Environmental stresses enhance the production of reactive oxygen species (ROS) in plant cells, namely superoxide (O_2\^-) and hydroxyl radical (\cdot OH). These ROS at lower concentration participate in signaling events, but at higher concentration, they oxidize cellular constituents such as lipids, proteins and nucleic acids and can initiate chain reactions triggering cellular apoptosis. To modulate the intracellular ROS concentrations to ensure minimum damage and optimal functioning, plants have highly regulated and controlled enzymatic mechanisms, consisting of superoxide dismutase (SOD), catalase (CAT), ascorbate peroxidase (APX) and glutathione reductase (GR) to counterbalance the free radical production. The balance between activities of SOD, APX and CAT in cells are crucial for determining the steady-state level of ROS.

At the Institute of Himalayan Bioresource Technology (IHBT), full length cDNAs of manganese superoxide dismutase (Mn-SOD) from *Camellia sinensis*, thermostable copper-zinc superoxide dismutase (Cu/Zn-SOD) from *Potentilla atrosanguinea* and ascorbate peroxidase (APX) from *Rheum australe* have been isolated by earlier workers. The present thesis is focused on the functional validation of expression of the above genes alone and in combination (Cu/Zn-SOD and APX as well as Mn-SOD and APX) in the model plant *Arabidopsis thaliana* ecotype Columbia 0.

Transgenic plants for Cu/Zn-SOD, Mn-SOD and APX were confirmed for single copy insert and raised to T3. Double gene transgenic plants were developed by crossing the single gene transgenic lines. All the transgenic lines were assessed for their tolerance against various abiotic stresses (NaCl, PEG and copper), in vitro at the seedling and plantlet stage. Seed germination experiments on different levels of copper stress showed that Cu/Zn-SOD, Mn-SOD and APX transgenics exhibited higher germination percentage than the WT at various concentrations of NaCl, PEG and copper stress. The Cu/Zn-SOD transgenics retained the novel autoclavable and thermostable properties of the original enzyme in *Arabidopsis thaliana*. This result implies the use of these transgenics as bioreactors for the production of autoclavable and thermostable SOD in *A. thaliana*. Over expression of Mn-SOD and APX showed enhanced tolerance to NaCl stress concomitant with the moderate increase in the level of enzyme. All the transgenic plants showed an increase in the root length, rosette area over the WT. Transgenics for Cu/Zn-SOD having
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increased enzyme activity and providing tolerance to NaCl stress show a concomitant increase of lignification in the inter-fascicular cambium. This clearly advances our insight to the relationship of H₂O₂ generation by SOD which up regulates genes in the lignin biosynthesis pathway.

The results of 2DE and MALDI-ToF data analysis of germinating seedlings under copper salt stress indicated that Cu/Zn-SOD transgenics under stress showed a similar protein profile to that of the WT without stress. Hence, the transgenic plants over expressing Cu/Zn-SOD increases the level of tolerance to stress. Transgenics having Cu/Zn-SOD alone and in combination with APX are more tolerant to cold stress in comparison to the WT as was evident by increased enzyme activity and decreased in situ ROS production when compared to the WT plants.

The transgenics for Cu/Zn-SOD, Mn-SOD and APX as well as the double transgenics for Cu/Zn-SOD and APX and Mn-SOD and APX also showed enhanced tolerance to various concentrations of NaCl in comparison to that of the WT plants in pots. This was corroborated by the NBT assay and the relative enzyme activity assay of SOD and APX. These assays reveal a lower impact of oxidative stress on the transgenics. The net photosynthesis rate (Pₚ) as well as root and shoot biomass were significantly (p≤0.05) improved in the transgenic lines over that of the WT. This result leads to a significantly higher number of seeds/siliqua (p≤0.05) giving rise to higher seed yield (p≤0.05) by 10% in the transgenics.

In conclusion, the present study effectively achieved most of the objectives laid down for the development and assessment of the single (Cu/Zn-SOD, Mn-SOD and APX) as well as the double transgenics (Cu/Zn-SOD and APX as well as Mn-SOD and APX) in A. thaliana. Analysis of the transgenic plants that over-express these genes also provided insights into their relative contribution to abiotic stress tolerance. The study may also help in understanding the relation of these genes (SOD and APX) with one another. On the basis of these results, relevant crop improvement strategies can be evolved.