchanged in coconut oil and no peroxide value found in mineral oil. For antioxidant soap bar with SCG scrubber made with 5 types of SCG infused oil and with oil carrier, samples were tested for preference test, 20 panelists graded liking score range from 1 to 9 followed by these attributes, color, fragrance, lather moistening, scrubbing, skin feel and over all liking. Data from the test was statistically proved by Duncan multiple range test. The result showed that infused mineral oil soap had highest moistening score as 6.75 ± 1.16.

Keywords: Spent coffee ground (SCG), Oil infusion, Melanoidins, Antioxidant activity, Soap bar

ACKNOWLEDGEMENT

I would like to express my special thanks of gratitude to my advisor; A Sireerat Laodheerasiri. Faculty of Biotechnology, Assumption University who helped me made this project happen and gave me great advices as well as broaden my knowledge. Including, providing room and raw materials.

Also, I would like to thank Dr.Siriwan Panprivech, Faculty of Biotechnology, Assumption University who guided me in part of total phenolic compounds and antioxidant activity.

And, I would like to thank Dr. –Ing. Tatsawan Tipvarakarnkoon, Faculty of Biotechnology, Assumption University; for guided me how to further develop this glycerin soap product.

Besides these, I would like to thank Miss Thipthida Kaewtathip, Institute of Food Research and Product Development (IFRPD) Kasetsart University who provided me Arabica spent coffee ground.

Finally, I would like to thank my family for supporting and encouraging me. Also thank to my friends for helping me and each other in all the semesters since we had been study together

TABLE OF CONTENT

LIST OF FIGURES

LIST OF FIGURES

LIST OF FIGURES

LIST OF FIGURES

LIST OF TABLE

CHAPTER I

INTRODUCTION

Coffee is the most popular beverage in the world with the second world trade community while the biggest is Petroleum. There are two main species of coffee that cultivate for consumption, *Coffea Arabica* (Arabica) and *Coffea canephora var. robusta* (Robusta). Because of Arabica its taste milder and more intense flavor, 70-70% of coffee's cultivation is Arabica.

The coffee after cultivation and preparation will get through roasting process then milling to form coffee powder use in brewing, after brewing process, a cup of espresso is made and the residue coffee is called "spent coffee ground" or SCG. Mostly it will be disposed as solid waste of fertilizer because it does not have commercial value. In some case, SCG are transformed into value added product by used as raw material of producing bioethanol or biofuel. Add referrence

The main component of phenol in green bean coffee is chlorogenic acid which is formed by esterification, consists of trans-cinnamic acids (caffeic, ferulic and *p*-coumaric acids) with hydroxyl groups on quinic acid. During coffee brewing process, chlorogenic acid is degraded by the heat and hydrolysis into smaller structure compounds such as caffeine and mostly are the soluble compounds. Besides this, the coffee beans during roasting process due to the high temperature applied in the process lead to transformation of chlorogenic acid into melanoidin which represents the brown colored compound, that come from Maillard reaction between reducing sugar and amino acid. Add refference Therefore, infusion oil process is used to extract the non-soluble melanoidin through the carrier oil and increase the total phenolic compound and antioxidant activity included with the change of color in infused oil.

In this study, the oil infusion method was used to extract nonsoluble compounds "melanoidin" by vary the type of oil carrier. The SCG-infused oil had potential to make antioxidant soap bar because of the phenolic compounds which isolated from spent coffee ground. Antioxidant compounds can protect skin from the sun light and helping the cell from the damag by free radical. Glycerin based soap was used for making soap bar due to its transparency. Besides this, SCG was applied in the soap bar as a scrubber in order to boost cleansing ability.

This project aims to study the effectiveness of oil infusion method to extract the phenolic compounds which contain antioxidant activity from SCG and to development the glycerin soap bar added with the SCG infused oil. Furthermore, the browning color and peroxide value were also determined to indicate the characteristic of SCG infused oil.

OBJECTIVES

1. To determine the amounts of total phenolic compounds antioxidant activity and color development in different types of oil carriers after oil infusion method.

2. To study the effect of infusion ratios and infusion times on the phenolic compounds, antioxidant activity, color and peroxide value development in spent coffee ground infused oil

3. To compare the attributes between different type of infused oil glycerin soaps

CHAPTER II

LITTERATURE REVIEW

1. Coffee

1.1 Robusta & Arabica

Coffee are cultivated in more than 50 countries, consist of Asia, Africa, America and Caribbean region. There are two spices of coffee those are the most cultivation in the world due to its unique taste and smell which are Arabica (*Coffea arabica*) 75% and Robusta (*Coffea canephora*) 25% of the world's cultivation.

When these two spices of coffees are made, their taste and aroma are different which Arabica has a much more intense flavor and complex aroma of those flowers, fruit, honey and chocolate. Robusta is more concentrated by its bitterness and is less aromatic. Although Arabica is more intense in aroma, Robusta contains almost twice amount of caffeine. The 100% Arabica is considered to be the best coffee in the world and much more superior than Arabica blend with Robusta (Jansen, 2006).

1.2 Spent coffee ground

After coffee beverages are made, a solid residue known as spent coffee grounds (SCG) is produced. Normally, SCG are disposed as a solid waste. It needs a proper management in order to get rid of SCG waste. Burning is not the good way to do, due to its bioactive compound which such as polyphenols, caffeine and tannins, can lead to production of greenhouse gas which directly bad effect to atmosphere. To avoid making the air pollution, there are many ways to make it more value, SCG can use as fertilizer for planting or raw material of biofuel production due to high organic compound in it. (Alessia Panusa, 2013)

1.3 Phenolic compound in coffee

Coffee contains large amount of phenolic compound. Phenolic compounds are found as a main composition of compound in plants, which has high in antioxidant activity to prevent or slow down oxidation reactions that destruct their cells. Mostly found in seeds and green leaves.

1.3.1 Chlorogenic acids (CGA)

Chlorogenic acid can be separated into two main groups which are mono-caffeoyl and di-caffeoyl acids. Robusta coffee contains more di-caffeoyl acids than Arabica coffee. Most of di-caffeoyl acids are hard to decompose in roasting process and remain barely unchanged in form that promote bitter taste in coffee and exhibit harsher taste profiles in Robusta, beside this the harsh taste present by di-caffeoyl acids in plant are effective protection of insects. Whereas, mono-caffeoyl acids are simply in structure and much more easier to decompose pass though roasting process

Chlorogenic acids formed by esterification consist of caffeoylquinic acids (CQA) with the main isomers (3-, 4- and 5-CQA), and related compounds such as caffeic acid, ferulic acid and p-coumaric acid; these compounds are the main of the phenolic fraction occurring in green coffee beans (Figure 1). (Alessia Punusa,2013) Chlorogenic acid content in green coffee bean was shown in Table 1.

1.3.2 Melanoidin

Polyphenols in coffee like Chlorogenic acids are changed in structure or composition pass though the roasting process that high in temperature promote Maillard reactions between reducing sugars and free amino group, some of phenolic compounds are deconstructed, meanwhile new phenolic compounds with antioxidant activity are formed. (www.coffeechemistry.com/news/health/antioxidants-in-coffee, 2010). The final products of Maillaard reactions are Melanoidins. Melanoidins are high molecular weight nitrogenous brown colored compounds formed during roasting process. Its structures are largely unknown due to its complexity. Melanoidins consist of soluble and non-soluble groups and account up to 25% of dried coffee beans (Figure 2). (Moreira AS, Nunes FM, Domingues MR, Coimbra MA. 2012.)

Table1 Chlorogenic acid content in green coffee bean, expressed in g%, dry matter basis (Adriana Farah, 2006,7/8/2017)

Samples	CQA	FQA	diCQA	Total CGA	References
C. arabica	5.76	0.25	0.87	6.88	Trugo & Macrae. 1984
C. arabica var Caturra	4.63	0.33	0.66	5.62	Clifford & Ramirez-Martinez .1991
C. arabica var. Bourbon	4.77	0.34	0.56	5.67	Clifford & Ramirez-Martinez .1991
Wild *C. arabica* (average)	3.26	0.19	0.60	4.10	Ky et al., 2001
C. arabica (Angola)	4.30	0.57	1.23	6.10	Correia et al., 1995
C. arabica (Angola)	4.84	0.28	0.53	5.65	Correia et al., 1995
C. arabica (Angola)	5.67	0.79	1.39	7.85	Correia et al., 1995
C. arabica var Boubon (Brazil)	4.2	0.28	0.77	5.25	Farah et al., 2005a
C. arabica cv. Longberry (Ethiopia)	4.6	0.29	0.84	5.73	Farah et al., 2005a
C. canephora cv Robusta	6.82	0.60	1.37	8.80	Trugo & Macrae .1984
C. canephora cv Robusta	5.33	0.79	1.05	7.17	Clifford & Ramirez-Martinez, 1991
C. canephora cv Robusta (Angola)	3.43	0.54	1.20	6.08	Correia et al., 1995
C. canephora cv Robusta (Angola)	4.97	0.75	1.46	7.18	Correia et al., 1995
C. canephora cv. *Conillon* (Brasil)	7.42	0.95	1.09	9.47	Farah et al., 2001
Wild *C. canephora (average)*	7.66	1.43	2.31	11.3	Ky et al., 2001
C. canephora var. Robusta (Uganda)	5.77	0.47	1.34	7.58	Farah et al., 2005a
Timor hybrid (*C.arabica x C. canephora*)	4.71	0.33	0.58	5.62	Clifford and Ramirez-Martinez, 1991
Catimor (Timor hybrid x *C. arabica*)	5.51	0.35	0.45	6.31	Clifford and Ramirez-Martinez, 1991
C. liberica cv. Dewevrei	5.39	0.48	1.1	6.97	Ky et al., 1977

CQA- caffeoylquinic acid; FQA – feruloyiquinic acid; diCQA dicaffeoylquinic acid. Total CGA – total chlorogenic acids. [a] Units may have been changed for consistency [a]

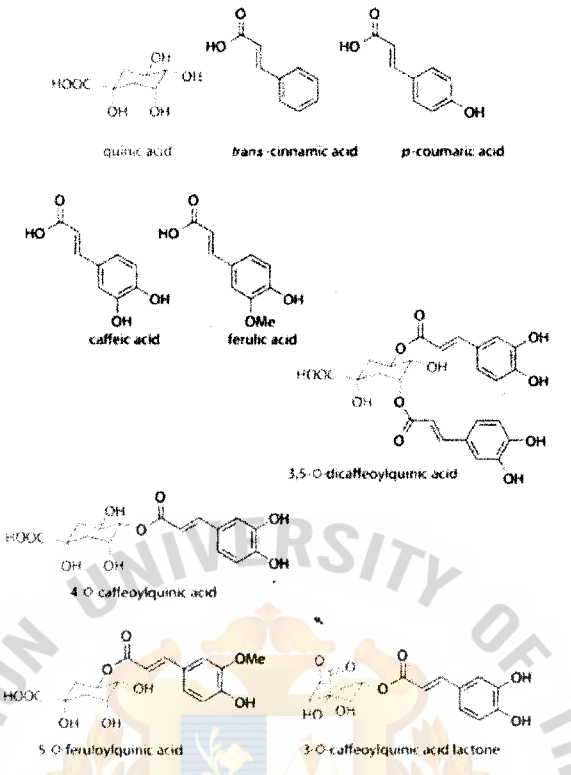


Figure 1 Phenolic compounds in coffee (www.denalihealthcaremi.com/80-chemicals-marijuana-coffee/. 7/8/2017)

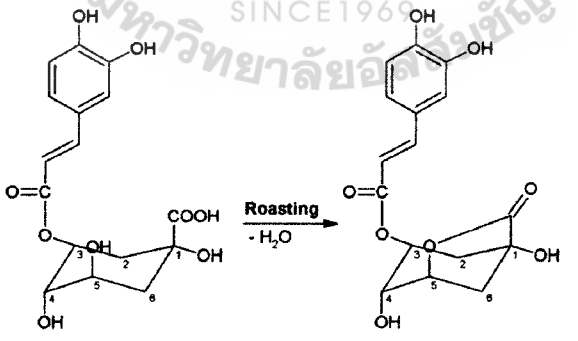


Figure 2 Melanoidins formed during roasting process of coffee bean.(Emma Eley, 2012. 1/8/2017)

2. Free radical and Antioxidant

Free radicals are unstable molecules that steal electrons from stable molecules, in order to stable their molecules lead to oxidation reactions which destruct the vegetative cells or occur in blood can induce acidity and cause of many diseases in result. Free radicals are normally produced by the body in order to aid the metabolic processes call as internal source and free radical also can promoted by external source such as smoking cigarettes, alcoholic beverages, pollutant in air. They are helpful in body functions but if too many, it is dangerous. (Figure 3)

Antioxidants could inhibit or slow down oxidation reactions by donate pair electron to free radical in order to make it stable instead of stealing from others molecule. (Figure 4) Antioxidants sources are often discussed in terms of their free radical scavenging abilities. (Susan Katchur, www.selfgrowth.com/articles/benefits-of-antioxidants-homemade-skin-care-and-more and Dr. Edward Group DC, NP, DACBN, DCBCN, DABFM, 2017. 1/8/2017)

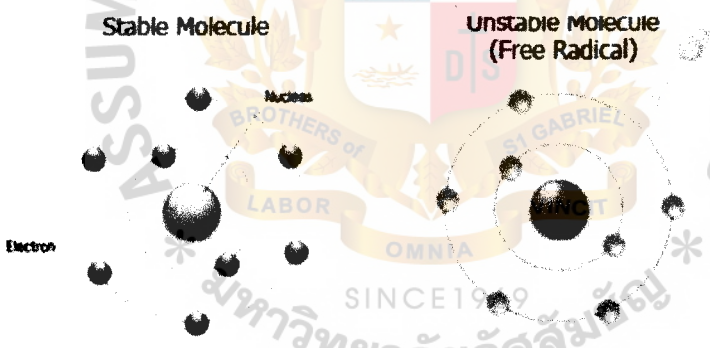


Figure 3 Stable and unstable molecule (free radical)
(Joan McDaniel, 2013 1/8/2017)

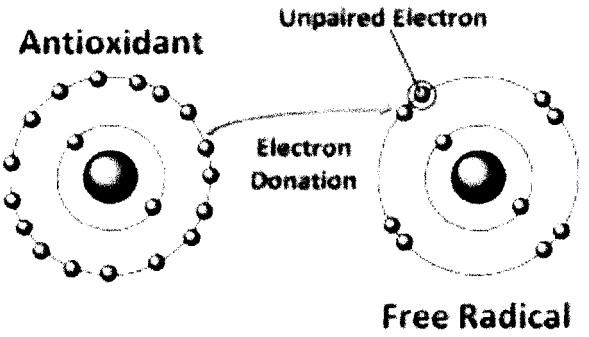


Figure 4 Antioxidant reacts with free radical
(bestofbothworldsaz.com/tag/free-radical-theory-of-aging/ 1/8/2017)

3. Oil infusion

Infusion is the method that extracting out of chemical compound or flavors in plant material pass though the solvent. Infused oil, also referred to as macerated oil, and consists of oil carrier that has been permeated "infused" or "macerated" with one or more herbs. Infused oils will contain the properties of essential compound in both used herbs and oil itself. In some case, oil infusion is not suitable for extracting some plants that are less in essential oils content.(www.aromaweb.com/articles/whatinfu.asp 1/8/2017) In spent coffee ground, due to its lipids and melanoidins content, oil infusion method is expected to carry out of phenolic compounds in coffee.

There are two methods to create infused oil, the cold method and the hot method. The cold method, which is done at room temperature, takes time but easy and requires little attention while the oil is soaking in the herb's qualities. The hot method requires a direct heat source and close attention, but takes shorter time than cold method. (Candace Hunter, 2008) From the study, oregano and rosemary were infused in olive oil by varying the period of infusion (24, 48 and 72 hours). Most of panelists judged the odor and flavor as medium strong to strong, however there was no significant different between infusion time. (Mayada Damechki, 2001, 7/8/2017)

3.1 Oil carrier

The various type of oil could be used as carrier oil for infusion and extraction the active compounds from herbs and plants.

3.1.1 Canola oil

Canola oil is light yellow and has a neutral taste of brassica plants. In general, canola seeds pressed either employing traditional cold-pressing methods or in large scale, by hexane extraction method. Color, taste, and odor of cold-pressed oil indeed more pronounced than that of refined oil. In addition, the content of saturated fatty acids of canola oil is the lowest among all common sources of vegetable oil. (http://www.nutrition-and-you.com/canola-oil.html, Fereidoon Shahidi and Udaya Wanasundara , 1994, 7/8/2017)

3.1.2 Corn oil

Corn oil is extracted from the germ of corn. These germs are rich in omega-6 and oils. There are various methods of extracting oil from the seed germs. The oil when extracted is dense and needs to be refined before it is used for cooking purposes. One can also use unrefined oil as it contains more of health boosting plant phytochemicals. (oilhealthbenefits.com/corn-oil/, 2017, 7/8/2017)

3.1.3 Coconut oil

Coconut oil is derived from the seeds of coconut palm, *Cocos nucifera*. Commercial coconut oil is made from copra or the dried kernel meat of coconut and goes through refining, bleaching and deodorizing processes. Coconut oil is considered a saturated fat because it contains more than 90% saturated fatty acids which is good for health. (A. M. Marina, Y. B. Che man, S. A. H. Nazimah & I. Amin, 2009, 7/8/2017)

3.1.4 Sunflower oil

Sunflower oil is produced from oil type sunflower seeds. There are two types of sunflower seeds – confection sunflower seeds and non-oil sunflower seeds. Confection sunflower seeds are edible and are used for the extraction of oil whereas non-oil sunflower seeds are used for feeding animals and are not suitable for human consumption. Sunflower oil is light in taste and appearance and supplies more Vitamin E than any other vegetable oil. It is a combination of monounsaturated and polyunsaturated fats with low saturated fat levels. (Saba 2017, www.sunflowernsa.com/oil, 7/8/2017)

3.1.5 Mineral oil

Mineral oil is clear and colorless, oily liquid that is a by-product of the distillation of petroleum. Mineral oil is used in medicine as a laxative and as an emollient. Mineral oil is completely indigestible and is not absorbed by the intestine. Its prolonged use may cause vitamin deficiencies. Mineral oil applied to the skin makes the latter softer and more pliable by retaining moisture within the epidermis. (The Editors of Encyclopædia Britannica, www.britannica.com/technology/mineral-oil, 7/8/2017)

4. Glycerin soap bar

Glycerin soaps have unique quality of moisturizing that is effective for all different kinds of the skins. It makes your skin moisturized and healthy. By these reasons, Glycerin soaps are considered to be one of the most moisturizing types of soap. Glycerin soaps promote moisture to your skin and hold it still. Beside this, glycerin soap keeps your skin feeling more hydrated than others types of soap that dried the skin, make it feel tight and even flaky. That why glycerin is a good skin humectant.

When the skins are moisturized, it is promote healthy skin by preventing of developing of wrinkles, stretch marks and tears in skins. Glycerin soap also suitable for using as facial washer more than harsh soap that dried skin and creating extra oil. Glycerin soap can help decrease or completely rid of acne problem.(RIPA AJMERA, 2017, 1/8/2017)

5. Lipid oxidation and peroxide value

Oxidation of lipid could be classified into positive and negative effect, in process of utilizing fatty acid to production of energy though β-oxidation.Oxidation is also involved in the production of signaling substances called eicosanoids. These are formed from the omega-3 fatty acid eicosapentaenoic acid (EPA) and the omega-6 fatty acid arachidonic acid (AA) by the action of specific enzyme systems. Besides this, oxidation also result of damaging the cells by free radicals are stealing electron in order to form a stable molecule and it also first step in the formation of several cytotoxic and mutagenic substances

Peroxide value is the measurement level of oxidation of a fat or oil containing polyunsaturated fatty acids, the cause of rancidity. A measure of hydroperoxides in oxidised oil. These are measured quantitatively on the basis of their ability to liberate iodine from acidic solutions of potassium iodide. This can be measured by titrating with sodium thiosulphate solution or electrochemically. .(Dr. Kristi Ekrann Aarak and Dr. Linda Saga, BioActive Foods, www.1life63.com/en/omega-in-your-body-oxidation-of-lipids/oxidation-of-lipids, 7/8/2017)

CHAPTER III

METHODOLOGY

Materials

1. **Samples**

Sample	**Source**
Oil	
Canola	TVO, Thailand
Corn	Golden drop, Thailand
Coconut	Neturel, Thailand
Sunflower	TVO, Thailand
Mineral	Chemipan, Thailand
Coffee	
Arabica SCG	Coffee shop in Kasetsart University

2. **Chemical substances**

Chemical substances	**Company**
Potassium iodide (KI MW = 166)	Ajax Finechem, New Zealand
Gallic acid monohydrate ($C_7H_6O_5$ MW = 170.12)	sigma-aldrich, China
2,2-Diphenyl-1-Picrylhydrazyl (DPPH) ($C_{18}H_{12}N_5O_6$ MW = 394.32)	Srichem, India
Ascorbic acid ($C_6H_8O_6$ MW=176.12)	Ajax Finechem, Australia
Sodium Thiosulphate ($Na_2S_2O_3$ MW = 158.11)	Ajax Finechem, Australia
Sodium carbonate (Na_2CO_3 MW = 105.98)	Ajax Finechem, New Zealand
Chloroform ($CHCl_3$ MW = 119.38)	RCI Labscan, Thailand
Acetone (C_3H_6O MW = 58.08)	RCI Labscan, Thailand
Acetic acid (CH_3COOH MW = 60.05)	RCI Labscan, Thailand
Ethyl acetate ($C_4H_8O_2$ MW = 88.11)	RCI Labscan, Thailand
Glycerin ($C_3H_8O_3$ MW = 92.09)	Chemipan, Thailand

3. **Other substances**

Substances	**Company**
Starch soluble	QRëC, New Zealand
Foiln&Ciocalteu's Phenol reagent	Srichem, India

4. **Equipments**

Equipments	**Company**
Balance	A&D Company Limited, Japan
Micropipette	Biohit, Germany
Vortex mixer	VELP, Italy
Spectophotometer	Milton Roy, US
Stirrer	VELP, Italy
Centrifugation machine	HERMLE LABORTECHNIK, Germany
Shaker	IKA LABORTECHNIK, Malaysia
Thermostat oven	Jebsen & Jessen (Thailand) Co., Ltd

5. **Miscellaneous**

Plasticwares	**Company**
Centrifuge tube (15 mL)	Quality Group Co., Ltd, Thailand
Pipette tip (100 μl, 1000 μl)	QSP, US

Glasswares	**Company**
Beaker	Pyrex, US
Funnel	Pyrex, US
Erlenmeyer Flask	Pyrex, US
Stirring rod	Pyrex, US

Methods

1. Preparation of spent coffee ground

The coffee ground passed from the brewing process which contain a lot of water content were collected and used further for drying process. SCG was collected from Art Coffee Shop at Kasertsart University; samples were collected 3 times and then blended together. The SCG was dried in hot oven at temperature 40 °C°C until the weight constant in order to prevent compound destruction. The dry spent coffee ground was kept in the aluminum bag at room temperature.

2. Spent coffee ground oil infusion

Essential compounds in SCG were extracted by oil infusion technique. Five types of oil were purchased from the Thailand grocery shop and applied for oil infusion (Canola, Corn, Coconut, Sunflower, and Mineral oil). The infusion ratio were prepared as 45 g : 5 g of oil : SCG (10% w/w), 40 g : 10 g of oil : SCG (20% w/w) and 35 g : 15 g of oil : SCG (30% w/w) in the nontransparent bottle. The SCG in each oil (total 50g) were extracted at room temperature for 24 hrs (day1), 48 hrs (day2), 72 hrs (day3), 96 hrs (day4) and 120 hrs (day5) and agitated one time per day. The SCG infused oils were collected by filtration method using multi-layer thin white cloth and kept in non-transparent container at room temperature for for chemical characteristic analysis

3. Sample preparation for chemical analysis

3.1 Sample preparation for total phenolic compounds assay

Weight 2.5g of infused oils sample and mixed with 15 ml of 70% acetone solvent (70:28:2, acetone/distilled water/acetic acid) then shakes 30 minutes in the dark place and centrifuged at 4000 rpm. for 10 minutes. After that, collected the watering part for analyzing by folin-ciocalteu's method. The control oil carrier was also prepared with the same procedure.

3.2 Sample preparation for DPPH scavenging Assay

Dissolved 1 g of SCG infused oil sample in 10 ml of ethyl acetate solvent, the concentration of prepared sample was 100 mg/ml, then diluted sample solution into concentration of 20, 40, 60 and 80mg/ml in ethyl acetate. These five concentrations were used to determine antioxidant activity and IC 50 by DPPH scavenging method. The oil carrier with have no SCG infused was used as a control to compare the increasing of antioxidant activity.

3.3 Sample preparation for peroxide value

Dissolved 5 g of SCG infused oil in 12 ml of chloroform and 18 ml of acetic acid.

4. Study of chemical characteristics

4.1 Total Phenolic compounds by Folin-Ciocalteu's method

Prepared fresh Folin-Ciocalteu's reagent by diluting with distilled water (folin-ciocalteu's reagent/distilled water, 1:5, v/v). One ml of sample and 2 ml of folin-ciocalteu's reagent were mixed in test tube and leave it for 3 min then 10 ml of 10% (w/v) of sodium carbonate (Na_2CO_3) was added and kept in the dark for 30 min. The absorbance was measure by spectrophotometer at 725 nm. The total phenolic compound was expressed as Gallic acid equivalent. Percentage total phenolic compound increased from carrier oil was calculated by this following formula.

$$\%TPC\ increased\ from\ carrier\ oil = \left(\frac{A_i - A_c}{A_c}\right) \times 100$$

$$A_c = Absorbance\ control\ (carrier\ oil carrier\ oil)$$

$$A_i = Absorbance\ infused\ oil$$

4.2 Anti-oxidation activity by DPPH scavenging method

The modified DPPH is scavenging technique that measured the anti-oxidant activity in the percentage reduction of DPPH. The 0.1 mM of DPPH in ethyl acetate were prepared in 100 ml Erlenmeyer flask, 0.0039 g of DPPH was dissolved in 10 ml of ethyl acetate (1mM DPPH) then topped up to 100ml with ethyl acetate (0.1mM DPPH).

Volume 1 ml of SCG infused oil prepared in ethyl acetate was mixed with 4 ml of 0.1 mM of DPPH solution in test tube, mix vigorously by vertex 10 second and the mixture was then leave in dark place for 30 minutes. The reaction was measured absorbance by spectrophotometer at 517nm. (1 ml of ethyl acetate + 4 ml of 0.1 mM of DPPH were used as control and ethyl acetate as a blank)

Percentage of antioxidant activity of each concentration of SCG infused oil samples were plotted in the graph versus with the concentration in order to examine IC50 value from linear equation, compared to ascorbic acid standard curve. Percentage of antioxidant activity and percentage of antioxidant activity increased from carrier oil were calculated by these formulas.

Note : 100 mg/ml SCG infused oil sample was used for calculating percentage of antioxidant activity increased

$$\%Inhibition = \left(\frac{A_b - A_i}{A_b}\right) \times 100$$

$$A_b = Absorbance\ blank$$

$$A_i = Absorbance\ infused\ oil$$

$$\%Inhibition\ increased\ from\ carrier\ oil = \left(\frac{A_i - A_c}{A_c}\right) \times 100$$

$$A_c = Absorbance\ control\ carrier\ oil$$

$$A_i = Absorbance\ infused\ oil$$

4.3 Color intensity

The SCG infused oil in different oil types were measured color intensity by spectrophotometry technique for determination the percentage of color intensity of SCG infused oils comparing with oil carrier (No SCG). All SCG infused oil samples and oil carrier samples were measured the absorbance at wavelength 420 nm. Percentage of color increased was calculated by this following formula.

$$\%color\ increased\ from\ carrier\ oil = \left(\frac{A_I - A_c}{A_c}\right) \times 100$$

$$A_c = Absorbance\ control$$

$$A_I = Absorbance\ infused\ oil$$

4.4 Peroxide Value

Peroxide value can be measured by using titration method which shown as concentration of peroxide value (meq/kg). One ml of saturated potassium iodide (KI) was added into prepared oils samples and flask was covered flask with aluminum foil and shakes for 5 min at room temperature. Then 30 ml of distilled water was added and following with the adding of 1 ml of 1% starch solution. The color change into dark brown or dark blue color was observed. After that the mixture was titrated with 0.1 N sodium thiosulphate ($Na_2O_3S_2$) until dark brown or dark blue color disappeared. The volume (ml) of used titrant (0.1N sodium thiosulphate) was recorded and used to calculate peroxide value by this following formula..

$$Peroxide\ value = \left[\frac{(V_1 - V_0) \times c}{m}\right] \times 1000$$

V_0 = consumption of 0.1N sodium thiosulphate at initial

V_1 = consumption of 0.1N sodium thiosulphate at the end point

C = molar concentration of sodium thiosulphate

m = weight oils in grams

5. Development of antioxidant glycerin soap bar

The soaps were made by using glycerin soap based, first layer was scrubbed soap which contained SCG as scrub beat and second layer was soap with SCG infused oil.

Glycerin was chopped into small pieces and melted in the double boiler hot water. The 30% w/w SCG infused oils at 120 hrs were used as ingredient to develop the glycerin soap bar as they contained the highest antioxidant activity value. Solid glycerin 50 g was boiled in double boiler hot water and0.2 g of SCG infused oil was added and mixed gently, after that 2g of dried SCG was added. The mixture was poured into a mold, leave until it cooling. For second layer, 50g of glycerin was melted with boiler and 0.2g of SCG infused oil was added and mixed gently then poured on top of first layer. Wait until the soaps were formed.

