Abstract

Cellular redox states mediate various physiological and developmental processes. Mechanisms involved in sensing cellular redox state and linking it to an appropriate physiological response remains poorly understood in plants. Arabidopsis bZIP68 was previously found to undergo reversible oxidation in its Cys320 in cells under oxidative stress. In this study, it was found that bZIP68 was localized in the nucleus in Arabidopsis seedlings under normal conditions. Upon treatment of oxidative stress, bZIP68 underwent nucleocytoplasmic shuttling and accumulated in the cytoplasm. This stress-dependent nucleocytoplasmic shuttling depends on the redox-sensitive Cys320 and its nuclear export signal. bZIP68 suppresses expression of stress response genes under normal conditions and its loss-of-function mutation of bZIP68 leads to elevated expression of genes involved in oxidative stress defense including genes encoding for antioxidant proteins and for enzymes involved in biosynthesis of small molecule antioxidants. The bzip68 mutant also showed enhanced responses to stress treatment such as the oxidative stress and cold stress. Our study suggests that bZIP68 directly or indirectly senses perturbation of cellular redox states and links the redox change to activation of oxidative stress defense genes through redox regulation of transcription.
Table of Contents

Chapter 1 General Introduction and the Objective.................................1

1.1 The Concepts of Redox Homostasis and Redox Signaling in Plants ...1

1.2 Production of ROS and RNS in Plants ........................................2

1.3 Antioxidants in scavenging ROS and repairing oxidized biomolecules

1.3.1 Characteristics of antioxidants ..............................................3

1.3.2 Superoxide dismutases .........................................................4

1.3.3 Catalases ..............................................................................5

1.3.4 Ascorbate and Glutathione ...................................................5

1.3.5 Others ..................................................................................7

1.4 Redox Regulation and Redox signaling in Plants .............................8

1.4.1 The term of redox regulation ..................................................8

1.4.2 Redox regulation during different biological processes ..........9

1.5 Redox signaling in environmental stress ......................................9

1.5.1 Redox signaling responding to abiotic stress ..........................10

1.5.2 Oxidative burst in response to biotic stress in plants ...............10

1.5.3 Redox–phytohormone interactions .........................................11

1.5.4 MAPK cascades in oxidative stress response .........................12

1.6 Examples of redox signaling components ....................................13

1.6.1 Redox sensor in bacteria and yeast .......................................13

1.6.2 Redox sensitive proteins in human .......................................14

1.6.3 Redox sensors in plants .......................................................14

1.7 bZIP transcription factors in Arabidopsis ...................................15
1.7.1 Overview of the bZIP factor family......................................................15
1.7.2 Functions of the Arabidopsis G-group bZIPS .................................16
1.7.3 Well known bZIPS in other groups .................................................17

1.8 Objective ............................................................................................18

Chapter 2 Materials and Methods.............................................................19
2.1 Plant Materials and Growth Conditions..............................................19
2.2 Transformation of Arabidopsis............................................................19
2.3 Constructs for Subcellular Localization Study of bZIP68-eYFP and its point mutations and seedling treatment ..................................................20
2.4 Confocal Microscopy for subcellular localization of eYFP ..............20
2.5 Phenotypic analysis under stress treatments ......................................21
2.6 Genetic complementation of the bzip68 mutant .................................21
2.7 GUS reporter assay ...........................................................................22
2.8 RNA isolation, RNA-seq and quantitative real-time RT-PCR ..........22
2.9 Accession Numbers ..........................................................................23

Chapter 3 Results .....................................................................................27
3.1 bZIP68 is localized in nuclei under normal conditions ....................27
3.2 Nucleocytoplasmic shuttling of bZIP68 is dictated by its redox-sensitive Cys320 and requires its nuclear export signal.................................34
3.4 The bzip68 mutant showed an enhanced response to the oxidative stress treatment .........................................................................................66
3.5 The bzip68-1 mutant showed an enhanced stress response under cold

Chapter 4 Discussion ..................................................................................75
4.1 bZIP68 directly or indirectly senses perturbation of cellular redox
4.2 bZIP68 links the redox change to activation of oxidative stress defense genes through redox regulation of transcription ........................................77
4.3 bZIP68 mediates plant abiotic stress responses ........................................77
4.4 Conclusion and Working model ..............................................................78

List of References ..........................................................................................88

Curriculum Vitae ............................................................................................109