APPLICATION OF BRYOPHYTE RHIZOID-ASSOCIATED BACTERIA INCREASES SILICON ACCUMULATION AND GROWTH IN MAIZE (*ZEa MAYS* L.) SEEDLINGS

HU, L.^{1,2} – XU, C. C.³ – WANG, J.¹ – CHEN, D. Q.¹ – ZENG, R. S.¹ – SONG, Y. Y.¹ – CHEN, D. M.^{1*}

¹Key Laboratory of Ministry of Education for Genetics, Breeding and Multiple Utilization of Crops, College of Crop Science, Fujian Agriculture and Forestry University, Fuzhou, China

²Key Laboratory of Beibu Gulf Environment Change and Resources Utilization of Ministry of Education, Nanning Normal University, Nanning, China

³College of Life Sciences, Fujian Agriculture and Forestry University, Fuzhou, China

*Corresponding author

e-mail: dongmeifj@163.com; phone: +86-130-1572-5866; fax: +86-591-8378-9483

(Received 19th Jun 2019; accepted 2nd Sep 2019)

Abstract. Silicon (Si) enhances plant resistance to various forms of stress. However, the availability of Si as a plant nutrient is frequently limited. Bryophytes are able to grow on the surface of rocks and buildings with high Si accumulation. Therefore, bryophyte rhizoids might harbor microorganisms capable of solubilizing Si. In this study, silicate solubilizing bacteria (SSB) were isolated from the rhizoids of the bryophyte *Hypnum plumaeforme* L. to examine their effects on Si weathering and growth of maize (*Zea mays* L.). Molecular phylogeny and 16S rRNA gene sequence analysis demonstrated that the dominant bacterial strain B1-5 was a member of the *Kosakonia* genus. The concentrations of soluble Si released from the feldspar and quartz powder in liquid media with strain B1-5 inoculation were higher than those of the control. B1-5 inoculation in pot soil significantly increased the water-extractable Si content in soil, improved Si uptake and accumulation in maize plants, and promoted seedling growth. These results not only prove the existence of SSB in bryophyte rhizoids, but also demonstrate a new approach to searching for effective biological Si fertilizers.

Keywords: silicate solubilizing bacteria, microbial weathering, molecular phylogeny, *Kosakonia*, biological fertilizer

Introduction

Silicon (Si) is the second most abundant element after oxygen in soil. It occurs mostly in the form of dioxide and silicates (Myshlyaeva and Krasnoshchekov, 1974), which cannot be directly used by plants. Only water-soluble monosilicic acid [Si(OH)₄], a product of the weathering of rocks and soil minerals, is available to plants (Epstein, 1994; Raven, 1983). Si has not yet been recognized as an essential nutrient element for plants, but its beneficial effects on resistance have been extensively reported in numerous crops (Epstein, 1999). In 2015 Si was listed as a “beneficial substance” by the International Plant Nutrition Institute, USA (Reynolds et al., 2016). Increasing evidences indicate that Si can alleviate both biotic and abiotic stresses including pathogens, herbivores, extreme temperature, water deficiency, high salinity and nutrient stresses (Debona et al., 2017; Ma, 2004; Wu et al., 2017). Nitrogen (N), phosphorus (P) and potassium (K) fertilizers are widely applied in agricultural production, but Si fertilizer is usually neglected. Si deficiency frequently occurs as a result of Si removal by continuous crop harvest (Liang et al., 2015). Thus, it is of significance to effectively convert unavailable Si in the soils into a form plants can utilize.

Soil microbes play a crucial role in nutrient availability and mineral weathering from primary minerals (Vessey, 2003). Most related studies focused on the P and K solubilizing abilities of some microorganisms as seen in *Table 1*.

Table 1. Studies of phosphate and potassium solubilizing bacteria in crops

Crop name	Solubilizing capacity	Citation
Barely (<i>Hordeum vulgare</i> L.)	P	Mehrvarz and Chaichi, 2008
Wheat (<i>Triticum aestivum</i> L.)	P	Afzal et al., 2005
Wheat (<i>Triticum aestivum</i> L.)	K	Sheng and He, 2006
Pepper (<i>Capsicum annuum</i> L.)	P and K	Han and Lee, 2006
Cucumber (<i>Cucumis sativus</i> L.)	P and K	Han and Lee, 2006
Tobacco (<i>Nicotiana tabacum</i> L.)	K	Zhang and Kong, 2014
Cotton (<i>Gossypium barbadense</i> L.)	K	Sheng, 2005
Rape (<i>Brassica campestris</i> L.)	K	Sheng, 2005
Peanut (<i>Arachis hypogaea</i> L.)	K	Youssef et al., 2010
Sesame (<i>Sesamum indicum</i> L.)	K	Youssef et al., 2010
Ryegrass (<i>Lolium rigidum</i> L.)	K	Xiao et al., 2017
Maize (<i>Zea mays</i> L.)	K	Singh et al., 2010

