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EFFECTS OF DROUGHT STRESS ON THE GROWTH, PHYSIOLOGY AND SECONDARY METABOLITE PRODUCTION IN *PINELLIA TERNATA* THUNB.

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Abstract

Pinellia ternata grown under four water regimes in a greenhouse. The morphology traits and photosynthetic pigment content were highest in *P. ternata* plants grown under the well-watered treatment. The antioxidant activities and malondialdehyde content were significantly greater in *P. ternata* under drought condition. The soluble protein content of *P. ternata* was significantly higher under moderate drought stress, but the soluble sugar content of *P. ternata* was significantly lower under severe and moderate drought stress. Additionally, the total alkaloid content of *P. ternata* showed the highest increase under severe drought stress, but the guanosine and succinic acid contents of *P. ternata* were significantly lower under drought stress. The maximum yield of total alkaloid, guanosine and succinic acid of *P. ternata* were obtained under the well-watered regime. The results suggest that a well-watered condition can contribute to the yield of tuber and secondary metabolites production in *P. ternata*.

Key words: *Pinellia ternata*, Growth, Physiological, Secondary metabolites, Drought.

Introduction

Drought stress is a crucial environmental factor that can influence plant growth, resulting in changes in its physiological and biochemical properties and secondary metabolite accumulation (Zhu *et al*., 2009; Chen *et al*., 2016). Under drought stress, plants experience changes in stomatal responses, osmotic adjustment and antioxidative defenses in order to alleviate damage caused by the drought stress. However, a long period of drought stress could cause stomatal closure, leaf size reduction, plant growth suppression, or even death (Dias *et al*., 2007; Christina & Gisela, 2013).

Previous studies indicate that drought stress is one type of oxidative stress that at the cellular level enhances the generation of reactive oxygen species (ROS), such as superoxide radicals (O_2^-), hydrogen peroxide (H_2O_2) and hydroxyl radicals (·OH) (Reddy *et al*., 2004; Zhu *et al*., 2009). Plants have evolved antioxidative defense systems that involve enzymatic antioxidants and non-enzymatic antioxidants to prevent ROS damage (Zhu *et al*., 2009; Li *et al*., 2011). Moreover, another mechanism to protect plants under drought stress is by promoting the accumulation of soluble substances, for instance soluble protein and sugars, and proline through osmotic adjustment (Xu *et al*., 2015).

Several studies indicated that plants under drought stress generated a greater concentration of secondary metabolites than those planted under well-watered conditions (Chen *et al*., 2011; Selmar & Kleinwachter, 2013; Alinian *et al*., 2016). In some medicinal plants, drought stress enhanced secondary metabolite production, namely the saikosaponin content in the roots of *Bupleurum chinense* (Zhu *et al*., 2009), phenolic compounds in the roots of *Salvia miltiorrhiza* Bunge (Liu *et al*., 2011) and total phenolics, flavonoids and anthocyanin contents in the leaves of *Labisia pumila* Benth (Jaafar *et al*., 2012). However, drought stress reduced bioactive compound (e.g., phenolic compounds) concentrations in tea leaves (Cheruiyot *et al*., 2007), rosmarinic acid in the roots of *S. miltiorrhiza* (Liu *et al*., 2011), and phenolic acid and organic acid contents in the roots of *Rehmannia glutinosa* (Chung *et al*., 2006).

Pinellia ternata (Thunb.) Breit is an important species of the genus *Pinellia* belongs to the Araceae, which are endemic to East Asia (Flora of China 1977). The dried tuber of *P. ternata* - "Pinelliae Rhizoma" or "Banxia" in Chinese - is commonly used by Chinese populations in the treatment of cough, infection, congestion with phlegm, vomiting and termination of early pregnancy (Chen *et al*., 2003; Kim *et al*., 2006). Furthermore, *P. ternata* demonstrated the following medicinal properties: expectorant, antitussive, antitumor, antiemetic, antibacterial, antioxidant, sedative-hypnotic and antiinflammatory (Wang *et al*., 2008).

