THE ROLE OF HYDROGEN PEROXIDE (H₂O₂) IN SENSING AND SIGNALLING ABIOTIC STIMULI IN RICE

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The Role of Hydrogen Peroxide (H₂O₂) in Sensing and Signalling Abiotic Stimuli in Rice

Nyuk Ling Ma

A Dissertation Submitted for the Degree of

Doctor of Philosophy

Department of Life Sciences Division of Cell and Molecular Biology Imperial College London 2012 To my dear husband

"The path to our destination is not always a straight one. We go down the wrong road, we get lost, we turn back.

Maybe it doesn't matter which road we embark on.

Maybe what matters is that we embark"

- Barbara Hall

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Statement of originality

This dissertation describes work performed in the Department of life sciences at the Imperial college London between February 2008 and January 2012. All the work outlined in this dissertation is my own except the where otherwise acknowledged and referenced. The work contained in the dissertation has not been previously submitted, in whole or in part, for a degree at any institution.

This dissertation is submitted in fulfilment of the requirements of the PhD. It does not exceed 100,000 words and contains fewer than 150 figures.

Abstract

This dissertation describes the integration of physiological, proteomic, and metabolomic changes in conjunction with the synthesis of H_2O_2 under various stress conditions in rice, *Oryza sativa* (Japonica var. *Koshihikari*). Dynamics of H_2O_2 production were observed in the leaves following the treatment with drought, salinity, osmotic and cold stress. The physiological parameters such as H_2O_2 content, photosynthetic activity, water content, and lipid peroxidation were used as indicators to correlate the physiological status of the plant with its metabolites and proteins profile/changes in response to stresses. A targeted proteomic approach has been developed to identify oxidative modified thiols, and a NMR-based analysis has been performed for metabolites profiling.

Activation of amino acids was observed in the treatment of short-term drought stress while deficit of sugar and lipids in addition to accumulation of amino acids were observed when treated with long-term drought stress. The high levels of lipid peroxidation and the irreversible oxidation of protein thiols observed under long-term drought stress indicate that the plants were likely to have suffered membrane damage. In osmotic stress treatment, the plants showed rapid dehydration compared to that treated with drought stress. The transient increase of both lipid peroxidation and reversible thiol oxidation suggest that signalling of adaptation had been activated, resulting in fluctuation of carbon and nitrogen metabolism in the plants. In salinity stress treatment, very low levels of lipid peroxidation, decline in photosynthetic activities, and very few metabolites changes were observed. This suggests that the dosage used in salinity test was insufficient to induce salt stress to the plants. Another possibility is that the changes in thiol proteins following the salt treatment were likely to have contributed to salt tolerance, hence keeping the metabolites in the control level. Moreover, the metabolite changes observed in the plants following exposure to exogenous H_2O_2 was found to overlap with those induced by other stresses, indicating that H2O2 is likely to have effects on the carbohydrate, fatty acid, and amino acids pathways. In summary, the results suggest that there were possible overlap of H₂O₂ mediated-signalling pathway in coordinating cellular changes at both proteomic and metabolomic level under abiotic stress conditions in rice plants. However, further studies are needed to confirm the role and the underlying mechanism of H2O2 in regulating the cellular changes.

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