Cell. Mol. Life Sci. 65 (2008) 3049 – 3057
1420-682X/08/193049-9
DOI 10.1007/s00018-008-7580-x

Cellular and Molecular Life Sciences

Review

Functions and transport of silicon in plants

J. F. Ma* **and N. Yamaji**

Research Institute for Bioresources, Okayama University, Chuo 2-20-1, Kurashiki (Japan),
Fax: +81-86-434-1209, e-mail: maj@rib.okayama-u.ac.jp

Received 21 December 2007; received after revision 29 April 2008; accepted 15 May 2008
Online First 14 June 2008

Abstract. Silicon exerts beneficial effects on plant growth and production by alleviating both biotic and abiotic stresses including diseases, pests, lodging, drought, and nutrient imbalance. Recently, two genes (*Lsi1* and *Lsi2*) encoding Si transporters have been identified from rice. Lsi1 (low silicon 1) belongs to a Nod26-like major intrinsic protein subfamily in aquaporin, while *Lsi2* encodes a putative anion transporter. Lsi1 is localized on the distal side of both exodermis and endodermis in rice roots, while Lsi2 is localized on the proximal side of the same cells. Lsi1 shows influx transport activity for Si, while Lsi2 shows efflux transport activity. Therefore, Lsi1 is responsible for transport of Si from the external solution to the root cells, whereas Lsi2 is an efflux transporter responsible for the transport of Si from the root cells to the apoplast. Coupling of Lsi1 with Lsi2 is required for efficient uptake of Si in rice.

Keywords. Silicon, stress, beneficial effect, transporter, accumulation.

Introduction

Silicon (Si) is the second most abundant element after oxygen in the earth's crust. It is an essential element for animals, and has been implicated in optimal bone and connective tissue development in the human body [1]. It is also essential for diatoms and plants in Equisetales. Diatom cell walls are made of amorphous silica that exhibits species-specific, mostly porous, patterns in the nanometer to micrometer range [2]. Since silicon dioxide comprises 50–70% of the soil mass, all plants grown in soil will contain some Si in their tissues. However, Si is not considered as an essential element for higher plants if the criteria required for essentiality established by Arnon and Stout [3] are adopted, because there is no evidence that Si is involved in the metabolism of plants. Recently, a new definition of essentiality was proposed by Epstein and Bloom [4]. According to this definition, an element is essential if it fulfills either one or both of two criteria: (1) the element is part of a molecule that is an intrinsic component of the structure or metabolism of the plant, and (2) the plant can be so severely deficient in the element that it exhibits abnormalities in growth, development, or reproduction, i.e., "performance", compared to plants with a milder deficiency. Since Si deficiency causes various abnormalities in a wide variety of plant species [5, 6], Si is "quasi-essential" for the growth of higher plants. In this review, we focus on the functions of Si in plants and the recent progress in Si transporters.

Functions of Si in plants

The function of Si is to protect the plant from various biotic and abiotic stresses [5, 6]. Therefore, the effect of Si on plant growth becomes obvious under stress conditions, but usually not under non-stressed con-

* Corresponding author.

ditions. This was well demonstrated by a recent study with an Arabidopsis-powdery mildew system using microarray analysis. In non-inoculated plants, the application of Si only altered the expression level of two genes of the nearly 40 000 transcripts [7]. However, inoculation with powdery mildew altered the expression of a set of nearly 4000 genes in plants treated with Si or not and application of Si resulted in >25 % attenuation of the magnitude of down-regulated genes, suggesting that Si alleviates the stress [7]. Si enhances resistance of plants to diseases caused by both fungi and bacteria in different plant species such as rice blast (Fig. 1A), powdery mildew, sheath blight, ring spot, rust, leaf spot, and gray leaf spot [8, 9]. Si also suppresses insect pests such as stem borer, brown planthopper, rice green leafhopper, whitebacked planthopper, and non-insect pests such as leaf spider and mites (Fig. 1B) [9]. Two mechanisms for Si-enhanced resistance to diseases have been proposed. One is that Si acts as a physical barrier. Si is deposited beneath the cuticle to form a cuticle-Si double layer (Fig. 2) [6]. This layer can mechanically impede penetration by fungi, thereby avoiding the infection process. The protective effects of Si in pest damage could also be related to the mechanical barrier provided by silica deposition in the cell wall (Fig. 2), which would make it difficult for the stylet to penetrate the plant tissues and for tissues to be chewed by insects [10].

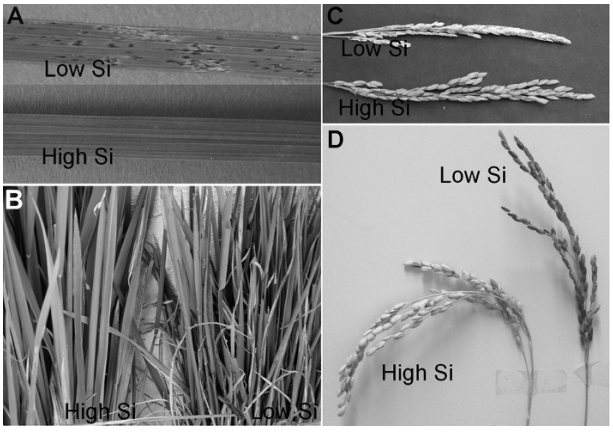

Figure 1. Some examples showing beneficial effects of Si in rice grown in the field. (*A*) Effect of Si on suppression of pathogen infection. (*B*) Effect of Si on suppression of pest damage. (*C*) Effect of Si on excessive transpiration from the panicles. The picture was taken after a typhoon. (*D*) Effect of Si on grain fertility of rice. Pictures are adopted from [49].

