Original Research Article

Growth and Ingredient Contents of Platycodon grandiflorum Roots under Sensor-based Soil Moisture Contents of Farmland Conditions

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Abstract - Growth characters and ingredient contents of two-year-old bellflower (*Platycodon grandiflorum*) roots were investigated under both control and soil moisture treatment condition using soil moisture control system including soil sensing and automatic water supply chain in this study. Root diameter, fine root number, root length, fresh weight and dry weight of the plant were significantly influenced by the automatic water treatment, 20%, 30%, 40% and 50%, respectively. Ingredient contents of the two-year-old roots in bellflower plants were detected in the 20% and 50% of controlled soil moisture content. Contents of amino acids were decreased by the soil moisture treatment, meanwhile, contents of minerals were not showed significant decrease except for phosphorus content. Showing no difference in proline and tyrosine, fourteen of the amino acid contents were gradually decreased by the increased soil moisture contents, with significant decrease in serine, glycine, alanine, leucine, lysine and histidine at 20% treatment.

Key words - Amino acid, Automatic irrigation, Bellflower, Mineral nutrients, Platycodon grandiflorum, Soil moisture

Introduction

It is well known that light, air temperature, air, soil texture and moisture are major limiting factors in crop production (Henderson et al., 2018; Jeong et al., 2020; Lee et al., 2010b, 2000). Soil moisture act a limiting factor in plant growth and development, and more than 50% content of soil induce severe reduction in crop productivity on upland fields (Henderson et al., 2018; Khalid, 2006; Lee et al., 2008; Lee et al., 1999; Osakabe et al., 2014). Moreover, water treatment influence not only growth but also chemical composition of plants (Khalid, 2006).

Soil moisture sensor-based systems are promising, however, there is not enough information on crop type, or field condition

(Kang et al., 2020; 2021). Recently development of sensing

technology and data analysis technique leads to automation of agronomy labors, and data based agriculture is enriched through cloud technology (Henderson et al., 2018; Kim et al., 2021). So far, limited automatic irrigation system that controlled and warned about under- and overwatering message is partly used in open field agriculture (Kang et al., 2020; Henderson et al., 2018).

As a well-known medicine food homology species, Platycodon grandiflorum (PG) has various pharmacological effects and health benefits, and is widely distributed in China, Korea, Japan and eastern Siberia (Huang et al., 2021; Lee et al., 2020). PG contains proteins, lipids, sugars, ash, iron, saponin, inulin and phytosterin. It contains multiple essential nutrients, including proteins, starches, vitamins, minerals and amino acids, and has been widely consumed as a dietary supplement for its various beneficial effects. Moreover, it has various pharmacological actions, such as expectorant, antitussive, antibacterial, hypotensive and hypoglycemic effects (Choi and Lee, 2018; Lee et al., 2010a; 2010b; Zhang et al., 2022). As an important

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traditional herbal medicine, its root is used to treat several diseases including, hyperlipidemia, hypertension, and diabetes (Lee *et al.*, 2010a, 2010b; Zhang *et al.*, 2022). The therapeutic effects of PG will be exerted in more than 2 years cultivated plants, and thus over two-year-old PG roots were used as traditional herbal medicine to relieve cough, excessive phlegm, sore throat, tonsillitis, chest congestion and other pulmonary or respiratory ailments.

The multiple chemical compositions and pharmacological activities of PG have been broadly investigated in the past decades (Huang *et al.*, 2021; Lee *et al.*, 2020; Wang *et al.*, 2017; Yan *et al.*, 2013; Zhang *et al.*, 2022). In recent decades, the biological activities of PG, including its anti-tumor, hepatoprotective, immunoregulatory and anti-oxidant effects, have resulted in the compositions of saponins, flavonoids, anthocyanins, phenolics, and polysaccharides, among other compounds from the plant (Huang *et al.*, 2021; Lee *et al.*, 2020; Zhang *et al.*, 2022).

The data for the growth characters and components of *P. grandiflorum* roots by the soil moisture conditions were investigated in this study using an automatic irrigation system that has constructed by our study (Lee *et al.*, 2021).

Materials and Methods

Crop cultivation and soil conditions of experimental field

Seedlings of one-year-old bellflowers (*Platycodon grandiflorum*) with 5 to 6 leaves were purchased from bellflower plantation (Bellflower Farm, South Korea) and used for cultivation. The one-year-old bellflower seedlings were transplanted to farmland fields which is controlled by the soil moisture control system (AcroTNS Com., South Korea) on May 26th, 2021 (Lee *et al.*, 2021). The soil components used in this experiment were shown by Lee *et al.* (2021). The farmland was mulched with black vinyl film with 100 cm in width. All agricultural practices, other than the experimental treatments, were done according to the recommendation of the RDA, Korea (Rural Development Administration, 2012).

