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Agriculture and Agricultural Science Procedia 11 (2016) 100 – 104

Agriculture and Agricultural Science Procedia

International Conference on Inventions & Innovations for Sustainable Agriculture 2016, ICIISA 2016

The Source of Silicon for Thai Riceberry Germinated on Top of an Aqueous Solution

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Abstract

Rice plants accumulate silicon (Si) in various parts of the body as a form of body protection as well as for fighting against stresses. In this study, we used Thai Riceberry grown on top of a Si-free water as a model to study Si accumulation. Scanning Electron Microscope and Energy Dispersive X-ray Spectrometer (SEM-EDX) together with Inductively Couple Plasma-Optical Emission Spectrometer (ICP-OES) were used. It was found that rice husks contained a large amount of Si in the form of silicon dioxide at an average level of 23,104.28 ± 2021.27 µg Si/g husk. Rice could grow well and accumulated Si in whole body tissues from 170.00 ± 126.48 µg Si/g tissue at the start of germination to 1,380.00 ± 667.16 µg Si/g tissue when the rice grew to 10 cm long. SEM-EDX analysis revealed that Si was accumulated the most in leaves and crystalized to form dumbbell shape-like bodies. Husks would therefore most likely be the source of Si during early germination and growth of the rice plant seedlings.

Peer-review under responsibility of the Faculty of Animal Sciences and Agricultural Technology, Silpakorn University

Keywords: Thai Riceberry; ICP-OES, Silicon; SEM-EDX; Silica bodies

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1. Introduction

Rice plants take up silicon into their bodies through their roots in the form of soluble silicic acid (Tamai and Ma, 2003). The silicic acid accumulates in rice plant organs, polymerizes to create intracellular or extracellular silica bodies

Peer-review under responsibility of the Faculty of Animal Sciences and Agricultural Technology, Silpakorn University
doi:10.1016/j.aaspro.2016.12.017

(Ma and Takahashi, 2002). It is well accepted that silica renders strength to the stalk and stem, helping to toughen stem and widen the leaves of rice plants (Yamanaka et al., 2009). Additionally, silica accumulated in husks to give protection to the seeds (Takahashi et al., 2006). There are evidences indicating that silica also guards against the attack by pests and fungi and offers protection to the plant from stresses such as heavy metal toxicity (Ma, 2004)

Most of the experiments on silicon and rice seedlings were done by adding silicon compounds to the growing media (Ma et al., 1989; Hossain et al., 2002; Hayasaka et al., 2005; Gong et al., 2006; Singh et al., 2011) or by adding rice hull ash (Sistani et al., 1997). Since there are a good number of studies showing that rice husks contain substantial amount of silicon in the form of silicon dioxide (Takahashi et al., 2006; Van Soest, 2006) we, therefore, are interested in investigating if the husks would serve as an important silicon reserve for growth during the germination of rice caryopses. The investigation was done by germinating Thai Riceberry on top of a Si-free water. The growth of rice seedlings was followed while silicon accumulation in the whole plant body parts were examined. Husks from different stages of germination were separated from the rice plant bodies to examine Si contents at the same time.

2. Materials and Methods

2.1. Thai Riceberry cultivation on an aqueous solution

Thai Riceberry seeds were purchased from a commercial supplier in Nakorn Pathom. The rice seeds were incubated in water overnight and were dispersed on a plastic sieve and covered by wet tissue papers. The rice seeds were sprinkled with Si-free water once a day until the rice grew to about 1-1.5 cm. The rice seedlings were placed on top of 4 L of Si-free water in triplicate plastic boxes and allowed to grow under a normal light condition, having only the roots submerged in water. Rice seedlings were grouped according to their body lengths as 0, 2, 4, 6, 8 and 10 cm. Husks were separated from all other germinated parts of the rice at the time of harvesting. The husks and the other rice parts were dried in an oven at 90 ^{0}C for 3 days. Samples were taken for analysis using a Scanning Electron Microscope-Energy Dispersive X-ray spectrometer and an Inductively Couple Plasma-Optical Emission Spectrometer.

2.2. Treatments of husks and the seedling parts for the analysis with an Inductively Couple Plasma-Optical Emission Spectrometer (ICP-OES)

The dried husks and seedling parts were heated at 750 ^{0}C in a furnace for 4 hours to yield ashes. The fusion process for each ash sample was done by adding 2.5 g NaOH and heated with a Bunsen burner for 15 min. After the sample cooled down to room temperature, 20 ml of DI water was added, then followed by 15 ml of H_2O:50% (v/v) HNO_3 (1:1 by volume). The solution was adjusted to a final volume of 100 ml using DI water. The sample solutions were analyzed by an ICP-OES (PerkinElmer Optima 7300).

2.3. Treatments of the husks and the seedling parts for the examination with a Scanning Electron Microscope-Energy Dispersive X-ray Spectrometer (SEM-EDX)

The dried husks and seedling parts were mounted on aluminium stubs via double-sided conductive carbon tapes. The specimens were examined under a Tabletop Hitachi Scanning Electron Microscope Model 3030Plus without metal sputtering. The specimen was positioned at 4.5 mm below the BSE detector and examined at 15 keV. The scanning electron microscope images were taken under the mixed mode of BSE and SE. X-ray microanalysis was done by using the built-in silicon drift detector (SDD) to obtain elemental compositions of the samples. EDX spectra were obtained using a sensitive mode with the scanned time of 3 min. The identification of the elements was done using an auto identification against the built-in elemental database library.

2.4 Statistical analysis

Numerical data were analysed using One-way analysis of variance (ANOVA) while the statistical insignificance of the data was set at $p<0.05$. SPSS software version 18 was used to process the data.