Another mechanism proposed recently is that soluble Si acts as a modulator of host resistance to pathogens [8]. Studies in monocotyledons (rice and wheat) and dicotyledons (cucumber) have shown that plants

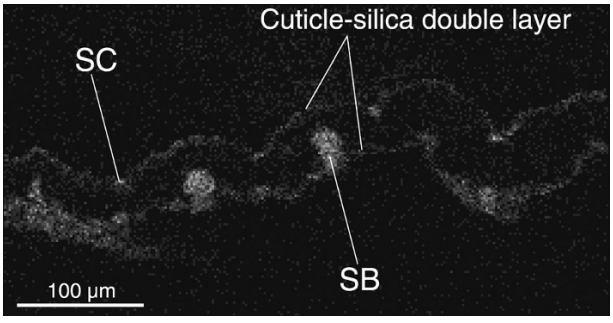

Figure 2. Deposition of Si in rice leaf blade. Silica is deposited beneath the cuticle, forming cuticle-silica double layers, and also deposited on specific cells as silica bodies (SB) and silica cells (SC). Silica is detected by scanning electron microscopy–energy dispersive X-ray spectroscopy.

supplied with Si produce phenolics and phytoalexins in response to fungal infection such as those causing rice blast and powdery mildew [11–14]. Si also can activate some defense mechanisms. For example, in roots of cucumber plants infected and colonized by *Pythium*, Si enhanced the activity of chitinases, peroxidases and polyphenoloxidases [15]. In rice, differential accumulation of glucanase, peroxidase, and PR-1 transcripts were associated with limited colonization by the fungus *Magnaporthe grisea* in epidermal cells of a susceptible rice cultivar supplied with Si [16]. These biochemical responses are only induced by soluble Si, suggesting that soluble Si may play an active role in enhancing host resistance to diseases by interacting with several key compounds of plant stress signaling systems [8], in addition to the increase in synthesis of plant defense compounds.

Si has beneficial effects under abiotic stresses including chemical stress (salt, metal toxicity, nutrient imbalance) and physical stress (lodging, drought, radiation, high temperature, freezing, UV) (Fig. 1C, D) [6, 17]. These effects have been attributed to: (1) Si-reduced cuticular transpirational water loss by deposition of Si beneath the cuticle; (2) Si-decreased apoplastic flow and uptake of toxic minerals due to deposition of Si in the root; (3) chelation with toxic metals; and (4) Si-enhanced strength of the stem (for a review, see [5]).

Si accumulation in different plant species

Although all plants contain Si in their bodies, there are wide variations in the Si accumulation in the shoot among species. The Si concentration in the shoot ranges from 0.1 % to 10 % Si in dry weight, depending on the plant species [5, 18, 19]. In the plant kingdom, Si is highly accumulated in Bryophyta, as well as Lycopsida and Equisetopsida of Pteridophyta, but

hardly accumulated in Filicopsida in Pteridophyta and Gymnospermae [5]. In Angiospermae, only Cyperaceae and Gramineae show high Si accumulation [5, 19].

There is also a genotypic variation in the Si concentration in the shoot within a species, although the variation is usually not as large as the one among species. For example, in a survey of about 400 cultivars of barley, the Si concentration in barley grain showed a large variation, ranging from 1.24 to 3.80 mg/g in hulled barley cultivars [20]. In sugarcane grown in the field, the Si concentration in the shoot varied with the variety, ranging from 6.4 to 10.2 mg/g [21]. In rice, *japonica* rice cultivars usually have a higher Si concentration than *indica* rice cultivars [22–24].

Uptake modes of Si in different plant species

The difference in Si accumulation of different plant species as described above has been ascribed to the ability of the roots to take up Si [25]. Plant roots take up Si in the form of silicic acid [$Si(OH)_4$], an uncharged monomeric molecule, when the solution pH is below 9 [26]. Recently, the uptake system of Si was investigated in terms of radial transport from external solution to root cortical cells and release of Si from cortical cells to xylem in rice, cucumber and tomato, which differ greatly in the shoot Si concentration [27]. The concentrations of Si in the root-cell symplast in all species were higher than that in the external solution, although the concentration in rice was three- and fivefold that in cucumber and tomato, respectively. A kinetic study showed that the radial transport of Si was mediated by a transporter with a K_m value of 0.15 mM in all species, but with different V_{max} values in the order of rice > cucumber > tomato. In the presence of the metabolic inhibitor 2,4-dinitrophenol and at low temperature, the Si concentration in the root-cell symplast decreased to the level in the apoplastic solution. These results suggest that both transporter-mediated transport and passive diffusion of Si are involved in the radial transport of Si and that the transporter-mediated transport is an energy-dependent process.

The subsequent process, i.e., the release of Si from the cortical cells to the xylem (xylem loading) was also compared among rice, cucumber, and tomato. The Si concentration of xylem sap in rice was 20- and 100-fold higher than that in cucumber and tomato, respectively. In contrast to rice, the Si concentration in the xylem sap was lower than that in the external solution in cucumber and tomato. A kinetic study showed that xylem loading of Si was also mediated by a kind of transporter in rice, but by passive diffusion in cucumber and tomato [27]. These results indicate that xylem loading is the most important determinant for a high level of Si accumulation in the shoots of rice. The much lower accumulation of Si in cucumber and tomato might be explained by a lower density of the transporter from the external solution to the cortical cells, and a defective or an absence of the transporter from cortical cells to the xylem.

Si transporters in rice

Influx transport of Si

Cloning of Si influx transporter gene. The first gene encoding a Si transporter was identified from rice, which requires high Si for healthy growth and high production [28]. The gene was cloned using a rice mutant (*lsi1*, *low silicon 1*) defective in Si uptake [29], which was isolated by screening mutagenized seeds in a solution containing germanium (Ge). Si and Ge are chemically similar and plant roots cannot discriminate Ge and Si in terms of uptake. However, in contrast to Si, Ge taken up is toxic to plant, which is characterized by brown spots on the leaves. The gene *Lsi1* isolated by map-based cloning is localized on chromosome 2 and consists of five exons and four introns [28, 30]. The cDNA of this gene is 1409 bp long and the deduced protein consists of 298 amino acids. Blast search and ClustalW analysis revealed that Lsi1 belongs to a Nod26-like major intrinsic protein (NIP) subfamily of aquaporin-like proteins. The predicted amino acid sequence has six transmembrane domains and two Asn-Pro-Ala (NPA) motifs, which are well conserved in typical aquaporins. There are two close homologs in maize (ZmNIP2–1 and 2–2) with identity of 77–83 % and one homolog in rice (Os06 g0228200, named Lsi6) with identity of 77 %.

