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Banat's Journal of Biotechnology

2020, XI(21)

Effects of titanium and silicon nanoparticles on antioxidant enzymes activity and some biochemical properties of *Cuminum cyminum* L. under drought stress

DOI: 10.7904/2068–4738–XI(21)–19

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Abstract. Environmental stresses change antioxidant and non–enzymatic activities of plant cells. Nanoparticles may help plants against drought stress. This study was conducted to evaluate the effect of titanium and silicon nanoparticles on antioxidant enzymes activity and some biochemical properties of *Cuminum cyminum* L. under drought stress. This study was conducted based on randomized complete block design in an arrangement of split plot with three replications, and irrigation content (50, 100, and 150 mm evaporation), and foliar applications of titanium and silicon nanoparticles, and soil application of superabsorbent polymer as factors. The results showed that levels of water, titanium, silicon nanoparticles, superabsorbent polymer, and their interactions were efficient on growth of *Cuminum cyminum* L. Drought stress decreased relative humidity, but increased antioxidant enzyme activity, leaf proline, and carbohydrate. Application of titanium nanoparticles increased leaf proline. It was observed a significant interaction between water availability, and fertilizers on ascorbate enzyme activity ($P<0.05$), so that most activity (0.193 μmol/mg) was observed in 100 mm evaporative pan and foliar application of titanium. In sum, drought stress increased carbohydrate, antioxidant enzymes activity and proline contents, and decreased relative water contents.

Keywords: Antioxidant enzymes, *Cuminum cyminum*, Evaporative pan, Nanotechnology, Proline.

Introduction

Drought stress is an environmental challenge that occurs in plants that live in arid and semi–arid regions [CHAVES *et al.*, 2003].

The water shortage increases production of reactive oxygen species (ROS) such as $^1O_2$, O_2^- and H_2O_2, that are very reactive and increase damages on tissues and macromolecules (e.g. DNA, lipids, proteins and carbohydrates) [GILL and TUTEJA, 2010]. The stress condition increases injuries within green tissues, due to production of ROSs [REDDY *et al.*, 2004].

Plants have several mechanisms for increasing their capability for survive and grow during short–and long–term drought stress, such as antioxidant defense systems and osmotic adjustment [YIN *et al.*, 2005].

The antioxidant enzymes, such as superoxide dismutase (SOD), glutathione reductase (GR), ascorbate peroxidase (APX), dehydroascorbate reductase (DHAR), and monodehydroascorbate reductase (MDHAR) have important roles in scavenging ROSs [GILL and TUTEJA, 2010].

Nanotechnology has provided much attention as fertilizer carriers or controlled release vectors for preparing smart fertilizers that increase nutrient use efficiency and decrease costs of environmental protection [SHALLAN *et al.*, 2016].

The metal and metal oxides nanoparticles have some physiochemical properties and are different than their native bulk compounds in some aspects, such as their surface, optical, thermal, and electrical properties [RASTOGI *et al.*, 2017].

Titanium oxide nanoparticles have biological activities on plants, such antioxidant activities [MINGYU *et al.*, 2008].

It also increases shoot, root length, and growth, yield and yield components in radish, corn, lettuce and cucumber [LIN and XING, 2007], Canola plant [MAHMOODZADEH *et al.*, 2013] and wheat plant [JABERZADEH *et al.*, 2013].

The silicon is an important trace element for inducing resistance against distinct stresses, diseases, and pathogens of plants.

SiO_2 nanoparticles increased content in all the photosynthetic pigments [RAD et al., 2014], amounts of total soluble proteins [ABDUL QADOS, 2015], and proline contents [KALTEH et al., 2014].

It also increased antioxidant enzymes activities in faba bean [KALTEH et al., 2014], tomato plant [SIDDIQUI and AL–WHAIBI, 2014] and alfalfa plant [WANG et al., 2011].

The silicon application increased wheat biomass under rice seedlings [AZIMI et al., 2014].

Medicinal plants are used for treating different diseases. Cumin *(Cuminum cyminum L.)* is a medicinal plant in the Apiaceae or Umbelliferae family [TAKESH et al., 2019].

It contains major compounds, such as myrtenal, trans–carveol, O–cymene, cuminique alcohol and other major compounds [REBEY et al., 2012].

