Annals of Agri-Bio Research **25** (1) : 103-109, 2020

Stomatal Behaviour of Soybean under Drought Stress with Silicon Application

YONNY KOENTJORO*, SUKENDAH[1], EDI PURWANTO[2] AND DJOKO PURNOMO[2]

Department of Agricultural Science, Graduated School, Sebelas Maret University, Surakarta, 57126, Indonesia
**(e-mail : yonny_k@upnjatim.ac.id; Mobile : +62817586978)*

(Received : June 25, 2019; Accepted : August 20, 2019)

ABSTRACT

The limited available water is becoming the main phenomenon of drought stress, so the water becomes a very important factor in the agricultural business. The drought-tolerant plant is very necessary for this situation. Plants have different adaptability to environmental conditions that occur, including the mechanism of opening and closing the stomata. One effort that can be done to reduce excessive water loss in conditions of limited water content is engineering physiology which can be done through the arrangement of the stomata opening and closing system, by giving silicon as an element that has been known to increase plant resistance to abiotic stress. This research evaluated the behaviour of stomata in soy under drought stress with silicon application. The results showed that the treatment of silicon was closely related to the size of the opening of the stomata, the increasing concentration of silicon, the wider stomata opening width (R^2 = 0.52 at 9 a. m., R^2 = 0.92 at 11 a. m., and R^2= 0.98 at 01.00 p. m.). In terms of concentrations of 15 ml/l was able to produce the widest opening stomata compared to concentrations of 0.5 and 10 ml/ l. Silicon also affected the density of stomata, the percentage of open and closed stomata. Applying the higher silicon increased the density of stomata. However, silicon had a varied influence against the percentage of stomata abaxial and adaxial.

Key words : Drought stress, stomata, silicon, soybean

INTRODUCTION

Drought stress in plants occurs because water is available in limited quantities. This drought occurs due to a decrease in plant water content from the field capacity to a permanent wilting point. During drought (wilting plants), the water vapour pressure in the air is lower than in leaf tissue so transpiration increases to the limit of the rate of transmission of ground water to the root, unable to compensate for the rate of transpiration (Radwan *et al.,* 2017). Water is the main limiting factor for plant growth and production (Chen *et al.,* 2018). Plants have the ability to adapt to drought conditions that are different in terms of morphology, physiology and biochemical and molecular processes (Haisel *et al.,* 2006; Fariduddin *et al.,* 2009). Drought for plants will inhibit evapotranspiration, interfere with growth by influencing several physiological and biochemical processes such as leaf respiration, leaf chlorophyll content, gas exchange, leaf water content, and relative plant growth rates (Okunlola *et al.,* 2017), such conditions may result in a significant production decrease (Lei and Yin, 2006; Farooq *et al.,* 2012). Under drought stress, photosynthesis rates, stomatal conductance and transpiration rate of soybean plants decrease significantly, while the CO_2 concentration between cells changes only slightly at the start of drought stress (Zhang *et al.,* 2016). One mechanism of plant adaptation to drought stress conditions is the regulation of stomatal guard cell control system (Sterling, 2004).

Stoma (plural : stomata) is a gap that is flanked by two epidermal cells which are limited by special cells called guard cells. Stomata have an important role as an exit door for water vapour, oxygen, CO_2, pollutant gases, or certain compounds (Dias *et al.,* 2014; Engineer *et al.,* 2016). When plants experience drought stress, one of the initial symptoms is losing water control through the stomata. The level of intensity of high drought and increasing duration causes plants to experience drought, and results in accumulation of ABA. Potassium

[1]Department of Agrotechnology, Faculty of Agriculture, University of Pembangunan National "Veteran" Jawa Timur, Surabaya, 60294, Indonesia.
[2]Department of Agrotechnology Science, Faculty of Agriculture, Sebelas Maret University, Surakarta, 57126, Indonesia.

flows into the body of the plant is disrupted because ABA blocks the proton pump, which flows K+ into the guard cell. This condition affects the regulation system of opening the stomata because the opening and closing activities of the stomata are closely related to the osmotic humidity in the leaves (Shinohara and Leskovar, 2014).

