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Determination of Cholesterol and β -Carotene Content in some Selected Edible Oils

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Abstract :- Determination of cholesterol and beta carotene content in some selected edible oil samples were analyzed using double beam UV-Vis spectroscopy technique at a wave length range 500nm and 445nm respectively. For the study, four edible oil samples were collected, two from palm oil variety (Viking and avena) and two from noug oil variety (selam and nur). The concentration of β-carotene and cholesterol content in four edible oil samples were determined. β-carotene content in Viking and avena oils which has 5.917 ± 0.3395mg/l, 7.107 ± 0.8083mg/l respectively, cholesterol content in Viking and avena oils which has 126.140 ± 0.2598mg/l ,66.187 ± 0.5838mg/l respectively and β-carotene content in selam and nur oils which has 8.587 ± 0.2957mg/l ,5.220 ± 0.2400mg/l respectively and cholesterol content in selam and nur oils which has 330.340 ± 0.7500mg/l ,125.290 ± 0.3122mg/l respectively reported. The result revealed that level of cholesterol and beta carotene contents in edible oil samples were in agreement with FAO/WHO recommended index.

Key words: *Beta carotene, Cholesterol, Edible oil, UV-Vis spectroscopy.*

I. INTRODUCTION

The human body uses oils and fats in the diet for three purposes as an energy source, as a structural component and to make powerful biological regulators. Oils and fats also play an important role in metabolic reactions in the human body (Khan et.al, 2007). Vegetable oils are beneficial and popular due to their cholesterol-lowering effect. In contrast to animal fats, which are predominantly saturated and hence do not react readily with other chemicals, especially oxygen, unsaturated vegetable oils are more reactive. Vegetable oils are essential in global nutrition depending on the regional conditions a variety of oils are produced in different qualities (Matalgyto & Al-Khalifa, 1998).

Oil and fats are important parts of human diet and more than 90 percent of the world production from vegetable, animal and marine source is used for food or as an ingredient in food products. Oil and fats are rich in source of dietary energy and contain more than the twice the calorie value equivalent amount of sugar .Their functional and textural characteristics contribute to the flavor and palatability of natural and prepared foods. They contain certain fatty acid which plays an important role in nutritional and are also carries of fat soluble vitamins (Hand book of food analysis part XIII, 1984).

The word “cholesterol” may quickly be associated with chronic heart disease & other heart problems. However, cholesterol also has essential functions in the body such as providing essential components of membrane and serving as a precursor of bile acids, steroid hormones and vitamin D. But there is no need to consume cholesterol in our diet. It increases the level of Low Density Lipoproteins (LDLs) (Attarde et.al, 2004). Cholesterol is a lipid play vital role in the physiological regulation of membrane fluidity and proper functioning of cells. Metabolic studies have shown that trans-fat have adverse effect on blood lipid level increasing LDL (bad) cholesterol while decreasing HDL(Good) cholesterol. This combined effect on the ratio of LDL to HDL cholesterol is double that of saturated fatty acids (Mensink et.al, 2004).

The carotenoids, whose name is derived from the fact that they constitute the major pigment in the carrot root, *Daucus carota* are undoubtedly among the most widespread and important pigments in living organisms. Carotenoids are a group of more than 700 compounds (e.g alpha-carotene, beta-carotene). The human body uses carotenoids as Vitamin A which enhances eye health. Carotenoids also play an important potential role by acting as biological antioxidants protecting cells and tissues from the damaging effects of free radicals, which could also cause cancer (Koh, 2006).

Studies also suggest that carotenoids enhance immune function by a variety of mechanisms and improve cardiovascular health. Carotenoids are present in numerous vegetable oils, including yellow maize (corn) oil, groundnut oil, soya-bean oil, rape seed oil, linseed oil, olive oil, barley oil, sunflower oil and cotton seed oil. (Ong et al., 1980).

Beta carotene is the most available and therefore important source of pro-vitamin A in the diet of most people living in developing countries, providing about 66% of vitamin A in

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their diets. The carotenoids (e.g. beta-carotene and lycopene) are micronutrient antioxidants that have integral role in regulating vital metabolic reactions in the body (Abiaka et al., 2002).

Nowadays, the major interest in carotenoids which are found in plants is not only due to their pro-vitamin A activity but also their antioxidant action of scavenging oxygen radicals and reducing oxidative stress in the organism (Rho & Honglie, 2002). Epidemiological evidence also suggests that carotenoids-rich foods protect against some chronic diseases; including certain type of cancer, cardiovascular disease and age-related macular degeneration (National Research Council, 982). In West Africa much carotene is obtained from red palm oil, which is widely used in cooking (Latham, 1997).