6. Preference test

20 panelists were assigned to test the soap with 3 steps

1. Observed the color (transparency) and smell soaps sample and score color and fragrance attributes.
2. Washed hand by using soaps sample, scrubbed soap part on hand until the lathers formed and scrubbed the coffee scrub side on back hand. Then score lather and scrubbing attributes.
3. Rubbed the hands dry and score skin feel and over all liking attributes

The data from preference test was statistically prove by Duncan multiple range test

7. Statistical analysis

The results of %increase in phenolic compound and antioxidant activity were statistically analyzed by using R-program v2-15-3 and the results of preference test were statistically analyzed by SAS program.

CHAPTER IV

RESULTS AND DISCUSSIONS

1. Total Phenolic Compounds in SCG infused oil

Five types of carrier oil (canola oil, corn oil, coconut oil, sunflower oil and mineral oil) were selected for study the effectiveness of the extraction of phenolic compounds from SCG by oil infusion method. Three concentration of SCG infusion in each oil carriers were prepared into 10% (w/w), 20% (w/w) and 30% (w/w) and extracted for 24, 48, 72, 96 and 120 hrs at room temperature. Determination of total phenolic compounds (TPC) in SCG infused oils were done by Folin-Ciocalteu's method and expressed as standard Gallic acid equivalent (mg/ml). The amount of TPC in the carrier oils, various concentration of SCG infused oils were shown in Table 2 and Table 3, respectively.

After oil infusion process, TPC in all oil carriers were increased according to time and concentration ratio of SCG. The highest amount of TPC of non-infused oil (pure oil) was found in canola oil 22.30±0.232 mg/ml, corn oil was 20.02±0.63 mg/ml, sunflower oil was 19.46±0.004 mg/ml,coconut oil was 19.23±0.003 mg/ml and there was no TPC found in mineral oil as shown in Table 2. Mostly, 10% (w/w) and 20% (w/w) infusion ratio at 24 hrs to 72 hrs could showed the development of TPC, however at 96 hrs to 120 hrs of all infusion ratio showed higher amount of TPC, especially for 30% (w/w). By this result, it explained that as more ratio and time of infusion applied, the more TPC absorbed in oil carrier as shown in Table 3.

The percentage development of TPC in all SCG infused oils were determined by comparing to non-infused oils, the result indicated that in all oil sample 30% (w/w) infusion ratio had highest percentage development followed by 20% (w/w) infusion ratio and lowest percentage development is 10% (w/w) infusion ratio.

Table 2 Total phenolic compounds of carrier oils (canola oil, corn oil, coconut oil, sunflower oil and mineral oil

Carrier oil Type	TPC (mg/ml)
Canola	22.30 ± 0.232
Corn	20.02 ± 0.63
Coconut	19.23 ± 0.003
Sunflower	19.46 ± 0.004
Mineral	18.44 ± 0.00

Table 3 Total phenolic compounds in the various concentration (10%, 20%, and 30% w/w) of SCG infusion in different oil carriers

Canola						
		24hours	48hours	72hours	96hours	120hours
10%	TPC mg/L	22.78±0.56	23.79±0.55	28±0.99	26.29±0.06	26.48±0.87
20%		24.35±0.28	24.84±1.24	28.15±1.35	25.85±0.93	30.63±1.12
30%		30.37±0.5	28.92±0.31	36.1±0.93	33.35±1.24	31.99±0.06
Corn						
		24hours	48hours	72hours	96hours	120hours
10%	TPC mg/L	20.72±0.37	20.63±0.53	21.57±0.35	22.87±1.18	22.65±1.12
20%		22.71±0.48	23.61±0.35	23.53±0.12	27.71±0.88	27.5±1.67
30%		25.5±1.05	26.64±0.19	29.71±0.31	30.89±1.36	35.53±0.87
Coconut						
		24hours	48hours	72hours	96hours	120hours
10%	TPC mg/L	21.2±1.18	21.27±0.45	22.87±0.56	21.33±0.27	23.5±1.08
20%		22.17±0.56	23.88±0.5	25.13±0.4	25.69±0.8	27.65±0.7
30%		26.16±1.01	28.04±0.93	30.57±0.88	28.75±1.43	29.05±0.74
Sunflower						
		24hours	48hours	72hours	96hours	120hours
10%	TPC mg/L	20.94±0.31	22.04±0.66	22.59±0.22	22.39±0.72	24.49±0.87
20%		21.6±0.37	22.69±0.19	25.54±0.25	23.53±0.4	26.27±0.18
30%		24.32±0.5	24.71±0.31	26.03±0.81	28.22±0.56	30.37±0.5
Mineral						
		24hours	48hours	72hours	96hours	120hours
10%	TPC mg/L	0±0	0±0	20.25±0.31	21.29±0.19	21.95±0.12
20%		0±0	0±0	20.72±0.4	21.63±0.66	20.84±0.37
30%		19.9±0.35	20.34±0.22	20.15±0.19	20.41±0.56	22.04±0.72

The percentage of TPC increasing after infusion the SCG in oil carriers were calculated, the results were shown in Table 4.

Table 4 The percentage of total phenolic compound increasing from carrier oils in all SCG infused oils

Canola						
		24hours	48hours	72hours	96hours	120hours
10%	% TPC increased	2.16±1.67	11.47±2.46	19.67±4.45	17.9±0.28	21.24±3.89
20%		9.44±1.39	20.09±5.56	29.18±6.05	15.93±4.17	37.37±5.01
30%		28.13±2.23	29.8±1.39	54.78±4.17	49.57±5.56	43.47±0.28
Corn						
		24hours	48hours	72hours	96hours	120hours
10%	% TPC increased	3.5±1.86	9.36±2.67	12.88±1.77	14.42±5.89	13.14±5.58
20%		15.75±2.41	19.26±1.75	20.31±0.62	36.26±4.39	31.37±8.33
30%		14.36±4.73	19.47±0.83	33.24±1.39	38.55±6.12	59.4±3.89
Coconut						
		24hours	48hours	72hours	96hours	120hours
10%	% TPC increased	13±2.26	14.39±2.34	19.64±2.9	10.95±0.65	25.32±2.26
20%		15.28±2.9	24.18±2.58	30.72±2.06	33.61±4.14	43.8±3.62
30%		36.04±5.26	45.84±4.84	59±4.59	49.5±7.42	51.1±3.87
Sunflower						
		24hours	48hours	72hours	96hours	120hours
10%	% TPC increased	7.59±1.59	13.22±3.4	16.08±1.13	15.02±3.69	25.84±4.46
20%		10.97±1.91	16.6±0.96	31.25±1.27	20.88±2.07	35.01±0.94
30%		24.94±2.55	26.97±1.59	33.73±4.14	45±2.87	56.04±2.55
Mineral						
		24hours	48hours	72hours	96hours	120hours
10%	% TPC increased	0±0	0±0	20.25±0.31	21.29±0.19	21.95±0.12
20%		0±0	0±0	20.72±0.4	21.63±0.66	20.84±0.37
30%		20.06±0.31	20.46±0.12	20.15±0.19	20.41±0.56	22.34±0.68

For canola oil (Figure 5 and Figure 6), at 72 hrs infusion period, TPC in 30% (w/w) infusion ratio had highest percentage development which was 54.78±4.17% (36.1±0.931mg/L) and TPC decreased at 96 hrs and 120 hrs to 49.57±5.56% (33.35±1.241 mg/ml) and 43.47±0.28% (31.990.061 mg/ml), compared to others infusion period. For 20% (w/w) infusion ratio, the percentage TPC increasing developed from 24 to 72 hrs and dropped down at 96 hrs to 15.93.±4.17 (25.85±0.93 mg/ml) which less than 10% 96 hrs 17.9.±0.28 (26.29±0.06 mg/ml) however, except 96 hrs infusion time, 20% infusion ratio had higher percentage total phenolic compound increase than 10% infusion ratio.

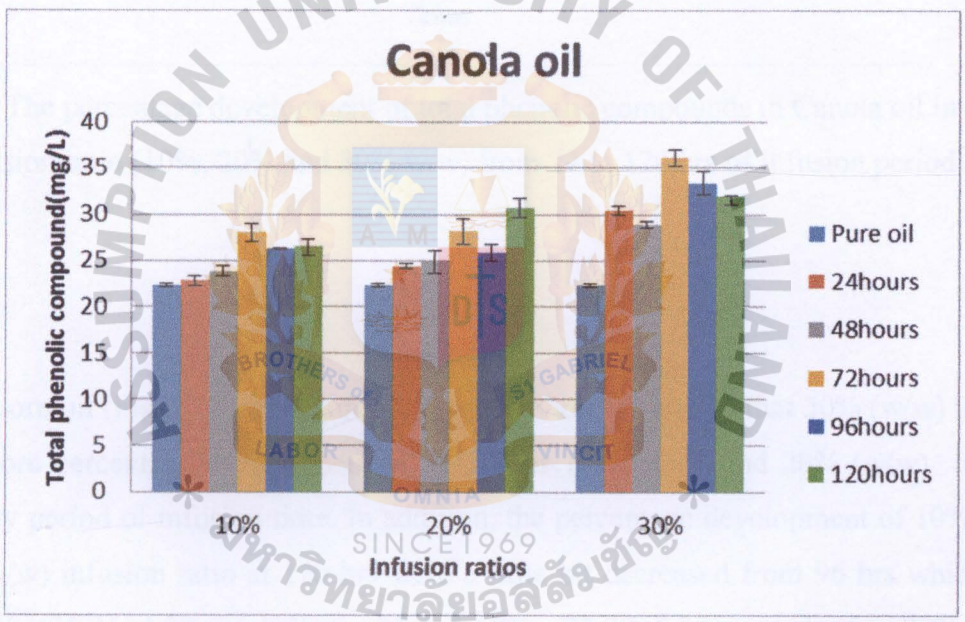


Figure 5 Total phenolic compounds in Canola oil compared between infusion ratio (10%, 20% and 30% w/w) and infusion period (24 to 120 hrs)

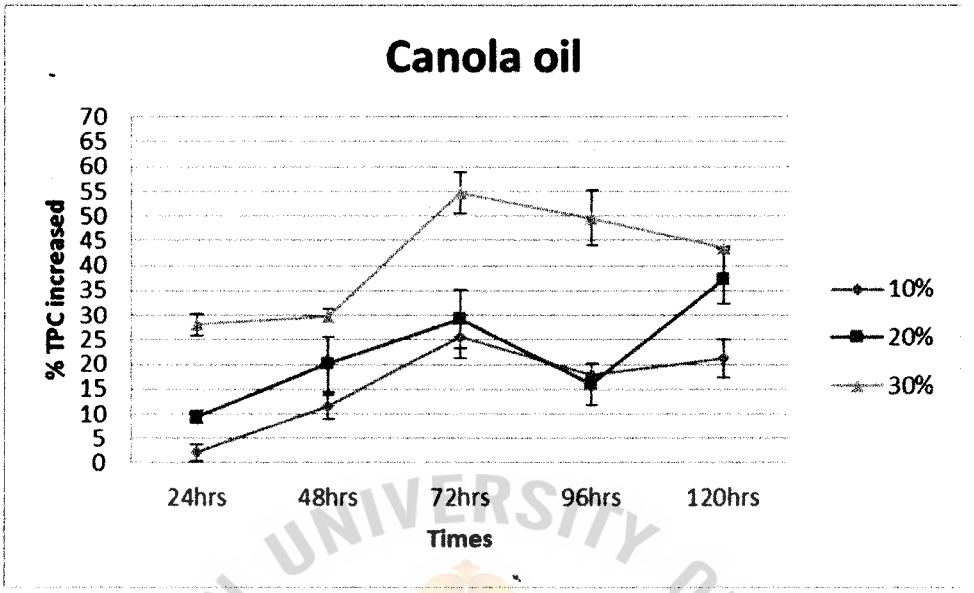


Figure 6 The percentage development of total phenolic compounds in Canola oil in each infusion ratio (10%, 20% and 30% w/w) from 24 to 120 hrs of infusion period

For corn oil (Figure 7 and Figure 8), it was obviously showed that 30% (w/w) infusion ratio had more percentage development of TPC over 10% (w/w) and 20% (w/w) infusion ratio in every period of infusion time. In addition, the percentage development of 10% (w/w) and 20% (w/w) infusion ratio at 120 hrs were a little bit decreased from 96 hrs which were 27.71±0.88% (36.26±4.39 mg/ml) to 27.5±1.67% (31.37±8.33 mg/ml) for 20% (w/w) infusion ratio and 14.42±5.89 (22.87±1.18 mg/ml) to 13.14±5.58% (22.65±1.12 mg/ml) for 10% (w/w) infusion ratio. Moreover, 10% (w/w) infusion ratio had poor TPC compared to others infusion ratio. However, the highest percentage development of total phenolic compound for infused corn oil is 59.4±3.89% (35.53±0.87 mg/ml) found in 30% (w/w) infusion ratio at 120 hrs according to table 4.

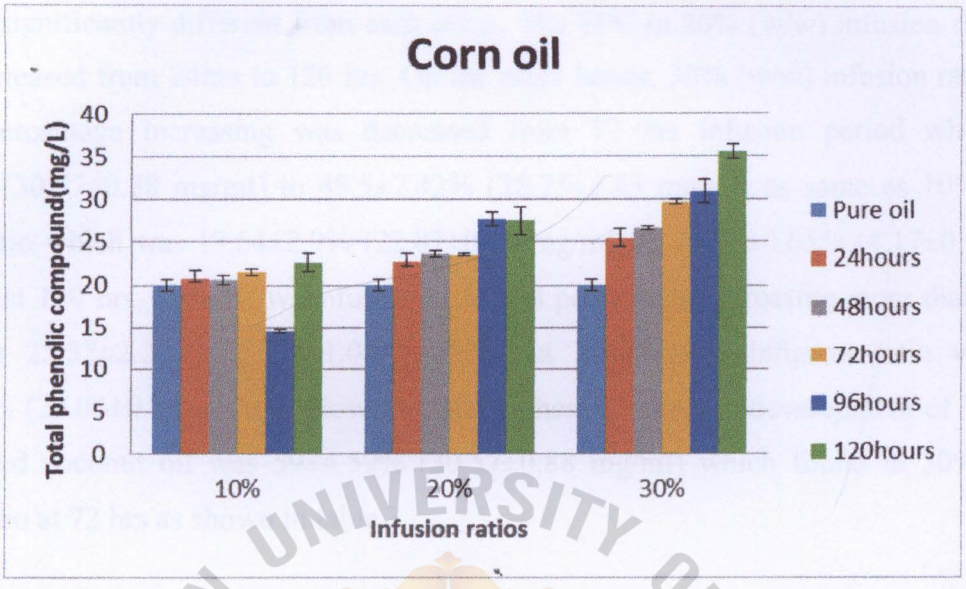


Figure 7 Total phenolic compounds in Corn oil compared between infusion ratio (10%, 20% and 30% w/w) and infusion period (24 to 120 hrs)

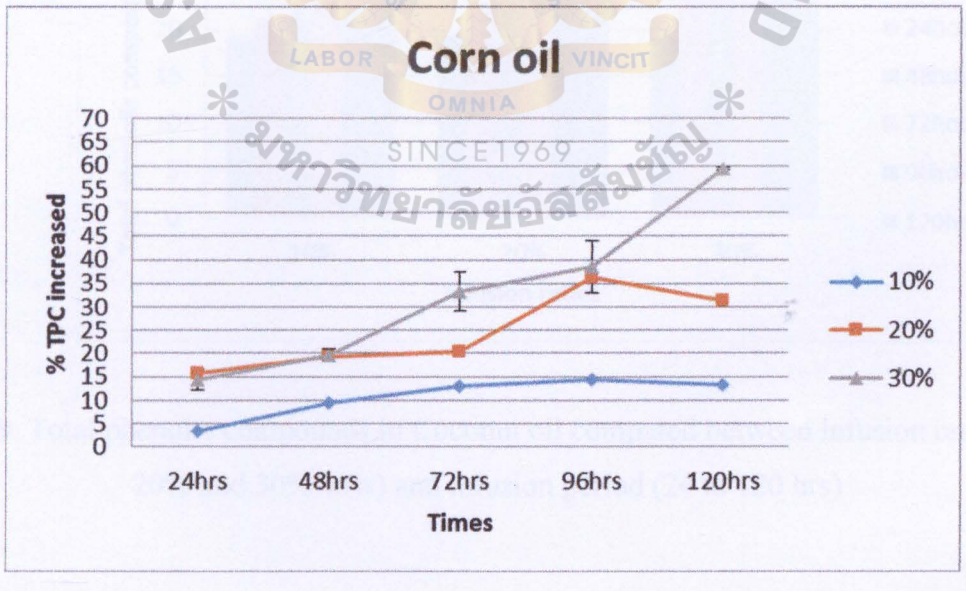


Figure 8 The percentage development of total phenolic compounds in Corn oil in each infusion ratio (10%, 20% and 30% w/w) from 24 to 120 hrs of infusion period

For coconut oil (Figure 9 and Figure 10), the development of TPC of all infusion ratio were significantly different from each other. The TPC in 20% (w/w) infusion ratio was slightly increased from 24hrs to 120 hrs. On the other hands, 30% (w/w) infusion ratio at 96 hrs, the percentage increasing was decreased from 72 hrs infusion period which was 59±4.59% (30.57±0.88 mg/ml) to 49.5±7.42% (28.75±1.43 mg/ml) as same as 10% (w/w) infusion ratio which was 19.64±2.9% (22.87±0.56 mg/ml) to 10.95±0.65% (4.17±0 mg/ml). Moreover, at 120 hrs, 10% (w/w) infusion ratio had percentage increasing more than 72 hrs which was 25.32±2.26 % (23.5±1.08 mg/ml) but 30% (w/w) infusion ratio was not, 51.1±3.87% (29.05±0.74 mg/ml). However, the highest percentage development of TPC for SCG infused coconut oil was 59±4.59% (30.57±0.88 mg/ml) which found in 30% (w/w) infusion ratio at 72 hrs as shown in table 3.

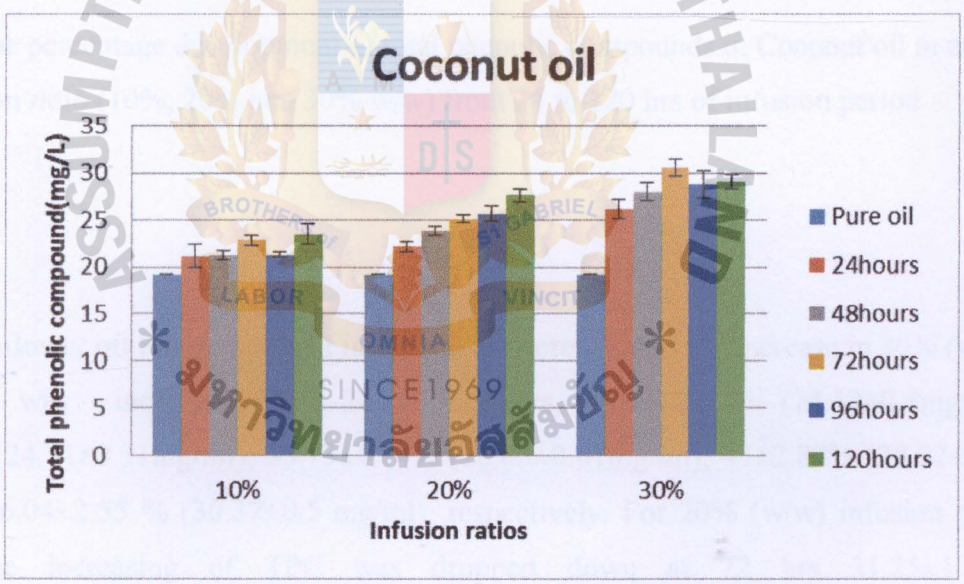


Figure 9 Total phenolic compounds in Coconut oil compared between infusion ratio (10%, 20% and 30% w/w) and infusion period (24 to 120 hrs)

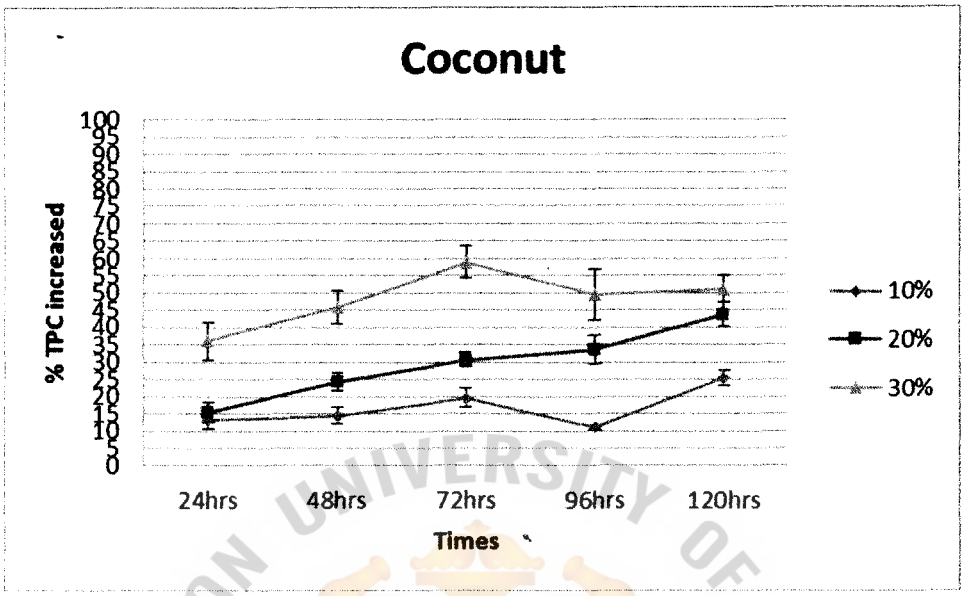


Figure 10 The percentage development of total phenolic compounds in Coconut oil in each infusion ratio (10%, 20% and 30% w/w) from 24 to 120 hrs of infusion period

For sunflower oil Figure 11 and Figure 12, the percentage TPC increase in 30% (w/w) infusion ratio was increased from 24 to 120 hrs; 24.94±2.55% (24.32±0.5mg/ml), 26.97±1.59% (24.71±0.31mg/ml), 33.73±4.14% (26.03±0.81mg/ml), 45±2.87% (28.22±0.56 mg/ml). and 56.04±2.55 % (30.37±0.5 mg/ml), respectively. For 20% (w/w) infusion ratio, the percentage increasing of TPC was dropped down at 72 hrs 31.25±1.27% (25.54±0.25mg/ml) to 20.88±2.07% (23.53±0.4 mg/ml) at 96 hrs and increased to 35.01±0.94% (26.27±0.18mg/ml) at 120 hrs. The 10% (w/w) showed the lowest percentage increasing of TPC compared to others infusion ratio in every period of infusion time. Lastly, the highest percentage development of total phenolic compound for infused sunflower oil was 56.04±2.55 % (30.37±0.5 mg/ml) which found in 30% (w/w) infusion ratio at 120 hrs as shown in table 4.

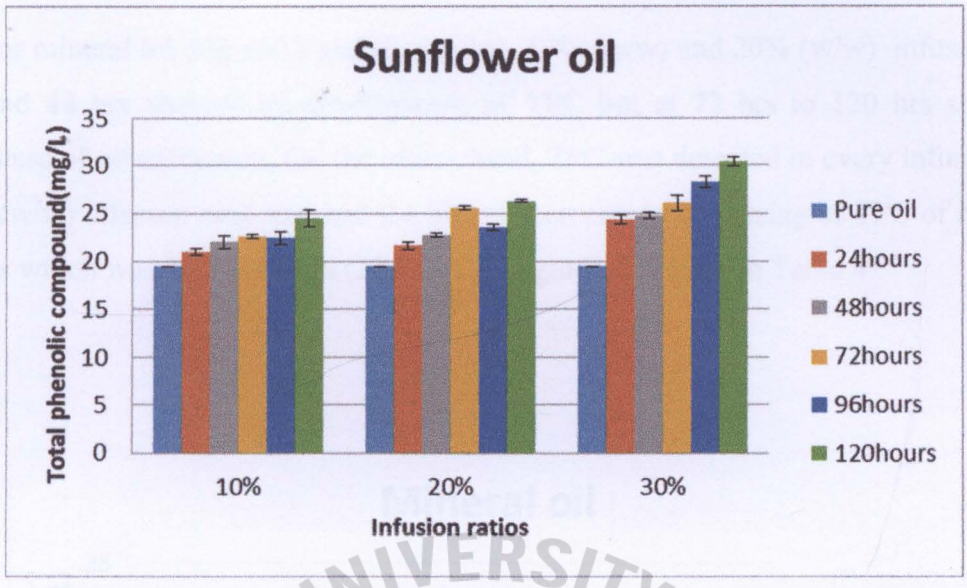


Figure 11 Total phenolic compounds in Sunflower oil compared between infusion ratio (10%, 20% and 30% w/w) and infusion period (24 to 120 hrs)

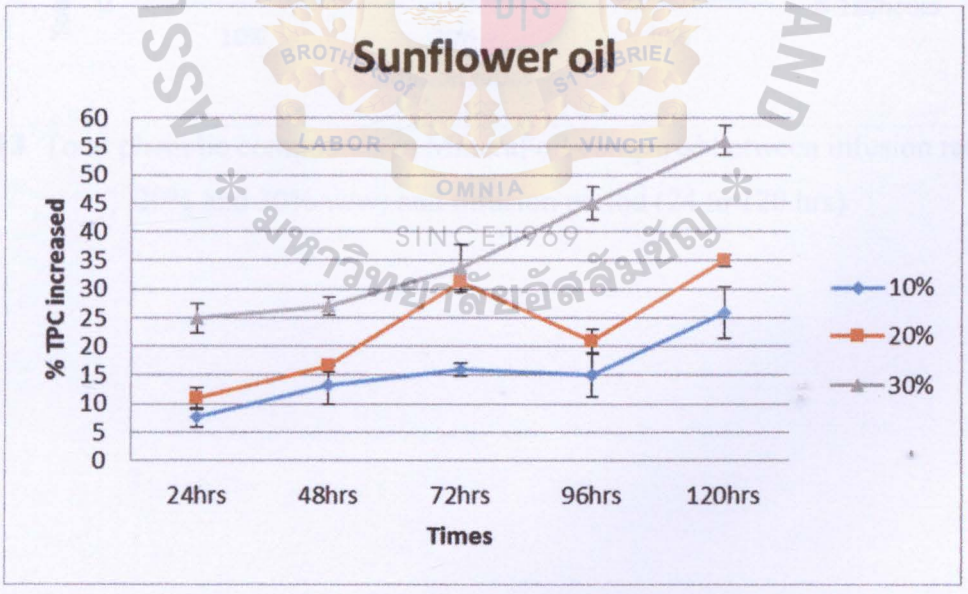


Figure 12 The percentage development of total phenolic compounds in Sunflower oil in each infusion ratio (10%, 20% and 30% w/w) from 24 to 120 hrs of infusion period

For mineral oil (Figure13 and Figure 14), 10% (w/w) and 20% (w/w) infusion ratio at 24 hrs and 48 hrs showed no development of TPC but at 72 hrs to 120 hrs showed the effectiveness of oil extraction. On the others hand, TPC was detected in every infusion period for 30% (w/w) infusion ratio and had the highest percentage increasing of TPC of mineral oil at 120 hrs which was 22.34±0.68% (22.04±0.72 mg/ml) as shown in Table 4.

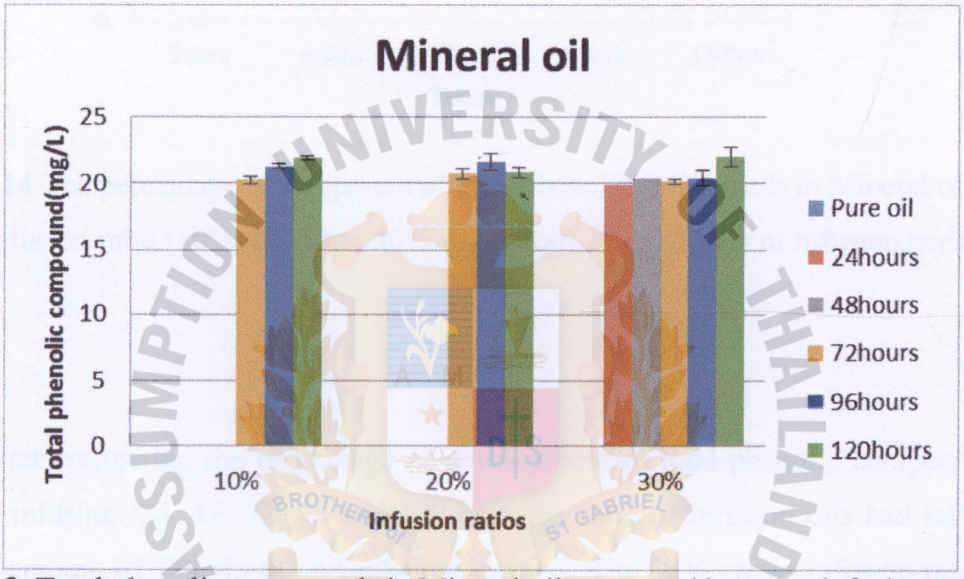


Figure 13 Total phenolic compounds in Mineral oil compared between infusion ratio (10%, 20% and 30% w/w) and infusion period (24 to 120 hrs)

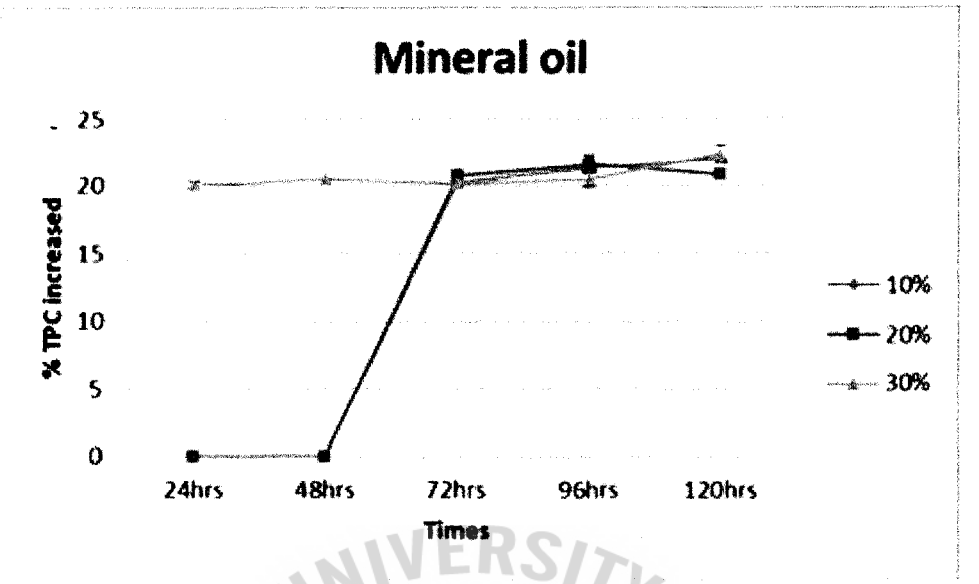


Figure 14 The percentage development of total phenolic compounds in Mineral oil in each infusion ratio (10%, 20% and 30% w/w) from 24 to 120 hrs of infusion period

After comparing the percentage of development of total phenolic compound in each period of infusion (24, 48, 72, 96 and 120 hrs), the ratio of infusion oils had influence the extracted amount of TPC in the oil infusion process. The 30% (w/w) infusion ratio showed the highest percentage of TPC development, followed by 20% (w/w) infusion ratio and the lowest was 10% (w/w) infusion ratio. The highest percentage development of TPC was found in SCG infused corn oil at 30% (w/w) infusion ratio,120 hrs

The comparison between 30% (w/w) infusion ratio of all carrier oils samples, as 30% (w/w) infusion ratio had more percentage development of phenolic compound than others ratio, so this ratio was selected to compare between each type of oil carriers.

