

Application on Biological System with Sulfur K-edge X-ray Absorption Spectroscopy

I-Jui Hsu (許益瑞)¹, Mei-I Li (李美儀)², U-Ser Jeng (鄭有舜)³,
Ling-Yun Jang (張凌雲)³, and Yu Wang (王瑜)¹

¹Department of Chemistry, National Taiwan University, Taipei, Taiwan

²National Nano Device Laboratories, Hsinchu, Taiwan

³National Synchrotron Radiation Research Center, Hsinchu, Taiwan

This report describes the possible applications of sulfur K-edge x-ray absorption spectroscopy (XAS) to biological system. Sulfur element is essential in biological system, and it often exist thiols (R_1-S-R_2) and disulfide ($R_1-S-S-R_2$) forms. There are two sulfur-containing amino acids, cysteine (Cys, $C_3O_2NH_6-SH$) and methionine (Met, $C_4O_2NH_8-S-CH_3$)¹. Cysteine can be further oxidized to the disulfide form cystine ($C_3O_2NH_6-S-S-C_3O_2NH_6$). The XAS experiments of these amino acids and the cytochrome c (Cyt-C) are carried out at BL16A. In order to resolve the differences between solid state and solution state, all spectra of standard sample, including Met, Cys, cystine and Cyt-C, are collected in both states. As shown in Figure 1, the main transition peak around 2473.5 eV in solution state spectrum is much stronger than that of solid state one, and the shoulder peak around 2474 eV is more obvious than that of solutions. However, the main features of this region are similar in both compounds. Based on the report by Szilagyi et al., the peaks in this region are assigned to the transition $1s \rightarrow \sigma^*(S-C/H \text{ bond})$. In Figure 2, there are two distinguishable peaks in cystine, which have been assigned as $1s \rightarrow \sigma^*(S-S \text{ bond})$ and $1s \rightarrow \sigma^*(S-C \text{ bond})$, for the lower and higher energy peaks⁴, respectively.

Obviously, we can tell the disulfide bonding character based on the feature of two clearly resolved peaks. According to the spectra of standard compounds, there is no disulfide bonding character in the solution or powder state of Cyt-C protein. These results are consistent with the X-ray single crystal and NMR solution state structures.

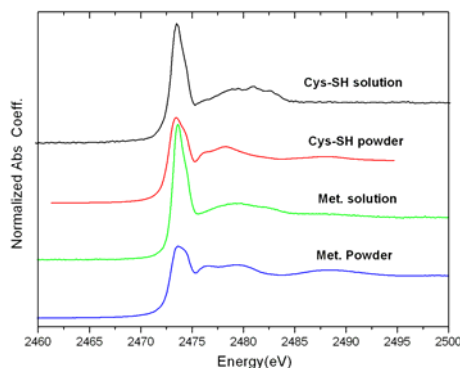


Figure 1

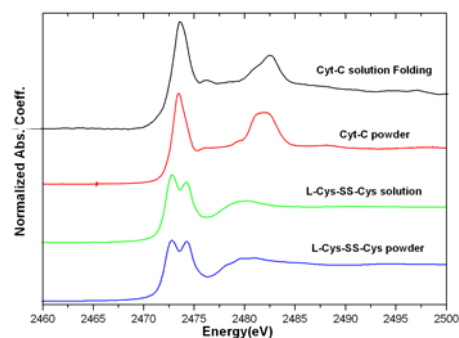


Figure 2

Another selected XAS results of one metallothionein protein with rich of coppers and cysteines are depicted in the Figure 3. Based on the previous discussion, there exist disulfide bonds in the solution state of this protein. Moreover, the pre-edge peak³ due to the bonding between sulfur and Cu(II) are displayed around 2465 eV, but quickly disappeared within 20 minutes. The detailed reasons of such variation are still under investigation.

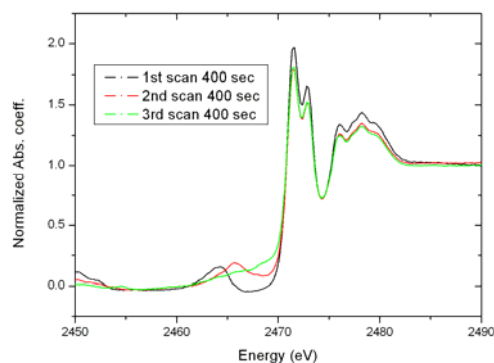


Figure 3

Reference

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