



## Amino Acid Content in Different Cultivars of *Liriope platyphylla*

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*Liriope platyphylla* is a well-known herb in traditional Chinese medicine and its root has been clinically demonstrated to be effective in the treatment of metabolic and neural disorders. The present study investigated the variation in amino acids from different cultivars of *L. platyphylla*. Twenty amino acids were detected in four different cultivars of *L. platyphylla*. The presence and amounts of amino acids varied greatly among the cultivars. Seongsoo contained the highest total amount of amino acids with 2.18, 1.77 and 1.762 times the total amino acid levels observed in the Liriope Tuber No. 1, Cheongsim and Cheongyangjaerae cultivars, respectively. The Seongsoo cultivar contained the highest levels of 13 different amino acids, with a marked variation observed in the amount of glutamine. The Seongsoo cultivar contained 2.84, 2.01 and 1.85 times the amount of glutamine detected in the Liriope Tuber No. 1, Cheongyangjaerae and Cheongsim cultivars, respectively. Furthermore, in the Seongsoo cultivar, the levels of asparagine, arginine, threonine,  $\gamma$ -aminobutyric acid, leucine and lysine were 3.04, 2.75, 1.89, 1.73, 1.74 and 1.68 times, respectively, higher than that in the Cheongsim cultivar, which showed the lowest amino acid content of all cultivars. The cultivar Cheongyangjaerae contained the highest levels of six different amino acids: histidine, alanine, tyrosine, cysteine, valine and isoleucine. The levels of tyrosine, alanine, valine, isoleucine, cysteine and histidine were 1.79, 1.72, 1.67, 1.61, 1.36 and 1.28 times, respectively, higher than that in the Cheongsim cultivar. Although the Cheongsim cultivar contained the lowest total amount of amino acids and the levels of most of the amino acids were the lowest, the tubers of this cultivar contained the highest level of glutamate. Our results therefore demonstrate that *L. platyphylla* cultivars contain varying amounts of amino acids and suggested that the Cheongsim cultivar shows excellent potential to serve as a source of amino acids.

**Keywords:** Amino acids, Cultivars, *Liriope platyphylla*, Herbaceous perennial plant.

### INTRODUCTION

*Liriope platyphylla* Wang et Tang is a herbaceous perennial plant of the lily family (Liliaceae) and is commonly used for medicinal purposes. "Radix Liriope" the tuber of the liriopis plant, is a medicinal ingredient traditionally used in several Asian countries. Based on ancient records, these tubers have been utilized for hundreds of years to relieve the symptoms of a dry hacking cough, dry tongue and mouth, insomnia and constipation. Modern scientific studies have also confirmed that Liriope root extract reduces inflammation and alleviates symptoms of metabolic and neurodegenerative disorders, bacterial illness, diabetes and pathological obesity [1-6]. It is also believed to improve learning and memory and to delay aging [7]. In Korea, it has traditionally been used as a tonic, as an antitussive agent and as an expectorant [5]. Several steroidal saponins have been identified as the main active constituents of *L. platyphylla* [8,9]. One of the major steroidal saponins, spicatoside A, has been reported to induce neurite outgrowth

[10]. Constituents isolated from Liriope have recently been demonstrated to possess anticancer activity [11,12].

About 65 % of the human protein requirement comes from plant sources [13]. The most important function of amino acids is their role as protein building blocks. Numerous researchers have also reported antioxidant effects by several amino acids [14-17]. Free amino acids are needed in secondary plant metabolism and in the biosynthesis of compounds such as the glucosinolates and phenolics that play important roles, either directly or indirectly, in plant-environment interactions and human health [18].

Although there are many amino acids that exist in nature, not all are essential for human health. Approximately 24 different amino acids are reported to be essential to human nutrition [19,20]. Several studies have addressed the medicinal values of the tuber of *L. platyphylla*. Up to now, no clearly documented information regarding the amino acid content in the different cultivars of *L. platyphylla* has been reported. The objectives of this study were to examine the different cultivars of

*L. platyphylla* in order to identify the amino acids present and to assess their levels.

## EXPERIMENTAL

The four cultivars of *L. platyphylla* (Seongsoo, Cheongyangjaerae, Cheongsim and Liriope Tuber No. 1) were maintained in a greenhouse at the Cheongyang Boxthorn Experiment Station, Cheongyang, Korea. Twenty-five plants of each cultivar were grown under the same conditions in the greenhouse. The tubers of the four cultivars of *L. platyphylla* were harvested on April 30, 2015. Immediately after being harvested, the samples were freeze-dried at -80 °C for at least 72 h and then ground with a mortar and pestle into a fine powder for amino acid analysis.