In Figure 15, coconut carrier oil showed highest percentage development of TPC in 24, 48 and 72 hrs infusion times, canola carrier oil showed the highest percentage development TPC at 96 hrs, corn carrier oil showed the highest percentage development TPC at 120 hrs. Every carrier oils had potential to extract TPC from SCG though infusion process. In addition, the phenolic compound in initial oils carrier had influent to amount of extracted TPC after infusion process which the TPC could be absorbed well if less TPC contented in initial oil carrier like mineral oil, corn oil and sunflower oil.

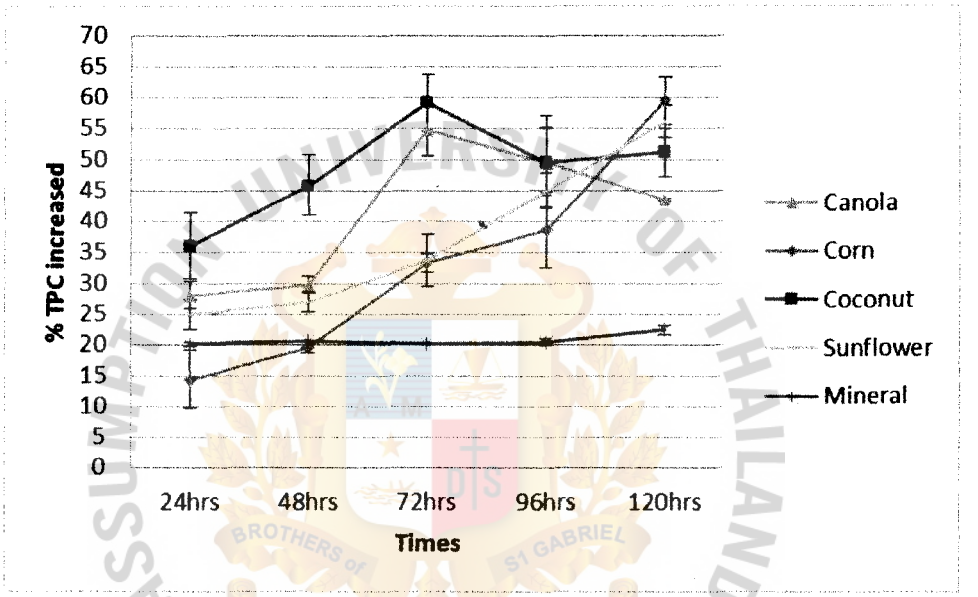


Figure 15 The percentage increasing of total phenolic compound compared with oil carrier from 24 to 120 hrs, in 30% (w/w) infusion ratio

From the previous study of extraction phenolic compound antioxidant by oil infusion, Rosemary and Oregano were used as herbal materials infused in olive oil. Total polar phenol content in oregano flavoured oils increased about 3.5 times in comparison with that of the control. For the rosemary flavoured oil an increase of 1.7 times was observed. Besides this peroxide value changes were higher for Oregano flavoured oil compared with Rosemary, both were higher than the control sample. In addition, there were development of pigment from Pheophytin α, β-Carotene and Lutein, oregano was higher than Rosemary. (Mayada Damechki, 2001).

2. Anti-oxidation activity by DPPH scavenging method

The antioxidant activity of SCG infused oil was measured by DPPH scavenging assay. Table 5 indicated the antioxidant activity of all oil carriers before adding SCG in the oil infusion process, the highest antioxidant activity was found in sunflower oil (90.5%) and the antioxidant activity in the other oil carriers were canola oil (89.7%), corn oil (74.9), coconut oil (13.4%) and the lowest was mineral oil (4.3%).

The antioxidant activity of SCG infused in the various types of oil carriers were also determined as shown in Table 6. Furthermore, the percentage of antioxidant activity increased from oil carrier was also studies and showed in Table 7.

Table 5 The percentage antioxidant activity of oil carriers by DPPH scavenging

Type of Oil carriers	% antioxidant activity
canola	89.7±0.77
corn	74.9±1.44
coconut	13.4±1.25
sunflower	90.5±0.87
mineral	4.3±1.07

Table 6 The percentage of antioxidant activity of SCG infused in the various type oil carriers

Canola						
	% antioxidant activity	24hours	48hours	72hours	96hours	120hours
10%		85.6±0.9	88.35±0.8	87.92±0.7	85.65±1.1	84.63±1.1
20%		86.7±1.4	89.25±1.1	88.16±0.5	87.14±0.8	86.2±1
30%		88.2±0.8	91.02±1.2	90.2±1	90.63±0.8	92.31±1.1
Corn						
	% antioxidant activity	24hours	48hours	72hours	96hours	120hours
10%		67.14±1.2	74±1.8	75.37±0.9	79.84±1.3	81.02±1
20%		70.08±1.1	79.06±1.3	80.78±0.7	87.33±0.8	89.76±1.6
30%		74.78±2	80.98±2.1	82.75±1.6	88±1	91.25±1.9
Coconut						
	% antioxidant activity	24hours	48hours	72hours	96hours	120hours
10%		18.27±1	20.35±0.9	23.73±0.9	24.67±1.1	28.82±0.7
20%		17.69±1.7	22.51±0.8	26.51±0.6	29.06±1.6	35.96±0.8
30%		18.94±0.7	24.24±0.6	33.18±0.6	37.57±0.5	48.94±0.4
Sunflower						
	% antioxidant activity	24hours	48hours	72hours	96hours	120hours
10%		81.33±0.4	87.33±0.5	88.71±1	88.35±0.6	93.8±0.3
20%		82.67±0.6	90.67±0.6	89.92±0.4	89.45±0.4	94.35±0.4
30%		85.29±0.4	90.94±0.5	90.9±0.7	91.57±1	94.71±0.3
Mineral						
	% antioxidant activity	24hours	48hours	72hours	96hours	120hours
10%		2.86±2	5.96±4.4	8.47±5.7	7.18±5.2	8.82±6
20%		3.57±3.6	6.59±6.6	9.96±0.7	9.69±0.3	10±1.2
30%		4.31±0.6	8.35±1.3	9.06±0.5	9.76±1.5	10.51±0.4

Table 7 The increasing of percentage antioxidant activity of SCG infused in the various type of oil carriers compared with oil carrier

Canola						
	% Increase of antioxidant activity	24hours	48hours	72hours	96hours	120hours
10%		0	0	0	0	0
20%		0	0	0	0	0
30%		0	0.92±0.3	0.79±0.8	1.03±0.9	2.91±1.2
Corn						
	% Increase of antioxidant activity	24hours	48hours	72hours	96hours	120hours
10%		0	0.49±0.8	0.77±1.1	6.6±1.8	8.17±1.3
20%		0	5.55±1.7	7.86±0.9	16.6±1	19.85±2.1
30%		0.87±2	8.12±2.1	10.47±1.6	17.49±1	21.84±1.9
Coconut						
	% Increase of antioxidant activity	24hours	48hours	72hours	96hours	120hours
10%		36.38±7.5	51.89±6.9	77.06±6.6	84.08±8.5	26.72±5.4
20%		31.99±12.6	67.98±5.6	97.83±4.8	116.86±11.9	168.36±5.7
30%		41.35±3.5	80.86±5.8	147.59±12.2	180.36±9.7	265.23±5.8
Sunflower						
	% Increase of antioxidant activity	24hours	48hours	72hours	96hours	120hours
10%		0	0	0	0	3.65±0.3
20%		0	0	0	0	4.26±0.4
30%		0	0.49±0.5	0.58±0.6	1.18±1.2	4.65±4.6
Mineral						
	% Increase of antioxidant activity	24hours	48hours	72hours	96hours	120hours
10%		0	38.62±11.1	96.99±17.9	66.89±8.2	105.2±8.2
20%		0	53.21±27.4	131.65±16.7	125.26±6.9	132.56±27.8
30%		14.91±0	94.25±29.6	110.67±12.5	127.09±34.3	144.41±10.4

For canola oil as shown in figure 16, the antioxidant activity in oil carrier was higher than 20% (w/w) and 10% (w/w) infusion ratio in every period of infusion time and also higher than 30% (w/w) infusion ratio at 24 hrs. At 48 to 120 hrs, it was obviously found that the antioxidant activity of 30% (w/w) infusion ratio was more than oil carrier. The highest percentage of antioxidant activity of SCG infused oil increased from oil carrier was observed in 30% (w/w) at 120 hrs that indicated 92.31%±1.1 increasing shown in Table 7 and Figure 17. According to Figure 5, the TPC in 20% (w/w) and 10% (w/w) infusion ratio were higher than carrier oil which is unrelated with lesser antioxidant activities performed by DPPH method. By these results, the 20% (w/w) and 10% (w/w) infusion ratio of oils and spent coffee ground was not promoted antioxidant activity. However, 30% (w/w) infusion ratio showed the increasing of antioxidant activity started from 48 hrs to 120 hrs. In addition, the antioxidant activity of SCG infused canola oil in each period of infusion time were increased significantly by more infusion ratio applied. In Figure 17, the increasing of percentage antioxidant activity was only found in 30% infusion ratio from 48 to 120 hrs which had the highest value of 2.91±1.2% at 120hours infusion ratio.

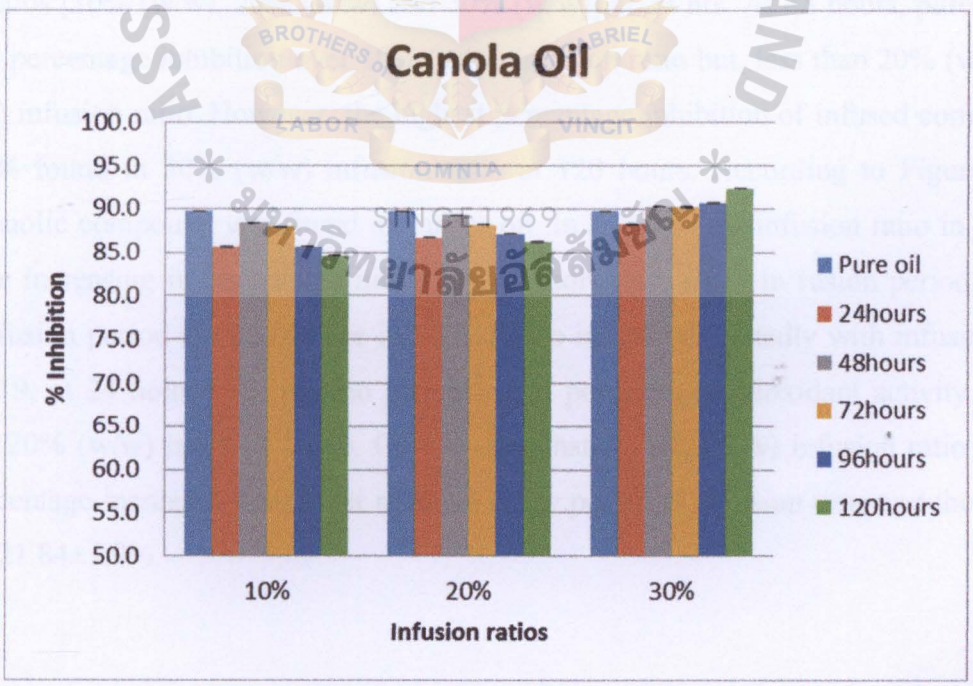


Figure 16 The percentage inhibition of DPPH scavenging assay in canola oil compare between infusion ratio (10%, 20% and 30% w/w) and infusion period (24, 48, 72, 96 and 120 hours)

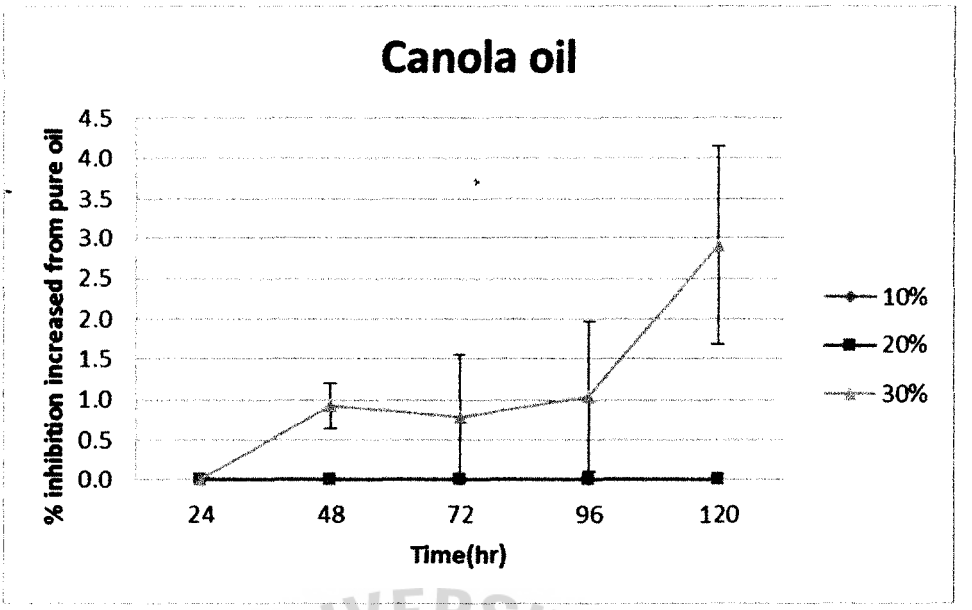


Figure 17 The percentage inhibition increased from carrier oil in canola oil in each infused ratio (10%, 20% and 30%) from 24 houres to 120 hours infusion period

For corn oil (Figure 18), corn oil carrier had more antioxidant activity than all SCG infusion ratios [10% (w/w), 20% (w/w) and 30% (w/w)] at 24 hrs. At 48 hours, pure oil also had higher percentage inhibition over 10% (w/w) infusion ratio but, less than 20% (w/w) and 30% (w/w) infusion ratio. However, the highest percentage inhibition of infused corn oil was 91.25±1.9% found in 30% (w/w) infusion ratio at 120 hours. According to Figure 7 The lowest phenolic compound was found in carrier oil. In addition, all infusion ratio in corn oil showed the increasing in percentage inhibition relatedly with more in fusion period applied and all in fusion period the percentage inhibition also increased relatedly with infusion ratio. In Figure 19, at 24 hour there was no increasing in percentage antioxidant activity in 10% (w/w) and 20% (w/w) infusion ratios. On the other hand, 30% (w/w) infusion ratio showed higher percentage increased over other ratios in every period of infusion time and the highest value was 21.84±1.9%.

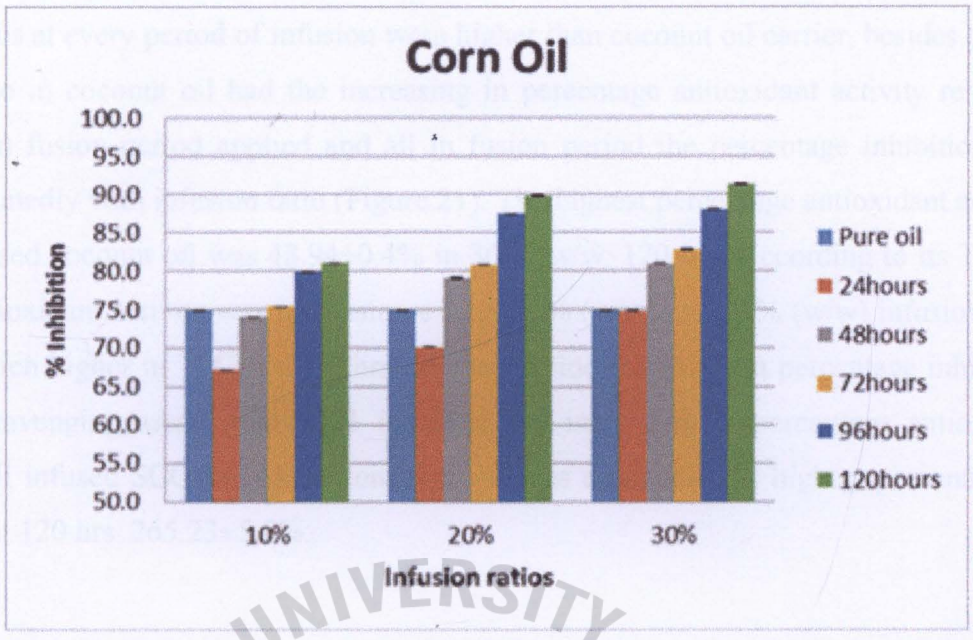


Figure 18 The percentage inhibition of DPPH scavenging assay in corn oil compare between infusion ratio (10%, 20% and 30% w/w) and infusion period (24, 48, 72, 96 and 120 hours)

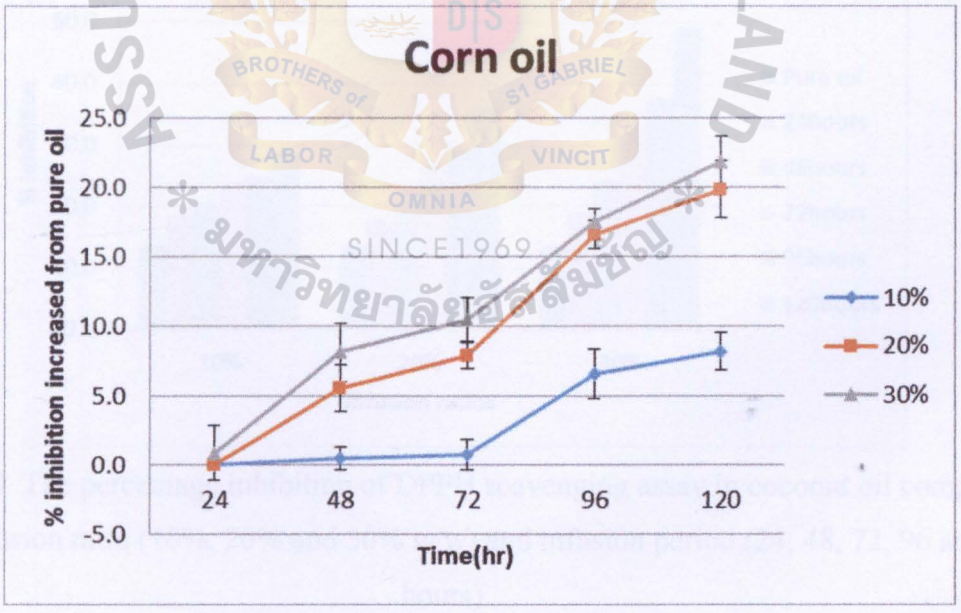


Figure 19 The percentage inhibition increased from carrier oil in corn oil in each infused ratio (10%, 20% and 30%) from 24 houres to 120 hours infusion period

For coconut oil in (Figure 20), it was obviously found that antioxidant activity of all infusion ratios at every period of infusion were higher than coconut oil carrier, besides this all infusion ratio in coconut oil had the increasing in percentage antioxidant activity relatedly with more in fusion period applied and all in fusion period the percentage inhibition also increased relatedly with infusion ratio (Figure 21). The highest percentage antioxidant activity of SCG infused coconut oil was 48.94±0.4% in 30% (w/w, 120 hrs. According to its TPC in figure 9, antioxidant activity was increasing except 10% (w/w) and 30% (w/w) infusion ratio at 72 hrs which higher in TPC than 96 hrs infusion period but lower in percentage inhibition of DPPH scavenging assay. Figure 21 indicated the increasing in percentage antioxidant activity in all infused SCG infused coconut oil samples especially the highest percentage in 30% (w/w) at 120 hrs 265.23±5.8%.

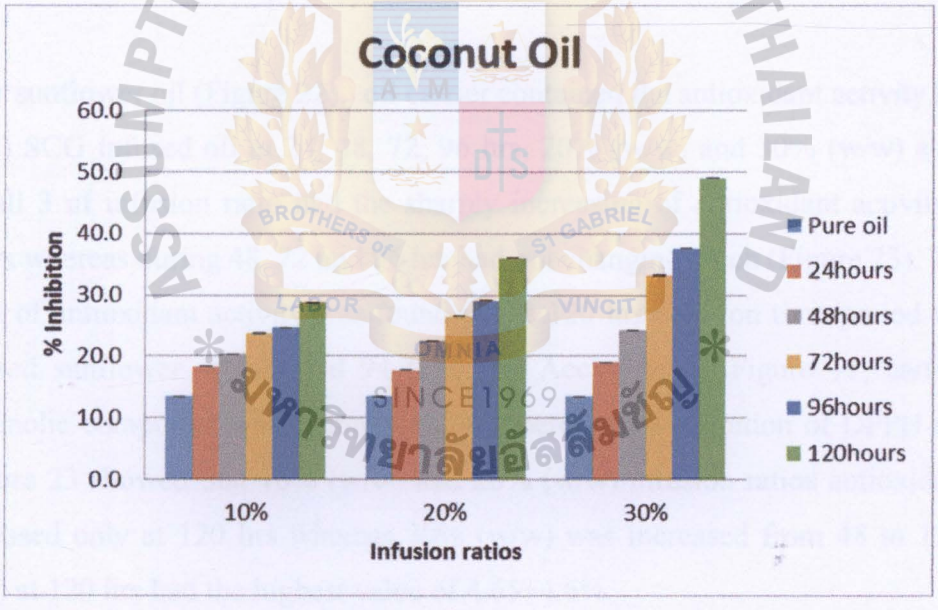


Figure 20 The percentage inhibition of DPPH scavenging assay in coconut oil compare between infusion ratio (10%, 20% and 30% w/w) and infusion period (24, 48, 72, 96 and 120 hours)

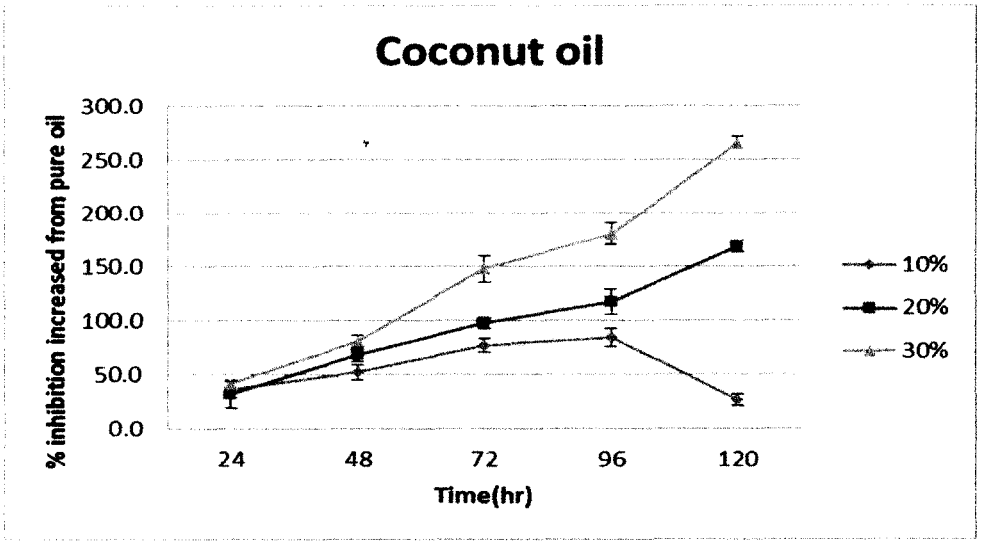


Figure 21 The percentage inhibition increased from carrier oil in coconut oil in each infused ratio (10%, 20% and 30%) from 24 houres to 120 hours infusion period

For sunflower oil (Figure 22), oil carrier contained the antioxidant activity higher than 10% (w/w) SCG infused oil at 24, 48, 72, 96 hrs, 20% (w/w) and 30% (w/w) at 24 hrs. In additiion all 3 of infusion ratio had the sharply increasing of antioxidant activity at 48 hrs from 24 hrs whereas during 48, 72 and 96 hrs had not changing much (Figure 23). The highest percentage of antioxidant activity was found in the 120 hrs infusion time period which 30% SCG infused sunflower oil showed 94.71±0.3%. According to Figure 11, carrier oil had lowest phenolic compound which unrelated with percentage inhibition of DPPH scavenging assay. Figure 23 showed that 10% (w/w) and 20% (w/w) infusion ratios antioxidant activity were increased only at 120 hrs whereas 30% (w/w) was increased from 48 to 120 hrs. So, 30% (w/w) at 120 hrs had the highest value of 4.65±4.6%.

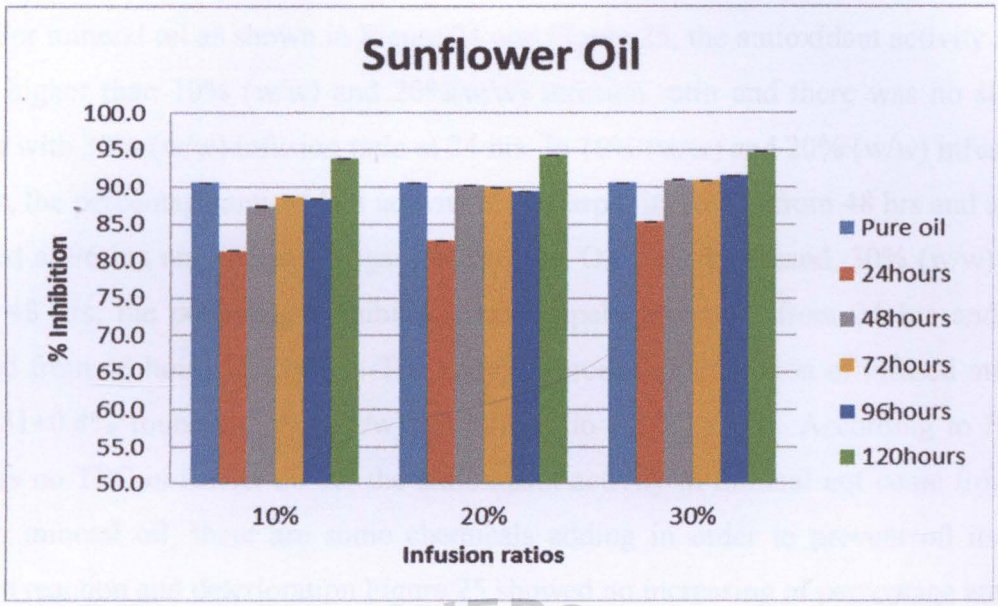


Figure 22 The percentage inhibition of DPPH scavenging assay in sunflower oil compare between infusion ratio (10%, 20% and 30% w/w) and infusion period (24, 48, 72, 96 and 120 hours)

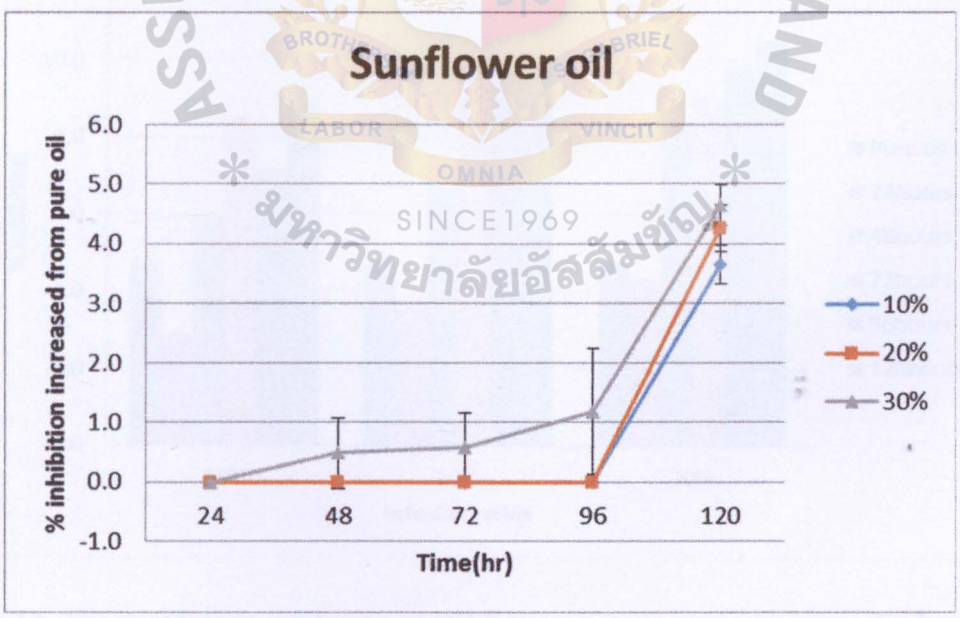


Figure 23 The percentage inhibition increased from carrier oil in sunflower oil in each infused ratio (10%, 20% and 30%) from 24 houres to 120 hours infusion period

For mineral oil as shown in Figure 24 and Figure 25, the antioxidant activity in carrier oil was higher than 10% (w/w) and 20%(w/w) infusion ratio and there was no significant different with 30% (w/w) infusion ratio at 24 hrs. In 10% (w/w) and 20% (w/w) infusion ratio at 72 hrs, the percentage antioxidant activity was sharply increased from 48 hrs and a little bit decreased at 96 hrs and increased again at 120 hrs. On the others hand, 30% (w/w) infusion ratio at 48 hrs, the percentage inhibition was shapely increased from 24 hrs and slightly increased from 48 hours to 120 hrs. The highest percentage inhibition of infused mineral oil was 10.51±0.4% found in 30% (w/w) infusion ratio at 120 hours. According to Figure13, there was no TPC in carrier oil so, the antioxidant activity in mineral not come from TPC's itself. In mineral oil, there are some chemicals adding in order to prevent oil itself from oxidation reaction and deterioration Figure 25 showed no increasing of percentage antioxidant activity in 10% (w/w) and 20% (w/w) infusion ratios. The 30% (w/w) infusion ratio at 120 hrs indicated the highest percentage antioxidant activity increased at 144.41±10.4%.

