6th Int. Conference on Silicon in Agriculture

Stockholm, SWEDEN, 26-30 August, 2014





OUR SPONSORS





Organisation committee

Assoc. Prof. Maria Greger (Main organizer), Stockholm University, **Sweden**, Prof. Alexander Lux, Comenius University in Bratislava, **Slovakia** PhD. Marek Vaculik, Comenius University in Bratislava, **Slovakia** PhL. Tommy Landberg, Stockholm University, **Sweden** Prof. Sylvia Lindberg, Stockholm University, **Sweden** Prof. Catherine Keller, Cerege Université Aix-Marseille III, **France** PhD. Rivka Elbaum, Hebrew University of Jerusalem, **Israel** PhD. Mary Provance-Bowley, Harsco Metals and Minerals, **USA** PhD. Martin Hodson, Oxford Brookes University, **UK** PhD. Michal Martinka, Comenius University in Bratislava, **Slovakia**

Invited Speakers

Plenary Speakers

Prof. Jonathan J Powell, University of Cambridge, UK Prof. Catherine Keller, Cerege Université Aix-Marseille III, France

Key Note Speakers

Prof. Carol Perry, Nottingham Trent University, UK

Prof. Richard R. Bélanger, Université Laval, Québec, Canada

Prof. Jian Feng Ma, Okayama University, Japan

Prof. Miroslav Nikolić, University of Belgrade, Serbia

Acknowledgements

Stockholm University Conference Service: Ami Hedblad, Beghol Alinia, Per Sterner and Karin Löfroth Arifin Sandhi Springer Verlag (Poster price) Stockholm Visitors board – Congress Stockholm (Maps)

© 2014 Maria Greger Stockholm University Printed by Stockholm Printcenter, Stockholm, Sweden ISBN: 978-91-637-6572-8



Agripower owns the world's largest deposit of diatomite, a natural source of amorphous silica. The deposit located in north Queensland, Australia where it is mined and processed into a range of high quality products used in agriculture and in other industries.

Agripower produces quality granulated Silica Fertiliser products, which are rich in Plant Available Silicon (PAS). Agripower's Silica Fertilisers are unique in that they also have high cation exchange capacities, significantly improving nutrient uptake. Agripower Silica fertilisers are available in two granule sizes, 2-5mm and 1-2mm and are suited for use in Agriculture and Turf.

Agripower has been at the forefront of trialling Silica Fertilisers on a wide range of soil types and crops in Australia and in many international locations. Trials using Agripower Silica Fertiliser have consistently shown improvements in crop yields and crop quality and improved utilisation of applied nutrients resulting in the potential to reduce applied fertilisers.

Agripower has also developed SIRA Soil Amendment for use in landscaping and sporting field construction. When incorporated into the soil SIRA will provide a source of Plant Available Silicon, improve the water holding capacity and nutrient retention capacity of most soils.

Over the past five years Agripower Australia Limited (Agripower) has spent in excess of AU\$40M on product research, testing, trials and construction of new processing facilities to supply the growing need for Silicon products in agriculture and industry.



Holistic review Presentation of new publication

'SILICON SOLUTIONS' Bent Edward

Hortcom, independent horticultural writer and consultant, Via Legionari in Polonia 33, 24128 Bergamo, Italy (hortcom@tin.it)

The use of silicon has the potential to become a fundamental tool in support of sustainable agriculture, biological/organic production and in safeguarding the environment. It can also render intensive agriculture more sustainable.

However, few publications (if any) have attempted to transform the large body of scientific research, experimentation and knowledge into a form more accessible to the practical interests and needs of growers and agronomists.

Comparative information on silicon-rich amendments; types, methods, timing and frequency of application, dose rates, is scarce. There are difficulties in defining the functions of silicon in crop production - the fact that silicon is excluded from the registered list of fertilizers and essential elements results in limited interest (or scepticism) to routinely use silicon amendments.

The effects of silicon on plants cannot be considered separately from its influence on soil structure, soil microorganisms and in the context of plant stress dynamics - because silicon has beneficial effects in ALL of these areas.

When testing the effects of silicon, experiments often finish by measuring external quality parameters such as weight, volume and appearance. This is insufficient since the effects of silicon can be even more pronounced in terms of post-harvest internal quality parameters: nutritional content, chemical residues (where crops are treated with fungicides and pesticides) and characteristics that positively influence the resistance of produce to transport, handling, storage and its behaviour in food transformation and processing.

The new book 'Silicon Solutions' takes a holistic approach to the above aspects, aiming to sensibilize modern-day agriculture and the food industry to the important role that silicon can play. As such it is the first of its type. The work is divided into sections addressed to agricultural, horticultural and floricultural crops together with a series of tables showing positive results obtained from the use of silicon. It is written in a straighforward style and amply illustrated.

Summary

Title: 'SILICON SOLUTIONS' Author: Edward Bent Language: English Publisher: Sestante Edizioni ISBN: 978-88-6642-151-1 Format: Paperback 17 x 24cm Pages: 184 plus cover Price: €39 (*net of postage & packaging, import duty and taxes*).

Discounts are available on quantity



Availability

The book will be available from Sestante Edizioni, Italy (despatch starting September 2014).

A pdf file (flipbook) will also be available at the same price. Orders should be made via e-mail until there is a direct online link. Sestante Edizioni, Via dell'Agro 10, 24124 Bergamo, Italy. Tel: +035 4124205 E-mail: info@sestanteedizioni.it www.sestanteedizioni.it

Foreword

Welcome to Stockholm and the 6th International Conference on Silicon in Agriculture

Dear Colleague of Silicon,

Welcome to the first Conference on Silicon in Agriculture to be held in Europe. The conference is given each 3rd year and the previous onces were held 1999 in Florida, US, 2002 in Tsuruoka, Japan, 2005 in Uberlandia, Brazil, 2008 in Durban, South Africa and 2011 in Beijing, China. We are pleased to host this 6th Conference at the Stockholm University, in Stockholm, the Nobel prize and capital town of Sweden.

The aim of this conference is to, during three days discuss the latest knowledge and the state of the art of the important issue of using application of Silicon in agriculture. Application of Silicon was shown to increase biomass production, resistance to several abiotic and biotic stressors, as well as to diminish the content of several toxic elements and increase the uptake of some nutrients in crops. Its possible role as an essential nutrient element to plants, animals and humans has long been discussed. Its possible essentiality can be put in relation to a decreased available Silicon level in arable soils, due to crop harvest and no Silicon application. The conference will therefore span from the chemistry of Silicon fertilizers via Silicon influence on soil and plants to the influence of Silicon on crop nutrition for human and animals.

The interest in Silicon in agriculture has increased a lot during the last six years, both in science and commercially. This book contains 76 exciting and important presentations from all over the whole world. The importancy of Silicon in agriculture is also shown by the interest to form an official Global Silicon Community whose main objectives are to gain full acceptance of silicon in the scientific, governmental, regulatory, and industrial communities, worldwide.

You are very welcome to participate in an important and exciting event!

Maria Greger

Chairperson, Silicon Conference Organising Committee 2014

Table of Content

Programme	9
Oral sessions	12
Poster sessions	20
Papers	23
Plenary	25
Papers in alphabetic order (except plenary)	32
Lists	185
List of Authors	187
List of Participants	193
Map of Stockholm University campus	198

Programme

PROGRAMME

Venues:

Stockholm University, Svante Arrenius väg 20 C, Biology Building, House E
 G-Hall (Oral sessions)
 G-Hall lobby (Registration, Welcome reception, Poster presentation, Swedish food tasting, Exhibition)

City Hall, Hantverkargatan 1, Stockholm City City Hall Reception hosted by The City of Stockholm

Stora Skuggans värdshus (Inn), Stora Skuggans väg 12, Norra Djurgården Conference dinner

Tuesday 26 August

(G-Hall lobby)

- 18.00 21.00 Registration
- 19.00 20.00 Welcome reception

Wednesday 27 August

(G-Hall)

Oral sessions

8.00 Registration

9.00	Opening and Welcome
9.15	Astrid Söderbergh Widding, Rector of Stockholm University
9.30	Information

PLENARY SESSIONS

Chair: Alexander Lux

9.45	Catherine Keller Silicon in agricultural soils: is availability an issue?	1
10.30	COFFEE AND POSTERS	
11.15	Jonathan Powell The human health value of the silicon content of food crops	2

12.00 LUNCH AND POSTERS

THEME 1 — Chemistry and analysis of silicon in soil, plant andenvironmentAbstract noAbstract no

13.15 Keynote	Carole Perry Chemical studies of biosilicification and analytical methods for analysing silicon	55
13.45	Mary Provance-Bowley A method for determining soluble silicon concentrations in non-liquid fertilizer materials	59
14.00	Meunier Jean-Dominique The 1% Na ₂ CO ₃ method as an indicator of bioavailable Si in crop land	50

Paper no

14.15	Tapasya Babu Dynamics of monosilicic acid in solid and solution phases of different soils of Louisiana	3
14.30	Daniel Conley Emerging understanding of anthropogenic impact on the ecosystem silica filter	14
THEME 2 — Chair: Catheri	Silicon cycle in agro-systems and Si-fertilizers ne Keller Abstrac	ct no
14.45	Wim Clymans Mineral Si fertilisation in forests – what can be learned from agro-systems?	13
15.00	Alex Höhn The importance of biogenic pools to Si cycling	X
15.15	COFFEE AND POSTERS	
15.45	Eric Struyf Silicate fertilization, crop production and carbon storage: an enhanced weathering approach	67
16.00	Floor Vandevenne Grazers: bio-catalysts of terrestrial silica cycling	73
16.15	Richard Haynes Unravelling the enigma of the effect of soil pH on Si availability	26
16.30	Thimo Klotzbücher Silicon cycling and budgets in rice production systems of Laguna, the Philippines	38
16.45	Brenda Tubana Changes on plant-available silicon level of selected soils from the Midwest and South USA applied with different rates of silicate slag	69
17.00	Mônica Sartori de Camargo Silicate fertilization on tropical soils: silicon availability, uptake and recovery by two sugarcane cultivars	10

17.15	Anika Marxen Controls on Si uptake by rice in Southeast-Asian paddy soils	46
17.30	Petra Tallberg Silicon fertilization and its impacts on phosphorus availability: a way to mitigate phosphorus fertilizer scarcity in the future?	68
17.45	Malcolm Keeping Provision of nitrogen as ammonium rather than nitrate increases silicon uptake in sugarcane	35

18.00 SWEDISH FOOD TESTING (G-Hall lobby)

BUSINESS MEETING (for all) — Forming a silicon society (G-Hall)

Chair: Lawrence Datnoff

18.30 - 19.30	Mary Provance-Bowley	58
	Formation of a Global Silicon Community	

Paper no

Thursday 28 August

Oral sessions (G-Hall)

THEME 3 — Molecular and physiological mechanisms of Si uptake and accumulation in plants		
Chair: Sylvia Line	-	aper no
9.00 Keynote	Jian Feng Ma Silicon transporters and their role in plants	43
9.30	Rivka Elbaum Characterization of silica accumulation in maize cell culture	17
9.45	Santosh Kumar The mechanism of biosilicification in <i>Sorghum</i> leaves	39
10.00	Sylwia Głazowska <i>Brachypodium</i> mutants defective in silicon transport as a tool for investigation of silicon effects on cell wall composition	24
10.15	COFFEE AND POSTERS	
11.00	Scott Leisner Finding what shouldn't be there: the discovery of silicon transporters in low Si accumulator plants	42
11.15	Oshry Markovich Silicification in sorghum: insights from a mutant defective in silicon uptake	45
11.30	Naoki Yamaji Transcriptional regulation of Si transporter genes involved in uptake and distribution in rice	75
11.45	Wendy Zellner Identification of Putative Plant Silicon Binding Proteins	77
12.00	LUNCH AND POSTERS	

THEME 4 — Silicon influence on plant growth and developmentChair: Marek VaculikPaper no

13.30	Miroslav Nikolic	52
Keynote	Role of silicon in plant nutrient acquisition and transport	

14.00	Gisele Silva Hormonal changes and growth of rice plants in response to bioagents and silicon fertilization application	65
14.15	Brandon White Effect of enhanced soil silicon on grain yield and agronomic parameters of wheat under sufficient and high nitrogen application rates	74
14.30	Mahendran Peyandi Paraman Fixing critical limit for silicon fertilization in the intensively rice growing soils of Periyar Vaigai Command area, Tamil Nadu, India	44

14.45	COFFEE AND	POSTERS

THEME 5 — Silicon influence on abiotic stress Chair: Maria Greger Paper no **Tommy Landberg** 15.15 41 Cadmium uptake in wheat protoplasts is reduced by silicon 15.30 Marek Vaculík 70 Influence of silicon on zinc toxicity in wheat 7 15.45 **Boris Bokor** Ionome and Si transporters affected by zinc and silicon interaction in maize 16.00 Naeem Igbal 31 Effect of silicon in modulating vigour of wheat (*Triticum aestivum* L.) seedlings under saline conditions 16.00 Waqas ud Din Khan 36 Screening and categorization of maize cultivars for silicon acquisition under salinity stress

CITY HALL RECEPTION — Hosted by the City of Stockholm

(City Hall in the town center, Hantverkargatan 1, Underground station T-Centralen)

19.00 SHARP!

Friday 29 August (G-Hall)

Oral sessions

THEME 6 — Silicon influence on biotic stress <i>Chair:</i> Malcolm Keeping		Paper no
9.00 Keynote	Richard Bélanger Silicon influence on biotic stress in plants	6
9.30	Ofir Katz Silica phytoliths in Asteraceae species: formation patterns and potential antiherbivory role	34
9.45	Marta Cristina Filippi Silicon and bioagents in leaf rice blast suppression	21
10.00	Sabine Holz Insights into molecular impact of silica on virus infected cucumber cultures	29
10.15	COFFEE AND POSTERS	
11.00	Olivia Reynolds Silicon alters the volatile profile of pest-infested grapevines and increases attractiveness to predators	61
11.15	David De Vleesschauwer Hormonal regulation of silicon-induced brown spot resistand in rice	16 ce
11.30	Jonas Van Bockhaven Transcriptome analysis of silicon-induced brown spot resistance in rice reveals central role of photorespiration	72
12.00	LUNCH AND POSTERS	

	Silicon influence on nutritional value of crops, in a d animal health aspects	human
Chair: Jonathan	-	Paper no
13.30	Valentin Kindomihou The effect of seasonal variations, covariations with minerals and forage value on Itchgrass [<i>Rottboellia cochinchinensis</i> (Lour.) W.D. Clayton]' foliar silicification from sudanian Beni	<i>37</i> n
13.45	Maria Greger Si decreases Cd and As in crops — a field study	25
THEME 8 — E Chair: Mary Pro	Effects of silicon fertilizer products mance-Bowley	Paper no
14.00	Peter Prentice From the pot to the plot: An extensive investigation into crop responses to a silica fertiliser	57
14.15	Prakash Nagabovanalli Behaviour of different levels and grades of Diatomite as silico source in acidic and alkaline soils	51 on
14.30	Neeru Jain Ortho Silicic Acid (OSA) based formulations facilitates improvement in plant growth and development	32
14.45	Ratna Kumar Pasala Assessment of silixol (osa) efficacy on wheat physiology: growth and nutrient content under drought conditions	54
15.00	COFFEE AND POSTERS	
15.30	Wiesław Ciecierski Impact of silicon based fertilizer OPTYSIL on abiotic stress reduction and yield improvement in field crops.	12
15.45	Henk-Maarten Laane The efficacy of the use of foliar sprays with silicic acid	40
16.00	Vladimir Matichenkov New generation Si fertilizers: activation of the natural plant defense system	47

 16.15 Jose Cristancho Silicon in agriculture new developments in Latin America
 16.30 Edvard Bent Presentation of new publication Holistic review 'SILICON SOLUTIONS'

Discussion, Prices/Awards, Next Conference, Closure

16.45

CONFERENCE DINNER AT STORA SKUGGAN

(Stora Skuggans värdshus (Inn), Stora Skuggans väg 12)

19.00

Saturday 30 August

POST CONFERENCE STOCKHOLM SIGHTSEEING

8.30-ca. 17

8.30 Bus from the Metro station "Universitetet" to Vaxholm, a typical archipelago town. On the way to Vaxholm you will among other things see a runestone and an old silicon mine. In Vaxholm (or on the boat) you will have the possibility to have lunch on your own.

13.30 SHARP! Ferry from Vaxholm to Stockholm city (Djurgården). The sightseeing ends with a visit to Vasa museum and if there is time also the old town.

15

Poster sessions

(G-Hall lobby)

THEME 2 — Silicon cycle in agro-systems and Si-fertilizers

	Poster nr	Paper nr
Ana Lucia Barão Pedogenic silica in two croplands of the Belgian loam belt	1	4
Leonardo Büll Residual effects of steel slags on the availability of silicon in the soil	2	9

THEME 3 — Molecular and physiological mechanisms of Si uptake andaccumulation in plantsPoster nrPaper nr		
Paula Cartes Gene expression of superoxide dismutase isoforms and RUBISCO in <i>Lolium perenne</i> in response to silicon supply	3	11
Sofia Pontigo Identification of putative Si transporters in ryegrass plants	4	56
Gen Sakurai The role of Casparian strips and Lsi transporter distribution in efficient Si transport in rice	5	64

THEME 4 — Silicon influence on plant growth and develo	opment	
	Poster nr	Paper nr
Martin Hinrichs Transcriptomic analysis of Si-induced casparian band development in rice	6	28
Abbas Ghanbari-Malidarreh Effect of silicon and phosphorus rates on Si and P content and uptake of rice (<i>Oryza sativa</i> L. cv. Tarom) and disease	7	23

THEME 5 — Silicon influence on abiotic stress	Poster nr	Paper nr
Muhammad Ansar Farooq Silicon-mediated oxidative stress tolerance and genetic variability in rice (<i>Oryza sativa</i> L.) grown under combined stress of salinity and boron toxicity	8	18
Ivana Fialová The effect of silicon on the antioxidative response of maize young plants to salinity and zinc stress	9	20
Dirceu Maximino Fernandes Influence of silicon on castor beans plants under aluminium stress	10	19
Lucas Barbosa de Freitas Effect of silicon on mitigation of aluminum stress in upland rice plants	11	22
Lourdes Hernández-Apaolaza Effect of silicon in soybean (<i>Glycine max</i> L.) plants grown under zinc deficiency conditions	12	27
Vagnum Flaumbert Rivera Varela Effect of silicon addition on rice <i>(Oryza sativa)</i> plants grown under zinc deficiency	13	62
Iqbal Hussain Silicon decreases mobility and accumulation of cadmium in wheat grains	14	30
Dragana Nikolić Silicon has a protective effect on cucumber grown in copper Excess	15	53
Lenka Šimková The effect of silicon on some physiological parameters of maize leaves under antimony stress	16	66
Miroslava Vaculíková Growth and antioxidative response of maize roots exposed to antimony and silicon	17	71
Ivan Zelko Silicon effect on growth dynamics, macro and microelements content in cadmium and arsenic treated poplar calli	18	76

THEME 6 — Silicon influence on biotic stress Poster nr Paper nr Telma Batista Diversity of insect fauna associated with rice fertilized with 19 5 silicon and bioagents Fabricio de Ávila Rodrigues Histochemical aspects of wheat resistance to leaf blast 20 63 mediated by silicon Xiaolei Jin The role of Si and potassium bicarbonate in controlling 21 33 epidemics of *Podospheara aphanis* on strawberry in the field **Emma McLarnon** The role of silicon in tall fescue leaf characteristics 22 48 **Renata Sousa Resende** Sorghum resistance to anthracnose mediated by silicon 23 60

THEME 8 — Effects of silicon fertilizer products

	Poster nr	Paper nr
Anna Botta Català Evaluation or Armurox® (Complex of peptides with soluble silicon) on mechanical and biotic stresses in graminae	24	8
Radosław Wilk Indirect effect of silicon product against apple scab and strawberry diseases	25	49

Papers

PLENARY

1

Silicon in agricultural soils: is availability an issue?

Keller C, Meunier J-D

Aix-Marseille Université, CNRS, Collège de France, IRD, CEREGE, UMR 7330, Technopôle de l'Environnement Arbois-Méditerranée, BP 80, 13545 Aix-en-Provence, Cedex 4, FRANCE (keller@cerege.fr)

Silicon cycle is closely related to the weathering of silicates and terrestrial plants make significant contributions to the weathering budget and soil formation. The perturbation of the terrestrial Si cycle by human occupation has become a challenging issue because of possible impact on the equilibrium of aquatic ecosystems and agriculture sustainability. Indeed, while in terrestrial plants the concentration of Si is highly variable (from below 1 to higher than 100 mg/g dry weight (DW)) depending on the plant type and the growth medium, crops such as rice and wheat may contain more than 10 mg Si/g in shoots DW as phytoliths and, as a result, are considered as Si accumulators. Si concentration in those plants falls within the same range as values measured for some macronutrients such as Ca, Mg or P, but no Si fertilization is used, at least in Europe. More importantly the beneficial effect of Si in European agriculture has never been assessed probably because it is assumed that Si is not a limited element contrary to depleted soils found in countries under the influence of tropical climate regimes (i.e. Asia, Australia, South America) where Si fertilization is commonly performed. However, in Europe, the stagnation of wheat yields is observed since the early 90's and the reasons of it are still not clearly established (Brisson et al., 2010). The hypothesis that the depletion of plant-available silica in soils could lead to yield decline has been proposed by Savant et al. (1997). No recent studies have been published to support Savant et al.'s hypothesis, which remains of utmost interest considering the question of the sustainability of food production. Indeed, the budget of silica in cultivated areas is poorly documented and it might strongly differ from the natural ecosystems depending on agricultural practices, i.e. if straws are reincorporated or not to the soils.

However, although Si is not generally considered as an essential element, a large set of literature highlights its beneficial effects on crops (Jones and Handreck, 1967; Epstein, 1999; Datnoff et al., 2001). This has been demonstrated in a whole range of pots, hydroponic and field experiments (e.g. Epstein, 1994; Datnoff et al., 2001) and Si alleviating effect is particularly remarkable in plants exposed to biotic or abiotic stresses. Many supporting evidences will be presented within the course of this conference. Si acts at several levels in the plant functioning. It has 1) a structural function, as Si increases cell wall resistance and rigidity; 2) a physiological function, as it reduces evapotranspiration, increases root oxygenation and enhances interactions with other nutrients; 3) a protective function as it induces a resistance to many pathogens and animals. However, the mechanisms responsible for alleviating stresses remain unclear because they may act in the soils, at the root surface and *in planta* (roots and shoots).

Plants take up silicic acid from the soil solution (Ding et al., 2005; Henriet et al., 2008), which concentration is not correlated with total soil Si concentration (Savant et al., 1997).

Several studies have evaluated the fluxes of Si between plants, soil solutions and soil minerals in natural ecosystems (Keller et al., 2012). The major finding of these studies was that plants absorb a significant fraction of dissolved Si that originates from litterfall decomposition i.e phytolith dissolution. More recently Cornelis et al. (2011) showed that adsorbed Si on Fe oxides might be a significant pool of readily soluble Si pool besides phytoliths. Assuming that phytoliths are the only source of Si for crops, a simple calculation shows that the phytoliths pool would be depleted within a few decades taking into account an exportation of 50-100 kg Si/ha/yr and an initial pool of Si from phytoliths of 1t Si/ha (Bartoli, 1983). However, correlations were also observed between the Si concentration in rice and the percentage of clay in soils (Cheng, 1982), and between the Si concentration in rice or banana and the stock of weatherable minerals (Henriet et al., 2008; Makabe et al., 2009).

These findings raise several questions: do we observe losses of bioavailable Si stocks in soils experiencing straw (where Si is mostly accumulated in crops) exportation? Are phytoliths the only bioavailable Si compartment for plants in soils or do other soil minerals, as fine clay minerals notably, participate to this compartment? How is the extent of Si adsorbed on soil minerals? In other words, what is the bioavailable Si pool for plants, how can you assess it and how does it evolve with plant uptake?

In the presentation we review evidences from recent literature and we present original data that suggest that crops modify silicon biogeochemical cycle through both accelerated Si uptake due to crop plants being more efficient than natural vegetation at taking up nutrients and other elements, and Si export through total or partial shoots removal from the field. The resulting trend is a potential decrease in long term phytoavailable (less phytoliths) and short term phytoavailable Si (soluble) leading to lower Si concentrations in plants and probable lower Si exports out of the soil. The consequences are: a) putative changes in the various Si pools in soil and their respective contribution to Si cycle (Struyf et al., 2010; Carey and Fulweiler, 2011); b) a possible impact on crop yield and plant health (Savant et al., 1997).

The literature review shows also that the extent of these changes is not enough documented, neither is the impact of Si soil depletion on the long term. There is thus a need for characterizing the various Si pools in soil including their dissolution kinetics and their relative contribution to amorphous silicon and phytoavailable silicon. The relative contribution of clays minerals, phytoliths and other amorphous Si forms to the phytoavailable Si pool needs to be clarified. This would allow 1) using Si as a putative tracer of soil element depletion in European soils where silicon losses are not balanced by fertilizers inputs and 2) assessing the extent of plant-available Si depletion in soils.

- Brisson, N <u>Gate, P., Gouache, D., Charmet, G., Oury, F.X., Huard, F.</u> (2010) Why are wheat yields stagnating in Europe? A comprehensive data analysis for France. Field Crops Research 119, 201-212.
- Carey, J.C., Fulweiler, R.W. (2011) Human activities directly alter watershed dissolved silica fluxes. Biogeochemistry. DOI 10.1007/s10533-011-9671
- Cornelis, J.T., Titeux, H., Ranger, J., Delvaux (2011) Identification and distribution of the readily soluble silicon pool in a temperate forest soil below three distinct tree species. Plant Soil 342, 369-378.
- Datnoff, L.E., Snyder G.H., Korndorfer G.H., Ed. (2001) Silicon in agriculture. Studies in Pl. Sci. 8. Elsevier.
- Cheng B.T. (1982) Some significant functions of silicon to higher-plants. J.Pl. Nutr. 5, 1345-1353.
- Ding T.P., <u>Ma, G.R.</u>, <u>Shui, M.X.</u>, <u>Wan, D.F.</u>, <u>Li, R.H.</u> (2005) Silicon isotope study on rice plants from the Zhejiang province, China. Chem. Geol. 218, 41-50.
- Epstein, E. (1999) Silicon. Annual Review of Plant Physiology and Plant Molecular Biology 50, 641-664.

Epstein E. (1994) The anomaly of silicon in plant biology. PNAS 91, 11-17.

Henriet C., Bodarwe L., Dorel M., Draye X., Delvaux B. (2008) Leaf silicon content in banana (Musa spp.) reveals the weathering stage of volcanic ash soils in Guadeloupe. Plant Soil 313, 71-82.

Bartoli F. (1983) The biogeochemical cycle of silicon in two temperate foresty ecosystems, in Hallberg R. ed., Environmental Biogeochemistry, Ecol. Bull. (Stockholm), 35, 469-476.

Jones L.H.P., Handreck K.A. (1967) Silica in soils, plants, and animals. Adv. Agron. 19, 104-149.

- Keller C., F. Guntzer, D. Barboni, J. Labreuche, J.-C. Meunier (2012) Impact of agriculture on the chemical weathering of silicates: evidences from the study of the Si biogeochemical cycle. CR Geoscience 344, 739-746.
- Makabe, S., Kakuda, K., Sasaki, Y., Ando, T., Fujii, H., Ando, H. (2009) Relationship between mineral composition or soil texture and available silicon in alluvial paddy soils on the Shounai Plain, Japan. Soil Sci. Plant Nutr. 55, 300-308.
- Savant N.K., Datnoff L.E., Snyder, G.H. (1997) Depletion of plant-available silicon in soils: a possible cause of declining rice yields. Comm.Soil Sci. Plant Anal. 28, 1245-1252.
- Struyf, E., Smis, A., Van Damme, S., Garnier, J., Govers, G., Van Wesemael, B., Conley, D.J., Batelaan, O., Frot, E., Clymans, W., Vandevenne, F., Lancelot, C., Goos P., Meire, P. (2010) Historical land use change has lowered terrestrial silica mobilization. Nature Communications 1.

PLENARY

2

The Human Health Value of the Silicon Content of Food Crops

Powell J J

MRC Human Nutrition Research, Elsie Widowson Laboratory, Fulbourn Road, Cambridge CB1 9NL, UK (Jonathan.Powell@mrc-hnr.cam.ac.uk)

In considering a balanced discussion around the title of this presentation, a state-of-art understanding of silicon's role in human health must first be developed.

Four strong threads of evidence point to silicon's (Si) key site of bio-activity as collagen (i.e. in connective tissues such as bone, skin, blood vessels and joints). First, analysis indicates that >80% of Si is found in the connective tissue [1]. Secondly, in bone for example, decalcification leaves only the residual connective tissue materials (>90% collagen) and of all elements analysed Si retains by far the strongest presence [2]. Thirdly, supplementation sees loading of Si into connective tissues – in equilibrium with the circulation – whilst robust deficiency studies show an adverse influence on connective tissue health [3-6]. Finally, the pattern of changing Si concentrations in bone with age clearly follows that of collagen turnover [1]. Nonetheless the actual amount of Si in connective tissues (associated with collagen) is very small. In the rat for example, 40 µg Si appears sufficient for the entire connective tissue of bone [1]. This tissue accounts for about 50% of their overall collagen content and 50% of their overall Si content. Even extreme Si deficient rodent diets contain ~ 2 ug Si/g feed and the average adult rat will consume 20-30 g feed per day. Moreover, in a state of imposed dietary Si deficiency urinary losses of the element approach zero [6]. Obligatory losses - perhaps a few µg/day- will occur but it is abundantly evident that the amount of Si gained from a Si deficient diet is more than enough to offset this. Indeed, in a large multinational, collaborative study that was led by my group, we have recently identified the first mammalian Si transporter (exporter) [7]. This, and a likely corresponding importer, probably maintain Si homeostasis very tightly [6].

The Si deficient animal models that do exist come in two forms. The most detailed and recent study is from Jugdaohsingh *et al* in rats [6]. In this study, dietary levels were $\sim 3 \ \mu g$ Si/g feed and, as noted above, Si levels were conserved *versus* supplemented animals. Nonetheless, bone was compromised: in particular phosphate levels were low and the growth plate did not close so the long bones were abnormally long [6]. Rats, however, were healthy. In contrast Carlisle in 1972, and Schwarz & Milne in the same year, studied the impact of extreme Si deficiency in chicks and rats respectively [4,5]. In neither paper is there sufficient detail to judge robustness of study design but both reported extreme phenotypes – failure to thrive and consistently under-developed connective tissues [4,5]. Animals were deprived of Si at birth and, based on the work of Jugdaohsingh *et al* [6] noted above and the extensive studies of Nielsen [8] where no visible phenotype is ever observed in Si-deficient animals, it seems likely that these early studies had achieved an ultra-low dietary Si content that today cannot be easily, or perhaps ethically, replicated. However, there appears to be no analogous human disorder that mimics these extreme Si deficient phenotypes of experimental animals in the

1970s. Indeed, the tiny amounts of Si apparently required to support collagen health, the ubiquitous nature of Si in food, drinking water and other ingested fluids make it inconceivable that a frank Si deficient state exists in humans. If the story ended here then the title of this presentation would merit little discussion. However, this is not the case.

