ISSN 1996-0875 ©2011 Academic Journals

DOI: 10.5897/JMPR11.285

## Full Length Research Paper

# Composition of the essential oil and petroleum ether extract of *Lycium chinense* Miller fruits and antioxidant activity of its several extracts

III-Min Chung, Praveen Nagella, Young-Sup Ahn<sup>2</sup> Seok-Ju Kim<sup>1</sup> and Ateeque Ahmad\*

<sup>1</sup>Department of Applied Life Science, Konkuk University, Seoul 143-701, South Korea. <sup>2</sup>Department of Herbal Crop Research, NIHHIS, RDA, Eumseong 369-873, South Korea.

Accepted 17 May, 2011

The essential oil and petroleum ether extract from the fruits of Lycium chinense Mill. (Solanaceae), were analyzed by gas chromatography-mass spectrometry (GC-MS). Out of 42 peaks, 40 components, which constituted 99.81%, were identified in the oil. The oil was dominated by acid and esters which accounted for 66.61 and 8.43%, respectively. The major constituents were hexadecanoic acid methyl ester (4.92%), hexadecanoic acid ethyl ester (9.92 %), ethyl linoleate (3.45%), tetradecanoic acid (3.72%) and hexadecanoic acid (62.89%). Forty components, which constitute 99.50 %, were identified in the petroleum ether extract. The major components of petroleum ether extract were hexadecanoic acid (17.43%), hexadecanoic acid ethyl ester (9.51%), hexadecanoic acid methyl ester (3.72 %), ethyl linoleate (10.26%), and phytol (3.58%). The petroleum ether extract components were identified for the first time. The identity of constituents of essential oil and petroleum ether extract was confirmed on the basis of retention time and mass and supplemented library. Extracts of different polarity from fruits of L. chinense were investigated for their antioxidant activity. Two different bioassays were used, namely scavenging of the diphenylpicrylhydrazyl (DPPH) radical method and the other reducing power of Fe<sup>3</sup> method. The total phenolic content was quantified of each extract as well. Butanol, ethyl acetate and methanolic extracts contributed to the strongest antioxidant activity. The fruit extracts of medicinal plant (L. chinense) has potential as a natural antioxidant and thus inhibit unwanted oxidation process.

**Key words:** *Lycium chinense*, Solanaceae, fruits, essential oil, petroleum ether extract, gas chromatographymass spectrometry (GC-MS), antioxidant activity.

#### INTRODUCTION

Lycium chinense Miller fruits (Fructus Lycii) known as "Gou-Qi-Zi" in Chinese, has long history of application as a valuable tonic and health food supplement for improving vision and maintaining good health. It is reputed to have the properties of nourishing the blood, enriching the yin, tonifying the kidney and liver, and moistening the lungs (Pharmacopeia, 2000; Peng et al., 2005). In almost every herbal eye remedy in Chinese medicine, Lycium fruit remains one of the core

ingredients. Fruits of *L. chinense* (Solanaceae), distributed in northeast Asia, have been used as a tonic in traditional Oriental medicine and were reported to exhibit hypotensive, hypoglycemic and antipyretic activities (Funayama et al., 1980). *L. chinense* has been used especially in China for its emmenagogue, diuretic, antipyretic hepatoprotective effects and reducing the risk of certain disease as arteriosclerosis, essential arterial hypertension, diabetes and night blindness (Zargari, 1992; Yahara et al., 1993; Yang et al., 1987; Dafini and Yaniv, 1994; Rivera and Obon, 1995; Davis, 1972). The dried ripe fruit of *L. chinense* has been widely used in folk medicine as a tonic and is still used in Korea as an ingredient of herbal drugs and functional foods. Several

<sup>\*</sup>Corresponding author. E-mail: ateeque97@gmail.com. Tel: +82-2-450-3730. Fax: +82-2-446-7856.

volatile, steroidal and alkaloidal compounds in this plant are known to have various bioactivities (Sannai et al., 1983; Kim et al., 1997a; Noguchi et al., 1984; Funayama et al., 1980; Yang et al., 1987; Itoh et al., 1978). Four volatile flavor compounds (Sannai et al., 1983) were identified as 3-hydroxy-7, 8-dihydro-β-ionone, 3-hydroxy-7, 8-dihydro- $\beta$ -ionol, 3-hydroxy- $\beta$ -ionone and 3-hydroxy- $\beta$ -ionol from L. chinense leaves. The volatile oil components were reported from dichloromethane extract of leaves (Sannai et al., 1983). Forty five volatile flavor components were identified from box thorn (L. chinense) leaves (Kim et al., 1997b). The main components were (23.65)%), 2-methylbutanal (9.37%),hexadecene (8.34%), 9, 12, 12-octadecatrienal (2.10%), dodecanoic acid, methyl ester (2.65%), linoleic acid and methyl ester (2.65%).