The role of silicate solubilizing bacteria (SSB), which can solubilize silicon from silicate bearing mineral and rocks (Bosecker, 1997), is largely ignored. Use of highly-efficient SSB is a promising approach for enhancing crop yield and defense in Si deficient soil.

An analysis of the Si contents in 175 plant species collected from a botanical garden shows that Bryophyta (3.46%) contains the highest Si, following by Pteridophyta (1.4%), Gymnospermae (0.5%), Angiospermae (0.13%) (Takahashi and Miyake, 1976a, b, c), suggesting that bryophytes are Si accumulators with high Si content. Therefore, we speculated that bryophyte rhizoids harbor certain SSB capable of solubilizing Si from poor nutrient habitats.

In the present study, our aims were to: (1) isolate and characterize bryophyte rhizoids-associated silicate bacteria, (2) determine the Si-solubilizing ability of selected bacteria, and (3) examine the effect of soil inoculation with selected bacteria on Si availability in the soil, Si uptake and plant growth in maize.

Materials and methods

Plants, soil and mineral

Bryophyte *Hypnum plumaeforme* was collected from rock walls in the back hill on Jinshan Campus of Fujian Agriculture and Forestry University, Fuzhou, China (119°54' E, 26°05' N) in July, 2018. Bryophytes from eight random sampling locations were kept in sterilized plastic Petri-dishes until the isolation experiment began in the same day.

The soils for maize potting were collected from a hillside near the university campus. The soil was a red soil (Typic Hapludults) (Soil Survey Staff, 2010). The properties of the soil were: pH (1:2.5 w/v water) 5.5; organic C 10.03 g·kg⁻¹; total N 1.28 g·kg⁻¹; available N 19.3 mg·kg⁻¹; total P 0.13 g·kg⁻¹; available P 12.6 mg·kg⁻¹; total K 2.05 g·kg⁻¹; available K 154.6 mg·kg⁻¹, water-extractable Si 34.3 mg·kg⁻¹. Seeds of

maize (*Zea mays*, cultivar Yuecai) were obtained from the Crop Research Institute, Guangdong Academy of Agricultural Sciences, Guangzhou, China.

Mineral powders, feldspar (KAlSi_3O_8 , SiO_2 68.09%) and quartz (SiO_2 98.81%), were manufactured by Foshan Hongda Ceramics Co., Ltd and passed through 0.075 mm mesh sieve. The powders were rinsed three times in pure water to eliminate soluble Si.

Isolation of silicate bacteria from bryophyte rhizoids

All collected bryophytes were slightly shaken to remove sands loosely adhering to the rhizoids. The rhizoids with the remaining adhered soil were swayed in 25 mL sterile water with tweezers, and the water was transferred to a 50 mL plastic tubes. The tubes were shaken with a vortex mixer for 15 s. The mixture was kept at room temperature for 30 min. The serially diluted suspension was plated on the surface of Aleksandrov's medium for silicate bacterial culture containing 0.5% sucrose, 0.05% $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, 0.5% FeCl_3 , 0.2% Na_2HPO_4 , 0.01% CaCO_3 , 0.1% KAlSi_3O_8 , and 2% Agar, pH 7.2 (Hu et al., 2018). The Petri-dishes were placed in an incubator at 28 °C for 5 d. The growth of bacteria was checked every day. Fast-growing isolates with different morphology were further purified. The pure colonies were inoculated into liquid medium and preserved in 25% glycerol solution at -80 °C for future use.

Mineral dissolution experiment

The dominant strain was selected to test the ability to solubilize the mineral. The liquid medium consisted of 1% sucrose, 0.05% yeast extract, 0.1% NH_4SO_4 , 0.2% Na_2HPO_4 , 0.05% $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, 0.01% NaCl , 0.01% CaCO_3 , 5 g mineral powder, pH 7.2. Each conical flask (250 mL) contained 100 mL autoclaved liquid medium and 5 mL bacterial inocula (10^6 cfu mL^{-1}) as inoculation treatment. Five mL of sterilized inoculum were also added to the liquid medium to serve as control.