A single nucleotide substitution occurred from G in the wild type to A in the *lsi1* mutant, resulting in an amino acid change from alanine in the wild type to threonine in the mutant at position 132 [28]. Alanine at 132 seems to be a critical residue, because substitution of this amino acid in the mutant significantly alters the conformation according to the modeling of the native and mutant proteins [28]. Thus, the substitution of Thr for Ala132 provoked severe steric interactions with Val55 and Val59 in helix 1 (H1), facilitating a movement of H1. This unfavorable interaction would affect the conformation of Asn108, the pore-forming residue in the P-loop.

Expression pattern of Si transporter gene *Lsi1*. *Lsi1* is constitutively expressed in the roots (Fig. 3A), but its expression is decreased to one fourth by Si supply

(Fig. 3B) [28]. Within a root, the expression of *Lsi1* was much lower in the root tip region between 0 and 10 mm than that in the basal regions (>10 mm) [31]. Si uptake in the root tip region (0–10 mm) comprising both the apical meristem and the elongation zone was also much lower than that in the basal regions (>10 mm from the root tips). These observations indicate that Lsi1 plays an important role in Si uptake and that the site of Si uptake is located in the mature regions of the roots rather than the root tips. The expression of *Lsi1* displayed a distinct diurnal pattern [31], which is characterized by the high expression from 12:00 to 24:00 (light from 5:00), but low from 4:00 to 8:00. However, the fluctuation of *Lsi1* expression during the day was not as large as that reported for other aquaporin genes.

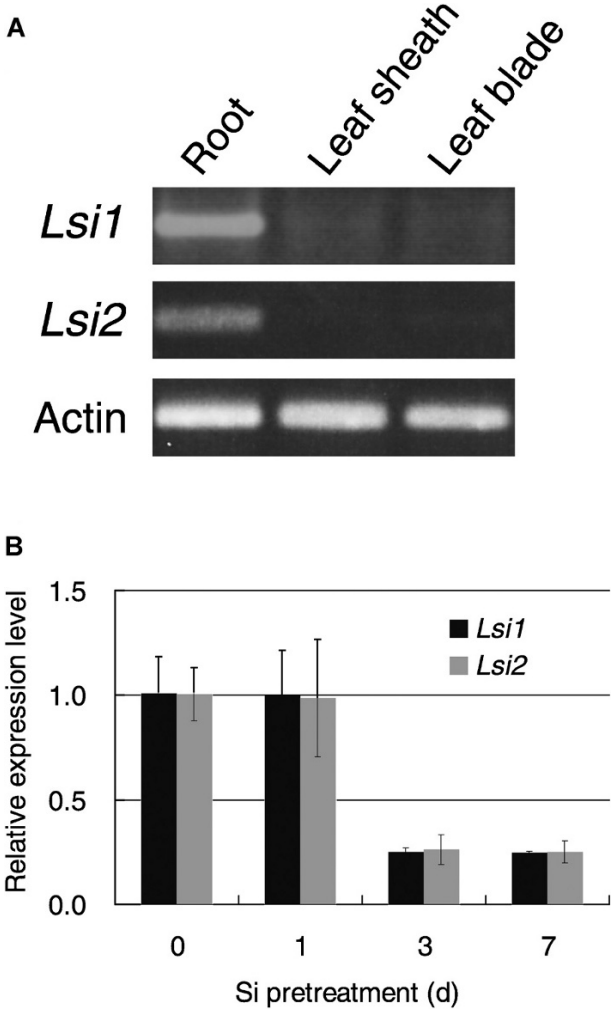

Figure 3. Expression of two Si transporter genes in rice. (*A*) Expression of *Lsi1* and *Lsi2* in different plant tissues. Both *Lsi1* and *Lsi2* are expressed in the roots. (*B*) effect of Si supply on the expression of *Lsi1* and *Lsi2*. The expression of *Lsi1* and *Lsi2* is decreased by Si supply.

Investigation of Lsi1 expression at different growth stages showed that the expression was transiently enhanced around the heading stage. A previous study showed that 67% of total Si was taken up during the reproductive stage from panicle initiation to heading in rice [32]. Deficiency of Si during this stage resulted in a significant reduction in the grain yield, suggesting that a high Si uptake during this period is required for producing a high yield. Therefore, the increased expression of *Lsi1* during the heading stage coincides with a high Si requirement during this growth stage. The expression of *Lsi1* was down-regulated by dehydration stress and abscisic acid (ABA) [31]. ABA is known to accumulate in response to water stress. Therefore, the expression of *Lsi1* may be regulated by ABA. Some ABA-responsive motifs exist in the promoter region of *Lsi1*. How ABA regulates *Lsi1* expression remains to be examined.

Cellular and subcellular localization of Si transporter Lsi1. Using transgenic rice plants carrying the open reading frame (ORF) for Lsi1 fused with GFP under the control of the Lsi1 promoter region (2 kb), Lsi1-GFP was found to be localized in the main and lateral roots, but not in root hairs [28]. This is consistent with the results of a previous physiological study that root hairs do not play any demonstrable role in Si uptake, but that lateral roots contribute significantly to Si uptake [33]. Within a root, Lsi1-GFP is at both the exodermis and endodermis, where Casparian strips exist. A subcellular study showed that Lsi1-GFP is localized on the plasma membrane. Results of *in situ* hybridization also showed that mRNA of *Lsi1* was localized on the exodermis and endodermis [28]. Immunostaining with an anti-Lsi1 polyclonal antibody confirmed the cellular location of Lsi1 (Fig. 4A–C). Furthermore, interestingly, Lsi1 is localized on the distal side of both exodermis and endodermis cells, irrespective of different roots including seminal, crown and lateral roots [31].