Drought conditions in soybean will inhibit the process of opening the stomata (Liu *et al.,* 2014; Sreenivasulu *et al.,* 2014). Drought stress also limits symbiosis of nitrogen fixation (Quintana *et al.,* 2013). In order for soybean to be cultivated on dry land to remain capable of high production, physiological engineering and soil manipulation need to be done, namely, by regulating stomata work. Plants physiologically respond to light water stress with stomata closure, which reduces stomatal conductance during short response times (Spinelli *et al.,* 2016).

Based on the results of several studies it is known that silicon can increase plant resistance to drought stress (Ahmad *et al.,* 1992; Arsenault-Labrecque *et al.,* 2012). In plants, silicon is thought to be involved in fortification which affects the oxidation of cell membranes and causes protection for various functions of plant organs that experience drought. Silicon also seems to be part of the regulation of osmolites in cells under conditions of drought stress (Janislampi, 2012). Silicon is known to be able to increase plant biomass and the concentration of absorbed silicon (Ahmad *et al.,* 2007), reduce the stress of Zn and Mn and streamline the translocation system of the silicon itself (Miyake and Takahashi, 2012; Mansano Sarto *et al.,* 2014). Silicon can reduce the influence of various abiotic stresses including salt stress, metal poisoning, dryness, radiation damage, nutritional imbalances, high temperatures and freezing (Ma, 2004), other researchers also reported that silicon was able to reduce water stress, salt stress, iron poisoning (Gonzalo *et al.,* 2013). Transpiration of the leaves occurs mainly through the stomata and partly through the cuticle. This study aimed at evaluating the behaviour of soybean stomata in drought conditions with silicon applications.

MATERIALS AND METHODS

This research was carried out in the Greenhouse. The planting material used was soybean cultivar Grobogan. The soybean plants were grown in pots media with drought stress conditions simulated by the application of Poly Ethylene Glycol (PEG 600) with a concentration of 15% which was equivalent to -0.67 Mpa. Applications of silicon at various concentrations of 0 ml/l (control), 5, 10 and 15 ml/l are given after the plants are 14 days after planting and then every seven days. This study aimed at evaluating the stomatal response to drought conditions using silicon applications. Observations on stomata behaviour in plants aged 50 days after planting with observation hours at 9 a. m., 11 a. m. and 1 p. m.

Analysis of stomata used the intact preparations (whole mount) based on the method of Johansen (1940) that have been modified. Soybean leaves that have been opened perfectly washed with distilled water and immediately sliced across using a sharp knife. The leaves were paradermally sliced on the upper (adaxial) and lower (abaxial) surfaces using a razor blade to form a transparent thin layer. The leaves of the paradermal incision were dipped in a 2% sodium hypochlorite bleach solution for 60 min with the aim of bleaching. Furthermore, the paradermal incision was rinsed with distilled water and stained with 2% safranin for 2 min. The incision was washed again with distilled water and then placed on top of the glass object and dripped with 30% glycerine and then covered with a lid around the edge of the cover glass glued with nail polish. Furthermore, the number of stomata was observed and calculated under a microscope with a magnification of 40 x 10. Stomata were calculated by considering the number of stomata that were closed and open in the abaxial and adaxial part.

The percentage of stomata :

$$\frac{\text{The number of stomata}}{\text{The overall number of stomata}} \times 100\%$$

RESULTS AND DISCUSSION

Correlation analysis between the concentration of silicon and the size of the stomata openings showed that the concentration of silicon was closely related positively to the measurement at 9.00 (r : 0.72), while the measurements at 11 a. m. and 1 p. m. were closely related (r :

0.99 and 0.96, respectively). This meant that the higher the concentration of silicon, the greater the opening of the stomata, then through the regression test showed that the concentration of silicon determined the size of the stomata both at 9 a. m. and 11 a. m. and 1 p. m. (R^2 each 0.52, 0.92 and 0.98; Fig. 1).