Interestingly, Jugdaohsingh et al. has found that in the same strain of adult female rats, consuming slightly different laboratory (chow) diets, the Si level of bone varies four fold [2]. The levels in the collagen fraction of bone are very similar and the difference is all about loading into the mineral phase [2]. Bone mineral is biological apatite and varies in its precise nature with age and probably diet as well as other environmental and/or 'early life programming' events. Silicon's role in this respect is not understood. Nonetheless, in a large epidemiological study with the US Framingham Cohort, there was a remarkable positive association shown between dietary Si intakes and bone mineral density (BMD) for premenopausal women [9]. In short, the higher the intake of Si then the higher the BMD which, within such a population, is a marker for bone health (*i.e* a higher BMD = a lower risk of osteoporosis development). This dietary effect was also observed, albeit less dramatically, in men but not at all in post-menopausal women [9]. A follow up study has confirmed the requirement for sex hormones, probably estradiol, for the Si-BMD interaction to be observed since hormone replacement therapy restores the association in post-menopausal women [10]. A murine intervention study confirmed the positive association between fasting serum Si levels (a marker of Si status) and BMD in adult female rats [3]. Taken together these data indicate that, in men and certainly up until the menopause in women, dietary Si levels do partially determine BMD and thus the adequacy of bone health in later life years when osteoporosis is so prevalent. Further work is required to really confirm this: namely a prospective long term intervention study in humans with follow through to later life years although it would take decades for incontrovertible evidence to be arrived at. In the meantime, more mechanistic studies are required that really identify the effect that Si-doping of biological apatite (bone mineral) is having and why this should impart advantage to bone. Nonetheless, given that both epidemiological and murine intervention studies support a relationship between higher dietary Si levels and superior bone mineral quality [3], the question of which foods are contributing to optimal Si nutrition is important.

Silicon is found almost entirely in (i) ingested fluids and (ii) in the plant-food-based portion of the diet [11]. In meat, *i.e.* muscle and fat, Si is at irrelevantly low levels [8,12]. The Si intake from fluids is mostly attributable to (i) drinking water where levels vary from typically 4-14 mg/L although they may be much higher in certain mineral waters and (ii) beer where the macerated grain may release Si to levels of saturation for orthosilicic acid in water (~ 50 mg/L). On average adult humans ingest about 20-50 mg Si/day and ~ 20% is derived from fluids, the remainder coming from plant-based foods [8,13]. We have reported food-Si contents in a large survey and analysis of UK foods [11]. Cereal crop-based foods (bread, pasta, breakfast cereals etc.) are a major dietary source not only due to Si content but because of the quantities of such foods eaten. Based upon studies that traced Si through the brewing process of beer making, it seems likely that both the Si content of the crop plant, and the way in which the plant is processed for ingestion, are key parameters in determining the Si content (value) of the final food [14,15]. More work in this area would be needed. However, cereal crops are not the only Si-accumulating plants that are ingested by humans: green beans, some fruits and legumes all showed high levels of Si and when regularly consumed in the diet they will significantly contribute to overall Si intakes.

Finally, a word on "bioavailability" of Si from foods. Technically, bioavailability concerns the extent to which the ingested nutrient is available to its biological site of activity (e.g.

absorbed iron being incorporated into haemoglobin of the red cell). In the case of Si this biological site of activity is likely to be bone mineral for optimal nutrition purposes but, as noted above, further work is required and biomarkers for this don't exist. In this paper, therefore, bioavailability will mean the amount of Si reaching the circulation (i.e. absorbed). Urinary output over the 6-8 h following ingestion of Si is an excellent measure of its absorption [16]. Indeed, this is one of the common ways that mineral/trace element haemostasis is achieved: absorption is high and then urinary excretion dictates retention – phosphate, for example, is handled in this way. In healthy adults, to maintain balance, the amount of a nutrient absorbed must very closely match the amount excreted. Using this 'urinary output' approach in humans, Jugdaohsingh et al [13], and Sripanyakorn et al [17], have shown that Si is best absorbed as orthosilicic acid from drinking water (circa 60% of the overall dose) although this is almost equalled by that from green beans. Cereals show high bioavailability at ~ 40 % whereas bananas, for example, have an apparently uniquely low bioavailability at $\sim 2-5\%$ [13]. Thus, whilst the Si levels of the plant food and its subsequent processing will determine Si intake, the additional issue of bioavailability needs to be considered when determining nutritional benefit that humans might derive.

In summary, Si must be essential to humans. It is highly conserved during imposed deficiency in mammals and at least one transporter is now known. It is all but impossible to deplete from collagen but severe phenotypes appear when this is achieved. Humans are unlikely to lack Si to this level. Nonetheless, there appears to be a second tier of silicon requirements in humans – not essential but contributing to optimal (long-term) bone health. In this case diet *does* impact. Some plant-based foods are a very significant source of bioavailable silicon and the extent of their ingestion can separate out humans with low, moderate or high levels of dietary silicon intake.

I acknowledge the support of the MRC (grant number MC_US_A090_0008/Unit Programme number U1059) and am indebted to my long-term friend and colleague, Ravin Jugdaohsingh, for the brainstorming over the years, his commitment to understanding how dietary silicon impacts human health and our shared passion for all things silicon!

- [1] Jugdaohsingh R, Watson A, Pedro LD, Powell. The decrease in silicon concentration of the connective tissues with age in rats is a marker of connective tissue turnover. *Submitted*.
- [2] Jugdaohsingh R, Pedro LD, Watson A, Powell. Silicon and boron differ in their localisation and loading in bone. *Submitted*.
- [3] Jugdaohsingh R, Watson A, Bhattacharya P, van Lenthe GH, Powell. Positive association between serum silicon levels and bone mineral density in female rats following oral silicon supplementation with monomethylsilanetriol. *Submitted*.
- [4] Carlisle EM. Silicon: an essential element for the chick. Science 1972;178:619-621.
- [5] Schwarz K, Milne DB. Growth-promoting effects of silicon in rats.Nature 1972;239:333-334.
- [6] Jugdaohsingh R, Calomme MR, Robinson K, Nielsen F, Anderson SHC, D'Hease P, Geutsens P, Loveridge N, Thompson RPH, Powell JJ. Increased longitudinal growth in rats on a silicon-depleted diet. Bone 2008;43:596-606.
- [7] Ratcliffe S, Jugdaohsingh R, Vivancos J, Marron A, Deshmukh R, Ma JF, Mitani-Ueno N, Boekschoten MV, Muller M, Mawhinney RC, Kinrade SD, Isenring P, Belanger RR, Powell JJ. Identification of a mammalian silicon transporter. *Submitted*.
- [8] Jugdaohsingh R. Silicon and bone health, J. Nutr. Health Aging. 2007;11:99-110.
- [9] Jugdaohsingh R, Tucker KL, Qiao N, Cupples LA, Kiel DP, Powell JJ. Dietary silicon intake is positively associated with bone mineral density in men and premenopausal women of the Framingham Offspring Cohort. J Bone Miner Res. 2004;19:297–307.

- [10] Macdonald HM, Hardcastle AC, Jugdaohsingh R, Fraser WD, Reid DM, Powell JJ. Dietary silicon interacts with oestrogen to influence bone health: evidence from the Aberdeen Prospective Osteoporosis Screening Study. Bone. 2012;50:681-87.
- [11] Powell JJ, McNaughton SA, Jugdaohsingh R, Anderson SH, Dear J, *et al.* A provisional database for the silicon content of foods in the United Kingdom. Br J Nutr 2005;94:804–12.
- [12] Pennington JAT. Silicon in food and diets. Food Addit Contam 1991;8: 97–118.
- [13] Jugdaohsingh R, Anderson SH, Tucker KL, Elliott H, Kiel DP, Thompson RP & Powell JJ. Dietary silicon intake and absorption. Amer J Clin Nutr 2002;75:887-893.
- [14] Walker C, Freeman G, Jugdaohsingh R and Powell JJ. Silicon in Beer: Origin and Concentration. In: Beer in Health and Disease Prevention. Preedy VR, Watson RR (Eds). *Elsevier Inc. New York*. 2008
- [15] Sripanyakorn S, Jugdaohsingh R, Elliott H, Walker C, Mehta P, Shoukru S, Thompson RP & Powell JJ (2004) The silicon content of beer and its bioavailability in healthy volunteers. Bri J Nutr 91(3), 403-9.
- [16] Pruksa S, Siripinyanond A, Powell JJ, Jugdaohsingh R. Silicon balance in human volunteers; a pilot study to establish the variance in silicon excretion versus intake. Nutr Metab 2014;11:4.
- [17] Sripanyakorn S, Jugdaohsingh R, Dissayabutr W, Anderson SHC, Thompson RPT, Powell JJ. The comparative absorption of silicon from different foods and food supplements. Br J Nutr. 2009;102:825-34.

Table 1. Silicon content of food Food Groups		Si conter	nt (mg/1	$(00g)^{l}$	Si content ¹
		Range			(mg/portion)
			Mean		
		Ν			
Cereals G	rains & Products	1.34-23.4	7.79	16	2.92 (37.5 g
	Breakfast	0.34-6.17	2.87	15	1.45 (50.5 g
Cereals		1.05-2.44	1.56	5	0.406 (26 g
corouis	Breads/Flour	0.88-3.76	1.54	8	1.85 (120 g
	Biscuits	0.62-1.84	1.11	7	2.55 (230 g
	Rice				2
	Pasta				
Fruits					
	Raw& canned	0.1-4.77	1.34	33	1.35 (101 g
	Dried	6.09-16.61	10.54	3	3.51 (33.3 g
Vegetable	S	0.1-8.73	1.79	49	1.25 (70 §
Legumes		0.38-4.42	1.46	11	0.759 (52 g
Nuts & Se	eds	0.28-1.99	0.78	4	0.174 (22.3 g
Milk & M	ilk Products	0.07-0.47	0.31	3+TDS	0.288 (93 g
Meat & M	eat Products	0.1-1.89		TDS	0.125-2.36 (125 g
Beverages	(non-alcoholic)				
	Tap water	0.10-0.61	0.37	11	0.740 (200 g
	Mineral/Spring	0.24-1.46	0.55	14	1.82 (330 g
waters		0.24-0.86	0.51	6	1.33 (260 g
	Tea & Coffee	0.05-1.5	0.38	11	0.866 (228 g
	Fruit juices	0.11-0.19	0.15	6	0.507 (338 g
-	Fizzy/Carbonate	0.2-3.96	1.30	6	3.38 (260 g
d^5	-				
	Milk based ⁵				
Beverages	(alcoholic)				
	Beers	0.9-3.94	1.92	76	6.37 (can/bottle
	. 5				11.0 (1 pint
	Wines ⁵	0.68-2.31	1.35	3	1.69 (125 g
	Port/Sherries'	1.24-1.26		2	0.62-0.63 (50 g
	Liquor/Spirits'	0.06-0.20	0.13	1	0.052 (40 g

¹Silicon content of foods is from Powell et al. [11]. N = number of samples. TDS: sample from the UK Food Standard Agency Total Diet Study. For each food group the range, mean level and number of foods analysed is given. Mean portion sizes are given in parentheses.

Papers in alphabetic order

Theme 1 ORAL

3

Dynamics of Monosilicic Acid in Solid and Solution Phases of Different Soils of Louisiana

<u>Babu T¹</u>, Tubana B¹, Datnoff L², Yzenas J³, Wang J¹

¹ School of Plant, Environment and Soil Sciences, 104 Sturgis Hall, Louisiana State

University, Baton Rouge, 70803, USA (tbabul@tigers.lsu.edu)

² Department of Plant Pathology and Crop Physiology, LSU AgCenter, Baton Rouge, LA, USA

³ Plant Tuff Inc., Edward Levy Corporation, Dearborn, Michigan, 48120, USA

The concentration of monosilicic acid (H_4SiO_4), the form of silicon (Si) taken up by plants, in soil solution changes due to adsorption (to mineral surfaces), polymerization, and dehydration. Studies have shown that active Fe and Al fractions, clay content, pH, and phosphorus levels in soils influence the adsorption of H_4SiO_4 . A series of laboratory experiments were initiated using an array of soils from Louisiana with varying clay content and chemical properties to: 1) quantify the adsorbed fraction of added H_4SiO_4 in soil solution with time, and 2) document the effect of addition of wollastonite and silicate slag on the concentration of H_4SiO_4 in soil solution within a 240-day period.

The data needed to generate the isotherm adsorption curves were obtained by incubating two sets of six 1-g samples weighed into centrifuge tubes with 25 mL of 0.1 M NaCl solutions with varying concentrations of H₄SiO₄ (0, 10, 20, 40 and 50 *u*g Si g⁻¹) at the speed of 15 rpm for 30 days. Sequential samplings of the solutions were carried out after 4, 7, and 30 days and analyzed for Si concentration using Molybdenum Blue Colorimetry (MBC). The difference between the amount of Si in solution for each sampling time and the initial concentration represented the amount of Si adsorbed by the soil from the H₄SiO₄ + 0.1 M NaCl solution. Separate experiments were established to address the second objective of this experiment wherein silicate slag, wollastonite and soil only were weighed and incubated into separate centrifuge tubes with 25 mL of 0.1 M NaCl for a 240-day period, during which supernatant solutions were subsampled at pre-determined intervals and analyzed for Si concentration using MBC. Another experiment was conducted, but this time, the six soils from Louisiana was incubated with 100 and 135 mg wollastonite and silicate slag, respectively.

Across soil types, the amount of adsorbed H_4SiO_4 increased with increasing concentration of added H_4SiO_4 . The amount of adsorbed H_4SiO_4 generally increased from 4 to 7 days after incubation but decreased at 30 days after incubation. The Si adsorption data of the six soils tend to follow the Freundlich isotherm with coefficient of determination (r^2) values ranging from 0.79 to 0.92. The H_4SiO_4 concentration in solution for the succeeding sampling times will be reported later. Based on our initial results, the changes on H_4SiO_4 in solution due to added Si varied between soils and were not related to soil pH only.

4

Pedogenic Silica in two croplands of the Belgian Loam Belt

Barão L¹, Vandevenne F¹, Ronchi B², Gerard G², Meire P¹, Struyf E¹

¹ University of Antwerp, Department of Biology, Universiteitsplein 1C, 2610 Wilrijk, BELGIUM (lucia.barao@uantwerpen.be)

² KULeuven, Department of Earth and Environmental Sciences, Celestijnenlaan 200 E, 3001 Leuven. BELGIUM

Silicon (Si) is gaining importance in agriculture since the majority of the most produced crops worldwide are Si accumulators. Several benefits can be associated with Si incorporation by crops such as the increased resistance to insects, alleviation of drought, alleviation of salt stress or the improvement of K and Ca uptake. When crops take up Si it is deposited in the cell walls in the form of phytoliths (biogenic Si – BSi). Due to efficient harvesting, BSi normally never returns to soil creating a loop in the Si cycle and a constant decrease in Si soil concentrations. The BSi presence in the top soil is crucial because phytoliths are one order of magnitude more soluble than silicate minerals and therefore dissolve faster in shorter time scales. However, other non-BSi fractions are also present in soils. Pedogenic processes in mineral horizons can originate pedogenic opal silica, Si adsorbed to Al and Fe hydro/oxides, allophane and imogolite or Si-Al amorphous precipitates. At present, less in known about the size and importance of the pedogenic Si pool (PSi) in the soils, especially in systems where BSi is decreasing over time.

We collected two soil profiles ($\sim 150 - 200$ cm total) from two croplands located in the Belgian Loam Belt (Luvisol with eolian loess as parent material). As a reference system, two soil profiles from two deciduous forests from the same location where also collected. A continuous analysis in 0.5 M NaOH of Si and Al was performed in samples from different horizons to separate BSi and PSi based on the Si:Al ratios of the different fractions present. An extraction with CaCl₂ on the same samples was performed to evaluate the amount of readily soluble silica (Si_{CaCl2}) as a proxy for the available Si in the pore water.

The results showed that both croplands are almost depleted in BSi $(7 - 11 \text{ ton } ha^{-1})$ compared to the reference forests (21-23 ton ha^{-1}) while the PSi pool is significant (121 – 128 ton ha^{-1}) especially in the deeper layers where it becomes dominant. Additionally, the CaCl₂ extraction showed that the PSi pool can be as important as the BSi in the short term in the agro-systems analysed: Si_{CaCl2} is constant through the whole profile (0.4 - 0.8 % of the BSi + PSi) and higher compared to the organic horizons in the forests (<0.4 % of the BSi + PSi).

Theme 6 POSTER

5

Diversity of insect fauna associated with rice fertilized with silicon and bioagents

Batista F V T¹, Silva B G¹, Rêgo C F M¹, Filippi M C²

¹ Amazonia Federal Rural University, Avenue Presidente Tancredo Neves, Belém, 66077-830, BRAZIL (telma.batista@ufra.edu.br)

² EMBRAPA Rice and Beans, Highway GO-462, Santo Antônio de Goiás, 74800-000, BRAZIL

Silicon fertilization is a technique with the potential to decrease the frequency of insect pests in grass, and when in combination with bioagents, besides suppressing diseases also acts as a growth promoter. The aim was to study the diversity of insect pests and predators, in rice fields fertilized with calcium silicate and magnesium and treated with bioagents, by abundance, dominance and diversity of orders and the functional groups present.

The experiment was conducted at BIC, with 4 replicates and factorial arrangement with sub sub plots. The main treatments consisted of 4 calcium silicate and magnesium fertilization doses (1, 2, 4 and 8 t ha-1.) and control; sub plots represents the residual and new silicon fertilization effect; the sub sub plots consisted in treated and untreated seeds with bioagent. Scanning the network 45 days after sowing collected samples, and all the insects sorted into orders and families. Jaccard similarity index (0.05) was calculated with software Past 2:07.

It was detected 5 groups of diversity orders. The groups formed by treatments 1, 2 t ha⁻¹ and control presented a higher insect diversity, probably due to plants biomass. The orders Diptera (53.77%) and Hemiptera (17.04%) were the most abundant and dominant once, as it were present in all treatments, however, the number of individuals decreased in the sub sub plots treated with bioagent. The major agricultural importance functional group found was the herbivores (22%), however, with lower incidence (37%) in the plots fertilized with 1 t ha⁻¹ of calcium silicate and magnesium, and in the sub sub plots treated with bioagent, indicating to be the best combination for this group control.

Funded by CNPQ/Fapespa

Theme 6 **KEYNOTE**

6

Silicon influence on biotic stress in plants

Bélanger R R¹, Vivancos J¹, Wilkinson J-A¹, Belzile F¹, Menzies J G²

¹ Department of Plant Science, Faculty of Agriculture and Food Science, Laval University, Quebec, G1V 0A6, CANADA (richard.belanger@fsaa.ulaval.ca) ² Agriculture and Agri-Food Canada, 101 Route 100, Morden, MB, R6M 1Y5, CANADA

The notion that silicon (Si) feeding offers plant protection against biotic stresses in general, and fungal pathogens in particular, has been convincingly demonstrated. In spite of the many scientific reports to that effect, the properties, spectrum of efficacy and mode of action of Si remain largely speculative. These controversies have hampered an optimal exploitation of Si in agriculture and have raised questions about the true benefits of Si. It is therefore essential that current and future research endeavors take advantage of the latest developments toward defining a universal understanding on how Si can best be integrated into sustainable management practices.

The latest molecular tools in genomics and high throughput sequencing have greatly assisted in Si research. For instance, with the discovery of Si transporters, they have moved our definition of Si absorption in plants from an empirical to a scientific classification. As a result, it is now possible to properly associate Si feeding with plant species, and even particular genotypes within a species, a situation that is likely to improve the benefits of Si. One can not understate the importance of understanding and maximizing Si absorption by plants as it is directly linked to its efficacy. It is therefore not surprising that species such as rice, wheat, cucumber, known as active Si absorbers, have now been precisely characterized for their presence of Si transporters and are also the most commonly associated with the prophylactic effects of Si. Another interesting association is the seemingly stronger efficacy of Si against biotrophic and hemibiotrophic pathogens (e.g. rice blast, powdery mildews) compared to necrotrophs. This observation revives the debate concerning the mode of action of Si in planta, and the balance between passive and active role. There is overwhelming evidence that priming and induced resistance are contiguous to Si feeding in protected plants. but no clear proof that Si is biochemically involved in the process. A better understanding of this phenomenon would certainly help in defining the plant-pathogen interactions that are the most amenable to be affected by Si. As stated earlier, new tools in genomics allow exploiting biological material that was hitherto unavailable for the study of Si mode of action. Recent use of such material has proven useful in highlighting specific properties of Si that could lead to optimizing Si prophylactic role against biotic stresses.

Theme 5 ORAL

7

Ionome and Si transporters affected by zinc and silicon interaction in maize

Bokor B¹, Bokorová S², Ondoš S³, Švubová R¹, Lukačová Z¹, Szemes T², Lux A¹

¹Comenius University in Bratislava, Faculty of Natural Sciences, Department of Plant Physiology, Mlynská dolina B2, Bratislava, 84215, SLOVAKIA (boris.bokor@gmail.com)
²Comenius University in Bratislava, Faculty of Natural Sciences, Department of Molecular Biology, Mlynská dolina B2, Bratislava, 84215, SLOVAKIA

³Comenius University in Bratislava, Faculty of Natural Sciences, Department of Human Geography and Demography, Mlynská dolina B2, Bratislava, 84215, SLOVAKIA

Zinc is an essential microelement involved in various physiological processes. Zinc excess in soil represents serious problem for plants resulting in Zn toxicity symptoms and decreasing biomass production. An interesting question about zinc toxicity is how to mitigate this stress in crops. We investigated the possibility of amelioration of maize stress resulted from elevated zinc concentration in cultivation medium by silicon.

Maize plants (hybrid Novania) were hydroponically cultivated 10 days in Hoagland solution in a controlled environment chamber. Various treatments were compared – control (C), Si (5 mM concentration of sodium silicate solution), Zn (800 μ M ZnSO₄ x 7H₂O) and Zn + Si (combination of Zn and Si). The expression level of *ZmLsi* genes was performed by real-time PCR method (ABI Fast 7900 HT). The root and shoot ionome was evaluated by multiple regression analysis and principal component analysis (PCA).

Growth of young maize plants was significantly inhibited in the presence of high Zn concentration in cultivation media. Silicon addition to Zn treated plants (Zn + Si treatment) was not protective and resulted in even more reduced growth parameters. The root ionome of Zn treated plants was significantly altered. High Zn application and also zinc and silicon interaction induced great imbalance of the root ionome and elemental composition. The shoot ionome of Zn and Zn + Si treated plants was less affected than the root ionome. Silicon applied alone (Si treatment) caused minimal effect on root and shoot ionome. Expression levels of ZmLsi1 and ZmLsi2 genes were downregulated in roots of Si treated plants. High Zn concentration caused decrease in expression level of these genes in roots of Zn and Zn + Si treatment. On the other hand, expression level of *ZmLsi6* was up-regulated in the presence of high Zn in the 1st leaf of maize plants in Zn and Zn + Si treatments in comparison to control treatment. In the 2nd leaf, Si and Zn + Si treated plants showed higher expression level of *ZmLsi6* gene, when compared with control. In conclusion, high Zn and Zn/Si interaction caused imbalance and alteration in ionome homeostasis and also changed the expression of Si transporter genes which resulted in increased physiological stress.

Theme 8 POSTER

8

Evaluation of Armurox® (complex of peptides with soluble silicon) on mechanical and biotic stresses in gramineae

Botta A¹, Rodrigues F A², Sierras N¹, Marín C¹, Cerdà J M¹, Brossa R¹

¹ Bioiberica, S.A. Research Department, Plant Physiology Division, Palafolls, 08389 Barcelona, SPAIN (abotta@bioiberica.com)

² Viçosa Federal University (UFV), Department of Plant Pathology, Laboratory of Host-Parasite Interaction, 36570-000 Viçosa (MG), BRAZIL

Silicon (Si) has proven its effectiveness in controlling both soil-born and foliar fungal diseases in cucumber, rice, sugarcane, turf, wheat and several other crops. It is absorbed as silicic acid and it is accumulated as amorphous hydrated silica ((SiO₂) x nH₂O) in the plant body, thus limitation of its soluble forms leads to difficult assimilation by plants. While most plants do not accumulate silicon, silica deposition is one of the important characteristics of the Poaceae family (gramineae), among others. In this family of high agronomic importance its role has been widely studied: it is involved in the mechanical stability of tissues, protection against fungi, insects and herbivores, resistance to drought, facilitation of light interception, and alleviation of problems caused by nutrient deficiency and excess. Silicon importance on crop protection lies on the fact that infection mechanism of most Fungi affecting gramineae is through the cuticle, thus the thickness and resistance of the cuticle is the most important physical barrier that fungi spores must overcome to infect the subjacent tissue. Moreover the structural reinforcement caused by Si deposition is also of agronomical interest for enhanced mechanical stress resistance of this plant family that lacks highly lignified structures. In addition it is also known that certain peptides strengthen gramineae natural defenses. These peptides have been proved to activate and reinforce the metabolic signaling pathways associated to cell signaling and synthesis of antimicrobial compounds (increase in the activity of the PAL enzyme, accumulation of callose and lignin polymers...).

Therefore, in the last 3 years, many researchers and field technicians has been involved in evaluating the effectiveness of Armurox, a product containing soluble silicon and specific peptide compounds that has been developed by Bioiberica, either for foliar spray or soil application.

The treatment with Armurox, decreased blast symptoms on rice, and revealed higher accumulation of Si in comparison with a potassium silicate standard in studies carried out at Viçosa University in Brazil. In several sugar cane trials, performed in Mexico, the treatment with two applications of Armurox showed higher production, stimulated a better stalk sprouting and increased the number of productive stalks, both by foliar and soil application. Recent trials in wheat also revealed positive results in increasing lodging resistance.

Theme 2 POSTER

9

Residual effects of steel slags on the availability of silicon in the soil

Büll T L, Deus C F A, Catojo P R

University of São Paulo, Crop Sciences College 1780 José Barbosa de Barros Street, P.O.Box 237, Zip Code 18610-307, Botucatu-SP, BRAZIL (bull@fca.unesp.br)

Brazilian soils are characterized by high acidity requiring proper correction for crop production. Some studies point to steel slags, by-products of the steel industry, as promising materials for soil acidity correction, increasing P and Si contents in the soil. This study investigated the release of Si by steel slags used as correctives of soil acidity in three incubation periods.

Trails consisted of three incubation periods: 2; 12 and 23 months with lime (L) and five silicate sources: steel slag (SS), Blast furnace slag (BFS), ladle furnace slag (LFS), stainless steel slag (SSS), wollastonite (W) and the control without corrective application. The dose of each material applied was calculated to raise the base saturation to 70. The Si contents obtained with lime and silicates application were 0.15; 1.3; 5.2; 1.6; 1.0; 1.2; Mg ha⁻¹ with L; SS; BFS; LFS; SSS and W, respectively. The Si content and pH in the soil were analyzed 2, 12 and 23 months after application, at 0-5; 5-10; 10-20; 20-40 cm. Bean was sown two months after material application and soybean was sown 12 and 23 months after material application.

The Si contents were higher in surface layers, but there was an increase at 20-40 cm, showing that Si moved in the soil. The materials favored the Si release, which was absorbed by the bean plants leading to the decrease observed at 12 months, however, Si started to be released again after 23 months. On the other hand, the surface application of materials provided constant increase of Si in the soil during the trails. The increase of Si contents in the soil varied with the source used. SS, LFS, and SSS were more efficient in releasing Si to the soil. Because the dose calculation was based on the increase of bases saturation to correct soil acidity, the Si doses released by each material varied. BFS was applied at a greater dose of Si, 5.2 Mg ha⁻¹, however, because it was a coarser material, it delayed to start Si release. BFS showed more pronounced effects at 23 months. Lime also increased Si contents in the soil due to the pH increase, which provided the release of existing Si in the soil. W, SS, and BFS provided the largest increases of Si in shoots of the bean plant, however, for sovbean, increased Si contents were observed in the second culture cycle, with the application of SS, and LFS. The materials increased grain production of bean and soybean in the second cycle, however, with no significant difference between the sources used. Steel slags can be used in agriculture as correctives of soil acidity, under these conditions, as they increase the soil pH and promote increased productivity of bean and soybean, similarly to lime that is usually used as corrective in Brazilian agriculture, however, steel slags have the differential to release Si into the soil. The slags showed residual effect on Si in the soil 23 months after application.

Theme 2 ORAL

10

Silicate Fertilization on Tropical Soils: silicon availability, uptake and recovery by two sugarcane cultivars

<u>Camargo M S¹, Korndörfer G H², Rocha G³</u>

 ¹ São Paulo Agency for Agribusiness Technology (APTA), P.O. Box 28, Piracicaba, 13412-050, BRAZIL (mscamarg@yahoo.com.br)
 ² Federal University of Uberlândia, P.O. box, 593, Uberlândia, 38400-902, BRAZIL

³ Faculdade de Tecnologia, 13414-155, Piracicaba, BRAZIL

Several classes of soils in Brazil have been used for sugarcane cultivation. New potential producers are located at highly weathered soils, characterized by low soluble silicon contents. Although the benefits of silicon fertilization for sugarcane have already been demonstrated, only a few studies have focused on the effects of silicate fertilization applied in the furrow at planting under field conditions. The objective was to evaluate the effects of a silicate applied in the furrow at planting on Si availability, Si uptake and Si recovery index (RI) by two sugarcane cultivars after plant cane and first ratio crops of sugarcane.

The study was conducted during the plant cane and first ratoon on a low silicon (Si) soil (Rhodic Hapludox-RH) located at Piracicaba, São Paulo state, Brazil. It was conducted in a completely randomized factorial scheme with four replicates, four Si application rates (0, 55, 110 and 165 kg ha⁻¹ Si) and two sugarcane cultivars (IAC 87-3396 and SP 89-1115). The source of silicon was a Ca-Mg silicate, which was applied in the furrow at planting such that all plots received the same quantity of Ca and Mg. The soil was fertilized based upon the soil analyses with N, P and K. Silicon concentration were determined on top-visible dewlap (TVD) leaves at eight months, on straw (old and new leaves + tips) and stalks after both harvest. Soluble Si in acetic acid (0.5 mol L⁻¹) and CaCl₂ (0.01 mol L⁻¹) were determined in soil samples (0-25 cm in depth) collected after plant cane and first ratoon harvest. The Si uptake by sugarcane from silicate (SiFF) was calculated using the following equation: SiFF = Si uptake by the control (rate zero). The percentage of the recovery index of Si (RI) by the two cultivars with Si application was also calculated using RI (%) = (Si uptake by sugarcane from silicate/Si applied) *100.

