Effects of Simulated Drought Stress on Seed Germination and Seedling Growth of Manglietia patungensis (Postprint)

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Abstract

The native population of *Manglietia patungensis* exhibits poor natural regeneration, with few regenerated seedlings or saplings observed in the understory. To investigate the tolerance of its seed germination and seedlings to drought stress during the growth period, this study used *Manglietia patungensis* seeds as experimental material and employed different mass concentrations of polyethylene glycol (PEG-6000) to simulate drought stress, analyzing the effects on seed germination, seedling growth, and related physiological and biochemical indices. The results showed that different mass concentrations of PEG solution exerted significant effects on seed germination and seedling growth of *Manglietia patungensis* (P<0.05), with germination rate and survival rate gradually decreasing as PEG mass concentration increased; when PEG mass concentration \$ \$301 g · L-1, seed germination was inhibited and germination time was significantly delayed. With increasing drought stress intensity, chlorophyll content in *Manglietia patungensis* seedlings gradually decreased; soluble protein content and superoxide dismutase (SOD) activity exhibited varying degrees of initial increase followed by decreasing trends; while proline (Pro) and malondialdehyde (MDA) contents and peroxidase (POD) and ascorbate peroxidase (APX) activities showed gradually increasing trends. Based on comprehensive analysis, seed germination and seedling growth of *Manglietia patungensis* have obvious water requirements, and although they can adapt to a certain degree of drought stress through osmotic adjustment and enhanced protective enzyme activity, drought damage should be prevented during the cultivation process. The research results can provide a theoretical basis for seed breeding and regeneration of *Manglietia patungensis*, contributing to its population expansion.

Full Text

Title and Authors

Effects of Simulated Drought Stress on Seed Germination and Seedling Growth of *Manglietia patungensis*

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Abstract

The native populations of *Manglietia patungensis* exhibit poor natural regeneration, with few seedlings or saplings observed in the understory. To investigate the tolerance of its seed germination and seedling growth to drought stress, this study utilized *M. patungensis* seeds exposed to different concentrations of polyethylene glycol (PEG-6000) to simulate drought conditions, analyzing the effects on seed germination, seedling growth, and related physiological and biochemical indicators. The results demonstrated that PEG solutions at various concentrations significantly affected seed germination and seedling growth of *M. patungensis* ($P < 0.05$). Both germination rate and survival rate decreased progressively with increasing PEG concentration; seed germination was inhibited when PEG concentration reached \$ $301g \cdot L$^{-1}\$, and germination time was significantly delayed. As drought stress intensified, chlorophyll content in seedlings gradually decreased; soluble protein content and superoxide dismutase (SOD) activity showed initial increases followed by decreases, while proline (Pro) and malondialdehyde (MDA) contents and peroxidase (POD) and ascorbate peroxidase (APX) activities increased progressively. In summary, *M. patungensis* seeds and seedlings have a clear water requirement. Although they can adapt to moderate drought stress through osmotic adjustment and enhanced protective enzyme activity, drought damage must be prevented during cultivation. These findings provide a theoretical basis for seed propagation and population restoration of *M. patungensis*, contributing to population expansion.

Keywords: *Manglietia patungensis*, drought stress, seed germination, seedling growth, seedling physiology

Classification Code: Q945

Introduction

Seed germination and seedling growth represent the initial stages of a plant's life cycle, profoundly influencing its overall fitness and adaptive capacity (Donohue et al., 2010). During these early developmental phases, seeds lack fully developed systems for acquiring and utilizing resources, making them particularly vulnerable to environmental stresses (James et al., 2011; Larson et al., 2015). Water is essential for seed germination and plays a critical role in seedling establishment. Research indicates that drought stress disrupts seed metabolism, reduces root activity, soluble sugar content, and respiration, leading to insufficient energy production for germination and negatively impacting both germination and early seedling survival (Doupis et al., 2011). Under drought conditions, germination rate and vigor decrease significantly (Wu et al., 2019; He et al., 2019), while prolonged drought severely inhibits seedling growth (Peng et al., 2018), causing water loss and inducing osmotic stress and reactive oxygen species damage.