Feldspar and quartz were used as Si mineral respectively in two separate experiments. The inoculation and corresponding control were replicated three times. Each flask was maintained at 180 rpm for 7 d at 28 °C. The fermented broth was centrifuged at 8000 g for 20 min. The solubilized Si concentration in the supernatant was determined using the colorimetric molybdenum blue method described by Pettersson and Karlberg (1999).

Molecular identification

Genomic DNA was extracted using the DNA extraction Kit (Qiagen, USA) according to the manufacture's protocol. The universal primers for 16S rRNA sequence amplification were 27F (5'-GTTTGATCCTGGCTCAG-3') and 1492R (5'-GGTTACCTTGTTACGACTT-3') (Lin et al., 2012). The PCR reaction system had a volume of 25 μL , including Taq PCR Master Mix 12.5 μL , primer 2 μL , ddH₂O 9.5 μL , and DNA template 1 μL . The PCR was performed as follows: hot start at 94 °C for 5 min, followed by 30 cycles of denaturation at 94 °C for 30 s, annealing at 55 °C for 30 s, and extension at 72 °C for 60 s. A final extension step lasted 8 min at 72 °C. The PCR products were purified and sequenced by Shanghai Biosune Biological Technology Co., Ltd. The sequencing results were analyzed using NCBI database to find the closely related homologous sequences.

The sequence and the reference sequences with the highest similarities were aligned using Clustalx 1.83 and phylogenetic analyses was done using MEGA version 4 with

the neighbor-joining method, p-distance and 1000 bootstrap replicates (Tamura et al., 2007; Thompson et al., 1997).

Pot experiment

Maize seeds were surface-sterilized with 10% H₂O₂ for 10 min, and then rinsed 5 times with distilled water. The seeds were soaked in distilled water for 24 h, and then pre-germinated for 48 h at 28 °C. One seedling was transplanted to a 1 L plastic pot with 750 g dry sterilized soil. The soil were autoclaved thrice in three consecutive days. Meanwhile, the cells in cultured bacterial broth were collected by centrifugation at 8000 g for 20 min at 4 °C and washed with sterilized tap water. The pelleted cells were resuspended with sterilized water and then the cells were adjusted to around 10⁶ cfu mL⁻¹ as determined by a standard growth curve based on optical density reading at 600 nm using a spectrophotometer (Ling et al., 2009). The experiment was established with two groups: one group was inoculated with 10 mL bacterial suspension as inoculation treatment, and the other group was added with 10 mL distilled water as control. A total of 20 pots with maize plants were divided into two groups. After 7 d, all plants were fertilized with 50 mL Hoagland nutrient solution (Hoagland and Arnon, 1950).

All plants were harvested 5 weeks after transplanting. The roots were washed and cleaned, and stem diameters were measured. All plants samples were dried at 70 °C for 3 d, then weighed. The Si contents of dry samples were then analyzed by molybdenum blue colorimetric method described by Dannon and Wydra (2004). The water-extractable Si content of air-dry soil samples (1:10 w/v water) was determined by colorimetric molybdenum blue method described by Wang et al. (2004).

Statistical analysis

Si concentrations in culture solution, growth indexes and Si content of plants and soils were analyzed using t-test ($P < 0.05$) to determine difference between bacterial inoculation and control. All analyses were performed with the SPSS 13.0 software package (SPSS Inc.).

Results and discussion

Isolation silicate bacteria from the bryophyte rhizoids

Si-solubilizing bacterial isolates were obtained from the bryophyte *Hypnum plumaeforme* rhizoids on rock walls (Fig. 1A), and one of them showed the dominant colony which was named as B1-5, and kept for further study. On the Aleksandrov plates, the strain B1-5 formed round, smooth, convex, slimy, elastic, translucent and colorless colonies (Fig. 1B). Silicate solubilizing bacteria (SSB) can solubilize silicon from silicate mineral (Bosecker, 1997). Bryophyte with high Si-accumulation mostly can grow on the surface of rocks and buildings. So it raises the possibility that some SSB may adhere to the bryophyte rhizoids.