In recent years, the demand for *P. ternata* has increased steadily in the market. It is estimated that China consumes 5-6 million kilograms of *P. ternata* each year (Zhang *et al*., 2010). The wild population of *P. ternata* in China is sufficient to meet only one third of this growing consumption, which is why large scale plantation cultivation of *P. ternata* has been proposed for southwest China since the 1970s (Guo *et al*., 1993). However,

during the cultivation process, farmers always have problems controlling the water supply, especially in the Karst Mountain area of Guizhou Province (Guo *et al*., 1993). Recently, some reports have focused on setting quality control standards for Pinelliae Rhizoma in terms of total alkaloid, guanosine and particularly succinic acid (Chinese Pharmacopoeia 2015). However, there is little information about the effect of drought stress on the growth, physiological and secondary metabolite in *P. ternata*. The objective of this study was to investigate the influence of drought stress on growth, chlorophyll content, antioxidant defenses, and secondary metabolites content and yields in *P. ternata*. These results may help inform the control of irrigation of *P. ternata* to improve its yield and medicinal quality.

Materials and Methods

Plant materials and growth conditions: The experiment was carried out under natural conditions at the Institute of Morden Chinese Medical Materials of the Guizhou Academy of Agricultural Sciences, Guiyang, PR. China. *Pinellia ternata* tubers were collected from the identical species group in Weining County, Guizhou Province, and identified by Prof. Qiaosheng Guo of Nanjing Agricultural University. Good tubers of a similar size and weight were selected and cultivated into plastic pots containing a nutrient-rich soil. Every tuber was maintained in the soil below 3 cm and 7 tubers planted in each pot.

Watering treatment: The experiment start from 28th February 2013, all of the pots were irrigated to 33.79% (The field capacity (FC) of the soil). The pots were then assigned to four groups under the following conditions: (1) well-watered (75% FC; Control); (2) slight drought (60% FC; LD); (3) moderate drought (45% FC; MD); and (4) severe drought (30% FC; SD). The levels of soil moisture were maintained by manual irrigation with distilled water in which plants were watered every day from 17:00 - 18:00 h; the FC for the different treatments was verified by soil weighing. A completely randomized experimental design was applied to 9 repeats (pots) per treatment. Plants were harvested to measure plant growth parameters, malondialdehyde content, antioxidative enzyme activities and chlorophyll content on the 25th day after the initiation of drought treatment. The plant biomass parameters (tuber fresh and dry weights), soluble protein and sugar contents, and bioactive component content (total alkaloids, guanosine and succinic acid) were determined on the 35th day after the initiation of drought treatment.

Plant growth parameters: The plant height was measured by a ruler, and the leaf size was measured with a portable area meter (LI-COR, Lincoln, NE, USA). After recording the tuber fresh weight, the remaining tubers were dried at 70°C until a constant mass was reached, and they were then measured using an electronic balance (Sartorius Bp221S, Germany). Finally, collected and counted all of the tubers.

The propagation index was determined using the following equation:

$$\text{Propagation index} = \frac{\text{Tubers number after harvest}}{\text{Planting tuber number}}$$

Chlorophyll content: The chlorophyll content in *P. ternata* leaves was determined as previously described method (Jin *et al*., 2015).

Antioxidant enzyme activities and malondialdehyde content: Peroxidase (POD) activity was measured using the method described by Shannon *et al*., (1996) with some modification. Catalase (CAT) activity was measured using the method by Pukacka & Ratajczak (2005). The determination of malondialdehyde (MDA) content was based on the method of Stewart & Bewley (1980).

Soluble protein and soluble sugar contents: Soluble protein and sugar content were measured by the previously described method (Xue *et al*., 2015).

Total alkaloid, succinic acid and guanosine contents: Total alkaloid content was measured by acid dye colorimetry method (Önal *et al*., 2005). Succinic acid content was measured by the potentiometric titration method (Jie *et al*., 2015). Guanosine content was determined by RP-HPLC (Yu *et al*., 2006).

Total alkaloid, succinic acid and guanosine yields: To calculate the total yield from the dry weight of the tuber mass multiplied by the total alkaloid, succinic acid and guanosine concentrations divided by the dry weight per plant tuber.

Statistical analyses: Data were subjected to an analysis of variance, correlation and Duncan's multiple range tests ($p<0.05$) using SPSS 17.0 software (SPSS, Chicago, IL, USA). Data were also expressed as the means ± SD ($n \geq 3$).

Results

Plant growth, morphology and biomass: Drought stress had a strong effect on the morphology of *P. ternata* plants (Table 1). Compared to the control group, the fresh tuber weights of *P. ternata* plants under the SD and MD treatments were reduced by 24.53% and 21.38%, respectively, and the dry tuber weights of *P. ternata* plants under the SD and MD treatments were declined by 24.42% and 13.95%, respectively. Compared to the control group, the plant heights of plants under the SD and MD treatments were reduced by 32.28% and 29.43%, respectively, and the tuber numbers of plants under the SD and MD treatments were declined by 34% and 50%, respectively. Compared to the control group, the propagation indexes of plants under the SD and MD treatments were decreased by 33.57% and 50.35%, respectively. However, there were no significant differences in these five growth parameters (except for leaf area) between the LD and Control group. The leaf areas of plants under the SD, MD and LD treatments were declined by 31.90%, 25.97% and 7.91%, respectively, compared to the control group.