Trichloroacetic acid (TCA, 99.0 %) was obtained from Samchun Pure Chemical Co., Ltd. (Pyeongtaek, Korea). Sixteen amino acid standards and four amino acid supplements were obtained from Agilent Technologies (Waldbronn, Germany). Vitamin U (DL-methionine methylsulfonium chloride) standards and sodium phosphate monobasic monohydrate (NaH<sub>2</sub>PO<sub>4</sub>) were purchased from Sigma-Aldrich (St. Louis, MO, USA). High performance liquid chromatography (HPLC)-grade acetonitrile (ACN) and methanol (MeOH) were supplied by J.T. Baker (Phillipsburg, NJ, USA). Ultrapure water with a resistivity of 18.2 MΩ/cm was produced by PureLab Option system from ELGA LabWater (Model LA 621, Marlow, UK).

**Extraction and high performance liquid chromatography analysis of free amino acids:** One hundred milligram of freeze-dried plant powder was measured and placed in a 2 mL Eppendorf tube and 1.2 mL of 5 % (v/v) trichloro acetic acid solution was then added. After being vortexed, the mixture was allowed to stand for at least 1 h at room temperature and was then centrifuged at 15,000 rpm, at 4 °C for 15 min. The supernatant was filtered through a 0.45 μm pore size hydrophilic polyvinylidene difluoride (PVDF) syringe filter (Ø 13 mm, Cat. no. 6779-1304, Whatmann Int. Ltd.) into an HPLC vial. HPLC analysis of the free amino acids was conducted according to the "Rapid, accurate, sensitive and reproducible HPLC analysis of amino acids analysis" (<http://www.chem.agilent.com/Library/chromatograms/59801193.pdf>).

In brief, 20 different free amino acids were identified using an Agilent Technologies 1200 series HPLC system equipped with Zorbax Eclipse AAA analytical columns (150 × 4.6 mm i.d., particle size 5 μm) and Zorbax Eclipse AAA guard column (12.5 × 4.6 mm i.d., 5 μm, 4-pack). The HPLC conditions were set at 338 nm wavelength, 40 °C oven temperature and 2.0 mL/min flow rate. The mobile phase consisted of 40 mM NaH<sub>2</sub>PO<sub>4</sub> (pH 7.8, solvent A) and MeCN:MeOH:H<sub>2</sub>O (45:45:10, v/v/v) (solvent B). The gradient programs were as follows: a linear step from 0 to 57 % of solvent B from 1.9 to 21.1 min, from 57 to 100 % of solvent B from 21.1 to 21.6 min, isocratic conditions with 100 % solvent B from 21.6 to 25.0 min, followed by a rapid drop to 0 % solvent B at 25.1 min and then isocratic conditions with 0 % solvent B to 30.0 min (total 30 min). A solution (50 pmol/μL [0.05 mM]) of the 20 amino acids was prepared as the standard. The quantification of free amino acids was based on the HPLC peak areas and calculated as equivalents of the standard compounds and all amounts were expressed as milligram per 100 g fresh weight. All samples were run in triplicate.

## RESULTS AND DISCUSSION

A total of 20 amino acids were compared to investigate the presence and amount of the amino acids from the four different cultivars of *L. platyphylla* (Table-1). No histidine was detected in Cheongsim cultivar of *L. platyphylla*. Tryptophan was detected only in Seongsoo and Cheongyangjaerae cultivars. Seongsoo cultivar contained the highest total amount of amino acids (970.58 mg/100 g dry wt.) exhibiting 2.18, 1.77 and 1.762 times the total amino acid of that in Liriope Tuber No. 1, Cheongsim and Cheongyangjaerae cultivars, respectively. Seongsoo cultivar not only contained the highest total amino acid, it also contained the highest levels of 13 different amino acids, *i.e.*, aspartic acid, asparagine, serine, glutamine, glycine, threonine, arginine, γ-aminobutyric acid, methionine, tryptophan, phenylalanine, leucine and lysine. A marked variation was observed in the content of glutamine in the cultivars. Among the cultivars, Seongsoo contained 2.84, 2.01 and 1.85 times the amount of glutamine in the Liriope Tuber No. 1, Cheongyangjaerae and Cheongsim cultivars, respectively. The variation in the levels of asparagine, threonine, arginine, γ-aminobutyric acid, leucine and lysine was considerably higher among the cultivars. The levels of asparagine, arginine, threonine, γ-aminobutyric acid, leucine and lysine in the Seongsoo cultivar were 3.04, 2.75, 1.89, 1.73, 1.74 and 1.68 times, respectively, higher than that in the Cheongsim cultivar, which had the lowest content. The cultivar Cheongyangjaerae contained the second highest total amount of amino acids and it contained the highest levels of six different amino acids, *i.e.*, histidine, alanine, tyrosine, cysteine, valine and isoleucine. The levels of tyrosine, alanine, valine, isoleucine, cysteine and histidine in the Cheongyangjaerae cultivar were 1.79, 1.72, 1.67, 1.61, 1.36 and 1.28, respectively, times higher than that of the Cheongsim cultivar. Although the cultivar Cheongsim contained the lowest total amount of amino acids and the levels of the various amino acid were the lowest for most of the amino acids, the tubers of this cultivar contained the highest level of glutamate, which was 1.49, 1.33 and 1.29 times, respectively, higher than that in the Cheongsim, Cheongyangjaerae and Seongsoo cultivars.