However, some plants have evolved strategies to cope with drought through physiological adaptations such as reducing cellular osmotic potential and altering enzyme synthesis in protective systems. Polyethylene glycol (PEG) is an ideal high-molecular-weight organic compound for simulating drought environments and is widely used in experiments investigating osmotic and drought stress effects on seeds and seedlings (Muscolo & Iglesias, 2014).

Manglietia patungensis is an evergreen tree species in the Magnoliaceae family, sporadically distributed in Hubei, Chongqing, and Hunan provinces. Due to its narrow distribution and small population size, it is considered endangered (IUCN, 2015). As the northernmost species in the *Manglietia* genus, *M. patungensis* holds significant value for research on the phylogeny and evolution of Magnoliaceae, as well as for ornamental and medicinal applications (Ge et al., 2009; Li et al., 2004). Natural regeneration is poor in native populations, with few seedlings or saplings observed in the understory, suggesting that soil moisture may be a primary factor limiting seed germination (Li et al., 2006). Previous studies on *M. patungensis* seed biology indicate that under artificial conditions, optimal germination occurs at 20–30 °C with rates reaching 100%, whereas natural germination rates are extremely low, with moisture likely being a key limiting factor (Chen et al., 2007; Chen et al., 2012). Therefore, investigating the drought tolerance of *M. patungensis* seed germination and seedling growth is crucial for population recovery. This study employs artificial drought simulation to examine the relationship between seed germination, seedling growth, and soil moisture, providing a scientific basis for seedling cultivation and population restoration of *M. patungensis*.

Materials and Methods

1.1 Experimental Materials

Manglietia patungensis seeds were collected on October 5, 2016, from Xiaoxi National Nature Reserve in Hunan Province. After removing the outer seed coat, seeds were stored in sand at 4 °C. In March of the following year, PEG-simulated drought stress experiments were conducted at the Biotechnology Research Center of China Three Gorges University.

1.2 Experimental Design

Distilled water served as the control (CK). Seven PEG-6000 solutions were prepared at concentrations of 95, 140, 170, 221, 250, 301, and 345 $g \cdot L^{-1}$ (Mai et al., 2009). Two layers of filter paper were placed in petri dishes as germination beds, moistened with different PEG solutions. Sterilized *M. patungensis* seeds were evenly distributed on the germination beds (30 seeds per dish, four replicates). Dishes were incubated at 25 °C in darkness for 25 days, then transferred to light conditions (2,000 lx, 16 h/d).

1.3 Measurement Indicators

1.3.1 Seed Germination and Growth Indices Germination was defined as radicle emergence through the seed coat. Germinated seed counts and seedling growth status were recorded every 3 days. Germination was considered complete when no seeds germinated for 5 consecutive days, and germination rate was calculated. Embryonic axis and radicle lengths were measured with a ruler for each germinated seed. Survival rate was determined on day 55. Germination rate = (number of germinated seeds / total seeds tested) × 100%; Survival rate = (number of surviving seedlings / total seedlings) × 100%.

1.3.2 Seedling Physiological Indices On day 55, 10 g of cotyledons and true leaves were collected from each treatment group, snap-frozen in liquid nitrogen, and stored at −80 °C. Soluble protein content was determined using the Coomassie brilliant blue method (Qu et al., 2006). Proline (Pro) content was measured via ninhydrin colorimetry (Zhang et al., 1990; Li et al., 2005). Malondialdehyde (MDA) content was assessed using the thiobarbituric acid (TBA) method (Wang et al., 2013). Chlorophyll content was determined by UV spectrophotometry (Li, 2006). Superoxide dismutase (SOD) activity was measured using the nitroblue tetrazolium (NBT) reduction method (Li, 2006). Peroxidase (POD) activity was quantified via guaiacol colorimetry (Zheng et al., 2010). Ascorbate peroxidase (APX) activity was determined by UV spectrophotometry (Song et al., 2011).