Silicate applied in the furrows at planting increased linearly extracted Si (using two different extractants) in soil samples, Si concentration in the leaves, and Si uptake for both the plant cane and the first ratoon, showing residual effects. Cultivars showed differences on Si uptake. The average of recovery index of applied silicon (RI) in plant cane was 30.7 and 41.7 % for IAC 87 3396 and SP89 1115, respectively. In the first ratoon crop, the values 5.9 and 10.2 % for IAC 87 3396 and SP89 1115, respectively. Considering these results, it can be concluded that the practice of silicate placement in the furrow at planting in low rates (< 200 kg ha⁻¹ Si) can increase Si availability, uptake in sugarcane with residual effects. Additionally, the recovery index of applied silicon (RI) was less than 50% and it was dependable of cultivar.

Т	heme	3
Ρ	OSTE	R

11

Gene expression of superoxide dismutase isoforms and RUBISCO in *Lolium perenne* in response to silicon supply

<u>Cartes P^{1,2}</u>, Jiménez H³, Pontigo S⁴, Ribera A^{2,3}

¹Department of Chemical Science and Natural Resources, La Frontera University, Temuco, 4811230, CHILE (paula.cartes@ufrontera.cl)

²Scientific and Technological Bioresource Nucleus (BIOREN), La Frontera University, Temuco, 4811230, CHILE

³Department of Agricultural and Livestock Production, La Frontera University, Temuco, 4811230, CHILE

⁴Doctoral Program in Science of Natural Resources, La Frontera University, Temuco, 4811230, CHILE

Silicon (Si) is recognized as a beneficial element for vascular plants due to it increase the tolerance to different biotic and abiotic stresses. Some reports showed that Si is able to counteract oxidative damage triggered by aluminium (Al) toxicity, which is one of the main factors that limit the growth of forage species like ryegrass in acid soils of Southern of Chile. Despite of the advances in research on this subject, there are few studies about the Si effect on the antioxidant system as well as on the photosynthetic performance of plants. The aim of our study was to evaluate the gene expression of superoxide dismutase (SOD) isoforms (Cu/Zn-SOD, Mn-SOD and Fe-SOD) and ribulose-1,5- biphosphate carboxilase (RUBISCO) in ryegrass (*Lolium perenne* cv. Jumbo) grown at different Si doses.

During 21 days, plants were hydroponically cultivated at increasing Si supply $(0, 2, 5 \text{ or } 10 \text{ mM}; \text{ applied as Na}_2\text{SiO}_3)$ at pH 6.0. At the end of the assay, dry weight (DW) and Si concentration of shoot and roots were determined. The relative gene expression of SOD isoforms and RUBISCO were also measured by real-time quantitative PCR (qRT-PCR).

According with the results, Si concentration in shoots and roots was progressively raised at increasing Si supply, and no difference in ryegrass yield was observed irrespective of the Si treatment. SOD isoforms were differentially expressed in shoots and roots, and the expression pattern was similar among isoforms in each plant organ. Even though Si supplies up to 5 mM down-regulated the gene expression of SODs in the shoots, we found an overexpression of these isoforms in the roots of plants growing at 2 mM Si. Interestingly, Si addition up to 10 mM did not greatly affect the gene expression of RUBISCO, which denotes that Si seems not affect the photosynthetic performance of ryegrass plants.

Acknowledgements: FONDECYT Project 1120901.

Theme 8 ORAL

12

Impact of silicon based fertilizer OPTYSIL on abiotic stress reduction and yield improvement in field crops.

Ciecierski W, Kardasz H

Intermag Sp. z o. o., Osiek 174 A, Olkusz, 32-300, POLAND (wieslaw.ciecierski@intermag.pl)

Silicon is the second most abundant element in the Earth crust. Its beneficial role in plant growth was discovered years ago but mostly using soil application of solid compounds. INTERMAG R&D team developed technology of production liquid product for foliar application. The aim of this work was to evaluate impact of liquid silicon application on leaf on plant growth.

The effect of the application of liquid silicon based fertilizer OPTYSIL (Si) on drought stress reduction was conducted in laboratory on wheat and on small plots field trials on soya. The trials were conducted in Department of Plant Physiology of Polish Academy of Science. The yield quality and quantity was checked on winter wheat (*Triticum aestivum* L.) and winter oilseed rape (*Brasssica napus* L.) in Experimental Stations of Research Center of Cultivar Testing located in four different parts of Poland.

The effect of OPTYSIL application on drought stress reduction resulted in very promising effects. The number of pods on soya increased by 18% and average seed weight per plant increased by 21%. Laboratory tests on wheat in stimulated drought condition showed lower electrolyte leakage by 41% and in the same time protein production increased by 40%. A field trials showed yield increase by 8% to 20.5% on winter wheat, depend on location and variety. At the same time increase of thousand-grain weight (TGW) was increased from 1.6 to 7%. The yield of oilseed rape was increased by 1.7 to 17% depend on variety and location. TGW was also modified by OPTYSIL and showed increase by 1.4 to 19%. The plots were located on well-fertilized soil, spatially prepared for each crop. The field trials were conducted already second year in a row and results confirmed the positive effect of Si based fertilizer on yield. However, the experiments have shown that the more severe stress, the higher yield increase can be observed

Theme 2 ORAL

13

Mineral Si fertilisation in forests – what can be learned from agro-systems?

<u>Clymans W¹</u>, Conley D J¹, Buso D C², Johnson C E³, Likens G E²

¹ Department of Geology, Lund University, Lund, 223 62, SWEDEN (wim.clymans@geol.lu.se)

² Cary Institute of Ecosystem Studies, Millbrook (NY), 12545, USA

³ Department of Civil and Environmental Engineering, Syracuse University, Syracuse (NY), 13244, USA

Long-term studies at the Hubbard Brook Experimental Forest (HBEF, USA) have shown that acid deposition has led to reduced growth rates in natural forest stands. Mineral wollastonite (CaSiO₃) was added in autumn 1999 to an 11.8-haforested watershed to ameliorate soil acidification. Subsequently, soil and stream water became less acidic, Al toxicity decreased, the health of forest stands improved and the water balance was altered. These effects have mainly been attributed to Ca effects, while the potential importance of Si has been neglected. This is surprising as we know from agronomic research that Si has positive biotic and abiotic effects controlling nutrient exchange and water regulation in crops. Unfortunately, similar knowledge is lacking for tree species. We aim to study the potential importance of Si following its addition to forest stands through: (1) its role on nutrient balances in soils; and (2) investigating Si uptake in trees after Si fertilization.

Several plots were installed at the HBEF to differentiate the response of terrestrial ecosystems to experimental additions of wollastonite, Aerosil (SiO₂) and sodium metasilicate (Na₂SiO₃). The latter treatments were used in an attempt to separate Ca and Si effects. Pore water samples within different soil horizons were analysed for geochemistry. In a separate study we determined the Si content in archived sugar maple, beech and birch leaves from the wollastonite-treated watershed, collected before and after the 1999 addition.

Our experimental plots showed increased pH after wollastonite and sodium metasilicate additions and Al was immobilized in the Aerosil- and wollastonite-treated plots. Similar positive effects on the nutrient balance, independent of treatment, challenge the hypothesis of a Ca-specific effect. However, visible degradation of the forest floor occurred, with increased leaching of cations, Al and DOC, after sodium metasilicate application, indicating its potentially destructive effects on acidic forest soils. This contrasts with the traditional use of sodium metasilicate as a Si fertiliser in less acidic agriculture fields. The time-series of leaf Si content in the watershed with wollastonite application showed an increased Si uptake ($\pm 2 - 3$ times) for all tree species, followed by a decrease concomitant with major changes in biomass production, plant health and hydrological changes. There is a clear need to create a sound scientific basis for the beneficial effects of Si additions across various terrestrial ecosystems. Knowledge acquired from agronomic studies may be useful in investigating the beneficial effects of Si additions to forested ecosystems. Further experiments and monitoring will advance our understanding of the biotic and abiotic effects of Si on nutrient balances, tree health and water balance regulation in forests.

Theme 1 ORAL 14

Emerging understanding of anthropogenic impact on the ecosystem silica filter

Conley D J¹, Struyf E²

 ¹ Department of Geology, Lund University, 22362 Lund, SWEDEN (daniel.conley@geol.lu.se)
 ² Department of Biology, University of Antwerp, 2610 Wilrijk, BELGIUM

The annual fixation of dissolved silica (DSi) into terrestrial vegetation ranges from 60 to 200 Tmole, 10–40 times more than the yearly export of DSi from the terrestrial geobiosphere to the coastal zone, and 3-6 times more than annual weathering of silicates. Ecosystems form a large filter between mobilization of DSi by silicate weathering and transport by rivers. A large reservoir of biogenic amorphous Si (mostly plant phytoliths) and pedogenically reworked amorphous Si (ASi) accumulates in soils. Although ASi is substantially more soluble than mineral Si, a major challenge is presented by the difficulty to separate pedogenic and biogenic ASi phases in the soil. Here, we discuss the key controls on the ability of ecosystems to filter and control the export of DSi.

We will utilize literature data to show that land use is the most important controlling factor of Si mobilization with changes in land use causing abrupt shifts in the biogeochemical Si cycle in terrestrial ecosystems.

Land clearance and the development of agriculture may result in an enhanced flux of DSi coupled with enhanced erosion losses of ASi contained in phytoliths. In temperate watersheds with sustained cultivation there is however a two- to three-fold decrease in baseflow DSi delivery. Our results also show that turnover rates of ASi in temperate cultivated soils are strongly reduced compared to forests, and stocks of pedogenic ASi are depleted. Human harvest of crop ASi has created a parallel anthropogenic Si cycle, which has received little attention to date. It shows that anthropogenic impact and agriculture should be recognized as an important controlling factor of terrestrial Si fluxes.

Theme 8 ORAL

15

Silicon in Agriculture New Developments in Latin America

Cristancho R J A, Restrepo F

Department of Research Mejisulfatos, Carrera 41 No. 46-114, Medellín, +57, COLOMBIA (jose.cristancho@mejisulfatos.com)

Most of the soils in tropical America are highly weathered soils. These soils have been used for growing rice, sugarcane, banana, coffee and oil palm. These soils are high in aluminum (Al) concentrations, with low phosphorous (P), magnesium (Mg), potassium (K) and mono-silicic acid availability. The low fertility and Al toxicity have been associated with pest and disease susceptibility and low yield. In Brazil and Colombia there are some sources like Ca and Mg silicates that have been proved to be effective in correcting surface and subsoil acidity. They have proven to be effective increasing silicon, calcium and magnesium content in the soil and in the plant tissue. The purpose of this paper is to summarize the progress on silicon to improve soil fertility, growth and suppress plant disease in oil palm crop.

The treatments tested were two sources of silicon (magnesium silicate and acidulated magnesium silicate) and two rates (150 and 300 kg SiO₂/ha) arranged in a randomized complete block design with experimental unit of 24 palms (4 row \times 6 palms in each row = 24 palms), with 8 recording palms. The various parameters recorded in this experiment were growth measurements, leaf and soil analysis, bud rot incidences and recovery. The experiment was run in a nursery and young oil palm stages.

The results showed that the acidulated magnesium silicate application increased P, Mg, B and Si uptake. The treatments improved the leaf area index and petiole cross section. The treated plants accelerated plant recovery from bud rot disease attack. Treatments with acidulated magnesium silicate showed a healthier growth and a higher yield. The early studies have shown that silicon applications had significant potential to improve plant nutrient efficiencies, improve plant disease recovery and to increase yields and business sustainability.

Theme 6 ORAL

16

Hormonal regulation of silicon-induced brown spot resistance in rice

<u>De Vleesschauwer D</u>¹, Kikuchi S², Höfte M¹, Van Bockhaven J¹

¹ Lab of Phytopathology, Ghent University, Coupure Links 653, Ghent, 9000, BELGIUM (david.devleesschauwer@ugent.be)

² Plant Genome Research Unit, National Institute of Agrobiological Sciences Tsukuba 305, Ibaraki, 8602, JAPAN

Over the past decades, various studies have shown the ability of silicon to mitigate a wide variety of abiotic and biotic stresses. However, despite this relative wealth of knowledge, much remains to be discovered about the mechanistic basis and regulation of Si-afforded stress protection. Aiming to shed further light onto the prophylactic effect of Si, we have investigated the role of hormone defense pathways in governing silicon-induced resistance to the rice brown spot pathogen *Cochliobolus miyabeanus* (Si-BSR).

To delineate the involvement of multiple hormone pathways, we have pursued a multidisciplinary approach combining exogenous hormone applications, pharmacological inhibitor experiments, time-resolved hormone measurements, and bioassays with hormone-deficient and/or –insensitive mutant lines.

Contrary to many other types of induced plant resistance, we found that Si-BSR functions independently of the archetypal stress hormones salicylic acid (SA), jasmonic acid (JA) and abscisic acid (ABA). Similarly, our data rule out a major involvement of the developmental hormones gibberellic acid (GA), auxin and cytokinin (CK). In contrast, several lines of evidence suggest that Si steers its positive effect on C. mivabeanus resistance through negative crosstalk with the rice ET pathway. Consistent with ET functioning as a virulence factor of C. miyabeanus, exogenous ET application increased susceptibility, whereas genetic and pharmacological disruption of ET signalling rendered plants less vulnerable to brown spot, thereby inducing a level of resistance similar to that observed on Si-treated wild-type plants. Moreover, ET emission levels and transcript levels of the ET-responsive marker gene OsEBP89 were markedly lower following Si application. Moreover, Si failed to further increase the already high levels of resistance observed in ET-insensitive rice lines, suggesting that Si triggers brown spot resistance by preventing the fungus from hijacking the host ET machinery. Interestingly, rather than antagonizing rice ET signalling per se, Si likely interferes either directly or indirectly with production of ET by C. miyabeanus. In conclusion, our findings favour a scenario whereby Si induces rice brown spot resistance by disarming fungal ET and argue that impairment of pathogen virulence factors is a core resistance mechanism underpinning Si-induced plant immunity.

Theme 3 ORAL

17

Characterization of silica accumulation in maize cell culture

Elbaum R¹, Nissan H², Menachem M²

 ¹ RH Smith Institute of Plant Sciences and Genetics in Agriculture, Faculty of Agriculture, POB 12 Rehovot, 7610001, ISRAEL (Rivka.elbaum@mail.huij.ac.il)
 ² RH Smith Institute of Plant Sciences and Genetics in Agriculture, Faculty of Agriculture, POB 12 Rehovot, 7610001, ISRAEL

Silica is accumulated in large quantities by grasses. The plants absorb silica in the form of silicic acid through dedicated root transporters. The silicic acid is then transported to the shoot via the transpiration stream, and distributed in the leaf tissue. However, very little is known on the absorption and accumulation of silica by cells embedded in a tissue.

In order to unravel cellular mechanisms that stand at the basis of plant bio-silicification we chose to expose maize cell culture, black Mexican sweet (BMS) to two levels of silica (0.5 ppm, and 50 ppm), and follow the silicification processes using light and electron microscopy, Raman micro-spectroscopy, and thermo-gravimetric and chemical analysis.

The two silicon levels did not change the development of the cells. However, the lowsilicon treatment showed significant increase in the expression of all three silicon transporters Low Silicon 1 (LSi1), LSi2, and LSi6. This sample had very low silicon content, 0.03 mg per gram dry weight tissue. The high-silicon treatment cells contained six times more silicon, where 25% was bound at the cell wall. We found that silicon binds spontaneously to cell walls, and increases their thermal stability, independent of their synthesis. Using Raman micro-spectroscopy we detected increased content of proteins in cell walls synthesized under high silicon treatment, with no other effect on the cell wall composition. The silicon in the cytoplasm was equally divided between free silicic acid form and bound silicon oxide. However, all absorbed silicon was rinsed after one day exposure to low silicon treatment. Transmission electron microscopy showed that the silicon accumulates cell walls, in vesicles, possibly as part of endocytosis processes, and around starch granules. Our results indicate that cells absorb silicic acid from the apoplasm through endocytosis, as well as through dedicated transporters. Once the levels of transporters is low, the silicic acid leaks from the cells. The silicic acid incorporates into the cell wall polymers and stabilizes them. It is accumulated in vesicles and other cell organelles. Our next aim is to explore how the silicic acid alters the transcriptome of the cells, and to test whether the cell culture treated with high silicon concentration has an advantage under stress.

Theme 5 POSTER

18

Silicon-mediated oxidative stress tolerance and genetic variability in rice (*oryza sativa* L.) grown under combined stress of salinity and boron toxicity

Farooq M A^{1,2}, Dietz K-J¹

² Institute of Soil & Environmental Sciences, University of Agriculture, Faisalabad-38040, PAKISTAN

Benefits of silicon (Si) in improving crop fitness by biotic and abiotic stress resistance are widely reported. However, investigations about its protective mechanisms for plants facing multiple stresses are very limited.

Two contrasting rice cultivars; KS-282 (salt-tolerant) and IRRI-6 (salt sensitive) were grown in a pot experiment to study the inter-relation between Si supplementation (0 and 150 mg kg⁻¹) and boron (B) toxicity (0 and 2.5 mg kg⁻¹) under salinity stress with emphasis on growth response, mineral contents, physiology and enzymatic antioxidant system response.

Results revealed that adverse growth conditions particularly combined stress of salinity and B toxicity severely affected physiological attributes of rice. It reduced plant biomass by damaging the membrane, reducing SPAD values and photosynthetic efficiency but Si application counteracted the adverse effects of stress by reducing uptake of toxic ions such as sodium (Na⁺) and B, lowering transpiration rate, increased relative water contents and photosynthetic efficiency due to more Si and K⁺ uptake ultimately better growth performance. Si significantly affected activities of enzymatic antioxidants in both genotypes, with increased ascorbate peroxidase (APX), guaiacol peroxidase (GPX) and reduced catalase (CAT) activity suggesting relieved stress by reduced oxidative damage. The response to stress and Si differed genotypically, with maximum damage to salt sensitive genotype (IRRI-6) particularly under combined stress of salinity and B toxicity. Conclusively, these results support the protective role of Si in the regulation of salinity and/or B toxicity stress by improving growth, K⁺/Na⁺ ratio, physiology and antioxidant capacity; suggesting it as a potential candidate for crops grown under such deteriorated soil conditions. However, field trails should be carried out before setting any recommendation for farmers.

¹Biochemistry and Physiology of Plants, Bielefeld University, D-33501 Bielefeld, GERMANY (ansar_1264@yahoo.com)

Theme 5 POSTER

19

Influence of silicon on castor beans plants under aluminium stress

Fernandes D M¹, Freitas L B¹, Moniz A¹, Maia S C M²

¹ Department of Soil Sciences and Environmental Resources, 1780 José Barbosa de

Barros, Botucatu-SP, 18610-330, BRAZIL (dmferandes@fca.unesp.br)

² Department of Crop Science, 1780 José Barbosa de Barros, Botucatu-SP, 18610-330, BRAZIL

Among the benefits of silicon (Si) for plants, the mitigation of abiotic stress has been gaining importance recently. However this effect is not well known especially in Si non-accumulator plants like castor beans. The majority of research on this role of silicon had been done on Si accumulators like rice and sugarcane, and therefore the aim of this study was to evaluate the influence of Si on a non-Si -accumulator plant (castor bean) grown under aluminum (Al^{3+}) stress.

The experiment was conducted in nutrient solution and was laid out in a 2x4 randomized factorial block design, with five replications. The treatments included two castor bean cultivars, CRZ 1 (Al³⁺ susceptible); CRZ 6 (Al³⁺ tolerant) and four combinations of Si and Al³⁺; 1) 0 Si and 0 Al³⁺; 2) 1.7 mmol L⁻¹ Si and 0 Al³⁺; 3) 1.7 mmol L⁻¹ Si and 1.1 mmol L⁻¹ Al³⁺; 4) 0 Si and 1.1 mmol L⁻¹ Al³⁺.

The treatment without Si and Al^{3+} ; and Si alone showed maximum shoot dry weight. Si supplementation was not seen to be contributing towards improving the root dry weight. The combinations without Al^{3+} showed better plant growth. Number of leaves showed the same trend in results as of the dry weight, with the best treatments being the one without Si and Al^{3+} and the one with Si alone. Stem diameter and shoot length did not show differences among the treatments. Therefore, Si did not have any mitigation effect on the Al^{3+} stress in castor beans, which is a Si non-accumulator. Future research might shed some light on the subject of as to why there is no Al^{3+} stress mitigating effect of Si on non-accumulators plants and only on accumulator plants as rice.

Theme 5 POSTER 20

The effect of silicon on the antioxidative response of maize young plants to salinity and zinc stress

Fialová I, Šimková L, Vaculíková M, Sedláková B, Luxová M

Institute of Botany SAS, Dúbravská cesta 9, Bratislava, 845 23, SLOVAKIA (ivfiala@gmail.com)

Salinity stress is an increasing environmental problem for crop production worldwide. Zinc is an essential component of thousands of proteins in plants, although it is toxic in excess. Silicon is the second most prevalent element in the earth crust and has beneficial effects in enhancing the tolerance of plants to biotic and abiotic stresses. The aims of our work were to study antioxidative responses as the activities of antioxidative enzymes catalase (CAT) and peroxidase (POD), lipid peroxidation by the content of the oxidation stress marker MDA, content of antioxidant ascorbate (AsA) and osmoprotectant proline. The isoenzyme pattern of POD was also determined.

The individuals of *Zea mays* L. (hybrid Novania) were grown under controlled conditions: light/dark 16/8 hours, temperature 24°C/22°C, relative humidity 60% and light intensity 300 μ mol m⁻² sec ⁻¹ PAR. The 3day-old seedlings were transferred to Hoagland nutrient solution treated by Si or/and NaCl and Zn as follows: 0 – control (1), 50 μ M ZnSO₄ (2), 500 μ M ZnSO₄ (3), 2.5 mM Si (4), 50 μ M ZnSO₄ + 2.5 mM Si (5), 500 μ M ZnSO₄ + 2.5 mM Si (6), 150 mM NaCl (7), 150 mM NaCl + 50 μ M ZnSO₄ (8), 150 mM NaCl + 500 μ M ZnSO₄ (9), 150 mM NaCl + 2.5 mM Si (10), 150 mM NaCl + 50 μ M ZnSO₄ + 2.5 mM Si (11), 150 mM NaCl + 50 μ M ZnSO₄ + 2.5 mM Si (11), 150 mM NaCl + 50 μ M ZnSO₄ + 2.5 mM Si (12). The pH of nutrient solution was adjusted to 6.2. As the source of silicon sodium silicate solution (27% SiO₂ in 14% NaOH) was used. After 9 days the apical segments of roots, primary and secondary leaves were harvested and stored at -70°C until they were analyzed. Activity of enzymes, content of proline, ascorbate and MDA were determined spectrophotometrically. To detect the isoforms of POD the 10% polyacrylamide gels under native conditions was used.

The changes of CAT and POD activities demonstrated positive influence of Si in the first leaf of the Zn treated plants (5, 6, 11) and in the second leaf of the variants with higher concentration of Zn (6, 12). The MDA value is the most increased in the case of the treatment with 500 μ M Zn (3) in both studied aboveground organs. The changes of MDA level indicated that Si decreased the negative effect of Zn and NaCl on the leaves. As for AsA, the positive effect of Si was apparent in the presence of Zn (5, 6). The proline level showed positive effect of Si at the variant with NaCl (10) and NaCl together with 500 μ M Zn (12) in the older first leaf. The differences in the POD isoenzyme pattern were observed.

This work was supported by the Slovak Research and Development Agency under the contract No. APVV-0140-10 and VEGA project No. 2/0022/13.

Theme 6 ORAL

21

Silicon and bioagents in leaf rice blast suppression

Filippi M C¹, Souza A C A², Pereira S T³, Prabhu S A¹ Silva B G³

- ¹ EMBRAPA Rice and Beans, Highway GO-462, Santo Antônio de Goiás, 74800-000, BRASIL (cristina.filippi@embrapa.br)
- ² Master in Agronomy, Street José Needermeyer, Goiânia, 74360-340, BRAZIL Goiás Federal University, Highway GO-462, Goiânia, 74800-000, BRASIL
- ³ Amazonia Federal Rural University, Avenue Presidente Tancredo Neves, Belém, 66077-830, BRASIL

Rice is considered a cereal that has the potential to eradicate hunger and malnutrition. However, during its cultivation, is submitted to biotic and abiotic stresses, which lowers productivity in key growing regions, followed by the rising cost of inputs such as fertilizers and pesticides. The aim of this work was to study the effect of silicon, alone and in combination with bioagents on leaf blast suppression of on upland rice.

Two experiments, E1 and E2 were conducted in a factorial design at greenhouse conditions. E1: aimed to select the best treatments for rice blast (*Magnaporthe_oryzae*) suppression. It was composed by 5 plots (control; 1 ton SiCaMg ha⁻¹; 2 ton SiCaMg ha⁻¹; 4 ton SiCaMg ha⁻¹; 8 ton SiCaMg ha⁻¹), 5 subplots (control; *Burkholderia pyrrocinia*; *Pseudomonas fluorescens*, *Trichoderma asperellum*; mixture of all these three bioagents) and 8 replication. E2: aimed to investigate the defense mechanisms of the best treatment selected in E1. It was composed of 2 plots (control; 2 ton SiCaMg ha⁻¹) and subplot 3 (control; *Trichoderma asperellum*; mixture of all these three bioagents) and 4 replication.

Silicon fertilization and bioagents interaction proved to be promising. In E1, the combination of all three bioagents with 2 ton ha of SiCaMg plants fertilization was the best treatment reducing 96% of leaf blast. In E2, the two best treatments in E1 statistically increased chitinase (CHI), glucanase (GLU), peroxidase (POX) and phenyl ammonia lyase (PAL) activity as well SA content in the absence of *Magnaporthe oryzae*. However, 24 and 48 hours after challenger inoculation with *M. oryzae*, only CHI and GLU activity and SA content statistically increased in the treatment that combined all three bioagents and 2 tonnes ha⁻¹ of SiCaMg plants fertilization. The reduced leaf blast followed by the active participation of defense mechanisms elect the silicon fertilization and bioagents as important strategies for rice blast sustainable management.

Finacial Support: CNPq/Fapespa

Theme 5 POSTER

22

Effect of silicon on mitigation of aluminum stress in upland rice plants

Freitas L B¹, Fernandes D M¹, Moniz A¹, Maia S C M²

¹ Department of Soil Sciences and Environmental Resources, FCA-UNESP, 1780 José

Barbosa de Barros, Botucatu-SP, 18610-330, BRAZIL (lucasbarbosaf@yahoo.com.br)

² Department of Crop Science, FCA-UNESP, 1780 José Barbosa de Barros, Botucatu-SP, 18610-330, BRAZIL

Although silicon (Si) is not considered to be an essential nutrient for plants, it is known to effectively mitigate various abiotic stresses in plants, such as aluminum (Al^{3+}) stress common in acids soil. However the mechanism of this interaction between Si and Al^{3+} is not well understood, therefore the aim of this work was to elucidate the role of Si in mitigating the Al^{3+} stress in upland rice.

The experiment was conducted in nutrient solutions, and was laid out in randomized blocks, (Factorial design, 2x4) with six replications. The treatments included four combinations of Si and Al^{3+} ; 1) 0 Si and 0 Al^{3+} , 2) 1.7 mmol L^{-1} Si and 0 Al^{3+} , 3) 1.7 mmol L^{-1} Si and 1.4 mmol L^{-1} Al3⁺, 4) 0 Si and 1.4 mmol L^{-1} Al³⁺; imposed on two upland rice cultivars Maravilha (Al³⁺ susceptible); ANA 7007 (Al³⁺ tolerant).

Differences were observed in shoot length, shoot and root dry matter among treatments. The treatments without Al^{3+} showed better growth over all other treatments. However, there was no significant difference between Si applied with Al^{3+} and the Al^{3+} alone treatments in root dry matter production. Therefore, Si application did not improve the root development under Al^{3+} stress. The number of tillers was also not improved by Si supplementation under Al^{3+} stress. But, Si applied with Al^{3+} showed improved shoot length and shoot dry matter production in both cultivars over Al^{3+} alone treated plants. The improved shoot growth can be attributed to the effect of silicon in alleviating Al^{3+} stress. The nuch needed and ongoing research works might bring into light the actual reason behind the alleviation of Al^{3+} stress by silicon supplementation in plants.

Theme 4 POSTER

23

Effect of silicon and phosphorus rates on Si and P content and uptake of rice (*Oryza sativa* L. cv. Tarom) and disease

<u>Ghanbari-Malidarreh A</u>¹, Dastan S²

 ¹ Department of Agronomy and Plant Nutrition, Jouybar Branch, Islamic Azad University, Jouybar, 47715-195, IRAN (aghanbari@jouybariau.ac.ir)
 ² Department of Agricultural Science, Payame Noor University, IRAN.

Silicon and phosphorus are two important elements in rice production areas with frequent cultivation that phosphorus pollution due to excessive use and immobility and the amount of silicon store in the soil because to fire and lack of use silicon fertilizer has been reduced. The aimed was determine the uptake and content two elements and to evaluate use of silicon and phosphorus on yield grain of rice.

Factorial experiment based on randomized complete block design with two factors phosphorus and silica with four replications was conducted. Rice was growth in field without silicon (check) and with silicon (500 and 1000 kg ha⁻¹) in three levels and with three phosphorus levels (0, low; 50 kg ha⁻¹ P medium; and 100 kg ha⁻¹ P high).

The results showed that the maximum plant height, panicle stage, panicle per m² and grain yield were produced with 1000 kg Si ha⁻¹, as in this treatment compare to control grain yield was increase about 23%. Also, the most silicon content and uptake in grain and straw and phosphorous content and uptake in grain and straw were obtained by 1000 kg Si ha⁻¹ treatment. Medium phosphorus use cause to increase panicle length, number of spikelet per panicle, 1000-grain weight and grain yield. Also, this treatment increase phosphorus content of grain. Infected leaf blast percentage, leaf blast diameter, panicle infected blast and grain infected to blast had decrease with 1000 kg Si ha⁻¹, but only panicle infected blast and grain infected to blast with phosphorus application compare to control treatment. The highest grain yield and biological yield were produced at interaction of 1000 kg Si ha⁻¹ and medium phosphorus use. Generally, silicon land use was increase phosphorus uptake, as this element has synergism affect with together that increase that cause to increase Si and P uptake and grain yield in rice, also decrease disease parameters in rice.

