

## *Full length Research Paper*

# **Effect of different concentration of red palm olein on blood lipid profile in rat**

**Eqbal M. A. Dauqan<sup>\*1</sup>, Halimah Abdullah Sani<sup>1</sup>, Aminah Abdullah<sup>2</sup> and Zalifah Mohd Kasim<sup>2</sup>**

<sup>1</sup>School of Biosciences and Biotechnology, Faculty of Science and Technology, Universiti Kebangsaan Malaysia, 43600 Bangi Selangor, Malaysia

<sup>2</sup>School of Chemical Sciences and Food Technology, Faculty of Science and Technology, Universiti Kebangsaan Malaysia, 43600 Bangi Selangor, Malaysia

Accepted 15 August 2011

In this paper, we aimed to investigate the effect of different concentration of red palm olein (RPO) on blood lipid profile in rat. Seventy eight Sprague Dawley male rats were randomly divided into thirteen groups of 6 rats per group with 6 rats for T<sub>0</sub> group which kill before any treatment. Treated groups were given with different concentrations of RPO (5%, 10% and 15%) for 2, 4 and 8 weeks. Rats in control group were given normal rat pellet only while in treated groups 5%, 10% and 15% of additional RPO were given. Results showed a decline in Low Density Lipoprotein Cholesterol (LDL-C) values whereas the High Density Lipoprotein Cholesterol (HDL-C) values increased. Rats treated with 15% RPO for 8 weeks showed an increased in the mean LDL-C level and decreased in HDL-C compared to the control group. At 2 and 4 weeks of treatment, the total cholesterol was no significant difference ( $p \geq 0.05$ ) between control group and groups fed with different concentration (5%, 10%, and 15%) RPO but At 8 weeks, the total cholesterol level decreases in all concentrations compared to control group. The results of triglyceride (TG) in all treated rats were within the normal range. There was no significant difference ( $p \geq 0.05$ ) in TG of rats treated with RPO compared to the control group.

**Keyword:** Red Palm Olein, Total Cholesterol; Triglyceride; High Density Lipoprotein Cholesterol; Low Density Lipoprotein Cholesterol.

## **INTRODUCTION**

Palm oil contains approximately an equal amount of saturated and unsaturated fatty acids. Amongst the former, palmitic and stearic acid account for 45% and 5% of the total fatty acids, respectively. Palm olein, a liquid fraction obtained from the refining of palm oil, is rich in oleic acid (42.7– 43.9%),  $\beta$ -carotene and vitamin E (tocopherols and tocotrienols) (Deepak et al., 2004). It is rich in tocotrienol which has been reported to be natural inhibitors of cholesterol synthesis. Tocopherols are very important minor components of oils and fats because of their antioxidant properties (Che Man et al., 2005). Palm oil has a wide range of applications and it is commonly

fractionated into olein and stearin (Pramod, 2006). The different properties of palm oil and its fractions allow the products to be used for different purposes (Flingoh and Chong, 1992). Red Palm Oil (RPO) contains 50% saturated fatty acids, 40% monounsaturated fatty acids and 10% polyunsaturated fatty acids. The RPO is the only vegetable oil with a balanced composition of saturated and unsaturated fatty acids both in the processed and unprocessed forms (Edem and Akpanabiatu, 2006). It contains carotenoids, phosphatides, sterols, tocopherols and trace metals. They have shown to be effective against oxidative stress in vitro and in vivo (Aboua et al., 2009). Red Palm Oil is the richest natural source of  $\beta$ -carotene (500 - 700 mg/L), which is responsible for the characteristic colour of the oil. Most of the  $\beta$ -carotene is destroyed during

---

<sup>\*</sup>Corresponding author E-mail: [edouqan@yahoo.com](mailto:edouqan@yahoo.com)

processing at the palm oil refineries (Edem and Akpanabiatu, 2006). The carotenoids, together with vitamin E, ascorbic acid, enzymes and proteins, are members of the biological antioxidant network converting highly reactive radicals and free fatty peroxy radicals to less active species (Peng and Stanley, 2001) thus, protecting against oxidative damage to cells. Besides providing high energy density in the diet,  $\beta$ -carotene is the most abundant carotenoids which can be converted to vitamin A which plays an important role in the visual process. In addition, it is an antioxidant that destroys singlet oxygen and free radicals. Red Palm Oil is also a rich source of vitamin E, which is about 559 to 1000 ppm. Vitamin E acts as a potent antioxidant serving to protect cellular membranes from free radical-catalyzed lipid peroxidation (Edem, 2002). Commercial red palm olein contains 18-25  $\mu\text{g kg}^{-1}$  of co-enzyme Q10. Co-enzyme Q10 is claimed to exhibit 10 times greater antioxidant property than vitamin E (Radhika et al., 2010).

