Pergamon

Tetrahedron: *Asymmetry* 10 (1999) 281–295

TETRAHEDRON: *ASYMMETRY*

Functional mimics of copper enzymes. Synthesis and stereochemical properties of the copper(II) complexes of a trinucleating ligand derived from L-histidine

Laura Santagostini,[a] Michele Gullotti,[a] Roberto Pagliarin,[b,*] Emanuela Bianchi,[a] Luigi Casella [c] and Enrico Monzani [c]

[a]*Dipartimento di Chimica Inorganica, Metallorganica e Analitica, Università di Milano, Centro CNR, Via Venezian 21, 20133 Milano, Italy*

[b]*Dipartimento di Chimica Organica e Industriale, Università di Milano, Via Venezian 21, 20133 Milano, Italy*

[c]*Dipartimento di Chimica Generale, Università di Pavia, Via Taramelli 12, 27100 Pavia, Italy*

Received 12 November 1998; accepted 17 December 1998

Abstract

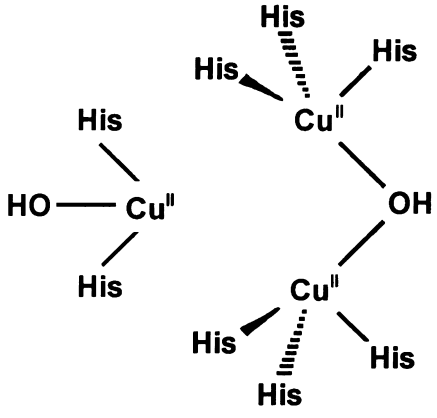
The ligand piperazine-1,4-bis[4-(*N*-(1-acetoxy-3-(1-methyl-1*H*-imidazol-4-yl))-2-propyl)-*N*-(1-methyl-1*H*-imidazol-2-ylmethyl)aminobutyl] (PHI) was synthesized by a multistep procedure starting from N^{τ}-methyl-L-histidine, piperazine-1,4-bis[4-(4-oxo-4-butanoic) acid] and 1-methyl-1*H*-imidazole-2-carbaldehyde. This ligand has two potential tridentate, aminobis(imidazole) (A sites), and one bidentate, piperazine (B site), binding sites for metal ions and was employed for the synthesis of the binuclear $[Cu_2PHI]^{4+}$ and the trinuclear $[Cu_3PHI]^{6+}$ complexes, the latter of which features a coordination environment mimicking that present in the trinuclear clusters of the blue copper oxidases. For comparison purposes, the mononucleating ligand L-N^{α}-(1-methyl-1*H*-imidazol-2-ylmethyl)-N^{τ}-methylhistidine methyl ester (IH) and its complex $[CuIH]^{2+}$ have been also prepared. These copper(II) model complexes are the first reported which are directly derived from chiral L-histidine residues. A detailed analysis of the UV–vis, CD and EPR spectra of the complexes has established that the Cu(II) centers bound to PHI A sites are square-pyramidal in solution, with the amino and one imidazole donor in the equatorial plane and the additional imidazole group bound axially. This arrangement implies the adoption of an unusual conformation of λ chirality by the L-histidine residue and is determined by the attempts to minimize steric interference between the substituents at the tertiary amine donor group and the histidine residue bearing the C-α substituent acetoxymethylene group of the bound PHI ligand. For the less sterically crowded secondary amine group of the bound IH ligand, the histidine C-α substituent can occupy a pseudoaxial position, so that in the complex $[CuIH]^{2+}$ the 'normal' arrangement with three equatorial nitrogen donors and δ chirality in the L-histidine chelate ring occurs.

