Development of selective Cu^+-responsive protein labeling reagent towards conditional proteomics

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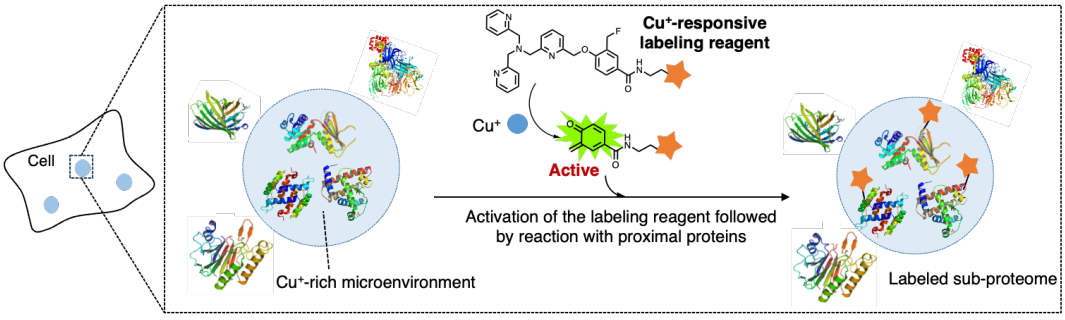
Keywords: *Cu^+-responsive protein labeling reagent*

Copper (Cu) is one of the most essential metals in all living systems, and its dynamics are precisely regulated in cells to maintain various physiological responses.[1] Cu^+-responsive probes have emerged as powerful tools in tracking the dynamic changes in Cu^+ to characterize its contribution to health and disease states.[2] However, most probes are diffusible hence the spatiotemporal resolution is limited, and they are incapable of identifying the proteins at Cu^+-rich sites.

The conditional proteomics approach has emerged as a powerful method for identifying proteins in specific condition-rich microenvironments and mapping the localization of those microenvironments. This strategy has been applied to the proteome profiling of zinc ion- and hydrogen peroxide-rich regions in live cells. [3] In this presentation, we report a Cu^+-responsive protein labeling strategy for the detection of Cu^+ with preserved spatial information and the identification of proteins at Cu^+-rich sites in living cells. Specifically, we designed quinone methide (QM) precursor-based fluorescent reagents that undergo Cu^+-dependent oxidative cleavage to release a highly electrophilic QM for subsequent labeling of proximal proteins *in situ*. The Cu^+-responsiveness of these reagents was evaluated by *in vitro* assay, and the optimal reagent showed high selectivity for Cu^+ over other biological metal ions and reactive oxygen species. The capability of this reagent in cellular contexts was demonstrated by Cu-dependent protein modification in HeLa cells overexpressing copper transporter 1.



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