1.4 Data Analysis

Data processing and graphing were performed using Excel. Statistical analysis was conducted with SPSS 21.0 software, with significance set at $P < 0.05$.

Results

2.1 Effects of Drought Stress on Seed Germination

Germination results [Figure 1: see original paper] showed that control group seeds initiated germination at day 10. Germination onset in treatment groups was progressively delayed with increasing PEG concentration: 15, 20, 20, 28, and 40 days for concentrations of 95, 140, 170, 221, and 250 $g \cdot L^{-1}$, respectively. No germination occurred at PEG concentrations $ 301*g* · *L*^{-1}$. These results indicate that low PEG concentrations delayed germination, with more severe drought stress causing significant germination postponement.

As shown in Table 1 , both germination rate and survival rate of *M. patungensis* seeds decreased significantly with increasing PEG concentration ($P < 0.05$). At 250 $g \cdot L^{-1}$ PEG, germination and survival rates were only 21.67% and 17.50% of control values, respectively, demonstrating that high PEG concentrations inhibited seed germination. Although germination rate declined with PEG concentration, some seeds still germinated and grew at concentrations $ 250*g* · *L*^{-1}$, indicating that *M. patungensis* seeds possess certain drought resistance capabilities.

Seedling radicle and hypocotyl lengths decreased with increasing PEG stress. Control seedlings exhibited the best growth, with average hypocotyl and radicle lengths of 6.88 cm and 5.44 cm, respectively. At 250 $g \cdot L^{-1}$ PEG, these values decreased to 0.14 cm and 0.65 cm, with significant differences observed among all treatments ($P < 0.05$). As drought stress intensified, some seedling root tips exhibited blackening, root rot, and mortality, indicating that drought stress inhibits *M. patungensis* seedling growth in a concentration-dependent manner.

2.2 Effects of Drought Stress on Seedling Physiological Indices

No physiological measurements were conducted for the 301 and 345 $g \cdot L^{-1}$ treatments as seeds failed to germinate at these concentrations.

2.2.1 Effects on Chlorophyll Content Chlorophyll content in *M. patungensis* seedlings decreased progressively with increasing PEG concentration [Figure 2: see original paper]. The control group showed the highest chlorophyll content at 0.026 $mg \cdot g^{-1}$, while the 250 $g \cdot L^{-1}$ treatment exhibited the lowest value at only 27% of the control. Significant differences were observed between the control and all treatment groups. No significant difference was found between the 95 and 140 $g \cdot L^{-1}$ treatments, nor between the 221 and 250 $g \cdot L^{-1}$ treatments; however, the 170 $g \cdot L^{-1}$ treatment differed significantly from both the 221 and 250 $g \cdot L^{-1}$ treatments ($P < 0.05$). These results demonstrate that drought stress impairs chlorophyll synthesis in *M. patungensis* seedlings, with increasing PEG concentrations reducing chlorophyll content and consequently affecting photosynthesis and normal growth.

2.2.2 Effects on Osmotic Regulation Soluble protein content initially increased then decreased with intensifying drought stress [Figure 3A: see original paper]. Content rose gradually from 0 to 221 $g \cdot L^{-1}$ PEG, reaching a maximum of 3.93 $mg \cdot g^{-1}$, then declined at 250 $g \cdot L^{-1}$. The control group showed the lowest soluble protein content at 1.00 $mg \cdot g^{-1}$. All treatments differed significantly except between the 170 and 250 $g \cdot L^{-1}$ treatments ($P < 0.05$). Proline (Pro) content increased progressively with PEG concentration [Figure 3B: see original paper]. Pro content remained low at \$ 95$g \cdot L$^{-1}\$ PEG, increased slowly at 140, 170, and 221 $g \cdot L^{-1}$ (all higher than control), and reached its maximum of 0.19 $mol \cdot g^{-1}$ at 250 $g \cdot L^{-1}$ (approximately 3.3 times the control value). The 250 $g \cdot L^{-1}$ treatment differed significantly from all other groups ($P < 0.05$), while other treatments showed no significant differences among themselves. These findings indicate that *M. patungensis* seedlings employ osmotic adjustment by accumulating soluble proteins and proline to enhance cellular water retention and mitigate drought damage.

