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Chemical Composition of Mongolian Sea-buckthorn (*Hippophae rhamnoides* L.) Fruits

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Abstract

Sea-buckthorn fruits (*Hippophae rhamnoides* L.) are used in Mongolia as traditional medicine due to the health-benefits associated with its bioactive components. The purpose of this study was to investigate the chemical composition of Mongolian sea-buckthorn fruits. In terms of proximate composition, crude fat content was the highest, whereas its crude ash content was the lowest. In organic acid contents, malic acid content (6.30±0.005 mg/g) was the highest. Free sugars were composed of sucrose, xylose, glucose and mannitol. Vitamin C and carotenoid content were 320 mg/100 g and 715.25 mg/100 g, respectively. The major fatty acids were palmitic (C16:0), palmitoleic (C16:1), caprylic (C8:0) and linoleic (C18:2) acid.

Key words : **Mongolian sea-buckthorn fruits, proximate composition, malic acid, palmitic acid.**

Introduction

Sea-buckthorn (*Hippophae rhamnoides* L.), a member of the family *Elaeganaceae*, is a hardy shrub. Sea-buckthorn is a dioecious species which means that the male and female flowers are on separate trees. It bears delicious and nutritious orange colored berries,widely distributed throughout the temperate zone of Asia, Europe and all over subtropical zones at high altitudes. The genus name *Hippophae* is classical Latin for “shining horse” (‘Hippo’- horse and ‘phaos’- to shine) a name that was given in ancient times after it was found that feeding the leaves to horses improved the health and luster of their hair (Lu 1992). The sea-buckthorn industry has been thriving in Russia since 1940 when scientists began to study bioactive substances found in its berries, leaves, and bark. The first Russian factory for sea-buckthorn product development was located in Bisk. These products were utilized in the diet of Russian cosmonauts and as a cream for protection from cosmic radiation (Delabays and Slacanin 1995). Sea-buckthorn cultivation in Mongolia began in the 1960s and related studies were begun (Ochirbat *et al.* 1986). In 1977, sea-buckthorn was officially listed in the Chinese Pharmacopoeia by the Ministry of Public Health as a traditional medicine and more than ten different drugs have been developed from sea-buckthorn. Its berries are a rich source of phytochemicals such as ascorbic acid, tocopherols, fatty acids, and organic acids (Beveridge *et al.* 1999, 2001), phenols and carotenoids (Gao *et al.* 2000; Yang and Kallio 2001). These chemical components of the berries are influenced by growing conditions, cultivars, ripeness and harvest time (Beveridge *et al.* 1999). In Mongolia, sea-buckthorn berries are widely cultivated in spite of severely adverse growing condition. Even so, the chemicals and nutrients of the fruits have not been well studied. Therefore, the proximate composition and organoleptic chemical components of Mongolian sea-buckthorn fruits have been studied and are reported in this paper.

Materials and Methods

Raw materials

Sea-buckthorn fruits(Chatsargana) were picked at an orchard in the Khovdo region in Mongolia, at the late-August in 2008, and stored at a refrigerator (-20℃). After eliminating the seeds, peels and pulps of the fruits were lyophilized using a freeze dryer and used as the analytical sample.

Table 1. Proximate composition of Mongolian sea-buckthorn fruit

Proximate component (g/100 g)					
Moisture	Crude protein	Crude fat	Crude fiber	Crude ash	Nitrogen free extract
4.67 ±0.647[1)]	4.49 ±0.5	50.3 ±0.735	9.17 ±0.5	1.7 ±0.042	29.67 ±0.356

1)Data are exhibited as the mean±standard (n=3).

Proximate composition determination

Moisture content was determined by the standard vacuum oven method, and crude protein was determined by the Kjedhal method. The crude fat content was determined by the Soxhlet method. Crude fiber, and crude ash determination were also executed (AOAC 1998).