Soil moisture sensing was executed by the sensors (WT1000B and WT1000A, MiraeSensor Com., South Korea) on the surface of soil with 6 cm to 11.5 cm in depth (Lee $\it et al.$, 2021). Moreover, EC (Electric conductivity) and soil temperature were also

checked. Soil moisture was controlled by 20%, 30%, 40% and 50%, respectively, according to manual of AcroTNS Company (AcroTNS Com., South Korea). Soil moisture was monitored and recorded using moisture sensor plugged into data loggers at 2-min intervals over the entire growth cycle (Lee *et al.*, 2021). Sensors were inserted into the substrate from the top, with sensor prongs close to the center of the ridge (Henderson *et al.*, 2018; Lee *et al.*, 2021).

Measurement of growth and development characteristics of root

After full maturity, all plants of each plot harvested and measured for determining below ground growth characteristics, such as root length, root diameter, fresh weight and dry weight (Kwon *et al.*, 2019; Rural Development Administration, 2012).

Constituent of major and minor mineral nutrients, inorganic substance

Reagents: HNO₃ and H_2O_2 for CRM hydrolysis were used EP-S (electronic grade, DongWooFineChem Company, Korea). Ca and Na standards (1,000 mg/L) were purchased from Accu-Standard company (U.S.A). Distilled water was deionized by equipment (MILLIPORE, Milli-Q, USA) and used for analysis. All other chemicals were used as analytical reagent grade. Equipment for microwave digestion were Multiwave PRO (Anton Paar, USA). ICP-AES Avio 500 (Perkin Elmer, USA) was used for measurement of Ca and Na.

Analysis of major and minor mineral nutrients

One mg of samples were weighing and set on Tefron beaker for disassemble by microwave digestion. And then added 8 ml HNO₃ and 2 ml H₂O₂. Samples were hydrolysed and 50 g of sample solution was mixed. The sample solution was used for analysis. Minerals were analyzed by ICP-AES (L-8900, Hitachi High Tech, Tokyo, Japan). The conditions for analyses of minerals by ICP-AES (L-8900, Hitachi High Tech, Tokyo, Japan) were shown in Table 1.

Analysis of constituent amino acid

Constituent amino acids in *Platycodon grandifflorum* were analyzed by acid hydrolysis method. The sample, 0.2 g to 0.5 g, was thoroughly mixed with 3 ml of 6 N-HCl in a test tube and

tightly sealed. The mixture was subjected to acid hydrolysis in a heating block (Thermo-Fisher Scientific Co., Rockford, IL, USA) at 105°C for 22 hr. The hydrolyzed sample, 10 ul, and sodium dilution buffer (pH 2.2), 990 µl, were mixed and subsequently filtered through a 0.2 µm PTFE membrane filter. An amino acid analyzer (L-8900, Hitachi High Tech, Tokyo, Japan) was used to determine the profiles of constituent amino acids. The conditions for analyses of amino acids by ICP-AES (L-8900, Hitachi High Tech, Tokyo, Japan) were shown in Table 2.

Statistical analysis

All of the experiments were executed with three (ingredient content) to five (plant-growth factors) replications. Using the SAS program (SAS, 9.2, Institute Inc, USA), statistical analysis was conducted by Duncan's multiple range test (DMRT, p=0.05). Frequency and percentage were used to analysis the qualitative characters, whereas mean and standard deviation were used for quantitative data analysis (Lee *et al.*, 2021; 2022).

Table 1. Conditions for analyses of minerals by ICP-AES

Instrument	ICP-AES, Avio 500, Perkein Elmer
Power (W)	1500
Use Gas	Argon gas
Pump Speed (rpm)	2.50
Nebulizer Flow (L/mir	0.7
Wavelength Na	589.592
(nm) Ca	317.933

Results and Discussion

Condition of soil on cultivation field

Monitoring for the soil moisture content, EC and soil temperature were executed on the base of automatic soil moisture content control system under farmland conditions and the trend of environmental data were reported by Lee *et al.* (2021, 2022). In addition, the trends of environmental data on farmland were checked after cultivation of plants in the field and shown in Table 3.