Atherosclerotic lesions in man and in animals appear to be related to elevated plasma total cholesterol (TC), low-density lipoprotein cholesterol (LDL-C), decreased high-density lipoprotein cholesterol (HDL-C) and excess fat consumption (Jaarin et al., 2001; Kamisah et al., 2006). Therefore the main aim of the present study was to investigate the effect of different concentration of red palm olein (RPO) on lipid profile of rats fed with red palm olein (RPO) until 8 weeks of treatment.

## MATERIALS AND METHODS

### Instruments

A Reflotron (ROCHE, 10007908, germany) was used for the measurement of blood lipid profile. A centrifuge (KUBOTA 2010, Malaysia) with speed 3000 r.p.m at room temperature for 10 min was used to separate the plasma from the whole blood.

### Experimental diets

The evaluated red palm olein (RPO) samples consisted of carotenes (576 ppm), vitamin E (>800 ppm) and free fatty acids (0.045%) was provided by Carotino SDN BHD company. The test diet was prepared by mixing RPO with normal commercial rat pellet to contain 5%, 10% and 15% of the red palm olein (RPO). The 5% diet was prepared by adding 5g RPO to 95g rat pellet, and mixed manually and the diets were then left to absorb the RPO at room temperature overnight and stored at 20° C before the feeding trial was conducted. Similar process was conducted with 10%, and 15% RPO.

### Animals

Seventy eight Sprague Dawley male rats each weighing between 170-250 g and approximately 80 days old were obtained from the animal house of the Faculty of Science and Technology, Universiti Kebangsaan Malaysia. Rats were fed *ad libitum* with commercial rat's food containing 5, 10 and 15% red palm oil. At the end of the experiment, after 2, 4 or 8 weeks of treatment, the feeding of rats was stopped and the rats were fasted for 18 h. They were anesthetized using chloroform. Blood samples were collected from post vena cava and transferred into vacuette EDTA tubes immediately. All procedures were reviewed and approved by the Universiti Kebangsaan Malaysia Animal Ethics committee (FST/SBB/2010/HALIMAH/24-AUGST/322).

### Lipid Analysis of the Blood

Total cholesterol (TC) and triglyceride (TG) were measured by strips with reflotron-machine using 32  $\mu\text{l}$  whole blood. High density lipoprotein cholesterol (HDL-C) was determined by strips (Roche, Germany) with reflotron-machine using 32  $\mu\text{l}$  plasma blood. Plasma blood was prepared using a centrifuge (KUBOTA 2010, Malaysia) with speed 3000 r.p.m at room temperature for 10 min to remove red blood cells and recover plasma. Low density lipoprotein cholesterol (LDL-C) was calculated from TC, HDL-C and TG values using the Friedewald equation. According to Wilai and Donpichit (2004) the Friedewald equation for serum triglyceride less than 400mg/dL should be calculated as follows:  $\text{LDL (mg/dL)} = \text{Total Cholesterol} - \text{HDL Cholesterol} - \text{Triglycerides}/5$

All analyses were completed within 24 h of sample collection.

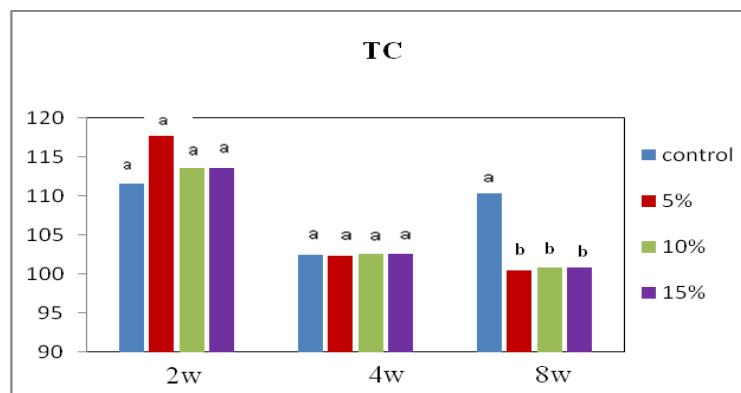
### Statistical Analysis

Results were expressed as mean values  $\pm$  SEM (n=6). Means of six samples were compared by analysis of variance (ANOVA). Significant differences between means were determined by Tukey's least significant difference ( $p \leq 0.05$ ). The software used was MINITAB® (14.20).