Water distilled essential oils composition from the fruits of L. barbarum and L. ruthinicum have been reported (Altintas et al., 2006). The main components in the essential oil of L. barbarum were hexadecanoic acid (47.5%), linoleic acid (9.1%),  $\beta$ -elemene (5.4%), myristic acid (4.2%), and ethyl hexadecanoate (4.0%). The essential oil of L. ruthenicum has heptacosane (14.3%), ethyl linoleate (10.0%), hexacosane (7.0%), and nonacosane (6.2%), and ethyl hexadecanoate (5.8%) as the main compounds (Altintas et al., 2006).

The essential oil composition of the fruits of *L. chinense* has previously been reported with ethyl hexadecanoate, 1-octadecanone, tetrapyrizine, 2-furanocarboxaldehyde, and ethyl linoleate as main constituents (Park et al., 1997). The air-dried berries were extracted twice with dichloromethane and the extract gave neutral oil by ordinary fractionation. It was then steam-distilled and the volatile oil was chromatographed on silica gel and obtained 1, 2-dehydro-α-cyperone and solavetivone (Sannai et al., 1982). Thirty-three odor compounds were identified from Lycii fructus (kukija), the small berries of L. chinese Miller. The most contributing odor compounds in L. fructus were (E)-2-heptenal, 1-heptanol, hexanal, 3octanol, 1-octen-3-ol, and 2-methyl-2-butenoic acid (Lee et al., 2008). L. chinense fruits essential oil composition previously reported with very few constituents and in this paper, detailed study of maximum number of possible forty two volatile components are reported. Petroleum ether extract components has not been investigated previously and this is a new approach for identified nonpolar components of *L. chinense*.

The objective of our research was to study full chemical composition of essential oil and petroleum ether extract of *L. chinense* fruits. The identity of constituents of essential oil and petroleum ether extract was confirmed on the basis of retention time and mass and supplemented library. Antioxidant activity of several extracts of medicinal plant (*L. chinense*) fruits was reported here.

#### **MATERIALS AND METHODS**

#### Plant materials

The dried ripe fruits of *L. chinense* were purchased from the herbal drug market in Seoul, Korea in September 2010 and the voucher specimen was deposited in the herbarium of our Department, College of Life Science and Environmental, Konkuk University.

#### Isolation of volatile components

Ripe fruits of L. chinense (500 g) were subjected to hydrodistillation in Clevenger-type apparatus for a minimum of 5 h. The resulting essential oil was obtained in a yield of 0.01% w/w after drying over anhydrous sodium sulphate and stored at 4  $^{\circ}$ C until use.

# Gas chromatography-mass spectroscopy (GC-MS) analysis of essential oil

Samples of essential oil were diluted in hexane (spectroscopic grade) and analyzed in a Finnigan Focus GC/ Finnigan Focus DSQ MS system (Thermo Co., Germany) apparatus equipped with VB-WAX bonded PEG capillary column (30 m x 0.25 mm internal diameter, 0.25 µm film thickness). Helium (1 ml/min) was used as a carrier gas. Sample volume was injected in the split mode 10 µl (split less). The injector was kept at 150°C. The column was maintained at 50°C for 10 min and then programmed to 200 at 2°C and held for 30 min at 200 °C. Detector temperature was held at 250 °C. The MS was operated in EI mode at 70 eV in the in the m/zrange 25 to 350. The identification of the compounds was performed by matching their recorded mass spectra of the GC-MS data system. Quantitative data were obtained from electronic integration peak areas and comparing their retention time and mass spectra library with those found in the literature and supplemented by the Wiley (Wiley 7th Mass Spectral Library) and NIST MS Search 2.0 (National Institute of Standards and Technology) GC-MS libraries.

#### Petroleum ether extract

The *L. chinense fruit* (50 g) were immersed in petroleum ether (250 ml, 35 to 60°C) for overnight at room temperature and then the supernatant was concentrated under vacuum to yield 1.5 g of the extract, which was small sample dissolved in hexane (spectroscopic grade) and prepare sample after filtration for GC-MS analysis.