2.2.3 Effects on Cell Plasma Membrane Malondialdehyde (MDA) content correlated positively with PEG concentration, increasing progressively with drought stress [Figure 4: see original paper]. At \$ 95$g \cdot L$^{-1}\$ PEG, MDA content increased slowly without significant difference from the control. From 140 to 221 $g \cdot L^{-1}$, MDA content increased gradually with no significant differences among these treatments. At 250 $g \cdot L^{-1}$, MDA content reached 0.37 $g \cdot g^{-1}$, approximately 2.2 times the control value, showing a sharp increase that differed significantly from all other treatments ($P < 0.05$). These results suggest that *M. patungensis* seedlings can resist membrane lipid peroxidation damage from moderate drought stress, but severe drought causes substantial plasma membrane injury, impairing normal growth.

2.2.4 Effects on Protective Enzyme Activity Superoxide dismutase (SOD) activity initially increased then decreased with PEG stress [Figure 5A: see original paper]. Activity increased sharply at 95 $g \cdot L^{-1}$ PEG, reaching its maximum of 0.34 $U \cdot g^{-1}$, then declined with further stress intensification. No significant difference was observed between the 95 and 140 $g \cdot L^{-1}$ treatments, nor between the 170 and 221 $g \cdot L^{-1}$ treatments. At 250 $g \cdot L^{-1}$, SOD activity fell below control levels, showing no significant difference from the control but differing significantly from other treatments ($P < 0.05$). Peroxidase (POD) activity increased overall with PEG concentration [Figure 5B: see original paper], rising gradually initially then increasing rapidly at 250 $g \cdot L^{-1}$ to 2.48 $U \cdot$

g^{-1}, which differed significantly from all other groups ($P < 0.05$). The control showed the lowest POD activity at 0.81 $U \cdot g^{-1}$, not significantly different from the 95 $g \cdot L^{-1}$ treatment but differing significantly from 140, 170, and 221 $g \cdot L^{-1}$ treatments ($P < 0.05$). Ascorbate peroxidase (APX) activity increased progressively with PEG concentration [Figure 5C: see original paper], with all treatments showing higher activity than the control. All treatments differed significantly except between the 140 and 170 $g \cdot L^{-1}$ treatments ($P < 0.05$). These findings indicate that under low PEG concentrations, *M. patungensis* seedlings cope with stress through elevated SOD activity. As stress intensifies, SOD activity is suppressed while POD and APX activities increase and remain high, with these protective enzymes scavenging reactive oxygen species to mitigate drought-induced damage.

Discussion and Conclusion

Drought stress affects not only seed vigor but also germination timing and rate. Under simulated drought conditions, adverse effects on *M. patungensis* seed germination and seedling growth became evident at PEG concentrations between 140 and 250 $g \cdot L^{-1}$, with germination rates declining as PEG concentration increased. Seeds lost viability and failed to germinate at concentrations >250 $g \cdot L^{-1}$, consistent with findings for *Catalpa ovata* (He et al., 2019). The study also revealed that increasing stress severity not only reduced germination rates but also significantly delayed germination onset, suggesting that *M. patungensis* possesses adaptive mechanisms to cope with drought—an evolutionary response to its native environment. This delayed germination strategy may disperse the risks associated with drought stress, benefiting population persistence. Some studies have reported that low PEG concentrations promote germination in woody species such as *Quercus variabilis* (Li et al., 2013) and *Metasequoia glyptostroboides* (Wu et al., 2019), possibly reflecting species-specific biological characteristics.