Table 1. Effects of drought stress on different growth parameters of *P. ternata* plants.

Water treatment	Tuber fresh weight (g)	Tuber dry weight (g)	Plant height (cm)	Leaf area (cm^2)	Tuber number	Propagation index
Control	3.18 ± 0.04 a	0.86 ± 0.01 a	20.35 ± 0.42 a	23.64 ±1.89 a	10.0 ± 0.04 a	1.43 ± 0.05 a
LD	3.20 ± 0.01 a	0.86 ± 0.02 a	18.17 ± 0.04 a	21.77 ±1.41 b	10.1 ± 0.03 a	1.45 ± 0.23 a
MD	2.50 ± 0.01 b	0.74 ± 0.02 b	14.36 ± 0.47 b	17.50 ±1.88 b	5.0 ± 0.03 d	0.71 ± 0.02 d
SD	2.40 ± 0.10 b	0.65 ± 0.04 c	13.78 ± 0.29 b	16.10 ±2.06 b	6.6 ± 0.01 c	0.95 ± 0.05 b

Each value is presented as the mean ± SD (n=3). Different letters in columns indicate statistically significant differences (p<0.05)

Chlorophyll content: The total chlorophyll content in *P. ternata* leaves showed no remarkable difference between the well water and LD treatments, whereas Chl content, Chl b content, and total Chl content significantly decreased in *P. ternata* leaves in the SD and MD treatments compared with the well water treatment. Under the SD treatment, the Chl a, Chl b and total Chl contents were declined by 22.11%, 22.64% and 22.27% compared with the well water treatment, respectively. Similar results were observed in the MD treatment where the Chl a, Chl b and total Chl contents were declined by 18.52%, 21.58% and 19.47% compared with the well water treatment, respectively. Moreover, there were no significant differences in the ratio of Chl a/b content among the four treatments with the highest value observed for the MD treatment (Fig. 1).

Antioxidant enzyme activity and malondialdehyde content: We observed significant differences in antioxidant enzymes (POD and CAT) activities and in MDA content among the four treatments (Fig. 2). The plants in the SD treatment revealed the highest POD and CAT activities and MDA content, whereas the lowest values for these variables were observed for the Control treatment. Meanwhile, there were no remarkable differences in these variables between the SD and MD treatments, respectively. Compared with the well water treatment, the activities of POD and CAT and the MDA content were increased in the LD treatment; however, there were no significant differences in these variables between the treatments.

Soluble protein and sugar contents: The soluble protein content in *P. ternata* tubers in the MD treatment differed significantly from that in the Control treatment (Fig. 3). The soluble protein content was lower in the SD and LD treatments, showing a decrease of 5.54% and 17.32%, respectively, compared with the well water treatment; differences among the other three treatments were not significant. The Control treatment showed a substantial increase in soluble sugar content in the *P. ternata* tubers, while there was no significant difference in this variable between the LD and Control treatments. Moreover, the soluble sugar contents in the SD and MD treatments were significantly lower than in the Control treatment, although there was no significant difference in this variable between the two treatments (Fig. 3).

Total alkaloid, guanosine and succinic acid contents: The total alkaloid contents of *P. ternata* tubers were highest under the SD treatment and lowest under the Control treatment. When the water level decreased from 75% FC to 30% FC, a significant increase in the total alkaloid contents were observed. Reverse results were observed for the guanosine content with the highest and lowest concentrations recorded in the Control treatment and the SD treatment, respectively. When the water availability increased from 30% FC to 75% FC, a remarkable increase in guanosine content was observed. In addition, the highest and lowest succinic acid contents were found in the Control treatment and SD treatment, respectively. When the water availability increased from 30% FC to 75% FC, a significant increase in succinic acid contents were observed (Table 2).

Total alkaloid, guanosine and succinic acid yields: Total alkaloid yields of *P. ternata* tubers were highest under the LD treatment and lowest under the SD treatment. The highest and lowest guanosine yields of *P. ternata* tubers were found under the Control treatment and SD treatment, respectively. Moreover, the yields of succinic acid in the Control treatment and LD treatment were significantly higher than the yields in the other two treatments, while the difference between the LD treatment and the well water treatment was not significant (Table 3).