Absorption of visible and ultraviolet (UV) radiation is associated with excitation of electrons in both atoms and molecules from lower to higher energy levels. Since the energy levels of matter are quantized only light with the precise amount of energy can cause transitions from one level to another will be absorbed (www.rsc.org, 2009). The conventional instrument for absorption spectroscopy is the double-beam spectrophotometer. In the present study UV-Vis spectrometer was selected for determination of cholesterol and beta-carotene in edible oil sample. Beta-carotene absorbs throughout the UV region but particularly and strongly in the visible region between 400 - 500 nm with a peak at 470 nm and cholesterol absorbs with peak at 500nm.

The objective of present study was determination of cholesterol and beta carotene contents in some selected edible oil samples by using double beam UV-Vis spectroscopy technique. Before carried the analysis some laboratory quality control procedure were applied.

II. MATERIALS AND METHODS

2.1. Equipments

Digital analytical balance (AA-200DS, deriver instrument company, Germany) was used to weigh oil sample. Refrigerator (RDP6900, Japan) was used to preserve prepared samples until analysis. Conical flasks, beakers, watch glass and pipette were used in the measurement and preparation of sample and standard solutions. Volumetric flask of 25, 50, 100 and 250 mL were used to dilute sample solutions and prepare standard solutions. Double Beam UV-Vis Spectrophotometer (SL160, India) was used for determination of β- carotene and cholesterol contents.

2.2. Reagents (Chemicals)

All reagents are of analytical grade unless otherwise it will be stated. Deionized water was used for preparation and dilutions of standard solutions. Nitric acid (HNO_3) and sulfuric acid (H_2SO_4) were used for cleaning of glassware. Representative edible oils samples were used for analysis. N-hexane solvent was used for analysis β- carotene. Ferric chloride and glacial acetic acid mixture and concentrated sulfuric acid were used for determination of cholesterol in edible oil sample.

2.3. Sample collection and sampling

In the present study, the two varieties of edible oil samples were selected. Now days in Ethiopia, among different oil seed varieties greatly processed edible oil has nuog seed oil, so it was selected for the study. Under this variety two different oil samples such as selam and Nur oils were selected. These oils are branded and industrially processed. Currently in Ethiopia, vastly imported edible oil has palm oil variety, so it was selected for the study. Under this variety two oil samples such as Viking and Avena oils were selected, they were also branded and industrially processed. Representative edible oil samples were collected from merkato commercial center.

2.4. Laboratory Quality Control

2.4.1. Cleaning of Glassware

All the glassware and apparatus to be used through the entire analysis were first washed with tap water and detergent in order to keep the analyte from contamination. Next, it was soaked with deionized water and followed by rinsing with 0.5N HNO_3. Then, it was rinsed again with deionized water and a corresponding analyte solution to be analyzed based on its necessity.

2.4.2. Calibration of apparatus

AA-200DS Model laboratory analytical balance (±0.00001 g precision) was calibrated first properly using a known 100 g weighing standard provided with the instrument by the manufacturer. Seven replicate measurements were taken to check the analytical balance performance. Mean and % Er was calculated. If the calculated % Er exceeds ±5 this step should be repeated before continuing with on-going calibration of other glass wares to minimize the error tolerated with poor calibration of glass wares as well as the analytical balance.

2.5. Method for determination of β-carotene in edible oils

The absorption of solution of fatty material in cyclohexane is measured at 445nm. The percentage content of total carotenoids is calculated as beta carotene.

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One gram of each oil samples were weighing in 100mL of volumetric flask then the oil samples were dissolved in cyclohexane solvent and make up to the mark. The solution was transferred into 1cm quartz cuvette and the absorbance was measured at 445nm against hexane .The carotene content of each vegetable oil was defined and calculated at β-carotene in part per million(ppm).

$$Carotenoids\ content = \frac{V \times 383 \times (AS - Ab)}{100 \times W} \ldots (1)$$

Where, V= volume used for analysis, 383=extinction coefficient for carotenoids, As = absorbance of the sample Ab = absorbance of the blank, W= weight of the sample (British standards methods of analysis B.S-684, 1977).

2.6. Method for determination cholesterol in edible oil

1gram of each oil samples and the standard cholesterol were dissolved in 10mL of chloroform and 3mL of glacial acetic acid and 3mL of color reagent (a solution of ferric chloride and glacial acetic acid and sulfuric acid) were added to the standard cholesterol and the sample then shake vigorously to dissolve the oil. Blank contained 2mL of chloroform, 3ml of glacial acetic acid and 3mL of color reagent without the oil samples. After cooling for 30min at room temperature absorbance of the standard and the sample were read at 500nm. Cholesterol content in edible oil sample estimated using the equation:

$$Cholesterol\ (mg/L) = AB/AS \times CS \ldots . (2)$$

Where, AB= absorbance of the oil sample, AS = absorbance of standard cholesterol CS = concentration of cholesterol (Ojiako &Akubugwo, 1997).

2.6.1. Preparation of standard series for calibration curve of cholesterol

The solutions of five different cholesterol concentrations (80,100,120,140 and160ppm) were prepared by using a cholesterol standard.