Characteristics of Si transporter Lsi1. The transport activity of Lsi1 has been investigated using a *Xenopus* oocyte assay system. Lsi1 shows both influx and efflux transport activity for Si [28, 34], indicating that Lsi1 is a bidirectional transporter, although it only functions as an influx transporter in rice roots [28]. This is because silicic acid transported into the root cells from the external solution by Lsi1 is immediately transported out of the cells by another transporter Lsi2 in rice as described below, generating a concentration gradient from the external solution to the root cells. Both the influx and efflux transport activity were inhibited by $HgCl_2$, an inhibitor of aquaporin [34]. The transport activity for Si was unaffected by low temper-

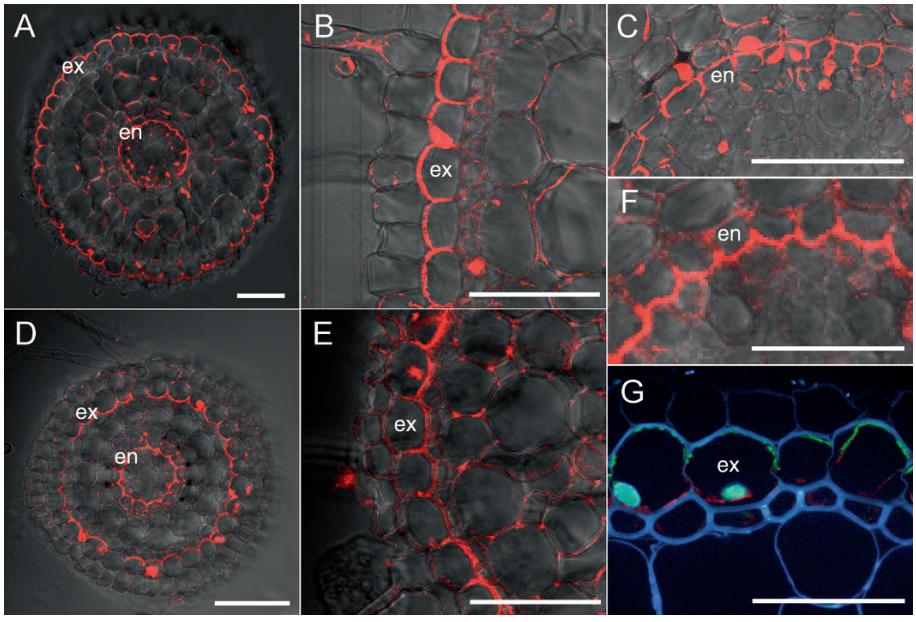

Figure 4. Localization of Si transporters. (*A*–*C*) Immunostaining of Si influx transporter Lsi1. Lsi1 is localized on the distal side of both exodermis (ex) and endodermis (en). (*D*–*F*) Immunostaining of Si efflux transporter Lsi2. Lsi2 is localized on the proximal side of both exodermis and endodermis. (*G*) Double staining of Lsi1 and Lsi2 on exodermis. Green shows Lsi1and red shows Lsi2.

ature (4°C) treatment [34]. Lsi1 is permeable to water, urea and boric acid, but not to glycerol [28, 34]. However, in the presence of equimolar concentrations of urea and boric acid, the transport activity of Si was not or only slightly affected. This indicates that Lsi1 is a highly specific transporter for silicic acid.

Lsi1 is an NIP, which are unique to plants, with 9 members in *Arabidopsis* and 10–13 members in rice [35]. Functional analysis of NIPs has revealed diversity in their transport substrates including glycerol [36], lactic acid [37], urea [38], formamide [38], and boric acid [39]. The substrate selectivity of aquaporins is mainly controlled by the ar/R (aromatic/arginine) selectivity filter [35, 40], which is located in the narrowest region on the extramembrane mouth of the pore. It is formed by four residues typically including aromatic residues and an Arg residue, one each from helix 2 (H2) and helix 5 (H5), as well as two residues from loop E (LE1 and LE2) [41]. The properties of the four residues making up the ar/R selectivity filter appear to govern the substrate specificity of the pore. Based on the ar/R regions of aquaporins, NIPs have been newly divided into three groups, NIP I, II and III (Fig. 5) [34]. NIP I proteins in Arabidopsis have been reported to transport water, glycerol [36], and lactic acid [37], while NIP II proteins are permeable to larger solutes than NIP I protein, such as urea [38], formamide [38], and boric acid [38]. Different from NIP I and NIP II, NIP III including Lsi1 has a unique selectivity filter, which consists of Gly (G), Ser (S), Gly (G), and Arg (R). It is predicted that the smaller size of the residues form a larger constriction size compared with other NIP groups [35, 42], which allows silicic acid with a larger diameter (4.38 Å) to permeate.

A number of studies indicated that the activity of both plant and animal aquaporins may be regulated by phosphorylation [43]. Recently, the involvement of phosphorylation site in the regulation of Lsi1 was investigated using two inhibitors; okadaic acid and K252a as protein phosphatase and protein kinase inhibitors, respectively. However, neither okadaic acid nor K252a affected the transport activity for silicic acid in oocytes. It is possible that, unlike in other aquaporins, phosphorylation is not involved in the regulation of Lsi1 [34]. Unlike other minerals, Si does not show excess toxicity to plants because silicic acid auto-polymerizes at higher concentrations. Furthermore, the beneficial effects of Si on plant growth are enhanced with more accumulation of Si. Therefore, it may not be necessary to regulate the activity of Si transporters by phosphorylation.

Based on the localization and transport activity of Lsi1, it is clear that Lsi1 is an influx transporter for Si, which is responsible for the transport of Si from the external solution to the root cells.