Organic acidscontent

The content of organic acids were determined by a modified method (Tiitinen *et al.* 2005), using anHPLC (Waters, Milford, MA, USA) associated with refractive index (RI) detector (KI-410, Waters, USA) with a Shodex Rspak KC-811 column (8.0×300 mm, i.d). The mobile phase was 0.1% phosphoric acid with the flow rate of 1.0 mL/min at 40℃.

Table 2. Organic acid contents of Mongolian sea-buckthorn fruit

Organic acid (mg/g)				
Malic acid	Tartaric acid	Quinic acid	Citric acid	Succinic acid
6.30±0.005[1)]	5.55±0.004	1.67±0.007	0.31±0.002	0.29±0.005

1)Data are exhibited as the mean±standard (n=3).

Free sugar contents

The content of free sugars was determined by a HPLC (Waters, Milford, MA, USA) with RI detector (KI-410, Waters, USA) in a Sugar-Pak I Column (6.5×300 mm, id.) at 90℃. The mobile phase was 0.005% Ca-EDTA in the flow rate of 0.5 mL/min.

Table 3. Free sugar content of Mongolian sea-buckthorn fruit

Free sugar (mg/g)				
Glucose	Fructose	Sucrose	Xylose	Mannitol
3.66±0.01[1)]	-[2)]	38.3±0.02	9.13±0.03	0.90±0.02

1)Data are exhibited as the mean±standard (n=3)
2)Not detected.

Vitamin C content

Vitamin C content was determined by the 2, 4-dinitrophenyl hydrazine (DNP) method.

Total carotenoid content

Total carotenoid content was examined by the spectrophotometric method (AOAC 1998). Fatty acid composition

The lipid extraction from the dried fruits was carried out by the method of Bligh and Dyer (1959), and the lipid extract was saponified with 15 mL of 1N KOH in ethanol. Fatty acid methyl ester (FAME) was prepared by 14% BF3/MeOH after acidification of the saponifiables, and fatty acid composition was determined using a gas chromatograph (GC-17A, Shimadzu CO., Kyoto, Japan) equipped with a flame ionization detector (FID). A capillary column (SP 2560 fused silica column 30 m × 0.25 mm ID, 0.20 µM film) was used. The chromatographic conditions were as follows: initial temperature, 80℃, for 10 min; program rate, 4℃/min; final temperature, 240℃; injector temperature, 220℃; detector temperature, 240℃; carrier gas, H_2 at 0.6 mL/min. By comparing the retention time of standard FAME compounds under the same conditions, each of the fatty acids were identified and the contents were expressed as the ratio of total fatty acids.

Table 4. Vitamin C and total carotenoid content of Mongolian sea-buckthorn fruit

Component	Vitamin C	Total carotenoid
Content (mg/100g)	320±8.60[1)]	715.25±5.50

[1)]Data are exhibited as the mean±standard (n=3)

Statistical treatment

All determinations were carried out in triplicate, and the experimental results were expressed as mean± standard. Statistical analysis was performed with the Statistical Analysis System (SAS version 9.1, Institute Inc., Cary, NC, USA).

overview of the nutritional value of a food and includes analysis of the ash, moisture, lipid, fiber and protein content (Kirk 1991). In chemical composition, the crude fat content reached much higher values than those of the crude protein, crude fiber, and crude ash. Sea-buckthorn fruits have a lot of oil content both in their seeds and soft parts including the peels and pulps (Yang and Kallio 2001). Moreover, their oils have become popular as health food ingredients and supplements (Johansson *et al.* 2000) because of their high contents of tocopherol and carotenoids. Nitrogen free extract was calculated by the subtraction from the total content.