Concentration(mg/L)	Absorbance
0	0.074
80	0.638
100	0.782
120	0.926
140	1.072
160	1.214

Source: own data

Each standard cholesterol solutions were treated with ferric chloride and acetic acid mixture and sulfuric acid, the absorbance of each solution was read at 500nm in UV-Vis spectrometer (Zlatikis et al., 1953) Absorbance of standard series of cholesterol solutions and blank for calibration curve at different concentrations were tabulated in table 1.

Calibration curves (absorbance versus concentration) were prepared to determine the instrument detection limit. The instrument was calibrated using a calibration blank and five series of respective working standards. The correlation coefficient of the calibration curves of the standard cholesterols presented in table.1 were in the acceptable range (0.999) under the study

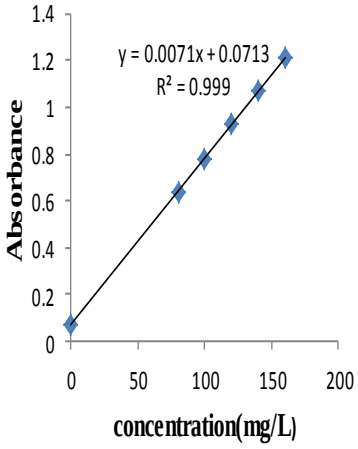

Fig .1 Calibration curve for standard cholesterol by using series of cholesterol solutions and blank

Source: own data

2.7. Statistical analysis

The significance of variation within each concentration of analyzed parameter was determined by paired comparison (t-test) between the mean. Means were followed by standard deviation throughout to indicate variation between measurements of the parameters. Microsoft Excel and statistical packages for social science version 16.0 were used for graphic illustration of edible oil parameters. The significance was set at 0.05.

III. RESULT AND DISCUSSION

β-carotene and cholesterol content in four edible oil samples were analyzed using UV-Vis spectroscopy technique at appropriate wave length range. Results were tabulated in table-2.

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Types of oil sample	β-carotene(mg/L)	Cholesterol(mg/L)
Viking oil	5.917±0.3395	126.140 ± 0.2598
Avena oil	7.107±0.8083	66.187± 0.5838
Selam oil	8.587±0.2957	330.340± 0.7500
Nur oil	5.220±0.2400	125.290± 0.3122

3.1.β-carotene

The compositions of beta carotene in four edible oil samples were analyzed using UV-Vis spectroscopy technique. The technique was carried out using blank determination at the specified range of wave length. The maximum permissible limit of beta carotene content in edible oils was 25mg/L (Codex Stan19-1981). In the present study the analyzed edible oil samples were under the recommended value. According to the analysis, within the oil samples avena oil has high beta carotene content (7.107 ±0.8083mg/L) than Viking oil (5.917 ±0.3395mg/L) from the palm oil variety and from niger/noug oil variety, selam oil has high beta carotene content (8.587 ± 0.2957mg/L) than nur oil (5.220 ± 0.2400mg/L). Beta-carotene has existed in four edible oils according to the present study. So from the observation suggest that, the analyzed edible oil samples were a good source of vitamin-A and natural antioxidants which are prevent the oxidative stress of the edible oil. According to the analysis of comparison paired t-test at 95% confidence interval ($P<0.05$) result of Viking oil and avena oil were statistically significant different ($P=0.034$) from each other and also result of selam oil and nur oil were significant different ($P=0.007$) from each other.

3.2. Cholesterol

The cholesterol content in four edible oil samples were determined using UV-Vis spectroscopy techniques at the entire wave length range. Representative analytical data for concentration of cholesterol in the edible oil samples were presented in table.6. According to the analysis, within the oil samples Viking oil has recorded high cholesterol level (126.140 ± 0.2598mg) than avena oil (66.187± 0.5838mg) from the palm oil variety and from niger/noog oil variety, selam oil has recorded high cholesterol level (330.340± 0.7500mg) than nur oil (125.290± 0.3122mg). According to the analysis of comparison paired t-test at 95% of confidence interval ($P<0.05$) result of Viking and avena oils are strongly significant different ($P=0.000$) from each other and also result of selam and nur oils are significant different ($P=0.000$) from each other.

IV. CONCULUSION

Beta carotene and cholesterol content in some selected edible oil samples were determined using spectroscopic technique. Under the study, beta-carotene has present in all edible oil samples, and the highest concentrations were reported in avena oil from palm oil variety and in selam oil from noug oil variety. Therefore possible to conclude that, the vegetable oils can be used as an alternative medicine in hostility serious health situations.

Although in all edible oil samples logo the manufacturer itemized that free from cholesterol, but the researcher reported the presence of cholesterol in all edible oil samples during the study. Viking oil from the palm oil variety shown high concentration of cholesterol and from noug oil variety selam oil has revealed high concentration of cholesterol content.

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Conflict of interest

The author(s) declare(s) that there is no conflict of interest regarding the publication of this paper."

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