Molecular identification based on 16S rRNA analysis

About 1500 bp band was obtained from the amplification of 16S rRNA gene sequence from the strain B1-5. The 16S rRNA gene sequence of B1-5 clones was submitted to GenBank with accession number MH051262. Phylogenetic analysis

showed that B1-5 and all 6 *Kosakonia* species formed a branch, and that other outgroup species formed different branches (Fig. 2). Thus, the B1-5 strain belongs to the *Kosakonia* genus. Figure 2 also showed that strain B1-5 shared the highest sequence similarities with *Kosakonia cowanii*. Thus, strain B1-5 was identified as a member of *Kosakonia*.

Several *Kosakonia* species isolated from soils and trees promote plant growth (Brady et al., 2013). Diazotrophic bacteria, *K. pneumoniae* and *K. radicincitans* were found to be associated sugarcane at Northeast Region of Brazil (Antonio et al., 2016). *K. radicincitans* improves fruit yield and quality of *Solanum lycopersicum* (Berger et al., 2017). *K. radicincitans* alters *Arabidopsis thaliana* root and root exudate metabolism (Witzel et al., 2017). *K. pseudosacchari* sp. nov., an endophyte of *Zea mays* was isolated (Kaempfer et al., 2016).

Si solubilizing activity of the bacteria

The ability of strain B1-5 to solubilize Si was assessed in liquid media containing mineral powders of KAlSi_3O_8 and SiO_2 (Fig. 3). After 7 days of incubation, a significant increase in soluble Si concentration of the medium with KAlSi_3O_8 ($t = -5.344$, $df = 4$, $P = 0.006$) or SiO_2 ($t = -43.466$, $df = 4$, $P < 0.001$) supplement was observed in inoculated versus control seedlings.

Figure 1. Habitat of bryophyte *Hypnum plumaeforme* on rock walls (A). Characteristics of strains B1-5 and A2-1 colonies after grown on Aleksandrov medium for 48 h (B)

Figure 2. Phylogenetic dendrogram based on comparative analysis of the 16S rRNA gene sequence, showing the relationship among strain B1-5 and other related species and genera. The GenBank sequence accession numbers were indicated in brackets after the strain names. The dendrogram was generated using the neighbour-joining method in the bootstrap test (1000 replicates)

Figure 3. Si content in KAISiO₈ - (A) and SiO₂ -containing (B) liquid medium with and without inoculation with the isolated silicate bacterium strain B1-5. Si content was determined 7 d after inoculation with strain B1-5. Significant differences ($P < 0.05$) between inoculation and non-inoculated control are indicated by asterisk above bars. Values are mean $\pm$ standard error from three replicates

Here we found the B1-5 bacterial strain was able to solubilize silicate from Si-bearing mineral in fermentation liquid. Indeed, microbial leaching is a simple and effective technology for metal extraction from low-grade ores and mineral concentrates, and this method is being increasingly applied in mining industry (Bosecker, 1997). Mineral weathering mechanism has been increasingly clarified in terms of chemical weathering processes. These microbes have been indicated to accelerate the dissolution of silicates by perturbing mineral–water equilibria and reaction dynamics, the production of chelating agents, proton, organic ligands, hydroxyl anion, extracellular polysaccharides and enzymes, and silicate precipitation by metal sorption at the cell membrane (Bennett et al., 2001). However, the influence of bacteria in this process and its molecular mechanisms are still unclear (Uroz et al., 2009).

Effect of strain B1-5 on maize growth and Si content in soil and plants

Maize seedlings grown in soils inoculated with B1-5 showed greater shoot diameter ($t = -4.745$, $df = 18$, $P < 0.001$) and dry biomass ($t = -2.634$, $df = 18$, $P = 0.017$) than un-inoculated plants (Fig. 4). Soil inoculation with B1-5 strain significantly increased the growth of maize plants.

Figure 4. Shoot diameter (A) and biomass (B) of maize seedlings grown in the soil with and without inoculation of strain B1-5. Significant differences ($P < 0.05$) between inoculation and non-inoculated control are indicated by asterisk above bars. Values are mean $\pm$ standard error from ten replicates

The soluble Si content in the soil inoculated with B1-5 strain was higher than that in un-inoculated control ($t = -3.67$, $df = 18$, $P = 0.001$) (Fig. 5A). Maize plants grown in inoculated soil showed significantly higher Si level in shoots ($t = -2.55$, $df = 18$, $P = 0.020$) and roots ($t = -2.228$, $df = 18$, $P = 0.039$) than those in un-inoculated plants (Fig. 5B). Inoculation with B1-5 strain significantly increased soluble Si release from the soil and Si uptake by maize plants.