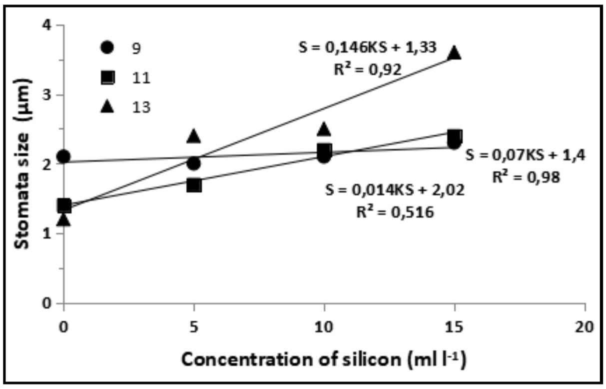

Fig. 1. Regression between stomatal size (Abaxial) and concentration of silicon at 9 a. m., 11 a. m. and 1 p. m.

The increase in the concentration of increased silicon concentration indicated that silicon was involved in the regulation of osmolites in leaf cells (Janislampi, 2012), including the process of opening and closing stomata, increasing stomatal conductance, photochemical effectiveness and gas exchange (Habibi and Hajiboland, 2013). Furthermore, it was reported that silicon had an important role in improving plant defense systems against abiotic stress (Adrees *et al.,* 2015; Rizwan *et al.,* 2015), and the positive role of silicon was confirmed to increase plant resistance to biotic stress (Fauteux *et al.,* 2006; Fallah, 2012).

Every increase in the concentration of silicon affected the opening of the stomata, both in the abaxial or adaxial stomata. Each silicon concentration treatment showed an opening size that increased at each observation time. The highest opening size was shown under concentrations of 15 ml/Si (3.6 µm in abaxial stomata, and 3.8 µm in adaxial stomata) at 1 a. m. (Figs. 2 and 3). The involvement of silicon on the mechanism of stomatal movement was thought to have an indirect effect. Etesami and Jeong (2018) explained that the addition of silicons to soybean cultivation increased levels of endogenous GA, reduced levels of abscisic acid (ABA) and proline, which ABA and proline are compounds that play a role in stomata hole movement. In drought stress conditions, ABA (Chen *et al.,* 2018) and proline (Cha-Um and Kirdmanee, 2009) will increase.

Soybean under drought stress without being given silicon (control) showed the size of the stomata hole was reduced (at 11 a. m. and 1 p. m.). The increasing concentration of silicon caused the size of abaxial stomata (Fig. 2) and adaxial (Fig. 3) to be wider. The concentration of 15 ml/l resulted in the widest size of the abaxial stomata, both at 1100 (2.4 µm) and at 1300 (3.6 µm). While the size of the widest adaxial stomatal size at the silicon concentration of 15 ml/l at 11 a. m. and 1 p. m. (2.5 and 3.8 µm). Xia (1994) suggested that the drought that occurred in all reproductive phases caused a decrease in photosynthetic rate, chlorophyll content, leaf area, stomatal opening width, biomass and seed yield, but the stomata frequency and respiration rate increased (Zhang *et al.,* 2015). The mechanism of the reaction caused by plants to defend themselves against drought and water loss could be minimized, manifested in the form of

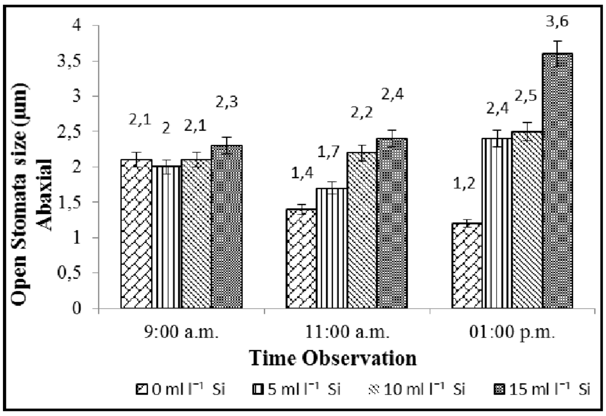

Fig. 2. The size of the stomata open (Adaxial) resulting from the treatment of silicon at various times of observations.