On the base of automatic soil moisture control system, the soil moisture was controlled from 20%, 30%, 40% and 50%, respectively (Lee et al., 2021), and the moisture contents were measured from May to December, 2021 (Lee et al., 2022). The soil moisture content was controlled by the system showing over the controlled-soil moisture contents except 40% treatment. The 40% treatment showed 30% in the moisture which is lower than that of the system controlled. Lee et al. (2021) assumed that the reason showing the higher soil content might be resulted from differences by rainfall, drainage and soil condition such as, uniformity of soil textures. And the lower soil content might be resulted from operating of automatic irrigation system or insertion position of sensor on the surface of farmland fields. Therefore, the position of sensors at the ridges is very important when moisture content is monitored on open farmland condition (Henderson et al., 2018; Kang et al., 2020; 2021).

As shown Fig. 1, additional data for the soil moisture content, EC and soil temperature were measured for 1 month,

Table 2. Conditions for analyses of amino acids by ICP-AES

Standard	An undiluted soln.: Amino acid Standard soln. (Sigma AAS18-10ML) Standard soln.: Diluted soln. of amino acid standard with 10, 25, 50, 100, 250 times, respectively.
Column	Ion exchange column Packed with Hitachicustom ion exchange resin (4.6 mm \times 60 mm)
Detection	Visible Detector Wavelength: 570 nm, 440 nm (for Proline)
Column Flow	0.999 mL/min
Temp. program	Column oven: 20 to 85°C (increase 1°C/step) Reaction unit: 50 to 140°C (increase 1°C/step)
Mobile phase	 Buffer Solution: pH-1, pH-2, pH-3, pH-4, pH-RG (Mitsubishi Chemical Corporation) Ninhydrin solution, Buffer (Wako, Ninhydrin Coloring Solution Kit for HITACHI)

Table 3. Characteristics of soil components of the cultivation field after the plant culture at Sunchon National University (SCNU), South Korea

Item Site	pH (1:5)	Organic Matter (g/kg)	P (mg/kg)	K (cmol+/kg)	Ca (cmol+/kg)	Mg (cmol+/kg)	Electric Conductivity (dS/m)
A ^z	5.7	28	513	1.07	6.0	2.1	0.7
\mathbf{B}^{y}	5.9	19	351	0.91	6.7	2.7	0.8
$\mathbf{C}^{\mathbf{x}}$	5.9	14	242	0.85	7.6	2.6	0.4
Mean	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Optimal Range	6.0~7.0	20~30	300~550	0.5~0.8	5.0~6.0	1.5~2.0	< 2

^zA, ^yB and ^xC: Three different soil sampling sites of the upland fields.

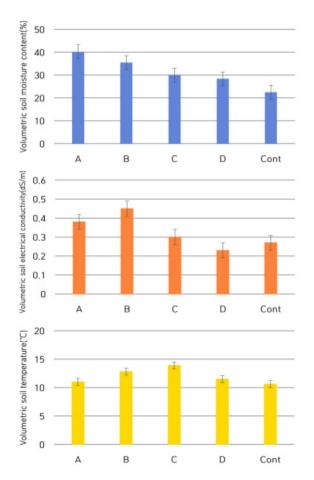


Fig. 1. A month (March, 2022) soil moisture (upper), EC (middle) and soil temperature (bottom) at the field condition. *Upper: Soil moisture contents were set as A: 50%, B: 40%, C: 30%, D: 20% by the soil moisture control system with soil sensing and automatic water supply chain, and control was shown at 22.5% on the field condition, respectively.

March, 2022. As a result of monitoring of the soil moisture by using automatic irrigation control system, even though the

sensing data were not matched directly, the system was worked properly (Lee *et al.*, 2021; 2022). Thus, we sampled roots for growth and ingredient contents analyses on 20% and 50% moisture content, respectively.

Root growth by soil moisture content

Growth of two-year-old roots by soil moisture contents in bellflower plants were shown in Table 4 and Fig. 2. Root diameter, root number and root length of the plants were significantly influenced by water treatment with 20%, 30%, 40% and 50%, respectively. Moreover, fresh weight and dry weight of the plant roots were significantly influenced by water treatment, 20%, 30%, 40% and 50%, respectively. In detail, main root and fine root diameter, main root and root number have a tendency that showing more increase from 20% to 40% of the water contents compared with control, and those were decreased in 50% of the water content. Especially, fine root numbers were decreased in 50% treatment (Fig. 2). Also, fresh weight, dry weight, shoot fresh weight and shoot dry weight have showing increasing by the water content from 20% to 40% compared with control, and these were decreased in 50% of the water content.

