

Table 2. Assigned hyperfine shifted resonances in the ^1H NMR spectra of Cu(II) AZAMI-F, AZAMI, AZ, and AMI

Peak	Assignment*	δ , ppm				
		δ_{obs} Cu(II) AZAMI-F ^{†‡}	δ_{dia} Cu(I) AZAMI-F [§]	δ_{obs} Cu(II) AZAMI [†]	δ_{obs} Cu(II) AZ [†]	δ_{obs} Cu(II) AMI [¶]
a	His-46 C $^{\delta 2}$ H	≈ 45 (≈ 43)	7.75/6.15	≈ 46	≈ 52	43
b	His-115 C $^{\delta 2}$ H					50
c/d	His-46/115 C $^{\epsilon 1}$ H	≈ 32 (≈ 31)	7.09	≈ 30	≈ 30	≈ 35
e	His-46 N $^{\epsilon 2}$ H	32.3**		30.8**	26.9 ^{††}	27.5
f	Met-117 C $^{\gamma 2}$ H	≈ 21 (≈ 19.5)	3.35			12/11.1
g	Asn-47 C $^{\alpha}$ H	17.8 (16.7)	4.81	17.5	19.0	14.1
h	Met-117 C $^{\gamma 1}$ H	11.6 (9.5)		≈ 15		12/11.1
i	His-46 C $^{\beta 1}$ H	-3 (-2.5)		-3	-2	-2.5
j	Cys-112 C $^{\alpha}$ H	-9 (-8)		≈ -10	-7.8	-9.5

*In AZAMI, Met-118 is found in place of Met-117. In AZ, His-117 and Met-121 are found in place of His-115 and Met-117. In AMI, the residues for which hyperfine shifted signals are observed are His-54, Asn-55, Cys-93, His-96, and Met-99.

[†]Data recorded at 25°C in 10 mM phosphate (99.9% D₂O), pH* 8. The observed shifts (δ_{obs}) arise from the three contributing factors δ_{dia} , δ_{pc} , and δ_{c} { δ_{dia} is the shift in an analogous diamagnetic system [for example the Cu(I) protein], δ_{pc} is the pseudocontact (through space) contribution, and δ_{c} is the Fermi contact (through bond) contribution}. The δ_{pc} values are not so significant, because the small anisotropy of the g tensor, and, therefore, δ_{obs} minus δ_{dia} for a particular proton provides a good estimate of δ_{c} , which is a measure of the spin density.

[‡]The δ_{obs} values in parenthesis are those obtained at 40°C, at which temperature the saturation transfer experiments were performed.

[§]Positions identified from saturation transfer difference experiments on 1:1 mixtures of Cu(II) and Cu(I) AZAMI-F at 40°C (see Fig. 9).

[¶]For *Paracoccus versutus* AMI in 50 mM phosphate at pH 7.0 and 32°C.

^{||}The second His C $^{\epsilon 1}$ H resonance is found at 46.7 ppm at 800 MHz (5°C) in 10 mM phosphate, pH 8.0.

**Observed in 10 mM phosphate (90% H₂O/10% D₂O) at pH 8.0 (25°C).

^{††}Recorded at 800 MHz (5°C) in 10 mM phosphate (90% H₂O/10% D₂O) at pH 8.0.