Correlation analysis: The *P. ternata* growth indicators (plant dry weight, plant height and leaf area) were significantly and positively correlated with the photosynthetic pigment content variables (total Chl content) respectively. However, the antioxidant enzyme activities variables (POD and CAT) were significantly and strongly negatively correlated with the photosynthetic pigment content variables. Moreover, there were significant and positive correlations between the variables of photosynthetic pigment content and soluble sugar content, but significant and negative correlations among the variables of CAT activity, soluble sugar content and MDA content. There were significant and positive correlations between the variables of photosynthetic pigment content and succinic acid content, but significant and negative correlations between the variables of antioxidant enzyme activity, specifically POD with CAT and, MDA content with succinic acid content. Furthermore, there was a significant and strongly positive correlation between dry tuber weight and succinic acid content but a significant and negative relationship between total alkaloids content and guanosine content (Table 4).

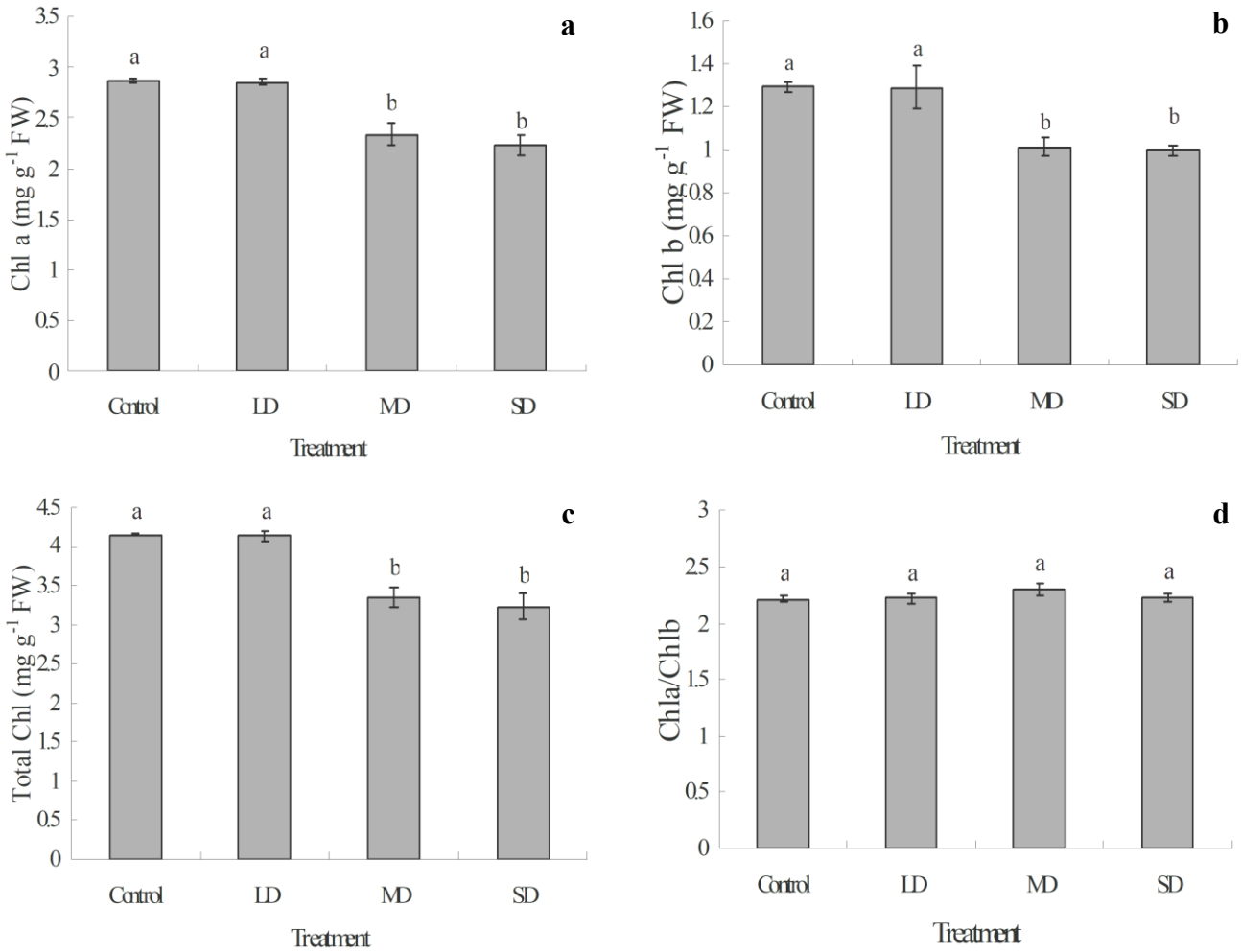

Fig. 1. Effects of drought stress on the leaf photosynthetic pigment contents (a Chl a; b Chl b; c total Chl; and d Chl a/b) of *P. ternata* on the 25th day after initiation of treatment. Values are expressed as the mean ± SD (n = 3). Bars carrying different letters indicate a significant difference at $p<0.05$ between the drought treatments.