Efflux transport of Si

Cloning of Si efflux transporter gene. The second transporter gene has been cloned using a novel rice mutant (*lsi2*) defective in Si uptake using the same approach as the first mutant *lsi1* [44]. This gene *Lsi2* is located on chromosome 3 and consists of two exons and one intron. The ORF of this gene is 1416 bp long and the deduced protein consists of 472 amino acids. Sequence comparison showed that a single base mutation in the first exon occurred from G in the wild type to A in the mutant, resulting in an amino acid

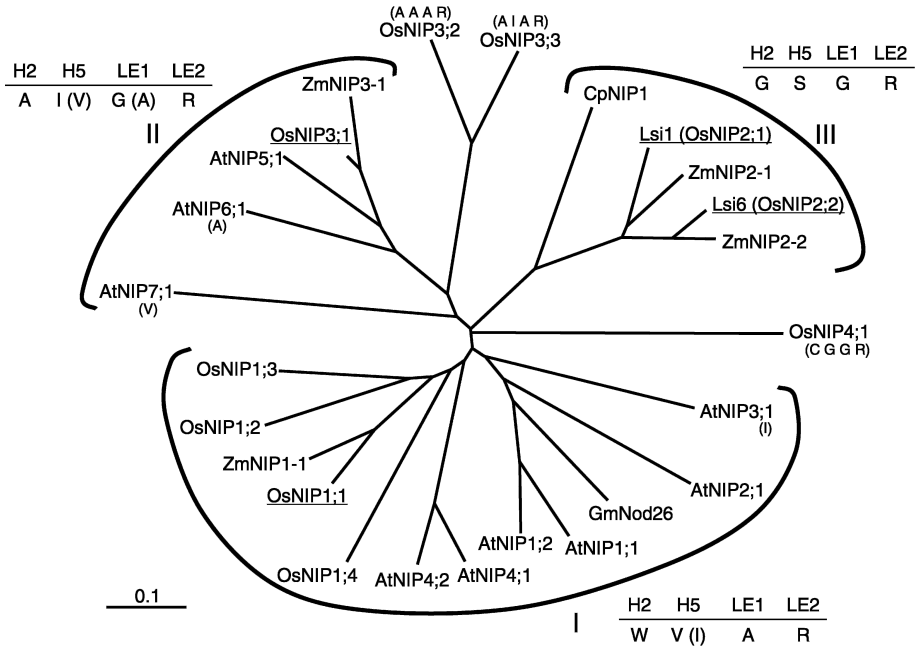

Figure 5. Polygenetic tree of Nod26-like major intrinsic proteins (NIPs) in plants. NIPs in rice (Os-), *Arabidopsis* (At-), maize (Zm-), soybean nodulin-26 (GmNod26) and zucchini CpNIP1 are shown. Plant NIPs are classified into three subgroups based on the ar/R selectivity filter formed by four amino acid residues (helix2: H2, helix5: H5, loop E1: LE1 and loop E2: LE2). The ar/R residues of each subgroup are indicated in the figure and minor substitutions of those residues are given in parentheses. For details, refer to the text.

change from serine in the wild type to asparagine in the mutant at position 115. The gene is predicted to encode a membrane protein with 11 transmembrane domains. BLAST search and ClustalW analysis revealed that Lsi2 belongs to a putative anion transporter without any similarity with the Si influx transporter Lsi1 [28, 44]. There are six full-length homologs in higher plants in the database, including one in *Arabidopsis* and five in rice. Some close homologs have also been reported as expressed sequence tag (EST) clones from other gramineous crops including wheat, barley, maize, sorghum and sugarcane. The closest homolog in rice (Os10 g0547500) has an identity of 81 % with Lsi2 [44].

Expression pattern of Lsi2. *Lsi2* was mainly expressed in the roots as *Lsi1* (Fig. 3A). There was no difference in the mRNA accumulation pattern between *Lsi2* (wild type) and *lsi2* (mutant) [44]. The mRNA accumulation was constitutive but it was decreased to one fourth by continuous Si supply for 3 days (Fig. 3B). Furthermore, there was little accumulation of *Lsi2* transcripts in the root tip (0–10 mm), but much accumulation in the mature parts of the roots [44]. In the field, similar to *Lsi1*, there was also a transient increase in the expression of *Lsi2* around the heading stage (Ma et al., unpublished data). These expression patterns are comparable to those of *Lsi1* as described above, suggesting that the expression of *Lsi1* and *Lsi2* may be regulated in a similar manner. Comparison of the promoter region revealed common domains in the *Lsi1* and *Lsi2* [44]. The expression of *Lsi2* is also regulated by ABA as seen in *Lsi1*.

Cellular and subcellular localization of Lsi2. Like Lsi1, Lsi2 is also localized on the exodermis and the endodermis cells of the roots (Fig. 4D–F). However, in contrast to Lsi1 localized on the distal side, Lsi2 was localized on the proximal side of these cells [44]. Subcellular study also showed that Lsi2 is localized on the plasma membrane [44].

Characteristics of Lsi2 transporter. In *Xenopus laevis* oocytes, Lsi2 did not show influx transport activity for silicic acid but did show efflux transport activity [44]. These results suggest that in contrast to influx transporter Lsi1, Lsi2 is a Si efflux transporter, capable of transporting Si out of the cells. Interestingly, the efflux of Si was inhibited at low temperatures and by three protonophores; 2,4-dinitrophenol (DNP), carbonylcyanide 3-chlorophenylhydrazone (CCCP) and carbonylcyanide *p*-(trifluoromethoxy)phenylhydrazone (FCCP) [44]. Furthermore, the efflux activity of Lsi2 was increased at lower external pH values [44]. These results suggest that transport of Si by Lsi2 is an energy-dependent active process, which is driven by the proton gradient (Fig. 6).