* Corresponding author. Tel: +39-02-2363469; fax: +39-02-2364369; e-mail: proibe@icil64.cilea.it

0957-4166/99/$ - see front matter
PII: S0957-4166(98)00505-9

1. Introduction

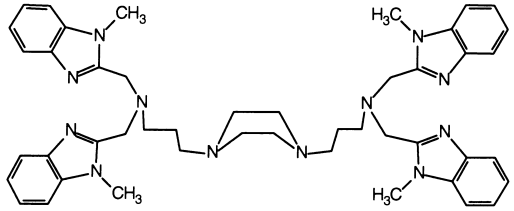
The multicopper 'blue' oxidases are a class of enzymes which couple the four-electron reduction of dioxygen to water with one-electron oxidation of a variety of substrates.[1] The best characterized members of this class of enzymes are laccase,[2] ascorbate oxidase,[3] and ceruloplasmin.[4] A combination of detailed spectroscopic studies[1] and the X-ray structural determination of ascorbate oxidase[5] and ceruloplasmin[6] have shown that the minimal functional unit of these enzymes contains one type 1 copper and a trinuclear cluster containing one type 2 and a couple of type 3 coppers, according to the classification by Malkin and Malmström.[7] Ceruloplasmin possesses additional copper centers, the function of which is unclear. Trinuclear copper clusters also appear to be involved in the membrane-bound form of methane monooxygenase[8] and, possibly, other copper monooxygenases.[1] Both the X-ray data and amino acid sequences of the multicopper oxidases show that the protein ligands of the trinuclear copper cluster are eight histidines, distributed as shown schematically in Scheme 1.[1a,5a,b,6] A terminal hydroxide group is bound to type 2 copper, and a bridging hydroxide or oxo group binds the two type 3 coppers in the oxidized form of the enzymes. The Cu···Cu distances are in the range 3.6–3.9 Å. However, upon reduction, the bridge between the type 3 coppers is broken and the Cu···Cu distances rise to 4–5 Å.[5c–e]



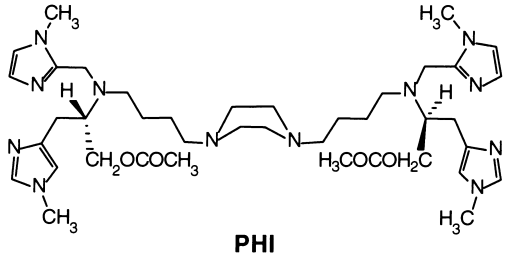
Scheme 1. Schematic representation of the trinuclear copper cluster of blue oxidases

In spite of substantial protein work, there is a need for synthetic studies aimed at clarifying the complex structures and the poorly understood chemistry exhibited by the trinuclear clusters of the blue oxidases. So far, studies on synthetic complexes containing trinuclear cores with nitrogen donor ligands have been scarce.[9] We have recently reported trinuclear and binuclear copper complexes of the ligand L, containing a piperazine residue modified with two aminobis(benzimidazole) arms,[10] which represent our first approach to the development of model complexes for the trinuclear copper clusters of the blue oxidases. The ligand L can, in fact, simulate the distribution of protein ligands in the cluster (Scheme 1) by binding one copper center at the piperazine site and two additional copper centers at the aminobis(benzimidazole) sites. In this paper we extend our investigation by reporting the synthesis and stereochemistry of the binuclear and trinuclear copper(II) complexes derived from the chiral ligand PHI, where two aminobis(imidazole) units, obtained by modification of L-histidine residues, are attached to the central piperazine nucleus. Clearly, the introduction of histidine residues in the ligand increases the significance of these compounds as synthetic models for the biological copper clusters. Although a few dicopper complexes with polyimidazole ligands have been reported,[11] the present biomimetic systems are the first ones directly derived from L-histidine. For assessing the stereochemical characteristics of

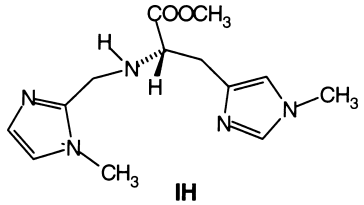
the new complexes we deemed it necessary to investigate the behaviour of the mononuclear analogue copper(II) complex derived from the ligand IH, which was also obtained from L-histidine.