Chlorophyll is fundamental to photosynthesis, and its content serves as an important indicator of plant drought tolerance (Sang et al., 2011). Drought stress inhibits chlorophyll synthesis while accelerating chloroplast degradation, leading to significant chlorophyll reduction (Ma et al., 2018). This study corroborates these findings, showing that *M. patungensis* seedling chlorophyll content decreased significantly with increasing PEG concentration, indicating severe impairment of photosynthesis under high PEG stress.

Plants under drought stress accumulate compatible solutes such as soluble sugars and proline to confer physiological protection and enhance drought tolerance (Azmat & Moin, 2019; Muscolo & Iglesias, 2014). In this study, soluble protein and free proline contents in *M. patungensis* seedlings increased with PEG concentration, suggesting that osmotic adjustment through solute accumulation helps maintain cellular water potential and turgor, thereby reducing stress damage. These results align with drought responses reported for many plant species (Wang et al., 2018; Wang et al., 2014). However, soluble protein content began to decline at $250g \cdot L^{-1}$ PEG, likely because high PEG concentrations denature protein synthesis enzymes, causing protein degradation to exceed synthesis.

MDA serves as an indicator of cell membrane damage, with elevated levels reflecting greater stress injury (Luo et al., 2017). This study found that MDA content in *M. patungensis* seedlings increased with PEG concentration, rising slowly at 0–221 $g \cdot L^{-1}$ but increasing sharply at 250 $g \cdot L^{-1}$, indicating severe plasma membrane damage at this stress level. These results are consistent with findings for *Cercidiphyllum japonicum* seedlings (Mai et al., 2009).

Plants have evolved protective enzymes including SOD, POD, and APX to combat reactive oxygen species damage (Li et al., 2013; Zheng et al., 2018). This study revealed that SOD activity in *M. patungensis* seedlings increased initially then decreased under low PEG stress, while POD and APX activities continued to rise. This suggests that under mild stress, seedlings enhance SOD, POD, and APX activities to eliminate reactive oxygen species. At $95g \cdot L^{-1}$ PEG, SOD primarily scavenged oxygen free radicals to protect membrane systems. As PEG concentration increased, SOD activity declined while POD and APX activities rose. At 95–250 $g \cdot L^{-1}$ PEG, POD catalyzed H_2O_2 degradation and APX participated in the ascorbate-glutathione cycle to scavenge H_2O_2 from chloroplasts, cytoplasm, and microbodies, thereby mitigating drought damage. However, at $250g \cdot L^{-1}$ PEG, despite elevated POD and APX levels, accumulation of harmful substances exceeded the scavenging capacity, causing seedling injury. These findings are similar to drought responses in *Magnolia wufengensis* (Sang et al., 2011) and *Polygala tenuifolia* (Peng et al., 2018), though protective enzymes could not completely eliminate drought effects.

In conclusion, *M. patungensis* seeds and seedlings have a clear water requirement. Under moderate drought stress (PEG 95–221 $g \cdot L^{-1}$), they can adapt through osmotic adjustment and protective enzyme activity, minimizing damage. However, severe drought stress (PEG $250g \cdot L^{-1}$) causes substantial harm. Therefore, moisture management is critical during *M. patungensis* cultivation to improve germination rates and seedling survival. These findings were obtained under specific experimental conditions and have certain limitations; further investigation is needed to identify the barriers to natural regeneration in native *M. patungensis* populations.

References

AZMAT R, MOIN S, 2019. The remediation of drought stress under VAM inoculation through proline chemical transformation action[J]. J Photochem Photobiol B, 193:155-161.

