Ecotoxicology and Environmental Safety 164 (2018) 562–570

Contents lists available at ScienceDirect

Ecotoxicology and Environmental Safety

journal homepage: www.elsevier.com/locate/ecoenv

Selenium protects rice plants from water deficit stress

Fabrício Ribeiro Andrade[a], Geanderson Nascimento da Silva[b], Karina Carvalho Guimarães[c], Herlon Bruno Ferreira Barreto[d], Kamila Rezende Dázio de Souza[e], Luiz Roberto Guimarães Guilherme[a], Valdemar Faquin[a], André Rodrigues dos Reis[f,*]

[a] Department of Soil Science, Federal University of Lavras, Postal Code 3037, 37200-000 Lavras, MG, Brazil
[b] Federal Institute of Education, Science and Technology of Sertão of Pernambuco, Postal Code 56304-060, Petrolina, PE, Brazil
[c] Department of Food Science, Federal University of Lavras, Postal Code 3037, 37200-000 Lavras, MG, Brazil
[d] Department of Engineering, Federal University of Lavras, Postal Code 3037, 37200-000 Lavras, MG, Brazil
[e] Department of Biology, Federal University of Lavras, Postal Code 3037, 37200-000 Lavras, MG, Brazil
[f] School of Science and Engineering, São Paulo State University - UNESP, Postal Code 17602-496, Tupã, SP, Brazil

ARTICLE INFO

Keywords:
Agronomic biofortification
Sodium selenate
Food composition
Antioxidant enzymes
Abiotic stress

ABSTRACT

Selenium (Se) is essential to humans and animals due to its antioxidant properties. Although it is not considered an essential nutrient for higher plants. Many studies show that Se in low concentrations (up to 0.5 mg kg^{-1}) provides beneficial effects to non-hyperaccumulating plants by participating in antioxidant defense systems and enhancing tolerance to abiotic stress. Therefore, this study aimed to evaluate the effects of Se application rates on rice plants under different soil water conditions. The experiment was conducted on an Oxisol using four Se rates (0, 0.5, 1.0 and 2.0 mg kg^{-1}) and two soil water conditions (irrigated and water deficit). Selenium application via soil up to 0.5 mg kg^{-1} increased the plant height, chlorophyll index, sulfur and copper accumulation in shoots, carbon dioxide assimilation, superoxide dismutase (EC 1.15.1.1) activity and decreased the hydrogen peroxide concentration in rice leaves. The accumulation of Se in shoot biomass and Se concentration in seeds increased linearly with the applied rates. Water deficit strongly decreased the plant growth and yield. However, rice plants treated with Se showed higher net photosynthesis, water use efficiency and antioxidant system. This study provides useful information about the roles of Se in protecting rice plants from water deficit stress.

1. Introduction

Cultivated crops are frequently exposed to adverse conditions that affect their growth and productivity. Among the various types of environmental stress, drought is considered the most devastating because of crop yield losses, which can reach billions of dollars annually worldwide (Lambers et al., 2008; Tardieu et al., 2014; Aon, 2015). It is estimated that by 2050, when the world population is expected to reach 9.7 billion people, approximately 49% of the global grain production will be cultivated in regions affected by water deficit (Rosegrant, 2016).

Rice (*Oryza sativa* L.) is the second most cultivated cereal in the world and the main food source for more than half of the world population (Reis et al., 2018). Approximately 75% of rice production worldwide comes from planting in an irrigated and/or flooded system (FAO, 2017). However, these cultivated area expansions are limited due to strong impacts on environmental problems (Sander et al., 2014). Therefore, upland rice cultivation tends to expand into arid regions, where dry spells are more consistent. These regions are characterized by long periods of soil water deficit due to interruptions in rainfall during the rainy season (Joy et al., 2015).

