

## Structural Biology Brussels

has the honor to invite you to the public defense of the PhD thesis of

**Bo WEI**

to obtain the degree of Doctor of Bio-Engineering Sciences

### Joint PhD with Universiteit Gent

Title of the PhD thesis:

*Site-specific discovery of protein S-sulfenylation in Arabidopsis thaliana: proteomics approaches*

#### Promotors:

Prof. dr. Joris Messens  
prof. dr. Frank Van Breusegem (UGent)

The defense will take place on  
**Tuesday November 24<sup>th</sup> 2020 from 15 h to 17 h**

The defense will be a ZOOM meeting:  
<https://us02web.zoom.us/j/81634510889>

#### Members of the jury:

Prof. dr. Wout Boerjan (chairperson, UGent)  
Prof. dr. Ive De Smet (secretary, UGent)  
Prof. dr. Geert Angenon  
Dr. Daria Ezerina  
Prof. dr. Lieven De Veylder (UGent)  
Dr. Jingjing Huang (UGent)  
Prof. dr. Claire Remacle (ULiège)  
Dr. Didier Vertommen (UCL)

#### Curriculum vitae

Bo Wei (Bo.We@vub.be) obtained his Master in Cell Biology from Hefei University of Technology (China) in 2016. In the same year, he got a CSC-Scholarship and started as a joined-PhD fellow with Frank Van Breusegem (UGent) and Joris Messens (VUB). His Ph.D. research focuses on the site-specific discovery of plant protein sulfenylation by means of the chemical probe (BTD) and genetically-encoded probe (YAP1C), which led to four publications in international peer-reviewed journals.

#### Abstract of the PhD research

Hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) is an important messenger molecule in diverse cellular processes. H<sub>2</sub>O<sub>2</sub> can oxidize proteinaceous cysteinyl thiols to sulfenic acid, also known as S-sulfenylation (-SOH), thereby affecting the protein conformation and functionality. Although many proteins have been identified as -SOH in plants, site-specific mapping and quantification remained largely unexplored at a comprehensive level. Therefore, I implemented two proteomic workflows to directly identify the oxidized cysteines of *Arabidopsis thaliana* cells under H<sub>2</sub>O<sub>2</sub> stress.

I implemented a chemical probe (BTD), which consists of a benzothiazine group for sulfenylated cysteine recognition with an alkyne handle for enrichment, within a quantitative chemoproteomic platform. Using this workflow, 1,537 S-sulfenylated sites were mapped on more than 1,000 proteins. Beside this chemical approach, the genetically encoded YAP1C probe also enables to trap -SOH. Here, I present a technological advancement to identify in situ sulfenylated cysteine sites by implementing an extra immunopurification step with anti-YAP1C-derived peptide (C<sub>598</sub>SEIWDR) antibody. With this approach, we identified 1,132 YAP1C-crosslinked peptides with at least two PSMs. Further, I introduced a nanobody-based enrichment specific for -SOH detection probes (dimedone, benzyl-BTD and YAP1C) to extend the exploration of sulfenylated cysteines. At last, a new method using the click-PEGylation technique was developed for -SOH visualization on immunoblot.

This PhD study contributes to the identification of redox-sensitive cysteines and delivers an unprecedented platform for the further understanding of redox signaling. All in all, I am convinced that this study will give rise to future studies on function and structure of redox-sensing proteins.