Table 2. Effects of different drought treatments on the total alkaloid, guanosine and succinic acid contents of *P. ternata* tubers.

Water treatment	Total alkaloids (%)	Guanosine (%)	Succinic acid (%)
Control	0.0023 ± 0.0004 d	0.0233 ± 0.0031 a	0.2711 ± 0.0024 a
LD	0.0027 ± 0.0002 c	0.0203 ± 0.0019 b	0.2704 ± 0.0030 a
MD	0.0028 ± 0.0003 b	0.0177 ± 0.0022 c	0.2592 ± 0.0011 b
SD	0.0029 ± 0.0001 a	0.0170 ± 0.0017 c	0.2541 ± 0.0022 c

Values are expressed as the mean ± SD (n=3). Different letters in columns indicate statistically significant differences ($p<0.05$)

Table 3. Effects of different drought treatments on the total alkaloid, guanosine and succinic acid yields of *P. ternata* tubers.

Water treatment	Total alkaloids (mg plant^{-1})	Guanosine (mg plant^{-1})	Succinic acid (mg plant^{-1})
Control	0.020 ± 0.000 c	0.20 ± 0.00 a	2.32 ± 0.03 a
LD	0.023 ± 0.000 a	0.17 ± 0.00 b	2.32 ± 0.04 a
MD	0.021 ± 0.001 b	0.13 ± 0.00 c	1.93 ± 0.05 b
SD	0.019 ± 0.001 c	0.11 ± 0.01 d	1.65 ± 0.09 c

Values expressed as the mean ± SD (n=3). Different letters in columns indicate statistically significant differences ($p<0.05$)

Discussion

In this experiment, significant differences in the growth parameters of *P. ternata* among the four water treatments were observed. Compared with the control condition, the drought treatments (SD and MD) markedly restrained plant growth, not only in terms of plant height, leaf area, tuber number and tuber propagation index but also in terms of tuber biomass accumulation (Table 1). Similar studies have been conducted on other medical plants subjected to drought stress (Chen *et al*., 2011; Miao *et al*., 2015). For one thing, the growth of plant roots was affected by low water availability and limited nutrient absorption, which directly and negatively impacted plant productivity (Razmjoo *et al*., 2008; Zhu *et al*., 2009). On the other hand, symptoms of slow growth, such as declines in plant height and leaf area were attributed mainly to cell growth and differentiation, which was seriously inhibited with water shortage, resulting in slow development of the lateral and apical meristems (Guerfel *et al*., 2009).

Table 4. Correlation analysis of variables of growth, chlorophyll content, antioxidant enzyme activity, MDA content, soluble protein content, soluble sugar content and secondary metabolites content of *P. ternata* plants.

	Dry tuber weight	Plant height	Leaf area	Tuber number	Chl a	Chl b	Chlorophyll	POD	CAT	MDA	Soluble protein	Soluble sugar	Total alkaloids	Guanosine	Succinic acid
Dry tuber weight	1														
Plant height	0.919	1													
Leaf area	0.957*	0.994**	1												
Tuber number	0.809	0.900	0.886	1											
Chl a	0.971*	0.961*	0.978*	0.926	1										
Chl b	0.945	0.962*	0.971*	0.957*	0.996**	1									
Chlorophyll	0.963*	0.962*	0.977*	0.937	1.000**	0.998**	1								
POD	-0.968*	-0.981*	-0.993**	-0.915	-0.996**	-0.991**	-0.995**	1							
CAT	-0.951*	-0.970*	-0.979*	-0.948	-0.997**	-0.999**	-0.999**	0.995**	1						
MDA	-0.915	-0.999**	-0.992**	-0.915	-0.964*	-0.968*	-0.966*	0.982*	0.975*	1					
Soluble protein	-0.158	-0.267	-0.233	-0.657	-0.362	-0.435	-0.386	0.311	0.404	0.302	1				
Soluble sugar	0.819	0.956*	0.931	0.977*	0.927	0.953*	0.936	-0.936	-0.952*	-0.966*	-0.526	1			
Total alkaloids	-0.773	-0.922	-0.899	-0.692	-0.789	-0.782	-0.787	0.842	0.802	0.912	-0.027	-0.825	1		
Guanosine	0.874	0.988*	0.973*	0.835	0.908	0.907	0.909	-0.943	-0.920	-0.983*	-0.159	0.923	-0.971*	1	
Succinic acid	0.993**	0.954*	0.980*	0.871	0.992**	0.977*	0.988*	-0.991**	-0.981*	-0.953*	-0.247	0.883	-0.804	0.909	1