L



PHI



IH

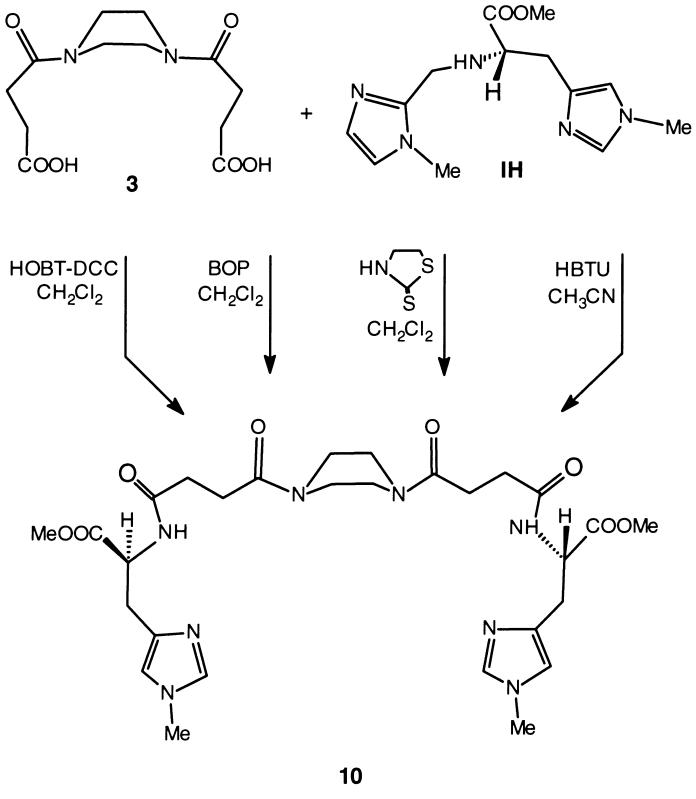
2. Results and discussion

2.1. Ligand design and synthesis

Mimicking the trinuclear cluster of the blue copper oxidases requires the design of polydentate ligands containing eight nitrogen donors, possibly imidazoles, with sufficient flexibility to allow geometric rearrangements required by metal redox chemistry and exogenous ligand binding to the metal centers. We have previously shown that ligand L gives access to binuclear and trinuclear copper complexes and, therefore, decided to proceed towards systems of more significance in the context of protein models by replacing the benzimidazole residues with biologically more relevant imidazole residues. Since we attribute special importance to the stereochemical properties of the models, as they are very typical features of enzyme active sites, we decided to introduce imidazole groups in the new ligand by using the histidine residue. This poses a synthetic challenge, because the imidazole groups of the ligand should preferably be *N*-methylated. This protection is essential for increasing the chemical stability of the resulting complexes when their oxidative reactions have to be studied.

The synthesis of ligand PHI was at first conceived through the reaction between the piperazine derivative **3** and the L-N^{α}-(1-methyl-1*H*-imidazol-2-ylmethyl)-N^{τ}-methylhistidine

methyl ester **1** (IH), as reported in Scheme 2. Several methods have been used for the activation of carboxy groups either using 1-hydroxybenzotriazole (HOBT),[12] benzotriazolyl-*N*-oxy-tris(dimethylamino)phosphoniumhexafluorophosphate (BOP),[13] and 1,3-thiazolidine-2-thione[14] in dichloromethane or *O*-(1*H*-benzotriazol-1-yl)-*N*,*N*,*N*′,*N*′-tetramethyluronium hexaflurophosphate (HBTU),[15] in acetonitrile. In all these cases, the analysis of the reactions showed several by-products and the presence of reagents even after a long reaction time.