Our study clearly illustrated that the growth and Si content of maize plants were improved by inoculation with B1-5 strain via enhancement of soil Si availability and plant Si uptake. This is consistent with the findings that SSB, *Bacillus circulans*, *Burkholderia eburnean* and *Enterobacter ludwigii*, increased the release of Si in soil and Si acquisition by plants (Kang et al., 2017; Lee et al., 2019; Zahra et al., 1984). Many studies have

showed that Si application improves the growth of monocot crops including rice, sugarcane (*Saccharum officinarum*) and turf (*Lolium perenne*) (Nanayakkara et al., 2008; Savant et al., 1999; Snyder, 2001). Moreover, two field experiments in India have also reported that silicon solubilizing bacteria improve silicon uptake and crop yield in rice and sugarcane (Brindavathy et al., 2012; Pedda et al., 2016).

Figure 5. (A) Soluble Si content in strain B1-5-inoculated and un-inoculated soils. (B) Si content in the shoots (left) and roots (right) of maize seedlings grown in strain B1-5-inoculated and un-inoculated soils. Significant differences ($P < 0.05$) between inoculation and control are indicated by asterisk by different letters above bars. Values are mean $\pm$ standard error from ten replicates

Conclusion

In conclusion, this study not only highlights the existence of bryophyte rhizoids-associated silicate solubilizing bacteria, but also opens up a new approach searching for Si-solubilizing bacteria to increase Si availability in soil. Use of silicate solubilizing bacteria biofertilizer may serve as an important approach to increase Si availability and crop production. More detailed mechanisms of microbiological molecular biology and metabonomics about mineral weathering await further research.

Acknowledgements. This work was supported by the National Natural Science Foundation of China (31670414, 31770474), Fujian Provincial Excellent Youth Science Foundation (2017J06010), and Fujian-Taiwan Joint Innovative Center for Germplasm Resources and Cultivation of Crop (FJ 2011 Program, NO.2015-75).

REFERENCES

- [1] Afzal, A., Ashraf, M., Asad, S. A., Farooq, M. (2005): Effect of phosphate solubilizing microorganisms on phosphorus uptake, yield and yield traits of wheat (*Triticum aestivum* L.) in rainfed area. – Int J Agric Biol 7: 207-209.
- [2] Antonio, C. D. S., Rouws, L. F. M., Teixeira, K. R. S., Reis, V. M. (2016): Diazotrophic bacteria associated to sugarcane varieties cropped at northeast region of Brazil. – Revista Brasileira de Ciencias Agrarias 11: 272-280.