Theme 3 ORAL

24

Brachypodium mutants defective in silicon transport as a tool for investigation of silicon effects on cell wall composition

Głazowska SE, Murozuka E, Baldwin L, Willats W G T, Schjoerring J K

Department of Plant and Environmental Sciences, Thorvaldsensvej 40, Copenhagen, 1871 Frederiksberg C, DENMARK (glaz@plen.ku.dk)

Silicon (Si) plays a role as beneficial element in plants by improving their growth and resistance towards biotic and abiotic stresses. Numerous members of the *Poaceae* family are capable of actively transporting silicon and accumulating it at a high level constituting even up to 10% of the plant dry matter. A major part of the Si is embedded in the cell walls. However, there is limited knowledge about the specific binding forms of Si and the functional properties of Si in relation to cell wall structure and composition. On this background, the objective of our research is to understand Si deposition mechanism and interactions with cell wall components in grass species.

We have isolated and characterized *Brachypodium* mutants defective in silicon influx (*bdLsi1-1*). Wild type (wt) plants and mutants were grown in soil until maturity and the elemental content of dry matter, including Si, measured by inductively coupled plasma optical emission spectrometry (ICP-OES). Cell wall composition was determined by comprehensive microarray polymer profiling (CoMPP). This method enables fast mapping of cell wall components by using monoclonal antibodies designed to recognize specific glycan-epitopes. Finally, glucose and xylose release were quantified by HPLC after acid hydrolysis and enzymatic digestion of the straw.

The Si concentration in *bdlsi1-1* mutants was on average 0.37% of the dry matter, which was 80% lower compared to the wt. At the same time, calcium and zinc levels were significantly elevated. Silicon and calcium are deposited in plant cell walls, inferring physical strength. Therefore, in the absence of Si, calcium may take over its role in order to maintain plant rigidity. The CoMPP profile was similar for both mutant and wt, except the signal from antibody LM6 which was higher in the *bdlsi1* mutant. LM6 recognizes $(1\rightarrow 6)$ - α -L-arabinan side chains of RG-I. Results indicate that the density of RG-I units may be higher when plants are lacking Si or that Si deposition may mask the epitopes recognized by the antibody. Enzymatic saccharification efficiency was similar for the wt and *bdlsi1-1* mutant. The release of glucose comprised about 16-20 % and that of xylose 11-14 % of the dry matter. The relatively low concentration of Si in the straw of both the wt and mutant may be part of the reason for lack of Si effects on the saccharification efficiency. Our preliminary data shows that Brachvpodium distachvon bdlsi1-1 mutant is a suitable tool for further studies of Si deposition and interactions with cell wall components, as well as the consequences for enzymatic degradability of cell walls. We are currently investigating these relationships in further details, focusing on soil grown plants with a larger span in cell wall Si concentrations between wt and mutants.

Theme 7 ORAL 25

Si decreases Cd and As in crops — a field study

<u>Greger M</u>, Landberg T

Department of Ecology, Environment and Plant sciences, Stockholm University, Lilla Frescati, Stockholm, 106 91, SWEDEN (maria.greger@su.se)

Cadmium (Cd) and arsenic (As) are toxic elements, which are known to create health problems. Those elements exist in elevated concentrations in alum shale soils. It is common to grow crops in Scandinavia in this type of soil, as it is fertile. Our recent research on wheat shows that the presence of silicon decreases Cd accumulation in grains of wheat grown in nutrient solution. The same result was found for As uptake in lettuce grown in solution culture. The aim of the present study was to find out if this effect by Si, decreasing the content of As and Cd, would be found in field-grown crops when adding silicon to the soil.

Winter potato (Solanum tuberosa), carrots (Dacus carota), yellow onion (Allium cepa) and spring bread wheat (Triticum aestivum) were grown in alum shale soil in Hedmark county, Norway. Silicon was applied as 500 kg Si per ha and different silicon additives were tested: A) liquid potassium silicate from YARA, B) amorphous SiO₂ Microsilica from Elkem and C) powder form of Solaritt (mixture of CaSiO₃, Ca₃Si₂O₇ and CaO with liming effect) from ELKEM. Silicon was applied, one month after sowing. Three months later, at maturity plants were harvested. Edible plant parts were dried, weighed, wet digested and analysed for As, Si and Cd content on AAS.

All plants investigated contained Cd and As. Generally, in all cases, either significantly ($p \le 0.05$), or as tendency, Si addition decreased the accumulation of both As and Cd in the edible plant part. This was the case for all three Si additives and all plant species. Thus, we can conclude that Si addition decreases Cd and As in edible parts of field grown crops. The reason can be that Si influences the 1) release of Cd and As from soil particles, 2) uptake and /or 3) translocation from roots to the edible parts of these elements. Si concentration in the analysed crops increased with up to approx. 0.1 - 0.3% and Si is may improve our health. Thus, Si can be used in biofortification of food.

The authors would like to thank the two companies YARA and ELKEM for providing us with silicon additives and Hedmark University College for funds for the analyses.

Theme 2 ORAL 26

Unravelling the enigma of the effect of soil pH on Si availability

Haynes R J, Zhou Y-F

School of Agriculture and Food Sciences/CRC CARE, The University of Queensland, St Lucia, Brisbane 4072, Queensland, AUSTRALIA (r.haynes1@uq.edu.au)

Although Si is considered as a beneficial rather than essential plant nutrient, economic yield increases have frequently been demonstrated for Si-accumulator plants such as sugarcane and rice. As a result, fertilizer Si applications (often using waste products such as blast and steel furnace slag) to sugarcane and rice are becoming increasingly common. Nevertheless, our understanding of Si fertility in soils is less than adequate. One controversial aspect of the fertility of Si is the effect of soil pH on Si availability. While some workers have shown increasing pH decreases Si availability others have recorded marked increases.

In this paper we use data from the literature plus that from our own laboratory incubation experiments (using Si-deficient soils taken from the Queensland sugar belt) to examine how soil pH can influence Si availability through a multitude of mechanisms. These mechanisms are explained and discussed.

When a group of soils from a region is surveyed there is a general positive relationship between pH and Si solubility/extractability since the more weathered soils with lower soluble Si were also those with lower pH values. This has been demonstrated in a number of studies. Adsorption of Si by Fe and Al hydrous oxide surfaces is highly pH dependant increasing up to a maximum at pH 9.8 (the pK_1 for H₄SiO₄). Thus, the solubility and availability of added Si in soils can decrease with increasing pH as Si adsorption increases. Nonetheless, in the normal pH range of agricultural soils (e.g. pH 4.5-6.5) Si is present in soil solution predominantly as silicic acid, which is not strongly adsorbed. As a result, adsorption to hydrous oxides is often of minor importance in relation to Si availability. Most fertilizer Si sources (e.g. Na, K or Ca silicate and blast furnace/steel slags) are liming materials and their application not only increases soluble Si but also markedly increases pH. Thus, for Si fertilized soils there is often a strong positive relationship between pH and Si solubility. In addition, at low pH (< pH 5.5) where non-liming Si sources are applied, precipitation of hydroxyl-alumino-silicates can occur thus lowering Si availability (also decreasing soluble and exchangeable Al concentations) and this also results in Si availability increasing with increasing pH. In recent times, the role of the pool of phytolith Si (SiO₂.nH₂O) present in soils in determining Si availability has become recognized and the solubility of this form of Si increases markedly with increasing pH. In the short-term this will increase Si availability with increasing pH but in the longer term it will result in a greater diminution of the phytolith Si pool in agricultural soils with increasing pH; and therefore decreased Si availability. Thus, the effect of soil pH on Si solubility and availability is the result of an interaction between a range of mechanisms and the overall effect in any specific situation is dependent on which particular mechanism is dominating.

Theme 5 POSTER

27

Effect of silicon in soybean (*Glycine max* L.) plants grown under zinc deficiency conditions

Hernández-Apaolaza L, Pascual-de Vega B, Lucena J J, Gonzalo M J

Agricultural Chemistry Department, Autónoma University, 28049 Madrid, SPAIN (lourdes.hernandez@uam.es)

Zinc (Zn) is an essential nutrient for plants and its deficiency is one of the most common nutritional disorders affecting vegetal crops in the Mediterranean area. Usually, the lack of this micronutrient is corrected by the chelates application. However, as occur with other nutritional deficiencies, the strategies to improve the mineral nutrition in plants are being developed to the addition of beneficial substances to enhance fertilization. Among these substances, Silicon (Si) has been reported as one of the elements which present favourable effects in plants, especially under stress conditions. In agriculture, Si is used as plant growth stimulant and tolerance builder to overcome abiotic and biotic stresses. Accordingly, the main objective of the present work is to study the potential positive effects of the Si addition in soybean plants growing under Zn deficiency.

Soybean (*Glycine max* L.) plants were grown under controlled conditions in hydroponic culture. The first two weeks of the experiment, plants were cultivated with 10 μ M Zn-EDTA in three batches with different Si doses (Si 0.0mM, Si 0.5mM and Si 1.0 mM). Then, Zn was eliminated from the nutrient solution and Si supply was maintained only in half of the plants of each batch (continues Si supply or initial Si supply). Soybean plants were monitored 21 days estimating chlorophyll content by a SPAD-502 (Minolta), every three days. Leaves, stems and roots were sampled at 7, 14 and 21 days after the zinc removal. Samples of the three tissues digested with nitric acid were evaluated for their mineral content. Total Si content was determined by ICP-MS and Zn was analysed by Atomic Absorption Spectrophotometry.

Soybean plants with Zn supply showed an enhancement of their plant biomass. Moreover, in soybean plants cultivated without Zn, deficiency symptoms were decreased and higher SPAD values were found. The growth of these plants supplemented with Si was also improved with an increase in leaves and roots dry weight. The results obtained were independent of the Si dose added to the nutrient solution. However, the Zn content had been enriched in leaves of plants where Si was added in an unique application at the beginning of the experiment.

Theme 4 POSTER

28

Transcriptomic analysis of Si-induced casparian band development in rice

Hinrichs M, Fleck A T, Schenk M K

Institute of Plant Nutrition, Herrenhäuser Str. 2, Hannover, 30419, GERMANY (hinrichs@pflern.uni-hannover.de)

Silicon is not an essential element but has a lot of beneficial effects on plants. In rice it was shown, that Si reduces the radial oxygen loss from roots growing under anaerobic conditions since the casparian band development in the exodermis enhanced (Fleck *et al.*, 2011). Transcriptomic analysis revealed that genes involved in lignin metabolism were upregulated under Si supply. These results were obtained by using a costume made array with 265 genes putatively involved in the casparian band formation. However the full molecular background for understanding the effect of Si supply and casparian band development is not yet known.

Thus, we aimed a whole genome approach and used the Affymetrix Rice (US) 1.0 ST microarray. Rice seedlings were cultivated in nutrient solution without Si (3 mg*l^{-1} Si) and with Si ($\approx 30 \text{ mg*l}^{-1}$) supply for four weeks in a climate chamber (28 °C, 70 % rel. humidity, 16 h light). For RNA isolation root sections from two to six cm behind the root tip were harvested. This section mirrors the most active zone in casparian band development in rice. The experiment consisted of four biological replications and two technical replications.

The results of the transcriptomic study will be shown and discussed.

Fleck AT, Nye T, Repenning C, Stahl F, Zahn M, Schenk MK. (2011): JEB 62, 2001–11.

Theme 6 ORAL

29

Insights into molecular impact of silica on virus infected cucumber cultures

Holz S¹, Zeise I², Bartoszewski G³, Kneipp J², Kube M¹, Büttner C¹

¹ Humboldt-University in Berlin, Division Phytomedicine, Lentzeallee 55/57, Berlin, 14195, GERMANY (sabine.holz@agrar.hu-berlin.de)

² Humboldt-University in Berlin, Department of Chemistry, Brook-Taylor-Straße 2, Berlin, 12489, GERMANY

³ Warsaw University of Life Sciences, Department of Plant Genetics Breeding and Biotechnology, 159 Nowoursynowska Street, Warsaw, 02-776, POLAND

The cell wall with incorporated SiO_2 is important as a strengthened mechanical barrier for plant defence. Also, silicic acid in a soluble form is reported to be important for defence. Up to now, the role of Si in relation to viral pathogens is not clarified. To address this topic, an *in vitro* approach using the model plant *Cucumis sativus* and Cucumber mosaic virus (CMV) was chosen for transcriptome analysis. In addition, molecular information obtained by Raman microspectroscopy was used for the overall biochemical characterization of the plant material under varying growth conditions. In the first step, this study aims to analyse the impact of silicic acid on the transcriptome of cucumber plants.

Therefore, *in vitro*-grown *Cucumis sativus* line B10 clones were generated derived from leaf microexplants and cultivated on Murashige & Skoog medium. The medium was supplemented with and without silicic acid. Control plants were cultivated on non-modified M & S medium, respectively. Per treatment, 6 clones were obtained. Half of the clones were experimentally inoculated with CMV. RNA isolation including DNase treatment was carried out on leaf and stem samples. Absence or presence of CMV-infection was confirmed by reverse transcription (RT)-PCR. mRNA enrichment was conducted by polyT-oligonucleotide hybridization. Initial RNA-Seq (Illumina) was performed on control and Si treated mRNA. CLC Genomics Workbench V7 was used for the mapping on the genomic draft of *C. sativus* line B10.

Clonal tissue cultures were established successfully and subsequently cultivated on regeneration medium. Furthermore, infection experiments using CMV were successfully applied in micropropagation experiments for follow-up experiments and mRNA of control, Si and NaCl treated plants were obtained. RNA-Seq analysis of the Cucumber transcriptome derived from mRNA samples of control and of Si treated plants indicated a shift in gene expression caused by Si supplementation. Transcripts of 18,000 cucumber genes were identified. A 2-fold change in expression was obtained for 1,180 genes. The confirmation by quantitative (q) reverse transcription (RT)-PCR is in progress. Subsequent analyses will focus on the role of Si on CMV infection in cucumber.

Theme 5 POSTER 30

Silicon decreases mobility and accumulation of cadmium in wheat grains

Hussain I, Ashraf M A, Javed T, Rasheed R, Asghar A, Iqbal M

Department of Botany, Govt. College University, Faisalabad, 38000, PAKISTAN (iqbalbotanist1@yahoo.com)

 Cd^{2+} is one of the most bio-toxic heavy metal due to its high mobility, tenacity, and toxicity even in small concentration. Mostly, Cd^{2+} is taken through symplasmic pathway and accumulates in specific tissues, where it hampers metabolism. The Cd^{2+} stress disturbed photosynthetic metabolism, stomatal opening, water homeostasis and ionic relations, mineral nutrient uptake, auxin metabolism or auxin carriers, photosynthetic pigments and enzymatic and non-enzymatic activity and resulted in reduced biomass and stunted growth. However, being mobile and transported through apoplasmic pathway, Cd^{2+} accumulation in wheat tissues could affect the synthesis and transport of photoassimilates to developing sinks, thereby resulting in major grain yield losses in wheat. Moreover, Cd^{2+} accumulation in grains could exert major threats to health. Therefore, the principle objective of the study was to assess whether Si application could affect the mobility, and thus accumulation of Cd^{2+} in the grains of wheat plants. We also determined the beneficial effects of Si on the production of wheat plants exposed to Cd^{2+} at the boot stage.

Seeds of two wheat cultivars, namely, AARI-2011 and FSD-2008 were obtained from Wheat Research Institute, AARI, Faisalabad, Pakistan. The seeds were sown in the plastic pots filled with 10kg sand. The plants were irrigated with half-strength Hoagland's nutrient solution. At boot stage, plants were exposed to different levels of CdCl₂ (0, 2.5, 5.0, 7.5 mg/L). The control plants were irrigated with half strength Hoagland's nutrient solution throughout the experiment. Sodium silicate (Na₂SiO₃) was used as Si source. Plants were supplied cadmium chloride along with two Si levels (0, 1.5 mM). Average day and night temperature ranged from 21 ± 8 °C to 13 ± 6 °C, relative humidity was 50% during day time and 88% during the night, and light intensity was in the range of 355 to 1395 µmol/ m² /s photon during the course of experiment. The experiment was conducted in completely randomized design with three replications. Sampling for different morpho-physiological and biochemicals attributes was done after 25 days of the treatments.

Cadmium stress resulted in a marked decline in various growth attributes, photosynthetic pigments and the activities of some enzymatic antioxidants, particularly in cv. AARI-2011. A consistent increase was recorded in relative membrane permeability, H_2O_2 contents, lipid peroxidation, total anthocyanin contents, peroxidase activity and Cadmium accumulation in shoots and seeds of Cd^{2+} stressed wheat plants. The addition of Si increased free proline concentration and the activity of superoxide dismutase in both wheat cultivars, while catalase in the AARI-2011 cultivar when under cadmium stress. Application of Si markedly mitigated adverse effects of cadmium on photosynthetic pigments (Chl.a, Chl.b and total carotenoids) and yields attributes by stimulating the antioxidant system of wheat plants. In nutshell, cv. FSD-2008 exhibited the potential to tolerate elevated cadmium levels in growth medium.

Theme 5 ORAL 31

Effect of silicon in modulating vigour of wheat (*Triticum aestivum* L.) seedlings under saline conditions

Iqbal N, Azeem M, Javed M T, Kausar S, Akram M S

Department of Botany, GC University, Faisalabad, 38000, PAKISTAN (drnaeem@gcuf.edu.pk)

Crop productivity is continuously threatened by the increasing levels of toxic salts in growth medium, particularly in arid and semi-arid regions of the world. Seed pre-conditioning, a short gun approach to modulate the effects of abiotic stresses on crop plants is gaining considerable attention of the researchers to induce salinity tolerance in agronomically important crops. The study was conducted to explore the efficacy of pre-sowing seed soaking with silicon in improving seedling growth of wheat.

The experiment was conducted in a growth chamber at $25/20^{\circ}$ C and 55/60% relative humidity with a light and dark period of 10 and 14 hours, respectively. The seeds of wheat variety (PUNJAB-11) sown in Petri plates were subjected to two levels of NaCl (0 and 120m*M*). Silicon was applied as Na₂SiO₃ (0, 10, 20, 30, 40 and 50 m*M*) as seed priming for 8 hours as well as in the rooting solution. The experiment was laid-out in a completely randomized design with three replications.

Application of silicon either in the growth medium or in the form of seed priming resulted considerable recovery from salinity stress. Priming of seeds with Na_2SiO_3 as a silicon source mitigated the adverse effects of salinity stress on germination percentage, root as well as shoot length, dry and fresh weight. Application of silicon either as pre-conditioning of seeds or addition in the growth medium resulted in reduced accumulation of Na^+ in wheat seedlings under saline environment. Whereas, seedlings K^+ and Ca^{++} levels were significantly increased in silicon treated seedlings growing under salt stress.

Theme 8 ORAL

32

Ortho Silicic Acid (OSA) based formulations facilitates improvement in plant growth and development

Jain N, Chidrawar S, Thorat V, Shah P, Rao V

Privi Life Sciences Pvt. Ltd., Privi House, A-71 TTC Industrial Area, Thane Belapur Road, Kopar Khairane, Mumbai-400706, INDIA (neeru.jain@privi.co.in)

Plants take up silicon equivalent to some macronutrients, in spite of this it is not widely recognized as an essential element. Perusal of literature ensures a pivotal role of Silicon in improving growth and development in various crops. The lower acceptance of essentiality of Silicon for plant could be due to lack of systemic studies. *Ortho Silicic Acid (OSA)*, the only bioavailable form of Silicon is a highly unstable molecule. Limited studies aimed to decipher the importance of silicon in agriculture employ adequate sources of Silicon. *Privi Life Sciences Pvt. Ltd.*, have developed and further improved unique proprietary formulas through a patented technology. The technology is being employed for the development of both liquid and (solid) powder formulas. The brand name of the product is *Silixol*. Availability of OSA facilitated us to dissect out pathways to understand the mode of action of this chemical compound within plants. We are involved in undertaking extensive studies to understand the role of OSA in various aspects of plant growth. In the present paper we would like to discuss studies aimed to enhance seed vigour and ultimately plant stand in various groups.

Studies were undertaken to exploit the role of OSA in improving seed vigour. Experiments were designed using different concentrations (1%, 2% or 4%) of powdered Silixol along with a seed coating polymeron maize seeds. Silicon coated seeds, when germinated, exhibited good seedling vigour coupled with 20% increment in seedling length and 27% increase in the biomass. Where coating is not preferred, seeds were soaked in a liquid Silixol solution prior to sowing. The application of Silixol had attributed to better seedling vigour along with 27% increase shoot length of seedling and 37% increase in the fresh weight over control seeds. To have a better insight of this response, rate of imbibition, mobilization efficiency of reserve material as well as starch-sugar content of the germinating seeds were analyzed.

It was found that the presence of OSA increased the rate of imbibition, which is in turn triggers better mobilization of reserve material (39.7% in treated seeds against 24.1% in control, after 2 days). Therefore, it plays an important role in nutrient partitioning as well mobilization. Foliar spray of Silixol had led to better nutrient uptake in the rice, when applied at the nursery stage. This facilitates early transplanting.Seedling had exhibited almost three fold increase in the length. The sprayed seedling also had a better nutrient status with regards to various nutrient ions especially P and K. Seedlings sprayed with Silixol showed a 17% increase in Chlorophyll content over control, ultimately contributing towards a higher rate of photosynthesis. To further enhance our understanding about essentiality of Silicon in crop improvement, we are on the way to dissect its role at the biochemical as well as molecular levels.

Theme 6 POSTER

33

The role of Si and potassium bicarbonate in controlling epidemics of *Podospheara aphanis* on strawberry in the field

Jin X, Hall A M, Huang Y, Fitt B D L

School of Life and Medical Sciences, University of Hertfordshire, College Lane, Hatfield, AL10 9AB, UK (x.jin3@herts.ac.uk)

Strawberry powdery mildew is a major disease of strawberries, which is caused by *Podosphaera aphanis*. The fungus mycelium is on the plant leaves, stems, shoots, buds; flowers and fruit surfaces, which causes yield loss. Whilst it can be controlled with fungicides, they cannot be used close to harvest because of the 'harvest interval', therefore growers need to used other products which limit disease but are not registered pesticides. Strawberry growers frequently use potassium carbonate in a S-based wetter in the interval immediately prior to harvest. The work reported here aimed to investigate the disease control achieved by the potassium carbonate and the Si nutrient, used singly and in combination.

The trial was set up in an open field with the susceptible variety Elegance. The trial used a randomised block design with the following treatments, which were untreated, potassium bicarbonate, 0.25% (v/v) concentration Si, 0.25% (v/v) concentration S with potassium bicarbonate, 0.5% (v/v) concentration Si and 0.5% (v/v) concentration Si with potassium bicarbonate.

The results show that the Si at low rate got greater number of colonies than the Si at high rate. However, the Si with potassium bicarbonate got less number of colonies than the Si without potassium bicarbonate. The potassium bicarbonate only compared with the Si got less control of the epidemic. Therefore, the Si with potassium bicarbonate achieved the best control of strawberry powdery mildew. A further field trial that shows reduction in disease levels where the silicon was only applied in the fertigation tubes will also be reported.

Theme 6 ORAL

34

Silica phytoliths in Asteraceae species: formation patterns and potential antiherbivory role

<u>Katz O¹, Lev-Yadun S², Bar K P¹</u>

¹ Department of Geography and Environmental Development, Ben-Gurion University of the Negev, Be'er-Sheva 84105, ISRAEL (katz.phyt@gmail.com)

² Department of Biology & Environment, Faculty of Natural Sciences, University of Haifa -Oranim, Tivon 36006, ISRAEL

Silica phytoliths are most abundant in the grass alliance, in which they occur in very high numbers due to seemingly unique active silica uptake, transport and deposition mechanisms. Studies of the formation of phytoliths in grass species have shown general positive dependencies on water availability and grazing intensity, and have demonstrated the antiherbivory role phytoliths play in grasses. However, non-grass species are seldom studied because of their lower phytolith concentrations. We studied whether phytolith formation in one such family, the Asteraceae, is dependent on water availability, grazing, and differences between plant organs, in order to assess the antiherbivory potential of phytoliths in this family.

We have studied variations in phytolith concentrations in 19 naturally growing non-spiny and spiny Asteraceae species along a large rainfall gradient in Israel (80 to 900 mm annual mean). In four of the five study sites plants were sampled in both grazed and ungrazed plots. In eight of the studied species, we have analysed inflorescences separately from the rest of the shoot in order to investigate whether reproductive organs have higher phytolith concentrations.

Most Asteraceae species formed more phytoliths under ungrazed conditions, a tendency which was more prominent in the more arid sites. In six out of eight species phytoliths concentrations in the inflorescences were considerably smaller than in the rest of the shoot. Hence, Asteraceae species in general tend to have lower phytolith concentrations when they are exposed to herbivory threat, in environments in which they have limited ability to recover, and in their most vulnerable and valuable organs. These results are inconsistent with an antiherbivory role, suggesting that the antiherbivory potential of phytoliths in the Asteraceae is lower than in grasses. Our study, alongside other studies, demonstrates some fundamental differences in phytolith formation patterns and functions between grass and non-grass species.

Theme 2 ORAL

35

Provision of nitrogen as ammonium rather than nitrate increases silicon uptake in sugarcane

Keeping M G^{1,2}, Rutherford R S^{1,3}, Sewpersad C¹

¹ South African Sugarcane Research Institute, Private Bag X02, Mount Edgecombe, 4300, SOUTH AFRICA (malcolm.keeping@sugar.org.za)

² School of Animal, Plant and Environmental Sciences, University of the Witwatersrand, Johannesburg, 2050, SOUTH AFRICA

³ School of Life Sciences, University of KwaZulu-Natal, Durban, 4000, SOUTH AFRICA

Field trials have shown that silicon (Si) uptake by sugarcane may be limited, even where provision of Si as calcium silicate (Ca₂SiO₄) greatly improved soil Si status. At low soil pH, dissolution of Ca₂SiO₄ is increased while adsorption of Si onto clay particles is decreased, both of which increase Si concentration in soil solution. Hence, Si uptake in grasses may be affected by rhizosphere pH, which can be manipulated through the use of different N-form fertilizers (ammonium or nitrate). Firstly, we tested whether fertilization with NH_4^+ (rhizosphere acidification) in the presence of a nitrification inhibitor (DCD) would increase Si uptake in sugarcane (cultivar N11) compared with NO_3^- (rhizosphere alkalinization). Secondly, we tested whether uptake would differ between an N-efficient, acid tolerant cultivar (N12) and an N-inefficient, acid intolerant cultivar N14.

Two pot trials (fully factorial randomized block designs, 12 and 10 replicates, respectively) were established using low-Si (8.4 mg/L, pH 5.0) soil, with Ca_2SiO_4 slag (10.3% Si) as a Si fertiliser incorporated at 7.4 t/ha. In Trial 1, cultivar N11 was supplied with N at 300 kg/ha as ammonium sulphate (T1), ammonium thiosulphate + DCD (T2), and calcium nitrate (T3). In Trial 2, N was adjusted to 210 kg/ha and treatments were ammonium thiosulphate (T1) and calcium nitrate (T2) only, with the cultivars N12 and N14. Trials were harvested at 4.5 months for plant Si and N, and leaf and stalk dry mass determination. Soil samples were taken for Si and pH analysis. ANOVA analyses were followed by Fisher's protected LSD tests.

 NH_4^+ treatments significantly reduced (p<0.03) soil pH and acid extractable Si (p<0.02) compared with NO_3^- . In both trials, however, NH_4^+ treatments significantly increased (p<0.05) leaf and stalk Si% compared with NO_3^- (with and without DCD in Trial 1). There was no effect of cultivar on leaf or stalk Si in Trial 2, and no significant interaction between cultivar and N treatments. Our results support the hypothesis that supplying N as NH_4^+ rather than NO_3^- increased Si uptake into leaf and stalk in sugarcane. We propose that reduced rhizosphere pH resulting either from bulk acidification of soil due to nitrification of NH_4^+ or from H^+ ions diffusing from the root to balance charges after NH_4^+ uptake, may solubilise Si from Ca_2SiO_4 , making silicic acid available in the immediate root environment. The use of NH_4^+ fertilizer combined with liming at appropriate rates to achieve long-term reduction of acid saturation and aluminium toxicity, may be an effective option for maximising Si uptake from dissolved Ca_2SiO_4 in the rhizosphere.

Theme 5 ORAL 36

Screening and categorization of maize cultivars for silicon acquisition under salinity stress

Khan W, Aziz T, Maqsood M A, Khalid M

Institute of Soil and Environmental Sciences, University of Agriculture, Faisalabad, Faisalabad–38040, PAKISTAN (waqas1919@gmail.com)

Poor seed germination is the major concern in soils having salinity problem. Better germination and seedling establishment may result in better economic yields. Silicon (Si) uptake in a salt stressed plant increases root activity for nutrient uptake and inhibits transpiration which reduces osmotic stress. The primary objective of the present study was to categorize the latest fifteen maize cultivars towards salinity stress and to screen out the salt tolerant and salt sensitive maize cultivars at different growth stages on the basis of their Si uptake ability.

Initially 15 maize cultivars were categorized as sensitive, medium and tolerant to salinity on the basis of germination parameters under control and 60 mM NaCl salinity. Four cultivars (Monsento-919, Golden cross, 32B33 and EV-1089) were categorized as salt sensitive, while four cultivars (Syngenta-8441, Pioneer-30R50, ICI hybrid and Dekalb) were categorized as salt tolerant. These eight cultivars were selected and seeds were germinated in petri plates as well as in pots with two levels of salinity (control and 60 mM NaCl) and two levels of Si (control and 2 mM K₂SiO₃).

Salinity stress significantly decreased germination and physiological parameters in all maize cultivars however effect was variable among tolerant and sensitive cultivars. The maximum decrease in germination percentage and seedling length was observed in 32B33 and EV-1089. Application of Si improved all of the germination parameters. Maximum increase in germination percentage (18 and 23%) and seedling length (22.5 and 21 cm) was recorded in Golden Cross and Dekalb, respectively. Exogenous Si application had increased the radicle, plumule and seedling length in all maize cultivars against salt stress. Similarly, root and shoot Si content were also significantly increased in slat tolerant cultivars with the application of 2 mM K₂SiO₃ as compared to salt sensitive. Availability of Si in plant tissues reduces Na⁺ uptake by making complex with Na⁺. When Si was applied, Na: K concentration ratio showed a significant reduction in all the cultivars as compared to control. EV-1089 was suggested as the most salt sensitive cultivar and Syngenta-8441 as the best salt tolerant. This study implies that Si application aiming at optimum seedling growth and seed germination depends on genotypic variation in maize cultivars and salinity stress.

