Supplementary material

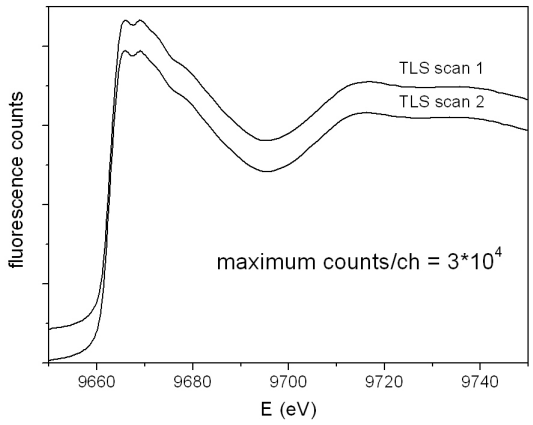

Figure 1 XANES regions of the two individual scans collected for thermolysin.

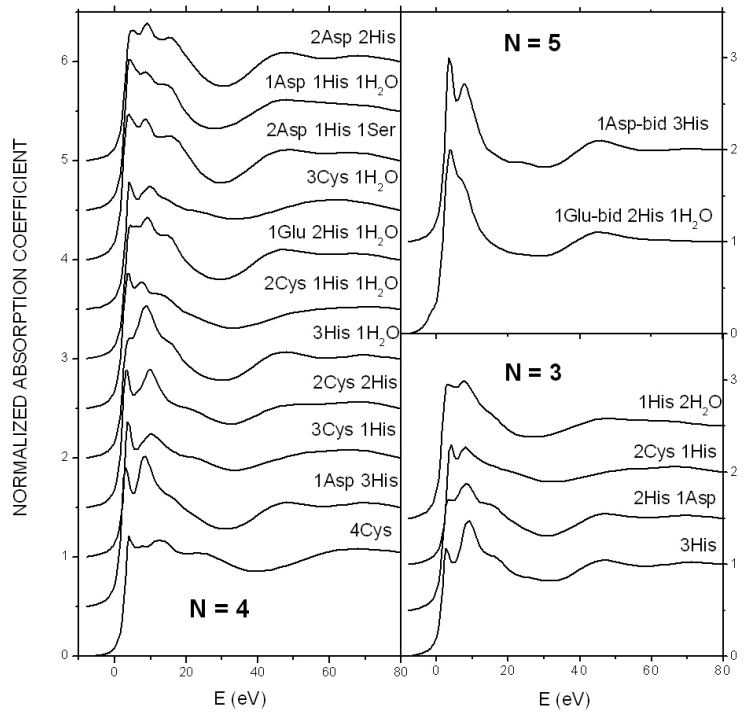

Figure 2 XANES simulations performed with the Finite Difference Method implemented in FDMNES for the 17 commonest Zn binding motives retrieved through a MDB search. The majority of the simulated spectra refers to 4-coordinated Zn sites (N=4), and their white line intensity is never higher than 1.5, as well as for 3-coordinated sites. To the contrary, when Zn is 5- or 6-coordinated (see also Figure 2 in the text), the white line intensity is much higher and in the examined cases it is never lower than 1.6. The upper limit of 1.5 when N=4 is reached by clusters where Cys residues are not involved, while Cys-containing

clusters show a much lower white line intensity. Moreover, in the presence of Cys the difference between maximum and minimum of the normalized absorption coefficient is lower than in absence of Cys and the post-edge minimum appears at higher energy values (up to a maximum of 40 eV after the edge for the 4Cys cluster). Nevertheless, these differences are not marked enough to establish quantitative criteria.

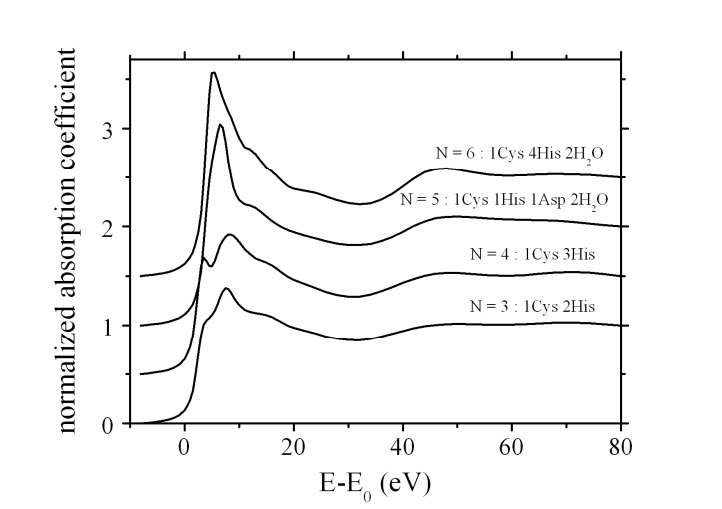

Figure 3 FDM simulations of mononuclear zinc binding sites (with coordination number ranging from 3 to 6), where 1 Cys residue is present. This set of simulation confirms that the correlation between white-line intensity and coordination number, established for the commonest Zn binding motives, holds also for the less common motives where a low number of S and a large number of N/O are present.