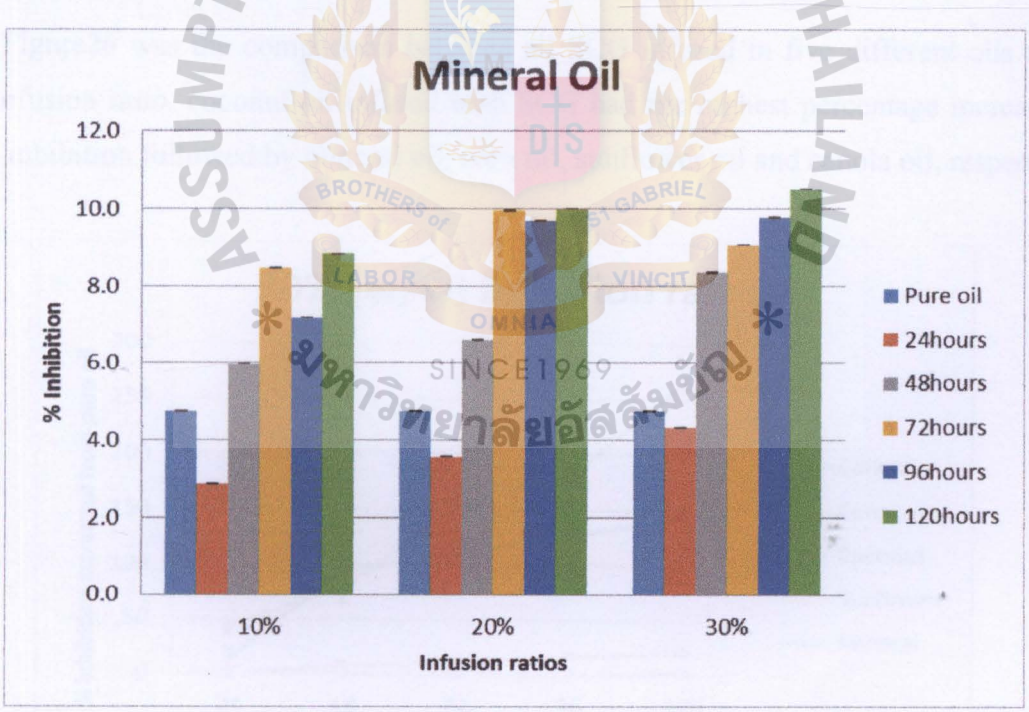


Figure 24 The percentage inhibition of DPPH scavenging assay in mineral oil compare between infusion ratio (10%, 20% and 30% w/w) and infusion period (24, 48, 72, 96 and 120 hours)

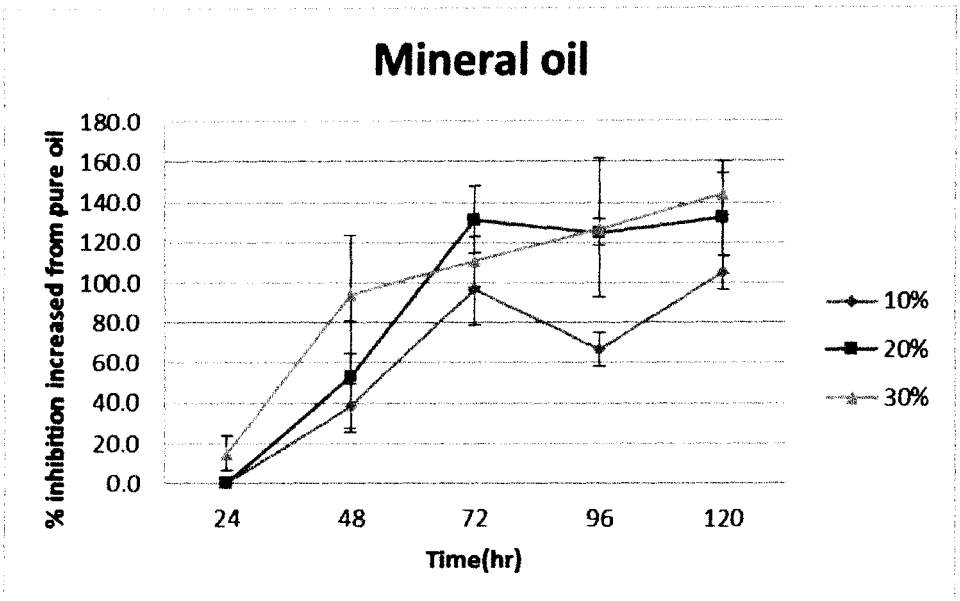


Figure 25 The percentage inhibition increased from carrier oil in mineral oil in each infuse ratio (10%, 20% and 30%) from 24 houres to 120 hours infusion period

Figure26 was the comparison between all SCG infused in five different oils at 30% (w/w) infusion ratio, coconut oil infused with SCG had the highest percentage increasing of percent inhibition followed by mineral oil, corn oil, sunflower oil and canola oil, respectively.

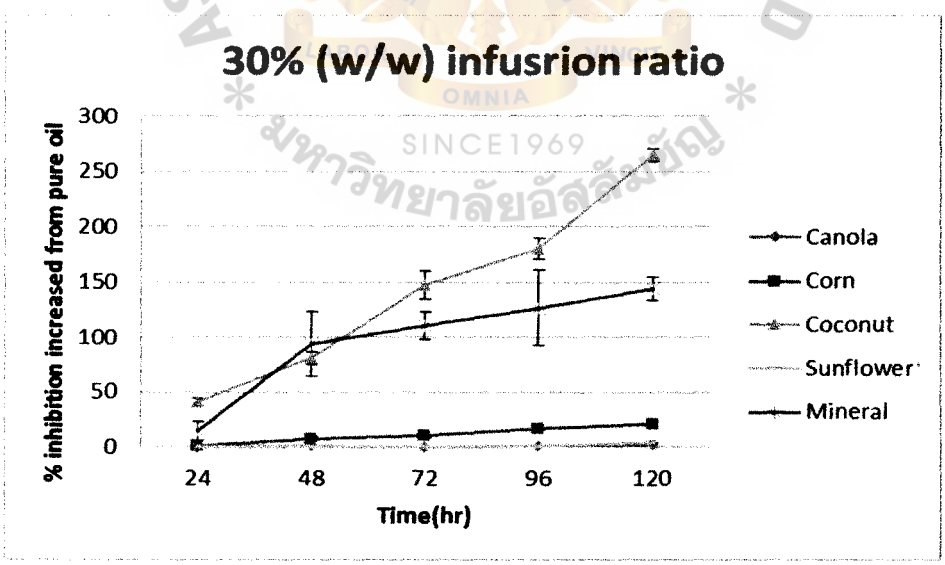


Figure 26 The increasing of percentage inhibition of DPPH scavenging assay from carrier oils in 30%(w/w) infusion ratio from 24 to 120 hours compare between each oils carrier

The result of IC50 was shown in Table 8, IC50 is the inhibition concentration at 50% according to the percentage inhibition, 30% infused sunflower oil at 120 hours exhibited strong antioxidant DPPH radical scavenging activity with the IC50 value of 29.07 mg/ml which had the highest percentage inhibition (94.71%±0.3) In addition, the other SCG infusion in canola and corn oils also exhibited the strong antioxidant activity as the IC 50 value were 27.93 mg/ml and 27.36 mg/ml, respectively. However, the standard ascorbic acid contained IC50 value of 2.97 mg/ml.

Table 8 the inhibition concentration of DPPH scavenging assay at 50% (IC50) in 30% (w/w) infusion ratio from 24 hours to 120hours in each oil

IC50 (mg/ml)						
Oil type	oil carrier	24hours	48hours	72hours	96hours	120hours
Canola	36.15	35.19	28.45	27.58	39.04	27.93
Corn	53.72	54.14	41.91	39.32	34.59	27.36
Coconut	1,119.20	330.72	262.36	179.1	163.4	102.86
Sunflower	41.05	42.98	36.56	32.17	35	29.07
Mineral	3,125.25	4,195.03	1,016.46	706.67	1,408.62	834.07

Note : IC 50 of ascorbic acid = 2.97mg/ml

3. Color intensity

The color of SCG infused oils were measured the absorbance by spectrophotometry method. The maximumwavelength was selected at 420 nm which suitable to observe the range of yellow color. The percentage increasing of color of all SCG infused oils were determined based on color of carrier oils (Table 9).

Table 10 and Table 11 showed the percentage increasing of color in oil after passed infusion process, the highest percentage increasing of color was found in coconut oil, which very different from the others.

Table 9 The measurement of color intensity of oil carrier at 420 nm

Carrier oil	Absorbance value at 420 nm
canola	0.196±0.01
corn	0.426±0.015
coconut	0.014±0.005
sunflower	0.072±0.004
mineral	0±0

Table 10 The percentage of color intensity increased from oil carrier

Canola						
		24 hours	48 hours	72hours	96 hours	120 hours
10%	% color intensity	238.78±15.71	280.1±10.98	336.9±12.03	399.15±31.01	494.73±17.62
20%		421.09±19.11	458.16±18.63	501.87±13.05	600.17±6.46	654.08±29.93
30%		615.31±10.04	690.31±15.37	697.11±14.05	756.63±20.35	804.59±13.72
Corn						
		24 hours	48 hours	72hours	96 hours	120 hours
10%	% color intensity	44.84±4	47.1±3.1	104.3±1.43	130.59±7.54	155.32±2.76
20%		81.22±6.92	106.03±6	183.26±3.56	199.61±5.51	277.23±3.88
30%		166.43±12.92	139.12±17.03	263.46±8.39	297.1±4.8	341.94±15.36
Coconut						
		24 hours	48 hours	72hours	96 hours	120 hours
10%	% color intensity	3323.81±185.49	3876.19±148.86	4550±72.49	6145.24±184.1	7109.52±257.18
20%		5619.05±111.65	6221.43±185.3	7526.19±227.04	8835.71±291.28	9697.62±185.76
30%		8919.05±69.81	9228.57±118.99	10304.76±154.61	11057.14±202.58	11966.67±120.49
Sunflower						
		24 hours	48 hours	72hours	96 hours	120 hours
10%	% color intensity	268.52±9.45	304.63±6.26	562.96±5.26	639.81±12.6	982.41±17.91
20%		418.519±9.85	443.981±4.88	725±9.11	1103.241±12.9	1518.519±37.71
30%		970.37±12.9	1031.481±11.56	1436.574±18.96	1714.352±15.61	2042.13±37.53
Mineral						
		24 hours	48 hours	72hours	96 hours	120 hours
10%	% color intensity	9.97±0.31	10±0.82	19.6±0.96	23.37±0.91	28.8±0.3
20%		24.5±3.3	34±0.3	39.47±0.71	42.03±0.76	46.57±1.25
30%		51.47±0.91	58.6±0.46	67.17±1.06	69.6±1.21	73.73±0.87

Table 11 The percentage color increasing (10^3) from oil carrier of all oil infused samples

Canola						
		Day1	Day2	Day3	Day4	Day5
10%	% color intensity(10^3)	0.24	0.28	0.34	0.40	0.49
20%		0.42	0.46	0.50	0.60	0.65
30%		0.62	0.69	0.70	0.76	0.80
Corn						
		Day1	Day2	Day3	Day4	Day5
10%	% color intensity(10^3)	0.04	0.05	0.10	0.13	0.16
20%		0.08	0.11	0.18	0.20	0.28
30%		0.17	0.14	0.26	0.30	0.34
Coconut						
		Day1	Day2	Day3	Day4	Day5
10%	% color intensity(10^3)	3.32	3.88	4.55	6.15	7.11
20%		5.62	6.22	7.53	8.84	9.70
30%		8.92	9.23	10.30	11.06	11.97
Sunflower						
		Day1	Day2	Day3	Day4	Day5
10%	% color intensity(10^3)	0.27	0.30	0.56	0.64	0.98
20%		0.42	0.44	0.73	1.10	1.52
30%		0.97	1.03	1.44	1.71	2.04
Mineral						
		Day1	Day2	Day3	Day4	Day5
10%	% color intensity(10^3)	0.01	0.01	0.02	0.023	0.029
20%		0.025	0.034	0.039	0.042	0.047
30%		0.051	0.059	0.067	0.07	0.074

For canola oil, in Table 11 and Figure 27, the percentage increasing of color related with infusion time, the highest percentage increasing of color in canola oil found in 30% (w/w) infusion ratio at 120 hours which is 0.8% (10^3). It was showed that the comparison of percentage increasing of color of canola oil in each infusion ratio, 30% (w/w) infusion ratio showed highest percentage increasing of color in every infusion period followed by 20% (w/w) and 10% (w/w) infusion ratio.

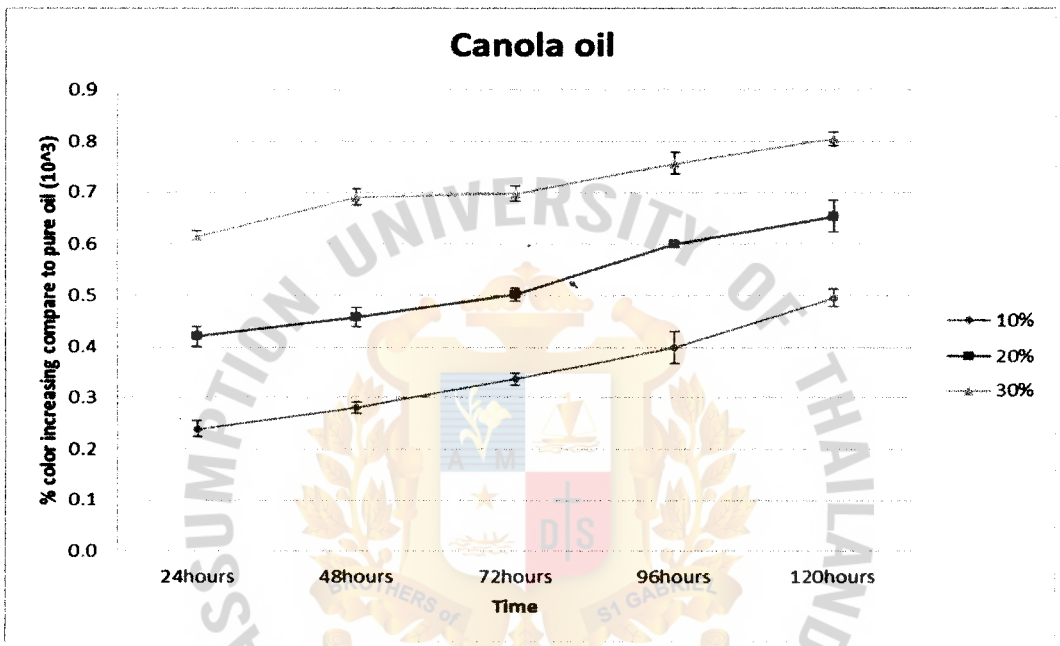


Figure 27 The percentage of color increasing compare to canola oil carrier in each SCG infused ratio (10%, 20% and 30% (w/w)) from 24 hours to 120 hours

For corn oil (Figure28), the20% (w/w) and 10% (w/w) infusion ratioshowed the percentage increasing of color had increase related with infusion time but, for 30% (w/w) infusion ratio at 48 hours was decreased from 24 hours, which is 0.17% (10^3) to 1.4% (10^3) and increased to 0.26% (10^3) at 72 hours. The highest percentage increasing of color in corn oil found in 30% (w/w) infusion ratio at 120 hours which is 0.34% (10^3) It was showed that the comparison of percentage increasing of color of corn oil in each infusion ratio, 30% (w/w) infusion ratio showed highest percentage increasing of color in every infusion period followed by 20% (w/w) and 10% (w/w) infusion ratio.

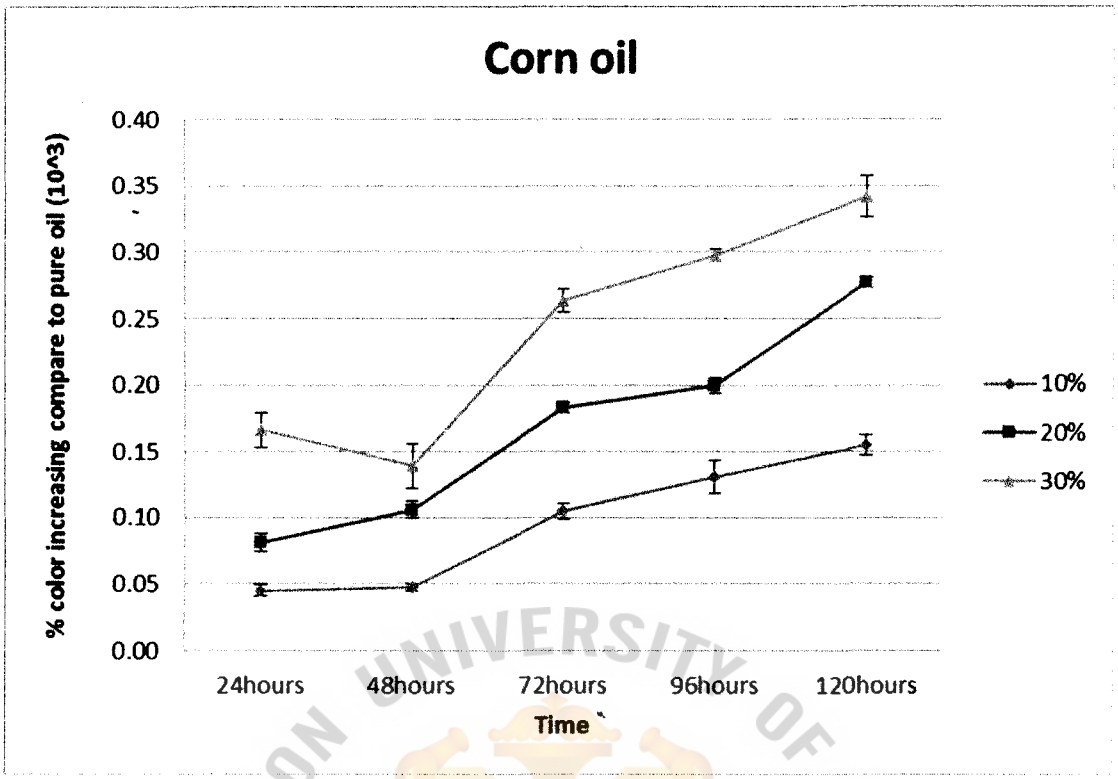


Figure 28 The percentage of color increasing compare to corn oil carrier in each SCG infused ratio (10%, 20% and 30% (w/w)) from 24 hours to 120 hours

For coconut oil, it was obviously found that coconut oil had highest percentage increasing of color compared to others oil carriers. As showin n Table 11 and Figure 29, the percentage increasing of color had increase related with infusion time, the highest percentage increasing of color in canola oil was 11.97% (10^3) which found in 30% (w/w) infusion ratio at 120 hours). Figure 29 showed the comparison of percentage increasing of color of corn oil in each infusion ratio, 30% (w/w) infusion ratio showed highest percentage increasing of color in every infusion period followed by 20% (w/w) and 10% (w/w) infusion ratio.

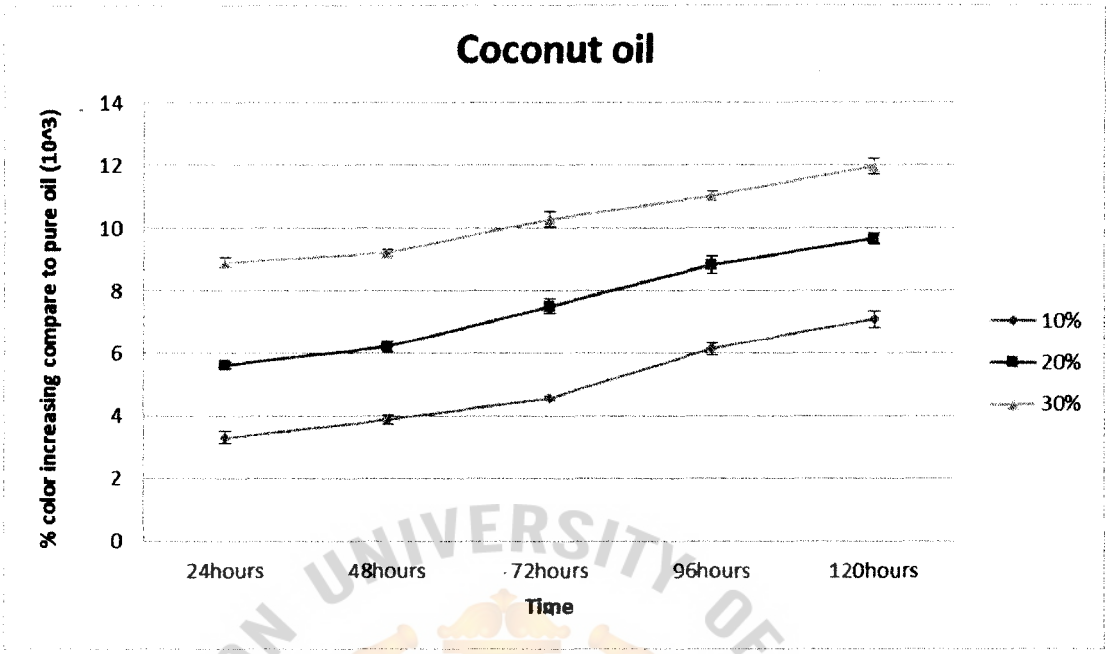


Figure 29 The percentage of color increasing compare to coconut oil carrier in each SCG infused ratio (10%, 20% and 30% (w/w)) from 24 hours to 120 hours

For sunflower oil, the percentage increasing of color had increase related with infusion time, the highest percentage increasing of color in sunflower oil found in 30% (w/w) infusion ratio at 120 hours which is 2.04% (10^3). In Figure 30, it indicated that the comparison of percentage increasing of color of canola oil in each infusion ratio, 30% (w/w) infusion ratio showed highest percentage increasing of color in every infusion period followed by 20% (w/w) and 10% (w/w) infusion ratio.

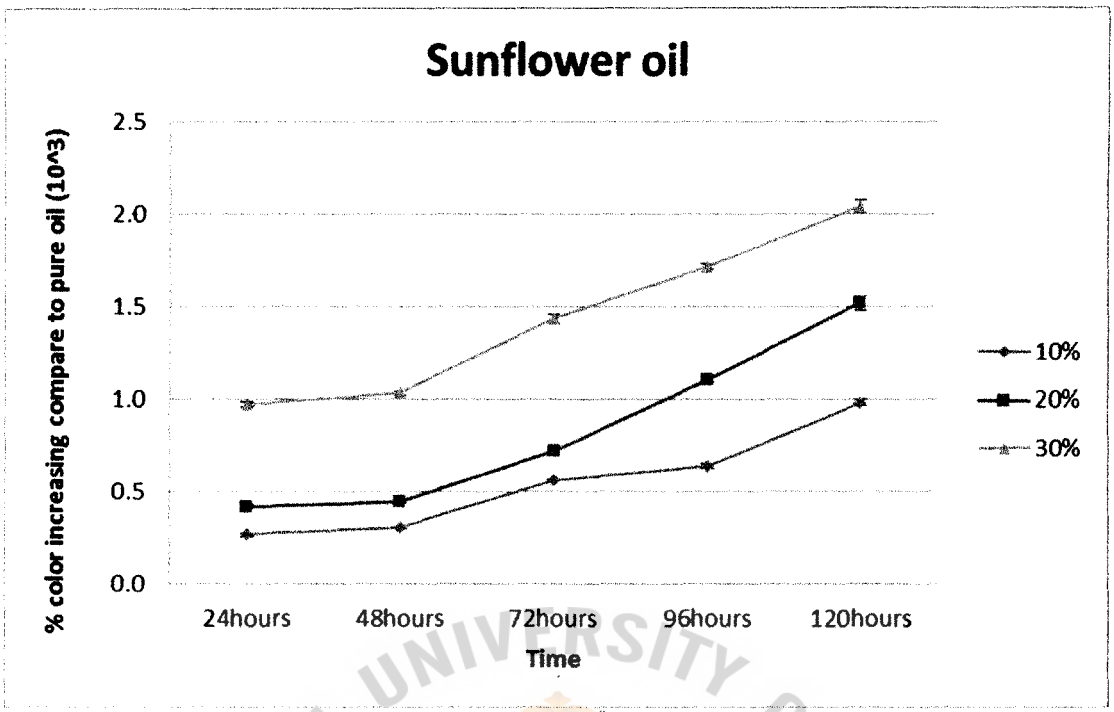


Figure 30 The percentage of color increasing compare to sunflower oil carrier in each SCG infused ratio (10%, 20% and 30% (w/w)) from 24 hours to 120 hours

For mineral oil, it had lowest percentage increasing of color compared to others oil carriers. In Table 11 and Figure 31, the percentage increasing of color had increase related with infusion time, the highest percentage increasing of color in canola oil found in 30% (w/w) infusion ratio at 120 hours which is 0.074% (10^3). In Figure 31 Showed the comparison of percentage increasing of color of canola oil in each infusion ratio, 30% (w/w) infusion ratio showed highest percentage increasing of color in every infusion period followed by 20% (w/w) and 10% (w/w) infusion ratio

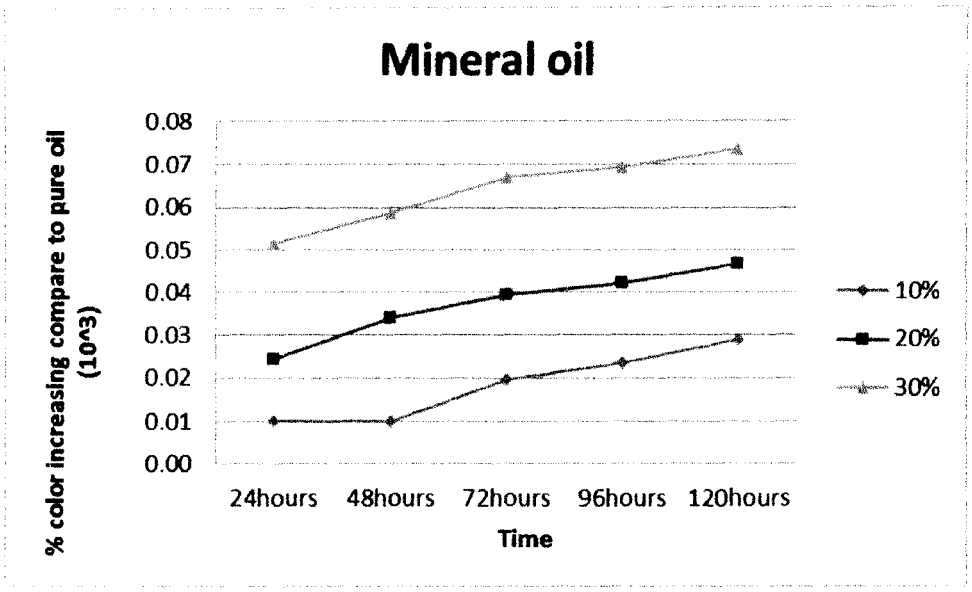


Figure 31 The percentage of color increasing compare to mineral oil carrier in each SCG infused ratio (10%, 20% and 30% (w/w)) from 24 hours to 120 hours

4. Peroxide value

Peroxide value of carrier oils and infused oils were determined by iodometric titration method. 30% ratio infusion at 120hours of all carrier oils was selected to use as samples, due to their phenolic compounds and antioxidant activities. The used of titrant volumes (0.1N sodium thiosulphate) were used for calculation of peroxide value in unit of milliequivalent/kilograms (meq/kg). The peroxide value of canola, corn. Coconut, sunflower and mineral oil were 10, 4, 2, 22, and 0 meq/kg, respectively (Table 12).