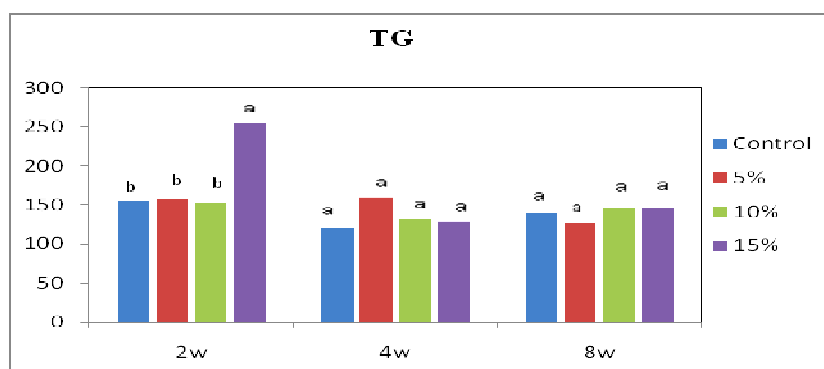
## RESULTS

### Total cholesterol

Figure 1 shows the results of TC level in blood samples



**Figure 1:** Total cholesterol (mg/dl) in rats fed different concentrations of RPO (5%, 10%, and 15%) for 2, 4 and 8 weeks. Bars are mean  $\pm$  SEM (n=6). Different alphabet on different histogram within each group indicate significant different ( $p \leq 0.05$ ).



**Figure 2:** Triglycerides (mg/dl) in rats fed different concentrations of RPO (5%, 10%, & 15%) for 2, 4 and 8 weeks. Bars are mean  $\pm$  SEM (n=6). Different alphabet on different histogram within each group indicate significant different ( $p \leq 0.05$ ).

of rats that were treated with different concentrations of RPO (5%, 10% and 15%) for different times (2, 4 and 8 weeks) of treatment. After 2 weeks of treatment the results of total cholesterol values were within the normal range ( $< 200$ mg/dl). However, the total cholesterol level in blood sample was no significant different ( $P \geq 0.05$ ) between control group and rat blood treated with 5%, 10% and 15% RPO groups. At 4 weeks, there was no significant difference ( $p \geq 0.05$ ) between control group and groups fed with different concentration (5%, 10%, and 15%) RPO. At 8 weeks, the total cholesterol level decreases in all concentrations compared to control group.

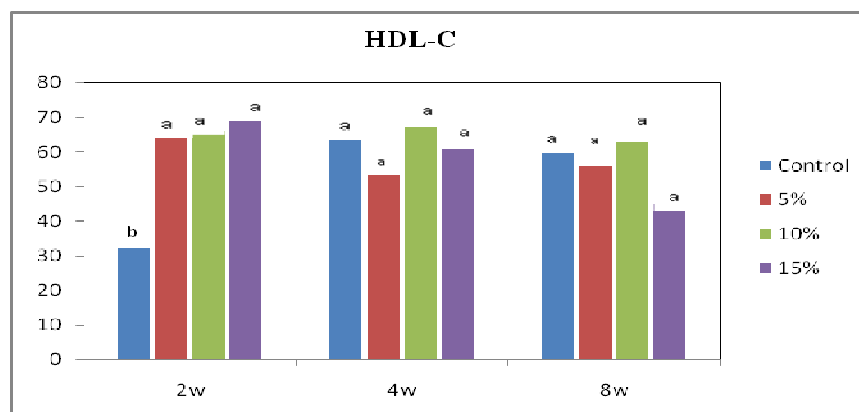
### Triglycerides

Figure 2 shows the results of TG in different concentration of RPO (5%, 10% and 15%) for different

times for of treatment. After 2 weeks of treatment the results in this group had triglyceride values within the normal range ( $< 150$ mg/dl). TG level in blood sample was the highest in rat blood fed with 15% RPO and significantly higher ( $P \leq 0.05$ ) than the control or 5% and 10% treated sample. This findings could be attributed to the period of treatment (2 weeks) was not enough to observe the effect of 15% of RPO on TG level. There were no significant different ( $P \geq 0.05$ ) between control and rat blood treated with 5% or 10% RPO. At 4 and 8 weeks, there was no significance difference ( $P \geq 0.05$ ) between control group and different concentrations groups (5%, 10%, and 15%).