Water deficit reduces soil water potential. This reduction has direct implications for transpiration, photosynthesis, leaf temperature, stomatal opening and antioxidant metabolism, all of which affect the growth, development and especially the yield of economically important crops (Nawaz et al., 2015; Reis et al., 2015). Application of selenium (Se) can increase growth and minimize the effects of abiotic stresses (e.g., drought, salinity, high temperatures and potentially toxic element) in plants (Djanaguiraman et al., 2010; Kumar et al., 2012;

Abbreviations: A, CO_2 assimilation rate; APX, ascorbate peroxidase; Ci, internal CO_2 concentration; CAT, catalase; E, transpiration; EiC, instantaneous carboxylation efficiency; FW, fresh weight; gs, stomatal conductance; H_2O_2, hydrogen peroxide; MDA, malondialdehyde; ROS, reactive oxygen species; SOD, superoxide dismutase; WUE, water use efficiency

* Corresponding author.
E-mail address: andre.reis@unesp.br (A.R.d. Reis).

https://doi.org/10.1016/j.ecoenv.2018.08.022
Received 22 March 2018; Received in revised form 27 July 2018; Accepted 6 August 2018

Nawaz et al., 2015; Jiang et al., 2017; Reis et al., 2017, 2018). There are conclusive evidences of Se deficiency in Brazilian soils (Reis et al., 2018; Silva et al., 2018). Selenium variation in Brazilian soils range from 10 to 150 μg kg^{-1} (Reis et al., 2017). Therefore, Se supplementation is needed in order to reduce The harmful effects on plant metabolism and physiology caused by drought stress can be decreased by Se supplementation. Selenium increases the activity of antioxidant enzymes and reduces both the generation of reactive oxygen species (ROS) and the lipid peroxidation rates in the leaf cell tissue (Habibi, 2013; Nawaz et al., 2015; Mostofa et al., 2017; Reis et al., 2018).

An increase in CO_2 assimilation rate and stomatal conductance is promoted by application of Se in wheat (*Triticum aestivum* L.) and barley (*Hordeum vulgare* L.) plants exposed to drought stress (Habibi, 2013; Nawaz et al., 2015). Similarly, in olive (*Olea europaea* L.) under water stress, foliar applications of Se can increase photosynthesis, regulate the water status of trees and maintain a sufficiently high level of leaf water content in plants (Proietti et al., 2013). Yao et al. (2009) observed significant increases in root activity, catalase activity and chlorophyll concentration as well as decreased malondialdehyde concentration in wheat seedlings subjected to Se application under drought stress.

Therefore, studies that demonstrate the positive effects of Se application on rice plants under water deficit conditions are highly relevant. This study aimed to validate the effects of Se doses applied to the soil under two soil water conditions on the growth, yield, antioxidant system and nutritional quality status of rice plants.

2. Materials and methods

2.1. Experimental design and rice cultivation

The experiment was conducted in a greenhouse from July to December 2014 at the Soil Science Department of Federal University of Lavras, Brazil. Pots with 4 kg of Oxisol (clay textured) collected from the 0–0.20-m layer were used. The soil was air-dried, sieved using a 4-mm mesh and characterized for its major chemical and physical properties according to the methodology suggested by EMBRAPA (1997). The chemical and physical properties as follows: pH-H_2O = 5.1; organic matter = 46 g kg^{-1}; P (Mehlich-1) = 0.84 mg kg^{-1}; K = 1.7 mmolc dm^{-3}; Ca = 1.5 cmolc dm^{-3}; Mg = 0.4 cmolc dm^{-3};Al = 0.5 cmolc dm^{-3}; HAl = 6.3 cmolc dm^{-3}; P-rem = 11.37 mg L^{-1}; clay = 710 g kg^{-1}; silt = 140 g kg^{-1}; and sand = 150 g kg^{-1}. The content of Se was 0.2 ± 0.04 mg kg^{-1}, which was determined after soil digestion according the USEPA 3051A method (USEPA, 1998).

Based on soil chemical analysis, liming was carried out to raise the base saturation to 60% using $CaCO_3$ and $MgCO_3$ at a ratio of 4:1. After 30 days of soil incubation with a humidity close to 60% of the total pore volume (TPV), doses of Se (sodium selenate, Sigma-Aldrich, Saint Louis, USA) were applied to the soil. Each pot received a macronutrient fertilizer containing: 80 mg of N, 250 mg of P, 90 mg of K, and 50 mg of S kg^{-1} soil. Micronutrient applications consisted of 0.5 mg of B, 1.5 mg of Cu, 0.1 mg of Mo and 5.0 mg of Zn kg^{-1} soil.

Afterward, 15 rice seeds (*O. sativa* L. cv. IAC 202) were sown per pot, and nine days after seedling emergence, the rice seedlings were thinned three plants per pot. In addition, during the rice cultivation period, blanket fertilizations of 473 mg of N and 436 mg of K kg^{-1} soil were carried out and divided into five applications.