Organic acids contents

Organic acids are major organoleptic components of sea-buckthorn fruit. Large variations in the organic acid contents of sea-buckthorn fruit have also been reported fromdifferent origins. The organic acid of sea-buckthorn fruits were composed of malic acid (6.30 mg/100 g), tartaric acid (5.55 mg/100 g), quinic acid (1,67 mg/100 g), citric acid (0.31 mg/100 g), and succinic acid (0.29 mg/100 g). Malic acid and tatraic acid were the major organic acids in sea-buckthorn fruits which

Table 5. Fatty acid content of Mongolian sea-buckthorn fruit

SFA[1)]	C8:0	C14:0	C15:0	C16:0	C18:0	C20:0	C22:0	Sum
Content (g/100g)	2.2±0.01[2)]	0.2±0.2	0.1±0.05	7.2±0.02	0.4±0.02	0.1±0.01	0.1±0.01	10.3±0.32
USFA[3)]	C14:1	C16:1	C18:1	C18:2	C18:3	C20:1	C22:1	Sum
Content (g/100g)	0.4±0.04	2.9±0.1	0.7±0.04	2.2±0.02	0.1±0.02	0.2±0.01	0.1±0.1	6.2±0.24

[1)]Saturated fatty acid
[2)]Data are exhibited as the mean±standard (n=3)
[3)]Unsaturated fatty acid.

Results and Discussion

Proximate composition of Mongolian sea-buckthorn fruit

The proximate composition provides a general

accounted for about 90% of total organic acids. The organic acid profile and the compositions of both quinic and malic acid could be used as a chemical marker for the evaluation of the quality of sea-buckthorn juice (Beveridge *et al.* 2002).

Free sugar contents

Free sugars can also be important orgenoleptic chemicals of fruits. Therefore, free sugar content playsa useful role in determining the sweetness of fruits and their processed products. Sucrose (38.3 mg/100 g) was a major sugar component in Mongolian sea-buckthorn fruit. In addition, glucose (3.66 mg/100 g), xylose (9.13 mg/100 g), and mannitol (0.90 mg/100 g) were analyzed. Although glucose and fructose accounted for around 90% of the total sugar content in sea-buckthorn fruits originating from China and Russia (Ma *et al.* 1989), fructose was not detected in this study. The high content of malic acid with low sugars has been commonly known to the typical organoleptic quality of sea-buckthorn fruit (Tiitinen *et al.* 2005).

Vitamin C and total carotenoid content

The Vitamin C content varied from 28 to 310 mg/100 g in the fruits of European subspecies *rhamnoides* (Gao *et al.* 2000), and from 200 to 2500 mg/100 g in Chinese subspecies sinesnsis (Yao *et al.* 1992). The vitamin C content of the Mongolian sea-buckthorn fruit was 320mg/100g. Various colors of ripened sea-buckthorn fruit, from yellow to bright red, are related to the occurrence of carotenoid compounds. Plant carotenoid compounds are the precursor of vitamin A and take a role in preventing several cardiovascular and degenerative eye diseases such as macular degeneration and cataracts. The total carotenoid content of the Mongolian sea-buckthorn fruit was 715.25 mg/100 g. The carotenoid contents of the fruits may differ depending on their growth conditions and the maturity (Ochirbat *et al.* 1986).

Fatty acid composition

The fatty acid composition of seeds and berries of *H. rhamnoides* growing in different regions of the world have been extensively reported (Yang and Kallio 2001). In contrast to the fatty acid composition of seed oils which are rich in linoleic and α-linoleic acid, sea-buckthorn fruit oils have been reported to be rich in palmitic acid (C16:0) and palmitoleic acid (C16:1) (Yang and Kallio 2002). The saturated fatty acid (SFA) and unsaturated fatty acid (USFA) composition of Mongolian sea-buckthorn fruit are shown in Table 5. The major fatty acids werepalmitic (C16:0), palmitoleic (C16:1), caprylic (C8:0) and linoleic (C18:2) acid. Palmitic acid (C16:0) in the Mongolian sea-buckthorn fruits reached about 40 % of the total fatty acids.

Conclusion

The chemical composition of Mongolian sea-buckthorn fruits was investigated. Crude fat accounted for the highest percentage of the proximate composition. Malic acid and sucrose were major organoleptic components in Mongolian sea-buckthorn fruits. The vitamin C and carotenoid contents were fairly high. The major fatty acids were palmitic and palmitoleic acids.

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