### HDL-C and LDL-C

Figure 3 shows the results of HDL-C in different



**Figure 3:** HDL-C (mg/dl) in rats fed different concentrations of RPO (5%, 10%, & 15%) for 2, 4 and 8 weeks. Bars are mean  $\pm$  SEM (n=6), Different alphabet on different histogram within each group indicate significant different ( $p \leq 0.05$ ).

**Table 1:** The mean  $\pm$  SEM values of body weight (n=6).

Group	Weight (g)		
	2 weeks	4 weeks	8 weeks
Control	238.4 $\pm$ 6.8 b	347.1 $\pm$ 6.8 b	451.8 $\pm$ 19.1 a
5%	331.9 $\pm$ 11.8 a	383.6 $\pm$ 5.3 a	398.8 $\pm$ 9.6 a
10%	348.8 $\pm$ 9.2 a	389.9 $\pm$ 16.4 a	407.4 $\pm$ 8.8 a
15%	351.6 $\pm$ 8.3 a	371.2 $\pm$ 7.8 a	387.0 $\pm$ 4.9 a

Different alphabet indicates significant different ( $p \leq 0.05$ )

concentrations of RPO (5%, 10% and 15%) for different times of treatment. The results showed that after 2 weeks of treatment the HDL-C increased parallel to the increasing concentration of RPO compared to control group. However there was significant difference of HDL-C. At 4 weeks, the results of HDL-C showed no significance difference between control group and different concentrations groups (5%, 10%, and 15%) of RPO and the duration of treatment compared to the control group. After 8 weeks the results of HDL-C showed no significance difference ( $P \leq 0.05$ ) between control group and groups receiving different concentrations (5% and 10%) of RPO while decreased in HDL-C were observed in 15% group compare to the control group. Fig. 4 shows the results of LDL-C in different concentrations of RPO (5%, 10% and 15%) for different times of treatment. The results of LDL-C were decreasing parallel to the increasing concentration of RPO compared to control group. However there was significant difference of LDL-C. After 2 weeks of treatment, the results of LDL-C were the lowest in rat blood fed with 5%, 10% and 15% RPO and significantly

lower ( $p \leq 0.05$ ) than control group. At 4 weeks, the results of LDL-C decreased with increasing concentration of RPO and the duration of treatment compared to the control group. Rats treated with 15% treated RPO for 8 weeks showed an increased in LDL-C value compared to the control group. The amount of LDL-C seemed to decrease with increasing concentration of RPO and duration of treatment compared to the control group.

## DISCUSSION

**Table 1** showed the effect of different concentrations of RPO on body weight of rats. The body weight increased in each group (2, 4 and 8 weeks) compared with  $T_0$  and control groups. Where  $T_0$  (243.9  $\pm$  7.3) is the mean  $\pm$  SEM of body weight of rats before treatment. The increase in mean body weight of rats could be attributed to the high total body fat content in the groups fed different concentrations of RPO in comparison with  $T_0$  and control groups. This is consistent with the work carried out by Oluba et al. (2008) in which rats that were fed with palm

oil enriched diet had a significantly higher body weight compare to  $T_0$  rats. This finding were similar to that of Hamid et al. (2010) who reported the increased in the body weight of rats fed palm olein. The use of palm oil in the diet should be safe and will not increase the risk of cardio vascular disease (CVD). Toxicological and pharmacological studies showed that supplementation with palm tocotrienols up to 2,500 milligrams per day per kilogram of body weight does not produce any significant side effects (Sutapa and Analava, 2009).

Total cholesterol in 5%, 10% and 15% RPO treated rat blood was significantly lower ( $P \leq 0.05$ ) than the control group after 8 weeks of treatment. This could be due to the fact that RPO is rich in antioxidant ( $\beta$ -carotene and vitamin E). Studies have shown that palm oil, saturated fatty acid-rich oil, has better positive impact on serum lipids than soybean oil (unsaturated fatty acid-rich oil) (Oluba et al., 2008). It is shown that different concentration of RPO did not effect on the blood cholesterol after 2 and 4 weeks of treatment but there was decreased in total cholesterol after 8 weeks of treatment. However, the effect of RPO on blood cholesterol appears to depend on the period of treatment.