Efficient transport system in rice by coupling of Lsi1 with Lsi2. As described above, both Si transporters (Lsi1 and Lsi2) are localized on the exodermis and endodermis, where Casparian strips exist (Fig. 4G). Casparian strips prevent solutes from passing freely from the external solution to the stele. Si is highly deposited on the exodermis and endodermis, and both Lsi1 and Lsi2 are required for transcellular transport of Si to the stele. Moreover, rice roots are charac-

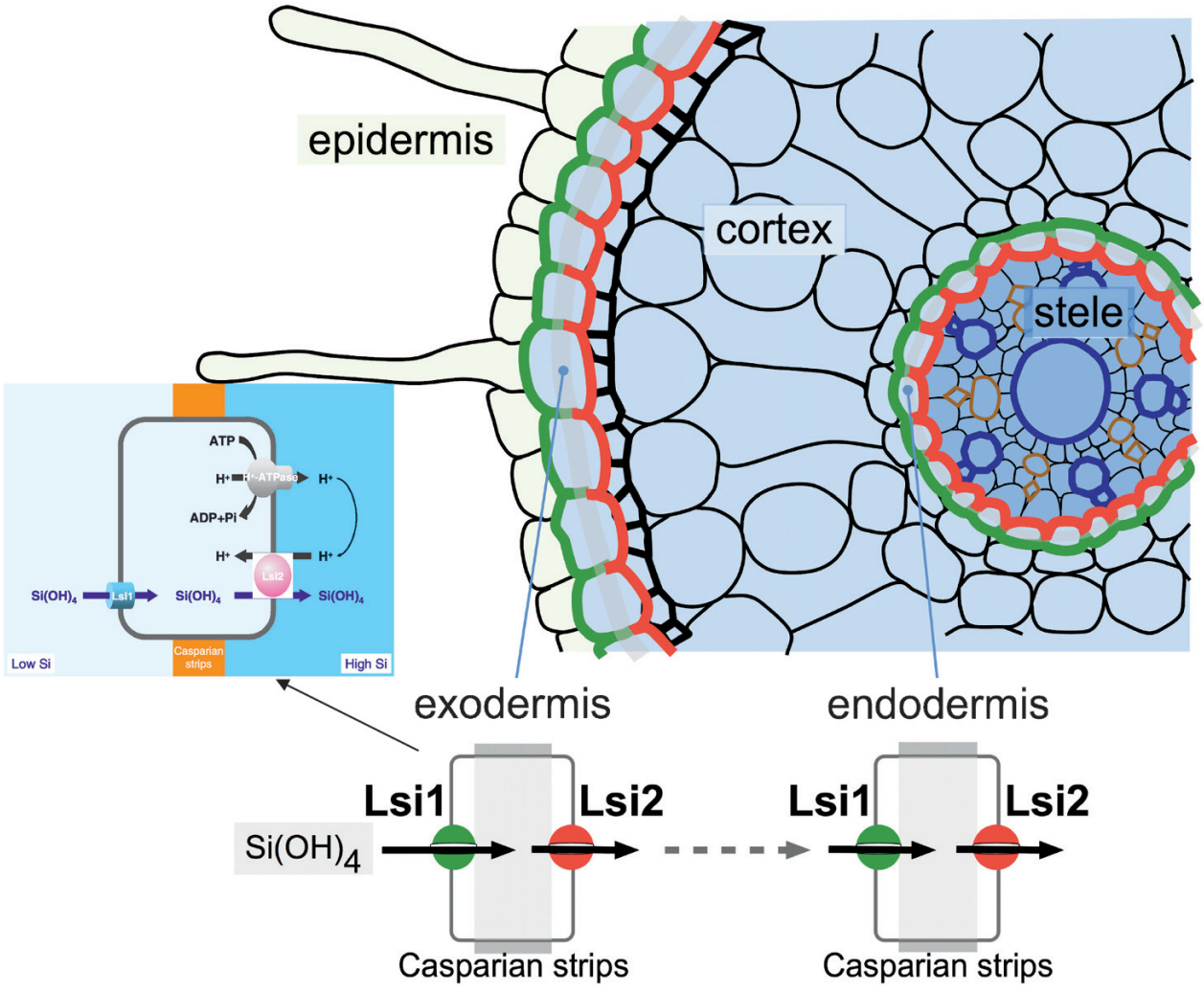

Figure 6. A schematic presentation of Si transport system in rice roots. Si as silicic acid is transported passively from the external solution to the root exodermal cells by Lsi1 and then released actively to the apoplast of the cortex by Lsi2. Silicic acid is then transported into the endodermis cells by Lsi1 and released to the stele by Lsi2. Coupling Lsi1 with Lsi2 is required for the efficient transport of Si in rice roots.

terized by the formation of the aerenchyma, which is accompanied by the destruction of almost all cortex cells except the exodermis and the endodermis. Therefore, Si transported into the exodermis cells by Lsi1 is released by Lsi2 into the apoplast of a spoke-like structure across the aerenchyma. Si is then transported into the endodermis cells by Lsi1 and released into the stele by Lsi2 (Figs. 4G and 6). Therefore, coupling of Lsi1 and Lsi2 in the same cell in the Casparian strips is required for efficient transport of Si across the cells into the stele.

A recent study showed that the genotypic difference in the Si accumulation results from the difference in the expression of Si transporter genes in rice roots [45]. The expression of both Lsi1 and Lsi2 was lower in a variety with low Si uptake capacity than in a variety with high Si uptake capacity. However, there is no difference in the cellular localization of these two transporters between low and high Si uptake varieties. Therefore, high expression of both Lsi1 and Lsi2 is required for high Si uptake in rice roots.

Future prospectives

Diatoms require Si as an essential element, and their Si uptake may include active transport by Si transporters at low Si and diffusional transport controlled by the capacity of intracellular pools in relation to cell wall silica incorporation at high Si [46]. A gene family encoding Si transporters in diatoms has been identified [47, 48]. However, no similarity between these genes and *Lsi1* and *Lsi2* has been identified. This suggests that higher plants have a distinct transport system for Si, which is different from that in diatoms. However, so far, only the two transporters described above have been identified in higher plants. Cloning of

more genes from different plant species will help in gaining further insight into the molecular mechanisms of Si uptake. For example, the Si concentration in the cell saps of rice root tips is higher than that of external solution [27], suggesting that other uncharacterized active transporters are involved in the uptake of Si. Some dicots also accumulate Si, although compared to rice the extent is lower, but it is unknown whether Si transporters are involved in Si uptake in these plant species. Furthermore, since many plants are not able to accumulate Si at levels high enough to be beneficial, so that genetic manipulation of the root's Si uptake capacity may help plants to accumulate more Si, thereby improving the plants ability to overcome biotic and abiotic stresses.