Theme 7 ORAL

37

The effect of seasonal variations, covariations with minerals and forage value on Itchgrass [*Rottboellia cochinchinensis* (Lour.) W.D. Clayton]' foliar silicification from sudanian Benin

<u>Kindomihou V</u>¹, Holou R², Adjolohoun S¹, Houinato M¹, Sinsin B¹, Meerts P³

¹ Department of Animal production, Faculty of Agronomic Sciences, University of Abomey-Calavi, 03 BP 1974, Cotonou, BENIN. (vkindomihou@yahoo.fr; valentin.kindomihou@fsa.uac.bj)

² Diaspora Engagement LLC, 7723 Carleton Ave, University City, St Louis, MO 63130, USA.

³ Department of organisms Biology of Faculty of Sciences, Free University of Brussels,

Campus de La Plaine, CP244, boulevard du Triomphe, 1050 Brussels, BELGIUM.

Since tropical grass species were found to be highly silicified, how to reduce leaf silica concentration to improve their palatability, digestibility and nutrient value for animal high productivity become questionable. Indeed, silica (SiO₂) in forage grasses has been found in reducing cell-wall digestibility. This study investigates whether: (i) the seasonal variability affects the silica and minerals accumulation and forage values of leaves of Itchgrass (*Rottboellia cochinchinensis*) and (ii) silica concentration is correlated with minerals and fodder value.

In an Itchgrass population selected in the W Biosphere Reserve, leaves were collected on 90 marked plants from May to October 2003 and 2004, at 15 days intervals except May, June and October. Some 300 g of fresh blades from the 3rd most recently expanded leaves were oven dried and analyzed for dry mass, SiO₂, ash, N, Na, Ca, P, K, Mg. Digestible Nitrogen Matter (DNM) and Fodder Energetic Value (FEV) were calculated using Demarquilly formula. Data except SiO₂, Ash and nutritional traits were log-transformed to restore homoscedasticity before Statistical analyses.

SiO₂ ranges from 5.69% to 9.95%, i.e. varying 1.4 fold between May and October, reaching 1.75 fold at mid-September. SiO₂ positively related to Ca but negatively to K, P, N, DNM and FEV. The negative correlations suggest that SiO₂ concentration in *R. cochinchinensis* could be reduced with a significant increase in energy and accumulation of important nutrients such as N, P and K. Therefore, leaf silicification and nutritive value relationship should be conclusive in the case of Itchgrass.

Theme 2 ORAL

38

Silicon cycling and budgets in rice production systems of Laguna, the Philippines

<u>Klotzbücher T¹</u>, Leuther F¹, Marxen A², Vetterlein D², Jahn R¹

¹ Matin-Luther University Halle-Wittenberg, Halle, 06120, GERMANY (thimo.klotzbuecher@googlemail.com) ² Helmholtz-Centre for Environmental Sciences-UFZ, Halle, 06120, GERMANY

Rice is a Si accumulator plant. Improving the Si supply to rice plants might help to mitigate major problems in rice production, including insect pests, fungal diseases and the accumulation of carcinogenic As. We assessed Si transformations (i.e., temporal changes in different forms of potential plant-available Si and uptake of Si by plants) and Si budgets for paddy rice fields of Laguna, one of the important rice producing regions of the Philippines.

We studied altogether 5 fields situated within an area of about $15x15 \text{ km}^2$ during the rainy season of 2013 (Apr-Oct). Studies were managed by researchers or local farmers. Agricultural practices varied in some aspects between fields; hence, this study not only detects the relevant fluxes and processes in the Si cycle but also how they can vary within the study region. We measured dissolved Si (dSi) in soil solutions, acetate-extractable Si in soils (to estimate current plant-available Si), and Si in plants in intervals of ~10 days throughout the cropping season. In addition, we quantified dSi and amorphous Si-oxide particles in irrigation, rain, and drainage water. Conductivity of the plough pan was assessed to calculate Si outputs via percolation. For two of the fields it was not possible to obtain reliable data on water fluxes, hence, we calculated budgets for three fields. Si in wet samples was analysed using ICP-OES, Si in plant samples with X-ray fluorescence analysis.

Si concentrations in plants continuously increased during the cropping season. Before harvest, they were 6.2 - 7.9% of plant-dry-weight, suggesting the plants were well supplied with Si (the critical value proposed in literature is 5%). The total Si uptake by plants at harvest was 51.4-70.8 g Si/m². The bulk of Si was stored in harvest residues (>86%). Hence, residue management is a main factor for budgets; at our sites the residues are left on the field, but it is known for other region that farmers remove part of the straw permanently from rice fields. Si concentrations in rain water were below detection limit, suggesting that Si inputs by rain are not significant. The water management (amounts of irrigation water input; draining of fields) differed considerably between the fields: as a result, inputs of dSi with irrigation water ranged from 7.7 to 62.8 g/m², and outputs with drainage plus percolation ranged from 0.2-69.8 g/m². Fluxes of Si via amorphous Si-oxide particles were less relevant: irrigation inputs were 0.2 – 1.6 g Si/m² and drainage outputs 0.0 - 10.9 g Si/m². Our data suggest that the largest part of Si supplied to plants during the rainy season was from soil constituents, and recent literature suggests that phytoliths in harvest residues are an important Si source. The situation probably differs in the dry season when the fields are primarily flooded by irrigation waters with high Si concentrations. Taken together, Si budgets ranged from -25.3 to +2.8 g Si m⁻² for the observed season. Water and harvest residues are the main factors to manage Si budgets.

Theme 3 ORAL 39

The Mechanism of Biosilicification in Sorghum Leaves

Kumar S, Elbaum R

Robert H Smith Institute for Plant Sciences and Genetics in Agriculture, The Hebrew University of Jerusalem, Rehovot-76200, ISRAEL (skg.research@gmail.com)

Silicon is beneficial to plants in many ways, but there is hardly any explanation of the molecular factors or mechanism that controls biosilicification in plants. To understand biosilicification in plants, we need to have a system in which we have control over silica deposition. Sorghum is a typical silicon accumulator with leaf silica content of 3.5% or even higher on dry mass basis. A sorghum line mutant for a silicon transporter primarily expressed in roots (homologous to the rice silicon transporter OsLSi1) has been isolated in our lab. We expect the leaf silicification machinery to be functional in the mutant and the silica deposition in the leaf tissue (or its pieces) directly proportional to the exposure to silica. Our aim is to identify the candidate gene(s) and/or the molecular factor(s) responsible for the silicification in sorghum leaves.

To ascertain that the mutant leaf silicification machinery is intact, leaf explants from the wild type as well as the mutant was kept in known strength of Si solution (double distilled water (ddw) for control) for a specific period of time, and burnt in between two glass slides. The ash was washed with HCl and ddw and the slide was observed under light microscope. Epidermal layer of live leaf tissues exposed to Si solution was observed under binocular. This allowed us to identify the initiation of silicification in the leaves and the areas of higher silicification. Leaf tissues were taken to Air-SEM (Air-Scanning Electron Microscope) where live tissues can be scanned. Higher magnification (up to 10,000 X) coupled with EDX allows us to identify specific locations of the tissues with high silicon concentration. WT and mutant leaf explants will be sent for RNA-seq analysis (i) before immersion in Si solution, (ii) after immersion but before massive silicification, and (iii) after massive silicification. We will test the candidate gene(s) for their role in silicification in maize cell culture.

We observed the typical dumbbell shaped silica bodies in the WT and in the mutant when the mutant explants were exposed to Si solution. The extent of silicification in the mutant leaf was proportional to the duration it was kept in Si solution. In 15 days of silicon treatment, the mutant leaves were as highly silicified as the WT without Si treatment. Examination of the mutant's live leaf tissue pieces after silicon treatment under binocular and Air-SEM showed that the leaf base of sorghum could be a good tissue system to work with. Spatial qRT PCR analysis of the WT plants hydroponically grown in Si solution revealed that the sorghum analogue of *OsLSi1* and *OsLSi2* are primarily expressed in the root system while the *OsLSi6* analogue is primarily expressed in the leaf tissues. We aim to decipher the molecular mechanism of silica deposition in sorghum leaves using our current system, bioinformatic analysis of sorghum microarray data available in the database in conjunction with our RNA-seq data and experimentally verifying the role of candidate gene(s) and/or molecular factors.

Theme 8 ORAL

40

The efficacy of the Silicic Acid Agro Technology: the use of stabilized silicic acid.

Laane H-M

R&D Department, ReXil Agro BV, Demmersweg 92A, 7556 BN, Hengelo, the Netherlands (hm.laane@rexil-agro.com)

Because silicon's abundant presence in the soil as silicates or silicon dioxide it is still the common opinion that plants do not suffer from Si deficiency. But silicates aren't plant available. Only mono silicic acid (SA) can be absorbed by the plant. But due to its instability the concentration of this 'biosilicon' is (too) low causing a 'relative silicic acid deficiency' in most soils. This instability is also the reason why there was previously no silicic acid available product. This situation has changed with a (patented) production method to make stabilized silicic acid. The application of this stabilized SA as foliar spray, hydroponically or to the soil is called the Silicic Acid Agro Technology (SAAT). The effects of trials using SAAT from 2001 on will be presented.

When foliar sprays are used the concentrated silicic acid (with/without other nutrients like boron, molybdenum, zinc, etc.) is diluted 500 times and applied several times during the vegetative stage with 2 weeks interval.

The application of SA induces in almost any crop (compared to control) an increase of root mass, more and thicker tillers, larger leaf area together with a higher chlorophyll content. The increase in root mass results in an increase in growth parameters due to the higher uptake of nutrients (P, Ca, K, Si and Bo) from the soil (1). SAAT also increases the resistance of the plant to both as well abiotic as biotic stress factors. Significant increases in yield, lower infection rates and higher quality are seen in many staple foods (rice (2), sugarcane, potatoes, etc.), vegetables (peppers, tomatoes, carrots, etc.), cereals (wheat, finger-millet, etc) and other crops. The sprayed silicic acid induces a top-down mechanism resulting in enhanced plant metabolism starting with an increased uptake of nutrients. This shows that SA should be regarded as a *biostimulant* next to silicon's function as fertilizer. Biostimulants are "substances and materials, with the exception of nutrients and pesticides, which, when applied to plants, have the capacity to modify physiological processes in a way that provides potential benefits to growth, development and/or stress response" (3). SAAT induces in almost any crop increased growth, more biomass, higher yield, higher quality and longer shelf life. The efficacy of SAAT is higher than that of almost any other conventional or silicon fertilizer. SAAT is safe, eco-friendly and cost effective. This promising technology for the 21th century needs much more attention (4).

^{1.} H.K. Bhavya, V. Nache gowda, S. Jaganath, K.N. Sreenivas & N.B. Prakash (2011): Effect of foliar silicic acid and boron acid in Bangalore blue grapes. Proc. 5th Int. Conf. on Silicon in Agr., Beijing, China, 7-8.

N. B. Prakash, N. Chandrashekar, C. Mahendra, S. U. Patil, G. N. Thippeshappa & H. M. Laane (2011): J. of Plant Nutrition, 34:12, 1883-1893.

^{3.} P. du Jardin: The Science of Plant BIOSTIMULANTS, April 2012.

^{4.} E. Bent (2014): Silicon Solutions. Sestante Edizioni.

Theme 5 ORAL 41

Cadmium uptake in wheat protoplast is reduced by silicon

Landberg T, Maity P J, Greger M, Lindberg S

Department of Ecology, Environment and Plant Sciences, Stockholm University, Lilla Frescati, Stockholm, 106 91, SWEDEN (tommy.landberg@su.se)

Silicon (Si) affects Cd uptake and transport in plants, especially the distribution of Cd within the plant. However, it is less known how Si influences Cd at the cellular level. This study investigates silicon's role on Cd uptake into wheat protoplasts, living cells without cell walls.

The analyses were performed on individual protoplasts from wheat shoots using fluorescence microscopy. Protoplasts were prepared from plants cultivated in solution with or without Si (1 mM). Uptake of Cd into the protoplasts was detected continuously during 5-10 min using a Cd-specific fluorescent dye, Leadmium Green, AM. For Ca-uptake, Fura 2, AM, a calcium binding benzofuran, was used. Protoplasts were treated with a series of Cd-concentrations (1 – 100 μ M) in combinations with Si (0 – 2 mM). Also the uptake of Ca were analysed in the presence of 1 or 10 mM CaCl₂ and different concentrations of Si (0 – 2 mM). Furthermore, vanadate was used in some measurements as an inhibitor of active uptake.

Our results showed that Si decreased the uptake of Cd into the protoplasts. Higher Sitreatment gave stronger reduction of the Cd uptake. At the highest Si-treatment (2 mM) the Cd-uptake was reduced with 65 - 90 % depending on Cd-treatment (1 - 100 μ M). Silicon treatment both during cultivation and at the uptake experiments almost abolished the Cd uptake. The kinetic pattern of Cd uptake indicates that the protoplasts have multiple uptake systems where Si interacts with Cd. It is less likely that Si interacts with the passive uptake of Ca and Cd via channels, as Si did not affect the Ca uptake. Moreover, the inhibition effect by vanadate on Cd uptake was not dependent on Si. The interpretation may be that Si operates on the active uptake of Cd. We can, thus, conclude that silica do influence uptake of Cd at cellular level and that Si likely influence multiple uptake systems for Cd.

Theme	3
ORAL	

42

Finding What Shouldn't Be There: The Discovery Of Silicon Transporters In Low Si Accumulator Plants

Leisner S¹, Zellner W L², Khandekar S¹, Frantz J³

 ¹ Department of Biological Sciences, The University of Toledo, Toledo, OH, 43606, USA (sleisne@utnet.utoledo.edu)
 ² USDA-ARS Greenhouse Production Research Group, 2801 W. Bancroft St. M.S. 604,

² USDA-ARS Greenhouse Production Research Group, 2801 W. Bancroft St. M.S. 604, Toledo, OH 43606, USA

³ Pioneer Hi-Bred International, 7305 NW 62nd Avenue, Johnston, IA 50131, USA

The beneficial element silicon (Si) confers many advantages on plants. Therefore, the ability of a plant to effectively respond to stress depends on the ability of the plant to acquire Si. Plants vary widely in their ability to accumulate Si ranging from high (>1% of the dry weight, similar to macronutrients) to low accumulators (<1% of the dry weight, similar to micronutrients). In order to acquire Si, plants have transporters to carry the element into the plant. Indeed, Si transporters were identified in a variety of plants that are high Si accumulators, while low Si-accumulating plants were proposed to obtain the element through non-specific means. We had previously found that challenging *Nicotiana tabacum* (classified as a low Si accumulator) with either *Tobacco ringspot virus* or copper (Cu) toxicity caused an increase in Si uptake into leaves. These data suggest that Si uptake is regulated, even in low Si accumulators and that growth conditions can strongly influence the levels of Si within plants.

Initially, our work employed a bioinformatics approach to identify potential *N. tabacum* Si transporters. This work used a variety of programs to determine relatedness of the putative Si transporters to known proteins. We then examined the regulation of any putative transporters, identified in our bioinformatics analyses, in response to Si by real-time reverse transcriptase-polymerase chain reaction (RT-PCR) analysis. The expression of these transporters in response to stress is currently being examined by RT-PCR. We are also examining the ability of these *N. tabacum* proteins to transport Si in *Xenopus* oocytes. Finally, we are performing pull-down experiments using the N- and C-termini of one of the putative *N. tabacum* Si transporters to identify interacting partners.

Our analyses identified a putative tobacco gene that could be a Si transporter of the aquaporin type. This predicted aquaporin contained the residues that are a signature of a Si transporter. RT-PCR analyses indicated that the RNA levels of this putative Si transporter gene are down-regulated by Si, like the rice Si transporter gene. The RNA levels of a putative standard tobacco aquaporin used as a control were unaffected by Si treatment. We are currently working to confirm that the putative Si transporter does indeed transport Si and examine how the expression of this protein is regulated in response to stress. Together these data will lead to a better understanding of Si uptake and transport in all plant species.

Theme 3 KEYNOTE 43

Silicon transporters and their role in plants

Ma J F, Yamaji N, Mitani-Ueno N

Institute of Plant Science and Resources, Okayama University, Kurashiki 710–0046, JAPAN (maj@rib.okayama-u.ac.jp)

Transport of silicon from soil to different organs of the above-ground part requires different transporters. Since the first Si transporter, Lsi1 was identified in rice in 2006, a number of Si transporters have been identified in different plant species. Homologs of rice Lsi1 has been isolated and functionally characterized in barley, maize, wheat, pumpkin and horsetail. They function as a silicon influx transporter, which is required for transporting Si from external solution to the root cells. On the other hand, Si efflux transporters (Lsi2) have also been identified in rice, barley, maize and pumpkin. They are also involved in the Si uptake, but are responsible for transporting Si out of the cells towards the root stele. Different plant species show distinct expression pattern and cellular localization of Lsi1 and Lsi2.

After Si uptake by the roots, the translocation of Si from the roots to the shoots is mediated by Lsi3, a homolog of Lsi2 in rice. It is localized to the root pericycle cells. On the other hand, a homolog of Lsi1, Lsi6 mediates xylem unloading of Si. Knockout of this gene resulted in disturbed Si deposition pattern in the leaves.

Final distribution of Si to the grain (mainly husk) is mediated by three different transporters, Lsi2, Lsi3 and Lsi6 in rice. All of them show high expression in the nodes, a place for mineral distribution. Lsi6 is localised at the xylem transfer cells of enlarged vascular bundle, whereas Lsi2 is localized at distal side of a parenchyma cell layer surrounding the enlarged vascular bundles, which is the next cell layer of xylem transfer cells and Lsi3 is localized at the parenchyma tissues between enlarged- and diffuse-vascular bundles. Knockout of *Lsi6, Lsi2* or *Lsi3* resulted in decreased Si accumulation on the husk, but increased Si accumulation in the flag leaf, indicating that cooperation of these transporters localized at the different cell layers are required for inter-vascular transfer of Si from enlarged vascular bundles to diffuse vascular bundles for preferential distribution of Si in the husk. Similar system on such inter-vascular transfer of Si in barley was also observed. In my talk, I will introduce recent progress made in Si transporter researches.

Theme 4 ORAL

44

Fixing critical limit for silicon fertilization in the intensively rice growing soils of Periyar Vaigai Command area, Tamil Nadu, India

Mahendran P P¹, Sreya U P², Balasubramaniam P³

¹ Dept. of Soil Science and Agricultural Chemistry, Agricultural College and Research Institute, Tamil Nadu Agricultural University, Killikulam-628252, INDIA (ppmahendran2002@yahoo.co.in)

² Department of Soil Science and Agricultural Chemistry, Agricultural College and Research Institute, Tamil Nadu Agricultural University, Madurai-628 252, INDIA

³ Dept of Soil Science and Agricultural Chemistry, Anbil Dharmalingam Agricultural College and Research Institute, Tamil Nadu Agricultural University, Tiruchirappalli-620009, INDIA

The Periyar Vaigai Irrigation System is one of the oldest and second largest irrigation systems in India existing since 1898 and rice is grown as monocrop. High accumulation of Si in rice has been demonstrated to be necessary for healthy growth and high stable production. Depletion of plant available Si in traditional rice soils from the continuous monoculture of high yielding varieties could be a possible limiting factor contributing to declining or stagnating yields in many rice growing countries. Though there is ample number of research work on macro and micro nutrients based on needs for sustaining rice productivity, the work related to the Si nutrient is very much limited in our country, in spite of its heavy feeding by rice crop. Hence the present investigation was ventured to assess the status of available silicon and fixed critical limit for silicon fertilization in the intensively rice growing soils of Periyar Vaigai Command (PVC) area.

Totally 100 surface soil samples were collected and analyzed for plant available silicon. In order to assess the response of rice to applied Si for arriving the optimum Si level and also to fix critical limit of Si, a pot culture experiment was conducted making use of the bulk soil samples collected from twenty locations (out of 100 locations) of rice growing tract of the PVC area with five levels of Si (0, 75,150, 225 and 300 ppm) using rice variety ADT-45 as test crop. The effect of Si on yield attributing characters (number of productive tillers, numbers of filled grains per panicle, per cent chaffiness) and rice grain yield were recorded.

The present investigation revealed that available silicon in the rice soils ranged from 4 to 250 ppm. The various yield attributes and grain yield of rice were significantly increased with Si application up to 225 ppm level. However, the optimum physical and economic dose of Si was found to be 368 and 310 ppm respectively in the rice growing soils of PVC area. The soil available silicon levels were categorized into low, medium and high based on per cent relative yield. Accordingly the soil available Si up to 122 ppm (<75% relative yield) was considered as low, 122 to 181 ppm (75 to 95 % relative yield) as medium and more than 181 ppm (>95 % relative yield) as high in Si availability. As per this categorization, out of 100 soil samples tested in PVC area, 64 per cent of the soil samples were deficient in Si supply cautioning the seriousness of Si deficiency. Hence the soils analyzing less than 122 ppm of available silicon need balanced fertilization including silicon to avoid its deficiency.

Theme 3 ORAL

45

Silicification in sorghum: insights from a mutant defective in silicon uptake

Markovich O, Cohen D, Fridman E, Elbaum R

Robert H. Smith Faculty of Agriculture, Food and Environment The Hebrew University of Jerusalem, POB 12, Rehovot, 76100, ISRAEL (oshrym@gmail.com)

Accumulation of silica is common in grasses, and may reach 10% of their dry weight and more. Silicon availability improves plants ability to accommodate to stress situations, mostly in unknown mechanisms. Sorghum *(Sorghum bicolor)*, an important staple grass crop in hot and dry regions, deposits silica in its roots and shoots. The aim of the work was to learn about the silicification mechanism in sorghum through the characterization of a mutation in the putative sorghum silicon transporter Low Silicon 1 (SbLSi1).

Mutagenesis by fast neutron radiation of 500 sorghum seeds resulted in a single mutant resistant to germanium oxide. We applied PCR-based methodologies to locate the mutation to the putative SbLSi1 root transporter. The plant was characterized by light and scanning electron microscopy, Raman micro spectroscopy, and silicon quantification by charring.

We performed a functional analysis for a candidate gene and demonstrated its active role in the import of silicon into sorghum. This gene resides on chromosome 4, and encodes for a protein sharing 97% identity with the maize root transporter ZmLSi1. A loss-of-function mutant contains 0.025 of the wild type plant total silicon. However, 80% of the mineral is deposited in the roots, in contrast to the wild-type plant, carrying only 16% of the silica in its roots. We applied Air-Scanning Electron Microscopy (Air-SEM), on intact leaves under ambient conditions. The mutant leaf epidermis displayed dumbbell-shaped silica cells, usually empty of silica. The initiation of the silicification in these bodies was from the cell's margins inwards. Raman spectroscopy showed that the empty silica cells carry highly crystalline wax, similar to their neighbouring cork cells. Once exposed to external silica, the mutant leaves uptake the mineral and deposit it ectopically. Our observations support two pathways of leaf silicification: one, under high control at the silica cells, and another, which is spontaneous, at the apoplasm of the epidermis.

Theme 2 ORAL

46

Controls on Si uptake by rice in Southeast-Asian paddy soils

Marxen A¹, Klotzbücher T², Vetterlein D¹, Jahn R²

¹ Helmholtz-Centre for Environmental Research, Department of Soil Physics, Theodor-Lieser-Str. 4, Halle, 06120, GERMANY (anika.marxen@ufz.de)

² Martin-Luther-University Halle-Wittenberg, Department of Soil Science, Von-Seckendorff-Platz 3, Halle, 06120, GERMANY

Rice (*Oryza sativa*) is the staple food for more than half of the world's population. Although it is one of the largest silicon (Si) accumulating crops, Si cycling in rice paddies is still understudied. Our objectives are (i) to assess plant-available Si (Si_{pa}) in soils and (ii) Si uptake by rice of paddies for altogether seven regions in Vietnam and the Philippines, which differ in climate, geology, soil, and agricultural practice. Additionally, we (iii) studied the effect of increasing Si supply to various soils on rice Si uptake and growth.

In each of the seven study regions of 15 km x 15 km area, we have chosen ten paddies. We sampled paddy topsoils (Ap-horizon) and extracted Si_{pa} by 0.18M Na-acetate, buffered to pH 4. We additionally sampled matured rice plants and measured Si concentration of straw, grains, and hulls by X-ray fluorescence analysis. Furthermore, we conducted a pot experiment to investigate the relationship between Si availability in soils and Si uptake by rice. Rice was grown in two Vietnamese and two Philippine soils with two treatments, with and without the addition of silica gel, a source of rapidly dissolvable Si. We continuously measured Si concentration in the soil solution. Biomass production and Si concentration in shoots were assessed after 29 days.

 $\mathrm{Si}_{\mathrm{pa}}$ concentrations in Ap-horizons were significantly higher in the Philippines than in Vietnam (222 \pm 92 mg kg⁻¹ versus 37 \pm 14 mg kg⁻¹). We refer this finding to differences in geoand pedologic conditions between the countries. In Vietnam, land surfaces are very old and soils are highly weathered and thus desilified; in the Philippines, rocks were recently formed due to active volcanism and high amounts of Si_{pa} are released during mineral weathering. Total Si uptake by rice was also significantly higher in the Philippines than in Vietnam $(709\pm144 \text{ kg ha}^{-1} \text{ versus } 201\pm102 \text{ kg ha}^{-1})$. However, within the countries, and even within individual regions, we found no correlation between Sipa and Si uptake. Reasons might be variations of agricultural practice (variety, crop residue management, water management etc.). Under controlled conditions in the laboratory, Si concentration of all soil solutions was significantly increased by the addition of silica gel, even in the Philippine soils with high initial concentration of Sipa. We found a positive relationship between Si in soil solution and plant Si uptake. In Vietnamese soils, rice growth was significantly increased by Si addition. We conclude that especially in Vietnam, an increase of Si availability might largely increase Si uptake, thus might improve rice growth and yield. Potential melioration strategies might be Si fertilization and changes in harvest residue management. Currently, we are conducting Si fertilization experiments in Vietnam; first results will be shown at the conference.

Theme 8 ORAL

47

New generation Si fertilizers: activation of the natural plant defense system

<u>Matichenkov V</u>¹, Bocharnikova E², Dåstøl M³, Wei X⁴, Zhan Q⁴

¹ Department of Ecology and Physiology of Autotrophic Organisms, Institute Basic Biological Problems, Pushchino, 142290, RUSSIA (vvmatichenkov@rambler.ru)

Problems in Soil Science, Pushchino, 149292, RUSSIA

Evolution of any type of agrochemicals implies increasing of their efficiency by using new technologies for product manufacturing and application according modern understanding of natural processes. Today traditional Si-rich substances used in practice are usually characterized by low efficiency (solid products) or short-term effect (liquid products). Recent investigations have shown the important role of active Si forms in the formation and function of plant defense system via Si influence on the plant signal system and production of stress ferments and stress proteins.

Greenhouse and field experiments were conducted with vegetables and rice to test the influence of elaborated Si fertilizers (Elkem AS, Silicon Materials) on plant stress resistance depending on application rate. Soluble Si and hydrogen peroxide were analyzed in plant sap in dynamic. Amounts of insect attacks and fungi infections were assessed as well.

The results obtained showed that the application of Si fertilizers had positive influence on plant growth. The numbers of infections and insect attacks were significantly decreased under Si fertilization. One of the mechanisms of beneficial Si effect on plant productivity and crop quality was supposed to be performed via activation of plant signal system providing synthesis of specific and non-specific antioxidants. As evident from our data, beside the uptake of monosilicic acid by roots, soluble Si can be adsorbed by leaf blades. Then this Si can be translocated from leaf to root. By this means, the Si transport inside plant can occur not only by root-shoot-leaf way but in opposite direction as well. Primary transport occurs through apoplasmic pathway. However, a positive Si influence on the synthesis of specific and non-specific stress proteins is probably realized in symplasmic media. The results showed a perspective of suggested Si fertilizers for activation or re-activation of the plant natural defense system. However, the mechanisms require further investigations.

²Department of Physical-Chemistry of Soil, Institute of Physical-Chemical and Biological

³ Elkem, Kristiansand, NO-4675, NORWAY

⁴ Hunan Institute of Economic Geography, Changsha, 410004, CHINA

Theme 6 POSTER 48

The Role of Silicon in Tall Fescue Leaf Characteristics

McLarnon E¹, Lenk I², McQueen-Mason S¹, Hartley S E¹

 ¹ Department of Biology, Wentworth Way, University of York, YO10 5DD, UK (em965@york.ac.uk)
 ² Dlf-Trifolium A/S Ny Østergade 9, 4000 Roskilde, DENMARK

Festuca arundinacea (Schreb.) is an allohexaploid, cool season perennial grass widely used as both a turf and forage grass. Some varieties of this grass have harsh leaf surfaces causing them to be unpalatable to grazing herbivores. This harsh texture may be due to silicon (Si) deposition. *F. arundinacea*, like many grasses is a Si accumulator and deposits Si as amorphous silica bodies (phytoliths) in the intercellular and intracellular spaces, within the cell wall and within leaf spines. Such Si deposition is an effective plant defence against herbivory. Si levels increase in damaged plants and plants with high Si content are less palatable to both vertebrate and invertebrate herbivores, more abrasive to herbivore mouthparts and their nutrients are less accessible to herbivores, reducing their growth rates. This study aims to investigate the relationship between leaf harshness and Si levels in *F. arundinacea* and to provide information relevant for plant breeders seeking to produce varieties with improved forage quality.

F. arundinacea varieties of contrasting harshness were grown in the glasshouse under high and low Si supply, with and without damage. Plants were analysed for leaf Si content using X-ray fluorescence (XRF) and Si deposition on the leaf surface was examined using a scanning electron microscope (SEM) combined with energy-dispersive X-ray analysis (EDX).

"Harsh" varieties of *F. arundinacea* had significantly higher leaf Si than the "soft" varieties. "Harsh" varieties deposited Si into the leaf spines on the leaf surface, in contrast to the "soft" varieties, which deposited Si around the leaf margin and in a regular pattern within the leaf cells; these varieties also had fewer leaf spines on the leaf surface. The higher Si content coupled with the higher density of leaf spines in the "harsh" varieties suggests these varieties deposit Si differently to the "soft" varieties. This has implications for palatability and digestibility and suggests that manipulation of Si uptake could be a target for plant breeders seeking to produce forage grasses with more accessible nutrients.