After infusion process, there were three types of oil carrier that had development in peroxide value which were canola oil from 10 to 16meq/kg, corn oil from 4 to 8meq/kg, sunflower oil from 22 to 36. From the result, it was obvious that sunflower oil had highest peroxide value 36meq/kg. Corn oil had highest percentage peroxide value increased from carrier oil 100%. Spent coffee grounds were not promoted inhibition of peroxide value in these carrier oils or acidity in coffee itself induced the oxidation to the carrier oils.

For coconut oil, there was not changing in peroxide value after infusion process. The reason was coconut oil was consist of saturated fatty acid which was the stable from of fatty acid and harder to oxidize than unsaturated fatty acid. And peroxide value in mineral oil was not found because it distillated from petroleum and did not have fatty acid to oxidize with oxygen.

Table 12 Peroxide value of carrier oils, SCG infused oils (30%(w/w)/120hours) and percentage peroxide value increase from carrier oil.

Peroxide Value(meq/kg)			
infused oil	carrier oil	30%(w/w)/120hours	% POV increased from pure oil
	POV	POV	
canola	10	16	60
corn	4	8	100
coconut	2	2	0
sunflower	22	36	63.64
mineral	0	0	0

5. Antioxidant soap bar with coffee scrub

5.1 Preference test

After infused oils used as essential oils for making glycerin soap. In addition, non-added glycerin soap was used as control, total of sample soaps were six types, they were graded liking score range from 1-9 based on panelists' preferences on these following attribute; color(transparency), fragrance, lather, moisturizing, scrubbing, skin feel and over all liking.

The data from preference test was statistically prove by Duncan multiple range test

Table13: preference scores of each type of infused oils soaps on seven attributes

Attributes	Oils					
	Canola	Corn	Coconut	Sunflower	Mineral	Control
Color	6.05 ± 1.00^{b}	5.95 ± 0.76^{b}	6.35 ± 1.23^{b}	6.45 ± 1.00^{b}	6.35 ± 1.31^{b}	7.35 ± 1.09^{a}
Fragrance	5.95 ± 1.70^{a}	6 ± 1.26^{a}	6.2 ± 1.54^{a}	5.3 ± 1.84^{b}	6.3 ± 1.42^{a}	6.35 ± 1.79^{a}
Lather	6.3 ± 1.59^{ab}	6.2 ± 1.79^{b}	6.55 ± 1.54^{ab}	7.1 ± 0.72^{ab}	6.6 ± 1.23^{ab}	7.2 ± 1.54^{a}
moisturizing	6.5 ± 1.15^{ns}	6.25 ± 1.29^{ns}	6.65 ± 1.09^{ns}	6.2 ± 1.32^{ns}	6.75 ± 1.16^{ns}	6.65 ± 1.35^{ns}
scrubbing	6.2 ± 1.40^{ns}	6.05 ± 1.64^{ns}	6.5 ± 1.61^{ns}	6.65 ± 1.27^{ns}	6.35 ± 1.87^{ns}	6.9 ± 1.52^{ns}
Skin feel	6.55 ± 1.10^{ab}	6.4 ± 1.14^{ab}	6.55 ± 1.36^{ab}	6.3 ± 1.26^{b}	6.75 ± 1.16^{ab}	7.1 ± 1.33^{a}
Over all liking	6.8 ± 0.83^{ab}	6.65 ± 0.93^{ab}	6.75 ± 1.02^{ab}	6.55 ± 1.00^{b}	6.95 ± 0.94^{ab}	7.3 ± 1.45^{a}

Remark: $^{a\ b}$ means with superscripts in the column and those without a common superscript were significantly different $\alpha=0.05$. ^{ns} superscript means no significantly different $\alpha=0.05$

From the results, there were no significantly different between each oil on these attributes; moisturizing and scrubbing. For color, control had highest score 7.35±1.09 significantly different from others soaps sample. For fragrance, infused sunflower oil soap had lowest score 5.3±1.84 significantly different from others soaps sample. For lather, control had 7.2±1.54 higher score than 6.2±1.79 infused corn oil soap significantly. For skin feel and over all liking, control had significantly higher scores than infused sunflower oil soap, 7.1±1.33 to 6.3±1.26 and 7.3±1.45 to 6.55±1.00, respectively.

5.2.Survey Questions

One hundred people were assigned to do survey questions for antioxidant glycerin soap bar with coffee scrub.

From the result, 62% were female and 38% were male. 35% of respondents were range between 41-50 years old and 23% were 51-60 years old. People who used soap bar and liquid soap were separated half by half which were 49% and 48%, respectively. Lux was the most used brand 26.3% followed by Shokubutsu 17.9% and Protex 14.7%. 30% of respondents had been using the same brand over 3 years, 17% had been using between 1 to 2 years and 16% had been using lower than 3 months. Fragrance was considered to be the most important attribute for respondents meanwhile other attributes were in range of important level. 58% of respondents did not know antioxidant soap but, 57% used to use scrub which most of natural scrubbers were rice (36.1%) and coffee (36.1%).

Lastly, 55.6% were not decided to purchase antioxidant glycerin soap bar with coffee scrub product yet, 31.3% were decided to purchase product meanwhile only13.1% were not response to this product. However, 55.6% was more than half of respondents that might or might not purchase the product which challenged to make them purchase product by marketing plans and development of product.

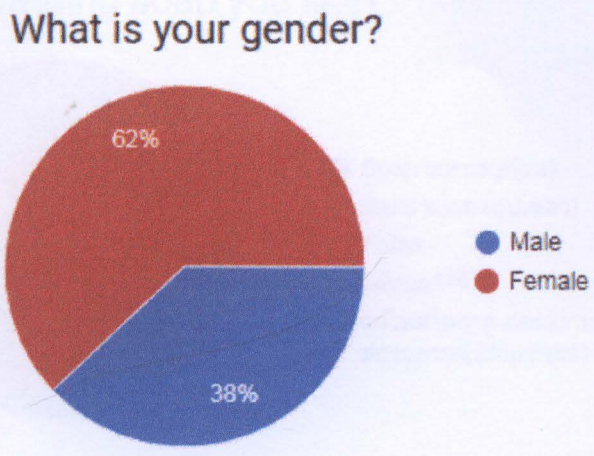


Figure 32 survey question; what is your gender?

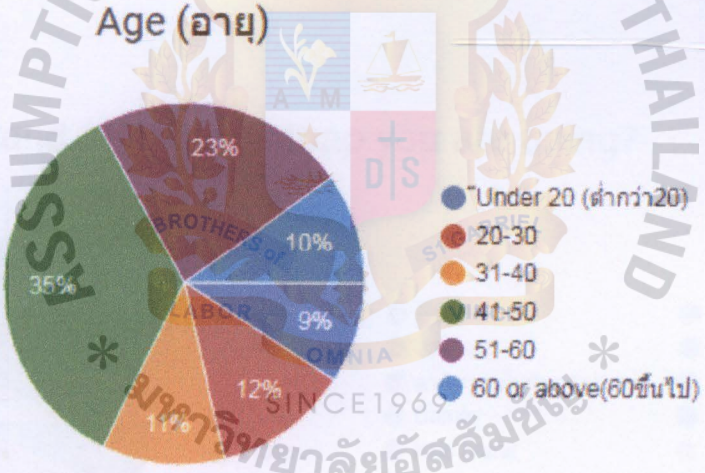


Figure33 survey question; age

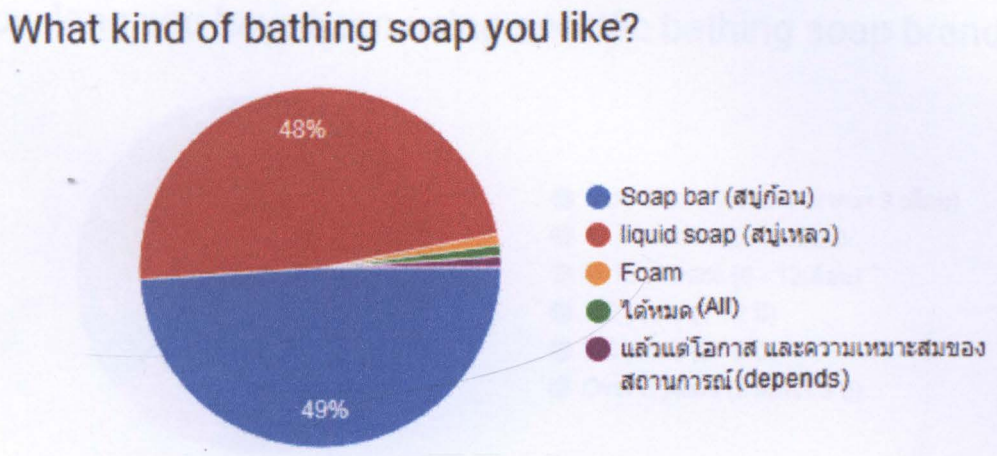


Figure34 survey question; What kind of bathing soap you like?

Figure35 survey question; Which brand of bathing soap you are using?

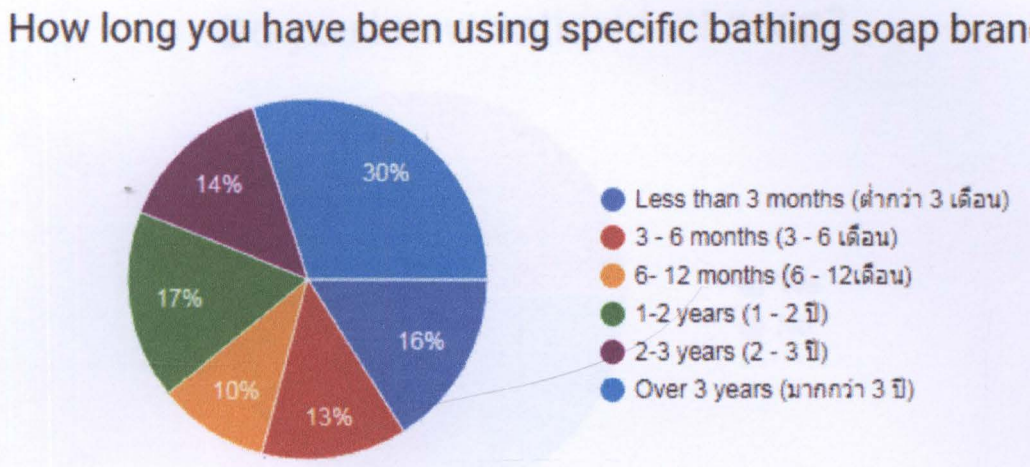


Figure36 survey question; How long you have been using specific bathing soap brand?

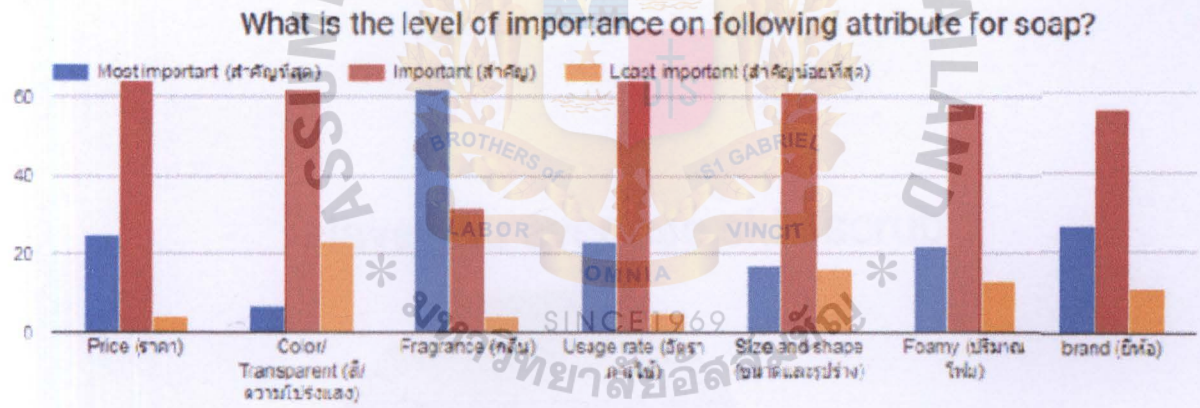


Figure37 survey question; What is the level of importance on following attributes for soap?

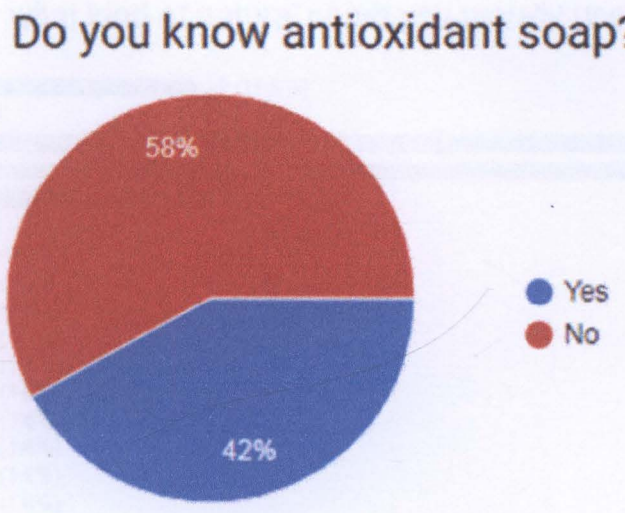


Figure38 survey question; Do you know antioxidant soap?

Figure39 survey question; Have you ever use skin scrub?

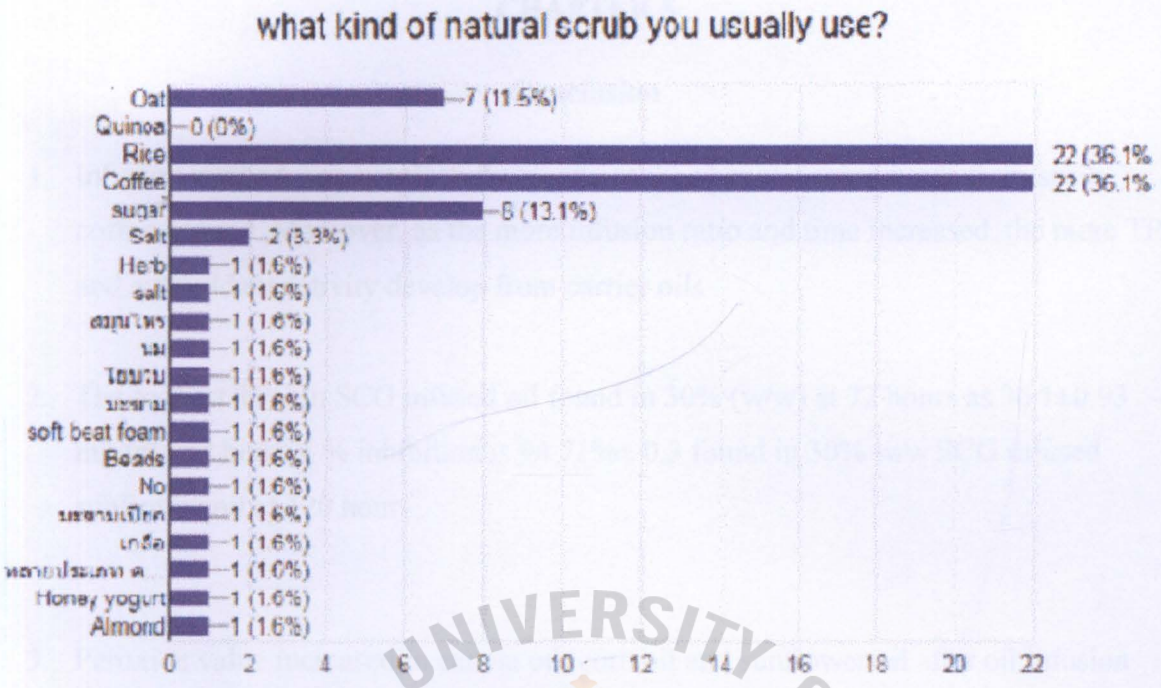


Figure40 survey question; If there is an antioxidant glycerin soap bar with coffee scrub available in market, will you buy it

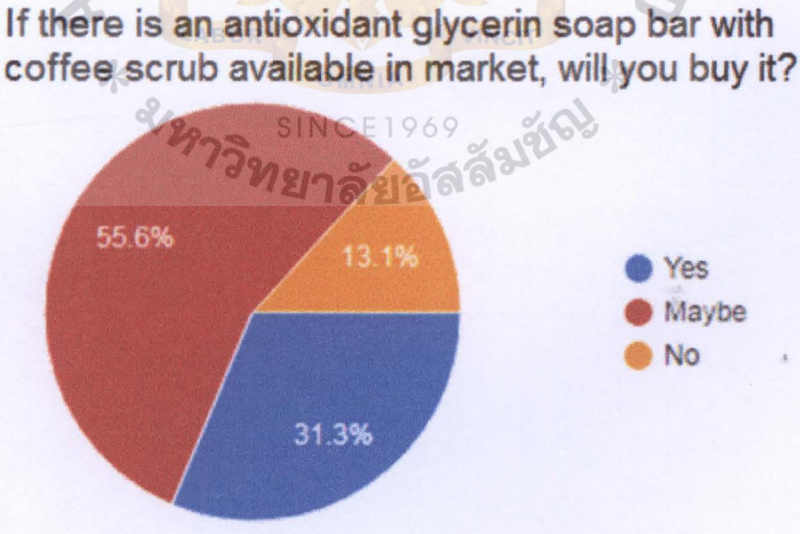


Figure 41 survey question; If there is an antioxidant glycerin soap bar with coffee scrub available in market, will you buy it

CHAPTER 5

Conclusion

1. Infusion method was successfully to extract the phenolic compounds from spent coffee ground. Moreover, as the more infusion ratio and time increased, the more TPC and antioxidant activity develop from carrier oils

2. The highest TPC in SCG infused oil found in 30% (w/w) at 72 hours as 36.1±0.93 mg/ml and highest % inhibition is 94.71%± 0.3 found in 30% w/w SCG infused sunflower oil at 120 hours.

3. Peroxide value increased in canola oil, corn oil and sunflower oil after oil infusion method whereas unchanged in coconut oil and not found in mineral oil

4. 30%(w/w) SCG infused sunflower oil at 120 hours was considered as the best infused oil to develop antioxidant soap due to antioxidant activity and result from preference test

REFERENCES

1. Ana S. P. Moreira,a Fernando M. Nunes,b M. Rosario Dominguesa and Manuel A. Coimbra, (2012). Coffee melanoidins: structures, mechanisms of formation and potential health impacts.

2. Viesturs Kreicbergs, Fredijs Dimins*, Velga Mikelsone, Ingmars Cinkmanis (2011). BIOLOGICALLY ACTIVE COMPOUNDS IN ROASTED COFFEE.

3. Selin Sahin, Mehmet Bilgin, Ezgi Sayım & Bedia Guvenilir, (2016) Effects of natural antioxidants in the improvement of corn oil quality: olive leaf vs. lemon balm.

4. I.A.R. Gray. OXIDATION INHIBITORS FOR INSULATING OILS.

5. Fereidoon Shahidi and Udaya Wanasundar, (1994) Stabilization of Canola Oil by Natural Antioxidants.

6. Alessia Panusa†, Antonio Zuorro§, Roberto Lavecchia§, Giancarlo Marrosu, and Rita Petrucci, (2013) Recovery of Natural Antioxidants from Spent Coffee Grounds

7. Carina Coelho, Miguel Ribeiro, Ana S. C. Cruz, Maria Rosario M. Domingues, Manuel A. Coimbra, Mirko Bunzel, and Fernando Milheiro Nunes, (2014) Nature of Phenolic Compounds in Coffee Melanoidins

8. Adriana Farah, Mariana Monteiro, Carmen M. Donangelo, and Sophie Lafay, (2008) Chlorogenic Acids from Green Coffee Extract are Highly Bioavailable in Humans

9. Coffeechemistry., Chlorogenic Acid

   (https://www.coffeechemistry.com/chemistry/acids/chlorogenic-acid)

10. Coffeechemistry The Chemistry of Organic Acids: Part 1

    (https://www.coffeechemistry.com/library/coffee-science-publications/the-chemistry-of-organic-acids-part-1)

11. Coffeechemistry The Chemistry of Organic Acids: Part 2 (https://www.coffeechemistry.com/library/coffee-science-publications/the-chemistry-of-organic-acids-part-2)

12. *Emma Sage, Coffee Science Manager, Specialty Coffee Association of America*, (2014) Coffee Roasting Chemistry: Chlorogenic Acids

13. Adriana Farah; Carmen Marino Donangelo, (2006) Phenolic compounds in coffee

14. Selfgrowth, Benefits of Antioxidants – Homemade Skin Care and More (http://www.selfgrowth.com/articles/benefits-of-antioxidants-homemade-skin-care-and-more)

15. Aromaweb, What are Infused Oils (also Known as Macerated Oils)? (http://www.aromaweb.com/articles/whatinfu.asp)

16. Mayada Damechki, Sofia Sotiropoulou and Maria Tsimidou, (2001). Antioxidant and pro-oxidant factors in oregano and rosemary gourmet olive oils

17. nutrition-and-you, Canola oil nutrition facts (http://www.nutrition-and-you.com/canola-oil.html)

18. Fereidoon Shahidi and Udaya Wanasundara , (1994). Canola Extract as an Alternative Natural Antioxidant for Canola Oil

19. Oilhealthbenefits, Corn oil. (oilhealthbenefits.com/corn-oil/)

20. A. M. Marina, Y. B. Che man, S. A. H. Nazimah & I. Amin, (2009). Antioxidant capacity and phenolic acids of virgin coconut oil.

21. Sunflowernsa, Sunflower Oil - Your Healthy Choice. (http://www.sunflowernsa.com/oil)

22. Britannica, Mineral oil.

(www.britannica.com/technology/mineral-oil)

23. Livestrong, Benefits of Glycerin Soap.

(http://www.livestrong.com/article/87954-benefits-glycerin-soap/)

24. Bioactive Food AS, Omega in your Body

(http://www.1life63.com/en/omega-in-your-body-oxidation-of-lipids/oxidation-of-lipids)

APPENDIX

I. Statistic analyzed

1. Total phenolic compound

Table1 Raw data of total phenolic compound in all carrier oils

	Canola	corn	coconut	sunflower	mineral
1st	0.047	0.01	0.012	0.016	0
TPC	22.56	19.32	19.49	19.84	0.00
2nd	0.043	0.024	0.007	0.008	0
TPC	22.21	20.54	19.05	19.14	0.00
3rd	0.042	0.02	0.008	0.011	0
TPC	22.12	20.19	19.14	19.40	0.00

Table2 Raw data of total phenolic compound in canola infused oil

Canola						
		24hours	48hours	72hours	96hours	120hours
10%	1st%	0.054	0.059	0.101	0.090	0.105
	TPC	23.175	23.614	27.298	26.333	27.649
	%TPC increase	3.935	5.902	22.425	18.097	23.998
	2nd%	0.039	0.056	0.117	0.089	0.079
	TPC	21.860	23.351	28.702	26.246	25.368
	%TPC increase	-1.966	4.722	28.719	17.704	13.770
	3rd%	0.045	0.068	0.087	0.044	0.091
	TPC	22.386	24.404	26.070	22.298	26.421
	%TPC increase	0.394	9.443	16.917	0.001	18.491
20%	1st%	0.065	0.083	0.121	0.124	0.130
	TPC	24.140	25.719	29.053	29.316	29.842
	%TPC increase	8.262	15.344	30.293	31.473	33.833
	2nd%	0.066	0.101	0.118	0.077	0.192
	TPC	24.228	27.298	28.789	25.193	35.281
	%TPC increase	8.656	22.425	29.112	12.983	58.224
	3rd%	0.071	0.063	0.093	0.092	0.148
	TPC	24.667	23.965	26.596	26.509	31.421
	%TPC increase	10.623	7.476	19.277	18.884	40.914
30%	1st%	0.140	0.089	0.203	0.180	0.154
	TPC	30.719	26.246	36.246	34.228	31.947
	%TPC increase	37.767	17.704	62.551	53.503	43.275
	2nd%	0.132	0.122	0.211	0.160	0.155
	TPC	30.018	29.140	36.947	32.474	32.035
	%TPC increase	34.620	30.686	65.698	45.635	43.668
	3rd%	0.091	0.117	0.190	0.216	0.227
	TPC	26.421	28.702	35.105	37.386	38.351
	%TPC increase	18.491	28.719	57.437	67.665	71.992

Table3 Raw data of total phenolic compound in corn infused oil

corn						
		24hrs	48hrs	72hrs	96hrs	120hrs
10%	1st%	0.013	0.021	0.035	0.037	0.014
	TPC	19.579	20.281	21.509	21.684	19.667
	%TPC increase	-2.193	1.312	7.447	8.324	-1.755
	2nd%	0.029	0.032	0.040	0.060	0.057
	TPC	20.982	21.246	21.947	23.702	23.439
	%TPC increase	4.818	6.133	9.638	18.402	17.088
	3rd%	0.023	0.022	0.032	0.041	0.039
	TPC	20.456	20.368	21.246	22.035	21.860
	%TPC increase	2.189	1.751	6.133	10.076	9.200
20%	1st%	0.043	0.059	0.078	0.105	0.084
	TPC	22.211	23.614	25.281	27.649	25.807
	%TPC increase	10.953	17.964	26.290	38.121	28.919
	2nd%	0.049	0.055	0.057	0.116	0.104
	TPC	22.737	23.263	23.439	28.614	27.561
	%TPC increase	13.582	16.211	17.088	42.942	37.683
	3rd%	0.054	0.063	0.059	0.096	0.122
	TPC	23.175	23.965	23.614	26.860	29.140
	%TPC increase	15.773	19.717	17.964	34.177	45.571
30%	1st%	0.072	0.095	0.131	0.104	0.188
	TPC	24.754	26.772	29.930	27.561	34.930
	%TPC increase	11.016	20.064	34.226	23.605	56.650
	2nd%	0.056	0.092	0.126	0.153	0.202
	TPC	23.351	26.509	29.491	31.860	36.158
	%TPC increase	4.722	18.884	32.260	42.881	62.158
	3rd%	0.089	0.057	0.085	0.131	0.153
	TPC	26.246	23.439	25.895	29.930	31.860
	%TPC increase	17.704	5.115	16.130	34.226	42.881

Table4 Raw data of total phenolic compound in infused coconut oil

Coconut						
		24hours	48hours	72hours	96hours	120hours
10%	1st%	0.022	0.038	0.046	0.032	0.068
	TPC	20.368	21.772	22.474	21.246	24.404
	%TPC increase	5.931	13.230	16.880	10.493	26.917
	2nd%	0.034	0.028	0.051	0.034	0.044
	TPC	21.421	20.895	22.912	21.421	22.298
	%TPC increase	11.406	8.668	19.161	11.406	15.968
	3rd%	0.041	0.031	0.055	0.028	0.061
	TPC	22.035	21.158	23.263	20.895	23.789
	%TPC increase	14.599	10.037	20.986	8.668	23.723
20%	1st%	0.047	0.058	0.072	0.084	0.108
	TPC	22.561	23.526	24.754	25.807	27.912
	%TPC increase	17.336	22.354	28.741	34.216	45.165
	2nd%	0.038	0.066	0.076	0.073	0.096
	TPC	21.772	24.228	25.105	24.842	26.860
	%TPC increase	13.230	26.004	30.566	29.198	39.690
	3rd%	0.062	0.079	0.081	0.091	0.111
	TPC	23.877	25.368	25.544	26.421	28.175
	%TPC increase	24.179	31.935	32.847	37.409	46.533
30%	1st%	0.089	0.102	0.149	0.153	0.165
	TPC	26.246	27.386	31.509	31.860	32.912
	%TPC increase	36.497	42.428	63.869	65.694	71.169
	2nd%	0.076	0.083	0.137	0.129	0.115
	TPC	25.105	25.719	30.456	29.754	28.526
	%TPC increase	30.566	33.760	58.395	54.745	48.358
	3rd%	0.099	0.117	0.129	0.106	0.127
	TPC	27.123	28.702	29.754	27.737	29.579
	%TPC increase	41.059	49.271	54.745	44.252	53.833