- [3] Bennett, P. C., Rogers, J. R., Choi, W. J., Hiebert, F. K. (2001): Silicates, silicate weathering, and microbial ecology. – *Geomicrobiol J* 18: 3-19.
- [4] Berger, B., Baldermann, S., Ruppel, S. (2017): The plant growth-promoting bacterium *Kosakonia radicincitans* improves fruit yield and quality of *Solanum lycopersicum*. – *J Sci Food Agric* 97: 4865-4871.
- [5] Bosecker, K. (1997): Bioleaching: metal solubilization by microorganisms. – *FEMS Microbiol Rev* 20: 591-604.
- [6] Brady, C., Cleenwerck, I., Venter, S., Coutinho, T., De Vos, P. (2013): Taxonomic evaluation of the genus *Enterobacter* based on multilocus sequence analysis (MLSA). – *Syst Appl Microbiol* 36: 309-319.
- [7] Brindavathy, R., Dhara, N., Rajasundari, K. (2012): Biodissolution of Silica by Silicon Bacteria in Sugarcane Rhizosphere. – *Res J Agri Sci* 3: 1042-1044.
- [8] Dannon, E. A., Wydra, K. (2004): Interaction between silicon amendment, bacterial wilt development and phenotype of *Ralstonia solanacearum* in tomato genotypes. – *Physiol Mol Plant Pathol* 64: 233-243.
- [9] Debona, D., Rodrigues, F. A., Datnoff, L. E. (2017): Silicon's role in abiotic and biotic plant stresses. – *Annu Rev Phytopathol* 55: 85-107.
- [10] Epstein, E. (1994): The anomaly of silicon in plant biology. – *P Natl Acad Sci USA* 91: 11-17.
- [11] Epstein, E. (1999): Silicon. – *Annu Rev Plant Biol* 50: 641-664.
- [12] Han, H. S., Lee, K. D. (2006): Effect of co-inoculation with phosphate and potassium solubilizing bacteria on mineral uptake and growth of pepper and cucumber. – *Plant Soil Environ* 52: 130-136.
- [13] Hoagland, D. R., Arnon, D. I. (1950): The water-culture method for growing plants without soil. – *Calif Agric Exp Stn Circ* 347: 357-359.
- [14] Hu, L., Xia, M., Lin, X., Xu, C., Zeng, R., Song, Y. (2018): Earthworm gut bacteria increase silicon bioavailability and acquisition by maize. – *Soil Biol Biochem* 125: 215-221.
- [15] Kaempfer, P., McInroy, J. A., Doijad, S., Chakraborty, T., Glaeser, S. P. (2016): *Kosakonia pseudosacchari* sp nov., an endophyte of *Zea mays*. – *Syst Appl Microbiol* 39: 1-7.
- [16] Kang, S. M., Waqas, M., Shahzad, R., You, Y. H., Asaf, S., Khan, M. A., Lee, K. E., Joo, G. J., Kim, S. J., Lee, I. J. (2017): Isolation and characterization of a novel silicate-solubilizing bacterial strain *Burkholderia eburnea* CS4-2 that promotes growth of japonica rice (*Oryza sativa* L. cv. Dongjin). – *Soil Sci Plant Nutr* 63: 233-241.
- [17] Lee, K. E., Adhikari, A., Kang, S. M., You, Y. H., Joo, G. J., Kim, J. H., Kim, S. J., Lee, I. J. (2019): Isolation and characterization of the high silicate and phosphate solubilizing novel strain *Enterobacter ludwigii* GAK2 that promotes growth in rice plants. – *Agronomy* 9: 144.
- [18] Liang, Y., Nikolic, M., Belanger, R., Haijun, G., Song, A. (2015): Silicon in Agriculture. From Theory to Practice. – Springer, Dordrecht.
- [19] Lin, W., Wang, Y., Li, B., Pan, Y. (2012): A biogeographic distribution of magnetotactic bacteria influenced by salinity. – *ISME J* 6: 475-479.
- [20] Ling, T. Y., Jong, H. J., Apun, K., Sulaiman, W. H. W. (2009): Quantifying *Escherichia coli* release from soil under high-intensity rainfall. – *T ASABE* 52: 785-792.
- [21] Ma, J. F. (2004): Role of silicon in enhancing the resistance of plants to biotic and abiotic stresses. – *J Plant Nutr Soil Sci* 50: 11-18.
- [22] Mehrvarz, S., Chaichi, M. R. (2008): Effect of phosphate solubilizing microorganisms and phosphorus chemical fertilizer on forage and grain quality of barely (*Hordeum vulgare* L.). – *Agri Environ Sci* 3: 822-828.
- [23] Myshlyayeva, L. V., Krasnoshchekov, V. V. (1974): Analytical Chemistry of Silicon. – Wiley, New York.

