CONCLUSION REMARKS

In this thesis, the author carried out the molecular characterization of all SOD isoforms and demonstrated that the phytohormone and AOS is closely associated with the regulation of rice SOD genes (CHAPTER I). In particular, the characterization of a novel type Fe-SOD, H_2O_2-insensitive Fe-SOD, is the first report in plants (Kaminaka et al., 1999a). Furthermore, the cloning and characterization of two cytosolic AOS-scavenging enzymes, GR and MDAR, and the coordinate gene regulation of AOS-scavenging enzymes under stress conditions were shown in CHAPTER II. In CHAPTER III, the analyses of transgenic tobaccos with the overexpression of SOD or APX in chloroplasts indicated the decrease of H_2O_2 concentration is important for the enhancement of stress tolerance. These findings in this thesis will be available to elucidate the molecular mechanisms for the gene regulation of AOS-scavenging enzymes, and to produce the stress tolerant plants using another plants.

In CHAPTER I, the author have isolated and characterized cDNA (Kaminaka et al., 1997) and gene for plastidic Cu/Zn-SOD, gene DNA for Mn-SOD, and cDNA for Fe-SOD (Kaminaka et al., 1999a). The characterization of the promoter regions and the comparison of exon/intron organizations in plastidic Cu/Zn-SOD and Mn-SOD genes were effective to propose the role of these SODs and the intron insertion event at evolutional pathway, respectively. The newly characterized rice Fe-SOD cDNA was analyzed, in the terms of metal contents and the sensitivities to inhibitors, using the recombinant protein expressed in *E. coli*. Furthermore, to elucidate the regulatory mechanisms of rice SOD genes under various stress conditions, the author examined the transcripts of SOD isoforms in rice vegetative tissues, and in seedlings with the various stress treatments using the gene-specific probes to all rice SOD isoforms, and demonstrated that phytohormone and AOSs may be strongly associated with the differential regulation of rice SOD genes (Kaminaka et al., 1999b).

In CHAPTER II, the author isolated and characterized cDNA and gene for GR (section 1) (Kaminaka et al., 1998b), and two cDNAs for MDAR (section 2). The isolated rice GR cDNA was characterized to encode cytosolic isoform using the antibody against the recombinant rice GR protein. The analyses of promoter region in rice cytosolic GR gene and the expression under ABA and ABA-associated environmental stresses suggested that rice GR gene is regulated by ABA-mediated signal transduction pathway. Rice cytosolic MDAR was seemed to be consisted a small gene family in rice nuclear genome, containing two genes. Furthermore, to elucidate the regulatory mechanisms of AOS-scavenging enzyme genes under various stress conditions, the transcripts of AOS-scavenging enzymes were examined in rice seedlings with the various stress treatments using

the gene-specific probes to all AOS-scavenging enzymes, and demonstrated that several genes may be coordinately regulated by ABA-mediated signal transduction pathway under stress conditions.

In CHAPTER III, the author evaluated the transgenic tobaccos, which overexpressed SOD or APX in chloroplasts not only to elucidate the correlation between the elevated SOD or APX activity and stress tolerance but also to clarify the important factors for the enhancement of the stress tolerance. The alteration of H_2O_2 concentration by the overexpression of SOD or APX affected the endogenous H_2O_2-sensitive SOD activities. APX-overexpressing plants was more tolerant against various stresses than SOD-overexpressing plants. The author concluded that the increase of H_2O_2-detoxification capacity is very important for the enhancement of stress tolerance.

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