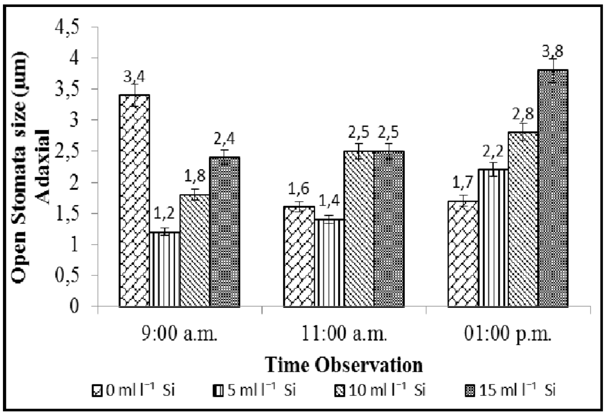

Fig. 3. Measurement of stomata open (Adaxial) as a result of silicon treatment at various times of observation.

the mechanism of opening and closing the stomata, the cuticle layer, the presence of fine hair on the leaf surface, etc. all acted as inhibiting factors for the rate of water loss from Davies *et al.* (1981).

Figs. 4 and 5 explain that at each observation hour, it appeared that the stomata opened wider as the concentration of silicon was given. Gas exchange activities were closely related to the size of the stomata and stomata density, generally smaller stomata sizes were associated with high density (Franks and Beerling, 2009; Giday *et al.,* 2013). Abaxial and adaxial stomata tended to respond differently to water stress and stomatal sensitivity differed depending on plant species (Hsiao, 1973). Stomata closure was believed to reduce water loss through transpiration (Torres-Ruiz *et al.,* 2013; Nemeskeri *et al.,* 2015; Clauw *et al.,* 2015).

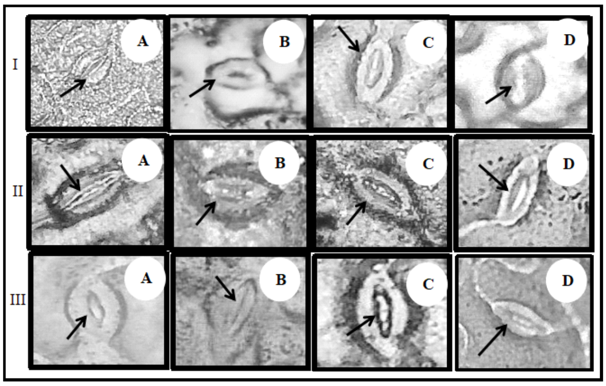

Fig. 4. The effect of silicon applications against the size of the stomatal opening width (Abaxial) at 9 a. m. observations (I), 11 a. m. (II) and (III) 1 p. m. (A : 0 ml/l Si, B : 5 ml/l Si, C : 10 ml/l Si and D : 15 ml/l Si).

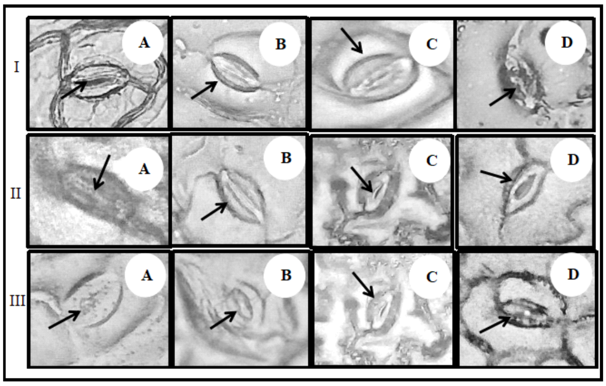

Fig. 5. The effect of silicon applications against the size of the stomatal opening width (Adaxial) at 9 a. m. observations (I), 11 a. m. (II) and (III) 1 p. m. (A : 0 ml/l Si, B : 5 ml/l Si, C : 10 ml/l Si and D : 15 ml/l Si).