It has been used for the treatment of diarrhea, indigestion and bloating, and also as an antimicrobial [KHODAIE and SADEGHPOOR, 2015].

It is used as an important seed spices used in India and other countries for flavoring dishes due to its strong and heavy flavor [RANA et al., 2018].

It is a small and slender annual herb that grows up to a height of about 45 cm with several branches and linear dark green leaves.

Cumin is a profitable medicinal plant, but drought stress can adversely its growth and antioxidant properties.

Titanium and silicon nanoparticles may improve antioxidant properties under drought stress.

This study investigates the effects of titanium and silicon nanoparticles on antioxidant enzymes activity and some biochemical properties of *Cuminum cyminum* L. under drought stress.

Material and methods

To investigate the effect of dioxide titanium nanoparticles, silicon, superabsorbent polymers on quantities, and qualitative properties of *Cuminum cyminum* L. under drought stress, a randomized complete block design was conducted in an arrangement of split plot with three levels of water availability of 50 (control), 100, and 150 mm as main factor, and foliar application of dioxide titanium nanoparticles, and silicon, and also soil application of superabsorbent polymer.

This study was conducted by using three replications.

This study was conducted in Pariz region in Sirjan town.

This region had desert climate based on Köppen–Geiger climate *classification, and had cold and dry weather based Amberge classification.*

Msin factor was drought stress, and other factors included dioxide titanium nanoparticles, silicon, and superabsorbent polymers.

A land piece was considered, and soil analysis and agriculture operations were conducted on plots with dimensions of 3×3.5 m.

Cumin seeds were planted on rows. Per plot included 7 rowsin distances of 35 cm. Seeds were planted in low depth (0.5–1 cm), and covered by a layer of sand.

Measurement of proline, soluble carbohydrates, and relative water content

Proline was assessed as reported by Bates and collab., [BATES et al., 1973], and carbohydrates were evaluated as reported by Keles and Oncel [KELES and ONCEL, 2004].

Relative water content was assessed as reported by Clavel and collab., [CLAVEL et al., 2005].

Assessment of antioxidant activities

Ascorbate peroxidase activity was assessed as reported by Sairam and collab., [SAIRAM et al., 2002].

Activities of catalase enzymes, and guaiacol peroxidase were assessed as gram of fresh weight as reported by Beers and Sizer [BEERS and SIZER, 1952], and Nakano and Asada, [NAKANO and ASADA, 1981], respectively.

Data analysis

The data were analyzed by Excel software. Statistical calculations were conducted by SAS software (version of

9.3), and means were compared at level of 0.05. Figures were illustrated by Graph Pad Prism Software (Version of 5.6).

Results and discussion

The results showed that drought stress had a significant effect on carbohydrate accumulation (Table 1).

Most carbohydrate rate (14.50 µg/g) was observed in drought stress treatments of 150 mm (Table 2).

Increased carbohydrate contents in drought stress could be attributed to increased osmotic pressure in plants for more absorption of soil, and water.

Table 1.

Analysis of variance some traits of cumin

	Df	Carbohydrate	Proline	RWC	Catalase	Guaiacol	Ascorbate
S.O.V.							
Replications	2	0.71	0.41	1.72	0.00003	0.0002	0.0001
Irrigation	2	131.02**	17.32**	2114.4**	0.005**	0.0086**	0.012**
Main error	4	2.67	0.33	6.23	0.00004	0.0002	0.0006
Fertilizer	7	0.52ns	1.60*	15.12ns	0.0007*	0.0003ns	0.001**
Irrigation* Fertilizer	14	1.46ns	0.45ns	16.08ns	0.0005ns	0.0002ns	0.0004*
Error	42	1.68	0.64	25.89	0.0003	0.0003	0.0002
CV%	–	10.69	13.54	7.54	9.69	12.44	10.22

RWC: Leaf relative water content. Superscripts *, **, and ns show significant differences at 0.05, 0.01, and non–significant, respectively.

Carbohydrate distribution is affected by stresses, such as water deficiency, and plant hormones.

It was reported that carbohydrates, and amino acids have important roles in regulating osmotic pressure [FLAGELLA *et al.*, 1995].

Additionally, carbohydrates are connected with proteins, and membrane, form hydrogen bond, and prevent form change under stress condition.

Hexokinases are first sugar sensors that are capable to change gene expression in drought–resistant pathway.

Table 2.