The total cholesterol lowering effect of RPO could be attributed to its high content of vitamin E (tocopherol and tocotrienol) and beta carotene. Tocotrienols had a hypocholesterolemic effect probably through the inhibition of cholesterol synthesis (Amr, 2010). This finding suggested that feeding with different concentration of RPO reduced blood total cholesterol after 8 weeks of the treatment. This findings were similar to that of Kamisah et al. (2006) who reported that the TC levels were unaffected in plasma rat that were fed different types of palm oil at three time 4, 8 and 12 weeks. The effect of RPO in this study was in agreement with Jaarin et al. (2001) who also reported that RPO reduced serum total cholesterol but Jaarin et al. (2001) used serum instead of whole blood.

The meaning of TG results suggested that feeding of red palm oil does not raise the level of blood triglycerides after long time of treatment. However, triglycerides are another important lipid. Like cholesterol, it is found in the foods but the body also makes its own triglycerides. People with high triglycerides often have high total cholesterol, high LDL cholesterol and a low HDL cholesterol level. Like high total cholesterol and a high LDL cholesterol, having high triglycerides is a risk factor for the build up of plaque and heart disease. People who have diabetes and those who are obese are more likely to have high triglycerides. In fact, no other vegetable oil has as much vitamin E as compared to palm oil (Mukherjee and Mitra, 2009).

These findings observed that feeding with RPO increased plasma HDL or kept within normal range. High density lipoprotein cholesterol protects against heart

disease, so for HDL higher numbers are better. A level less than 40 mg/dL is low and is considered a major risk factor because it increases the risk for developing heart disease. High density lipoprotein levels of 60 mg/dL or more help to lower the risk for heart disease.

Since red palm olein (RPO, mildly refined or crude) became available commercially, its role as a good natural source of antioxidant vitamins, namely tocopherols, tocotrienols, as well as  $\beta$ -carotene became all the more evident (Susanna et al., 2004). Natural vitamin E exists in eight different forms or isomers, four tocopherols and four tocotrienols. Palm oil contains alpha, beta, gamma, and delta tocopherols and alpha, beta, gamma, and delta tocotrienols. Tocotrienols in vitamin E have been found to have antioxidant and anti-cancer activities. Tocotrienols by its action on liver enzymes lowers blood cholesterol levels without reduce HDL-C (Mukherjee and Mitra, 2009). The antioxidant power of red palm oil can be of help in protecting against a variety of health problems (Bruce, 2010).

The group receiving 15% RPO had higher LDL-C than the control group. Red palm oil stimulates the synthesis of protective HDL cholesterol and suppression of harmful LDL cholesterol (Mukherjee and Mitra, 2009). The results in this study observed that feeding with RPO reduced plasma LDL-C. Low density lipoprotein builds up in the wall of the arteries. This causes a hardening of arteries, which blocks or slows down the blood flow to the heart. Red palm oil is also the only oil that contains high amounts of tocotrienols, which are very potent anti-oxidants. The high vitamin A (carotenoid) content (300 times higher than tomatoes for equivalent amounts), high vitamin E (tocopherols and tocotrienols) content and 45% oleic acid makes this oil a balanced and natural oil which can be used as a functional food (Jacques, 2005).

Palm oil is rich in vitamin E, (particularly tocotrienols), which appear to reduce serum cholesterol concentrations (Ebong et al., 1999; Sutapa and Analava, 2009). Palmitic acid increased the total: HDL cholesterol ratio more than other saturated fatty acids, including lauric acid and myristic acid, which are abundant in palm kernel oil and coconut oil; the other highly saturated tropical oil (Sutapa and Analava, 2009).

## CONCLUSION

There was no significant difference ( $p \geq 0.05$ ) the total cholesterol between control group and groups fed with different concentration (5%, 10%, and 15%) RPO but the total cholesterol level decreases in all concentrations compared to control group after 8 weeks of treatment. The TG were within the normal range with different concentration of RPO (5%, 10% and 15%) for different duration of treatment. However, after 2 weeks of

treatment the HDL-C increased parallel to the increasing concentration of RPO. In contrast, the results of LDL-C decreased with increasing concentration of RPO compared to the control group. After 4 weeks and 8 weeks, there were no significant difference ( $P \leq 0.05$ ) in HDL-C between the control group and groups receiving different concentrations of RPO (5%, 10%, and 15%) but LDL-C decreased with increasing concentration of RPO and duration of treatment compared to control group. However, rats treated with 15% RPO for 8 weeks showed an increased in LDL-C value compared to the control group. These results could be due to the high content of vitamin E (tocopherols and tocotrienols) and  $\beta$ -carotene in red palm olein.