Table5 Raw data of total phenolic compound in infused sunflower oil

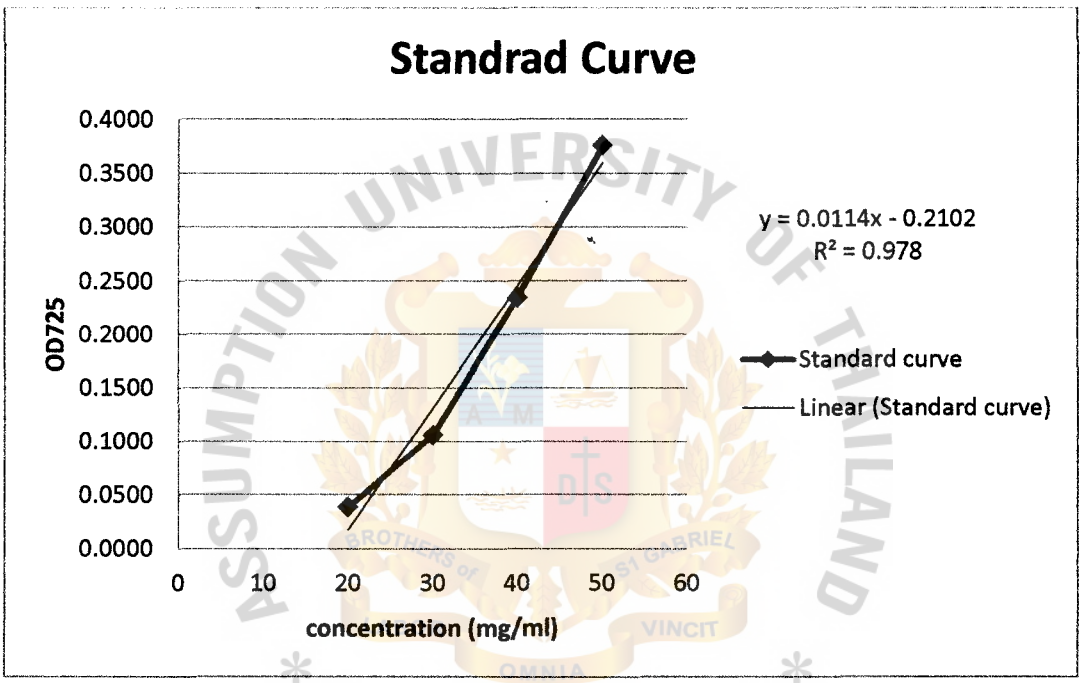
Sunflower						
		24hours	48hours	72hours	96hours	120hours
10%	1st%	0.031	0.042	0.050	0.038	0.076
	TPC	21.158	22.123	22.825	21.772	25.105
	%TPC increase	8.714	13.672	17.278	11.869	28.996
	2nd%	0.049	0.033	0.045	0.054	0.034
	TPC	22.737	21.333	22.386	23.175	21.421
	%TPC increase	16.827	9.615	15.024	19.080	10.066
	3rd%	0.026	0.048	0.047	0.043	0.062
	TPC	20.719	22.649	22.561	22.211	23.877
	%TPC increase	6.460	16.376	15.925	14.123	22.686
20%	1st%	0.033	0.035	0.100	0.053	0.087
	TPC	21.333	21.509	27.211	23.088	26.070
	%TPC increase	9.615	10.517	39.814	18.630	33.954
	2nd%	0.039	0.047	0.079	0.062	0.091
	TPC	21.860	22.561	25.368	23.877	26.421
	%TPC increase	12.320	15.925	30.348	22.686	35.757
	3rd%	0.057	0.050	0.083	0.059	0.090
	TPC	23.439	22.825	25.719	23.614	26.333
	%TPC increase	20.433	17.278	32.151	21.334	35.306
30%	1st%	0.053	0.069	0.135	0.128	0.111
	TPC	23.088	24.491	30.281	29.667	28.175
	%TPC increase	18.630	25.841	55.589	52.434	44.772
	2nd%	0.071	0.051	0.093	0.116	0.132
	TPC	24.667	22.912	26.596	28.614	30.018
	%TPC increase	26.743	17.728	36.659	47.025	54.237
	3rd%	0.063	0.074	0.080	0.107	0.140
	TPC	23.965	24.930	25.456	27.825	30.719
	%TPC increase	23.137	28.095	30.799	42.969	57.842

Table6 Raw data of total phenolic compound in infused mineral oil

Mineral						
		24hours	48hours	72hours	96hours	120hours
10%	1st%	0.000	0.000	0.024	0.031	0.019
	TPC	0.000	0.000	20.544	21.158	20.105
	%TPC increase	0.000	0.000	20.544	21.158	20.105
	2nd%	0.000	0.000	0.017	0.022	0.039
	TPC	0.000	0.000	19.930	20.368	21.860
	%TPC increase	0.000	0.000	19.930	20.368	21.860
	3rd%	0.000	0.000	0.021	0.034	0.041
	TPC	0.000	0.000	20.281	21.421	22.035
	%TPC increase	0.000	0.000	20.281	21.421	22.035
20%	1st%	0.000	0.000	0.030	0.029	0.024
	TPC	0.000	0.000	21.070	20.982	20.544
	%TPC increase	0.000	0.000	21.070	20.982	20.544
	2nd%	0.000	0.000	0.021	0.036	0.026
	TPC	0.000	0.000	20.281	21.596	20.719
	%TPC increase	0.000	0.000	20.281	21.596	20.719
	3rd%	0.000	0.000	0.027	0.044	0.032
	TPC	0.000	0.000	20.807	22.298	21.246
	%TPC increase	0.000	0.000	20.807	22.298	21.246
30%	1st%	0.013	0.024	0.018	0.018	0.034
	TPC	19.579	20.544	20.018	20.018	21.421
	%TPC increase	19.579	20.544	20.018	20.018	21.421
	2nd%	0.016	0.019	0.021	0.027	0.050
	TPC	19.842	20.105	20.281	20.807	22.825
	%TPC increase	19.842	20.105	20.281	20.807	22.825
	3rd%	0.021	0.022	0.032	0.041	0.039
	TPC	20.281	20.368	21.246	22.035	21.860
	%TPC increase	20.281	20.368	21.246	22.035	21.860

Table7 Standard curve of gallic acid investigation of folin-ciocalteu assay phenolic compound

OD725	20mg/ml	30mg/ml	40mg/ml	50mg/ml
1st	0.037	0.098	0.213	0.387
2nd	0.038	0.114	0.255	0.364
3rd	0.041	0.107	0.237	0.377
Average	0.0387	0.1063	0.2350	0.3760



2. Antioxidant activity DPPH scavenging assay

Table8 Raw data of absorbance OD517 investigation of DPPH scavenging assay

	Canola oil				
	20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml
1st	0.529	0.387	0.251	0.153	0.087
2nd	0.552	0.404	0.273	0.137	0.081
3rd	0.548	0.392	0.266	0.145	0.094
	corn oil				
1st	0.602	0.488	0.382	0.302	0.227
2nd	0.564	0.502	0.399	0.286	0.203
3rd	0.572	0.497	0.407	0.293	0.211
	Coconut oil				
1st	0.743	0.744	0.721	0.714	0.732
2nd	0.749	0.753	0.733	0.672	0.748
3rd	0.742	0.734	0.716	0.724	0.728
	Sunflower oil				
1st	0.574	0.416	0.272	0.188	0.072
2nd	0.558	0.432	0.298	0.204	0.086
3rd	0.580	0.422	0.279	0.192	0.083
	Mineral oil				
1st	0.806	0.794	0.802	0.808	0.824
2nd	0.826	0.816	0.788	0.813	0.807
3rd	0.797	0.821	0.812	0.796	0.810

Table9 Raw data of %inhibition in infused canola oil 24 and 48 hours

		24hours					48hours				
		20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml	20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml
10%	1st	0.558	0.422	0.262	0.191	0.130	0.573	0.410	0.227	0.129	0.099
	% inhibition	34.4	50.4	69.2	77.5	84.7	32.6	51.8	73.3	84.8	88.4
	2nd	0.601	0.415	0.266	0.156	0.115	0.552	0.396	0.210	0.140	0.106
	% inhibition	29.3	51.2	68.7	81.6	86.5	35.1	53.4	75.3	83.5	87.5
	3rd	0.580	0.420	0.278	0.185	0.122	0.560	0.425	0.236	0.136	0.092
	% inhibition	31.8	50.6	67.3	78.2	85.6	34.1	50.0	72.2	84.0	89.2
20%	1st	0.589	0.390	0.234	0.155	0.104	0.517	0.326	0.202	0.116	0.101
	% inhibition	30.7	54.1	72.5	81.8	87.8	39.2	61.6	76.2	86.4	88.1
	2nd	0.561	0.395	0.241	0.167	0.126	0.539	0.352	0.190	0.116	0.090
	% inhibition	34.0	53.5	71.6	80.4	85.2	36.6	58.6	77.6	86.4	89.4
	3rd	0.572	0.412	0.243	0.162	0.109	0.548	0.348	0.226	0.108	0.083
	% inhibition	32.7	51.5	71.4	80.9	87.2	35.5	59.1	73.4	87.3	90.2
30%	1st	0.534	0.400	0.222	0.144	0.102	0.545	0.330	0.160	0.078	0.088
	% inhibition	37.2	52.9	73.9	83.1	88.0	35.9	61.2	81.2	90.8	89.6
	2nd	0.562	0.389	0.231	0.163	0.093	0.530	0.342	0.176	0.089	0.072
	% inhibition	33.9	54.2	72.8	80.8	89.1	37.6	59.8	79.3	89.5	91.5
	3rd	0.549	0.395	0.216	0.151	0.106	0.542	0.326	0.163	0.096	0.069
	% inhibition	35.4	53.5	74.6	82.2	87.5	36.2	61.6	80.8	88.7	91.9

Table10 Raw data of %inhibition in infused canola oil 72 and 96 hours

		72hours					96hours				
		20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml	20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml
10%	1st	0.559	0.406	0.239	0.142	0.109	0.604	0.425	0.284	0.199	0.113
	% inhibition	34.2	52.2	71.9	83.3	87.2	28.9	50.0	66.6	76.6	86.7
	2nd	0.533	0.385	0.246	0.133	0.098	0.596	0.405	0.266	0.214	0.131
	% inhibition	37.3	54.7	71.1	84.4	88.5	29.9	52.4	68.7	74.8	84.6
	3rd	0.567	0.398	0.224	0.139	0.101	0.588	0.411	0.267	0.200	0.122
	% inhibition	33.3	53.2	73.6	83.6	88.1	30.8	51.6	68.6	76.5	85.6
20%	1st	0.543	0.353	0.196	0.112	0.100	0.592	0.425	0.234	0.153	0.110
	% inhibition	36.1	58.5	76.9	86.8	88.2	30.4	50.0	72.5	82.0	87.1
	2nd	0.561	0.321	0.201	0.101	0.097	0.611	0.399	0.251	0.143	0.102
	% inhibition	34.0	62.2	76.4	88.1	88.6	28.1	53.1	70.5	83.2	88.0
	3rd	0.558	0.344	0.226	0.126	0.105	0.620	0.381	0.251	0.166	0.116
	% inhibition	34.4	59.5	73.4	85.2	87.6	27.1	55.2	70.5	80.5	86.4
30%	1st	0.537	0.312	0.116	0.312	0.081	0.606	0.411	0.213	0.147	0.072
	% inhibition	36.8	63.3	86.4	63.3	90.5	28.7	51.6	74.9	82.7	91.5
	2nd	0.544	0.288	0.149	0.288	0.093	0.590	0.402	0.222	0.133	0.086
	% inhibition	36.0	66.1	82.5	66.1	89.1	30.6	52.7	73.9	84.4	89.9
	3rd	0.562	0.303	0.153	0.303	0.076	0.611	0.383	0.228	0.142	0.081
	% inhibition	33.9	64.4	82.0	64.4	91.1	28.1	54.9	73.2	83.3	90.5

Table11 Raw data of %inhibition in infused canola oil 120 hours

		120hours				
		20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml
10%	1st	0.566	0.425	0.324	0.207	0.140
	% inhibition	33.4	50.0	61.9	75.6	83.5
	2nd	0.581	0.444	0.303	0.190	0.121
	% inhibition	31.6	47.8	64.4	77.6	85.8
	3rd	0.563	0.448	0.291	0.216	0.131
	% inhibition	33.8	47.3	65.8	74.6	84.6
20%	1st	0.536	0.401	0.260	0.155	0.117
	% inhibition	36.9	52.8	69.4	81.8	86.2
	2nd	0.529	0.426	0.281	0.181	0.126
	% inhibition	37.8	49.9	66.9	78.7	85.2
	3rd	0.545	0.389	0.268	0.169	0.109
	% inhibition	35.9	54.2	68.5	80.1	87.2
30%	1st	0.506	0.306	0.222	0.095	0.058
	% inhibition	40.5	64.0	73.9	88.8	93.2
	2nd	0.522	0.323	0.236	0.108	0.062
	% inhibition	38.6	62.0	72.2	87.3	92.7
	3rd	0.518	0.333	0.219	0.090	0.076
	% inhibition	39.1	60.8	74.2	89.4	91.1

Table12 Raw data of %inhibition in infused corn oil 24 and 48 hours

		24hours					48hours				
		20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml	20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml
10%	1st	0.607	0.545	0.490	0.350	0.278	0.589	0.496	0.390	0.291	0.226
	% inhibition	28.6	35.9	42.4	58.8	67.3	30.7	41.6	54.1	65.8	73.4
	2nd	0.613	0.567	0.478	0.331	0.290	0.571	0.507	0.420	0.283	0.204
	% inhibition	27.9	33.3	43.8	61.1	65.9	32.8	40.4	50.6	66.7	76.0
	3rd	0.628	0.552	0.484	0.326	0.270	0.606	0.511	0.404	0.326	0.233
	% inhibition	26.1	35.1	43.1	61.6	68.2	28.7	39.9	52.5	61.6	72.6
20%	1st	0.581	0.498	0.461	0.323	0.256	0.572	0.449	0.371	0.307	0.187
	% inhibition	31.6	41.4	45.8	62.0	69.9	32.7	47.2	56.4	63.9	78.0
	2nd	0.599	0.518	0.455	0.301	0.263	0.588	0.432	0.355	0.281	0.166
	% inhibition	29.5	39.1	46.5	64.6	69.1	30.8	49.2	58.2	66.9	80.5
	3rd	0.590	0.526	0.462	0.312	0.244	0.566	0.448	0.369	0.267	0.181
	% inhibition	30.6	38.1	45.6	63.3	71.3	33.4	47.3	56.6	68.6	78.7
30%	1st	0.577	0.489	0.411	0.298	0.200	0.583	0.431	0.316	0.260	0.182
	% inhibition	32.1	42.5	51.6	64.9	76.5	31.4	49.3	62.8	69.4	78.6
	2nd	0.584	0.493	0.405	0.305	0.233	0.555	0.411	0.298	0.248	0.155
	% inhibition	31.3	42.0	52.4	64.1	72.6	34.7	51.6	64.9	70.8	81.8
	3rd	0.576	0.501	0.396	0.290	0.210	0.542	0.409	0.282	0.252	0.148
	% inhibition	32.2	41.1	53.4	65.9	75.3	36.2	51.9	66.8	70.4	82.6

Table13 Raw data of %inhibition in infused corn oil 72 and 96 hours

		72hours					96hours				
		20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml	20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml
10%	1st	0.556	0.469	0.381	0.289	0.216	0.520	0.470	0.334	0.266	0.184
	% inhibition	34.6	44.8	55.2	66.0	74.6	38.8	44.7	60.7	68.7	78.4
	2nd	0.546	0.458	0.369	0.299	0.201	0.549	0.452	0.351	0.236	0.167
	% inhibition	35.8	46.1	56.6	64.8	76.4	35.4	46.8	58.7	72.2	80.4
	3rd	0.558	0.477	0.374	0.274	0.211	0.553	0.462	0.322	0.241	0.163
	% inhibition	34.4	43.9	56.0	67.8	75.2	34.9	45.6	62.1	71.6	80.8
20%	1st	0.541	0.465	0.320	0.300	0.164	0.534	0.420	0.274	0.208	0.105
	% inhibition	36.4	45.3	62.4	64.7	80.7	37.2	50.6	67.8	75.5	87.6
	2nd	0.557	0.471	0.338	0.264	0.157	0.506	0.413	0.291	0.192	0.115
	% inhibition	34.5	44.6	60.2	68.9	81.5	40.5	51.4	65.8	77.4	86.5
	3rd	0.544	0.456	0.341	0.253	0.169	0.515	0.434	0.285	0.211	0.103
	% inhibition	36.0	46.4	59.9	70.2	80.1	39.4	48.9	66.5	75.2	87.9
30%	1st	0.523	0.401	0.289	0.265	0.141	0.518	0.399	0.255	0.149	0.104
	% inhibition	38.5	52.8	66.0	68.8	83.4	39.1	53.1	70.0	82.5	87.8
	2nd	0.538	0.422	0.281	0.232	0.137	0.523	0.421	0.235	0.132	0.109
	% inhibition	36.7	50.4	66.9	72.7	83.9	38.5	50.5	72.4	84.5	87.2
	3rd	0.540	0.438	0.297	0.235	0.162	0.527	0.407	0.241	0.158	0.093
	% inhibition	36.5	48.5	65.1	72.4	80.9	38.0	52.1	71.6	81.4	89.1

Table14 Raw data of %inhibition in infused corn oil 120 hours

		120hours				
		20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml
10%	1st	0.519	0.426	0.320	0.221	0.170
	% inhibition	38.9	49.9	62.4	74.0	80.0
	2nd	0.544	0.450	0.341	0.204	0.153
	% inhibition	36.0	47.1	59.9	76.0	82.0
	3rd	0.538	0.441	0.335	0.230	0.161
	% inhibition	36.7	48.1	60.6	72.9	81.1
20%	1st	0.508	0.356	0.188	0.140	0.102
	% inhibition	40.2	58.1	77.9	83.5	88.0
	2nd	0.512	0.331	0.221	0.149	0.082
	% inhibition	39.8	61.1	74.0	82.5	90.4
	3rd	0.523	0.360	0.206	0.158	0.077
	% inhibition	38.5	57.6	75.8	81.4	90.9
30%	1st	0.519	0.360	0.200	0.131	0.092
	% inhibition	38.9	57.6	76.5	84.6	89.2
	2nd	0.490	0.352	0.191	0.140	0.071
	% inhibition	42.4	58.6	77.5	83.5	91.6
	3rd	0.507	0.323	0.182	0.128	0.060
	% inhibition	40.4	62.0	78.6	84.9	92.9

Table15 Raw data of %inhibition in infused coconut oil 24 and 48 hours

		24hours					48hours				
		20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml	20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml
10%	1st	0.797	0.792	0.771	0.724	0.701	0.808	0.750	0.738	0.694	0.673
	% inhibition	6.2	6.8	9.3	14.8	17.5	4.9	11.8	13.2	18.4	20.8
	2nd	0.816	0.799	0.768	0.734	0.698	0.815	0.769	0.742	0.706	0.686
	% inhibition	4.0	6.0	9.6	13.6	17.9	4.1	9.5	12.7	16.9	19.3
	3rd	0.826	0.801	0.761	0.738	0.685	0.821	0.745	0.747	0.711	0.672
	% inhibition	2.8	5.8	10.5	13.2	19.4	3.4	12.4	12.1	16.4	20.9
20%	1st	0.779	0.723	0.788	0.689	0.712	0.775	0.703	0.717	0.675	0.656
	% inhibition	8.4	14.9	7.3	18.9	16.2	8.8	17.3	15.6	20.6	22.8
	2nd	0.791	0.733	0.767	0.706	0.703	0.783	0.694	0.725	0.683	0.666
	% inhibition	6.9	13.8	9.8	16.9	17.3	7.9	18.4	14.7	19.6	21.6
	3rd	0.802	0.745	0.777	0.714	0.684	0.771	0.712	0.735	0.679	0.654
	% inhibition	5.6	12.4	8.6	16.0	19.5	9.3	16.2	13.5	20.1	23.1
30%	1st	0.789	0.721	0.756	0.701	0.685	0.738	0.699	0.685	0.651	0.643
	% inhibition	7.2	15.2	11.1	17.5	19.4	13.2	17.8	19.4	23.4	24.4
	2nd	0.792	0.705	0.761	0.689	0.693	0.744	0.715	0.690	0.630	0.638
	% inhibition	6.8	17.1	10.5	18.9	18.5	12.5	15.9	18.8	25.9	24.9
	3rd	0.803	0.716	0.746	0.709	0.689	0.731	0.727	0.708	0.647	0.651
	% inhibition	5.5	15.8	12.2	16.6	18.9	14.0	14.5	16.7	23.9	23.4

Table16 Raw data of %inhibition in infused coconut oil 72 and 96 hours

		72hours					96hours				
		20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml	20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml
10%	1st	0.810	0.756	0.710	0.685	0.656	0.751	0.700	0.666	0.655	0.651
	% inhibition	4.7	11.1	16.5	19.4	22.8	11.6	17.6	21.6	22.9	23.4
	2nd	0.826	0.742	0.732	0.702	0.641	0.733	0.712	0.671	0.661	0.632
	% inhibition	2.8	12.7	13.9	17.4	24.6	13.8	16.2	21.1	22.2	25.6
	3rd	0.804	0.748	0.727	0.691	0.648	0.759	0.696	0.661	0.645	0.638
	% inhibition	5.4	12.0	14.5	18.7	23.8	10.7	18.1	22.2	24.1	24.9
20%	1st	0.751	0.700	0.646	0.639	0.619	0.719	0.697	0.629	0.636	0.602
	% inhibition	11.6	17.6	24.0	24.8	27.2	15.4	18.0	26.0	25.2	29.2
	2nd	0.733	0.712	0.658	0.648	0.630	0.704	0.703	0.637	0.630	0.590
	% inhibition	13.8	16.2	22.6	23.8	25.9	17.2	17.3	25.1	25.9	30.6
	3rd	0.759	0.696	0.641	0.650	0.625	0.722	0.689	0.640	0.642	0.617
	% inhibition	10.7	18.1	24.6	23.5	26.5	15.1	18.9	24.7	24.5	27.4
30%	1st	0.722	0.681	0.601	0.623	0.559	0.713	0.633	0.596	0.604	0.519
	% inhibition	15.1	19.9	29.3	26.7	34.2	16.1	25.5	29.9	28.9	38.9
	2nd	0.725	0.674	0.591	0.642	0.561	0.694	0.629	0.581	0.617	0.532
	% inhibition	14.7	20.7	30.5	24.5	34.0	18.4	26.0	31.6	27.4	37.4
	3rd	0.737	0.677	0.622	0.633	0.584	0.703	0.616	0.589	0.609	0.541
	% inhibition	13.3	20.4	26.8	25.5	31.3	17.3	27.5	30.7	28.4	36.4

Table17 Raw data of %inhibition in infused coconut oil 120 hours

		120hours				
		20mg/ml	40mg/ml	60mg/ml	80mg/ml	20mg/ml
10%	1st	0.707	0.687	0.673	0.629	0.707
	% inhibition	16.8	19.2	20.8	26.0	16.8
	2nd	0.699	0.690	0.656	0.651	0.699
	% inhibition	17.8	18.8	22.8	23.4	17.8
	3rd	0.711	0.703	0.661	0.640	0.711
	% inhibition	16.4	17.3	22.2	24.7	16.4
20%	1st	0.672	0.689	0.646	0.599	0.544
	% inhibition	20.9	18.9	24.0	29.5	36.0
	2nd	0.698	0.700	0.658	0.612	0.538
	% inhibition	17.9	17.6	22.6	28.0	36.7
	3rd	0.701	0.695	0.641	0.603	0.551
	% inhibition	17.5	18.2	24.6	29.1	35.2
30%	1st	0.683	0.602	0.550	0.489	0.441
	% inhibition	19.6	29.2	35.3	42.5	48.1
	2nd	0.667	0.598	0.570	0.502	0.428
	% inhibition	21.5	29.6	32.9	40.9	49.6
	3rd	0.685	0.616	0.561	0.485	0.433
	% inhibition	19.4	27.5	34.0	42.9	49.1

Table18 Raw data of %inhibition in infused sunflower oil 24 and 48 hours

		24hours					48hours				
		20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml	20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml
10%	1st	0.611	0.467	0.314	0.209	0.155	0.600	0.410	0.263	0.144	0.103
	% inhibition	28.1	45.1	63.1	75.4	81.8	29.4	51.8	69.1	83.1	87.9
	2nd	0.586	0.480	0.320	0.212	0.162	0.590	0.403	0.261	0.151	0.109
	% inhibition	31.1	43.5	62.4	75.1	80.9	30.6	52.6	69.3	82.2	87.2
	3rd	0.603	0.483	0.321	0.198	0.159	0.593	0.414	0.269	0.149	0.111
	% inhibition	29.1	43.2	62.2	76.7	81.3	30.2	51.3	68.4	82.5	86.9
20%	1st	0.599	0.450	0.289	0.200	0.148	0.584	0.380	0.242	0.108	0.084
	% inhibition	29.5	47.1	66.0	76.5	82.6	31.3	55.3	71.5	87.3	90.1
	2nd	0.602	0.444	0.293	0.192	0.142	0.590	0.385	0.235	0.129	0.080
	% inhibition	29.2	47.8	65.5	77.4	83.3	30.6	54.7	72.4	84.8	90.6
	3rd	0.611	0.467	0.288	0.186	0.152	0.598	0.392	0.239	0.127	0.074
	% inhibition	28.1	45.1	66.1	78.1	82.1	29.6	53.9	71.9	85.1	91.3
30%	1st	0.587	0.433	0.276	0.185	0.122	0.570	0.369	0.237	0.102	0.072
	% inhibition	30.9	49.1	67.5	78.2	85.6	32.9	56.6	72.1	88.0	91.5
	2nd	0.592	0.441	0.279	0.178	0.128	0.582	0.381	0.228	0.115	0.078
	% inhibition	30.4	48.1	67.2	79.1	84.9	31.5	55.2	73.2	86.5	90.8
	3rd	0.600	0.438	0.284	0.188	0.125	0.585	0.386	0.240	0.110	0.081
	% inhibition	29.4	48.5	66.6	77.9	85.3	31.2	54.6	71.8	87.1	90.5

Table19 Raw data of %inhibition in infused sunflower oil 72 and 96 hours

		72hours					96hours				
		20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml	20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml
10%	1st	0.605	0.432	0.258	0.191	0.086	0.577	0.468	0.289	0.182	0.093
	% inhibition	28.8	49.2	69.6	77.5	89.9	32.1	44.9	66.0	78.6	89.1
	2nd	0.618	0.451	0.260	0.179	0.099	0.602	0.471	0.301	0.201	0.101
	% inhibition	27.3	46.9	69.4	78.9	88.4	29.2	44.6	64.6	76.4	88.1
	3rd	0.585	0.450	0.241	0.187	0.103	0.594	0.470	0.286	0.196	0.103
	% inhibition	31.2	47.1	71.6	78.0	87.9	30.1	44.7	66.4	76.9	87.9
20%	1st	0.549	0.412	0.194	0.108	0.089	0.571	0.416	0.301	0.180	0.088
	% inhibition	35.4	51.5	77.2	87.3	89.5	32.8	51.1	64.6	78.8	89.6
	2nd	0.561	0.392	0.198	0.123	0.082	0.552	0.402	0.259	0.174	0.094
	% inhibition	34.0	53.9	76.7	85.5	90.4	35.1	52.7	69.5	79.5	88.9
	3rd	0.566	0.397	0.202	0.135	0.086	0.574	0.416	0.261	0.171	0.087
	% inhibition	33.4	53.3	76.2	84.1	89.9	32.5	51.1	69.3	79.9	89.8
30%	1st	0.565	0.370	0.199	0.115	0.084	0.545	0.400	0.240	0.175	0.073
	% inhibition	33.5	56.5	76.6	86.5	90.1	35.9	52.9	71.8	79.4	91.4
	2nd	0.533	0.355	0.202	0.111	0.076	0.538	0.377	0.222	0.167	0.079
	% inhibition	37.3	58.2	76.2	86.9	91.1	36.7	55.6	73.9	80.4	90.7
	3rd	0.543	0.361	0.197	0.109	0.072	0.540	0.382	0.236	0.181	0.063
	% inhibition	36.1	57.5	76.8	87.2	91.5	36.5	55.1	72.2	78.7	92.6

Table20 Raw data of %inhibition in infused sunflower oil 120 hours

		120hours				
		20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml
10%	1st	0.548	0.398	0.224	0.134	0.050
	% inhibition	35.5	53.2	73.6	84.2	94.1
	2nd	0.558	0.388	0.217	0.130	0.055
	% inhibition	34.4	54.4	74.5	84.7	93.5
	3rd	0.544	0.391	0.220	0.139	0.053
	% inhibition	36.0	54.0	74.1	83.6	93.8
20%	1st	0.559	0.376	0.186	0.131	0.045
	% inhibition	34.2	55.8	78.1	84.6	94.7
	2nd	0.564	0.371	0.197	0.139	0.051
	% inhibition	33.6	56.4	76.8	83.6	94.0
	3rd	0.561	0.383	0.192	0.142	0.048
	% inhibition	34.0	54.9	77.4	83.3	94.4
30%	1st	0.519	0.344	0.184	0.138	0.043
	% inhibition	38.9	59.5	78.4	83.8	94.9
	2nd	0.526	0.349	0.166	0.129	0.048
	% inhibition	38.1	58.9	80.5	84.8	94.4
	3rd	0.521	0.351	0.169	0.135	0.044
	% inhibition	38.7	58.7	80.1	84.1	94.8

Table21 Raw data of %inhibition in infused mineral oil 24 and 48 hours

		24hours					48hours				
		20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml	20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml
10%	1st	0.834	0.833	0.834	0.834	0.822	0.805	0.800	0.775	0.795	0.800
	% inhibition	1.9	2.0	1.9	1.9	3.3	5.3	5.9	8.8	6.5	5.9
	2nd	0.840	0.829	0.824	0.830	0.834	0.818	0.814	0.793	0.811	0.795
	% inhibition	1.2	2.5	3.1	2.4	1.9	3.8	4.2	6.7	4.6	6.5
	3rd	0.821	0.826	0.831	0.820	0.821	0.823	0.822	0.801	0.805	0.803
	% inhibition	0.832	0.829	0.830	0.828	0.826	0.815	0.812	0.790	0.804	0.799
20%	1st	0.844	0.803	0.833	0.826	0.816	0.807	0.782	0.801	0.794	0.784
	% inhibition	0.7	5.5	2.0	2.8	4.0	5.1	8.0	5.8	6.6	7.8
	2nd	0.830	0.821	0.832	0.831	0.819	0.813	0.805	0.812	0.783	0.794
	% inhibition	2.4	3.4	2.1	2.2	3.6	4.4	5.3	4.5	7.9	6.6
	3rd	0.829	0.830	0.835	0.835	0.824	0.817	0.816	0.795	0.799	0.804
	% inhibition	2.5	2.4	1.8	1.8	3.1	3.9	4.0	6.5	6.0	5.4
30%	1st	0.827	0.822	0.814	0.819	0.808	0.801	0.811	0.801	0.804	0.767
	% inhibition	2.7	3.3	4.2	3.6	4.9	5.8	4.6	5.8	5.4	9.8
	2nd	0.819	0.810	0.817	0.823	0.815	0.795	0.827	0.807	0.785	0.788
	% inhibition	3.6	4.7	3.9	3.2	4.1	6.5	2.7	5.1	7.6	7.3
	3rd	0.830	0.815	0.824	0.820	0.817	0.813	0.825	0.808	0.783	0.782
	% inhibition	2.4	4.1	3.1	3.5	3.9	4.4	2.9	4.9	7.9	8.0