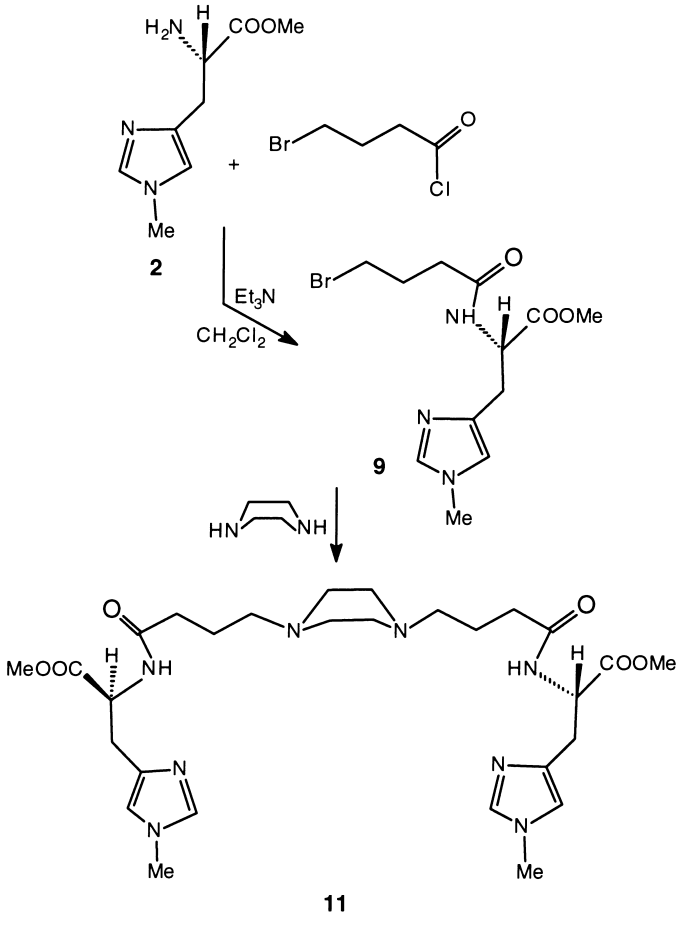


Scheme 2. Various routes to the ligand PHI precursor **10**

Following another approach, L-N^{τ}-methylhistidine methyl ester **2** was allowed to react with 4-bromobutanoylchloride in dichloromethane in the presence of triethylamine as base (Scheme 3), to produce compound **9** which, after purification, was expected to give compound **11** upon reaction with piperazine, potassium carbonate and acetonitrile as solvent.

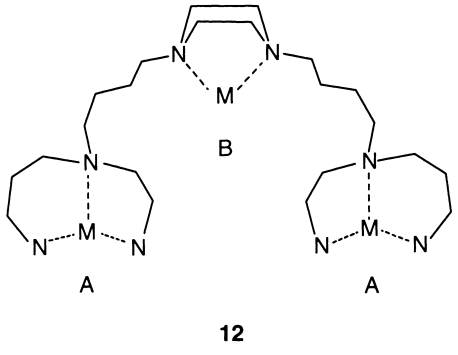
Also in this case the reaction sequence occurred in poor yields and significant amounts of by-products were present. At the end, the reactions reported in Scheme 4 proved to be a valid method for the preparation of compound PHI. Condensation of two molecules of succinic anhydride with piperazine afforded the diacid **3**,[16] which reacted with N^{τ}-methyl histidine methyl ester **2**[17] to give the diamide **4**. Reduction of the carbonyl groups of the diamide **4** with $BH_3 \cdot Me_2S$ led to compound **5**, which was reductively alkylated with 1-methyl-1*H*-imidazole-2-carbaldehyde **8**,[18] and $NaBH(OAc)_3$ to afford **6** and then *O*-acetylated to give the ligand PHI, **7**.

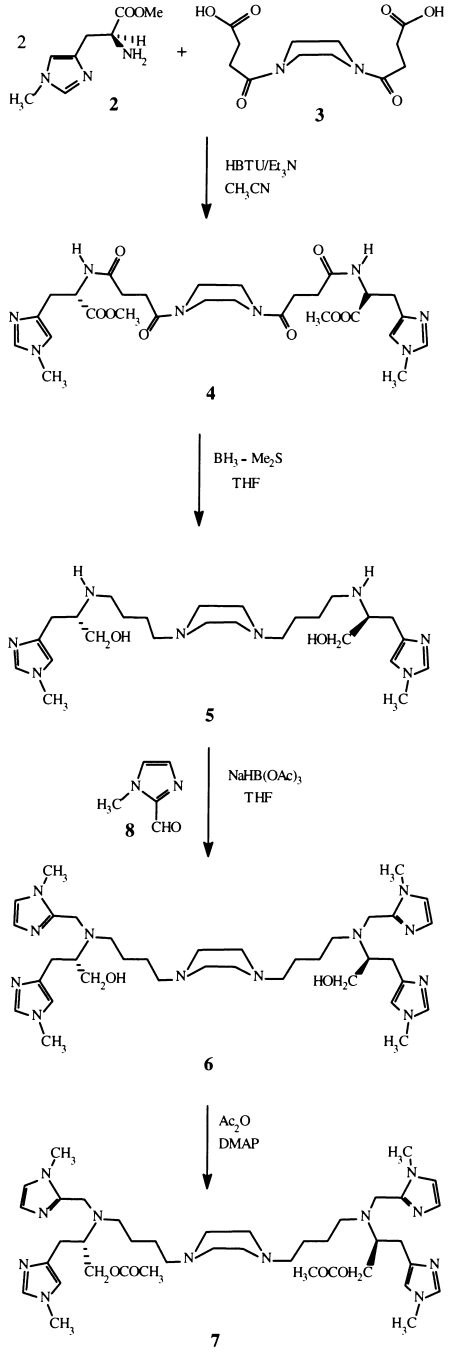
Besides replacement of benzimidazole groups with imidazole groups, there are additional differences between L and PHI which have been introduced through the synthesis and that may have structural consequences on complex formation. First, the piperazine and tertiary amine nitrogens are separated by four carbon atoms instead of three. The lengthening of this carbon chain was suggested by the fact that we