In general, approximately 50% of soil moisture is a maximum content for normal plant growth and development (Henderson *et al.*, 2018; Khalid, 2006; Lee *et al.*, 2008; Lee *et al.*, 1999; Osakabe *et al.*, 2014). However, no significant decrease in all growth factors in this study. This may be resulted from mismatching between sensor controlled data and the field data (Lee *et al.*, 2021). The result means higher soil moisture act on repressor plant growth, and it is consistent with other crop plants (Kang *et al.*, 2021; Khalid,

Table 4. Characteristics of growth of roots of bellflower (Platycodon grandiflorum) by soil moisture treatment with cultivation of two-year-old tuberous roots

Treatment	CONT	20%	30%	40%	50%
Main root diameter (mm)	23.0±2.65b	30.0±8.66ab	30.7±6.43ab	42.3±6.43a	24.7±6.43b
Fine root diameter (mm)	14.7±5.03b	22.3±4.51a	21.3±3.51a	16.7±3.06ab	16.0±4.00ab
Main root length (cm)	19.2±2.84b	25.5±4.27a	22.7±3.21ab	22.0±17.3ab	23.0±6.56ab
Fine root No.	2.3±1.15b	4.0±3.00a	4.0±1.00a	3.7±0.58ab	3.7±1.15ab
Fresh weight (g/root)	17.8±4.05b	35.1±12.96ab	42.0±11.5a	31.4±11.03ab	17.9±1.27b
Dry weight (g/root)	3.4±1.16b	7.2±2.43ab	8.6±3.08a	8.2±2.43ab	$4.1\pm0.47b$
Fr. weight of shoot (g/shoot)	9.8±2.98b	13.6±6.41a	11.9±1.84ab	9.7±6.19ab	7.4±1.00b
Dr. weight of shoot (g/shoot)	1.3±0.25b	2.6±0.21a	2.0±0.74ab	2.0±0.47ab	1.4±0.75b

^{*}Means within a line followed by the same letter are not significantly different base on the Duncan's Multiple Range Test (DMRT, p < 0.05).



Fig. 2. Growth profiles of two-year-old bellflowers (*Platycodon grandiflorum*) by soil moisture treatment at the field condition.

2006). Especially, Henderson *et al.* (2018) reported that approximately 50% of soil moisture is a maximum content for a normal plant growth and development and it leads severe reduction for crop productivity on upland fields.

Ingredient contents by soil moisture content

Ingredient contents of two-year-old roots in bellflower plants were detected in the 20% and 50% controlled soil moisture content.

Mineral nutrients

The mineral nutrients contents by soil moisture contents in two-year-old roots of bellflower plants were shown in Table 5. Phosphorus content of the root was 140.5~mg/100~g in control, and that of treatment was significantly decreased in

the soil moisture treatment with 20% and 50% showing 112.2 mg/100 g and 120.4 mg/100, respectively. However, except for phosphorus nothing shown significant decrease in mineral contents. Potassium content of the root was 475.8 mg/100 g in control, and that of treatments was decreased in 20% and 50% soil moisture showing 401.3 mg/100 g and 423.4 mg/100, respectively. Magnesium content of the roots was not showed difference by soil moisture contents compared with the control. Sodium, none-essential nutrient element, content was somewhat higher (11.3 mg/100 g) in the 50% soil moisture treatment compared with the control (9.5 mg/100 g). Contents of minor nutrients, iron and zinc, were showed few difference by the soil moisture treatment.

Chemical composition in bellflower was reported in leaves and stems of (Jeong and Shim, 2006), roots (Lee *et al.*, 2013)

Table 5. Mineral contents of roots of bellflower (*Platycodon grandiflorum*) by soil moisture treatment with cultivation of two-year-old tuberous root (mg/100 g)

			(0 2)
Treatment Inorganic component	CONT	20%	50%
Na	9.5±1.01 ^a	7.1±3.2 ^a	11.3±5.52 ^a
Ca	46.1 ± 12.96^{a}	39.3 ± 0.74^{a}	50.7 ± 10.84^a
K	475.8±25.15 ^a	401.3±41.11 ^a	423.4 ± 61.95^{a}
P	140.5±4.53 ^a	112.2±9.42 ^b	120.4 ± 11.06^{b}
Mg	34.5±4.41 ^a	35.6 ± 6.09^{a}	34.1 ± 6.85^{a}
Fe	6.1 ± 1.24^{a}	4.8 ± 1.99^{a}	6.2 ± 0.12^{a}
Zn	1.4±0.21 ^a	1.4 ± 0.36^{a}	1.4 ± 0.15^{a}

*Means within a line followed by the same letter are not significantly different base on the Duncan's Multiple Range Test (DMRT, p < 0.05).