Mean comparison of water availability on antioxidant enzymes activity and biochemical properties of cumin

Water availability	Carbohydrate (µmol/g)	Proline (µg/g)	Relative humidity (%)	Catalase (µmol/mg)	Guaiacol (µmol/mg)
50	9.65c	5.11c	77.18a	0.163c	0.129c
100	12.43b	5.85b	66.80b	0.183b	0.149b
150	14.50a	6.81a	58.45c	0.192a	0.167a

Superscripts (a–c) show significant differences in per column.

The results also showed that main effects of water, and fertilizer treatments on proline content was significant (P<0.01).

Most and lowest proline content were observed in 150 mm, and 50 mm treatments (6.81 µmol/g versus 5.11 µmol/g) (Table 2).

The results also showed that most and lowest proline content were observed in foliar application of titanium, and silicon treatments, respectively (6.67 µmol/g versus 5.37 µmol/g) (Figure 1).

Proline accumulation is a general response against environmental stresses in some organisms, such as plants [LIANG *et al.*, 2013].

Faulted water is a serious threat for plants, and causes physiological changes such as stomatal closure, decreased gaseous exchange and chlorophyll content [CHUN *et al.*, 2018].

Proline is an important amino acid that participates in membrane structures and scavenges ROS under drought stress [ASHRAF and FOOLAD, 2007].

Increased proline accumulation is attributed to an activation of proline production by glutamate pathway.

The results also showed a significant effect between water availability on relative water content (*P*<0.01) (Table 1).

Highest, and lowest relative water content were observed in 50, and 150 mm, respectively (Table 2).

Drought stress decreased relative water content and plant growth. It is a positive correlation between decreased relative water content and soil water.

Leaf relative water content is an important parameter for assessing water status in plants that shows balance between water supply to the leaf tissue and transpiration rate [LUGOJAN and CIULCA, 2011].

It was reported that drought stress increases relative foliar water content, and decreases flower, seed, and essence [BOWEN *et al.*, 1992].

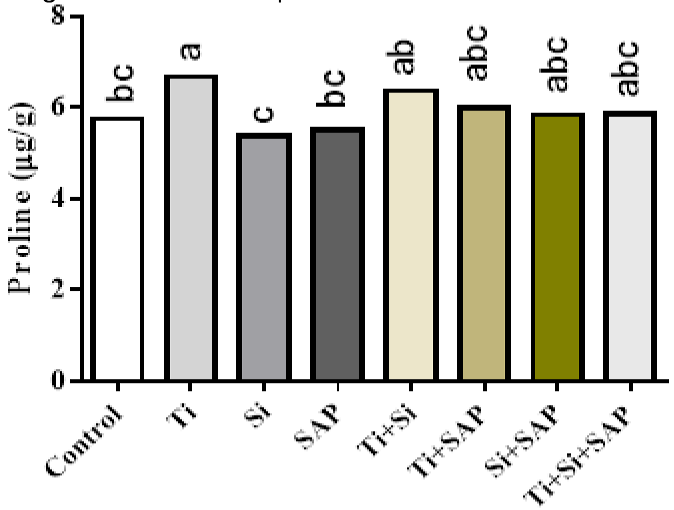

Figure 1. The effects of treatments on proline content of cumin. Ti=Titanium, Si= Silicon, SAP= Superabsorbent polymer

The results showed that main effects of drought, and fertilizer on antioxidant activity were significant (*P*<0.01).

Most antioxidant activity of catalase was observed in 150 mm treatment (0.192 µmol/mg), and titanium nanoparticles fertilizer (0.193 µmol/mg) (Figure 2).