# Gas chromatography-mass spectroscopy (GC-MS) analysis of petroleum ether extract

Samples of petroleum ether extract were diluted in hexane (spectroscopic grade) and analyzed in a Finnigan Focus GC/Finnigan Focus DSQ MS system (Thermo Co., Germany) apparatus equipped with Vesteck rtx-50 capillary column (30 m x 0.25 mm internal diameter, 0.25  $\mu m$  film thickness). Helium (1 ml/min) was used as a carrier gas. Sample volume was injected in the split mode 10  $\mu l$  (split less). The injector was kept at 150 °C. The column was maintained at 50 °C for 10 min and then programmed to 200 °C for 2 min and held for 30 min at 200 °C. Detector temperature was held at 250 °C. The MS was operated in El mode at 70 eV in the in the m/z range 25 to 350. The identification of the compounds was performed by matching their recorded mass spectra of the GC-

MS data system. Quantitative data were obtained from electronic integration peak areas and comparing their retention time and mass spectra library with those found in the literature and supplemented by the Wiley (Wiley 7<sup>th</sup> Mass Spectral Library) and NIST MS Search 2.0 (National Institute of Standards and Technology) GC-MS libraries.

#### Total phenolic content

The total phenolic content was determined by the Folin-Ciocalteu (FC) method (Singleton and Rossi, 1965) and expressed as grammes of gallic acid equivalents per 100 g plant extract. Distilled water (3.16 ml) was mixed with a DMSO solution of the test compound (200  $\mu$ l). Then, 200  $\mu$ l of FC reagent was added. After 5 min, 600  $\mu$ l of 20% sodium carbonate solution was added and the solutions were mixed again. The solutions were left at room temperature for 2 h. Then the absorption of the developed blue colour was determined at 765 nm, using a Macasys Optizen 2120UV plus UV-spectrophotometer (Mecasys, Korea).

#### Free radical scavenging activity

The antioxidant activity of the different extracts (Hexane, Ethyl acetate, Methanol, Butanol and Water), based on the scavenging activity of the stable 1,1-diphenyl-2-picrylhydrazyl (DPPH<sup>-</sup>) free radical, was determined by the method described by Keterere and Eloff (2005). The different concentrations (25, 50, 100, 200 and 500 μg) of the tested samples (0.2 ml; extracts and tocopherol) were taken in different test tubes with 4 ml of a 0.006% MeOH solution of DPPH<sup>-</sup>. Water (0.2 ml) in place of the extract was used as control. Absorbance at 517 nm was determined after 30 min. Radical scavenging activity was expressed as the inhibition percentage and was calculated using the following formula:

Percentage (%) Radical scavenging activity =  $[(A_0 - A_1)/A_0] \times 100$ , where  $A_0$  is the absorbance of the control, and  $A_1$  is the absorbance of the extract/standard.

#### Reducing power

The reducing power of the *Lycium* fruit extracts was determined according to the method of Oyaizu (1986). Different extracts of concentration (25, 50, 100, 200 and 500  $\mu$ g) in 1 ml of distilled water was mixed with phosphate buffer (2.5 ml, 0.2 M/L, pH 6.6) and potassium ferricyanide [K $_3$ Fe (CN) $_6$ ] (2.5 ml, 1%). The mixture was incubated at 50°C for 20 min. A portion (2.5 ml) of trichloroacetic acid (10%) was added to the mixture, which was then centrifuged at 1000 rpm for 10 min. The upper layer of the solution (2.5 ml) was mixed with distilled water (2.5 ml) and FeCl $_3$  (0.5 ml, 0.1%) and the absorbance was measured at 700 nm. Increased absorbance of the reaction mixture indicated increased reducing power. All analysis were run in triplicate and averaged.

#### **RESULTS AND DISCUSSION**

The essential oil and petroleum ether extract of *L. chinense* fruits have been subjected to gas chromatography-mass spectroscopy (GC-MS) analysis and identified 42 and 40 constituents respectively were

reported in this paper. To the best of our knowledge, this is the complete study of their essential oil and petroleum ether extract of medicinal plant (*L. chinense*). Petroleum ether extract components are identified from *L. chinense* fruits. This is a new approach for non-polar components of *L. chinense*. Previously, petroleum ether extract has not been investigated.

Two different bioassays were described, namely; scavenging of the diphenylpicrylhydrazyl (DPPH) radical method and the other reducing power of Fe3+ method. The total phenolic content was quantified of each extract as well. Butanol, ethyl acetate and methanolic extracts contributed to the strongest antioxidant activity. The fruit extracts of *Lycium* has potential as a natural antioxidant and thus inhibit unwanted oxidation process.