The percentage of abaxial stomata that were open at each silicon concentration showed an increase along with the addition of silicon concentration at 9 a. m., 11 a. m. and 1 p. m. the same pattern was observed, which was a decrease in the percentage of the number of open abaxial stomata, but at a concentration of 15 ml/l the increase was relatively insignificant. At 0900 the concentration of 15 ml/l silicon produced the highest percentage of abaxial stomata, which were 34.26 and 21.62%, respectively, but at 1 p. m. the highest percentage of open abaxial stomata was produced by control without silicon (29.03%; Fig. 6). The highest percentage of closed abaxial stomata was generated by treatment without silicon (control) at 9 a. m. (78.03%) and concentration of 10 ml/l at 11 a. m. (83.67%), while at 1 p. m. the highest percentage of closed stomata was produced by silicon concentration of 10 ml/l (88.04%; Fig. 7). The results of Zhao *et al.* (2015) reported that drought stress conditions caused an increase in stomatal density and decreased stomatal size. There was a close relationship between environmental

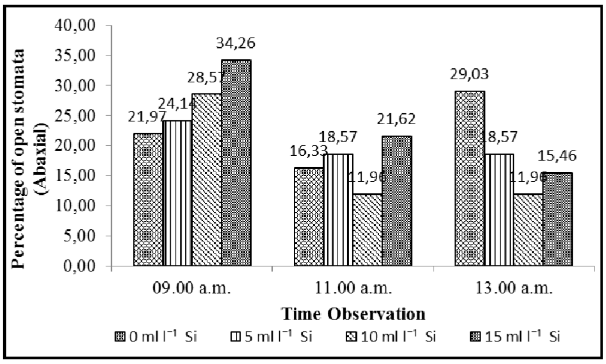

Fig. 6. The percentage of open stomata (abaxial) at 9 a. m., 11 a. m. and 1 p. m. by silicon application.

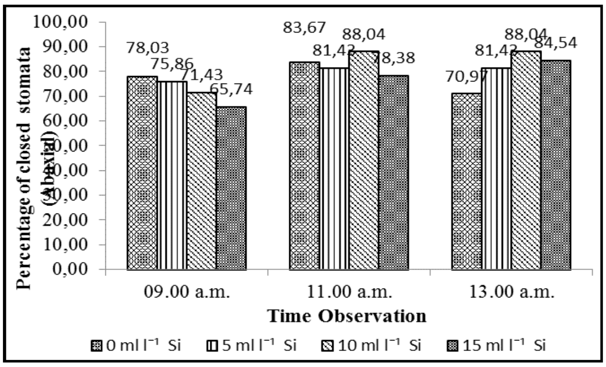

Fig. 7. The percentage of closed stomata (abaxial) at 9 a. m., 11 a. m. and 1 p. m. by silicon application.

conditions with stomata size and density and responses that varied from plant type.

The percentage of open adaxial stomata showed differences in each silicon concentration test. The concentration of 0 ml/l showed the highest percentage of adaxial stomata which was open at 1 p. m. (27.27%), but different patterns were shown at 9 a. m. (highest percentage 26.47% at concentrations of 5 ml/l) and 11 a. m. (highest percentage 26.47% at the concentration of silicon 15 ml/l; Fig. 8). The percentage of closed adaxial stomata showed different results in each silicon treatment. The concentration of 15 ml/l, in general, produced the highest percentage of closed adaxia stomata 78.26% (at 9 a. m.) and 87.10% (at 1 p. m.), while at 11 a. m. the treatment without silicon (control) resulted in the highest percentage of closed stomata (89.13%; Fig. 9).

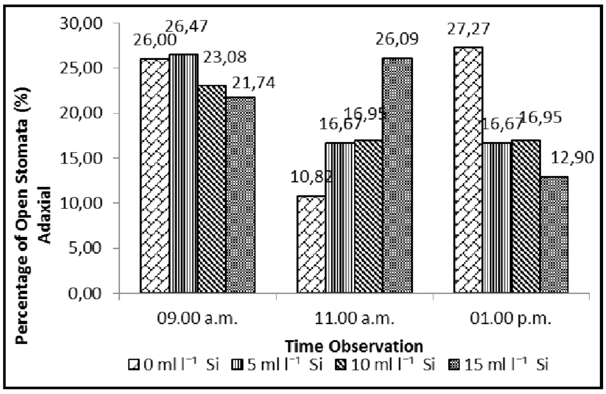

Fig. 8. The percentage of open stomata (adaxial) at 9 a. m. 11 a. m. and 1 p. m. by silicon application.