Following uptake by the roots, Si is translocated to the shoot *via* the xylem. In the shoot, silicic acid is further concentrated through loss of water (transpiration) and is polymerized and finally deposited on specific cells. Therefore, it is likely that transporters for the xylem loading and unloading of Si and the distribution of Si are also required. Their transporters still need to be identified.

Both influx and efflux Si transporters show polar localization at the plasma membrane. The mechanisms involved in the polar localization are unknown. Elucidation of these mechanisms will help us to gain a better understanding of the transport system not only of Si but also other minerals.

Acknowledgements. Work on silicon transporters in the laboratory of J.F.M. is supported by a Grant-in-Aid for Scientific Research on Priority Areas from the Ministry of Education, Culture, Sports, Science and Technology of Japan (No. 17078008 to J.F.M.) and a grant from the Ministry of Agriculture, Forestry and Fisheries of Japan (Genomics for Agricultural Innovation IPG0006 to J.F.M.).

1 Sripanyakorn, S., Jugdaohsingh, R., Thompson, R. P. H. and Powell, J. J. (2005) Dietary silicon and bone health. Nutr. Bull. 30, 222–230.
2 Lopez, P. J., Descles, J., Allen, A. E. and Bowler, C. (2005) Prospects in diatom research. Curr. Opin. Biotechnol. 16, 180–186.
3 Arnon D. I., and Stout, P. R. (1939) The essentiality of certain elements in minute quantity for plants with special reference to copper. Plant Physiol. 14, 371–375.
4 Epstein, E. and Bloom, A. J. (2003). Mineral Nutrition of Plants: Principles and Perspectives, 2nd edn, John Wiley & Sons, New York.
5 Ma, J. F. and Takahasi, E. (2002) Soil, Fertilizer, and Plant Silicon Research in Japan. Elsevier Science, Amsterdam.
6 Ma, J. F. and Yamaji, N. (2006) Silicon uptake and accumulation in higher plants. Trends Plant Sci. 11, 392–397.
7 Fauteux, F., Chain, F., Belzile, F., Menzies, J. G. and Belanger, R. R. (2006) The protective role of silicon in the Arabidopsis-powdery mildew pathosystem. Proc. Nat. Acad. Sci. USA 103, 17554–17559.
8 Fauteux, F., Remus-Borel, W., Menzies, J. G. and Belanger, R. R. (2005) Silicon and plant disease resistance against pathogenic fungi. FEMS Microbiol. Lett. 249, 1–6.
9 Savant, N. K., Snyder, G. H. and Datnoff, L. E. (1997) Silicon management and sustainable rice production. Adv. Agron. 58, 151–199.
10 Moreas, J. C., Goussain, M. M., Basagli, M. A. B., Carvalho, G. A., Ecole C. C. and Sampaio, M. V. (2004) Silicon influence on the tritrophic interaction: Wheat plants, the greenbug *Schizaphis graminum* (*Rondani*) (*Hemiptera : Aphididae*), and its natural enemies, *Chrysoperla externa* (Hagen) (*Neuroptera : Chrysopidae*) and *Aphidius colemani viereck* (*Hymenoptera : Aphidiidae*). Neotrop. Entomol. 33, 619–624.
11 Belanger, R. R., Benhamou, N. and Menzies, J. G. (2003) Cytological evidence of an active role of silicon in wheat resistance to powdery mildew (*Blumeria graminis f.* sp *tritici*). Phytopathology 93, 402–412.
12 Fawe, A., Abou-Zaid, M., Menzies, J. G. and Belanger, R. R. (1998) Silicon-mediated accumulation of flavonoid phytoalexins in cucumber. Phytopathology 88, 396–401.
13 Rodrigues, F. A., Mcnally, D. J., Datnoff, L. E., Jones, J. B., Labbe, C., Benhamou, N., Menzies, J. G. and Belanger, R. R. (2004) Silicon enhances the accumulation of diterpenoid phytoalexins in rice: A potential mechanism for blast resistance. Phytopathology 94, 177–183.
14 Remus-Borel, W., Menzies, J. G. and Belanger, R. R. (2005) Silicon induces antifungal compounds in powdery mildew-infected wheat. Physiol. Mol. Plant Pathol. 66, 108–115.
15 Cherif, M., Asselin, A. and Belanger, R. R. (1994) Defense responses induced by soluble silicon in cucumber roots infected by *Pythium* spp. Phytopathology 84, 236–242.
16 Rodrigues, F. A., Jurick, W. M., Datnoff, L. E., Jones, J. B. and Rollins, J. A. (2005) Silicon influences cytological and molecular events in compatible and incompatible rice-*Magnaporthe grisea* interactions. Physiol. Mol. Plant Pathol. 66, 144–159.
17 Ma, J. F. (2004) Role of silicon in enhancing the resistance of plants to biotic and abiotic stresses. Soil Sci. Plant Nutr. 50, 11–18.
18 Epstein, E. (1999) Silicon. Annu. Rev. Plant Physiol. Plant Mol. Biol. 50, 641–664.
19 Hodson, M. J., White, P. J., Mead, A. and Broadley, M. R. (2005) Phylogenetic variation in the silicon composition of plants. Ann. Bot. 96, 1027–1046.
20 Ma, J. F., Higashitani, A., Sato, K. and Tateda, K. (2003) Genotypic variation in Si content of barley grain. Plant Soil 249, 383–387.
21 Dern, C. W. (2001) Plant genotype, silicon concentration, and silicon-related responses. In: Silicon in Agriculture, pp. 149–158, Datnoff, L. E., Snyder, G. H. and Korndorfer, G. H. (eds.); Elsevier Science, Amsterdam.
22 Deren, C. W., Datnoff, L. E. and Snyder, G. N. (1992) Variable silicon content of rice cultivars grown on Everglades Histosols. J. Plant Nutr. 15, 2363–2368.
23 Winslow, M. D. (1992) Silicon, disease resistance, and yield of rice genotypes under upland cultural conditions. Crop Sci. 32, 1208–1213.
24 Winslow, M. D., Okada, K. and CorreaVictoria, F. (1997) Silicon deficiency and the adaptation of tropical rice ecotypes. Plant Soil 188, 239–248.
25 Takahashi, E., Ma, J. F. and Miyake, Y. (1990) The possibility of silicon as an essential element for higher plants. Comments Agric. Food Chem. 2, 99–122.
26 Takahashi, E. and Hino, K. (1978) Silicon uptake by plant with special reference to the forms of dissolved silicon. Jpn. J. Soil Sci. Manure 49, 357–360.
27 Mitani, N. and Ma, J. F. (2005) Uptake system of silicon in different plant species. J. Exp. Bot. 56, 1255–1261.
28 Ma, J. F., Tamai, K., Yamaji, N., Mitani, N., Konishi, S., Katsuhara, M., Ishiguro, M., Murata, Y. and Yano, M. (2006) A silicon transporter in rice. Nature 440, 688–691.
29 Ma, J. F., Tamai, K., Ichii, M. and Wu, K. (2002) A rice mutant defective in active Si uptake. Plant Physiol. 130, 2111–2117.
30 Ma, J. F., Mitani, N., Nagao, S., Konishi, S., Tamai, K., Iwashita, T. and Yano, M. (2004) Characterization of Si uptake system

