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## **EPR of Co(ii) as a structural and mechanistic probe of metalloprotein active sites: characterisation of an aminopeptidase** Brian Bennett

Co(II) can often be substituted for Zn(II) in zinc-dependent metalloenzymes to provide spectroscopically accessible forms of the enzymes. Co(II) is an excellent spectroscopic probe as it is both optically active and EPR active. Further, its fast relaxation properties make it a useful paramagnetic shift reagent in NMR. In EPR, the dependence of the spectra of high-spin Co(II) on E/D and the sensitivity of the resolvability of the 59Co hyperfine structure to strain terms allow structural information to be inferred from the EPR spectra. In addition to its useful spectroscopic properties, Co(II) is often an extremely good functional mimic of Zn(II), and Co(II)-substituted zinc-dependent enzymes often display catalytic activities analogous to the native Zn(II)-containing enzyme forms. It is therefore somewhat surprising that there are few examples of EPR studies of Co(II)-substituted enzymes. The most detailed studies carried out to date are those on the aminopeptidase from *Aeromonas proteolytica*. Thesrefore, the methodology of extracting structural information from EPR of Co(II)-containing proteins is described using studies on *A. proteolytica* as an example.