Theme 8 POSTER 49

Indirect effect of silicon product against apple scab and strawberry diseases

Meszka B¹, Wilk R²

¹Research Institute of Horticulture, Skierniewice, POLAND (beata.meszka@inhort.pl)

² Institute of Inorganic Technology and Mineral Fertilizers, Wroclaw University of Technology, Wroclaw, POLAND

Integrated Pest Management (IPM) is at present the mandatory standard for all professional users of plant protection products. It involves all available methods of plant protection, but special emphasis is put on alternatives. One of such a possibility is application of growth stimulator OPTYSIL containing active silicon, which is fully assimilable by plants. OPTYSIL induces resistance of plants to stress and supports the natural defense system. Cell walls become more resistant to enzymes produced by pathogens. In our study this preparation was used for foliar application on apple cv. Golden Delicious against apple scab (*Venturia inaequalis*) and on strawberry plants cv. Senga Sengana against grey mould (*Botrytis cinerea*) and leather rot (*Phytophthora ccatorum*). Experiments were conducted in 2013 in which, the weather conditions were very favorable (over 200 mm rainfall during May and June) for the development and spread of the diseases.

The severity of apple scab on unprotected trees has reached in early June up to 40% and in July 82% on leaves and 60% on fruits. The effectiveness of OPTYSIL applied at 0.5 and 1.0 l/ha six times from 'pink bud' (BBCH 57) ranged from 67% to 81% and from 78% to 80%, respectively in control of scab on leaves and fruits and it was similar (on leaves) or lower (on fruits) to that obtained with standard fungicides Captan 80 WG (captan) and Delan 700 WG (ditianon).

The severity of gray mold and leather rot on unprotected plants of strawberry was respectively 10% and 14%. OPTYSIL used at 0.5 and 1.0 l/ha four times from beginning of flowering (BBCH 61) reduced the severity of both diseases. Its efficacy ranged from 40 to 80% depending on dose and term of assessment and it was lower (1st assessment) or similar (2nd assessment) to that obtained with standard fungicides Switch 62,5 WG (cyprodinil + fludioxonil), Signum 33 WG (piraclostrobin and boskalid) and biological product Polyversum WP (*Pythium oligandrum*). The yield from plots treated with OPTYSIL was significantly higher than that from control plots. The preliminary results indicate the possibility of using OPTYSIL in IPM program against fungal diseases.

Theme 1 ORAL

50

The 1% Na₂CO₃ method as an indicator of bioavailable Si in crop land

Meunier J D, Keller C, Riswan M, Barboni D

AMU, CNRS, CEREGE, Europole Mediterraneen de l'Arbois, 13545 Aix en Provence, France (meunier@cerege.fr)

Several simple and rapid extractions have been proposed for estimating bioavailable Si in agronomy. They have been validated with the values of Si obtained in plant grown on the same soil. These extractants include e.g. H_2O , HCl, CaCl₂, ammonium citrate, acetic acid or exchange with anionic resins. The obtained dissolved Si fraction corresponds to the fraction of Si loosely bound to the solid phase. However those protocols do not take into account the dynamics of silicate dissolution in soils, which may take several weeks to months to dissolve. This is the case for phytoliths and other amorphous silica particles (ASi), which are poorly dissolved by the extractants cited above but represent a source of silica amongst the most soluble of the soil solids at slightly acid to neutral pH.

In order to determine if the Si uptake by plant is correlated with the pool of ASi from natural soils, we conducted a pot experiment on 3 different French soils: a Silandosol, an aric Podzol and a Calcisol. Two acid extraction methods were used to quantify the short-term bioavailable Si: 0.5 M acetic acid and 0.2 M NH₄ oxalate buffered at pH 3.0. ASi was analysed using alkaline extraction with 1% Na₂CO₃ and physical extraction by heavy liquid flotation. We also conducted a series of 9 successive croppings using Durum wheat with straw exportation where key parameters (ASi in soil and plants; dissolved silica i.e. DSi in soil solutions) were measured over time.

The 1% Na₂CO₃ method extracted larger amounts of Si than the acid methods except for the Silandosol characterized by the highest values using NH₄ oxalate. The presence of poorly crystalline Al-Si phases in Silandosol partially explained the high Si values using oxalate and 1% Na₂CO₃ extractions. Physical extraction shows that ASi is mostly composed of phytoliths with higher values in Silandosol. We observed significant correlations between ASi_{1% Na2CO3} and Si_{shoots} (R² = 0.45) and ASi_{1% Na2CO3} and DSi (R² = 0.63). However the correlation between DSi and Si_{shoots} was not significant (R² = 0.26). Our results suggest that DSi may not be a relevant indicator for assessing bioavailable Si because it does not take into account the dynamics of Si at the scale of the crop development. On the contrary, ASi_{1% Na2CO3} integrates the various sources of bioavailable Si accumulated in crops during growth such as biogenic silica particles (e.g. phytoliths), poorly crystallized Al-Si phases, and adsorbed Si.

Theme 8 ORAL

51

Behaviour of different levels and grades of Diatomite as silicon source in acidic and alkaline soils

Nagabovanalli B P, Anitha M S, Sandhya K

Department of Soil Science and Agricultural chemistry, UAS, GKVK, Bangalore, PIN – 560065, INDIA (nagabovanalliprakash@rediffmail.com)

Diatomite is a sedimentary rock that results from the deposition of Si rich unicellular life forms of aquatic algae found in both salt and fresh water known as diatoms. Given the rice as a silicon accumulator plant, information on the use of diatomite as a source of silicon, application rates and their effects on growth and yield are of great importance. Hence, greenhouse studies were conducted to know the effect of different levels of AgriPower diatomite on availability of nutrients in acidic and alkalinesoils under field capacityand submergenceconditions.

An incubation study was conducted by using acidic and alkali soils in a plastic pot with different levels (0, 150, 300 and 600 kg ha⁻¹) of diatomite under field capacity and submerged conditions. In another study, different grades of diatomite at different levels (0, 250, 500, 750, 1000 and 1500 kg ha⁻¹) were incubated under field capacity moisture regime. The soil samples were periodically analyzed on 7, 15, 30, 60, 90 and 120 days after incubation (DAI) with a special emphasis on Plant available silicon (PAS). Pot culture studies were also conducted by using different levels of diatomite maintained under field capacity and submerged conditions using rice as test crop and different grades of diatomite at different levels maintained at field capacity using aerobic rice as test crop. The soil and plant samples were collected at 60 days after transplanting (DAT) and at harvest and subjected for nutrient analysis by adopting standard procedure.

Irrespective of the application rates of diatomite, under both the moisture regimes there was an increase in the pH of both acidic and alkaline soils. Increase in PAS was observed at 15 DAI in acidic and alkaline soil with the increasing levels of diatomite, whereas the highest was observed at 30 DAI in both types of soil. Irrespective of application rates of diatomite, there was a decrease in PAS at 90 DAI and thereafter increased at 120 DAI in both soil types and higher than control. Among the different grades of diatomite, Agripower Silica grade-1 recorded significantly higher PAS in acidic soil under field capacity. There was a significant increase in the yield and yield parameters with the application RDF (Recommended Dose of Fertilizers) + varied levels of diatomite in submergence and field capacity and diatomite grades under field capacity in both soil types compared to control. Application of 600 kg diatomite ha⁻¹ along with the RDF increased the yield and yield attributes of crop grown in soil under submerged condition compared to field capacity moisture regime. Among the different grades of diatomite, application of 750 kg ha⁻¹ of Agripower Silica grade-1 recorded higher yield and yield attributes in acidic soil when compared to other grades of diatomite.

Theme 4 KEYNOTE

52

Role of Silicon in Plant Nutrient Acquisition and Transport Nikolić M

Plant Nutrition Group, Multidisciplinary Research Institute, University of Belgrade, Kneza Viseslava 1, Belgrade, 11030, SERBIA (mnikolic@imsi.bg.ac.rs)

Silicon (Si) is a unique mineral thought to paly a role in enhancing overall plant tolerance to stressful environment. Over the past decade rapid progress has been made in elucidating the mechanisms through which Si modulates mineral toxicities in higher plants. Yet, information on the relevance of Si under nutrient deficiency stresses is still lacking. This is partly due to the fact that root responses to lack of nutrients have so far mainly been studied and characterized in nutrient solution experiments in which Si was omitted. On the other hand, it has been reported from numerous field studies that Si fertilization might increase leaf nutrient content and/or improve nutrient balance.

The aim of my keynote is to provide updated review on the role of Si in nutrient acquisition and utilization by plants. I will focus on the physiological and molecular mechanisms of how Si influences root acquisition of phosphorus (P) and iron (Fe) in the rhizosphere. Beside possible interactions between Si and P in soil, application of Si in acidic soil resulted in enhancement of the expressions of P-deficiency related genes encoding phosphoenolpyruvate carboxylase (PEPC), the organic anion efflux transport proteins (belonging to ALMT and MATE families) and high-affinity Pi transporter (PHT1.2) in wheat roots, along with an increased exudation of malate and citrate and P uptake. Consequently, the leaf P concentration in Si-treated plants without P application achieved the range of that in Pfertilized plants. In Fe deficient cucumber plants, Si addition resulted in extension of the root apoplastic pools for Fe, together with the enhanced expression levels of the proteins (FRO2 and IRT1) involved in reduction-based Fe uptake. Also Si enhances biosynthesis of Fechelating compounds (organic acids and phenolics) for improved mobilization of Fe from the rhizosphere and reutilization of apoplastic Fe. In addition, I will present very recent results of my research group, demonstrating for the first time Si-induced expression of genes responsible for symplastic Fe unloading and phloem transport of Fe-nicotianamine complex from old to younger leaves.

In conclusion, the role of Si in enhancing nutrient efficiency appears to be more indirect, by affecting activation of genes responsible for enhanced root acquisition and tissue mobilization of nutrients.

Theme 5 POSTER

53

SILICON HAS A PROTECTIVE EFFECT ON CUCUMBER GROWN IN COPPER EXCESS

Nikolić S D¹, Samardžić J¹, Nikolić M²

¹ Institute of molecular genetics and genetic engineering, University of Belgrade, Vojvode Stepe 444a, 11010 Belgrade, SERBIA (dsnikolic@imgge.bg.ac.rs)

² Institute for multidisciplinary research, University of Belgrade, Kneza Viseslava 1, 11030 Belgrade, SERBIA

Silicon (Si) is the only known mineral element showing a protective effect for plants under stressful conditions. Copper (Cu) is a redox-active transition metal, as a protein cofactor involved in electron transfer reactions. Although being essential micronutrient for plants, in excess Cu initiates an oxidative stress. This study aiming to elucidate the mechanisms involved in Si protection of cucumber (*Cucumis sativus* L.) plants subjected to elevated Cu concentration. We analyzed the level of oxidative stress, expression of the related genes involved in an antioxidative defence, metal sequestration and phenolics biosynthesis, as well as the Cu concentration in different cell compartments.

Cucumber plants were grown hydroponically in a complete nutrient solutions supplied with either 0.2 μ M (control) or 10 μ M (high Cu) Cu, without or with addition of 1.5 mM Si. Lipid peroxidation levels were estimated by the TBARS assay. Gene expression were analysed by a Real time PCR method. Fractionated extraction of Cu in the tissues by subsequent centrifugations includes: water soluble, protein-bound, and cell wall-bound (weakly and strongly) fractions. Concentrations of Cu in all samples were determined by ICP-OES.

Silicon addition decreased lipid peroxidation, in high Cu-treated plants, as well as the expression of antioxidative enzymes, in roots and leaves, implying that the plants did not experienced the same level of oxidative stress. Key steps of the organic acids metabolism were not altered by Si-treatment, meaning that excess copper may not be deposited in vacuoles. PAL is the first enzyme in the biosynthesis of polyphenyl compounds and mainly involved in plant defence mechanisms. Interestingly, PAL expression level in leaves was increased manifold in high Cu + Si-treated plants. Copper content in roots and leaves was not affected by Si, while showed different distribution within the cell compartments. At the leaf level, Si promotes sequestration of Cu in protein and strongly bound cell wall fractions and reduces free Cu in water-soluble fraction. Our results indicate that Si nutrition stimulates synthesis of lignins and enhances deposition of Cu into the cell walls, thus preventing harmful effects of ROS in cucumber.

Theme 8 ORAL

54

Assessment of Silixol (OSA) Efficacy on Wheat Physiology: Growth and Nutrient content under Drought conditions

Passala R¹, Jain N², Deokate P P¹, Rao V², Minhas P S¹

 ¹ National Institute of Abiotic Stress Management (NIASM), ICAR, Malegaon, Baramati-Pune-413115, INDIA (ratnakumar@niam.res.in)
 ² Privi Life Sciences Pvt. Ltd, Privi House, A-71 TTC Industrial Area, Thane Belapur Road, Kopar Khairane, Mumbai-400706, INDIA

Drought is considered to be one of the most important agricultural predicament stress limiting wheat production. Silicon is present in the soil at concentrations ranging from 0.1 to 10%. It is beneficial for most of the higher plants as it has a proven role inmitigating stressful environments. Since, the mechanism in Si-alleviated damage caused by drought stress remains unclear, the present study was carried out to assess Silixol efficacy in alleviating drought stress in wheat.

To evaluate efficacy of Silixol on growth, physiology and nutrient content of wheat under drought, various concentration i.e. 0, 80, 160 and 320 ppm of foliar application of Silixol were used at different plant growth stages (vegetative, booting and seed development stage). Drought was imposed by withholding irrigation from booting to physiological maturity. Foliar application of 4 ml/L of Silixol (320 ppm) showed an impact in terms of increase in relative water content (RWC), leaf chlorophyll content and lower canopy temperatures. Root growth and Root length density were increased under both conditions with 4 ml/L foliar application and soil amended with Silixol.

K and P content in straw and seed of wheat were increased with application of Silixol and has strong significant (0.934) relationship in-between K content in straw and seed was observed under water stress conditions. The yield in terms of seed weight was increased with increasing Silixol treatment concentration in stress condition. The percent of increase was 5-10% over control under severe stress conditions. Therefore, Silixol had a strong impact on alleviating drought stress and minimizing the yield losses in wheat under drought conditions. Further studies are in progress to have an insight of mechanisms involved during the process.

Theme 1 KEYNOTE

55

Chemical studies of biosilicification and analytical methods for analysing silicon

Perry C C

Interdisciplinary Biomedical Research Centre, Nottingham Trent University, Clifton Lane, Nottingham NG11 8NS, UK (carole.perry@ntu.ac.uk)

Biosilicification in plants and other organisms is a complex process under biological control. As well as transport of the 'element' in some form through the plant, control over nucleation, growth, aggregation and cessation of growth is required in order to generate the wonderful silicified structures that are found in a wide range of plants. All processes occur in a largely aqueous environment and the silica structures that form vary from plant to plant and sometimes many different structures can be found within a single cell!

Using examples from the work of my research group and others, I will present information about the solution chemistry of 'silicon' pertinent to both the soil environment and to the environment in which silicified deposits form. I will provide background information on how silica structures form and how these structures can be modified by contact with an environment that includes ions and/or biomolecules. I will discuss methods that can be used to analyse for the presence of silicon in solution and the solid state. From our studies of silica produced in the laboratory and generated by living organisms I will highlight the importance of the interface between biomolecules and the mineral itself in controlling structure formation.

Theme	3
POSTE	R

56

Identification of putative Si transporters in ryegrass plants

Pontigo S¹, Gutiérrez-Moraga A^{2,3}, Ribera A^{2,3}, Cartes P^{3,4}

- ¹ Doctoral Program in Science of Natural Resources, La Frontera University, Temuco, 4811230, CHILE (<u>s.pontigo01@ufromail.cl</u>)
- ² Department of Agricultural and Livestock Production, La Frontera University, Temuco, 4811230, CHILE
- ³ Scientific and Technological Bioresource Nucleus (BIOREN), La Frontera University, Temuco, 4811230, CHILE
- ⁴ Department of Chemical Science and Natural Resources, La Frontera University, Temuco, 4811230, CHILE

Silicon (Si) provides benefits for many plants species, especially under stress conditions. These beneficial impacts can be variable due to plants differ in their ability to take up Si from the external solution, resulting in a great variation in Si accumulation among species. Molecular approaches underlying on Si uptake and transport in plants have revealed that these processes are mediated by different transporters that differ in their function, expression and localization in plant cells. Ryegrass, a forage species of great interest in Southern Chile, is able to accumulate relatively high Si concentrations. However, neither the uptake mechanisms have been characterized nor the transporters that mediate these processes have been identified yet. In order to identify the genes involved in the uptake and transport of Si in ryegrass, molecular cloning of genes codifying for Si transporters in this crop was performed.

Ryegrass (*Lolium perenne* L.) plants were grown in nutrient solution. After 15 days, roots were harvested to carry out the extraction of RNA and cDNA synthesis. Subsequently, partial fragments of cDNA were obtained by PCR using degenerated primers designed from conserved sequences among Si transporters identified in other plant species. PCR products were purified and sequenced.

A putative Si transporter gene that shared a sequence identity greater than 85% with homologues of the Si influx transporter Lsi1 of wheat, barley and maize was identified. We also found another putative Si transporter gene that presented a high homology (86%) with the Si efflux transporter Lsi2 from rice and barley. This work will contribute to the understanding of the Si uptake system of ryegrass plants. The knowledge of the molecular nature and the role these genes could lead to an improvement of productivity and quality of ryegrass cultivated under environmental stress conditions.

Acknowledgements: FONDECYT project 1120901, CONICYT Doctoral Scholarship 21120704 and Dirección de Investigación of Universidad de La Frontera (Temuco, Chile).

Theme 8 ORAL

57

From the pot to the plot: An extensive investigation into crop responses to a silica fertiliser

Prentice P

Agripower Australia Limited, 71 Macquarie Street, Sydney, 2011, AUSTRALIA (peterp@agripower.com.au)

Numerous studies on the beneficial effects of Silicon (Si) have been carried out worldwide historically focused on either a pot trial or a limited number of crops in particular regions. The increased yield with Si fertilisers has mainly been attributed to enhanced resistance to stress therefore field-based experiments are an important if not critical part of understanding the response of crops to a Si fertiliser. This field-based investigation aims to provide an overview of the responses of a variety of crops to Si under real-world stress and make recommendations for including Si fertilisers as a standard practice in crop production.

Over 100 field trials, the majority of which are replicated and managed by acknowledged research organisations, were conducted with a silica fertiliser in a variety of crops over two seasons and diverse regions and soils. These independently run field trials applied strict scientific protocols and experimental methods including soil and tissue analyses. In India, where crop nutrient and moisture stress are evident, 11 crops, repeated in 7 different regions, were trialled, ranging from well-known Si accumulators (e.g rice) to pomegranate and chilli. In Australia trials were carried out in in two distinctly different climatic regions and soils.

Results from two seasons of rice trials (total of 16 trials) with applied silica showed increased growth, yield and nutrient acquisition and availability. The uptake of N, P, K, Si and micronutrients in grain and straw in rice were significantly influenced by the silica treatment. In general, SFP (standard fertiliser practice) + 300kg/ha silica fertiliser gave significant increases in rice yield, with typically 600kg/ha required for the more acid soils. The application of silica to Bt cotton significantly (and positively) influenced the yield (increases of up to 20% were recorded), nutrient uptake, total chlorophyll content, percentage reddening and number of bolls per plant. In sugar cane trials where PAS levels were theoretically high, the application of Silica increased Si uptake by as much as 45% with an associated yield increase. Similar growth, yield and resistance to pest and disease trends to applied silica were also observed in non-Si accumulators such as tomato, chilli, onion, citrus and banana. The Plant Available Silicon (PAS) of the treated soils increased, irrespective of the starting Si status. The magnitude of yield increase over the SFP with the application of silica fertiliser varied between regions and varieties within each crop group. The magnitude of yield increase to applied silica could not be explained by the initial PAS status of the soil. It is concluded from this extensive field trial data set that environmental stress, including for example, the method of irrigation (moisture stress), plays an important role in determining the magnitude of yield and quality response to the applied silica. In order for Si fertilisers to be more widely applied in crop production a greater understanding is required of the relative importance of, for example, initial soil PAS and stress factors, on the resultant yield and quality of crops treated with Si fertilisers.

Business Meeting Aug 27th 2014 ORAL 58

Formation of a Global Silicon Community

Provance-Bowley M¹, Datnoff L²

¹ Harsco Metals and Minerals, 359 North Pike Road, Sarver, PA 16055, USA (mpbowley@harsco.com)

² Louisiana State University Agricultural Center, Department of Plant Pathology & Crop Physiology, Baton Rouge, LA 70803, USA

Since the first International Conference on Silicon in Agriculture was held in Fort Lauderdale, Florida, USA in 1999, knowledge of silicon's beneficial effects to plants and soils has increased dramatically. Assembled here is a diverse group of researchers and industry members, the torch bearers, united in a common goal, the time when silicon's use in agriculture becomes as commonplace as NPK fertilizers. Looking around the room you will notice that some colleagues are missing. This is not due to a lack of desire to attend, but mainly due to a lack of funding, affirming the need for further action in gaining full recognition and acceptance of silicon in agriculture.

What I am proposing today is the formation of an official Global Silicon Community whose main objectives are to gain full acceptance of silicon in the scientific, governmental, regulatory, and industrial communities, worldwide. This Global Silicon Community will act as a unified and influential voice for silicon.

It is our hope that you will join us in the creation of this community, realizing its necessity to the future of silicon research, marketability of silicon products, and sustainability of agricultural production worldwide. This community is long overdue.

Theme 1 ORAL

59

A Method for Determining Soluble Silicon Concentrations in Non-Liquid Fertilizer Materials

<u>Provance-Bowley M</u>¹, Sebastian D², Rodrigues H², Kinsey C², Korndörfer G³, Pereira H³, Buck G³, Datnoff L⁴, Miranda S¹

¹ Harsco Metals & Minerals, 359 North Pike Road Sarver, PA 16055, USA (mpbowley@harsco.com)

² Thornton Laboratories Testing & Inspection Services, Tampa, FL 33602, USA

³ Research Institute of Agricultural Sciences, Federal University, Uberlândia, 38400, BRAZIL

⁴ Louisiana State University Agricultural Center, Department of Plant Pathology & Crop

Physiology, Baton Rouge, LA 70803, USA

Silicon's beneficial effects in ameliorating abiotic and biotic stresses have been demonstrated in a wide range of agronomic and horticultural cropping systems. Despite this fact, the regulatory body that governs the labeling of fertilizers in the United States has been slow to recognize silicon as a plant nutrient. In 2012, this governing body, the Association of American Plant Food Control Officials (AAPFCO), designated silicon as a "plant beneficial substance". Prior to AAPFCO's approval, fertilizer and soil amendment labels were restricted to listing only "total" silicon content and crop managers had no way of selecting an appropriate silicon fertilizer based on plant available silicon. Further, regulators had no way of verifying and regulating fertilizer label claims of silicon content. A method was required to quantify the soluble (plant available) silicon concentrations and differentiate them from the insoluble SiO₂ (silica) portion in non-liquid silicon fertilizer sources.

Consequently, a number of extractors were compared in recovering plant available silicon from a number of solid sources and were correlated with plant silicon uptake. Sodium carbonate-ammonium nitrate ($Na_2CO_3 + NH_4NO_3$) was found to be the best extractant and a five-day method was developed using this extractant along with visible spectroscopy and heteropoly blue analysis for determining soluble silicon in non-liquid fertilizer products. The method can be applied to quantify the plant available silicon in solid fertilizer products at levels ranging from 0.2 to 8.4% silicon with an LOD of 0.06% and LOQ of 0.20%, but is not intended to quantify slow release silicon from nonliquid silicon fertilizers.

This method, recently published (2013) in the Journal of AOAC International Vol. 96 Issue 2, pp. 251-259, can now be used for quality control during production of silicon fertilizers, accurate fertilizer labeling, label content compliance monitoring, and in selection of an appropriate silicon fertilizer source to meet crop production needs. Fertilizers, now labeled to contain beneficial substances such as silicon may now be sold in the United States.

Theme 6 POSTER 60

Sorghum resistance to anthracnose mediated by silicon

<u>Resende R S</u>, Rodrigues F Á

Federal University of Viçosa, Department of Plant Pathology, Laboratory of Plant-Pathogen Interaction, Viçosa, Minas Gerais, Zip Code 36570-900, BRAZIL (resenders@yahoo.com.br)

The use of silicon (Si) in agriculture has attracted a great deal of interest from researchers because of the numerous benefits of this element to plants, especially when they are submitted to abiotic and/or biotic types of stress. The host's increased resistance to diseases, promoted by Si, is mainly associated with the deposition of this element in the tissues and the potentiation of defence mechanisms. However, the mechanisms involved in Si-mediated host resistance need to be further investigated. Thus, this study aimed to microscopically and biochemically elucidate the resistance of sorghum to anthracnose.

The severity was assessed at 2, 4, 6, 8 and 10 days after inoculation. Samples for the analysis for Si concentration, x-ray microanalysis of Si deposition in the leaf epidermis and scanning electron microscopy were collected at 120 hours after inoculation (hai). For the determination of enzyme activities like peroxidases, polyphenoloxidases, chitinases and β -1,3-glucanases, the concentrations of total soluble phenolics and lignin-like phenolic polymers, the samples were collected at 24, 48, 72 and 96 hai with *C. sublineolum*. For the determination of the concentration of anthocyanins, the leaf samples were collected at 120 hours hai. Leaf samples collected from non-inoculated plants served as the controls.

In the leaves of plants supplied with Si, in addition to a greater deposition of Si at the infection sites, the acervuli were smaller in number compared to the leaves of plants not supplied with Si. Additionally, the activities of the defence enzymes peroxidases and polyphenoloxidases and the concentration of anthocyanins were higher in the leaves of plants supplied with Si. It can be concluded that Si, in addition to participate in the physical barrier that slows or prevents *Colletotrichum sublineolum* penetration in sorghum leaves, also plays a role in the biochemical aspect of sorghum resistance to anthracnose.

Theme 6 ORAL

61

Silicon alters the volatile profile of pest-infested grapevines and increases attractiveness to predators

<u>Reynolds O L¹</u>, Connick V J², Simmons A T³, Guisard Y², Nicol H I⁴, An M⁵, Gurr G M^{6,4}

¹ Graham Centre for Agricultural Innovation (CSU and NSW Department of Primary Industries), Elizabeth Macarthur Agricultural Institute, Private Bag 4008, Narellan NSW 2567, AUSTRALIA (olivia.revnolds@dpi.nsw.gov.au)

² National Wine & Grape Industry Centre, Charles Sturt University (CSU), PO Box 883, Orange NSW 2800, AUSTRALIA

³ School of Agriculture & Wine Sciences, CSU, PO Box 883, Orange NSW 2800, AUSTRALIA

⁴ Graham Centre for Agricultural Innovation, CSU, PO Box 883, Orange NSW 2800, AUSTRALIA ⁵ Environmental and Analytical Laboratories, Faculty of Science, CSU, Wagga Wagga, NSW

2650

⁶ Fujian Agriculture & Forestry University, Fuzhou, CHINA

Plant induced defence mechanisms are generated upon attack by herbivore feeding, arthropod oviposition or disease. Silicon (Si) directly enhances plant resistance to such stressors by acting as a signal for induced chemical defences in plants. Two studies were conducted of the effects of Si fertilisation of grapevines on induced plant defence.

A Y-tube olfactometer was used to investigate the attraction of the predatory beetle, Dicranolaius bellulus (Guérin-Méneville) (Coleoptera: Melyridae) to plants infested with Epiphvas postvittana (Walker) (Lepidoptera: Tortricidae) larvae. In a separate study, the direct effect of silicon fertilisation on the volatile profile of grapevine plants infested with Phalaenoides glycinae (Lewin) (Lepidoptera: Noctuidae) was determined using solid-phase micro-extraction (SPME)/gas chromatography-mass spectrometry (GC-MS) analysis.

Insect responses showed significantly increased attraction of D. bellulus to E. postvittanainfested plants that had the highest silicon tissue content. GC-MS analysis identified seven volatile compounds emitted from *P. glycinae*-infested grapevines, with *n*-heptadecane produced in significant amounts only by silicon treated plants. Cis-thio rose oxide production was significantly lower in silicon treated grapevines. Results suggest that silicon fertilisation may affect the HIPV production and attractiveness of grapevines to natural enemies. These results are the first demonstration of the direct effect of silicon fertilisation on the volatile profile of a pest-infested plant.

Theme 5 POSTER

62

Effect of silicon addition on rice (Oryza sativa) plants grow under zinc deficiency

Rivera V, Hernández-Apaolaza L

Agricultural Chemistry Department, Autonomous University of Madrid, 28049 Madrid, SPAIN (lourdes.hernandez@uam.es)

Silicon is the second most abundant element in the earth's crust. In soil solution at pH below 9.0, the prevailing form is monosilicic acid $Si(OH)_4$. Is considered an essential element in several crops, enhancing grow and creating a guard from biotic and abiotic stresses. On the other hand, zinc is an essential micronutrient for normal growth and plants development and plays multiple roles in basic biochemical processes such as chlorophyll production. Zn deficiency is very common in calcareous soils and the most common symptoms are discoloration of lower leaves (brown strips and spots). Among others, one of the strategies that plants use to alleviate these symptoms is lowering the pH of the rhizosphere. In this work, the role of Si in the alleviation of Zn deficiency in rice plants has been studied.

For that purpose, rice (*Oryza sativa* cv marisma) was grown for two weeks in a hydroponic culture in which different concentrations of Si (0.0, 1.0 and 2.0mM) were applied at calcareous soil pH conditions. On the third week Zn was taken out from the nutrient solution and plants were collected daily for 5 days. Chlorophyll a and b, fresh and dry weight, pH of the nutrient solution, and Zn in the apoplasm were measured.

Statistical differences were observed in plant growth parameters (fresh and dry weight) and plants treated with Si 2mM show the highest values. Moreover a protective effect of Si in Chlorophyll a and b has been seen in Si 1mM and 2mM treatments, the first one shows the highest values. Apoplasmic Zn did not seem to be correlated with the beneficial Si effect, due to no differences on its content were detected with or without Si supply to the plants. The Si addition tends to decrease less the nutrient solution pH. In general terms we observed an important beneficial effect of Si in rice grow under Zn deficiency.

Theme 6 POSTER 63

Histochemical Aspects of Wheat Resistance to Leaf Blast Mediated by Silicon

Rodrigues F Á, da Silva W L, Cruz M F A, Fortunato A A

Viçosa Federal University, Department of Plant Pathology, Laboratory of Host-Parasite Interaction, Viçosa, Minas Gerais State, , Zip Code 36570-900, BRAZIL (fabricio@ufv.br)

Blast, caused by the fungus *Pyricularia oryzae*, has become an important disease of wheat in Brazil and in other countries in South America. This study aimed to investigate, at the histochemical level, whether silicon (Si) could enhance the production of flavonoids in the leaves of wheat plants in response to *P. oryzae* infection.