Table22 Raw data of %inhibition in infused mineral oil 72 and 96 hours

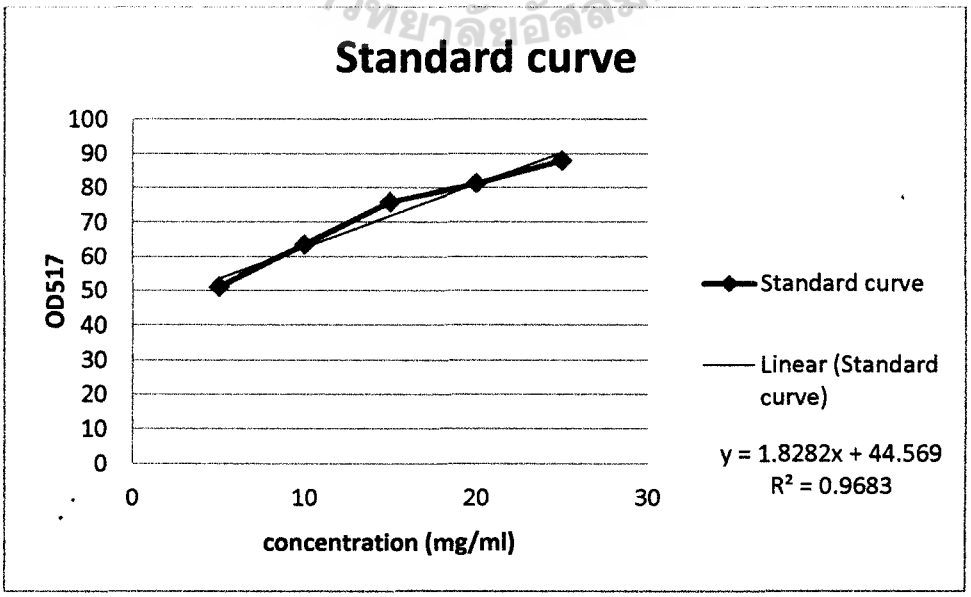
		72hours					96hours				
		20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml	20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml
10%	1st	0.819	0.801	0.788	0.802	0.777	0.803	0.801	0.776	0.799	0.786
	% inhibition	3.6	5.8	7.3	5.6	8.6	5.5	5.8	8.7	6.0	7.5
	2nd	0.823	0.807	0.792	0.790	0.785	0.790	0.812	0.794	0.804	0.789
	% inhibition	3.2	5.1	6.8	7.1	7.6	7.1	4.5	6.6	5.4	7.2
	3rd	0.820	0.813	0.791	0.793	0.772	0.801	0.796	0.796	0.811	0.792
	% inhibition	0.821	0.807	0.790	0.795	0.778	0.798	0.803	0.789	0.805	0.789
20%	1st	0.801	0.794	0.800	0.784	0.760	0.803	0.785	0.801	0.780	0.765
	% inhibition	5.8	6.6	5.9	7.8	10.6	5.5	7.6	5.8	8.2	10.0
	2nd	0.797	0.791	0.814	0.788	0.764	0.808	0.791	0.812	0.790	0.768
	% inhibition	6.2	6.9	4.2	7.3	10.1	4.9	6.9	4.5	7.1	9.6
	3rd	0.793	0.805	0.799	0.792	0.772	0.809	0.796	0.799	0.792	0.770
	% inhibition	6.7	5.3	6.0	6.8	9.2	4.8	6.4	6.0	6.8	9.4
30%	1st	0.803	0.789	0.779	0.745	0.769	0.804	0.770	0.782	0.789	0.755
	% inhibition	5.5	7.2	8.4	12.4	9.5	5.4	9.4	8.0	7.2	11.2
	2nd	0.813	0.784	0.776	0.755	0.778	0.788	0.777	0.801	0.780	0.780
	% inhibition	4.4	7.8	8.7	11.2	8.5	7.3	8.6	5.8	8.2	8.2
	3rd	0.809	0.792	0.781	0.748	0.772	0.794	0.785	0.795	0.774	0.766
	% inhibition	4.8	6.8	8.1	12.0	9.2	6.6	7.6	6.5	8.9	9.9

Table23 Raw data of %inhibition in infused mineral oil 120 hours

		120hours				
		20mg/ml	40mg/ml	60mg/ml	80mg/ml	100mg/ml
10%	1st	0.812	0.805	0.796	0.781	0.778
	% inhibition	4.5	5.3	6.4	8.1	8.5
	2nd	0.808	0.798	0.794	0.785	0.775
	% inhibition	4.9	6.1	6.6	7.6	8.8
	3rd	0.798	0.802	0.790	0.788	0.772
	% inhibition	0.806	0.802	0.793	0.785	0.775
20%	1st	0.808	0.795	0.793	0.787	0.756
	% inhibition	4.9	6.5	6.7	7.4	11.1
	2nd	0.803	0.800	0.787	0.750	0.776
	% inhibition	5.5	5.9	7.4	11.8	8.7
	3rd	0.807	0.791	0.781	0.773	0.763
	% inhibition	5.1	6.9	8.1	9.1	10.2
30%	1st	0.786	0.803	0.790	0.777	0.759
	% inhibition	7.6	5.5	7.1	8.6	10.7
	2nd	0.804	0.811	0.784	0.773	0.765
	% inhibition	5.4	4.6	7.8	9.1	10.0
	3rd	0.797	0.792	0.789	0.786	0.758
	% inhibition	6.2	6.8	7.2	7.5	10.8

Table24 Standard curve of ascorbic acid investigation of DPPH scavenging assay

OD517	5ug/ml	10ug/ml	15ug/ml	20ug/ml	25ug/ml
1st	0.418	0.312	0.212	0.178	0.094
2nd	0.412	0.306	0.195	0.136	0.109
3rd	0.416	0.309	0.208	0.164	0.102
Average	0.415	0.309	0.205	0.159	0.102



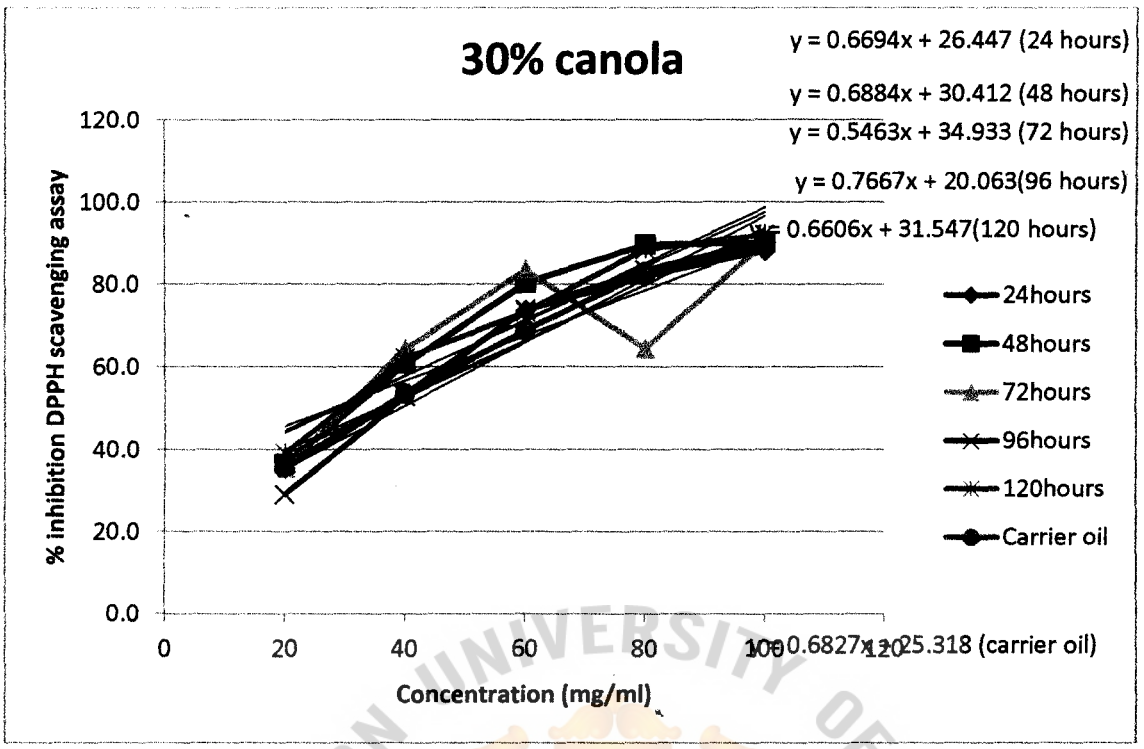


Figure1 Linear equation calculated for IC50 value of carrier oil and 30% infused canola oil

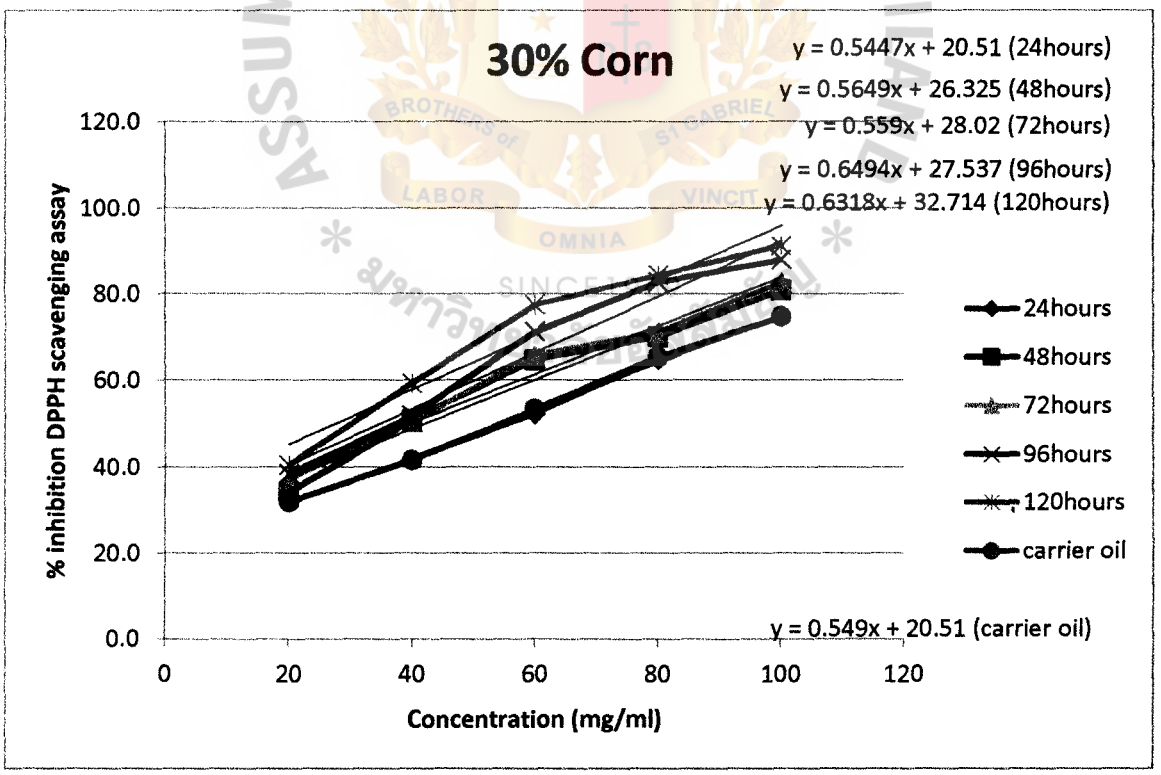


Figure2 Linear equation calculated for IC50 value of carrier oil and 30% infused corn oil

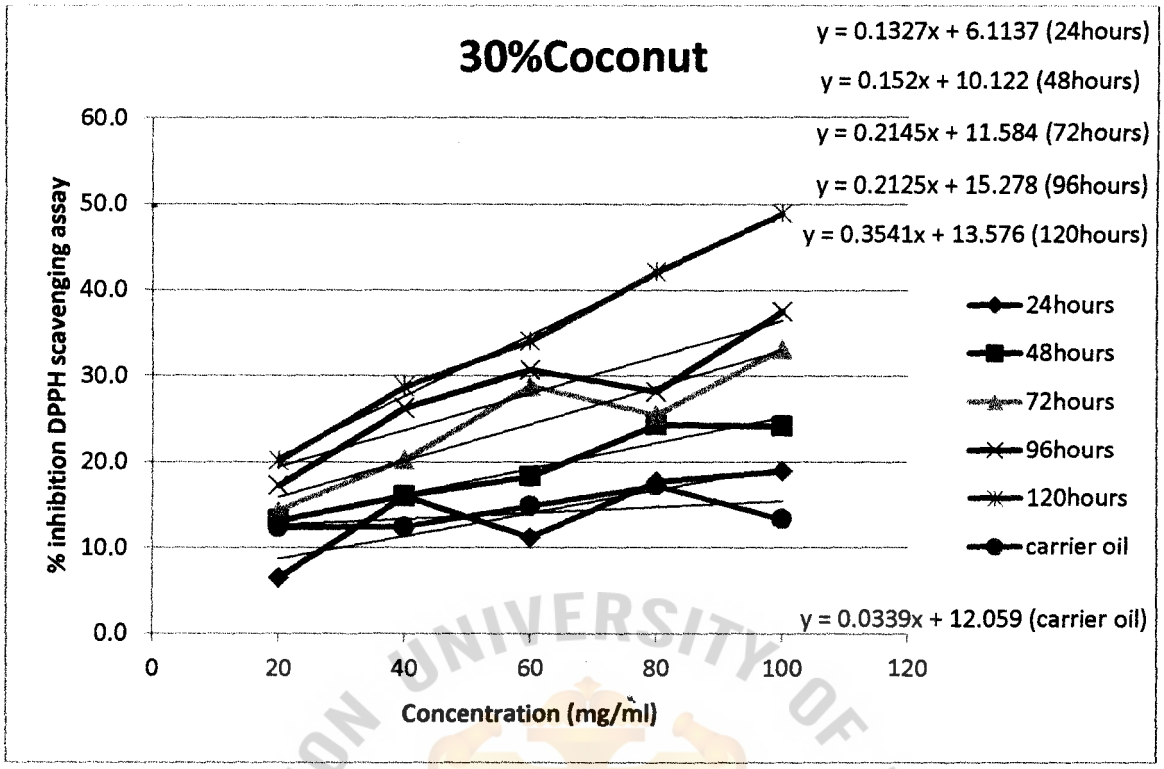


Figure3 Linear equation calculated for IC50 value of carrier oil and 30% infused coconut oil

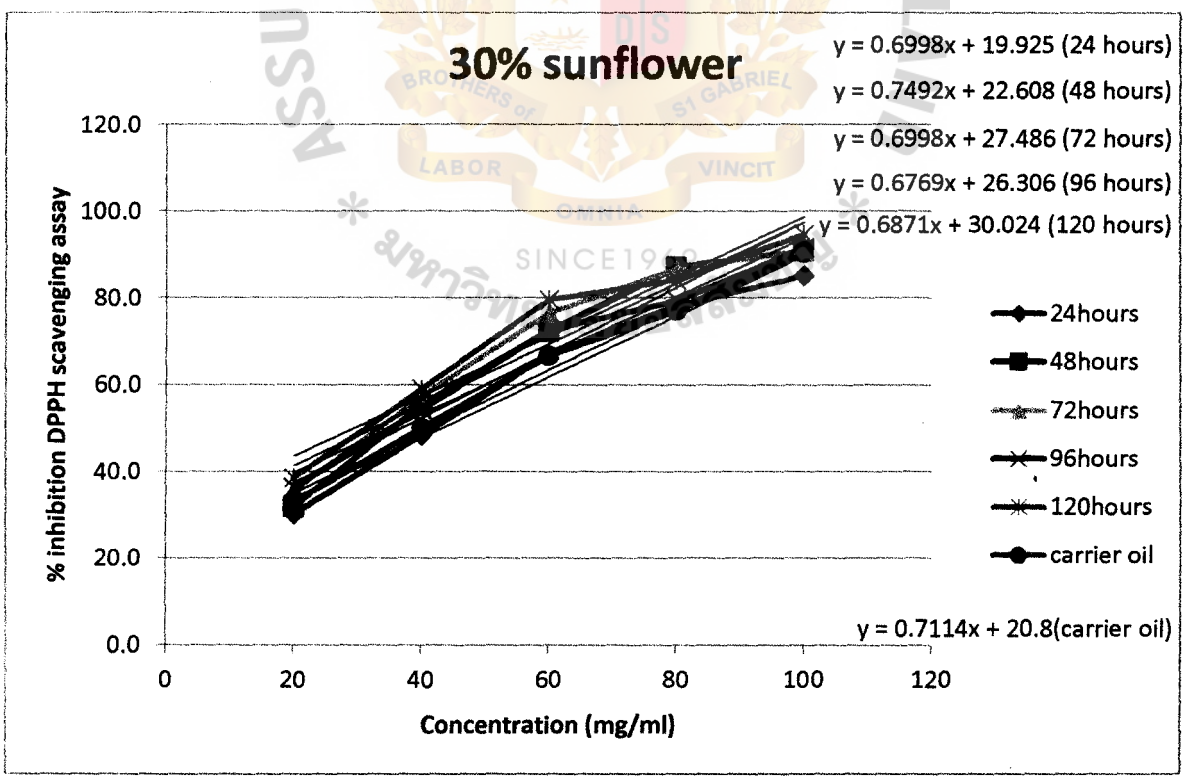


Figure4 Linear equation calculated for IC50 value of carrier oil and 30% infused sunflower oil

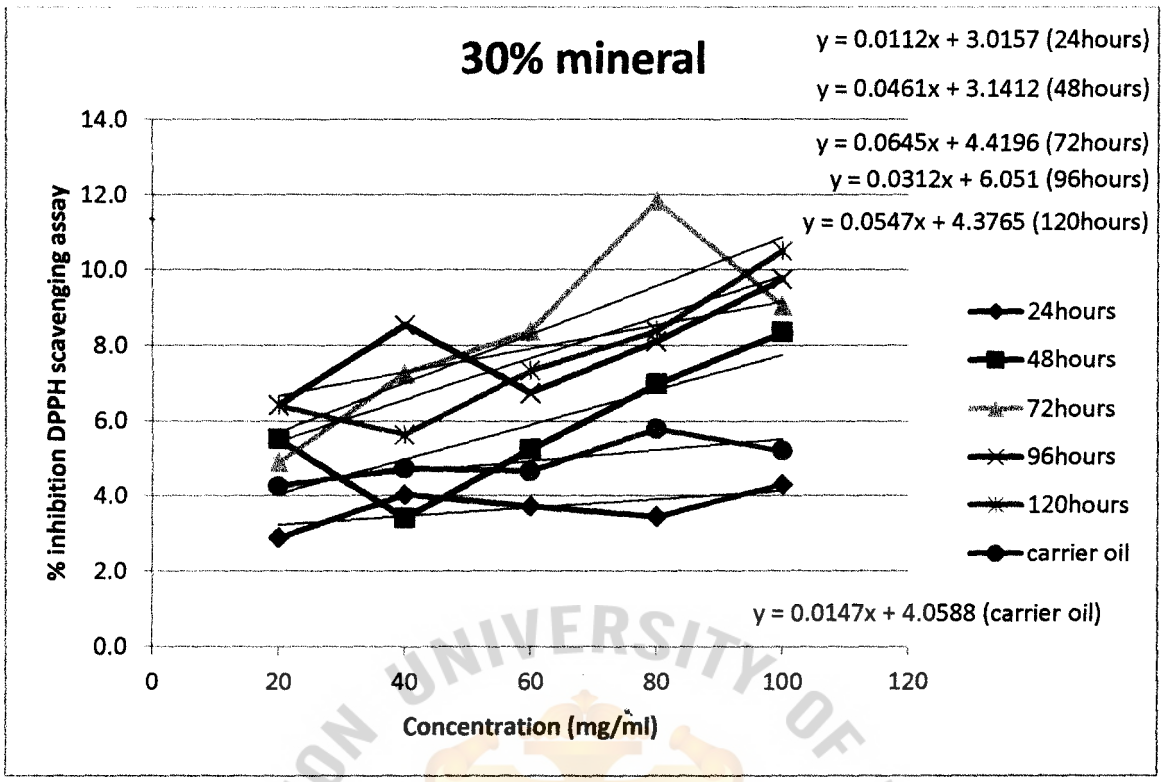


Figure5 Linear equation calculated for IC50 value of carrier oil and 30% infused mineral oil

3. Color increasing from carrier oils

Table25 absorbance value of all carrier oils OD420

	canola	corn	coconut	sunflower	mineral
1st	0.206	0.442	0.019	0.076	0
2nd	0.186	0.412	0.010	0.068	0
3rd	0.197	0.425	0.014	0.072	0
Average	0.196	0.426	0.014	0.072	0

Table26 Absorbance value of canola oil OD420

Canola					
	24hours	48hours	72hours	96hours	120hours
10%	0.638	0.724	0.854	0.926	1.132
	0.698	0.744	0.881	0.964	1.201
	0.656	0.767	0.834	1.045	1.164
Average	0.664	0.745	0.856	0.978	1.166
20%	1.053	1.082	1.153	1.358	1.473
	0.980	1.135	1.182	1.382	1.539
	1.031	1.065	1.204	1.377	1.422
Average	1.021	1.094	1.180	1.372	1.478
30%	1.399	1.542	1.589	1.693	1.804
	1.423	1.523	1.534	1.710	1.756
	1.384	1.582	1.564	1.634	1.759
Average	1.402	1.549	1.562	1.679	1.773

Table27 Absorbance value of corn oil OD420

Corn					
	24hours	48hours	72hours	96hours	120hours
10%	0.638	0.621	0.894	0.934	1.084
	0.602	0.641	0.845	1.040	1.123
	0.611	0.618	0.872	0.973	1.056
Average	0.617	0.627	0.870	0.982	1.088
20%	0.746	0.876	1.223	1.302	1.608
	0.804	0.904	1.193	1.256	1.590
	0.766	0.853	1.204	1.271	1.623
Average	0.772	0.878	1.207	1.276	1.607
30%	1.126	1.090	1.589	1.683	1.952
	1.194	0.945	1.534	1.715	1.822
	1.085	1.021	1.522	1.677	1.874
Average	1.135	1.019	1.548	1.692	1.883

Table28 Absorbance value of coconut oil OD420

coconut					
	24hours	48hours	72hours	96hours	120hours
10%	0.485	0.534	0.642	0.870	1.045
	0.451	0.561	0.649	0.851	0.973
	0.502	0.575	0.662	0.902	1.010
Average	0.479	0.557	0.651	0.874	1.009
20%	0.815	0.893	1.032	1.272	1.346
	0.784	0.856	1.078	1.204	1.398
	0.803	0.906	1.093	1.277	1.371
Average	0.801	0.885	1.068	1.251	1.372
30%	1.284	1.302	1.487	1.543	1.694
	1.266	1.295	1.417	1.584	1.721
	1.238	1.321	1.466	1.559	1.653
Average	1.263	1.306	1.457	1.562	1.689

Table29 Absorbance value of sunflower oil OD420

sunflower					
	24hours	48hours	72hours	96hours	120hours
10%	0.263	0.287	0.473	0.534	0.783
	0.273	0.291	0.480	0.523	0.790
	0.260	0.296	0.479	0.541	0.765
Average	0.265	0.291	0.477	0.533	0.779
20%	0.367	0.392	0.593	0.862	1.182
	0.381	0.388	0.601	0.877	1.134
	0.372	0.395	0.588	0.860	1.180
Average	0.373	0.392	0.594	0.866	1.165
30%	0.768	0.824	1.104	1.294	1.532
	0.781	0.808	1.094	1.309	1.573
	0.763	0.812	1.121	1.316	1.522
Average	0.771	0.815	1.106	1.306	1.542

Table30 Absorbance value of mineral oil OD420

mineral					
	24hours	48hours	72hours	96hours	120hours
10%	0.099	0.093	0.193	0.227	0.285
	0.103	0.109	0.206	0.244	0.291
	0.097	0.098	0.189	0.230	0.288
Average	0.100	0.100	0.196	0.234	0.288
20%	0.212	0.343	0.387	0.412	0.453
	0.278	0.337	0.401	0.427	0.478
	0.245	0.340	0.396	0.422	0.466
Average	0.245	0.340	0.395	0.420	0.466
30%	0.525	0.581	0.670	0.694	0.735
	0.508	0.587	0.662	0.709	0.747
	0.511	0.590	0.683	0.685	0.730
Average	0.515	0.586	0.672	0.696	0.737

4. Peroxide value

Table31 Usage volume of sodium thiosulfate

infused oil	Control	30% infused
	Used sodium thiosulfate	Used sodium thiosulfate
canola	0.5	0.8
corn	0.2	0.4
coconut	0.1	0.1
sunflower	1.1	1.8
mineral	0	0

II. Statistic analyzed

1. % total phenolic increase

Table32 summary(Fact.RCBD) canola oil

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
A	**4**	**3435**	**859**	**15.447**	**3.73e-06 *** **
B	**1**	**4772**	**4772**	**85.843**	**4.75e-09 *** **
rep	**1**	**183**	**183**	**3.294**	**0.0832 .**
A:B	**4**	**408**	**102**	**1.836**	**0.1579**
Residuals	**22**	**1223**	**56**		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Table33 summary(Fact.RCBD) corn oil

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
A	**4**	**3534**	**884**	**27.861**	**4.58e-09 *** **
B	**1**	**3638**	**3638**	**114.709**	**4.98e-11 *** **
rep	**1**	**1**	**1**	**0.017**	**0.89615**
A:B	**4**	**739**	**185**	**5.828**	**0.00174 ** **
Residuals	**26**	**825**	**32**		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Table34 summary(Fact.RCBD) coconut oil

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
A	**4**	**1563**	**391**	**15.768**	**1.14e-06 *** **
B	**1**	**6465**	**6465**	**260.808**	**4.52e-15 *** **
rep	**1**	**25**	**25**	**1.025**	**0.3207**
A:B	**4**	**245**	**61**	**2.472**	**0.0694 .**
Residuals	**26**	**645**	**25**		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Table35 summary(Fact.RCBD) sunflower oil

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
A	**4**	**2244.3**	**561.1**	**29.755**	**5.48e-09 *** **
B	**1**	**2588.5**	**2588.5**	**137.276**	**2.05e-11 *** **
rep	**1**	**10.7**	**10.7**	**0.568**	**0.4584**
A:B	**4**	**241.5**	**60.4**	**3.202**	**0.0305 * **
Residuals	**24**	**452.6**	**18.9**		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Table36 summary(Fact.RCBD) mineral oil

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
A	**4**	**2365.5**	**591.4**	**41.686**	**3.52e-11 *** **
B	**1**	**359.2**	**359.2**	**25.317**	**2.80e-05 *** **
rep	**1**	**22.3**	**22.3**	**1.568**	**0.221177**
A:B	**4**	**474.9**	**118.7**	**8.369**	**0.000158 *** **
Residuals	**27**	**383.0**	**14.2**		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Table37 summary(Fact.RCBD) 30% infusion ratio of all infused oils

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
A	**4**	**3589**	**897.2**	**37.452**	**1.19e-10 *** **
B	**4**	**4721**	**1180.3**	**49.270**	**5.04e-12 *** **
rep	**1**	**25**	**25.3**	**1.055**	**0.314**
A:B	**16**	**2389**	**149.3**	**6.234**	**1.75e-05 *** **
Residuals	**27**	**647**	**24.0**		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Table38 Duncan multiple comparison on effect of 24hours infusion period on every infused oils

	24hrs	std.err	replication
canola	**28.12887**	**9.6381864**	**2**
coconut	**36.04064**	**3.0375695**	**3**
corn	**14.36003**	**3.3438606**	**2**
mineral	**20.06140**	**0.2192982**	**2**
sunflower	**24.93983**	**1.8028835**	**2**

alpha: 0.05 ; Df Error: 6
Critical Range

2	3	4	5
15.86103	**16.43875**	**16.72492**	**16.86836**

Harmonic Mean of Cell Sizes 2.142857
Different value for each comparison
Means with the same letter are not significantly different.
Groups, Treatments and means

a	**coconut**	**36.04**
ab	**canola**	**28.13**
ab	**sunflower**	**24.94**
ab	**mineral**	**20.06**
b	**corn**	**14.36**

> duncan.test(model,"trt",alpha=0.05,console=TRUE)
Error in duncan.test(model, "trt", alpha = 0.05, console = TRUE) :
unused argument(s) (console = TRUE)

Table39 Duncan multiple comparison on effect of 48hours infusion period on every infused oils

	48hrs	std.err	replication
canola	**29.70245**	**0.9834884**	**2**
coconut	**45.84907**	**3.4215453**	**2**
corn	**19.47417**	**0.5900930**	**2**
mineral	**20.45614**	**0.0877193**	**2**
sunflower	**26.96807**	**1.1268022**	**2**

alpha: 0.05 ; Df Error: 5
Critical Range

2	3	4	5
6.147896	**6.339247**	**6.420346**	**6.449503**

Means with the same letter are not significantly different.
Groups, Treatments and means

a	**coconut**	**45.85**
b	**canola**	**29.7**
b	**sunflower**	**26.97**
c	**mineral**	**20.46**
c	**corn**	**19.47**

> duncan.test(model,"trt",alpha=0.05,console=TRUE)
Error in duncan.test(model, "trt", alpha = 0.05, console = TRUE) :
unused argument(s) (console = TRUE)

Table40 Duncan multiple comparison on effect of 72hours infusion period on every infused oils

	72hrs	std.err	replication
canola	**55.20758**	**5.0088224**	**3**
coconut	**59.00301**	**2.6514081**	**3**
corn	**33.24301**	**0.9834884**	**2**
mineral	**20.14912**	**0.1315789**	**2**
sunflower	**33.72889**	**2.9296857**	**2**

alpha: 0.05 ; Df Error: 7

Critical Range

2	3	4	5
12.11011	**12.59202**	**12.84948**	**12.99461**

Harmonic Mean of Cell Sizes 2.307692

Different value for each comparison
Means with the same letter are not significantly different.
Groups, Treatments and means

a	**coconut**	**59**
a	**canola**	**55.21**
b	**sunflower**	**33.73**
b	**corn**	**33.24**
c	**mineral**	**20.15**

> duncan.test(model,"trt",alpha=0.05,console=TRUE)
Error in duncan.test(model, "trt", alpha = 0.05, console = TRUE) :
unused argument(s) (console = TRUE)

Table41 Duncan multiple comparison on effect of 96hours infusion period on every infused oils

	96hrs	std.err	replication
canola	**49.56892**	**3.9339536**	**2**
coconut	**49.49872**	**5.2463695**	**2**
corn	**38.55385**	**4.3273490**	**2**
mineral	**20.41228**	**0.3947368**	**2**
sunflower	**44.99691**	**2.0282440**	**2**

alpha: 0.05 ; Df Error: 5

Critical Range

2	3	4	5
13.20748	**13.61856**	**13.79278**	**13.85542**

Means with the same letter are not significantly different.

