Plant Signaling & Behavior 6:7, 1024-1025; July 2011; © 2011 Landes Bioscience

Is reactive oxygen species (ROS) the underlying factor for inhibited root growth in *Osspr1*?

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Reactive oxygen species (ROS), like hydrogen peroxide (H_2O_2) and superoxide anion ($O_2^{\bullet-}$), are important plant cell signaling molecules involved in diverse physiological processes, such as programmed cell death, development, cell elongation and hormonal signaling. Recently, much attention has been paid to the role of ROS in regulating plant root development. Two ROS, superoxide and hydrogen peroxide, were shown to exhibit a typical accumulation pattern in the Arabidopsis root apex and play distinct roles in root development.[1] The latest study showed that UPBEAT1 (UPB1), a bHLH transcription factor, modulates the ROS balance by directly regulating the expression of a set of peroxidases, therefore, regulates the root cell proliferation and differentiation.[2] In this addendum, we proposed a possible hypothesis that OsSPR1 maintained the mitochondria function to restrict H_2O_2 production in root apex for normal root development.

Key words: reactive oxygen species, hydrogen peroxide, cell elongation, meristem, mitochondria

Abbreviations: ROS, reactive oxygen species; AOX, alternative oxidase; H_2O_2, hydrogen peroxide; bHLH, basic helix-loop-helix; OH, hydroxyl radical; $O_2^{\bullet-}$, superoxide anion; RAM, root apical meristem

Submitted: 03/22/11

Accepted: 03/22/11

DOI: 10.4161/PSB.6.7.15577

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Addendum to: Jia LQ, Wu ZC, Hao X, Carrie C, Zheng LB, Whelan J, et al. Identification of a novel mitochondrial protein, short postembryonic roots (SPR1), involved in root development and iron homeostasis in *Oryza sativa*. New Phytol 2011; 189:843–55; PMID: 21039568; DOI: 10.1111/j.1469-8137.

Reactive oxygen species (ROS), like superoxide anion ($O_2^{\bullet-}$), hydrogen peroxide (H_2O_2), are by-products constantly produced during normal metabolic processes, such as photosynthesis, photorespiration and cellular respiration. High level of ROS can be very lethal for the plant cell integrity. However, at lower concentrations, ROS function in signaling pathways that regulate plant development in response to physiological and environmental cues. Superoxide anion generated by one-electron reduction of oxygen by the plasma membrane NADPH oxidase play important role for root growth and root hair development.[3] The superoxide ion may be converted into H_2O_2 spontaneously which is also involved in many developmental and physiological process.[4] Hydrogen peroxide can further be reduced by peroxidases to generated more reactive free radicals hydroxyl radical (OH). This radical is very important for cell elongation by cell wall loosening mechanism.[5] Therefore, the maintenance of ROS homeostasis is crucial for plant development.

The root apex is a zone of active ROS production.[1] It comprises cells in very different states within a short distance, including cell division, elongation and differentiation zones. Different ROS distribution pattern in root apex is reported in several plants include Arabidopsis,[1,2] maize[5] and rice.[6] $O_2^{\bullet-}$ and H_2O_2 have both distinct accumulation zones and different roles in the extremity of the growing Arabidopsis.[1] The latest study indicates that UPBEAT1 (UPB1), a bHLH transcription factor, modulates the ROS balance by directly regulating the expression of a set of peroxidases, therefore, regulates the root cell proliferation and differentiation for root growth.[2] It was also proposed that maintenance of cellular proliferation requires an accumulation of $O_2^{\bullet-}$, whereas cellular differentiation requires elevated H_2O_2 levels.[2]

The *Osspr1* mutant was identified as a short root mutant with altered iron content in shoot and elevated H_2O_2 levels in the root tip. *OsSPR1* encodes a novel mitochondrial protein with the Armadillo-like repeat domain. It is well known that the mitochondria are one of the major sources for ROS production in plants. Therefore, we reasoned that OsSPR1 directly acts

in mitochondria of root meristem cells to affect respiratory electron chain for restricting the H_2O_2 production, which can maintain the ROS balance in root apex to direct normal root growth. While, in the *Osspr1* mutant, high content of H_2O_2 are generated from dysfunctional mitochondria as indicated by altered alternative oxidases (AOXs) expression levels. It altered ROS balance in the root apex and ultimately lead to a quick cellular differentiation with inhibited root cell elongation. However, we could not exclude other possibilities, such as OsSPR1 involved in the iron transport into the mitochondria to control H_2O_2 production, since a higher root iron concentration was detected in OsSPR1 overexpression lines. Although the *Osspr1* mutant root exhibited a normal iron concentration, the cellular distribution of iron in *Osspr1* root apex cells might still be altered. To determine the exact function of OsSPR1 in rice, detailed analysis of the mitochondria function, the ROS production and cellular iron distribution in *osspr1* still need to be investigated.

Acknowledgments

We thank Dr. keke Yi (Institute of Virology and Biotechnology, Zhejiang Academy of Agricultural Sciences) for critical reading and valuable suggestion. This work was supported by the Zhejiang Academy of Agricultural Sciences.

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