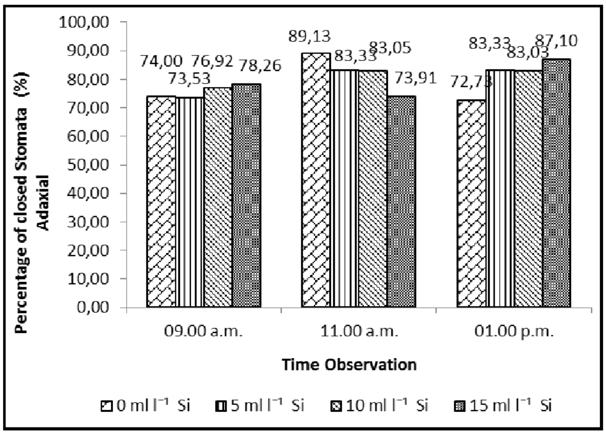

Fig. 9. The percentage of closed stomata (adaxial) at 9 a. m., 11 a. m. and 1 p. m. by silicon application.

CONCLUSION

The application of silicon to various variables showed that the increase in silicon concentration was closely related to stomata behaviour. The mechanism of opening of abaxial and adaxial stomata, in this case the size of the stomata opening increased with the concentration of silicon applied. Increased stomata density also occurred with given increasing silicon concentration. Silicons had a varied influence on the percentage of abaxial and adaxial stomata both open and closed. In general, the treatment of the concentration of silicon 15 ml/l had a large effect on almost all observed variables.

REFERENCES

Adrees, M., Ali, S., Rizwan, M., Zia-Ur-Rehman, M., Ibrahim, M., Abbas, F., Farid, M., Qayyum, M. K. and Irshad, M. K. (2015). Mechanisms of silicon-mediated alleviation of heavy metal toxicity in plants : A review. *Ecotoxicol. Environ. Saf.* **119** : 186-197.

Ahmad, F., Rahmatullah, T., Aziz, M. A., Maqsood, A., Mukkram, A., Tahir and Kanwal, S. (2007). Effect of silicon application on wheat (*Triticum aestivum* L.) growth under water deficiency stress. *J. Food Agric.* **19** : 01-07.

Ahmad, R. S., Zaheer and Ismail, S. (1992). Role of silicon in salt tolerance of wheat (*Triticum aestivum* L.). *Plant Sci.* **85** : 43-50.

Arsenault-Labrecque, G., Menzies, J. G. and Bélanger, R. R. (2012). Effect of silicon uptake on soybean resistance to *Phakopsora pachyrhizi* in different cultivars. *Plant Dis.* **96** : 37-42.

Cha-Um, S. and Kirdmanee, C. (2009). Proline accumulation, photosynthetic abilities and growth characters of sugarcane (*Saccharum officinarum* L.) plantlets in response to iso-osmotic salt and water-deficit stress. *Agric. Sci. in China* **8** : 51-58.

Chen, Z., Wang, Z., Li, M., Yang, Y. and Xu, B. (2018). Abscisic acid and brassinolide combined application synergistically enhances drought tolerance and photosynthesis of tall fescue under water stress. *Scientia Horticulturae* **228** : 1-9.

Clauw, P., Coppens, F., De Beuf, K., Dhondt, S., Van Daele, T., Maleux, K. and Inze, D. (2015). Leaf responses to mild drought stress in natural variants of *Arabidopsis thaliana. Plant Physiol.* **10** : 110-114.

Davies, M. T., Wilson, J. A., Sharp, R. E. and Osonubi, O. (1981). Control of stomatal behaviour in water stressed plants. *Biol. Seminar Series* **8** : 163-181.

Dias Gabrielen, M. G., Soares, J. D. R., Pasqual, M., Renata Alves, R., Silva, L., Rodrigues L. C. A., Pereira, F. J. and Castro, E. M. (2014). Photosynthesis and leaf anatomy of Anthurium cv. Rubi plantlets cultured *in vitro* under different silicon (Si) concentrations. *Aust. J. Crop Sci. (AJCS)* **8** : 1160-1167.