Scheme 3. Synthetic route to the ligand PHI precursor **11**

previously encountered some difficulty in introducing a third copper center in L, after binding of the first two,[10] due to the fact that the piperazine nitrogens are involved in the binding in the binuclear complex. Upon lengthening the carbon chain we expect to weaken or even eliminate these interactions, leaving the piperazine residue available for facile binding of a third metal ion. As a second difference between PHI and L, any metal ion bound at the aminobis(imidazole) sites of PHI will be chelated with one five-membered and one six-membered ring, whereas binding at the corresponding aminobis(benzimidazole) sites of L occurs with formation of two five-membered rings. Assuming that metal binding to PHI occurs with the same distribution of nitrogen donors as for L,[10] we can schematically represent the metal binding sites of PHI according to structure **12**.





Scheme 4. Synthetic scheme for the preparation of ligand PHI

The synthesis of the ligand IH, from L-N^{τ}-methylhistidine methyl ester and 1-methyl-1*H*-imidazole-2-carbaldehyde, was achieved by reductive condensation in the presence of palladium–charcoal. Surprisingly, the methyl ester was found largely hydrolyzed at the end of this reaction and, therefore, the acid group had to be re-esterified. Upon coordination of IH, the ligand environment around the metal center closely represents the situation at sites A of **12**, i.e. two chelate rings of five and six atoms are formed.

2.2. Characterization of copper(II) complexes

Both the binuclear complex $[Cu_2PHI][ClO_4]_4$ and the trinuclear complex $[Cu_3PHI][ClO_4]_6$ were easily obtained by reacting solutions of the ligand with corresponding amounts of copper(II) perchlorate hexahydrate. The UV–vis, CD and EPR characterization data for these complexes and for $[CuIH]^{2+}$ are summarized in Table 1. Rather similar spectra were obtained in acetonitrile or methanol solutions. The near-UV region of the optical spectra of the copper(II) complexes shows a well defined, but somewhat asymmetric band of moderate intensity, with maximum in the range 270–290 nm and a tail extending up to about 400 nm. This band has three corresponding CD features, with a prominent peak near the wavelength of the UV maximum and one shoulder at both sides of this peak, near 240 nm and between 350 and 400 nm. Interestingly, the CD peak is negative for $[CuIH]^{2+}$ but positive for $[Cu_2PHI]^{4+}$ and $[Cu_3PHI]^{6+}$. Since the ligands exhibit much weaker absorption and negligible CD activity in the range 250–400 nm, the origin of the electronic absorptions above 250 nm must be mostly of charge transfer type. The more intense absorption bands occurring at higher energy are largely due to intraligand transitions and, therefore, will not be analyzed further. The visible part of the electronic spectra of the copper(II) complexes contains the broad envelope of the *d–d* transitions. The maximum occurs at 650 nm for $[CuIH]^{2+}$ and at 620 nm for $[Cu_2PHI]^{4+}$ and $[Cu_3PHI]^{6+}$, but in the latter cases a prominent shoulder near 750 nm is clearly evident. At least two components are resolved in the CD spectra, near 600 nm and at or above 700 nm, but again while the CD activity is essentially positive for $[CuIH]^{2+}$, components of negative sign dominate the CD spectra of $[Cu_2PHI]^{4+}$ and $[Cu_3PHI]^{6+}$. The rather close similarity of the UV–vis and CD spectra of the latter two complexes throughout the spectral range indicates that, as in the trinuclear copper(II) complex derived from L,[10] the Cu(II) ion bound at the piperazine B site (structure **12**) gives little contribution to these spectra.