#### Chemical constituents of the essential oil

The essential oil of L. chinense fruits obtained on hydrodistillation was analyzed by gas chromatography-mass spectrometry (GC-MS) (Figure 1). Forty two components, representing 99.81% of the total oil were identified. The constituents identified by GC-MS analysis, their retention time, mass data and area percentage (concentrations) are summarized in Table 1. The oil was dominated by acids and esters which accounted for 66.61 and 22.25% of the oil, respectively. The major components were hexadecanoic acid (62.89%), hexadecanoic acid ethyl ester (9.92%), hexadecanoic acid methyl ester (4.92%), tetradecanoic acid (3.72%), ethyl linoleate (3.45%), 6-Isopropenyl-4,8α-dimethyl-1, 2, 3, 5, 6, 7, 8α-octahydronaphthalen-2-ol (2.19%), dodecyl acrylate (1.17%), ethyl oleate (1.23%), 9, 12-octadecadienoic acid, methyl ester (1.56%), 9-octadecenoic acid (1.19%). The identity of constituents of essential oils was confirmed on the basis of retention time and mass and supplemented library. The chemical composition of the essential oil was however different from previously reported constituents (Sannai et al., 1983; Kim et al., 1997a, b; Altintas et al., 2006; Park et al., 1997; Sannai et al., 1982). Table 1 are summarizes current composition on the analysis of the volatile oils from L. chinense fruits. However, the comparison of our results with literature shows qualitative and quantitative differences in the composition of L. chinense fruit oil.

#### Chemical constituents of the petroleum ether extract

The petroleum ether extract of *L. chinense* fruits was analyzed by gas chromatography-mass spectrometry (GC-MS) (Figure 2). Forty components, representing 99.50% of the total extract were identified. The constituents identified by GC-MS analysis, their retention

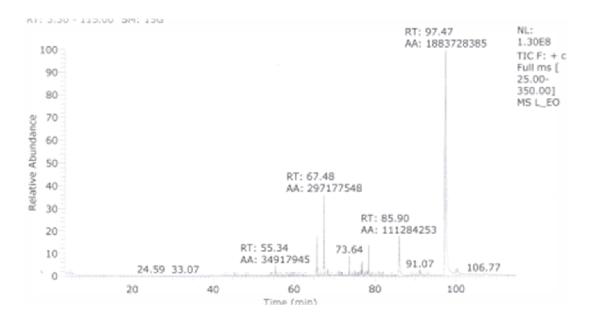


Figure 1. Gas chromatograph of *L. chinense* fruits essential oil.

time, mass data and area percentage (concentrations) are summarized in Table 2. The extract was dominated by acids, esters and higher alkanes which accounted 23.76, 31.77 and 9.52%, respectively. The major components petroleum ether of extract hexadecanoic acid (17.43%), hexadecanoic acid ethyl ester (9.51%), hexadecanoic acid methyl ester (3.72 %), ethyl linoleate (10.26 %), and phytol (3.58 %). The identity of constituents of petroleum ether extract was confirmed on the basis of retention time and mass and supplemented library. Table 2 are summarizes current composition on the analysis of the petroleum ether extracts from *L. chinense fruit*. The chemical composition of the petroleum ether extract was reported for the first time.

#### **Antioxidant activity**

## Total phenolic content

The distribution of phenolic compounds in *Lycium* fruits demonstrated that the ethyl acetate extract contained highest amount 9.25 g gallic acid equivalents (GAE) per 100 g extract, followed by water extract with amount of 6.67 g GAE per 100 g extract, butanol extract (5.69 g), methanol extract (2.94 g) and hexane extract (2.22 g). The Folin-Ciocalteu (FC) method is actually not an antioxidant test but instead an assay for the quantity of

oxidizable substances, that is, phenolic compounds. Correlations between the content of phenolic compounds and antioxidant activity are described (Escrig et al., 2001; Nuutila et al., 2003).