Variation in amino acids among cultivars was reported by Kim *et al.* [21]. They found that among all the amino acids isolated from *Momordica charantia*, arginine was present in remarkably high quantities, whereas cysteine and methionine were present at the lowest concentrations. Variation in amino acids content has also been observed in the different organs of *Scutellaria baicalensis* [22], in different species of aloe [23] and in different organs of green and red mustard [24]. Similar results regarding variation in the amino acid content in different cultivars of *L. platyphylla* were observed in this study. Li *et al.* [25] reported that amino acid contents, as well as γ-amino butyric acid content, varied according to the cultivars, which is supported by the findings of the current study.

## Conclusion

The levels of amino acids in the different cultivars of *L. platyphylla* varied greatly. The cultivar Seongsoo contained the highest total amount of amino acids and contained the highest levels of 13 different amino acids. This study demon-

TABLE-1  
AMINO ACID CONTENT IN DIFFERENT CULTIVARS OF *Liriope platyphylla*

Free amino acids	Cultivars (amino acid: mg/100 g dry wt.)			
	Seongsoo	Cheongyangjaerae	Cheongsim	Liriope Tuber No. 1
Aspartate	11.51 ± 0.30	10.47 ± 0.25	8.52 ± 0.26	9.64 ± 0.10
Glutamate	7.35 ± 0.45	7.13 ± 0.23	6.33 ± 0.26	9.47 ± 0.16
Asparagine	84.56 ± 0.37	30.10 ± 0.63	27.80 ± 0.55	30.89 ± 0.38
Serine	69.87 ± 0.60	57.39 ± 1.42	49.81 ± 0.33	50.44 ± 0.71
Glutamine	630.56 ± 3.90	313.42 ± 2.92	340.43 ± 3.83	221.19 ± 6.01
Histidine	2.70 ± 2.34	3.35 ± 0.14	Not detected	2.61 ± 0.03
Glycine	8.74 ± 0.65	8.48 ± 0.21	5.74 ± 0.04	7.09 ± 0.07
Threonine	6.56 ± 0.85	5.89 ± 0.21	3.47 ± 0.07	4.27 ± 0.07
Arginine	61.56 ± 1.05	22.32 ± 0.72	48.13 ± 0.27	49.16 ± 0.57
Alanine	42.31 ± 0.41	53.10 ± 1.17	30.93 ± 0.45	31.05 ± 0.30
γ-Aminobutyric acid	18.01 ± 0.30	15.80 ± 0.58	10.39 ± 0.07	10.75 ± 0.12
Tyrosine	0.89 ± 0.03	1.02 ± 0.03	0.73 ± 0.22	0.57 ± 0.07
Cystine	5.68 ± 0.19	6.27 ± 0.20	4.62 ± 0.13	4.77 ± 0.21
Valine	5.23 ± 0.24	5.43 ± 0.23	3.25 ± 0.19	3.26 ± 0.32
Methionine	4.17 ± 0.37	3.11 ± 0.40	2.64 ± 0.28	3.21 ± 0.43
Tryptophan	0.55 ± 0.09	0.14 ± 0.24	Not detected	Not detected
Phenylalanine	2.34 ± 0.15	1.54 ± 0.27	1.80 ± 0.03	1.55 ± 0.36
Isoleucine	1.73 ± 0.12	1.77 ± 0.06	1.10 ± 0.07	1.20 ± 0.17
Leucine	2.20 ± 0.09	2.02 ± 0.07	1.26 ± 0.04	1.46 ± 0.01
Lysine	4.05 ± 0.06	2.85 ± 0.04	2.41 ± 0.01	2.69 ± 0.03
Total	970.58 ± 4.49	551.62 ± 7.49	549.34 ± 2.22	445.29 ± 8.80

trates that cultivars of *L. platyphylla* exhibit varying amount of amino acid accumulation in the tubers. Cultivar-specific amino acid profiles suggest that *L. platyphylla* offers potential as a source of amino acids, particularly the tuber of the Cheongsim cultivar, which contains a large amount of glutamate.

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