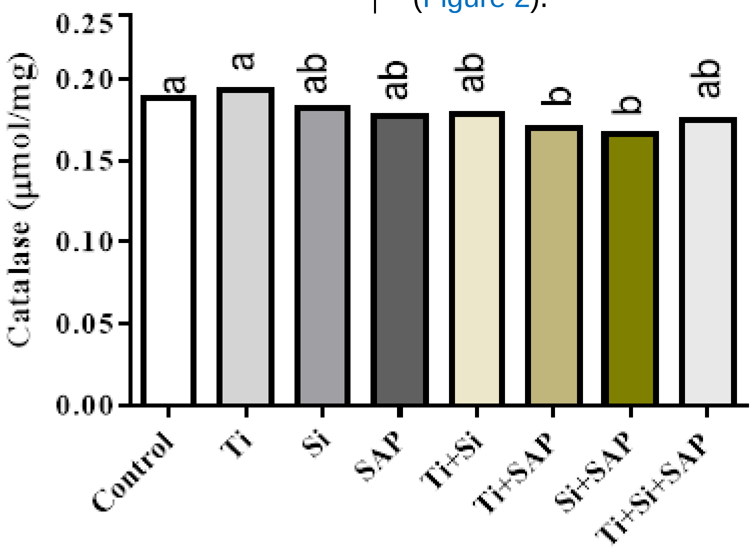

Figure 2. The effects of treatments on catalase activity of cumin. Ti=Titanium, Si= Silicon, SAP= Superabsorbent polymer

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Banat's Journal of Biotechnology
2020, XI(21)

Drought stress had significant effect on antioxidant activity of guaiacol (Table 1). Most activity was observed in 150 mm treatment (0.169 µmol/mg), and lowest was observed in 50 mm treatment (0.129 µmol/mg) (Table 2).

Fertilizer treatments, and interaction between experimental treatments were not significant on antioxidant activity of catalase (Table 1).

Experimental treatments had significant effects on ascorbate peroxidase activity.

Most, and lowest activity rate (0.173 µmol/mg) was observed in 100 mm treatment, and lowest activity was observed in 50 mm treatment (0.107 µmol/mg), and foliar application of silicon (Figure 3).

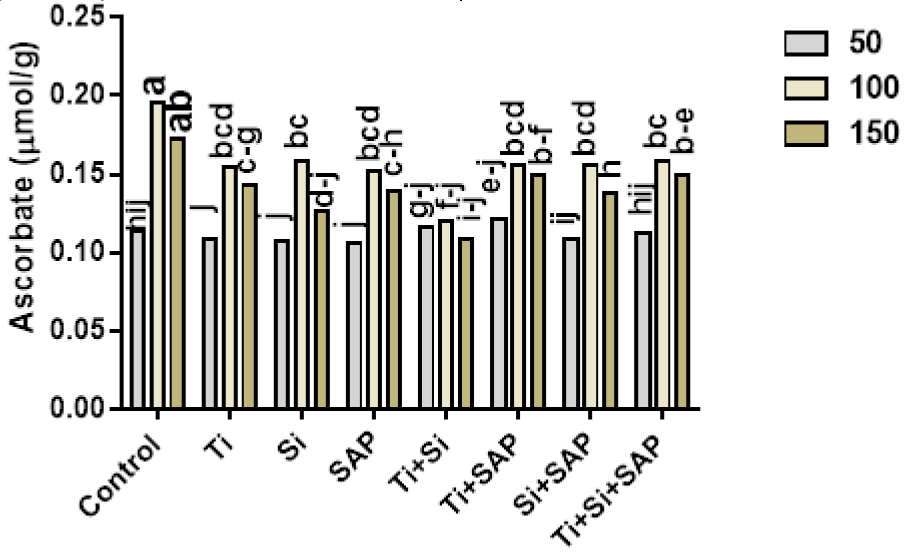

Figure 3. Interaction between water availability, and fertilizer treatments on ascorbate activity in cumin. Ti=Titanium, Si= Silicon, SAP= Superabsorbent polymer

Drought stress increases production of free radicals, and decreases inner cells carbon dioxide concentration that results in increased lipid peroxidation.

These happens increase production of antioxidant enzymes for fighting against free radicals [SHARMA and DUBEY, 2005].

Plants use antioxidant system composed of antioxidant enzymes for eliminating free radicals [BLOKHIN *et al.*, 2003].

The results showed that titanium decreased antioxidant activity of the enzymes. It means that theses did not act as antioxidants.

In contrast to our findings, the sensitivity of amino acids of a peptide is different to an oxidative attack and there are various forms of activated oxygen in terms of reactiveness potential.

Conclusions

In sum, drought stress increased carbohydrate, antioxidant enzymes activity and proline contents, and decreased relative water contents in *Cuminum cyminum*.

The results also showed that most and lowest proline content were observed in foliar application of titanium, and silicon treatments, respectively.

Acknowledgments

The authors thank the Islamic Azad University, Bam Branch (Iran) for financial support. The experiment was carried out as a research project, Contract 2019–B–5575.