* Indicates significant difference at probability of $0.01 < p < 0.05$

** Indicates significant difference at probability of $p < 0.01$

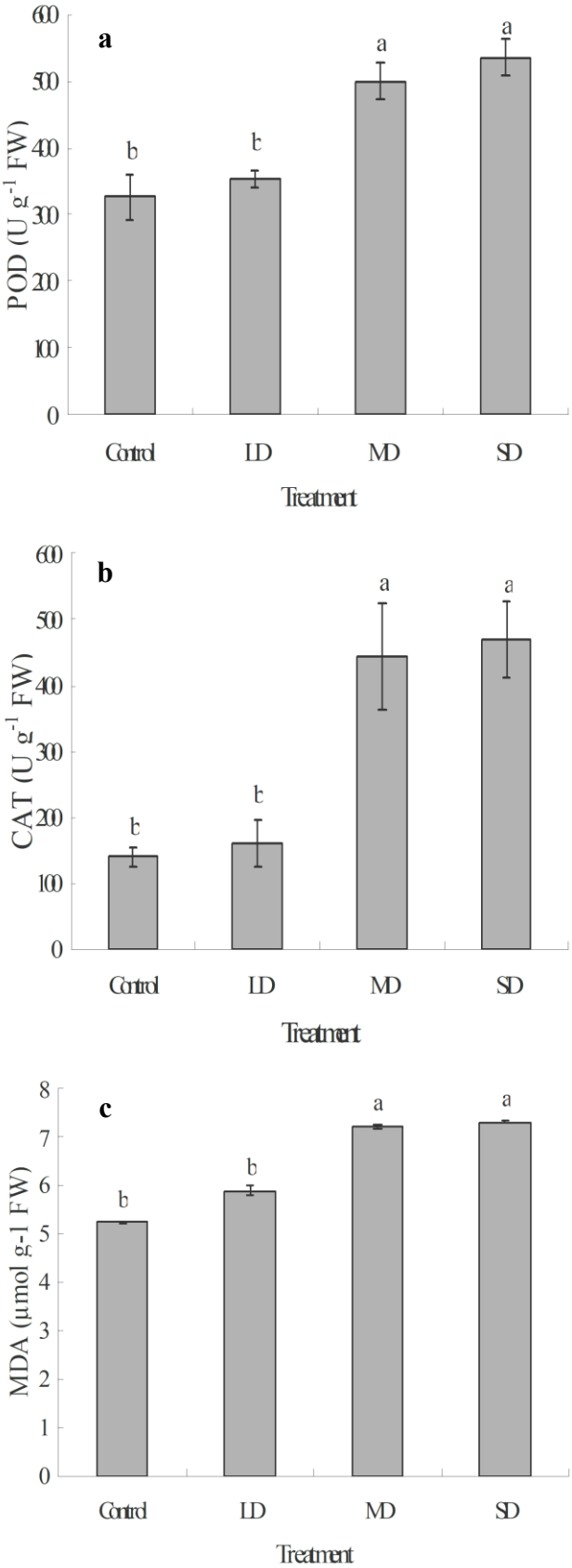

Fig. 2. Effects of drought stress on the a) peroxidase (POD) activity, b) catalase (CAT) activity and c) malondialdehyde (MDA) content in leaves of *P. ternata* after the 25th day of initiation of treatment. Bars are expressed as the mean ± SD (n = 3). Bars carrying different letters indicate a significant difference at $p<0.05$ between the drought treatments.