Engineer, C. B., Hashimoto-Sugimoto, M., Negi, J., Israelsson-Nordström, M., Azoulay-Shemer, T., Jan Rappel, W., Iba, K. and Schroeder, J. I. (2016). CO_2 sensing and CO_2 regulation of stomatal conductance : Advances and open questions. *Trends in Plant Science* **21** : 16-30.

Etesami, H. and Jeong, B. R. (2018). Silicon (Si) : Review and future prospects on the action mechanisms in alleviating biotic and abiotic stresses in plants. *Ecotoxicol. and Environ. Safety* **147** : 881-896.

Fallah, A. (2012). Silicon effect on lodging parameters of rice plants under hydroponic culture. *Int. J. Agric. Sci.* **2** : 630-634.

Fariduddin, Q. S., Khanam, S. A., Hasan, B., Ali, S., Hayat, A. and Ahmad (2009). Effect of 28 homobrassinolide on the drought stress-induced changes in photosynthesis and antioxidant system of *Brassica juncea* L. *Acta Physiol. Plant.* **31** : 889-897.

Farooq, M., Hussain, M., Wahid, A. and Siddique, K. H. M. (2012). Drought stress in plants : An overview. In : *Plant Responses–From Morphological to Molecular Features,* Aroca, R. (ed.). Springer, Heidelberg, New York, London. pp. 1-36.

Fauteux, F., Chain, F., Belzile, F., Menzies, J. G. and Bélanger, R. R. (2006). The protective role of silicon in the Arabidopsis powdery mildew pathosystem. *Proc. Natl. Acad. Sci., U. S. A.* **103** : 17554-17559.

Franks, P. J. and Beerling, D. J. (2009). Maximum leaf conductance driven by CO effects on stomatal size and density over geologic time. *Proc. Natl. Acad. Sci., USA* **106** : 10343-10347.

Giday, H., Kjaer, K. H., Fanourakis, D. and Ottosen, C. O. (2013). Smaller stomata require less severe leaf drying to close : A case study in Rosa hydrid. *J. Plant Physiol.* **170** : 1309-1316.

Gonzalo, M. J., Juan-Lucena, J. and Apaolaza, L. H. (2013). Effect of silicon addition on soybean (*Glycine max*) and cucumber (*Cucumis sativus*) plants grown under iron deficiency. *Plant Physiol. and Biochemi.* **70** : 455-461.

Habibi, G. and Hajiboland, R. (2013). Alleviation of drought stress by silicon supplementation in pistachio (*Pistacia vera* L.) plants. *Folia Hort.* **25** : 21-29.

Haisel, D., Pospíšilová, J., Synková, H., Schnablová, R. and Batková, P. (2006). Effects of abscisic acid or benzyladenine on pigment contents, chlorophyll fluorescence and chloroplast ultrastructure during water stress and after rehydration. *Photosynthetica* **44** : 606-614.

Hsiao, T. C. (1973). Plant responses to water stress. *Annu. Rev. Plant Physiol.* **24** : 519-570.

Janislampi, K. W. (2012). Effect of silicon on plant growth and drought stress tolerance. Thesis, Utah State Universtiy Logan, Utah.

Johansen, D. A. (1940). *Plant Microtechnique.* New York : Mc-Graw-Hillbook Company, Inc. M.

Lei, Y. C. and Yin, L. I. (2006). Differences in some morphological, physiological and biochemical responses to drought stress in two contrasting populations of *Populus przewalskii. Physiol. Plant.* **127** : 182-191.

Liu, C. L., Wang, M., Ma, X. Y. and Zhang, W. (2014). NRGA1, a putative mitochondrial pyruvate carrier, mediates ABA regulation of guard cell ion channels and drought stress responses in Arabidopsis. *Molecular Plant* **7** : 1508-1521.

Ma, J. F. (2004). Role of silicon in enhancing the resistance of plants to biotic and abiotic stresses. *Soil Sci. Plant Nutr.* **50** : 11-18.

Mansano Sarto, M. V., Lana, M. C., Rampim, L., Rosset, J. S., Wobeto, J. R., Ecco, M., Bassegio, D. and Da Costa, P. F. (2014). Effect of silicate on a nutrition and yield of wheat. *African J. Agric. Res.* **9** : 956-962.