References

1. Abdul Qados, M.M.S. Mechanism of Nanosilicon–Mediated Alleviation of Salinity Stress in Faba Bean (*Vicia faba* L.) Plants. *American Journal of Experimental Agriculture*, **2015**, 7(2), 78–95.
2. Ashraf, M.; Foolad, M.R. Roles of glycine betaine and proline in improving plant abiotic stress resistance.

Environmental and Experimental Botany, **2007**, 59, 206–216.
3. Azimi, R.; Borzelabad, M.J.; Feizi, H.; Azimi, A. Interaction of SiO_2 nanoparticles with seed prechilling on germination and early seedling growth of tall wheatgrass (*Agropyron elongatum* L.). *Polish Journal of Chemistry Technology*, **2014**, 16(3), 25–29.
4. Bates, L.S.; Waldren, S.P.; Teare, I.D. Rapid determination of free proline for water– stress studies. *Plant Soil*, **1973**, 39, 205–207.
5. Beers, R.F.; Sizer, I.W. A spectrophotometric method for measuring the breakdown of hydrogen peroxide by catalase. *Journal of Biological Chemistry,* **1952**, 195, 133–140.
6. Blokhin, O.; Virolainen, E.; Fagerstedt, K. Antioxidant oxidative damage and oxygen deprivation stress. *Annals Review Botany*, **2003**, 91, 179–194.
7. Bowen, P.; Menzies, J.; Ehret, D.; Samuel, L.; Glass, A.D.M. Soluble silicon sprays inhibit powdery development in grape leaves. *Journal of American Society Horticultural Science*, **1992**, 117, 906–912.
8. Chaves, M.M.; Maroco, J.P.; Pereira, J.S. Understanding plant responses to drought–from genes to the whole plant. *Functional Plant Biology*, **2003**, 30, 239–264.
9. Chun, S.C.; Paramasivan, M.; Chandrasekaran, M. Proline accumulation influenced by osmotic stress in carbuncular mycorrhizal symbiotic plants. *Frontiers in Microbiology*, **2018**, 5, 1–13.
10. Clavel, D.; Drame, N.K.; Roy–Macauley, H. Analysis of early responses to drought associated with field drought adaptation in four Sahelian groundnut (*Arachis hypogaea* L.) cultivars. *Environmental and Experimental Botany*, **2005**, 54, 219–230.
11. Flagella, Z.; Pastore, D.; Campanile, R.G.; Fonzo, N.D. The quantum yield or photosynthetic Application to precision agriculture and crop physiology. *The Journal of Agriculture Science*, **1995**, 170, 224–233.
12. Gill, S.S.; Tuteja, N. Reactive oxygen species and antioxidant machinery in abiotic stress tolerance in crop plants. *Plant Physiology Biocenology*, **2010**, 48, 909–930.
13. Keles, Y.; Oncel, I. Growth and solute composition on two wheat species experiencing. *Crop Science*, **2004**, 40, 470–475.
14. Liang, X.; Zhang, L.; Kumar Natarajan, S.; Becker, D.F. Proline mechanisms of stress survival. *Antioxidants & Redox Signaling*, **2013**, 19, 998–1011.
15. Lin, D.; Xing, B. Phytotoxicity of nanoparticles, inhibition of seed germination and root growth. *Environmental Pollution*. **2007**, 150(2), 243–50.
16. Lugojan, C.; Ciulca, S. Evaluation of relative water content in winter wheat. *Journal of Horticultural Forestry Biotechnology*, **2011**, 15, 173–177.
17. Jaberzadeh, A.; Moaveni, P.; Reza, H.; Tohidi, M.; Zahedi, H. Influence of bulk and nanoparticles titanium foliar application on some agronomic traits, seed gluten and starch contents of wheat subjected to water deficit stress. *Notulae Botanicae Horti Agrobotanici*, **2013**, 41(1), 201–207.
18. Kalteh, M.; Alipour, Z.T.; Ashraf, S.; Aliabadi, M.M.; Nosratabadi, A.F. Effect of silica nanoparticles on basil (*Ocimum basilicum*) under salinity stress. *Journal of Chemistry Health Risks*, **2014**, 4(3), 49–55.
19. Khodaie, L.; Sadeghpoor, O. Ginger from ancient times to the new outlook. *Jundishapur Journal of Natural Pharmaceutical Products*, **2015**, 10(1), 18402.
20. Mahmoodzadeh, H.; Nabavi, M.; Kashefi, H. Effect of nanoscale titanium dioxide particles on the germination and growth of canola (*Brassica napus*). *Journal of Ornamental and Horticultural Plants*, **2013**, 3(1): 25–32.
21. Mingyu, S.; Jie, L.; Sitao, Y.; Linglan, M.; Fashui, H. Effects of Nanoanatase on the Photosynthetic Improvement of Chloroplast Damaged by Linolenic Acid. *Biological Trace Element Research*, **2008**, 124, 173.
22. Nakano, Y.; Asada, K. Hydrogen peroxide is scavenged by ascorbate–specific peroxidase in spinach chloroplasts. *Plant Cell Physiology*, **1981**, 22, 867–880.
23. Rad, J.S.; Karimi, J.; Mohsenzadeh, S.; Rad, M.S.; Moradgholi, J. Evaluating SiO_2 nanoparticles effects on developmental characteristic and photosynthetic pigment contents of

