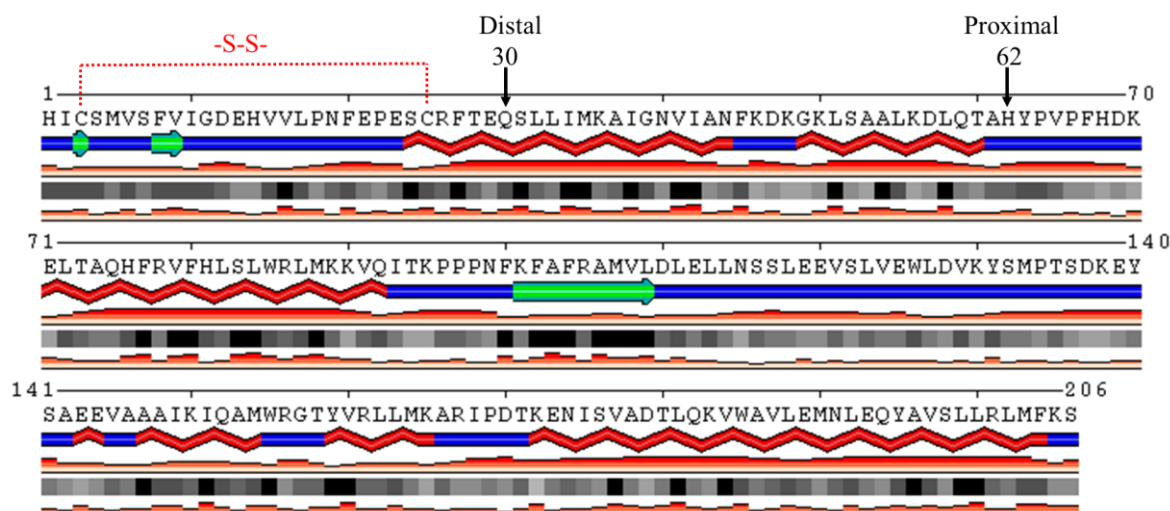
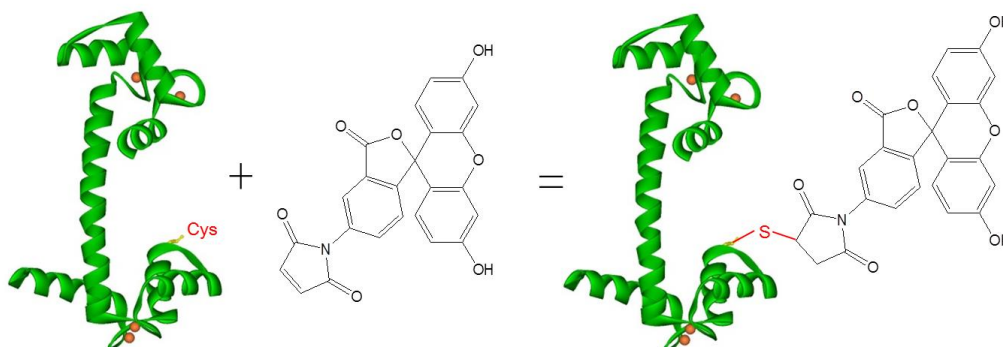


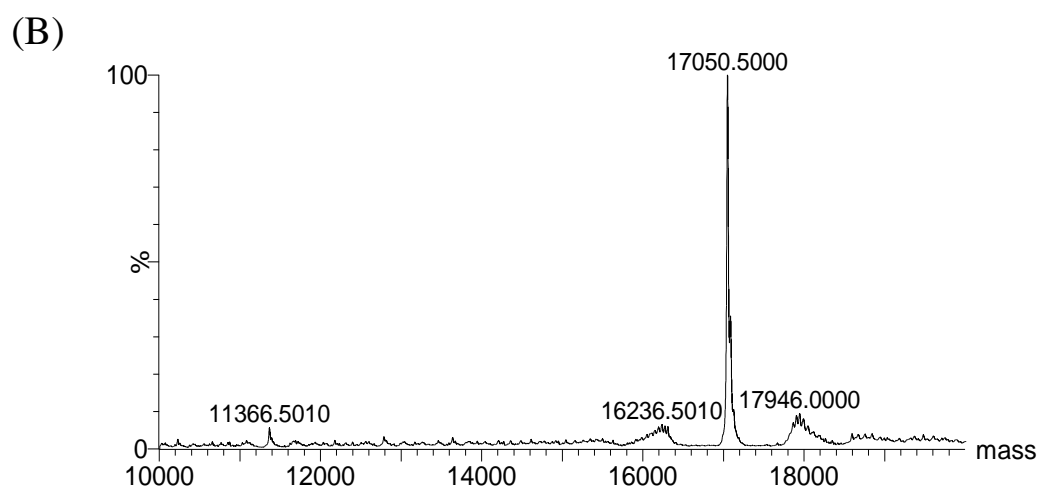
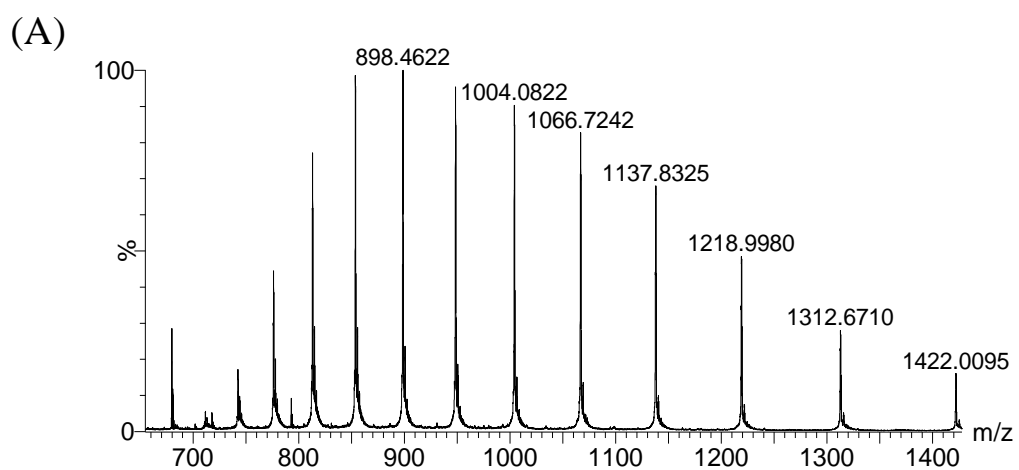
## Supporting Information