CHEN FJ, LIANG HW, WANG X, et al., 2007. Seed dormancy and germination characteristics of *Manglietia patungensis*, an endangered plant endemic to China [J]. Biodivers Sci, 15(5): 492-499. [Chen Faju, Liang Hongwei, Wang Xu, et al., 2007. Study on seed dormancy and germination characteristics of the endangered plant *Manglietia patungensis* [J]. Biodiversity Science, 15(5): 492-499.]

CHEN FQ, LIU L, CHEN FJ, et al., 2012. The ecological characteristics of seed germination and seedling establishment of *Manglietia patungensis*: implication for species conservation[J]. Am J Plant Sci, 3; 1455-1461

DONOHUE K, RAFAEL RDC, BURGHARDT L, et al., 2010. Germination, post-germination adaptation, and species ecological ranges[J]. Ann Rev Ecol Syst, 41(41): 293-319.

DOUPIS G, CHARTZOULAKIS K, BEIS A, et al., 2011. Allometric and biochemical responses of grapevines subjected to drought and enhanced ultraviolet-B radiation[J]. Aust J Grape Wine Res, 17(1): variation

HE RH, LI Y, XIAO Y, et al., 2019. Effect of simulated drought stress on seed germination and [J]. Guihaia. provenances geographical http://kns.cnki.net/kcms/detail/45.1134.Q.20190801.1807.006.html. [He Runhua, Li Yuan, Xiao Yao, et al., 2019. Study on seed germination characteristics and geographical variation of six provenances of *Catalpa ovata* under simulated drought stress [J]. Guihaia, http://kns.cnki.net/kcms/detail/45.1134.Q.20190801.1807.006.html.]

IUCN Red List, 2015. *Magnolia patungensis*. The IUCN Red List of Threatened Species. Available at http://dx.doi.org/10.2305/IUCN.UK.2015-2.RLTS.T15115907A15115911.en.

JAMES JJ, SVEJCAR TJ, RINELLA MJ, 2011. Demographic processes limiting seedling recruitment in arid grassland restoration[J]. J Appl Ecol, 48(4): 961-969.

LARSON JE, SHELEY RL, HARDEGREE SP, et al. 2015. Seed and seedling traits affecting critical life stage transitions and recruitment outcomes in dryland grasses[J]. J Appl Ecol, 52(1): 199-209.

LI HS, 2006. Principles and techniques of plant physiology and biochemistry experiments [M]. Beijing: Higher Education Press. [Li Hesheng, 2006. Principles and Techniques of Plant Physiology and Biochemistry Experiments [M]. Beijing: Higher Education Press.]

LI SJ, GONG YH, WANG JR, et al., 2005. Discussion on the reaction between proline and ninhydrin in the determination of proline content by ninhydrin method[J]. Plant Physiol Comm., 41(3): 365-368. [Li Shaojun, Gong Yuehua, Wang Junru, et al., 2005. Discussion on the reaction between proline and ninhydrin in the determination of proline content by ninhydrin method [J]. Plant Physiology Communications, 41(3): 365-368.]

LI XD, HUANG HW, LI ZZ, et al., 2004. Distribution and conservation strategy of endangered distribution and conservation strategies of the endangered *Manglietia patungensis* Hu [J]. J Wuhan Bot Res, 22(5): 421-427. [Li Xiaodong, Huang Hongwen, Li Zuozhou, et al., 2004. Distribution and conservation strategies of the endangered *Manglietia patungensis* [J]. Journal of Wuhan Botanical Research, 22(5): 421-427.]

LI XD, HUANG HW, LI JQ, et al., 2006. Community structure of *Manglietia patungensis* in Xiaoxi Natural Reserve, Hunan Province [J]. J Wuhan Bot Res, 24(1): 31-37. [Li Xiaodong, Huang Hongwen, Li Jianqiang, et al., 2006. Community structure of *Manglietia patungensis* in Xiaoxi Natural Reserve, Hunan Province [J]. Journal of Wuhan Botanical Research, 24(1): 31-37.]