#### DPPH<sup>-</sup> radical-scavenging activity

The free radical-scavenging activity the polysaccharides was tested through DPPH method (Katerere and Eloff, 2005) and the results were compared with tocopherol (Figure 3). DPPH is usually used as a substrate to evaluate antioxidative activity of antioxidants. The method is based on the reduction of methanolic DPPH solution in the presence of a hydrogen donating antioxidant, due to the formation of the non-radical form DPPH-H by the reaction. The extract was able to reduce the stable radical DPPH to the yellow-coloured diphenylpicrylhydrazine. It has been found that cysteine, glutathione, ascorbic acid, tocopherol, polyhydroxy aromatic compounds (e.g., hydroquinone, pyrogallol, gallic acid), and aromatic amines (e.g., p-phenylene diamine, p-aminophenol), reduce and decolorize 1, 1diphenyl-2-picrylhydrazyl by their hydrogen donating ability (Blois, 1958). The content of total phenolic compounds in the extracts might explain their high antioxidant activities. In this study different extracts from L. chinense fruits also showed a remarkable antioxidant activity, one of the possible mechanisms is polyphenolic-

**Table 1.** Composition of the essential oil (%) of *L. chinense* fruits.

S/N	Retention time	Compounds	Percentage (%)
1	4.52	Hexanal	0.15
2	4.60	2,4-Pentadienal	0.19
3	24.59	u.i	t
4	33.07	u. i	t
5	43.13	(E,E)-2,4-Decadienal	t
6	45.27	(E,Z)-2,4-Decadienal	0.29
7	48.07	(Z) 6,10-Dimethyl-5,9-undecadien-2-one	0.14
8	54.23	1-Dodecanol	0.31
9	55.34	Dodecyl acrylate	1.17
10	56.37	2-Pentadecanone	0.12
11	58.12	Tetradecanoic acid, ethyl ester	0.41
12	59.46	Nonanoic acid, 9-oxo- ethyl ester	0.23
13	59.87	Octanoic acid	0.13
14	60.57	9-Methoxycalamenene	0.11
15	61.50	6,10,14-Trimethyl-2-pentadecanone,	0.16
16	62.84	Pentadecanoic acid, ethyl ester	0.12
17	65.67	Hexadecanoic acid, methyl ester	4.92
18	66.60	(Z) 9-Hexadecenoic acid, methyl ester,	0.21
19	67.48	Hexadecanoic acid, ethyl ester	9.92
20	68.30	Ethyl 9-hexadecenoate	0.70
21	69.40	(Z) 9-Octadecenoic acid	0.13
22	71.01	Isopropyl palmitate	0.25
23	71.29	1-Napthalenamine 4-bromo	t
24	71.82	( <i>E,E</i> )-5,9,13-Pentadecatrien-2-one 6,10,14-trimethyl	0.35
25	73.64	6-Isopropenyl-4,8α-dimethyl-1,2,3,5,6,7,8α-octahydro-naphthalen-2-ol	2.19
26	74.44	Octadecanoic acid, methyl ester	0.21
27	75.05	(Z)-9-Octadecenoic acid, methyl ester	0.59
28	75.99	Octadecanoic acid, ethyl ester	0.29
29	76.55	Ethyl oleate	1.23
30	76.89	(Z,Z)-9,12-octadecadienoic acid, methyl ester	1.56
31	77.99	Dodecanoic acid	0.43
32	78.36	Ethyl linoleate	3.45
33	79.46	9,12,15-Octadecatrienoic acid, methyl ester	0.21
34	80.86	9,12,15-Octadecatrienoic acid, ethyl ester	0.40
35	81.99	Phytol	0.33
36	84.09	Dibutyl phthalate	0.14
37	85.90	Tetradecanoic acid	3.72
38	89.29	9-Hexadecenoic acid	0.11
39	91.06	Pentadecanoic acid	0.64
40	92.91	Oleic acid	0.22
41	97.47	Hexadecanoic acid	62.89
42	100.15	9-Octadecenoic acid	1.19

associated compounds (formation of non-extractable complex between high molecular weight phenolics and polysaccharides). Those kinds of phenolic compounds show antioxidant activity due to their redox properties,

which play an important role in absorbing and neutralizing free radicals, quenching singlet and triple oxygen or decomposing peroxide. As shown in Figure 3, the different extracts exhibited a concentration-dependent

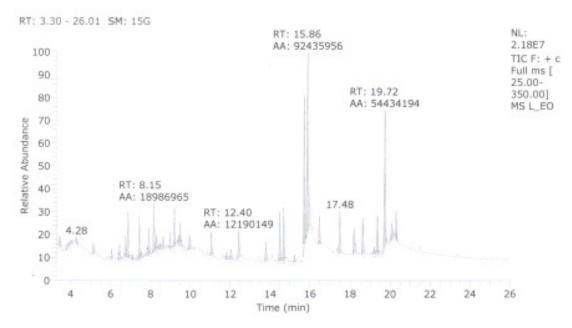


Figure 2. Gas chromatograph of *L. chinense* fruits petroleum ether extract.