Groups, Treatments and means

a	**canola**	**49.57**
a	**coconut**	**49.5**
a	**sunflower**	**45**
a	**corn**	**38.55**
b	**mineral**	**20.41**

> duncan.test(model,"trt",alpha=0.05,console=TRUE)
Error in duncan.test(model, "trt", alpha = 0.05, console = TRUE) :

unused argument(s) (console = TRUE

Table42 Duncan multiple comparison on effect of 120 hours infusion period on every infused oils

	120hrs	std.err	replication
canola	**43.47129**	**0.1966977**	**2**
coconut	**51.09544**	**2.7372363**	**2**
corn	**59.40380**	**2.7537675**	**2**
mineral	**22.34211**	**0.4824561**	**2**
sunflower	**56.03957**	**1.8028835**	**2**

alpha: 0.05 ; Df Error: 5
Critical Range

2	3	4	5
7.011148	**7.229367**	**7.321854**	**7.355105**

Means with the same letter are not significantly different.

Groups, Treatments and means

a	**corn**	**59.4**
ab	**sunflower**	**56.04**
b	**coconut**	**51.1**
c	**canola**	**43.47**
d	**mineral**	**22.34**

> duncan.test(model,"trt",alpha=0.05,console=TRUE)
Error in duncan.test(model, "trt", alpha = 0.05, console = TRUE) :
unused argument(s) (console = TRUE)

Table43 Duncan multiple comparison on %TPC increased of 30%infused canola oil on every infusion ratio

	canola	std.err	replication
120hrs	**43.47129**	**0.1966977**	**2**
24hrs	**28.12887**	**9.6381864**	**2**
48hrs	**29.70245**	**0.9834884**	**2**
72hrs	**55.20758**	**5.0088224**	**3**
96hrs	**49.56892**	**3.9339536**	**2**

alpha: 0.05 ; Df Error: 6
Critical Range

2	3	4	5
18.54559	**19.22109**	**19.55570**	**19.72341**

Harmonic Mean of Cell Sizes 2.142857

Different value for each comparison
Means with the same letter are not significantly different.
Groups, Treatments and means

a	**72hrs**	**55.21**
a	**96hrs**	**49.57**
ab	**120hrs**	**43.47**
b	**48hrs**	**29.7**
b	**24hrs**	**28.13**

> duncan.test(model,"trt",alpha=0.05,console=TRUE)
Error in duncan.test(model, "trt", alpha = 0.05, console = TRUE) :
unused argument(s) (console = TRUE

Table44 Duncan multiple comparison on %TPC increased of 30%infused corn oil on every infusion ratio

	corn	std.err	replication
120hrs	**59.40380**	**2.7537675**	**2**
24hrs	**14.36003**	**3.3438606**	**2**
48hrs	**19.47417**	**0.5900930**	**2**
72hrs	**33.24301**	**0.9834884**	**2**
96hrs	**38.55385**	**4.3273490**	**2**

alpha: 0.05 ; Df Error: 5

Critical Range

2	3	4	5
10.12771	**10.44293**	**10.57653**	**10.62456**

Means with the same letter are not significantly different.

Groups, Treatments and means

a	**120hrs**	**59.4**
b	**96hrs**	**38.55**
b	**72hrs**	**33.24**
c	**48hrs**	**19.47**
c	**24hrs**	**14.36**

> duncan.test(model,"trt",alpha=0.05,console=TRUE)
Error in duncan.test(model, "trt", alpha = 0.05, console = TRUE) :
unused argument(s) (console = TRUE)

Table45 Duncan multiple comparison on %TPC increased of 30%infused coconut oil on every infusion ratio

	Coconut	std.err	replication
120hrs	**51.09544**	**2.737236**	**2**
24hrs	**36.04064**	**3.037570**	**3**
48hrs	**45.84907**	**3.421545**	**2**
72hrs	**59.00301**	**2.651408**	**3**
96hrs	**49.49872**	**5.246370**	**2**

alpha: 0.05 ; Df Error: 7

Critical Range

2	3	4	5
11.49854	**11.95611**	**12.20057**	**12.33837**

Harmonic Mean of Cell Sizes 2.307692
Different value for each comparison
Means with the same letter are not significantly different.
Groups, Treatments and means

a	**72hrs**	**59**
ab	**120hrs**	**51.1**
ab	**96hrs**	**49.5**
bc	**48hrs**	**45.85**
c	**24hrs**	**36.04**

Table46 Duncan multiple comparison on %TPC increase of 30%infused sunflower oil on every infusion ratio

	sunflower	std.err	replication
120hrs	**56.03957**	**1.802884**	**2**
24hrs	**24.93983**	**1.802884**	**2**
48hrs	**26.96807**	**1.126802**	**2**
72hrs	**33.72889**	**2.929686**	**2**
96hrs	**44.99691**	**2.028244**	**2**

alpha: 0.05 ; Df Error: 5

Critical Range

2	3	4	5
7.355151	**7.584077**	**7.681102**	**7.715984**

Means with the same letter are not significantly different.

Groups, Treatments and means

a	**120hrs**	**56.04**
b	**96hrs**	**45**
c	**72hrs**	**33.73**
cd	**48hrs**	**26.97**
d	**24hrs**	**24.94**

> duncan.test(model,"trt",alpha=0.05,console=TRUE)
Error in duncan.test(model, "trt", alpha = 0.05, console = TRUE) :
unused argument(s) (console = TRUE)

Table47 Duncan multiple comparison on %TPC increase of 30%infused mineral oil on every infusion ratio

	mineral	std.err	replication
120hrs	**22.34211**	**0.4824561**	**2**
24hrs	**20.06140**	**0.2192982**	**2**
48hrs	**20.45614**	**0.0877193**	**2**
72hrs	**20.14912**	**0.1315789**	**2**
96hrs	**20.41228**	**0.3947368**	**2**

alpha: 0.05 ; Df Error: 5

Critical Range

2	3	4	5
1.104669	**1.139051**	**1.153623**	**1.158862**

Means with the same letter are not significantly different.

Groups, Treatments and means

a	**120hrs**	**22.34**
b	**48hrs**	**20.46**
b	**96hrs**	**20.41**
b	**72hrs**	**20.15**
b	**24hrs**	**20.06**

> duncan.test(model,"trt",alpha=0.05,console=TRUE)
Error in duncan.test(model, "trt", alpha = 0.05, console = TRUE) :
unused argument(s) (console = TRUE)

2. % inhibition increase

Table48 summary(Fact.RCBD) canola oil

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
A	**4**	**3.791**	**0.948**	**2.916**	**0.036002 ***
B	**1**	**8.272**	**8.272**	**25.451**	**1.62e-05 ***
rep	**1**	**0.109**	**0.109**	**0.336**	**0.566305**
A:B	**4**	**8.299**	**2.075**	**6.383**	**0.000641 ***
Residuals	**33**	**10.726**	**0.325**		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Table49 summary(Fact.RCBD) corn oil

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
A	**4**	**1598.1**	**399.5**	**81.036**	**< 2e-16 ***
B	**1**	**548.6**	**548.6**	**111.271**	**2.92e-12 ***
rep	**1**	**9.3**	**9.3**	**1.887**	**0.178576**
A:B	**4**	**139.1**	**34.8**	**7.054**	**0.000301 ***
Residuals	**34**	**167.6**	**4.9**		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Table50 summary(Fact.RCBD) coconut oil

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
A	**4**	**78749**	**19687**	**171.63**	**<2e-16 ***
B	**1**	**57889**	**57889**	**504.68**	**<2e-16 ***
rep	**1**	**6**	**6**	**0.05**	**0.824**
A:B	**4**	**50108**	**12527**	**109.21**	**<2e-16 ***
Residuals	**34**	**3900**	**115**		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Table51 summary(Fact.RCBD) sunflower oil

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
A	**4**	**114.15**	**28.537**	**218.482**	**< 2e-16 ***
B	**1**	**3.74**	**3.741**	**28.639**	**7.14e-06 ***
rep	**1**	**0.00**	**0.002**	**0.016**	**0.901**
A:B	**4**	**1.77**	**0.444**	**3.397**	**0.020 ***
Residuals	**32**	**4.18**	**0.131**		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Table52 summary(Fact.RCBD) mineral oil

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
A	**4**	**79162**	**19790**	**110.726**	**1.61e-15 ***
B	**1**	**8138**	**8138**	**45.530**	**4.52e-07 ***
rep	**1**	**355**	**355**	**1.984**	**0.171308**
A:B	**4**	**4915**	**1229**	**6.875**	**0.000708 ***
Residuals	**25**	**4468**	**179**		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Table53 summary(Fact.RCBD) 30% of every oils

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
A	**4**	**52244**	**13061**	**356.230**	**<2e-16 ***
B	**4**	**240660**	**60165**	**1640.976**	**<2e-16 ***
rep	**1**	**8**	**8**	**0.206**	**0.652**
A:B	**16**	**67442**	**4215**	**114.965**	**<2e-16 ***
Residuals	**41**	**1503**	**37**		

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Table54 Duncan multiple comparison on effect of 24hours infusion period on every infused oils

	24hrs	std.err	replication
canola	**0.0000000**	**0.0000000**	**3**
coconut	**41.3520632**	**2.0275690**	**3**
corn	**0.8743684**	**0.6298629**	**3**
mineral	**14.9110807**	**NA**	**1**
sunflower	**0.0000000**	**0.0000000**	**3**

alpha: 0.05 ; Df Error: 8
Critical Range

2	3	4	5
4.096280	**4.268711**	**4.365090**	**4.422873**

Harmonic Mean of Cell Sizes 2.142857

Different value for each comparison
Means with the same letter are not significantly different.
Groups, Treatments and means

a	**coconut**	**41.35**
b	**mineral**	**14.91**
c	**corn**	**0.8744**
c	**canola**	**0**
c	**sunflower**	**0**

> duncan.test(model,"trt",alpha=0.05,console=TRUE)
Error in duncan.test(model, "trt", alpha = 0.05, console = TRUE) :
unused argument(s) (console = TRUE)

Table55 Duncan multiple comparison on effect of 48hours infusion period on every infused oils

	48hrs	std.err	replication
canola	**0.6164339**	**0.3284800**	**3**
coconut	**80.8604039**	**3.3239147**	**3**
corn	**8.1180136**	**1.6281381**	**3**
mineral	**77.8385773**	**8.2079343**	**2**
sunflower	**0.4983209**	**0.3358249**	**3**

alpha: 0.05 ; Df Error: 9
Critical Range

2	3	4	5
9.539799	**9.957166**	**10.197593**	**10.347385**

Harmonic Mean of Cell Sizes 2.727273

Different value for each comparison
Means with the same letter are not significantly different.
Groups, Treatments and means

a	**coconut**	**80.86**
a	**mineral**	**77.84**
b	**corn**	**8.118**
b	**canola**	**0.6164**
b	**sunflower**	**0.4983**

> duncan.test(model,"trt",alpha=0.05,console=TRUE)
Error in duncan.test(model, "trt", alpha = 0.05, console = TRUE) :
unused argument(s) (console = TRUE)

Table56 Duncan multiple comparison on effect of 72hours infusion period on every infused oils

	72hrs	std.err	replication
canola	**0.7913087**	**0.4386111**	**3**
coconut	**147.5856014**	**7.0419721**	**3**
corn	**10.4740962**	**1.2178019**	**3**
mineral	**110.6703146**	**7.2387177**	**3**
sunflower	**0.8774781**	**0.2599935**	**2**

alpha: 0.05 ; Df Error: 9
Critical Range

2	3	4	5
16.10604	**16.81068**	**17.21659**	**17.46948**

Harmonic Mean of Cell Sizes 2.727273

Different value for each comparison
Means with the same letter are not significantly different.
Groups, Treatments and means

a	**coconut**	**147.6**
b	**mineral**	**110.7**
c	**corn**	**10.47**
c	**sunflower**	**0.8775**
c	**canola**	**0.7913**

> duncan.test(model,"trt",alpha=0.05,console=TRUE)
Error in duncan.test(model, "trt", alpha = 0.05, console = TRUE) :
unused argument(s) (console = TRUE)

Table57 Duncan multiple comparison on effect of 96 hours infusion period on every infused oils

	96hrs	std.err	replication
canola	**1.033948**	**0.5372245**	**3**
coconut	**180.362891**	**5.6064513**	**3**
corn	**17.489987**	**0.7422941**	**3**
mineral	**144.870041**	**15.0478796**	**2**
sunflower	**1.657459**	**0.6499838**	**2**

alpha: 0.05 ; Df Error: 8
Critical Range

2	3	4	5
18.55372	**19.33472**	**19.77127**	**20.03299**

Harmonic Mean of Cell Sizes 2.5

Different value for each comparison
Means with the same letter are not significantly different.
Groups, Treatments and means

a	**coconut**	**180.4**
b	**mineral**	**144.9**
c	**corn**	**17.49**
c	**sunflower**	**1.657**
c	**canola**	**1.034**

> duncan.test(model,"trt",alpha=0.05,console=TRUE)
Error in duncan.test(model, "trt", alpha = 0.05, console = TRUE) :
unused argument(s) (console = TRUE)

Table58 Duncan multiple comparison on effect of 120 hours infusion period on every infused oils

	120hrs	std.err	replication
canola	**3.613352**	**0.2623123**	**2**
coconut	**265.232660**	**3.3239147**	**3**
corn	**21.835650**	**1.4743975**	**3**
mineral	**150.341997**	**1.3679891**	**2**
sunflower	**4.647384**	**0.1985733**	**3**

alpha: 0.05 ; Df Error: 8

Critical Range

2	3	4	5
6.661540	**6.941954**	**7.098690**	**7.192659**

Harmonic Mean of Cell Sizes 2.5

Different value for each comparison

Means with the same letter are not significantly different.

Groups, Treatments and means

a	**coconut**	**265.2**
b	**mineral**	**150.3**
c	**corn**	**21.84**
d	**sunflower**	**4.647**
d	**canola**	**3.613**

> duncan.test(model,"trt",alpha=0.05,console=TRUE)

Error in duncan.test(model, "trt", alpha = 0.05, console = TRUE) :

unused argument(s) (console = TRUE)

Table59 Duncan multiple comparison on %TPC increased of 30%infused canola oil on every infusion ratio

	canola	std.err	replication
120hrs	**3.6133517**	**0.2623123**	**2**
24hrs	**0.0000000**	**0.0000000**	**3**
48hrs	**0.6164339**	**0.3284800**	**3**
72hrs	**0.7913087**	**0.4386111**	**3**
96hrs	**1.0339476**	**0.5372245**	**3**

alpha: 0.05 ; Df Error: 9

Critical Range

2	3	4	5
1.237204	**1.291332**	**1.322513**	**1.341939**

Harmonic Mean of Cell Sizes 2.727273

Different value for each comparison

Means with the same letter are not significantly different.

Groups, Treatments and means

a	**120hrs**	**3.613**
b	**96hrs**	**1.034**
b	**72hrs**	**0.7913**
b	**48hrs**	**0.6164**
b	**24hrs**	**0**

> duncan.test(model,"trt",alpha=0.05,console=TRUE)

Error in duncan.test(model, "trt", alpha = 0.05, console = TRUE) :

unused argument(s) (console = TRUE)

Table60 Duncan multiple comparison on %TPC increased of 30%infused corn oil on every infusion ratio

	corn	std.err	replication
120hrs	**21.8356501**	**1.4743975**	**3**
24hrs	**0.8743684**	**0.6298629**	**3**
48hrs	**8.1180136**	**1.6281381**	**3**
72hrs	**10.4740962**	**1.2178019**	**3**
96hrs	**17.4899866**	**0.7422941**	**3**

alpha: 0.05 ; Df Error: 10

Critical Range

2	3	4	5
3.795810	**3.966586**	**4.067111**	**4.131415**

Means with the same letter are not significantly different.

Groups, Treatments and means

a	**120hrs**	**21.84**
b	**96hrs**	**17.49**
c	**72hrs**	**10.47**
c	**48hrs**	**8.118**
d	**24hrs**	**0.8744**

> duncan.test(model,"trt",alpha=0.05,console=TRUE)
Error in duncan.test(model, "trt", alpha = 0.05, console = TRUE) :
unused argument(s) (console = TRUE)

Table61 Duncan multiple comparison on %TPC increased of 30%infused coconut oil on every infusion ratio

	coconut	std.err	replication
120hrs	**265.23266**	**3.323915**	**3**
24hrs	**41.35206**	**2.027569**	**3**
48hrs	**80.86040**	**3.323915**	**3**
72hrs	**147.58560**	**7.041972**	**3**
96hrs	**180.36289**	**5.606451**	**3**

alpha: 0.05 ; Df Error: 10

Critical Range

2	3	4	5
14.59249	**15.24902**	**15.63547**	**15.88268**

Means with the same letter are not significantly different.

Groups, Treatments and means

a	**120hrs**	**265.2**
b	**96hrs**	**180.4**
c	**72hrs**	**147.6**
d	**48hrs**	**80.86**
e	**24hrs**	**41.35**

> duncan.test(model,"trt",alpha=0.05,console=TRUE)
Error in duncan.test(model, "trt", alpha = 0.05, console = TRUE) :
unused argument(s) (console = TRUE)

Table62 Duncan multiple comparison on %TPC increased of 30%infused sunflower oil on every infusion ratio

	sunflower	std.err	replication
120hrs	**4.6473838**	**0.1985733**	**3**
24hrs	**0.0000000**	**0.0000000**	**3**
48hrs	**0.4983209**	**0.3358249**	**3**
72hrs	**0.8774781**	**0.2599935**	**2**
96hrs	**1.6574586**	**0.6499838**	**2**

alpha: 0.05 ; Df Error: 8

Critical Range

2	3	4	5
1.003419	**1.045657**	**1.069266**	**1.083421**

Harmonic Mean of Cell Sizes 2.5

Different value for each comparison
Means with the same letter are not significantly different.
Groups, Treatments and means

a	**120hrs**	**4.647**
b	**96hrs**	**1.657**
bc	**72hrs**	**0.8775**
c	**48hrs**	**0.4983**
c	**24hrs**	**0**

> duncan.test(model,"trt",alpha=0.05,console=TRUE)
Error in duncan.test(model, "trt", alpha = 0.05, console = TRUE) :
unused argument(s) (console = TRUE)

Table63 Duncan multiple comparison on %TPC increased of 30%infused mineral oil on every infusion ratio

	mineral	std.err	replication
120hrs	**150.34200**	**1.367989**	**2**
24hrs	**14.91108**	**NA**	**1**
48hrs	**77.83858**	**8.207934**	**2**
72hrs	**110.67031**	**7.238718**	**3**
96hrs	**144.87004**	**15.047880**	**2**

alpha: 0.05 ; Df Error: 5

Critical Range

2	3	4	5
36.83244	**37.97884**	**38.46471**	**38.63939**

Harmonic Mean of Cell Sizes 1.764706

Different value for each comparison
Means with the same letter are not significantly different.
Groups, Treatments and means

a	**120hrs**	**150.3**
ab	**96hrs**	**144.9**
bc	**72hrs**	**110.7**
c	**48hrs**	**77.84**
d	**24hrs**	**14.91**

> duncan.test(model,"trt",alpha=0.05,console=TRUE)
Error in duncan.test(model, "trt", alpha = 0.05, console = TRUE) :
unused argument(s) (console = TRUE

III. Preference test

Table64 SAS out put; Duncan comparison, class level information

Randomized Complete Block

The ANOVA Procedure

Class Level Information

Class	Levels	Values
Block	20	1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20
Treatment	6	A B C D E F

Number of Observations Read	120
Number of Observations Used	120

Table65 SAS out put; Duncan multiple comparison on color attribute

Randomized Complete Block

The ANOVA Procedure

Dependent Variable: Color

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	24	54.1666667	2.2569444	2.08	0.0066
Error	95	103.0000000	1.0842105		
Corrected Total	119	157.1666667			

R-Square	Coeff Var	Root MSE	Color Mean
0.344645	16.22734	1.041254	6.416667

Source	DF	Anova SS	Mean Square	F Value	Pr > F
Block	19	29.50000000	1.55263158	1.43	0.1306
Treatment	5	24.66666667	4.93333333	4.55	0.0009

Randomized Complete Block

The ANOVA Procedure

Duncan's Multiple Range Test for Color

Alpha	0.05
Error Degrees of Freedom	95
Error Mean Square	1.084211

Number of Means	**2**	**3**	**4**	**5**	**6**
Critical Range	.6537	.6879	.7106	.7272	.7401

Means with the same letter are not significantly different.

Duncan Grouping	**Mean**	**N**	**Treatment**
A	7.3500	20	D
B	6.4500	20	C
B			
B	6.3500	20	E
B			
B	6.3500	20	F
B			
B	6.0500	20	A
B			
B	5.9500	20	B

Table66 SAS out put; Duncan multiple comparison on fragrance attribute

Randomized Complete Block

The ANOVA Procedure

Dependent Variable: Fragrance

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	24	211.1666667	8.7986111	8.64	<.0001
Error	95	96.8000000	1.0189474		
Corrected Total	119	307.9666667			

R-Square	Coeff Var	Root MSE	Fragrance Mean
0.685680	16.77722	1.009429	6.016667

Source	DF	Anova SS	Mean Square	F Value	Pr > F
Block	19	196.3000000	10.3315789	10.14	<.0001
Treatment	5	14.8666667	2.9733333	2.92	0.0170

Randomized Complete Block

The ANOVA Procedure

Duncan's Multiple Range Test for Fragrance

Alpha	0.05
Error Degrees of Freedom	95
Error Mean Square	1.018947

Number of Means	**2**	**3**	**4**	**5**	**6**
Critical Range	.6337	.6669	.6889	.7050	.7175

Means with the same letter are not significantly different.

Duncan Grouping	**Mean**	**N**	**Treatment**
A	6.3500	20	D
A			
A	6.3000	20	E
A			
A	6.2000	20	F
A			
A	6.0000	20	B
A			
A	5.9500	20	A
B	5.3000	20	C

Table67 SAS out put; Duncan multiple comparison on lather attribute

Randomized Complete Block

The ANOVA Procedure

Dependent Variable: Lather

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	24	89.0000000	3.7083333	2.12	0.0055
Error	95	165.9916667	1.7472807		
Corrected Total	119	254.9916667			

R-Square	Coeff Var	Root MSE	Lather Mean
0.349031	19.85253	1.321847	6.658333

Source	DF	Anova SS	Mean Square	F Value	Pr > F
Block	19	72.15833333	3.79780702	2.17	0.0075
Treatment	5	16.84166667	3.36833333	1.93	0.0968

Randomized Complete Block

The ANOVA Procedure

Duncan's Multiple Range Test for Lather

Alpha	0.05
Error Degrees of Freedom	95
Error Mean Square	1.747281

Number of Means	**2**	**3**	**4**	**5**	**6**
Critical Range	.8298	.8733	.9021	.9232	.9396

Means with the same letter are not significantly different.

Duncan Grouping	Mean	N	Treatment
A	7.2000	20	D
A			
B A	7.1000	20	C
B A			
B A	6.6000	20	E
B A			
B A	6.5500	20	F
B A			
B A	6.3000	20	A
B			
B	6.2000	20	B

Table68 SAS out put; Duncan multiple comparison on moturizing attribute

Randomized Complete Block

The ANOVA Procedure

Dependent Variable: moisturizing

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	24	78.8666667	3.2861111	3.15	<.0001
Error	95	99.1333333	1.0435088		
Corrected Total	119	178.0000000			

R-Square	Coeff Var	Root MSE	moisturing Mean
0.443071	15.71573	1.021523	6.500000

Source	DF	Anova SS	Mean Square	F Value	Pr > F
Block	19	73.66666667	3.87719298	3.72	<.0001
Treatment	5	5.20000000	1.04000000	1.00	0.4241

Randomized Complete Block

The ANOVA Procedure

Duncan's Multiple Range Test for moisturizing

Alpha	0.05
Error Degrees of Freedom	95
Error Mean Square	1.043509

Number of Means	**2**	**3**	**4**	**5**	**6**
Critical Range	.6413	.6749	.6971	.7134	.7261

Means with the same letter are not significantly different.

Duncan Grouping	**Mean**	**N**	**Treatment**
A	6.7500	20	E
A			
A	6.6500	20	F
A			
A	6.6500	20	D
A			
A	6.5000	20	A
A			
A	6.2500	20	B
A			
A	6.2000	20	C

Table69 SAS out put; Duncan multiple comparison on scrubbing attribute

Randomized Complete Block

The ANOVA Procedure

Dependent Variable: scrubbing

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	24	105.3000000	4.3875000	2.29	0.0025
Error	95	182.2916667	1.9188596		
Corrected Total	119	287.5916667			

R-Square	Coeff Var	Root MSE	scrubbing Mean
0.366144	21.50420	1.385229	6.441667

Source	DF	Anova SS	Mean Square	F Value	Pr > F
Block	19	95.75833333	5.03991228	2.63	0.0011
Treatment	5	9.54166667	1.90833333	0.99	0.4254

Randomized Complete Block

The ANOVA Procedure

Duncan's Multiple Range Test for scrubbing

Alpha	0.05
Error Degrees of Freedom	95
Error Mean Square	1.91886

Number of Means	**2**	**3**	**4**	**5**	**6**
Critical Range	.8696	.9151	.9453	.9675	.9846

Means with the same letter are not significantly different.

Duncan Grouping	**Mean**	**N**	**Treatment**
A	6.9000	20	D
A			
A	6.6500	20	C
A			
A	6.5000	20	F
A			
A	6.3500	20	E
A			
A	6.2000	20	A
A			
A	6.0500	20	B

Table70 SAS out put; Duncan multiple comparison on skin feel attribute

Randomized Complete Block

The ANOVA Procedure

Dependent Variable: skinfeel

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	24	83.5666667	3.4819444	3.41	<.0001
Error	95	97.0250000	1.0213158		
Corrected Total	119	180.5916667			

R-Square	Coeff Var	Root MSE	skinfeel Mean
0.462738	15.29284	1.010602	6.608333

Source	DF	Anova SS	Mean Square	F Value	Pr > F
Block	19	75.42500000	3.96973684	3.89	<.0001
Treatment	5	8.14166667	1.62833333	1.59	0.1691

Randomized Complete Block

The ANOVA Procedure

Duncan's Multiple Range Test for skinfeel

Alpha	0.05
Error Degrees of Freedom	95
Error Mean Square	1.021316

Number of Means	**2**	**3**	**4**	**5**	**6**
Critical Range	.6344	.6676	.6897	.7058	.7183

Means with the same letter are not significantly different.

Duncan Grouping	**Mean**	**N**	**Treatment**
A	7.1000	20	D
A			
B A	6.7500	20	E
B A			
B A	6.5500	20	A
B A			
B A	6.5500	20	F
B A			
B A	6.4000	20	B
B			
B	6.3000	20	C

Table71 SAS out put; Duncan multiple comparison on over all liking attribute

Randomized Complete Block

The ANOVA Procedure

Dependent Variable: Overall

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	24	46.4000000	1.9333333	2.13	0.0053
Error	95	86.2666667	0.9080702		
Corrected Total	119	132.6666667			

R-Square	Coeff Var	Root MSE	Overall Mean
0.349749	13.94528	0.952927	6.833333

Source	DF	Anova SS	Mean Square	F Value	Pr > F
Block	19	39.33333333	2.07017544	2.28	0.0048
Treatment	5	7.06666667	1.41333333	1.56	0.1799

Randomized Complete Block

The ANOVA Procedure

Duncan's Multiple Range Test for Overall

Alpha	0.05
Error Degrees of Freedom	95
Error Mean Square	0.90807

Number of Means	**2**	**3**	**4**	**5**	**6**
Critical Range	.5982	.6295	.6503	.6655	.6773

Means with the same letter are not significantly different.

Duncan Grouping		Mean	N	Treatment
	A	7.3000	20	D
	A			
B	A	6.9500	20	E
B	A			
B	A	6.8000	20	A
B	A			
B	A	6.7500	20	F
B	A			
B	A	6.6500	20	B
B				
B		6.5500	20	C