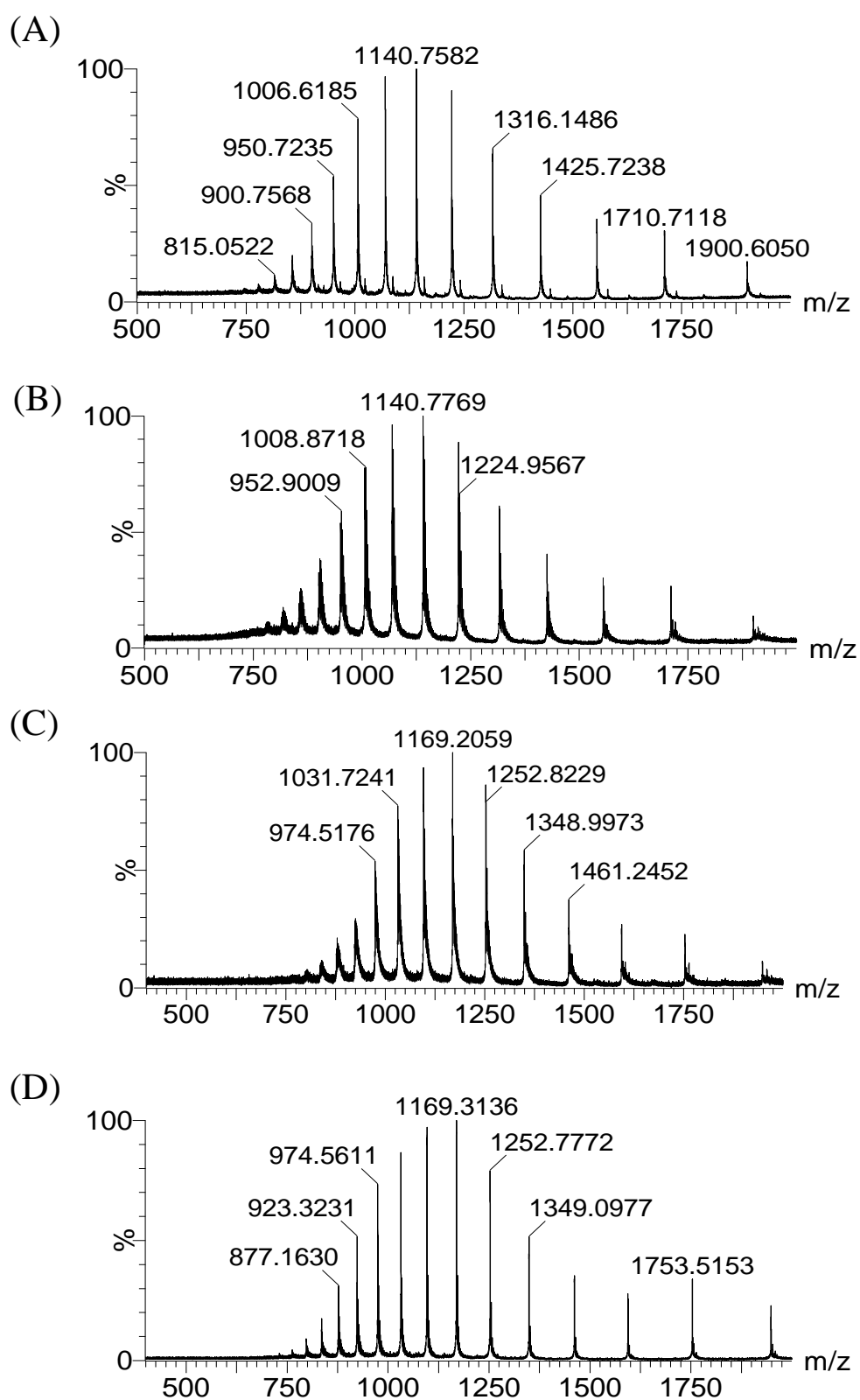
**Fig S1.** Amino acid sequence of the heme-binding domain of the C132S Adgb mutant, and the secondary structure predicted by the SABLE server (Adamczak *et al.* Proteins, 2005, 59:467-75). The possible formation of a disulfide bond (-S-S-), the distal and proximal ligands (Q30 and H62) and the mutation of C132S are highlighted.



**Fig S2.** Covalent modification of CaM by fluorescein-5-maleimide (FL) with a maleimide group to react with the Cys sulfur group.



**Fig S3.** ESI-MS spectra of CaM: (A) Original multiply-charged series. (B) The MaxEnt survey spectrum showing the major component. Calculated molecular weight of CaM 17049 Da; Observed,  $17050.5 \pm 1.5$  Da.



**Fig S4.** ESI-MS spectra of G41C CaM (A), G114C CaM (B), FL-G41C CaM (C) and FL-G114C CaM (D) showing the original multi-charge series.