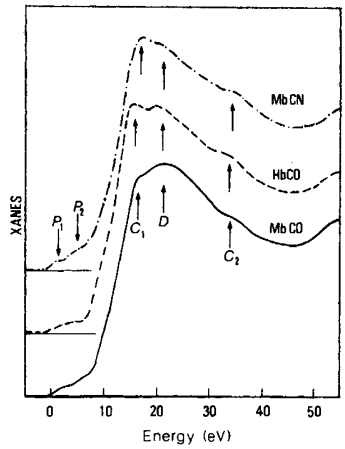

Fig. 3 Experimental XANES spectra of MbCO, HbCO and MbCN in solution. The main difference is the variation of the intensity ratio between peak C_1 and peak D which depends on the bonding angle Fe-C-O(N).

of 30 atoms have been deduced from diffraction data[5] and polarized extended X-ray absorption fine structure (Fe-N_P = 2.0 Å, F-N_ε = 2.09 Å Fe-C = 1.84 Å) (S. Hasnain, A.B. and S.P., unpublished), and an idealized symmetrical and plane porphyrin ring was used. Figure 2*a* shows that the theory predicts all the experimental features *A*, *B*, *D*, *E*, *F* due to the scattering in the haem plane, while Fig. 2*b*-*d* shows the large variation of the $\vec{E}//\hat{n}$ spectra obtained by changing the CO bonding geometry. The best agreement between the theoretical spectrum and the experimental results was found for the Fe-C-O bent configuration at $\theta = 150°$. Using the crystallographic coordinates, which have large errors[5], with Fe-C at 1.78 Å and C-O parallel but displaced from the haem normal axis, we obtain a large disagreement with the XANES experiment.

Finally, having demonstrated the XANES method for a single crystal, we next applied it to determine the bonding geometry of ligands in haem proteins in solution, where diffraction methods cannot be applied. Figure 3 shows the Fe XANES spectra of MbCO, HbCO and MbCN in solution with good signal-to-noise ratio. The main difference between these spectra is the variation of the intensity ratio between peaks *C* and *D*, which depends strongly on the Fe-C-O(N) bonding angle. Both by calculation and by comparison with crystal XANES spectra, we find the Fe-C-O angle in solution to be ~150°, as it is in MbCO crystals.

Theoretical XANES analyses indicate that the Fe-C-O(N) angle is 150° in MbCO, 165° in HbCO and 180° in MbCN in solution. Our results strongly support the Moffat hypothesis[12] that the CN configuration in HbCN is linear and tilted rather than bent. We find, within the sensitivity of the XANES method, that the Fe-C-O configuration is bent in both crystals and solutions, and we estimate that we can easily detect variations of $\Delta\theta = 10°$. We have obtained experimental evidence that the (Fe-C = 0) configuration is different in HbCO and MbCO in solution ($\Delta\theta = +15°$ going from MbCO to HbCO) and therefore that the CO bonding is modified by the protein. Crystal diffraction studies have shown a different bent configuration, with $\theta = 136°$ in HbCO (ref. 8). Our data therefore indicate that there is a difference in CO bonding in HbCO in the crystalline and solution phases.

In conclusion, our results show that there is a large dichroism in the X-ray range of the haem protein crystal which can be fully analysed and its bond angles determined quantitatively by XANES. This method can be used to improve our knowledge of the subtle variations that occur in the local structure of proteins and can thus help us to establish the relationship between localized structural changes and the mechanism of protein control of ligand binding in solution close to the protein native state.

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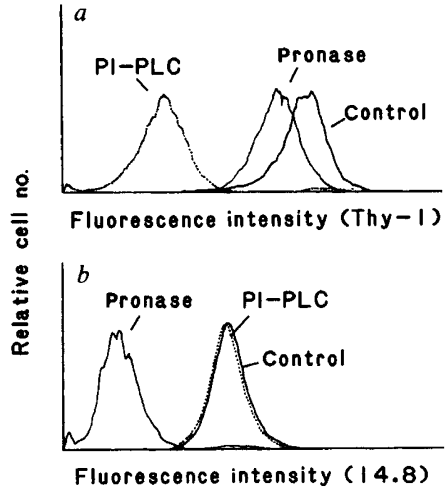
Errata

Phosphatidylinositol is the membrane-anchoring domain of the Thy-1 glycoprotein

M. G. Low & P. W. Kincade

Nature **318**, 62-64 (1985)

IN Fig. 1*a* and *b*, two lines representing the incubation with phosphatidylinositol-specific phospholipase C (PI-PLC) were not properly reproduced. The correct figure is shown here:



More help required on T and B cells

J. Shifflett

Nature **316**, 490 (1985)

THE word 'antigen' was omitted from the second sentence in paragraph 2, which should read: 'B lymphocytes might find it difficult to pinocytose a surface antigen of any pathogen unwilling to facilitate its own destruction . . .' In addition, the second sentence in the third paragraph is made clearer with the words 'of materials' deleted.

Shanghai Institute of Biochemistry: The molecular biology revolution

A. Anderson

Nature **318**, 217-218 (1985)

IN this item in the 'Science in China' feature, the deputy-director of the Shanghai Institute of Biochemistry was inadvertently referred to as director. The director is Professor Lin Qi-shui, and his deputy is Professor Zhang You-shang.