antiradical activity by inhibiting DPPH radical. Three of the selected extracts from the Lycium fruits exhibited good free radical scavenging activities that is, above 90% (Figure 3), the activity increased with increasing concentration. Of the different extracts from the Lycium fruits, butanol, ethyl acetate and methanol extract exhibited the highest activity of more than 90%. The DPPH activity of tocopherol showed higher degree of free radical-scavenging activity than that of the extracts at each concentration points. Similar to our results (Wangsteen et al., 2004) reported that ethyl acetate extract from the coriander leaves exhibited highest antioxidant activity when compared with other extracts. Whereas Li et al. (2007) reported that the polysaccharide fraction from the fruits of Lycium barbarum exhibited a weak DPPH activity. This is similar to other studies wherein they have reported that only 0.3 mg/ml tocopherol, 0.23 mg/ml BHT and 0.1 mg BHA exhibited a free radical scavenging activity equivalent to 3.9 mg/ml of red bean and 10 mg/ml of sesame coat extract (Chang et al., 2002; Chung et al., 2002).

### Reducing power

Antioxidant effect exponentially increases as a function of the development of the reducing power, indicating that the antioxidant properties are concomitant with the development of reducing power (Tanaka et al., 1988; Okuda et al., 1983) have reported that the reducing

power of tannins from medicinal plants prevents liver injury by inhibiting formation of lipid peroxides. Reductones are believed not only to react directly with peroxides but also prevent peroxide formation by reacting with certain precursors. As seen in Figure 4 reducing power of the different extracts of Lycium increased with increasing concentration from 25 to 500 µg. Reducing power of the Lycium extracts followed the order- ethyl acetate < water < butanol < methanol < hexane. The activity of tocopherol was pronouncedly higher than the test samples. This is in line with the observations of several other workers, where the reducing power of BHT and tocophero (Chang et al., 2002), and BHA (oktay et al., 2003) was higher than that of the extracts. In the present study, though the Lycium extracts exhibited a moderate reducing power, they did have an activity that reveals that the *Lycium* fruit extracts are electron donors and can react with free radicals and convert them to stable products thus, terminating the free radical chain reactions.

#### Conclusion

L. chinense has been used as traditional Chinense medicine and it has long history of applications. The chemical composition of volatile oil, and the petroleum ether extract and antioxidant activity of hexane, EtOAc, MeOH, BuOH and water extracts have been studied. L. chinense fruits essential oil composition previously

**Table 2.** Composition of the petroleum ether extract (%) of *L. chinense* fruits.

S/N	Retention time	Compounds	Percentage (%)
1	3.43	2-Methyloctyl benzene	1.04
2	3.83	Hexanal	0.89
3	3.98	u. i	1.10
4	4.28	7-Methyl tetracycloheptane	0.78
5	5.13	1,6-Anhydro-α-D-glucopyranose (levoglucosan)	1.01
6	6.03	1,3-Propanediol-2-ethyl-2-hydroxymethyl	0.75
7	6.44	Hexadecane	1.52
8	6.74	Tridecane	1.06
9	6.87	5-Bromo-3-methylidene-1-methoxycyclohexane	3.02
10	7.10	3-Octylundecyl benzene	0.34
11	7.43	Tetradecane	2.79
12	7.53	Limonen-6-ol pivalate	0.52
13	7.91	(E,E)-2,4-decadienal	2.34
14	8.15	Phytol	3.58
15	8.28	Docosane-11-decyl	0.99
16	8.44	3-Hydroxy dodecanoic acid	0.44
17	8.63	10-Methyl-8-tetradecen-1-ol acetate	1.44
18	8.97	2,6,10-Trimethyl tetradecane	1.08
19	9.18	(-)-Spathulenol	2.60
20	9.48	Oleic acid	2.16
21	9.94	Dotriacontane	1.40
22	11.04	3-(2-Pentenyl)-1,2,4-cyclopentanetrione	1.59
23	11.82	D-Streptamine-O-2-amino-2-deoxy-α-D-glucopyranosyl–(14)-O-[O-2,6-diamino-2,6-dideoxy-α-L-iodopyranosyl-13]-α-D-ribofuranosyl-(15)]-2-deoxy	0.93
24	12.02	Fumaric acid, ethyl 2-(2-methylenecyclopropyl) propyl ester	0.78
25	12.40	2-Benzofuranone, 5,6,7,7α-trimethyl	2.30
26	13.77	trans-Longipinocarveol	1.42
27	14.48	2-Amino-4,7-pteridinedione	3.24
28	14.67	Hexadecanoic acid, methyl ester	3.72
29	15.22	u.i	0.48
30	15.70	Hexadecanoic acid, ethyl ester	9.51
31	15.86	Hexadeacnoic acid	17.43
32	16.46	u.i	1.98
33	17.48	Heptyl p-toluenesulphonate	3.37
34	18.20	u. i	2.27
35	18.63	8,11-Octadecadienoic acid, methyl ester	2.60
36	19.20	Ethyl linoleolate	0.49
37	19.37	Ethyl oleate	3.04
38	19.72	Ethyl linoleate	10.26
39	20.08	(Z,Z)-9, 12-octadecadienoic acid	1.72
40	20.30	(Z,Z,Z)-9,12,15-octadecatrienoic acid	2.01