Miyake, Y. and Takahashi, E. (2012). Effect of silicon on the growth of soybean plants in a solution culture. *Soil Sci. Plant Nutr.* **31** : 625-636.

Nemeskeri, E., Molnar, K., Vigh, R., Nagy, J. and Dobos, A. (2015). Relationships between stomatal behaviour, spectral traits and water use and productivity of green peas (*Pisum sativum* L.) in dry seasons. *Acta Physiol. Plant* **37** : 1-16.

Okunlola, G. O., Olatunji, O. A., Akinwale, R. O., Tariq, A. and Adelusi, A. A. (2017). Physiological response of the three most cultivated pepper species (Capsicum spp.) in Africa to drought stress imposed at three stages of growth and development. *Scientia Horticulturae* **224** : 198-205.

Quintana, E. G., Larrainzar, E., Seminario, A., Leal, J. L. D., Alamillo, J. M., Pineda, M., Igor, C. A., Wienkoop, S. and González, E. M. (2013). Local inhibition of nitrogen fixation and nodule metabolism in drought-stressed soybean. *J. Exptl. Bot.* **64** : 2171-2182.

Radwan, A., Kleinwachter, M. and Selmar, D. (2017). Impact of drought stress on specialised metabolism : Biosynthesis and the expression of monoterpene synthases in sage (*Salvia officinalis*). *Phytochemistry* **141** : 20-26.

Rizwan, M., Ali, S., Ibrahim, M., Faridm, M., Adreesm, M., Bharwana, S. A., Rehman, M. Z., Qayyum, M. F. and Abbas, F. (2015). Mechanisms of silicon-mediated alleviation of drought and salt stress in plants : A review. *Environ. Sci. Pollut. Res.* **22** : 15416-15431.

Shinohara, T. and Leskovar, D. I. (2014). Effects of ABA, antitranspirants, heat and drought stress on plant growth, physiology and water status of artichoke transplants. *Scientia Horticulturae* **165** : 225-234.

Spinelli, G. M., Richard, L., Snyder, Blake, L., Sanden, A. Ken and Shackel (2016). Water stress causes stomatal closure but does not reduce canopy evapotranspiration in almond. *Agric. Water Manage.* **168** : 11-22.

Sreenivasulu, N., Harshavardhan, V. T., Govind, G., Seiler, C. and Kohli, A. (2014). Contrapuntal role of ABA : Does it mediate stress tolerance or plant growth retardation under long-term drought stress. *J. Gene.* **506** : 265-273.

Sterling, T. M. (2004). *Transpiration–Water Movement through Plants.* Department of Entomology, Plant Pathology and Weed Science, New Mexico State University.

Torres-Ruiz, J. M., Diaz-Espejo, A., Morales-Sillero, A., Martín-Palomo, M. J., Mayr, S., Beikircher, B. and Fernandez, J. E. (2013). Shoot hydraulic characteristics, plant water status and stomatal response in olive trees under different soil water conditions. *Plant Soil* **373** : 75-85.

Xia, M. Z. (1994). Effect of soil drought during the generative development phase of faba bean (*Vicia faba*) on photosynthetic characters and biomass production. *J. Agric. Sci.* **122** : 67-72.

Zhang, J., Liu, J., Yang, C., Du, S. and Yang, W. (2016). Photosynthetic performance of soybean plants to water deficit under high and low light intensity. *South African J. Bot.* **105** : 279-287.

Zhang, Q., Liu, J., Lu, H., Zhao, S., Wang, W., Du, J. and Yan, C. (2015). Effects of silicon on growth, root anatomy, radial oxygen loss (ROL) and Fe/Mn plaque of *Aegiceras corniculatum* (L.) Blanco seedlings exposed to cadmium. *J. Environmental Nanotechnology, Monitoring and Management* **4** : 6-11.

Zhao, W., Sun, Y., Kjelgren, R. and Liu, X. (2015). Response of stomatal density and bound gas exchange in leaves of maize to soil water deficit. *Plants, Soils and Climate Faculty Publications.* Utah State University. pp. 1-28.