LI ZP, ZHANG WH, CUI YC, 2013. Effects of PEG simulated drught stress on seed germination and growth physiology of *Quercus variabili* [J]. Acta Bot Boreal-Occident Sin, 33(10): 2043-2049. [Li Zhiping, Zhang Wenhui, Cui Yuchuan, 2013. Effects of PEG-simulated drought stress on seed germination and growth physiology of *Quercus variabilis* [J]. Acta Botanica Boreali-Occidentalia Sinica, 33(10): 2043-2049.]

LUO QH, NING HS, HE M, et al., 2017. Ecophysiological responses of five sandy shrubs to drought stress[J]. Sci Silvae Sinicae, 53(11): 29-42. [Luo Qinghong, Ning Husen, He Miao, et al., 2017. Ecophysiological responses of five sandy shrubs to drought stress [J]. Scientia Silvae Sinicae, 53(11): 29-42.]

MA XH, ZHENG J, ZHANG XL, et al., 2018. The influence of drought stress on physiological and biochemical characteristics of *Lagerstroemia speciosa* and *Lagerstroemia balansae*[J]. J Southwest forest Univ (Nature Sci Ed), 38(5): 205-209. [Ma Xiaohua, Zheng Jian, Zhang Xule, et al., 2018. Influence of drought stress on physiological and biochemical characteristics of *Lagerstroemia speciosa* and *Lagerstroemia balansae* [J]. Journal of Southwest Forestry University (Natural Sciences), 38(5): 205-209.]

MAI MM, SHI DX, WANG ML, et al., 2009. Seed germination and seedling growth of *Cercidiphyllum japonicum* with PEG treatment[J]. Sci Silv Sin, 45(10): 94-99. [Mai Miaomiao, Shi Daxing, Wang Mili, et al., 2009. Effects of PEG treatment on seed germination and seedling growth of *Cercidiphyllum japonicum* [J]. Scientia Silvae Sinicae, 45(10): 94-99.]

MANTOVANI A, IGLESIAS RR, 2010. The effect of water stress on seed germination of three terrestrial bromeliads from restinga[J]. Braz J Bot, 33(1): 201-205.

MUSCOLO A, SIDARI M, ANASTASI U, et al., 2014. Effect of PEG-induced drought stress on seed germination of four lentil genotypes[J]. J Plant Interact, 9(1):354-363.

PENG L, YANG BY, ZHANG G, et al., 2018. Seed germination and seedling growth physiological characteristics of *Polygala tenuifolia* Willd. under drought stress[J]. Acta Bot Boreal-Occident Sin, 38(4): 741-749.[Peng Liang, Yang

Bingyue, Zhang Gang, et al., 2018. Seed germination and seedling growth physiological characteristics of *Polygala tenuifolia* Willd. under drought stress [J]. Acta Botanica Boreali-Occidentalia Sinica, 38(4): 741-749.]

QU CX, SHEN YD, WANG XF, et al., 2006. Study on the determination of soluble protein content in crude extracts of plants by Coomassie Brilliant Blue [J]. J Suzhou Univ (Nature Sci Ed), 22(2): 82-85. [Qu Chunxiang, Shen Songdong, Wang Xuefeng, et al., 2006. Study on the determination of soluble protein content in crude extracts of plants by Coomassie Brilliant Blue [J]. Journal of Suzhou University (Natural Science Edition), 22(2): 82-85.]

SANG ZY, MA LY, CHEN FJ, et al., 2011. Growth and physiological characteristics of *Magnolia wufengensis* seedlings under drought stress[J]. Acta Bot Boreal-Occident Sin, 31(1): 109-115. [Sang Ziyang, Ma Lüyi, Chen Faju, et al., 2011. Growth and physiological characteristics of *Magnolia wufengensis* seedlings under drought stress [J]. Acta Botanica Boreali-Occidentalia Sinica, 31(1): 109-115.]