reported with very few constituents and in this paper, detailed study of maximum number of possible volatile components are reported. Petroleum ether extract components have not been investigated previously and this is a new approach for non-polar components. *L*.

chinense fruit might be rich in phytoconstituent that could act as strongest antioxidant. Antioxidant capacity was observed a good correlation between antioxidant capacity and total phenolic contents. Various extracts provide different antioxidants, which in general demonstrated

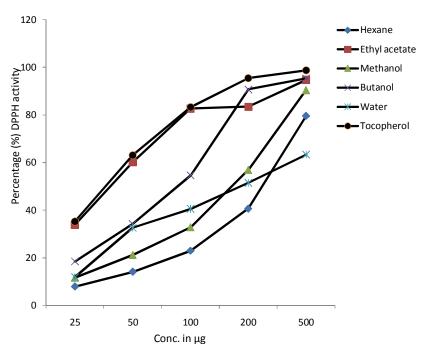


Figure 3. Free radical scavenging activity as determined by DPPH method.

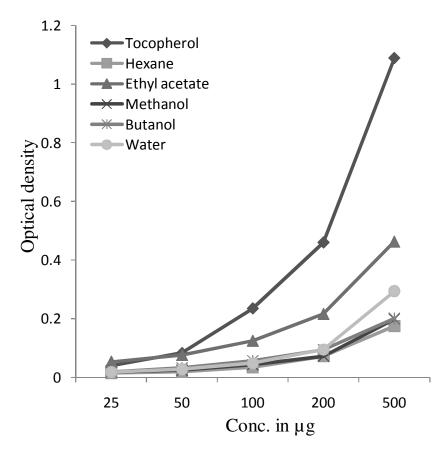


Figure 4. Reducing power of different extracts from Lycium fruits and tocopherol.

strong activities of butanol, ethyl acetate and methanol extracts. There is need of this *L. chinense* fruits for further investigation.

#### **ACKNOWLEDGEMENTS**

This work was carried out with the support of Cooperative Research Program for Agriculture Science and Technology Development (Project No. PJ 906938), Rural Development Administration, Republic of Korea.

#### **REFERENCES**

- Altintas A, Kosar M, Kirimer N, Baser KHC, Demirci B (2006). Composition of the essential oils of *Lycium barbarum* and *L. ruthenicum* fruits. Chem. Nat. Compds., 42: 24-25.
- Blois MS (1958). Antioxidant determinations by the use of a stable free radical. Nature, 181: 1199-1200.
- Chang LW, Yen WJ, Huang SC, Duh PD (2002). Antioxidant activity of sesame coat. Food Chem., 78: 347-354.
- Chung YC, Chang CT, Chao WW, Li CF, Chu ST (2002). Antioxidative Activity and Safety of the 50 Ethanolic Extract from Red Bean Fermented by *Bacillus subtilis* IMR-NK1. J. Agric. Food Chem., 50: 2454-2458.
- Dafini A, Yaniv Z (1994). Solanaceae as medicinal plants in Israel. J. Ethnopharmacol., 44: 11-18
- Davis PH (1972). Flora of Turkish and East Aegean Islands: Univ. Press; Edinburg, 6: 445-449.
- Escrig AJ, Jimenez IJ, Pulico RFS, Calixto FS (2001). Antioxidant activity of fresh and processed edible seaweeds. J. Sci. Food Agric., 81: 530-534.
- Funayama S, Yoshida SK, Konno C, Hikino H (1980). Structure of kukoamine A, a hypotensive principle of *Lycium chinense* root barks. Tetrahedron Lett., 21: 1355-1356.
- Itoh T, Tamura T, Matsumoto T (1978). Four new and other  $4\alpha$ -methylsterols in the seeds of Solanaceae. Phytochemistry, 17: 971- 977.
- Kim SY, Lee KH, Chang KS, Bock JY, Jung MY (1997a). Taste and flavor compounds in box thorn (*Lycium chinense* Miller) leaves. Food Chem., 58: 297-303.
- Kim SY, Choi Y, Huh H, Kim J, Kim YC, Lee HS (1997b). New Antihepatotoxic Cerebroside from *Lycium chinense* Fruits. J. Nat. Prods., 60: 274-276.
- Katerere DR, Eloff JN (2005). Antibacterial and antioxidant activity of Sutherlandia frutescens (Fabaceae), a reputed Anti-HIV/AIDS phytomedicine. Phytother. Res., 19: 779-781.
- Li XM, Li XL, Zhou AG (2007). Evaluation of antioxidant activity of the polysaccharides extracted from *Lycium barbarum* fruits *in vitro*. Eur. Polym. J., 43: 488-497.
- Lee GH, Shin Y, Oh MJ (2008). Aroma active components of *Lycii fructus* (kukija). J. Food Sci., 73: 500-505.