Table 6. Amino acid contents of radix of bellflower (*Platycodon grandiflorum*) by soil moisture treatment with cultivation of two-year-old tuberous root (mg/100 g)

			(mg/10
Amino acids	Treatment CONT*	20%	50%
Asp	179.4±31.75 ^{ay}	133.2±10.45 ^a	168.2±28.6 ^a
Thr ^z	97.8±15.81 ^a	68.0 ± 7.78^a	68.0 ± 7.78^{a}
Ser	95.1±15.59 ^a	69.9 ± 69.87^{b}	82.9±6.53ab
Glu	1301.5±495.72 ^a	780.6±206.99 ^a	1139.8 ± 41.58^{b}
Pro	17.1 ± 7.26^{a}	21.3±5.72 ^a	16.0±4.44°
Gly	64.3±3.03 ^a	51.4±1.29 ^b	59.8±5.03 ^a
Ala	81.5±5.98 ^a	59.4±7.37 ^b	74.6±5.66 ^a
Val ^z	86.9 ± 12.12^{a}	66.5 ± 6.07^{a}	80.1 ± 18.45^{a}
Met ^z	20.7 ± 11.06^{a}	11.2±3.50 ^a	13.4±2.85 ^a
Ile ^z	60.8 ± 6.22^{a}	46.6±5.39 ^a	57.1±12.63 ^a
Leu ^z	98.7 ± 9.06^{a}	80.0 ± 3.84^{b}	94.8 ± 12.00^{ab}
Tyr	52.2±5.75 ^a	53.3±9.53 ^a	52.3±11.67 ^a
Phe ^z	72.0 ± 4.75^{a}	63.5±9.64 ^a	70.8 ± 7.66^{a}
Lys ^z	103.6±5.50 ^a	81.2±1.73 ^b	96.3±9.77 ^a
His ^z	57.5±7.55 ^a	43.2±2.89 ^b	49.9 ± 7.72^{ab}
Arg	1025.7±141.58 ^a	910.8±96.85 ^a	886.6±267.89a
Total A.A	3414.8	2540.1	3010.6
Total E.A.A ¹⁾	598	460.2	530.4

^zEssential amino acids; Thr+Val+Met+Ile+Leu+Phe+His+Lys.

and raw ginseng (Kim *et al.*, 2002). The composition was different by genotype (Yan *et al.*, 2013), and cultivation year and soil texture (Lee *et al.*, 2000) of PG, and plant organs and cultivation year of roots in *Panax ginseng* C,A. Meyer (Park

et al., 2012). In addition, it was reported main components contents were different according to grow period in *Codonopsis lanceolata* (Im *et al.*, 2021). In this study, some of minerals were different by soil moisture contents.

^yMeans within a line followed by the same letter are not significantly different base on the Duncan's Multiple Range Test (DMRT, p < 0.05).

Amino acid

Amino acid contents of two-year-old roots of bellflower plants were shown in Table 6. Showing no difference in proline and tyrosine, all of the amino acid contents were gradually decreased by increased soil moisture contents, with significant decrease in serine, glycine, alanine, leucine, lysine and histidine at 20% treatment.

Total amino acid contents of the roots were 3,414.8 mg/100 g in control, and the contents of treatment were decreased in the soil moisture with 20% and 50% showing 2,540.1 mg/100 g and 3,010.6 mg/100 g, respectively. Essential amino acid contents of the root were significantly decreased by the treatment of soil moisture showing 598.0 mg/100 g, 460.2 mg/100g and 530.4 mg /100 g in the control, 20% and 50% treatment, respectively. In addition, the content of amino acid was the highest in glutamic acid, followed by arginine, and the proline was the lowest at control. And the content of amino acid was the highest in arginine, followed by glutamic acid, and the methionine was the lowest at 20% and 50% treatments.

Under water stress condition in paddy field, the main constituents of essential oil, proline and total carbohydrate content increased, and N, P, K, and protein decreased in ginseng (Khalid, 2006). As a heat shock, roasting induced diverse patterns of amino acid profiles in PG roots (Lee *et al.*, 2020). In this study, water content in soil affected to amino acid patterns, also. Jeong and Shim (2006) reported difference of amino acid contents between leaf and stem in ginseng showing glutamic acid, followed by arginine in leaf, lysine, followed by glutamic acid in stem parts of genseng plant. Moreover, their data shown that the amino acid contents were different from cultivation year, and the contents were high in glutamic acid, followed by arginine in 3-year cultivated roots, and in arginine, followed by alanine in 24-year cultivated roots.

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Conflicts of Interest

The authors declare that they have no conflict of interest.

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