- [24] Nanayakkara, U. N., Uddin, W., Datnoff, L. E. (2008): Application of silicon sources increases silicon accumulation in perennial ryegrass turf on two soil types. – *Plant Soil* 303: 83-94.
- [25] Pedda, K. S., Mohiuddin, G., Balasubramaniam, P., Mahendran, P. P. (2016): Performance of silicate solubilizing bacteria on silicon uptake and yield of rice under lowland ecosystem. – *J Appl Nat Sci* 8: 55-59.
- [26] Pettersson, Å. K., Karlberg, B. (1999): Simultaneous determination of orthophosphate and silicate in brackish water. – *Anal Chim Acta* 378: 183-189.
- [27] Raven, J. A. (1983): The transport and function of silicon in plants. – *Biological Reviews* 58: 179-207.
- [28] Reynolds, O. L., Padula, M. P., Zeng, R., Gurr, G. M. (2016): Silicon: potential to promote direct and indirect effects on plant defense against arthropod pests in agriculture. – *Front Plant Sci* 7. DOI: 10.3389/fpls.2016.00744.
- [29] Savant, N., Korndörfer, G., Datnoff, L., Snyder, G. (1999): Silicon nutrition and sugarcane production: a review. – *J Plant Nutr* 22: 1853-1903.
- [30] Sheng, X. F. (2005): Growth promotion and increased potassium uptake of cotton and rape by a potassium releasing strain of *Bacillus edaphicus*. – *Soil Biol Biochem* 37: 1918-1922.
- [31] Sheng, X. F., He, L. Y. (2006): Solubilization of potassium-bearing minerals by a wild-type strain of *Bacillus edaphicus* and its mutants and increased potassium uptake by wheat. – *Can J Microbiol* 52: 66-72.
- [32] Singh, G., Biswas, D. R., Marwaha, T. S. (2010): Mobilization of potassium from waste mica by plant growth promoting rhizobacteria and its assimilation by maize (*zea mays*) and wheat (*triticum aestivum*): a hydroponics study under phytotron growth chamber. – *J Plant Nutr* 33: 1236-1251.
- [33] Snyder, G. H. (2001): Calibration of soil and plant silicon analysis for rice production. – *J Plant Nutr* 24: 1071-1084.
- [34] Soil Survey Staff. (2010): Keys to Soil Taxonomy. 11th Ed. – USDA Natural Resources Conservation Service, Washington, DC.
- [35] Takahashi, E., Miyake, Y. (1976a): Distribution of silica accumulator plants in the plant kingdom (1) Monocotyledons. – *J Sci Soil Manu Jpn* 47: 296-300.
- [36] Takahashi, E., Miyake, Y. (1976b): Distribution of silica accumulator plants in the plant kingdom (2) Dicotyledons. – *J Sci Soil Manu Jpn* 47: 301-306.
- [37] Takahashi, E., Miyake, Y. (1976c): Distribution of silica accumulator plants in the plant kingdom (3) Gymnospermae, Pteridophyta, Bryophyta. – *J Sci Soil Manu Jpn* 47: 333-337.
- [38] Tamura, K., Dudley, J., Nei, M., Kumar, S. (2007): MEGA4: Molecular evolutionary genetics analysis (MEGA) software version 4.0. – *Mol Biol Evol* 24: 1596-1599.
- [39] Thompson, J. D., Gibson, T. J., Plewniak, F., Jeanmougin, F., Higgins, D. G. (1997): The CLUSTAL_X windows interface: flexible strategies for multiple sequence alignment aided by quality analysis tools. – *Nucleic Acids Res* 25: 4876-4882.
- [40] Uroz, S., Calvaruso, C., Turpault, M. P., Freyklett, P. (2009): Mineral weathering by bacteria: ecology, actors and mechanisms. – *Trends Microbiol* 17: 378-387.
- [41] Vessey, J. K. (2003): Plant growth promoting rhizobacteria as biofertilizers. – *Plant Soil* 255: 571-586.
- [42] Wang, Jim, J., Dodla, Syam, K., Henderson, Rodney, E. (2004): Soil silicon extractability with seven selected extractants in relation to colorimetric and ICP determination. – *Soil Science* 169: 861-870.
- [43] Witzel, K., Strehmel, N., Baldermann, S., Neugart, S., Becker, Y., Becker, M., Berger, B., Scheel, D., Grosch, R., Schreiner, M., Ruppel, S. (2017): Arabidopsis thaliana root and root exudate metabolism is altered by the growth-promoting bacterium *Kosakonia radicincitans* DSM 16656(T). – *Plant Soil* 419: 557-573.

- [44] Wu, G. Q., Liu, H. L., Feng, R. J., Wang, C. M., Du, Y. Y. (2017): Silicon ameliorates the adverse effects of salt stress on sainfoin (*Onobrychis viciaefolia*) seedlings. – Plant Soil Environ 63: 545-551.
- [45] Xiao, Y., Wang, X., Chen, W., Huang, Q. (2017): Isolation and identification of three potassium-solubilizing bacteria from rape rhizospheric soil and their effects on ryegrass. – Geomicrobiol J 34. DOI: 10.1080/01490451.01492017.01286416.
- [46] Youssef, G. H., Seddik, W. M. A., Osman, M. A. (2010): Efficiency of natural minerals in presence of different nitrogen forms and potassium dissolving bacteria on peanut and sesame yields. – J Am Sci 6: 647-660.
- [47] Zahra, M. K., Monib, M., Abdel-Al, S. I., Heggo, A. (1984): Significance of soil inoculation with silicate bacteria. – Zentralblatt für Mikrobiologie 139: 349-357.
- [48] Zhang, C., Kong, F. (2014): Isolation and identification of potassium-solubilizing bacteria from tobacco rhizospheric soil and their effect on tobacco plants. – Appl Soil Ecol 82: 18-25.