The better resolution of the CD spectra is helpful in the assignment of the transitions. The moderate absorptions and the CD activity in the 300 nm range comprize LMCT transitions of type π(imidazole) → Cu(II). In PHI complexes there are four contributions of this type. The assignment is based on the presence of similar UV and CD features in the spectra of copper(II)–imidazole[19] and copper(II)–L-histidine[20] complexes, respectively. The other LMCT components originating from imidazole donors, σ(imidazole) → Cu(II), which are electric dipole allowed, are much more intense and occur together with the intraligand absorptions in the 220 nm region.[19,20] At intermediate energy, near 250 nm, LMCT transitions from amino donor groups to Cu(II), of type σ(amino) → Cu(II) are located.[21] For instance, CD features in this range are typically observed in the spectra of copper(II)–L-N^{α},N^{α}-dimethylhistidine complexes.[20b] Furthermore, from the position of the *d–d* bands[22] and the EPR spectra to be discussed below, the Cu(II) centers of $[CuIH]^{2+}$, $[Cu_2PHI]^{4+}$ and those at sites A in $[Cu_3PHI]^{6+}$ are clearly in a tetragonal symmetry. The Cu(II) at site B in $[Cu_3PHI]^{6+}$ is also expected to be tetragonal, as discussed for $[Cu_3L]^{6+}$ and the complex Cu(II)–piperazine reported previously.[10] There are, however, significant differences between the Cu(II) center of $[CuIH]^{2+}$ and those bound to A sites in PHI complexes. The *d–d* band envelope of $[CuIH]^{2+}$ is more symmetric and has no resolved component at low energy. This indicates an essentially square planar Cu(II) chromophore, with only weak axial interaction, presumably by solvent molecules, and an energy order of the Cu(II) *d* orbitals: $d_{x^2-y^2} > d_{xy} > d_{z^2} > d_{xz}, d_{yz}$. Therefore, the following assignment of the visible CD bands can be made for $[CuIH]^{2+}$: the positive band at 670 nm to the transition $d_{xy} \rightarrow d_{x^2-y^2}$, the positive shoulder at 610 nm to $d_{z^2} \rightarrow d_{x^2-y^2}$, and the negative band at 540 nm to $d_{xz}, d_{yz} \rightarrow d_{x^2-y^2}$ (note that partial cancellation of adjacent CD bands with opposite sign can occur). In the case of Cu(II) complexes derived from PHI (A sites) a low-energy component of the *d–d* band (~750 nm) is observed in both the optical and CD spectra. This indicates that the structure of these Cu(II) centers is square-pyramidal,[22] with strong axial interaction by a donor ligand.

Table 1
UV–vis, CD and EPR[a] spectral data for copper(II) complexes in acetonitrile solutions

Complex	UV/Vis λ_{max}, nm (ε, M^{-1} cm^{-1})	CD λ_{max}, nm ($\Delta\varepsilon$, M^{-1} cm^{-1})	EPR		
			$g_{\parallel}$	$g_{\perp}$	$\lvert A_{\parallel}\rvert \times 10^4$ cm^{-1}
$[CuIH]^{2+}$	220 sh (8200)	270 (– 0.85)	2.231	2.085	173
	270 (2500)	320 (+ 0.50)			
	370 sh (250)	540 (– 0.05)			
		610 sh (+ 0.20)			
	630 (55)	670 (+ 0.32)			
$[Cu_2PHI]^{4+}$	225 sh (17500)	240 sh (+ 0.70)	2.252	2.086	143
	288 (5300)	285 (+ 1.65)			
		380 sh (+ 0.16)			
	620 (210)	600 (– 0.26)			
	750 sh (80)	700[b] (+ 0.10)			
$[Cu_3PHI]^{6+}$	225 sh (25000)	240 sh (+ 0.40)	2.256	2.088	143
	290 (5200)	290 (+ 1.71)			
	620 (220)	380 sh (+ 0.20)			
	750 sh (120)	605 (– 0.36)			
		700[b] (+ 0.09)			