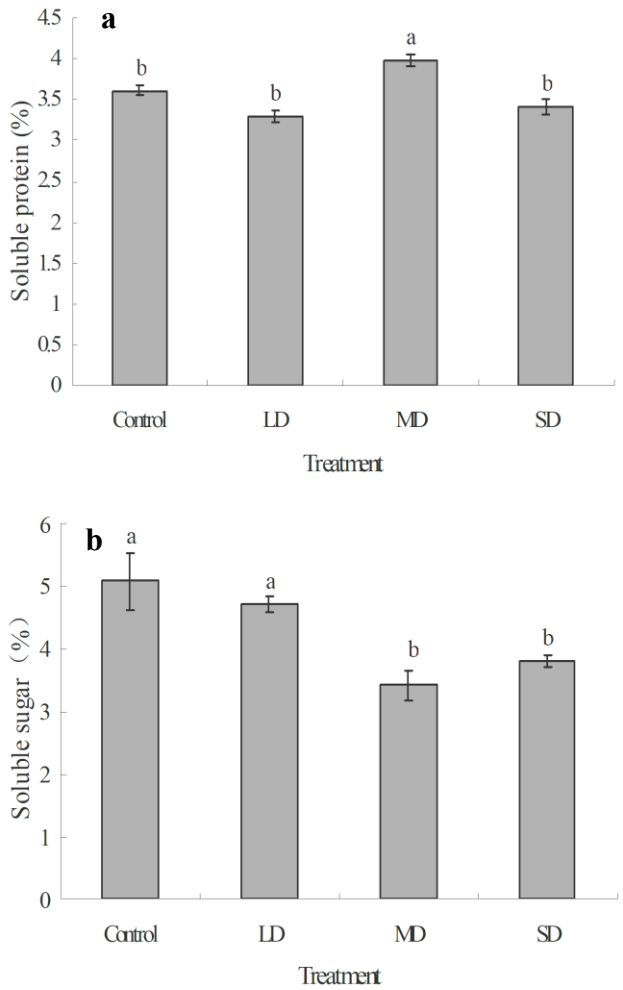

Fig. 3. Effects of drought stress on a) soluble protein content and b) soluble sugar content in the tubers of *P. ternata* on the 35th day after initiation of treatment. Bars are expressed as the mean ± SD (n = 3). Bars carrying different letters indicate a significant difference at $p<0.05$ between the drought treatments.

Our results showed that the photosynthetic pigments in *P. ternata* leaves were significantly reduced by drought stress (Fig. 1). Previous studies have demonstrated a drought stress induced significant decrease the Chl a content, Chl b content, and total Chl content, compared with the Control treatment, which could be caused by a disruption in chlorophyll biosynthesis or an accelerated degradation of chlorophyll due to drought stress (Kaewsuksaeng, 2011; Miao *et al*., 2015). Moreover, we found that the Chl a/b ratio increased slightly in the drought stress treatments (SD and MD) compared with the Control treatment. Our results suggest that a higher Chl a/b ratio in the *P. ternata* leaves indicates an adaptation to drought stress by maximizing energy efficiency, thus allowing to maintain an efficient photosynthesis under drought conditions.

The increase in the activity of antioxidant enzymes in plants is a common adaptive response under drought stress (Gill & Tuteja, 2010). In our study, drought stress generally caused an increase in the activities of POD and CAT, which are involved in the synthesis of ROS. A similar study reported an increase in the activities of SOD, POD and CAT in *P. ternata* leaves as a consequence of drought stress (Pi *et al*., 2007). Under drought stress, the POD and CAT activities were significantly and strongly positively correlated, indicating an integrated role of the two antioxidant enzymes to scavenge the ROS. Therefore, the increase in the POD and CAT activities during drought stress may act to protect plants against oxidative stress.

MDA is one of the markers for lipid peroxidation in plants under drought stress (Liu *et al*., 2014). In this present study, the content of MDA in *P. ternata* leaves increased significantly under drought stress, indicating membrane damage. This result is in line with previous research showing drought stress induced membrane lipid peroxidation (Naya *et al*., 2007; Filippou *et al*., 2011). Moreover, the MDA content increased considerably in *P. ternata* leaves under severe drought stress, indicating a more extreme peroxidation in the membrane.

The accumulation of soluble protein is considered as an adaptive response of plants to drought stress. In this study, osmotic stress induced by drought stress increased the soluble protein content of *P. ternata* tubers (Fig. 3), which is consistent with other plants (Vendruscolo *et al*., 2007; Xu *et al*., 2015). Previous studies have indicated that the activity of cysteine proteinase and protease play a crucial role in protein degradation, which is enhanced by a lack of water (Dhanda *et al*., 2004). Thus, the soluble protein content dramatically increased; however, it significantly decreased in *P. ternata* tubers with severe drought stress. This phenomenon indicated that the protein metabolism in *P. ternata* tubers was higher than catabolism under severe drought stress, leading to the dramatic breakdown of soluble protein. This result is consistent with those reported by Shan *et al*., (2015).