SHENG WB, XU LL, YE MB, et al., 1996. Study on determination of ASP activity[J]. Plant Phytophysiol Sin, (3): 203-205. [Shen Wenbiao, Xu Lailai, Ye Maobing, et al., 1996. Study on determination of APX activity [J]. Acta Phytophysiologica Sinica, (3): 203-205.]

WANG JY, SHI DH, BAI Y, et al., 2018. Effect of drought stress with PEG-6000 on Seed germination and physiological properties in *Abelmoschus esculentus*[J]. J Trop Subtrop Bot, 26(6): 611-616. [Wang Jiyue, Shi Denghong, Bai Yu, et al., 2018. Effect of drought stress with PEG-6000 on seed germination and physiological properties in *Abelmoschus esculentus* [J]. Journal of Tropical and Subtropical Botany, 26(6): 611-616.]

WANG ML, WEI X, KONG DX, et al., 2014. Effects of drought stress and rehydration on physiological characteristics of *Ardisia corymbifera* var. *tuberifera* seedlings[J]. Guihaia, 34(1): 105-110.[Wang Manlian, Wei Xiao, Kong Dexin, et al., 2014. Effects of drought stress and rehydration on physiological characteristics of *Ardisia corymbifera* var. *tuberifera* seedlings [J]. Guihaia, 34(1): 105-110.]

WANG Y, HU S, FU WC, et al., 2013. A new method for fast determination of total soluble sugars content in plant tissue: TBA-method[J]. J Jinggangshan Univ (Nature Sci Ed), 34(3): 37-40.[Wang Yan, Hu Sheng, Fu Wencheng, et al., 2013. A new method for fast determination of total soluble sugars content in plant tissue: TBA-method [J]. Journal of Jinggangshan University (Natural Science Edition), 34(3): 37-40.]

WU ML, ZHU J, AI XR, et al., 2019. Influences of simulating drought stress on seed germination of *Metasequoia glyptostroboides* under different temperature[J]. Guihaia. http://kns.cnki.net/kcms/detail/45.1134.Q.20190729.1413.002.html. [Wu Manling, Zhu Jiang, Ai Xunru, et al., 2019. Influences of simulating drought stress on seed germination of *Metasequoia glyptostroboides* under different temperature [J]. Guihaia, http://kns.cnki.net/kcms/detail/45.1134.Q.20190729.1413.002.html.]

ZHANG DT, TING PH, ZHAO XY, et al., 1990. Determination of the content of free proline in wheat leaves [J]. Plant Physiol Comm, (4): 62-65. [Zhang Dianting, Ting Peihong, Zhao Xiyi, 1990. Determination of the content of free proline in wheat leaves [J]. Plant Physiology Communications, (4): 62-65.]

ZHENG QL, YANG ZR, ZHANG FL, et al., 2018. Responses of reactive oxygen species scavenging system to drought stress in *Pugionium* Gaertn[J]. Acta Bot Boreal-Occident Sin,38(9): 1674-1682. [Zheng Qingling, Yang Zhongren, Zhang Fenglan, et al., 2018. Responses of reactive oxygen species scavenging system to drought stress in *Pugionium* Gaertn [J]. Acta Botanica Boreali-Occidentalia Sinica, 38(9): 1674-1682.]

ZHENG SY, SHANG XF, WANG JP, et al., 2010. Determination of antioxidant enzyme activity and contents of MDA in maize seedlings under salt stress with visible spectrophotometry[J]. Biotechnol Bull, (7): 106-109. [Zheng Shiying, Shang Xuefang, Wang Jingping, et al., 2010. Determination of antioxidant enzyme activity and contents of MDA in maize seedlings under salt stress with visible spectrophotometry [J]. Biotechnology Bulletin, (7): 106-109.]

Note: Figure translations are in progress. See original paper for figures.