Wheat plants of the cultivar Aliança, which are susceptible to blast, were grown in hydroponic culture containing 0 (-Si) or 2 mmol of Si (+Si) and inoculated with *P. oryzae*.

The foliar Si concentration was significantly higher in +Si plants (3.6 dag/kg) in comparison to -Si plants (0.26 dag/kg). The X-ray microanalysis revealed a different pattern of Si deposition between the leaves of -Si and +Si plants. In +Si plants, Si was deposited in greater amounts and in a linear pattern that corresponded to silica cells, compared to scarce Si deposition in the leaves of -Si plants. This increased Si reduced blast severity on leaves. At 72 hai, hyphae of *P. oryzae* grew successfully and formed an extensively branched mycelium in the first-penetrated epidermal cell and invaded many neighbouring cells in the leaves of -Si plants. In contrast, fungal hyphae were restricted to the first-penetrated epidermal cell in +Si plants. Strong fluorescence was detected in the epidermal and parenchyma cells as well as in the vascular vessels of the leaf sections of the +Si plants in contrast to the -Si plants after using Neu's and Wilson's reagents. There was no difference between the leaf sections of the – Si and +Si non-inoculated plants. The results of the present study support the notion that active Si uptake by wheat is extremely important in enhancing host resistance to blast, in which the phenylpropanoid pathway plays a major role.

Theme 3 POSTER

64

The role of Casparian strips and Lsi transporter distribution in efficient Si transport in rice

Sakurai G¹, Satake A², Yamaji N³, Mitani N³, Yokozawa M⁴, Ma J F³

¹ Ecosystem Informatics Division, National Institute for Agro-Environmental Sciences, 3-1-3 Kannondai, Tsukuba, 305-8604, JAPAN (sakuraigen@affrc.go.jp)

² Creative Research Initiative "Sousei", Hokkaido University, N21, W10, Kita-ku, Sapporo, 001-0021, JAPAN

³ Research Institute for Bioresources, Okayama University, 2-20-1 Chuo, Kurashiki, 710-0046, JAPAN

⁴ Graduate School of Engineering, Shizuoka University, 3-5-1 Johoku Naka-ku, Hamamatsu, 432-8561, JAPAN

Rice, a typical siliciphilous plant, can accumulate silicon to over 10% of its dry mass. In rice roots, the passive transporter Lsi1, which belongs to the aquaporin family, and the active transporter Lsi2 are important for silicon uptake from soil. Both transporters are localized to the plasma membrane of both exodermal and endodermal cells, where Casparian strips prevent apoplastic transport into the root stele. Lsi1 is located at the distal side of the cells, whereas Lsi2 is located at the proximal side. This pattern differs from that in other plant species; for example, in maize and wheat, Lsi2 homologs do not show polar localization and passive transporters are located in the root cortex. The following questions remain to be answered: (1) How can rice take up large amounts of silicon from soil? (2) Why do rice silicon transporters have such a characteristic localization pattern?

To simulate silicon uptake by rice roots, we developed a mathematical model based on a simple diffusion equation. The model parameters were calibrated by using *in vivo* experimental data on silicon concentration in the xylem sap and Markov chain Monte Carlo methods. The resulting model predicted the silicon concentration in the xylem sap of *lsi1* and *lsi2* mutants. Using this model in simulation experiments, we compared (1) the silicon uptake ability of the roots lacking Casparian strips with that of normal roots, and (2) the silicon uptake efficiency of various types of roots with different localization patterns of Lsi transporters.

The silicon uptake ability of the roots lacking external Casparian strips was lower than that of normal roots. This result suggests that the structure of the double layer of Casparian strips in rice is important for the high silicon concentration in the sap. The most efficient Lsi localization pattern was identical to that of wild-type roots. This suggests that evolution of the characteristic distribution of Lsi transporters in ricehas achieved a high cost-to-benefit ratio in silicon transport.

Theme 4 ORAL

65

Hormonal changes and growth of rice plants in response to bioagents and silicon fertilization application

<u>Silva B G¹</u>, Sousa P T¹, Souza A C A², Filippi M C³

¹ Amazonia Federal Rural University, Avenue Presidente Tancredo Neves, Belém, 66077-830, BRAZIL (gisele.barata@ufra.edu.br)

² Master in Agronomy, Street José Needermeyer, Goiânia, 74360-340, BRAZIL Goiás Federal University, Highway GO-462, Goiânia, 74800-000, BRAZIL

³ EMBRAPA Rice and Beans, Highway GO-462, Santo Antônio de Goiás, 74800-000, BRAZIL

The demand for increased productivity of rice requires sustainable agricultural practices such as the combination of bioagents and Silicon fertilization, which has been shown that promotes plant growth. The objective was to determine the changes in growth, physiological and biochemical patterns of rice plants when treated with rhizobacteria (*Burkholderia pyrrocinia*: R-46; *Pseudomonas fluorensces*: R-55), *Trichoderma asperellum* (Ta) and fertilized with calcium silicate and magnesium (Ca Si Mg).

Three assays, E1, E2 and E3 with three replications were conducted. E1: 16 treatments combined four Ca Si Mg (1,2,4 and 8 t ha-1) doses, three bioagents (R-46, R-55 and Ta) and control; E2 and E3: the E1 top six treatments were evaluated for total sugar content, nitrate reductase activity, four hormones (indole-3-acetic acid (IAA), gibberellic acid, salicylic acid and jasmonic acid) contents, physiological parameters (CO₂ assimilation, transpiration; stomatal conductance, water use efficiency) and photosynthetic pigments.

Plants fertilized with 2 t ha⁻¹ Ca Si Mg and treated with all three bioagents showed higher biomass (68%), total sugar and gibberellin content, CO₂ assimilation and improved efficiency in water use. However, this same treatment also presented, a lower nitrate reductase activity, plant transpiration and AIA contents. The best contents of jasmonic (28%) were observed in plants treated only with bioagents Ta. The synergistic interaction between silicon fertilization and bioagents promoted rice plants growth through physiological changes and hormonal modulation.

Finacial Support: CNPq/Fapespa

Theme 5 POSTER

66

The effect of silicon on some physiological parameters of maize leaves under antimony stress

Šimková L, Vaculíková M, Fialová I, Luxová M

Department of Plant Physiology, Institute of Botany, Slovak Academy of Sciences, Dúbravská cesta 9, Bratislava, 845 23, SLOVAKIA (sprekelia@gmail.com)

Antimony (Sb) is a naturally occurring toxic element present mainly as Sb_2S_3 and Sb_2O_3 in the Earth's crust. Sb gets into the soil mainly through mining and smelting of ores. In many regions of the world contamination of soils with Sb is a great problem. Antimony is potentially toxic at very low concentrations and inhibits plant growth. Heavy metals and toxic elements can induce oxidative stress in higher plants by producing reactive oxygen species. Silicon (Si) is, after the oxygen, the second most abundant element in the Earth's crust. Normally, Si is in the form of H_4SiO_4 . Silicon is not considered as an essential element for higher plants. Nevertheless, the research has shown that Si can mitigate the negative effect of biotic and abiotic stresses on plants. Alleviation of stress on plants by stimulating the activity of antioxidative enzymes was also confirmed. The aims of this work were to study the effect of Sb on the activity of two antioxidative enzymes catalase (CAT) and peroxidase (POX), the content of non enzymatic antioxidant ascorbate as well as soluble proteins and protein pattern in maize aboveground parts. Also the effect of Si on stressed 1st and 2nd leaves was observed.

The seeds of two maize hybrids (*Zea mays* L.), cv. Almansa and Novania, were germinated in dark for 3 days at 24 °C. Three day-old seedlings were transferred to Hoagland nutrient solution and grown under 16/8 h photoperiod with 300 μ mol m⁻² s⁻¹, temperature 24/22 °C. Young plants were treated by Sb and/or Si. (control, 10 mg l⁻¹ Sb, 50 mg.l⁻¹ Sb, 5 mM Si, 10 mg.l⁻¹ Sb + 5 mM Si, 50 mg.l⁻¹ Sb + 5 mM Si). We analyzed first and second fully developed leaves. CAT, APX activities, the content of ascorbate and soluble proteins were determined spectrophotometrically. Protein pattern was obtained by SDS-PAGE.

At higher Sb concentration the activity of CAT increased in Almansa and in the 1st leaf of Novania in comparison with control. The increase of POX activity was observed only in Novania. Differences in enzymes activities could be explained through determination of H_2O_2 content. Antimony increased the content of ascorbate in both maize hybrids. Higher concentration of Sb had a significant effect on studied parameters than lower Sb concentration. In some cases, Si mitigated the increase of CAT or POX activities and content of ascorbate, too. The opposite effect of Si was recorded in 2nd leaf of Novania. Although Sb affected the content of soluble proteins only slightly, significant changes in protein profiles in the aboveground parts of the stressed maize plants were observed. Higher accumulation of proteins with molecular weight around 65 kDa in the treatment with higher Sb concentration was recorded. Si had no effect on content of soluble proteins and Si treatment had no effect on protein accumulation in Sb stressed plants.

Acknowledgement: This work was supported by the Slovak Research and Development Agency under the contract No. APVV-0140-10 and was part of VEGA 1/0817/12 project.

Theme 2 ORAL

67

Silicate fertilization, crop production and carbon storage: an enhanced weathering approach

<u>Struyf E</u>¹, Schoelynck J¹, Belmonte D U¹, Goddéris Y², Weiss A³, Hartmann J³, Meire P¹

¹ University of Antwerp, Department of Biology, Ecosystem Management, Universiteitsplein 1C, 2610 Wilrijk, BELGIUM (eric.struvf@uantwerpen.be)

² Géoscience Environnement Toulouse, Avenue Edouard Belin 14, 31400 Toulouse, FRANCE

³ University of Hamburg, Bundesstrasse 55, 20146 Hamburg, GERMANY

Artificially enhanced weathering of olivine has recently been proposed as a potential geoengineering and terra-forming technique to reduce atmospheric CO_2 concentrations on relatively short timescales. Olivine (Mg₂SiO₄) is an easily weathered silicate, with wellknown dissolution kinetics. The silicate weathering process consumes CO_2 and is an important control on atmospheric CO_2 -concentrations. The associated Si fertilization can be of major importance for future crop production. In long-term cultivated land, soils have been depleted for bio-available silica pools, due to the long-term harvest of silica rich crops, such as wheat, rice, sugarcane, grass for hay and maize. Si-C interactions in cultivated soils have remained virtually unstudied. Detailed carbon balances in different enhanced weathering scenarios have never been performed, and state-of-the-art knowledge does not allow the construction of a mechanistic understanding of Si-C flows and interactions in different enhanced weathering scenarios. The aim of our experiment is to test whether the currently mostly theoretical concept of olivine fertilisation, can work in an actual soil environment. We test both the effect on soil biogeochemistry and crop production.

Since October 2013, we have initiated a fully-replicated setup (5 replicates per treatment combination), where application of olivine, olivine grain size, crop type (wheat and barley) and rain regime (continuous vs. event-like) are controlled factors. We are studying in detail soil pH, dissolved Mg, Fe, Al and Si in soil and infiltration water (as proxy for olivine weathering and formation of secondary silica precipitates), dissolved inorganic carbon and dissolved metals (to track potential release of toxic byproducts, including Ni, and to model weathering reactions) every month. We will also analyse C, Mg, Si, Fe, Ni, Al and other metal content in the biomass after harvest and soil reactive Si (biogenic Si, amorphous Si precipitates, adsorbed Si to Fe/Al hydroxydes, reactive secondary silicates) in the soil before and after the experiment, as well as particulate (in)organic C in the soil before and after the experiment. Surface area, micropore and macropore volume distribution as well as surface area composition and clay mineral abundance analysis are planned.

Here we present the first results on harvested biomass, soil and soil water biogeochemistry and soil characteristics for the experiment. Although the results are in full development, as the experiment is still running, strong alterations in soil Mg and Si availability are clear, and the formation of an amorphous Si layer in small grain olivine $(63\mu m)$ is apparent. By the time of the conference, it will be possible to provide an initial assessment of the applicability of this novel concept.

Theme 2 ORAL

68

Silicon fertilization and its impacts on phosphorus availability: a way to mitigate phosphorus fertilizer scarcity in the future?

Tallberg P

Department of Food and Environmental Sciences, PO Box 27, FIN-00014, University of Helsinki, FINLAND (petra.tallberg@helsinki.fi)

Feeding the world's growing population properly is one of the major challenges of the future. The availability of phosphorus (P, one of the major nutrients needed in crop production) for fertilizer use is a very important part of this challenge: As the availability of P decreases and its price increases, recycling and improvements in the efficiency of P use will become more and more important. As over-fertilization by P has also led to extensive water quality problems due to excess run-off, improving the crop utilization of P already present in the soil would have definite, multiple benefits. Silicon (Si) fertilization is an interesting method with potential to improve the utilization of P in-field through competitive ligand exchange. This process could decrease the need for external P fertilization and diminish P runoff. At the same time, the increased availability of Si has the potential to improve the condition of the plants involved and their resistance towards a multitude of diseases and pests. This study presents a conceptual framework for addressing this novel and promising but also very challenging question: "Can we enhance P availability through Si fertilization and at the same time improve crop quality while preventing watercourse eutrophication?"

In this pilot phase of the study the potential advantages of Si fertilization will be analysed utilizing existing data on Si-P interactions in soil, P fertilization practises and estimated P fertilizer price development and a simple model constructed. Later, the limited data on Si fertilization and its potential impacts on P will be augmented in laboratory and field experiments, where different Si fertilizers will be added to different soil types and the effects of Si and Si-P interactions on common agricultural crop plants included. Finally, the Si-P model will be improved and finalized for predictive use in fertilization application and the impacts of changes in Si and P runoff estimated.

The first results from the project will consist of a model providing a rough estimate of the potential economic usefulness of Si fertilization as a surrogate for P fertilization under different circumstances. Later on, the project will also provide us with a better understanding of the so far incompletely studied processes affecting the movements of Si in soil. Finally, we will provide some more insights into possible ways of ameliorating nutrient-induced eutrophication in water bodies subjected to agricultural runoff.

Theme 2 ORAL

69

Changes on Plant-Available Silicon Level of Selected Soils from the Midwest and South USA Applied with Different Rates of Silicate Slag

<u>Tubana B</u>¹, Datnoff L², Narayanaswamy C¹, White B¹, Babu T¹, Yzenas J³

¹ School of Plant, Environmental, and Soil Sciences, Louisiana State University, 104 MB

Sturgis Hall, Baton Rouge, Louisiana, 70803, USA (btubana@agcenter.lsu.edu)

² Department of Plant Pathology and Crop Physiology, Louisiana State University, 302 Life

Science Bldg., Baton Rouge, Louisiana, 70803, USA

³ Plant Tuff Inc., Edward Levy Corporation, Dearborn, Michigan, 48120, USA

The benefits of fertilizer application are optimized if done at times where the release of available nutrient coincides with the active growth of crop and the amount being at sufficient level to meet crop's demand. Plant-available silicon (Si) in soil can be raised through the application of silicate slag. This study was conducted to document the release pattern of plant-available Si from silicate slag applied to selected soils from the Midwest and South USA.

Bulk soil samples were collected from different field locations in Indiana, Mississippi, Ohio, Michigan, and Louisiana, USA. These soils were air-dried and processed to pass a 5-mm sieve before placing 2-kg portions to plastic pots. The treatments consisted of different Si rates of 0, 170, 340, 680, 1020, and 1360 Si kg ha⁻¹ applied as silicate slag (17% Si). Two checks were also included, with (2 Mt ha⁻¹) and without lime. All treatments were replicated four times and arranged in a randomized complete block design. Treated soils were grown to ryegrass for 90 days followed by wheat until 120 days after silicate slag application (DAI). Sequential sampling of soil and biomass was done within the 120-day period with 30-day interval. Silicon concentration in soil sample extracts (1:10 w/v, soil:0.5 M acetic acid solution) was quantified via Molybdenum Blue Colorimetry (MBC) while the amount of Si removed by plants was quantified by digesting 100 mg oven-dried, ground samples using the Oven Induced Digestion method with NaOH and H₂O₂ followed by MBC.

Initial plant-available Si levels were markedly different among soil types ranging from 22 to 165 ug Si g⁻¹. The plant-available Si of soils with high organic matter (OM, 3 to 5%) was raised by 0.47 ug g⁻¹ for every unit of Si (ug g⁻¹) applied, the highest rate achieved across soil types and sampling times. For soils with silt loam texture and moderate amount of OM (1 to 2%), the amount of Si applied transformed to plant-available Si increased with the number of DAI, peaked at 90 DAI then declined at 120 DAI. On the other hand, the pattern of plant-available Si released from silicate slag showed a distinct and steady decline with the number of DAI for soils containing large fractions of either sand or clay suggesting the reduction was perhaps related to either Si losses by leaching or high Si adsorption rate by the soil, respectively. Significant effect of Si application on Si uptake of ryegrass or wheat was observed only on several soils; those generally characterized as having low extractable Si (<67 ug g⁻¹), low pH, high OM, and coarse-textured soil. Both clay and OM content of the soil influence the required time at which the maximum amount of plant-available Si from applied silicate slag is achieved and the length of time to maintain this level.

Theme	5
ORAL	

70

Influence of silicon on zinc toxicity in wheat

Vaculík M^{1,2}, Vaculíková M^{3,4}, Lux A¹, Frossard E², Schulin R⁴

¹ Department of Plant Physiology, Faculty of Natural Sciences, Comenius University in

Bratislava, Mlynska dolina B2, SK-842 15, Bratislava, SLOVAKIA, (vaculik@fns.uniba.sk)

³ Institute of Botany, Slovak Academy of Sciences, Dubravska cesta 9, SK-845 23 Bratislava, SLOVAKIA

Although silicon (Si) is not an essential element for plants in general, the beneficial effect of this element has been demonstrated for many plant species, especially from Poaceae family. In addition to the importance of Si in plant nutrition, optimal growth and development, it plays a significant role in the alleviation of various symptoms of biotic as well as abiotic stresses. It was shown that Si could decrease the negative effects of various heavy metals and toxic elements like Cd, Pb, Cr or As and prevent their uptake into the plant tissues. Zinc (Zn) is an essential microelement and is required by plants for normal growth and development, however it becomes toxic when present in excess. Relatively less is known about the effect of Si on Zn in plants, and available results are contradictory. Although Si was shown to alleviate the toxicity caused by Zn in rice, opposite was observed in maize. Therefore, the aim of this contribution was to investigate the effect of Si on wheat – one of most important agricultural plants, when grown in excess of Zn.

Wheat seedlings (*Triticum aestivum*, cv. Sensas) were germinated for four days in wet filter paper in dark and cultivated in hydroponics in modified Hoagland solution in controlled environment for additional 18 days at 16/8 photoperiod, 24/18°C, and relative humidity 70%. Six treatments were used: Control, Si – addition of 1.25 mM sodium silicate solution (27% SiO2 dissolved in 14% NaOH), Zn100 and Zn 400 – addition of 100 or 400 mM ZnSO₄ x 7H₂O, respectively, Zn 100 + Si and Zn 400 + Si - combination of Zn and Si in the concentrations as mentioned previously. Fresh and dry biomass was recorded and the concentration of Zn was measured in roots and shoots by ICP-OES. Root morphology was analysed using Whin Rhizo system and in the leaves chlorophyll and carotenoids concentrations were evaluated.

We found that Zn significantly reduced both root and shoot biomass and application of Si to Zn-treated plants enhanced plant biomass, especially in the shoot. Similarly, addition of Si decreased the concentration of Zn in root and shoot tissues at lower Zn (Zn100), however no significant differences were observed in higher Zn (Zn400). Additionally, changes in the chlorophyll and carotenoids concentrations confirm our findings that Si can partially mitigate the negative effect of Zn in wheat plants.

The work was supported by APVV-0140-10, VEGA 1/0817/12 and Sciex Project 13.023.

² Institute of Agricultural Sciences, Department of Environmental System Science, ETH

Zürich, Eschikon 33, CH-8315, Lindau, SWITZERLAND

⁴ Institute of Terrestrial Ecosystems, Department of Environmental System Science, ETH Zürich, Universitaetstrasse 16, CH-8092 Zürich, SWITZERLAND

Theme 5 POSTER 71

Growth and antioxidative response of maize roots exposed to antimony and silicon

Vaculíková M^{1,3}, Vaculík M^{2,3}, Šimková L¹, Fialová I¹, Luxová M¹, Tandy S³

¹ Department of Plant Physiology, Institute of Botany, Slovak Academy of Sciences,

Dúbravská cesta 9, Bratislava, 845 23, SLOVAKIA (vaculikova.miroslava@gmail.com)

Pollution of antimony (Sb) raises a serious environmental problem. This element can accumulate in plants that grow at contaminated sites and in this way Sb can enter the food chain. Although this non-essential element can be taken up by roots and accumulated in plant tissues in relatively high concentrations, there is still a lack of knowledge about the effect of Sb on biochemical end metabolic processes in plants. It was shown that the application of silicon (Si) could decrease the toxicity of some heavy metals and toxic elements in various plants. The aim of this work was to assess how Si influences the growth and antioxidative response of young maize roots exposed to elevated concentrations of Sb.

Three days old seedlings of maize (*Zea mays* L., hybrid Novania) were cultivated hydroponically for 10 days in solutions with 10 or 50 mg L^{-1} Sb^{III} in the form of the tartrate complex and/or 5 mM Si (sodium silicate). Growth parameters of roots (the total length of primary seminal roots, fresh and dry weights of biomass) were determined. The content of proline, malonedialdehyde and activity of some enzymatic (catalase, ascorbate peroxidase, guaiacol peroxidase) and non-enzymatic (ascorbate) antioxidants were determined. The availability of Sb from the nutrient solution was evaluated by Visual MINTEQ aqueous speciation software. Concentrations of Sb and Si in root and shoot were measured.

We found that Sb toxicity in plants increased with increasing Sb concentration in the medium. Antimony reduced the root growth and induced oxidative stress and activates antioxidant defense mechanisms in maize. Silicon addition to Sb treated roots decreased lipid peroxidation, proline accumulation, and the activity of antioxidative enzymes (ascorbate peroxidase, catalase and guaiacol peroxidase), however it increased the activity of nonenzymatic antioxidant ascorbate in maize roots. Although no positive or negative effect of Si was observed on root length or biomass, changes in the oxidative response of plants exposed to Sb indicate a possible mitigation role of Si on Sb toxicity in plants.

Acknowledgement: The work was supported by grants Nr. APVV 0140-10 and was part of VEGA 1/0817/12 project. The presentation of the results is part of the Sciex project Nr. 13.024.

²Department of Plant Physiology, Faculty of Natural Sciences, Comenius University in

Bratislava, Mlynská dolina B2, Bratislava, 842 15, SLOVAKIA

³ Institute of Terrestrial Ecosystems, ETH Zürich, Universitätstrasse 16, 8092 Zürich, SWITZERLAND

Theme 6 ORAL

72

Transcriptome analysis of silicon-induced brown spot resistance in rice reveals central role of photorespiration

Van Bockhaven J¹, Kikuchi S², Asano T², Höfte M¹, De Vleesschauwer D¹

¹ Lab of Phytopathology, Ghent University, Coupure Links 653, Ghent, 9000, BELGIUM (jonas.vanbockhaven@ugent.be)

² Plant Genome Research Unit, National Institute of Agrobiological Sciences Tsukuba 305, Ibaraki, 8602, JAPAN

In face of the ever-increasing loss of arable land, shrinking fresh water resources and global climate change, feeding the world's burgeoning population in coming decades will be a daunting task. One of the solutions to safeguard global food security is to minimize yield losses caused by biotic and abiotic stresses. Over the past decades, application of silicon (Si) has emerged as a promising disease control strategy that is able to protect plants against a broad range of microbial pathogens, among which the necrotrophic rice brown spot fungus *Cochliobolus miyabeanus*. However, despite this huge potential, relatively little is known about the precise mechanisms by which Si induces broad-spectrum disease resistance

In an attempt to unveil the molecular underpinnings of silicon-induced disease resistance, we studied the transcriptome of control and silicon-treated rice plants subjected to *C. miyabeanus* infection using Agilent 44K *oligo DNA arrays*. The data obtained were analysed using a variety of bioinformatic approaches suitable for rice functional genomics.

Brown spot development is hallmarked by the formation of necrotic disease lesions surrounded by extensive chlorosis. Interestingly, analysis of brown-spot infected plants suggested that C. mivabeanus actively represses photosynthetic processes in order to trigger premature senescence and, hence, inflict disease. In silicon-treated plants, however, this pathogen-induced suppression of photosynthesis was strongly impaired, suggesting that Si alleviates biotic stress imposed by the pathogen. Further analysis of the effect of silicon in brown spot infected leaves demonstrated significantly increased levels of photorespiration in these plants. Even though photorespiration is often considered a wasteful process in C3 plants like rice, recent studies indicate that this metabolic bypass also indirectly enhances resistance during abiotic stress and pathogen attack by protecting the plant's photosynthetic machinery. Taking these facts into account, our findings favor a scenario whereby Si enhances brown spot resistance at least in part by boosting photorespiration-associated metabolism, thereby counteracting C. mivabeanus-induced senescence and cell death. Moreover, our results shed light onto the mechanistic basis of Si-afforded disease control and support the view that in addition to activating plant immune responses, Si may also reduce disease severity by interfering with pathogen virulence strategies.

Theme 2 ORAL

73

Grazers: bio-catalysts of terrestrial silica cycling.

Vandevenne F¹, Barao A L¹, Schoelynck J¹, Smis A¹, Ryken N², Van Damme S¹, Meire P¹, Eric Struyf¹

¹ Department of Biology, Universiteitsplein 1, Wilrijk, 2610, BELGIUM (floor.vandevenne@uantwerpen.be)

² Department of Soil Management, Coupure Links 653, Gent, 9000, BELGIUM

Silica is well known for its role as inducible defence mechanism countering herbivore attack, mainly through precipitation of opaline, biogenic silica (BSi) bodies (phytoliths) in plant epidermal tissues. Grasslands in particular play an important role in global silica cycling, with a high capacity to store BSi in soils and by enhancing Si mobilisation through mineral weathering. Even though grazing strongly interacts with other element cycles, its impact on terrestrial silica cycling has never been thoroughly considered. This study assessed the impact of grazing on the intrinsic reactivity and dissolvability of BSi in cultivated grasslands (pastures).

BSi content of ingested grass, hay and faeces of four common domestic herbivores was quantified by performing multiple chemical extraction procedures for BSi, allowing the assessment of chemical reactivity. Dissolution experiments (24 hours) with rainwater were carried out to measure direct availability of BSi for dissolution, both in fresh grass and faeces samples.

Average BSi and readily soluble silica numbers were two to four times higher in faeces as compared with grass or hay, and differences between herbivores could be related to distinct digestive strategies. Reactivity and dissolvability of BSi increases after digestion, mainly due to degradation of organic matrices, resulting in higher silica turnover rates and mobilization potential from terrestrial to aquatic ecosystems in non-grazed versus grazed pasture systems (2 versus 20 kg Si ha⁻² year⁻²). Our results suggest a crucial yet currently unexplored role of herbivores in determining silica export from land to ocean, where its availability is linked to eutrophication events and carbon sequestration through C-Si diatom interactions.

Theme 4 ORAL

74

Effect of Enhanced Soil Silicon on Grain Yield and Agronomic Parameters of Wheat under Sufficient and High Nitrogen Application Rates

White E B¹, Tubana S B¹, Dupree P¹, Yzenas J², Datnoff L³, Mascagni H Jr⁴

¹ School of Plant, Environmental, and Soil Sciences, Louisiana State University, 104 M.B.

Sturgis Hall, Baton Rouge, Louisiana 70803, USA (bwhit56@lsu.edu)

² Plant Tuff Inc., Edward C. Levy Corporation, Dearborn, Michigan, 48120, USA

³ Department of Plant Pathology and Crop Physiology, Louisiana State University, 302 Life

Science Bldg., Baton Rouge, Louisiana 70803, USA

⁴ Northeast Research Station, LSU Ag Center, St. Joseph Louisiana, 71366, USA

Silicon (Si) fertilization can potentially counteract the negative effects of excessive nitrogen (N) application by increasing plant resistance to lodging and biotic and abiotic stresses. Research has been conducted on the interactions of Si and N, but is limited to rice production. This study aims to establish the application rate for calcium silicate (CaSiO₃) slag for wheat production in Louisiana (LA) and elucidate the effects of Si fertilization on wheat supplied with sufficient and high rates of N fertilizer.

This study was initiated in 2012 on soils along the alluvial flood plain of LA. The treatments consisted of two N rates (urea) and five Si rates with two control treatments (lime and no lime) arranged in a randomized complete block design. Biomass samples were taken at different growth stages along with grain samples and yield components at harvest. All plant samples were analyzed for Si content via Oven Induced Digestion procedure followed by Molybdenum Blue Colorimetry and for nutrient content by ICP optical spectrometry. Soil samples taken at Feekes 10.5 and harvest were analyzed for Si content and Mehlich-3 extractable nutrients.

On average, higher grain yields were achieved at 101 kg N ha⁻¹ compared to 145 kg ha⁻¹ in 2013 while in 2014 higher yields were observed at the 145 kg ha⁻¹ rate. In 2013 the highest yield was obtained from plots which received 2 Mt ha⁻¹ CaSiO₃ slag and 101 kg N ha⁻¹. At the 145 kg ha⁻¹ N rate, grain yield was significantly raised from 1987 to 2123 kg ha⁻¹ with the addition of 4.5 Mt ha⁻¹ CaSiO₃ slag (P<0.05). Average leaf rust coverage for plots fertilized with 101 kg N ha⁻¹ was 8% lower than plots that received 145 kg N ha⁻¹. Within the 101 kg ha⁻¹ N rate, the weight of spikes was significantly higher for plots treated with 2 Mt ha⁻¹ CaSiO₃ slag than plots not treated with any CaSiO₃ slag (P=0.1). The initial results of our study show that wheat yield level was improved at certain combinations of N and Si rates. The outcomes of this research will help establish the links among Si fertilization rates, raised level of soil Si and plant-essential nutrients, increased resistance to lodging and diseases, and improved grain yield.

Theme 3 ORAL

75

Transcriptional regulation of Si transporter genes involved in uptake and distribution in rice

Yamaji N, Mitani-Ueno N, Ma J F

Institute of Plant Science and Resources, Okayama University, Kurashiki 710–0046, JAPAN (n-yamaji@rib.okayama-u.ac.jp)

Rice (*Oryza sativa*) is a typical Si-accumulating plant, which deposits up to 10% Si of dry weight in the shoots. We have identified two transporters (Lsi1 and Lsi2) responsible for Si uptake and a transporter (Lsi6) for Si distribution in the shoot of rice. However, the mechanism regulating these transporters is still poorly understood. In this study, we investigated the transcriptional regulation of these transporter genes in response to Si at the both vegetative and reproductive growth stage by using WT and *lsi6* mutant.