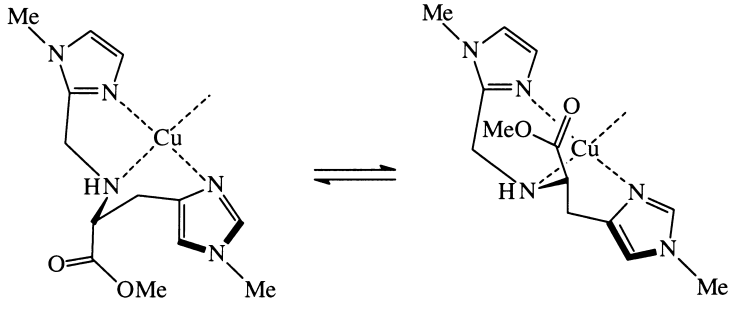
[a] Recorded in frozen solution at – 150 °C.

[b] The CD extremum is at longer wavelength, outside the range of the CD instrument.

This interaction raises the energy of the Cu(II) d_{z^2} orbital, leading to the following energy ordering: $d_{x^2-y^2} > d_{z^2} > d_{xy} > d_{xz}, d_{yz}$. Therefore, assignment of the positive CD activity above 700 nm to the transition $d_{z^2} \rightarrow d_{x^2-y^2}$ and the dominant CD activity of negative sign near 600 nm to unresolved contributions from the $d_{xy} \rightarrow d_{x^2-y^2}$ and $d_{xz}, d_{yz} \rightarrow d_{x^2-y^2}$ transitions can be made. The analysis of the EPR spectra of the complexes support the above interpretation.

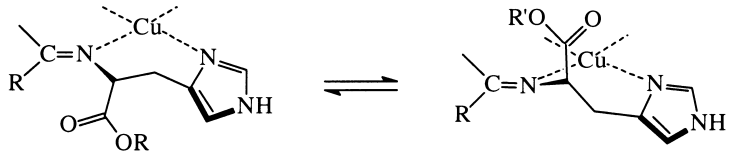
Further insight into the stereochemistry of the copper(II) complexes can be obtained from examination of the CD patterns exhibited by their CD spectra. As noted above, the dominant Cotton effects within the *d–d* bands are positive for $[CuIH]^{2+}$ but negative for $[Cu_2PHI]^{4+}$ and $[Cu_3PHI]^{6+}$, and also the prominent CD feature in the near-UV is opposite for IH and PHI complexes. The behaviour of the two PHI complexes is surprising and completely unusual, since L-histidine residues bound to Cu(II) through their amino and imidazole groups systematically give rise to opposite CD behaviour.[20,23] In fact, generally, and as confirmed here for $[CuIH]^{2+}$, CD activity of positive sign within the *d–d* envelope implies stabilization of the L-histidine chelate ring with δ conformation chirality, containing the C-α substituent oriented pseudoaxially with respect to the ring (structure **14**). This stabilization is quite evident for the Cu(II) complexes of histidine-imines,[23,24] where the (histamine-like) histidine chelate ring is fused to an imine chelate ring in the Cu(II) coordination plane. In these cases a C-α substituent oriented

pseudoequatorially to the L-histidine chelate ring would give steric interference with the azomethine substituent, destabilizing the λ conformation (structure **15**). For simple copper(II)–histidine complexes the main stabilizing factor for a given chelate ring conformation is the apical coordination of a polar group on the C-α substituent to Cu(II),[20] but even when this group is noncoordinating similar CD behaviour is generally observed.