Zea mays L. *Bulltin Environment, Pharmacology and Life Science*, **2014**, 3(6), 194–201.
24.Rana, A.; Kumar, V.; Mirza, A.; Panghal, A. Efficacy of cumin (*Cuminum cyminum* L.) as a bionutrient and its management. *Annals Biology*, **2018**, 34(2), 218–222.
25.Rastogi, A.; Zivcak, M.; Sytar, O.; Kalaji, H.M.; He, X.; Mbarki, S.; Brestic, M. Impact of Metal and Metal Oxide Nanoparticles on Plant: A Critical Review. *Frontiers Chemistry*, **2017**, 5, 78.
26.Rebey, I.B.; Jabri–Karoui, I.; Hamrouni–Sellami, I.; Bourgou, S.; Limam, F.; Marzouk, B. Effect of drought on the biochemical composition and antioxidant activities of cumin (*Cuminum cyminum* L.) seeds. *Indian Crop Production*, **2012**, 36(1), 238–45.
27.Reddy, A.R.; Chaitanya, K.V.; Vivekanandan, M. Drought–induced responses of photosynthesis and antioxidant metabolism in higher plants. *Journal of Plant Physiology*, **2004**, 16, 1189–1202.
28.Sairam, R.K.; Rao, K.V.; Srivastava, G.C. Differential response of wheat genotypes to long term salinity stress in relation to oxidative stress, antioxidant activity and osmolyte concentration. *Plant Science*, **2002**, 163(5), 1037–1046.
29.Shallan, M.A.; Hassan, H.M.M.; Namich, A.A.M.; Ibrahim, A.A. Biochemical and physiological effects of tio2 and sio2 nanoparticles on cotton plant under drought stress. *Research Journal of Pharmaceutical, Biological and Chemical Science*, **2016**, 7(4), 4–13.
30.Sharma, P.; Dubey, R.S. Drought induces oxidative stress and enhances the activities of antioxidant enzymes in growing rice seedlings. *Plant Growth Regulation*, **2005**, 46, 209–221.
31.Siddiqui, M.H.; Al–Whaibi, M.H. Role of nano–SiO2 in germination of tomato (*Lycopersicum esculentum* seeds Mill.). *Saudi Journal of Biological Science*, **2014**, 21, 13–17.
32.Takesh, A.; Fatahinia, M.; Zarei Mahmoudabadi, A. *In vitro* evaluation of antifungal activity of three traditionally used medicinal plants; *Umbilicus Intermedius Boiss*, *Cuminum Cyminum* and *Zingiber Officinale* extracts. *International Journal of Medicine Laboratory*, **2019**, 6(4), 266–274.
33.Wang, X.; Wei, Z.; Liu, D.; Zhao, G. Effects of NaCl and silicon on activities of antioxidative enzymes in roots, shoots and leaves of alfalfa. *African Journal of Biotechnology*, **2011**, 10, 545–549.
34.Yin, C.; Peng, Y.; Zang, R.; Zhua, Y.; Li, C. Adaptive responses of *Populus kangdingensis* to drought stress. *Physiology Plant*, **2005**, 123, 445–451.

Received: December 17, 2019
Article in Press: April 30, 2020
Accepted: Last modified on: May 20, 2020