Soluble sugar can accumulate to mitigate the negative effects of a decrease in water availability. However, our results indicated that soluble sugar content of *P. ternata* tubers was significantly higher in the Control treatment than the other drought treatments. Approximately 80% of the CO_2 assimilated during photosynthesis was soluble sugar (Koch, 2004), and a significant positive correlation between the total Chl content and soluble sugar was observed (Table 3), perhaps showing that drought stress decreased the photosynthesis rate leading to a decrease in the materials for organic carbon synthesis and ultimately in organic carbon.

Previous studies reported that nine alkaloids were isolated from *P. ternata* and that total alkaloids are often used as quality control markers for *P. ternata* tubers (Ji *et al*., 2014). In our experiment, it was observed that drought stress was beneficial to total alkaloids accumulation in *P. ternata* tubers, in particular when the plants in the severe drought environment; these results are similar to those of Kirk *et al*., (2010) and Çakira and Çebib (2010). Similarly, pharmacological research indicated that total alkaloids inhibited peroxidation in rat lung homogenate and enhanced the activity of antioxidases (e.g., SOD) against ROS (Zhao *et al*., 2016). Moreover, POD and CAT activities were positively correlated with total alkaloids (Table 4), perhaps indicating an integrated role of these two antioxidant enzymes along with total alkaloids in the elimination of ROS. This might indicate that total alkaloids undertake essential affect in protecting the plants under drought stress.

Guanosine is one of the most important secondary metabolites in *P. ternata* tubers. This compound is a well-known modulator of the glutamatergic system, which prevents glutamate-induced oxidative stress and has a neuroprotective function (Cristiane *et al*., 2016). In our experiment, the guanosine content in *P. ternata* tubers significantly decreased under drought stress, which is consistent with Zhang *et al*., (2014) who showed that glutamate synthetase (GOGAT) and glutamine synthetase (GS) activities in *P. ternata* were significantly inhibited in plants under drought stress. Due to the GS/GOGAT cycle there is an initial production of glutamine and aspartic acid - combined with other substances leading to the synthesis of hypoxanthine and then further synthesis of guanosine and adenosine (Zhang *et al*., 2014). Therefore, the guanosine content in tubers of *P. ternata* under drought stress was sharply reduced, which suggests that lower water availability is not useful to its synthesis.

Succinic acid is an important type of bioactive compound in *P. ternata* tubers. The Chinese Pharmacopoeia (2015 edition) considers the amount of succinic acid as the only criterion for quality control of Pinelliae Rhizoma. In this study, drought stress significantly inhibited succinic acid synthesis in *P. ternata* tubers, especially subjected to severe drought stress, which was also observed by Qin *et al*., (2011). Previous studies have indicated that the key enzyme activities of malate dehydrogenase (MDH), isocitrate dehydrogenase (IDH) and succinate dehydrogenase (SDH) have an important role in succinic acid synthesis of the tricarboxylic acid cycle (TCA) pathway, and they were significantly decreased as a result of drought stress (Qin *et al*., 2011). Thus, our results indicated that drought stress is disadvantageous to succinic acid accumulation.

In this study, total alkaloid accumulation significantly increased under drought stress. However, drought also reduced *P. ternata* growth and tuber biomass production. Meanwhile, *P. ternata* tuber biomass and bioactive compounds (i.e., guanosine and succinic acid) were significantly higher under a well-watered regime compared with the other drought treatments. Therefore, the maximum yields of total alkaloids, guanosine and succinic acid were obtained under the well-watered treatment, but they decreased dramatically under the drought treatments.

Conclusions

The results demonstrate that the dry and fresh tuber weights, plant height, leaf area, number of tuber, tuber propagation index, and photosynthetic pigment contents were highest in *P. ternata* plants grown under the well-watered regime (75% field capacity). The increase in POD and CAT activities and MDA content demonstrates their potential importance in the adaptation of the *P. ternata* plant to drought conditions. The soluble protein content of *P. ternata* tubers significantly increased under moderate drought stress, nevertheless, the soluble sugar content significantly decreased under severe and moderate drought stress. In addition, the total alkaloid content of *P. ternata* tuber increased under severe drought stress, whereas guanosine and succinic acid of *P. ternata* tubers significantly decreased under drought stress. The maximum yields of total alkaloids, guanosine and succinic acid were obtained under the well-watered treatment. These results demonstrate how *P. ternata* responds to drought stress and can be used to assist cultivation of *P. ternata* plants with the aim of increasing the medicinal qualities of their tubers.

Acknowledgments

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