## ACKNOWLEDGEMENT

This work was supported by the Organization for Women in Science for the Developing World (OWSDW) and the research was funded by UKM-GUP-NBT-27-103, UKM-HEJIM-Industri-16-2010 and UKM OUP-NBT-29-139/2011. We grateful thank to carotene sdn bhd Malaysia for providing red palm olein sample.

## REFERENCES

- Aboua YG, Brooks N, Awoniyi DO, and Plessis SS (2009). Red palm oil: A natural good Samaritan for sperm apoptosis. *Med. Technol.* 23:8-10
- Amr AR, Fatma AL and Abd Elrahman MA (2010). Effect of Some Dietary Oils and Fats on Serum Lipid Profile, Calcium Absorption and Bone Mineralization in Mice. *Pakistan J. Nutr.* 9(7): 643-650
- Bruce F (2010). Red palm oil. Coconut recherche centre (11/2/2011) <http://www.coconutresearchcenter.org/article%20red%20palm%20oil.htm>
- Che Man YB, Ammawath W and Mirghani MES (2005). Determining  $\alpha$ -tocopherol in refined bleached and deodorized palm olein by Fourier transform infrared spectroscopy. *Food Chem.* 90: 323-327
- Deepak N, Subeena S, Mathew KT, Amit KD, and Subir KM (2004). Effect of dietary palm olein oil on oxidative stress associated with ischemic-reperfusion injury in isolated rat heart. *BMC Pharmacol.* 47: 2210-429
- Ebong PE, Owu DU and Isong EU (1999). Influence of palm oil (*Elaeis guineensis*) on health. *Plant foods for Human Nutr.* 53:209-222
- Edem DO (2002). Palm oil: Biochemical, physiological, nutritional, hematological, and toxicological aspects. *Plant Foods for Human Nutr.* 57: 319-341
- Edem DO, and Akpanabiatu MI (2006). Effects of palm oil- containing Diets on enzyme activities of rats. *Pakistan J. Nutr.* 5: 301-305
- Flingoh, CHOH, and Chong CL, (1992). Surface tensions of palm oil, palm olein and palm stearin. *Elasia* 4(1)
- Jaarin Kamisah S, Nik Aziz S, Tan SI and Syed Z (2001). Changes in serum lipid profile and malondialdehyde following consumption of fresh or heated red palm oil. *Medical J. Islamic Academy of Sci.* 14: 79-86
- Jacques VR (2005). Can red palm oil protect the heart?. *Science in Africa*(11/2/2011) <http://www.scienceinafrica.co.za/2005/september/redpalmoil.htm>
- Kamisah J, Norhayati M, Norzana G, Nor Aini, U and Ima-Nirwana, S. 2006. Effects of heated vegetable oils on serum lipids and aorta of ovariectomized rats. *Pakistan J. Nutr.* 5: 19-29
- Mukherjee S. and Mitra A (2009). Health effects of palm oil. *J. Human Ecol.* 26: 197-203.
- Oluba OM, Adeyemi O, OJIEH GC, Aboluwoye CO and Eidangbe, GO (2008). Comparative effect of soybean oil and palm oil on serum lipids and some serum enzymes in cholesterol-fed rats. *Eur. J. Sci. Res.* 23: 559-566
- Peng Z, and Stanley TO (2001).  $\beta$ -Carotene: interactions with  $\alpha$ -tocopherol and ascorbic acid in microsomal lipid peroxidation. *J. Nutr. Biochem.* 12: 38-45
- Pramod K (2006). Palm Oil: a nutritional overview. *AgroFOOD industry hi-tech*, 3: 21-23
- Radhika L, Kanga RS, Ammu R, and Kalanithi N, (2010). Palm oil: Rich in health promoting phytonutrients. *Malaysian Palm Oil Board, Sec.* 1:16-26
- Sutapa M and Analava M (2009). Health effects of palm oil. *J. Hum. Ecol.* 26(3): 197-203
- Susanna, CS, Marlien P, Welma O, Johann CJ, Magdalena JCB, and Hester HV (2004). The effect of red palm olein and refined palm olein on lipids and haemostatic factors in hyperfibrinogenaemic subjects. *Thrombosis Res.* 113: 13-25
- Wilai PMD and Donpichit LMD (2004). Is Calculated LDL-C by using the new modified friedewald equation better than the standard friedewald equation?. *J. the Medical Association of Thailand* 87(6): 589-593