Understanding the regulation and quality control of protein *N*-glycosylation in *Chlamydomonas reinhardtii*

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The protein *N*-glycosylation pathway is one of the most important co- and post-translational modification of proteins in eukaryotes[1]. Glycoproteins are first produced in the endoplasmic reticulum (ER) where molecular actors are responsible for the proper folding and 3D conformation of proteins before their transfer into the Golgi apparatus[2]. The growing interest of using *C. reinhardtii* as an emerging cell biofactory for the industrial production of recombinant glycoproteins requires overall an in-depth understanding and analysis of protein *N*-glycosylation in this organism. Despite a relatively well-investigated *N*-glycosylation pathway in *C. reinhardtii*[3], many glyco-enzymes and lectins involved in the *N*-glycosylation pathway in the ER remain uncharacterized. This study aims to elucidate the regulatory mechanisms of *N*-glycosylation in the ER of *C. reinhardtii*, with a particular focus on protein quality control (QC). To do so, we are combining functional genetics of key molecular actors (e.g., enzymes, lectins) and multi-omics approaches (transcriptomics, proteomics, glycomics) to unravel the regulation of protein *N*-glycosylation in microalgae. Specifically, we will: (i) define the functional roles of ER molecular actors, such as ER-lectins calnexin and calreticulin, which are not yet functionally characterized in microalgae, using mutant approach strategy (ii) understand the interplay/regulation between the truncated glycan precursor ($Glc_3Man_5GlcNAc_2$), its monoglucosylated intermediates, and ER-resident lectins, which orchestrate protein folding and QC. This will be achieved through integrated approaches allowing identifying and mapping regulatory cellular pathways including the *N*-glycosylation. These results will provide insights 1- into the physiological role of protein *N*-glycosylation in microalgae and 2- into the molecular processes ensuring accurate protein folding, a critical step toward optimizing microalgae as efficient platforms for biopharmaceutical production.

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