- Noguchi M, Mochida K, Shingu T, Kozuka M, Fujitani K (1984). About the components of the Chinese drug "Ti-ku'pi. 'l. Isolation and constitution ofLyciumamid, a new dipeptide. Chem. Pharm. Bull., *32*: 3584-3587.
- Nuutila AM, Puupponen-Pimia R, Aarni M, Oksman-Caldentey KM (2003). Comparison of antioxidant activities of onion and garlic extracts by inhibition of lipid peroxidation and radical scavenging activity. Food Chem., 81: 485-493.
- Okuda T, Kimura Y, Yoshida T, Hatano T, Okuda H, Arichi HS (1983). Studies on the Activities of Tannins and Related Compounds from Medicinal Plants and Drugs. I. Inhibitory Effects on Lipid Peroxidation in Mitochondria and Microsomes of Liver. Chem. Pharma. Bull., 31: 1625-1631.
- Oktay M, Culcin I, Kufrevioglu OI (2003). Determination of *in vitro* antioxidant activity of fennel (Foeniculum vulgare) seed extracts. Food Sci. Technol., 36: 263-271.
- Oyaizu M (1986) Studies on product of browning reaction prepared from glucose amine. Jpn. J. Nutr., 44: 307-315.
- Park WJ, Bock JY, Baik SO, Han SB, Ju HK (1997). Volatile components of *Lycium chinense* Miller. Korean Soc. Foods Nutr., 10: 1-5.
- Peng Y, Ma C, Li Y, Leung KSY, Jiang ZH, Zhao Z (2005). Quantification of Zeaxanthin Dipalmitate and Total Carotenoids in *Lycium* Fruits (Fructus Lycii). Plant Foods Hum. Nutr., 60: 161-164.
- Pharmacopoeia of the People's Republic of China (2000). Chemical Industry Press, (Beijing).
- Rivera D, Obon C (1995). The ethnopharmacology of Madeira and Porto Santo Islands, a review. J. Ethnopharmacol., 46: 73-93.
- Sannai A, Fujimori T, Katie K (1982). Isolation of (–)-1,2-dehydro-α-cyperone and solavetivone from Lycium chinense. Phytochemistry, 21: 2986-2987.
- Sannai A, Fujimori TK, Kato K (1983). Isolation of 3-hydroxy-7,8-dehydro-β-ionone from *Lycium chinense* M. Agric. Biol. Chem., 47: 2397-2399.
- Singleton VL, Rossi JA (1965). Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. Am. J. Enol. Vitic., 37: 144-158.
- Tanaka M, Kuie CW, Nagashima Y, Taguchi T (1988). Application of antioxidative maillard reaction products from histidine and glucose to sardine products. Nippon Suisan Gakkaishi, 54: 1409-1414.
- Wangsteen H, Samuelsen AB, Malterud KE (2004). Antioxidant activity in extracts from coriander. Food Chem., 88: 293-297.
- Yang LL, Yen KY, Kiso Y, Kikino YH (1987). Antihepatotoxic actions of formosan plant drugs. J. Ethnopharmacol., 19: 103-110.
- Yahara S, Shigeyama C, Ura T, Wakamatsu K, Yashuhara T, Nohara T (1993). Cyclic Peptides, Acyclic Diterpene Glycosides and Other Compounds from Lycium chinense Mill. Chem. Pharm. Bull., 41: 703-709.
- Zargari A (1992). Medicinal Plants of the World: Chemical constituents, traditional and modern. Vol 3, 5<sup>th</sup> (eds), Tehran University Publications, No. 1810/3, Tehran, Iran, Book 3, P. 889.