

## Title

Exploring the structure of the light-stress sensor protein PsbS and the membrane interactions of PsbS with Light Harvesting Complex II

## Authors

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## Abstract

During photosynthesis, light energy is converted to chemical energy. To prevent photodamage in high-light conditions, plants dissipate excess energy as heat in a process known as non-photochemical quenching (NPQ). PsbS senses light stress via changes in luminal pH through protonation of two glutamates (E69 and E173). Protonation induces a movement its luminal amphipathic helix H2 into the membrane phase and folding of a luminal loop fragment into a  $3_{10}$  helix (H3) [1, 2, 3]. Understanding the molecular pH-response mechanism and interactions of PsbS with light harvesting complexes (LHCII) will advance our understanding of fast NPQ mechanisms in plants. This will create opportunities for optimization of biomass production.

In this project, optical and NMR spectroscopy were used to investigate the structural responses of PsbS to changes in pH. Moreover, the interactions of PsbS with light-harvesting complex II (LHCII) were investigated in a proteoliposome system using time-resolved fluorescence spectroscopy. Wildtype spinach PsbS and its E173Q and Y75W mutants were produced in *Escherichia coli* cells and refolded in n-Dodecyl-B-D-Maltoside (DDM) detergent micelles for NMR spectroscopy. NMR spectra collected at different pH show that the bulk of the Glu residues starts protonating below pH 6.5. Between pH 7.5-7.0, however, significant protein conformational changes occur and several few Glu residues protonate. Using a Y75W mutant, pH-dependent changes of the H3 fragment were monitored via Trp fluorescence spectroscopy. Finally, the proteoliposome system shows that PsbS induces fluorescence quenching of LHCII at low pH, allowing us to explore the effects of PsbS mutations, lipid composition and xanthophyll exchange.

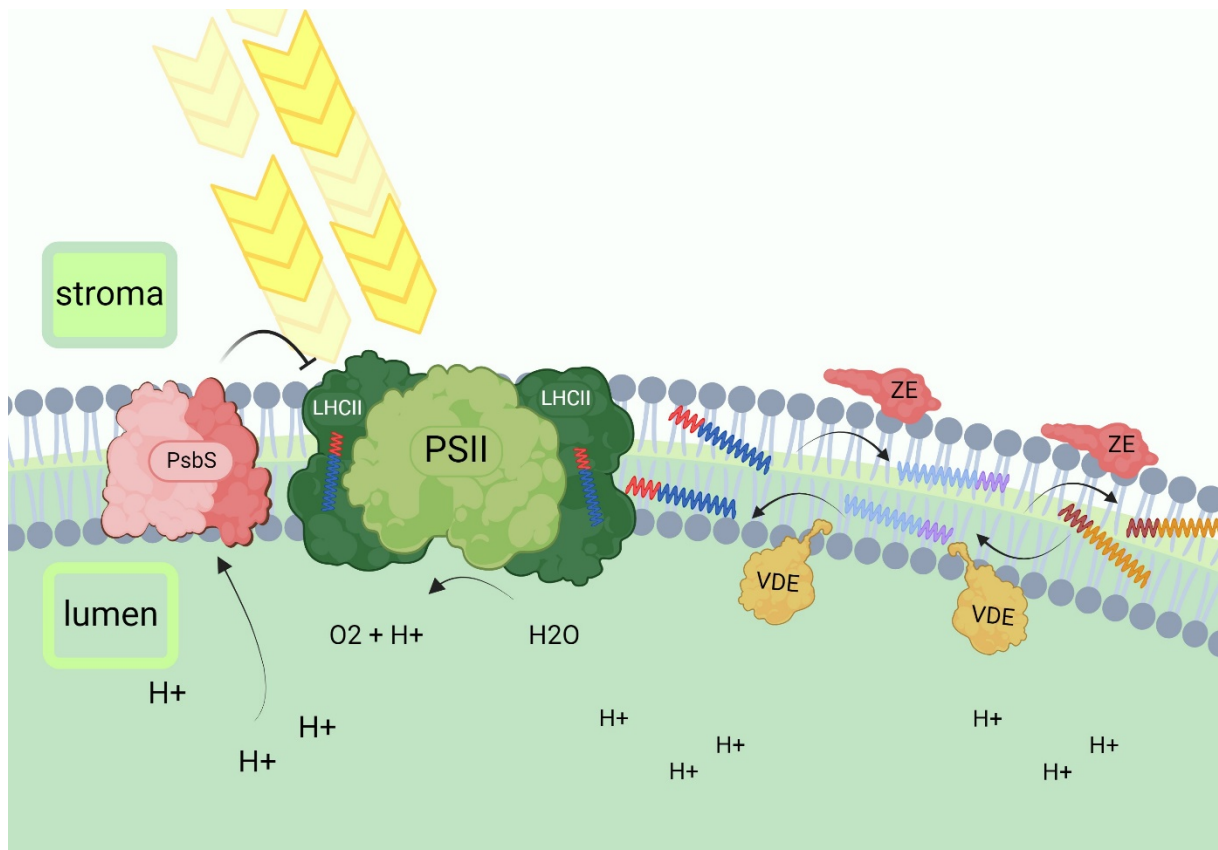


Figure 1: Schematic representation of NPQ in plant cells.

#### References (max 3)

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