and molecular mapping of Si transporter gene in rice. Plant Physiol. 136, 3284–3289.
31 Yamaji, N. and Ma, J. F. (2007) Spatial distribution and temporal variation of the rice silicon transporter Lsi1. Plant Physiol. 143, 1306–1313.
32 Ma, J. F., Nishimura, K. and Takahashi, E. (1989) Effect of silicon on the growth of rice plant at different growth stages. Soil Sci. Plant Nutr. 35, 347–356.
33 Ma, J. F., Goto, S., Tamai, K and Ichii, M. (2001) Role of root hairs and lateral roots in silicon uptake by rice. Plant Physiol. 127, 1773–1780.
34 Mitani, N., Yamaji, N. and Ma, J. F. (2008) Characterization of substrate specificity of a rice silicon transporter, Lsi1. Pflugers Arch. 456, 679–686.
35 Forrest, K. L. and Bhave, M. (2007) Major intrinsic proteins (MIPs) in plants: A complex gene family with major impacts on plant phenotype. Funct. Integr. Genomics 7, 263–289.
36 Dean, R. M., Rivers, R. L., Zeidel, M. L., Roberts, D. M. (1999) Purification and functional reconstitution of soybean nodulin 26. An aquaporin with water and glycerol transport properties. Biochemistry 38, 347–353.
37 Choi, W. G. and Roberts, D. M. (2007) Arabidopsis NIP2;1: A major intrinsic protein transporter of lactic acid induced by anoxic stress. J. Biol. Chem. 282, 24209–24218.
38 Wallace, I. S. and Roberts, D. M. (2005) Distinct transport selectivity of two structural subclasses of the nodulin-like intrinsic protein family of plant aquaglyceroporin channels. Biochemistry 44, 16826–16834.
39 Takano, J., Wada, M., Ludewig, U., Schaaf, G., von Wiren, N. and Fujiwara, T. (2006) The Arabidopsis major intrinsic protein NIP5;1 is essential for efficient boron uptake and plant development under boron limitation. Plant Cell 18, 1498–1509.
40 Wallace, I. S. and Roberts, D. M. (2004) Homology modeling of representative subfamilies of *Arabidopsis* major intrinsic proteins. Classification based on the aromatic/arginine selectivity filter. Plant Physiol. 135, 1059–1068.
41 Ilan, B., Tajkhorshid, E., Schulten, K. and Voth, G. A. (2003) Electrostatic tuning of permeation and selectivity in aquaporin water channels. Biophys. J. 85, 2884–2899.
42 Wang, Y., Schulten, K. and Tajkhorshid, E. (2005) What makes an aquaporin a glycerol channel? A comparative study of AqpZ and GlpF. Structure 13, 1107–1118.
43 Agre, P. and Kozono, D. (2003) Aquaporin water channels: Molecular mechanisms for human diseases. FEBS Lett. 555, 72–78.
44 Ma, J. F., Yamaji, N., Mitani, N., Tamai, K., Konishi, S., Fujiwara, T., Katsuhara, M. and Yano, M. (2007) An efflux transporter of silicon in rice. Nature 448, 209–211.
45 Ma, J. F., Yamaji, N., Tamai, K. and Mitani, N. (2007) Genotypic difference in Si uptake and expression of Si transporter genes in rice. Plant Physiol. 145, 919–924.
46 Thamatrakoln, K. and Hildebrand, M. (2007) Silicon uptake in diatoms revisited: A model for saturable and nonsaturable uptake kinetics and the role of silicon transporters. Plant Physiol. 146, 1397–1407.
47 Hildebrand, M., Higgins, D. R., Busser, K. and Volcani, B. E. (1993) Silicon-responsive cDNA clones isolated from the marine diatom *Cylindrotheca fusiformis.* Gene 132, 213–218.
48 Hildebrand, M., Volcani, B. E., Gassmann, W. and Schroeder, J. I. (1997) A gene family of silicon transporters. Nature 385, 688–689.
49 Tamai, K. and Ma, J. F. (2008) Reexamination of silicon effects on rice growth and production under field conditions using a low silicon mutant. Plant Soil (in press).