We first performed a root split experiment by dividing roots of a plant into two parts. When one root part was exposed to Si, but the other part was not, the expression of Lsi1 and Lsi2 was down-regulated in both root parts. This result suggests that the expression of Lsil and Lsi2 was regulated by some unknown long-distance signal from the shoots but not by root cells autonomously. Furthermore, at the vegetative stage, pre-treatment of 1 mM Si for 2 weeks decreased Si uptake by about half in WT. In contrast, this suppression was not observed in *lsi6* mutant with similar Si pretreatment. Pretreatment with Si also significantly decreased the expression of Lsil and Lsi2 in WT, but this down-regulation was hardly observed in lsi6 mutant. These results indicate that the expression of Lsi1 and Lsi2 is affected by the Si distribution pattern in the shoots because knockout of Lsi6 caused disturbed distribution of Si in the shoots. To confirm this hypothesis, we compared Si deposition pattern in WT and *lsi6* mutant. Lsi6 is expressed in the xylem parenchyma cells of both leaf sheath and leaf blade and polarly faced to the xylem vessels. This transporter is therefore required for Si distribution across the suberinized barrier at the mestome sheath of the leaf vascular bundles. Detailed analysis showed that in *lsi6* mutant, Si deposition was decreased in the leaf sheath, but increased in the leaf blade. This altered distribution of Si could be attributed to the lack of suberinized mestome sheath of small vascular bundles in the leaf blade, resulting in more Lsi6-independent deposition of Si in the leaf blade. Taken together, our results show that the expression level of *Lsi1* and *Lsi2* is regulated by lower Si level in the leaf sheath rather than higher Si level in the leaf blade.

At the heading stage, 13.5% of Si newly taken up by the root during two days was distributed to the panicle of WT, but only 1.4% was allocated to the panicles of the *lsi6* mutant, confirming that Lsi6 at the node play an important role in preferential distribution of Si to the panicles. However, the expression level of *Lsi1*, *Lsi2*, *Lsi6* were unaffected by Si in both WT and *lsi6* mutant at this stage and the uptake was similar between WT and *lsi6* mutant. These results suggest that the regulation of Si transporter genes is different between vegetative and reproductive growth stages.

Theme 5 POSTER

76

Silicon effect on growth dynamics, macro and microelements content in cadmium and arsenic treated poplar calli

Zelko I^{1,2}, Kollárová K², Vatehová Z^{2,3}, Lišková D²

¹Laboratory of Soil and Environment, Lorraine University, 2 av. de la Forêt de have, TSA 40602, Vandoeuvre-lès-Nancy, cedex F-54518, FRANCE (ivanzelko@yahoo.com)

² Department of Glycobiotechnology, Institute of Chemistry, Slovak Academy of Sciences, Dúbravská cesta 9, Bratislava 845 38, SLOVAKIA

³ Department of Plant Physiology, Faculty of Natural Sciences, Comenius University in Bratislava, Mlynská dolina B2, Bratislava 842 15, SLOVAKIA

Cadmium (Cd) and arsenic (As) are known as non-essential elements, toxic for plants, animals and humans. Contamination has been reported in many regions especially as a result of mining activities, and their accumulation in crops may pose a health risk to humans and animals. Mining heaps and waste dumps are often colonized by poplars - trees showing fast growth and high biomass production. Silicon (Si) is a beneficial element for plant growth, helping the plants to overcome toxic metal effects (Al, Mn, Zn or Cd). The aim of our work was to evaluate the Si effect on growth dynamics, macro and microelements content, Cd and As phytoextraction efficiency in poplar (Populus alba L., var. pyramidalis) calli grown in vitro.

Poplar callus was cultivated on medium [1] with addition of Cd(NO₃)₂.4H₂O (10 mM) or HAsNa₂O₄.7H₂O (1 mM), and/or silicon (5 mM Si as sodium silicate solution) with pH adjusted to 5.8 in controlled conditions. The duration of the experiment was 9 weeks (with 3 weeks subculture). Fresh and dry mass was taken every week and growth dynamics was determined [2]. The first and third subcultures were compared. The Cd, As and Si content as well as macro and microelements (Ca, Cu, Fe, K, Mg, Mn, Na, P, Si, Zn) content in calli were determined by Inductively coupled plasma mass spectroscopy (ICP-MS) analysis.

Cadmium and arsenic have increased callus fresh and dry mass. The differences in callus growth dynamics were dependent on culture duration, and on the toxic element used. Addition of silicon to media positively improved the fresh, dry mass, callus growth dynamics, and its effect was dependent on the toxic element used. The differences between macro and microelements content in calli under Cd, As and/or Si treatment may be linked with plant defence mechanisms against toxic elements.

Acknowledgements: This study was supported by the Slovak Research and Development Agency, contract No. APVV-0140-10 and by the Slovak Grant Agency for Science (VEGA No. 1/0817/12).

[1] Diaz-Colon et al., Physiol. Plant., 1972, 27, 60-64.

[2] Hlinková E. and Bobák M., J. App. Biomed., 2004, 2, 101-109.

Theme 3 ORAL

77

Identification of Putative Plant Silicon Binding Proteins

Zellner W L¹, Kim S¹, Krause C R²

¹ USDA-ARS Greenhouse Production Research Group, 2801 W. Bancroft St. M.S. 604, Toledo, OH 43606, USA (wendy.zellner@ars.usda.gov)

² USDA-ARS Application Technology Research Unit, 1680 Madison Ave., Wooster, OH 44691, USA

Silicon (Si) induced stress tolerance has been exemplified in a number of plants; however, the physiological pathways involved in the induced stress tolerance are not known. While Si is still identified as a beneficial element, the protection it provides both high and low accumulators suggests a fundamental role for the element within plants. While physiological changes in enzymatic activities and gene expression has been detected and widely reported in various plants treated with Si and an abiotic or biotic stress, the direct role of Si in plant defense(s) has yet to be determined. The aim of this study was to identify plant proteins that directly interact with Si to help identify pathways Si may be involved in.

In these studies, we used bioinformatics approaches to identify putative Si binding proteins from an identified domain in an *E. coli* ribosomal protein. BLAST search coupled with modelling and silicic acid docking predictions were performed using readily available programs, including Modeller, Patch Doc and PEARLs. The transcripts of the identified proteins were cloned into a Gateway cloning system and are being used to confirm and identify effects of direct Si binding on protein function. A pseudo-Si pull down was also performed with a crude plant extract and ran on an SDS-PAGE gel where four distinct bands were cut and sent for analysis to the University of Michigan Department of Pathology Proteomics Resource Facility.

Our bioinformatics results identified a ribosomal cytoplasmic protein having a high percent identity to the *E. coli* Si binding domains in tobacco, tomato and *A. thaliana*. In addition, predicted silicic acid interaction energies for this protein in tobacco were much higher than that of the rice Si transporter at $-13.1 \text{ kcal} \cdot \text{mol}^{-1}$ versus $-9.42 \text{ kcal} \cdot \text{mol}^{-1}$, respectively. We are currently purifying the cytoplasmic, chloroplast and mitochondrial homologs of the proteins to confirm direct binding to silicic acid. A pseudo Si pull-down assay was also performed with a tobacco root extract and the proteins from the silicic acid matrix were ran on an SDS-PAGE gel and stained with Commassie Blue. Four distinct bands were identified near 30; 20; 17 and 15 kDa. The bands were cut and sent for analysis, resulting in the presence of the predicted ribosomal protein in addition to other defence-related proteins containing Si binding motifs similar to the E. *coli* and plant ribosomal proteins. We are working on confirming the direct binding of these proteins in hopes to identify the fundamental role(s) of Si within plants, leading to the identification of defence pathways altered with Si supplementation.

Theme 2 ORAL

78

The importance of biogenic pools to Si cycling

<u>Höhn A</u>¹, Jochheim H¹, Breuer J², Zagorski Z³, Busse J¹, Kaczorek D^{1,3}, Sommer M^{1,4}

¹Leibniz-Centre for Agricultural Landscape Research, (ZALF), Müncheberg, GERMANY (hoehn@zalf.de)

² LTZ Augustenberg, Karlsruhe, GERMANY

³ Department of Soil Environment Sciences, Warsaw University, Warsaw, POLAND

⁴ University of Potsdam, Inst. of Earth and Environmental Sciences, Potsdam, GERMANY

The relevance of biological Si cycling for dissolved silica (DSi) export from terrestrial biogeosystems is still in debate. Even in systems showing a high content of weatherable minerals biogenic Si (BSi) might contribute >50% to DSi. Concerning the origin of DSi the major research questions are the following: 1. how large is the contribution of the BSi pool to DSi compared to litho-/pedogenic sources? 2. What are the main drivers of the relative importance of biogenic and mineral sources – climate, lithology, state of soil development, soil pattern, land use?

To cover one of the controlling factors on DSi (weatherable minerals content), we studied a forested site with absolute quartz dominance. Here we hypothesise minimal effects of chemical weathering of silicates on DSi. During a four year observation period, we quantified (i) internal and external Si fluxes of a temperate-humid biogeosystem (beech, 120 yr) by BIOME-BGC, (ii) related Si budgets, and (iii) Si pools in soil and beech, chemically as well as by SEM-EDX. For the first time two compartments of biogenic Si in soils were analysed: the phytogenic and the zoogenic Si pool (testate amoebae).

An average Si plant uptake of 35 kg Si ha $^{-1}$ yr⁻¹ was quantified – most of which is recycled to the soil by litterfall – and an annual biosilicification from idiosomic testate amoebae of 17 kg Si ha⁻¹ yr⁻¹ was calculated. The comparatively high DSi concentrations (6 mg L⁻¹) and DSi exports (12 kg Si ha⁻¹ yr⁻¹) could not be explained by chemical weathering of feldspars or quartz dissolution. Instead, dissolution of a relictic, phytogenic Si pool seems to be the main process for the DSi observed. We identified canopy closure accompanied by a disappearance of grasses as well as the selective extraction of pine trees 30 years ago as the most probable control for the phenomena observed. From the results it can be concluded, that the biogeosystem is in transient state in terms of Si cycling.

Lists

List of Authors

Author	Pape	r no
Adjolohoun S	37	
Akram M S	31	
An M	61	
Anitha M S	51	
Asano T	72	
Asghar A	30	
Ashraf M A	30	
Azeem M	31	
Aziz T	36	
Babu T	3	69
Balasubramaniam P	44	
Baldwin L	24	
Bar K P	34	
Barão A L	4	73
Barboni D	50	
Bartoszewski G	29	
Batista F V T	5	
Bélanger R R	6	
Belmonte D U	67	
Belzile F	6	
Bocharnikova E	47	
Bokor B	7	
Bokorová S	7	
Botta A	8	
Breuer J	78	
Brossa R	8	
Buck G	59	
Busse J	78	
Buso D C	13	
Büll T L	9	
Büttner C	29	
Camargo M S	10	
Cartes P	11	56
Catojo P R	9	
Cerdà J M	8	
Chidrawar S	32	
Ciecierski W	12	
Clymans W	13	
Cohen D	45	
Conley D J	13	14
Connick V J	61	
Cristancho R J A	15	

Cruz M F A	63			
da Silva W L	63			
Dastan S	23			
Datnoff L	3 58	59	69	74
De Vleesschauwer D	16 72			
	-			
Deokate P P	54			
Deus C F A	9			
Dietz K-J	18			
Dupree P	74			
Dåstøl M	47			
Elbaum R	17 39	45		
Farooq M A	18	-		
•	-			
Fernandes D M	19 22			
Fialová I	20 66	71		
Filippi M C	5 21	65		
••		00		
Fitt B D L	33			
Fleck A T	28			
Fortunato A A	63			
Frantz J	42			
Freitas L B	19 22			
Fridman E	45			
Frossard E	70			
Gerard G	4			
Ghanbari-Malidarreh A	23			
Głazowska S E	24			
Goddéris Y	67			
Gonzalo M J	27			
Greger M	25 41			
Guisard Y	61			
Gurr G M	61			
Gutiérrez-Moraga A	56			
-				
Hall A M	33			
Hartley S E	48			
Hartmann J	67			
Haynes R J	26			
-				
Hernández-Apaolaza L	27 62			
Hinrichs M	28			
Holou R	37			
Holz S	29			
Houinato M	37			
Höfte M	16 72			
Höhn A	78			
Hussain I	30			
lqbal N	30 31			
Jahn R	38 46			
Jain N	32 54			
Javed M T	30 31			

Jiménez H	11		
Jin X	33		
Jochheim H	78		
Johnson C E	13		
Kaczorek D	78		
Kardasz H	12		
Katz O	34		
Kausar S	31		
Keeping M G	35		
Keller C	1	50	
Khalid M	36	00	
Khan W	36		
Khandekar S	42		
Kikuchi S	16	72	
Kim S	77	12	
Kindomihou V	37		
Kinsey C	59		
Klotzbücher T	38	46	
	38 29	40	
Kneipp J	29 76		
Kollárová	-	50	
Korndörfer G H	10 77	59	
Krause C R	77		
Kube M	29		
Kumar S	39		
Laane H-M	40		
Landberg T	25	41	
Leisner S	42		
	48		
Leuther F	38		
Lev-Yadun S	34		
Likens G E	13		
Lindberg S	41		
Lišková D	76		
Lucena J J	27		
Lukačová Z	7		
Lux A	7	70	
Luxová M	20	66	71
Ma J F	64	75	43
Mahendran P P	44		
Maia S C M	19	22	
Maity P J	41		
Maqsood M A	36		
Marín C	8		
Markovich O	45		
Marxen A	38	46	
Mascagni H Jr	74		
Matichenkov V	47		

McLarnon E	48		
McQueen-Mason S	48		
Meerts P	37		
Meire P	4	67	73
Menachem M	17		
Menzies J G	6		
Meszka B	49		
Meunier J-D	1	50	
Minhas P S	54	50	
	-		
Miranda S	59		
Mitani N	64		
Mitani-Ueno N	75	-	
Moniz A	19	22	
Murozuka E	24		
Nagabovanalli B P	51		
Narayanaswamy C	69		
Nicol H I	61		
Nikolic M	52	53	
Nikolić S D	53		
Nissan H	17		
Ondoš S	7		
Pascual-de Vega B	27		
Passala R	54		
Pereira H	59		
Pereira S T	21		
Perry C C	55		
Pontigo S	11	56	
Powell J J	2		
Prabhu S A	21		
Prentice P	57		
Provance-Bowley M ¹	58	59	
Rao V	32	54	
Rasheed R	30	04	
Rêgo C F M	5		
•			
Resende R S	60		
Restrepo F	15		
Reynolds O L	61		
Ribera A	11	56	
Riswan M	50		
Rivera V	62		
Rocha G	10		
Rodrigues F Á	8	60	63
Rodrigues H	59		
Ronchi B	4		
Rutherford R S	35		
Ryken N	73		
-			
Sakurai G	64		

Samardžić J	53			
Sandhya K	51			
Satake A	64			
Schenk M K	28			
	20 24			
Schjoerring J K		70		
Schoelynck J	67 70	73		
Schulin R	70			
Sebastian D	59			
Sedláková B	20			
Sewpersad C	35			
Shah P	32			
Sierras N	8	.	~-	
Silva B G	5	21	65	
Šimková L	20	66	71	
Simmons A T	61			
Sinsin B	37			
Smis A	73			
Sommer M	78			
Sousa P T	65			
Souza A C A	21	65		
Sreya U P	44			
Struyf E	4	14	67	73
Švubová R	7			
Szemes T	7			
Tallberg P	68			
Tandy S	71			
Thorat V	32			
Tubana B	3	69	74	
Vaculík M	70	71		
Vaculíková M	20	66	70	71
Van Bockhaven J	16	72		
Van Damme S	73			
Vandevenne F	4	73		
Vatehová Z	76			
Vetterlein D	38	46		
Vivancos J	6			
Wang J	3			
Wei X	47			
Weiss A	67			
White E B	69	74		
Wilk R	49			
Wilkinson J-A	6			
Willats W G T	24			
Yamaji N	64	75	43	
Yokozawa M	64			
Yzenas J	3	69	74	
Zagorski Z	78			

Zeise I	29	
Zelko I	76	
Zellner W L	42	77
Zhan Q	47	
Zhou Y-F	26	

List of Participants

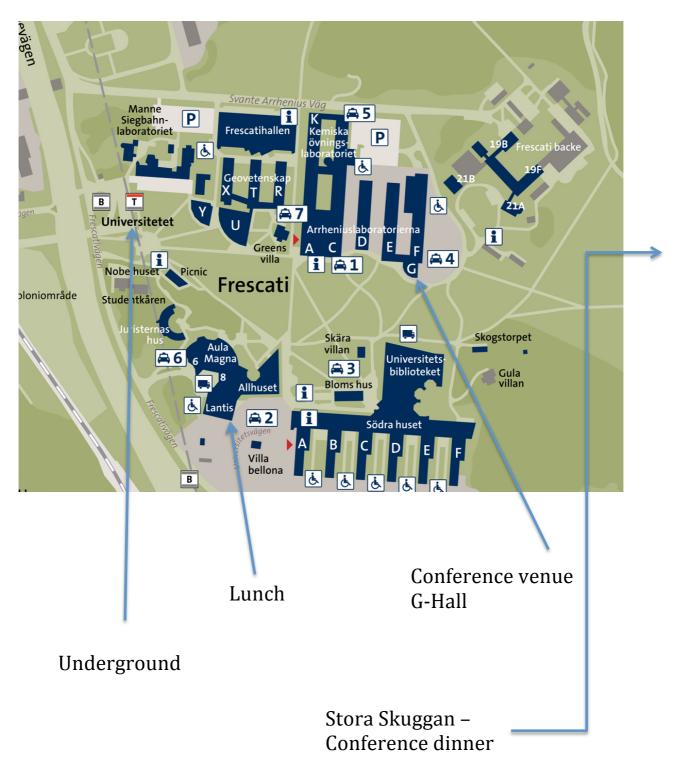
Name	Company	Country	E-mail
Aamodt, Arianeh	Elkem silicon materials	Norway	arianeh.aamodt@elkem.no
Aarup, Steen	Yara danmark gødning a/s	Denmark	steen.aarup@yara.com
Babu, Tapasya	Louisiana state university	USA	tbabu1@tigers.lsu.edu
Bahiri, Gidon	Private	UK	gidon@orionft.com
Barao, Ana Lucia	University of Antwerp	Belgium	luciabarao@gmail.com
Barbosa de Freitas, Lucas	Faculdade de Ciências Agronômicas - Unesp	Brazil	lucasbarbosaf@yahoo.com.b
Batista, Telma	Universidade Federal Rural da Amazônia	Brazil	telmabatistacoelho@yahoo.c om.br
Belanger, Richard	Universite laval	Canada	richard.belanger@fsaa.ulaval .ca
Bent, Edward	Hortcom	Italy	edbent@tin.it
Bernal Montolio, Meritxell	COMPO expert	Spain	meritxell.bernal@compo.co m
Bokor, Boris	Comenius University in Bratislava	Slovakia	boris.bokor@gmail.com
Botta Català, Anna M ^a	Bioiberica SA	Spain	abotta@bioiberica.com
Büll, Leonardo	FCA/UNESP	Brazil	bull@fca.unesp.br
Büttner, Carmen	Humboldt-Universität zu Berlin	Germany	carmen.buettner@agrar.hu- berlin.de
Camargo, Mônica Sartori de	São Paulo Agency for Agribusiness Technology (APTA)	Brazil	mscamarg@yahoo.com.br
Cartes, Paula	Universidad de La Frontera	Chile	paula.cartes@ufrontera.cl
Ciecierski, Wiesław	INTERMAG SP. ZOO	Poland	wieslaw.ciecierski@intermag .pl
Clymans, Wim	Lund university	Sweden	wim.clymans@geol.lu.se
Cocks, Simone	Agripower australia ltd	Australia	michela@agripower.com.au
Conley, Daniel J.	Lund university	Sweden	daniel.conley@geol.lu.se
Contartese, Valeria	Green Has Italia S.p.A.	Italy	v.contartese@greenhasitalia. com
Cristancho, Jose	Mejisulfatos	Colombia	jose.cristancho@mejisulfatos .com
Daoud, Abdel- monem	Soil,Water & Environment Research Institute -Agriculture Research Center of Egypt	Egypt	mobarak_salinlab47@yahoo. com
Datnoff, Lawrence	Lsu	USA	ldatnoff@agcenter.lsu.edu
De Avila Rodrigues, Fabricio	Viçosa federal university	Brazil	fabricio@ufv.br

De Vleesschauwer, David	Ghent university	Belgium	david.devleesschauwer@uge nt.be
Dåstøl, Magne	Elkem	Norway	magne.dastol@elkem.no
Elbaum, Rivka	Institute of Plant Sciences	Israel	rivka.elbaum@mail.huji.ac.il
Farooq, Muhammad Ansar	Department of Biochemistry and Physiology of Plants, Universität Bielefeld	Germany	ansar_1264@yahoo.com
Ferhout, Hicham	Agronutrition	France	h.ferhout@agro-nutrition.fr
Fialova, Ivana	Institute of Botany of SAS	Slovakia	ivfiala@gmail.com
Filippi, Marta Cristina	Embrapa	Brazil	cristina.filippi@embrapa.br
Frostgård, Gunilla	Yara AB	Sweden	gunilla.frostgard@yara.com
Galan Asensi, Luis Manuel	Agritecno fertilizantes	Spain	administracion2@agritecnofe rtilizantes.com
Ghanbari- malidarreh, Abbas	Islamic azad university	Iran	aghanbari@jouybariau.ac.ir
Goh, Geoffrey	Wastech multigreen S/B	Malaysia	ggohwtech@gmail.com
Greger, Maria	Stockholm university	Sweden	maria.greger@su.se
Głazowska, Sylwia	University of Copenhagen	Denmark	glaz@plen.ku.dk
Hamada, Afaf	Department of Ecology, Environment and Plant Sciences	Sweden	afaf.hamada@su.se
Haynes, Richard	The University of Queensland	Australia	r.haynes1@uq.edu.au
Hernandez- Apaolaza, Lourdes	University Autonomous of Madrid	Spain	lourdes.hernandez@uam.es
Hinrichs, Martin	Institute of Plant Nutrition	Germany	hinrichs@pflern.uni- hannover.de
Höhn, Alex	ZALF	Germany	hoehn@zalf.de
Holz, Sabine	Humboldt-Universitaet zu Berlin	Germany	sabine.holz@agrar.hu- berlin.de
Houben, Frank	Agro-solutions bv	Netherlands	info@agro-solutions.nl
Hussain, Iqbal	Govt. College university	Pakistan	iqbalbotanist1@yahoo.com
Iqbal, Naeem	Gc university faisalabad	Pakistan	drnaeem@gcuf.edu.pk
Jackson, Brian	Ignimbrite, inc.	USA	brian@montanagrow.com
Jacques, Denis	Meac	France	denis.jacques@meac.fr
Jain, Neeru	Privi life sciences pvt ltd	India	neeru.jain@privi.co.in
Janeczko, Robert	Vitrosilicon SA	Poland	r.j@vitrosilicon.com.pl
Jin, Xiaolei	University of hertfordshire	UK	x.jin3@herts.ac.uk
Kalimuthu, Rajendran	Viggi agro products	India	info@viggiagro.com

Kardasz, Hubert	INTERMAG SP. Z OO	Poland	hubert.kardasz@intermag.pl
Katz, Ofir	Ben-Gurion University	Israel	katz.phyt@gmail.com
	of the Negev	151401	
Keeping, Malcolm	South african sugarcane	South	malcolm.keeping@sugar.org.
1100p.118, 111000111	research institute	Africa	za
Keller, Catherine	Cerege - aix-marseille	France	keller@cerege.fr
	university		
Kindomihou,	University of Abomey	Benin	vkindomihou@yahoo.fr
Valentin	Calavi		· # •
Khan, ud Din Waqas	University of	Pakistan	waqas1919@gmail.com
,	Agriculture,		
	Faisalabad		
Klotzbücher, Thimo	University of Halle-	Germany	thimo.klotzbuecher@google
,	Wittenberg	5	mail.com
Ko, Gil Hong	Agchem Solution Co.	Korea,	agchem@unitel.co.kr
		South	
Konte, Makan	Mc agriprod	Mali	mkonte2@hotmail.fr
Kumar, Santosh	The Hebrew University	Israel	skg.research@gmail.com
,	of Jerusalem		
Laane, Henk-	ReXil Agro BV	Netherlands	hm.laane@rexil-agro.com
Maarten			
Landberg, Tommy	Stockholm university	Sweden	tommy.landberg@su.se
Leisner, Scott	University of Toledo	USA	sleisne@utnet.utoledo.edu
Lin, Zhen	Elkem silcon materials	China	lin.zhen@elkem.no
Lindberg, Sylvia	Stockholm university	Sweden	sylvia.lindberg@su.se
Lucena Marotta,	University Autonomous	Spain	juanjose.lucena@uam.es
Juan José	of Madrid	1	
Lux, Alexander	Dept.Plant Physiology,	Slovakia	lux@fns.uniba.sk;
	Faculty of Natural		luxalexander@hotmail.com
	Sciences, Comenius		<u> </u>
	University in Bratislava		
Ma, Jian Feng	Okayama university	Japan	maj@rib.okayama-u.ac.jp
Markovich, Oshry	Robert H. Smith	Israel	oshrym@gmail.com
	Faculty of Agriculture,		
	Food and Environment		
Martinsson, Mats	Yara AB	Sweden	mats.martinsson@yara.com
Marxen, Anika	Helmholtz Centre for	Germany	anika.marxen@ufz.de
	Environmental		
	Research - UFZ		
Matichenkov,	Institute basic	Russia	vvmatichenkov@rambler.ru
Vladimir	biological problems ras		
Maximino	FCA - UNESP	Brazil	dmfernandes@fca.unesp.br
Fernandes, Dirceu			
	University of York	UK	em965@york.ac.uk
McLarnon, Emma			
Meunier, Jean-	Cerege, CNRS, AMU	France	meunier@cerege.fr
Dominique			
Nagabovanalli,	University of	India	nagabovanalliprakash@rediff
Prakash	Agricultural Sciences,		mail.com
		1	

Nikolic, Miroslav	University of Belgrade	Serbia	mnikolic@imsi.bg.ac.rs
Nikolic, Dragana	Institute of molecular genetics and genetic engineering	Serbia	dsnikolic@imgge.bg.ac.rs
Pasala, Ratna Kumar	National institute for abiotic stress management	India	ratnakumar@niam.res.in
Perry, Carole	Interdisciplinary biomedical research centre	UK	carole.perry@ntu.ac.uk
Peters, Hans-Jacob	Fylkesmannen i Buskerud	Norway	hajape@gmail.com
Peyandi Paraman, Mahendran	Tamil nadu agricultural university	India	ppmahendran2002@yahoo.c
Pinto, João	ADP-fertilizantes SA	Portugal	castropinto@adp- fertilizantes.pt
Pontigo, Sofia	Universidad de La Frontera	Chile	s.pontigo01@ufromail.cl
Porterfield, John	Ignimbrite, inc.	USA	johnp@montanagrow.com
Pouw, William	Agripower australia ltd	Australia	michela@agripower.com.au
Powell, Jonathan	MRC HNR	UK	jonathan.powell@mrc- hnr.cam.ac.uk
Prekumar, Albert	Department of Ecology, Environment and Plant Sciences	Sweden	albertprem@gmail.com
Prentice, Peter	Agripower Australia ltd	Australia	michela@agripower.com.au
Preti, Michel	Ordinary	Belgium	michel.preti@gmail.com
Provance-Bowley, Mary	Harsco Metals and Minerals	USA	mpbowley@harsco.com
Rémus Borel, Wilfrid	Biotechnology consultant	France	wilfried.remusborel@gmail.c om
Reynolds, Olivia	Graham centre (NSW DPI & CSU)	Australia	olivia.reynolds@dpi.nsw.gov .au
Rivera Varela, Vagnum Flaumbert	Autonomous University of Madrid	Spain	vrivera07@hotmail.com
Rodriguez Ortega, Wilbert Michael	Atlantica agricola	Spain	wrodriguez@atlanticaagricol a.com
Sakurai, Gen	National Institute for Agro-Environmental Sciences	Japan	sakuraigen@affrc.go.jp
Sandhi, Arifin	KTH-Royal Institute of Technology	Sweden	asandhi@kth.se
Schenk, Manfred	University of Hannover	Germany	schenk@pflern.uni- hannover.de
Secci, Michela	Agripower Australia ltd	Australia	michela@agripower.com.au
Silva, Gisele	UFRA	Brazil	gibarata@bol.com.br
Simkova, Lenka	Institute of Botany of SAS	Slovakia	sprekelia@gmail.com

Sousa Resende,	Viçosa federal	Brazil	resenders@yahoo.com.br
Renata	university	Diuzii	resenders@yunoo.com.or
Struyf, Eric	University of Antwerp	Belgium	eric.struyf@uantwerp.be
Suthakar, Vikram	Marshall marine	India	vnayyadurai@gmail.com
	products		
Szeszo, Dobieslaw	Vitrosilicon SA	Poland	dobieslaw.szeszo@vitrosilico
			n.com.pl
Tallberg, Petra	University of Helsinki	Finland	petra.tallberg@helsinki.fi
Tripodi, Sergio	Green Has Italia S.p.A.	Italy	sving@greenhasitalia.com
Tubana, Brenda	LSU AgCenter	USA	btubana@agcenter.lsu.edu
Vaculik, Marek	Comenius University in	Slovakia	vaculik.marek@gmail.com
	Bratislava, Faculty of		
	Natural Sciences		
Vaculikova,	ETH Zurich	Switzerland	vaculikova.miroslava@gmail
Miroslava			.com
Van Bockhaven,	Ghent university	Belgium	jonas.vanbockhaven@ugent.
Jonas			be
Vandevenne, Floor	University of Antwerp	Belgium	floor.vandevenne@uantwerp en.be
Vetterlein, Doris	UFZ	Germany	doris.vetterlein@ufz.de
White, Brandon	LSU Ag Center	USA	b17white@hotmail.com
Wilk, Radosław	Wroclaw University of	Poland	radoslaw.wilk@pwr.edu.pl
	Technology		
Yamaji, Naoki	IPSR, Okayama	Japan	n-yamaji@rib.okayama-
	university		u.ac.jp
Yzenas, John	Edw C Levy Co	USA	jyzenas@levyco.net
Zelko, Ivan	Université de Lorraine	France	ivanzelko@yahoo.com
Zellner, Wendy	USDA-ASR	USA	wendy.zellner@ars.usda.gov
	Greenhouse Production		
	Research Group		



ISBN 978-91-637-6572-8