The experiments were set up as a completely randomized design in a 4 × 2 factorial scheme with four Se doses (0, 0.5, 1.0 and 2.0 mg kg^{-1} Se) and two soil water conditions (irrigated and water deficit), with four replicates, during the beginning of the reproductive stage. The control treatment consisted of maintaining the soil near field capacity, and water deficit treatment involved maintaining the soil at approximately − 50 kPa, as determined by for rice cultivation. Each experimental unit consisted of two pots: one used for the analyses performed (gas exchange, antioxidant system, SPAD index growth and nutrition) at the end of water deficit, and the other was used only at seed harvest.

To monitor soil water tension, tensiometers were installed at a 0.15-m depth in each experimental plot, and the replenishment of evapotranspired water was performed based on the tensiometric reading, which was performed twice daily at 9:00 and 16:00 h. Appropriate water replenishment was used to determine water retention curves for the soil. From emergence to the flag leaf/collar-formation stage, field capacity was maintained by soil moisture in all pots.

In the flag leaf/collar-formation stage, water deficit treatments were applied to the respective pots, and soil water potential was maintained at − 50 kPa for 14 days to simulate dry spells; samples not subjected to water deficit were maintained under normal irrigation conditions. At the end of this period, irrigation to plants under water stress was restored to field capacity until harvest.

2.2. Gas exchange measurements

On the 14th day of water stress at the end of water deficit period, which coincided with the initial panicle exsertion stage of rice, gas exchange evaluations were performed using a portable infrared gas analyzer (Infra Red Gas Analyzer - IRGA, brand LI-COR Biosciences, model LICOR 6400). The CO_2 assimilation rate expressed by area (*A* - μmol CO_2 m^{-2} s^{-1}), stomatal conductance (*gs* - mol H_2O m^{-2} s^{-1}), transpiration (*E* - mmol H_2O m^{-2} s^{-1}) and internal CO_2 concentration in the substomatal chamber (*Ci* - μmol CO_2 mol air^{-1}) were obtained. With those data, an estimation of both instantaneous carboxylation efficiency [*EiC*, (*A*/*Ci* - mol air^{-1})] and water use efficiency [*WUE*, (*A*/*E* - μmol CO_2 mmol^{-1} H_2O)] were determined. Readings were performed on a clear day between 9:00 and 11:00 a.m. using the flag leaf as a pattern, including the last fully developed leaf. The photosynthetically active radiation (PAR) was standardized to an artificial saturating light of 1000 μmol m^{-2} s^{-1} and an ambient CO_2 concentration. The average relative humidity was 30%, and the ambient temperature was between 35 and 42 °C.

2.3. Biometric and chlorophyll meter measurements

Using three leaves per pot on the same day, the SPAD index was determined using a portable chlorophyll meter (SPAD-502, Konica-Minolta, Japan). After SPAD readings, leaves were collected and immediately conditioned in liquid nitrogen, after which they were stored at − 80 °C for biochemical analyses. After all these determinations at end of the water deficit period, the height of plants at that time was obtained by measuring from the plant base to the end of last fully expanded leaf. The shoots and roots of plants were collected and dried in a forced-air oven for 72 h in order to obtain and determine the dry biomass and root/shoot relation. The other post treatments aimed at seed production were conducted until the cycle ended, after which the grains were harvested, dried in a forced-air oven, and weighed to obtain the dry mass of seeds in each pot.

2.4. Extraction and quantification of antioxidant enzymes

To quantify antioxidant enzyme activity in leaves, an extract was obtained by maceration of 0.1 g of leaves in liquid nitrogen. The extract was then added to an extraction buffer solution containing 0.1 M potassium phosphate (pH 7.8), 0.1 mM EDTA (pH 7.0), 0.01 M ascorbic acid and 22 mg of polyvinylpyrrolidone (PVPP) (Biemelt et al., 1998). The supernatant was then collected and used for enzymatic analyses of superoxide dismutase (SOD), catalase (CAT) and ascorbate peroxidase (APX).

2.4.1. Superoxide dismutase (SOD; EC 1.15.1.1)

SOD activity was evaluated by the ability of the enzyme to inhibit the photoreduction of nitroblue tetrazolium (NBT), as proposed by Giannopolitis and Ries (1977). An aliquot of the supernatant was added to an incubation medium composed of 50 mM potassium phosphate (pH