λ δ

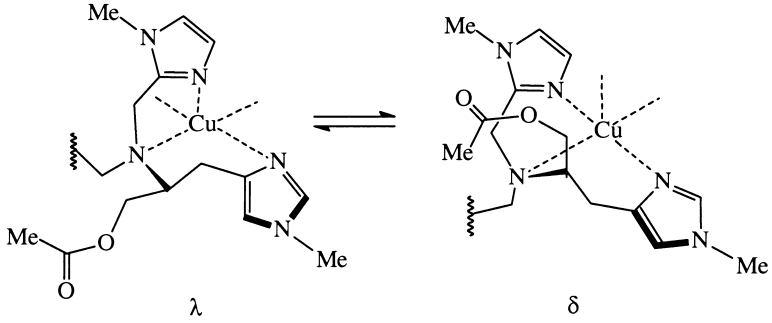
13 **14**



λ δ

15 **16**

The negative CD activity exhibited by the Cu(II) centers bound to PHI A sites (**12**) suggests the preference for a conformation of the L-histidine chelate ring of λ chirality, with a pseudoequatorial disposition of the acetoxymethylene side chain, as shown by structure **17**. It appears that, for the presence of an additional substituent chain at the tertiary amine nitrogen in the ligand PHI, some significant steric interference occurs with a pseudoaxial acetoxymethylene side chain, as shown in structure **18**. Minimization of this steric interference between the C-α and amino substituent groups leads to the preference for the conformational arrangement depicted in structure **17**, where the imidazole substituent is axially coordinated to the copper center.



λ δ

17 **18**

The EPR spectra of the complexes give further information on the structure of the Cu(II) centers (Table 1). In all cases, the spectra recorded in frozen acetonitrile solution at −150°C exhibit well resolved

signals attributable to a single species with tetragonal symmetry ($g_{\|}>g_{\perp}$).[22] For $[CuIH]^{2+}$ the rather large hyperfine splitting constant $|A_{\|}|$ indicates that the Cu(II) center is essentially square planar, with three strong donors in the plane and only weak axial binding by solvent molecules, confirming the analysis of the electronic and CD spectra. For $[Cu_2PHI]^{4+}$ and $[Cu_3PHI]^{6+}$ the EPR parameters of the species observed are practically identical and indicative of Cu(II) sites with square-pyramidal geometry, since the copper hyperfine constant $|A_{\|}|$ has a rather reduced value.[22,25] Such a reduced $|A_{\|}|$ value results from a strong axial interaction by a donor group, supporting the structural arrangement depicted in structure **17**. If the presence of a single Cu(II) species is expected for $[Cu_2PHI]^{4+}$, where the two coppers are presumably spatially well separated and non-interacting, such an observation is surprising for $[Cu_3PHI]^{6+}$, where an EPR spectrum with overlapping features from two distinct Cu(II) sites (A and B) should be expected. The integrated EPR signal for $[Cu_3PHI]^{6+}$, however, corresponds to only 1.6 paramagnetic centers (at −150°C), whereas it accounts for 2.0 paramagnetic centers in the case of $[Cu_2PHI]^{4+}$. A similar behaviour was exhibited by the trinuclear $[Cu_3L]^{6+}$ complex,[10] and it is therefore likely that some significant dipolar interaction in the frozen state between one of the Cu(II)-A centers and the Cu(II)-B center leads to broadening of the signals and an apparent reduction in EPR intensity.

In conclusion, the systems reported here represent a step forward in our studies aimed at mimicking the trinuclear copper clusters present in the blue oxidases. The introduction of L-histidine residues into the ligand framework and the possibility of exploiting the better resolution of CD spectra enables a more complete stereochemical description of the complexes to be achieved, besides producing a coordination environment that is closer to that of the enzymes, compared with the model compounds available to date.[9,10] We have shown here that, with respect to our previous systems derived from the ligand L, lengthening of the carbon chain connecting the piperazine residue to the two chiral arms of PHI is advantageous for two reasons: (i) it allows an easier access to the trinuclear complex $[Cu_3PHI]^{6+}$; and (ii) it prevents the binding of the piperazine nitrogens to Cu(II) in the binuclear $[Cu_2PHI]^{4+}$ complex. The latter complex, therefore, contains Cu(II) centers bound to genuine tridentate A sites (structure **12**) and represents a true model for the type 2 copper-depleted forms of the blue oxidases.[1,5] Binding of a third Cu(II) center to the piperazine moiety has no significant effect on the structural arrangement of the other pair of Cu(II) ions. The latter centers, bound to stereogenic A sites, adopt an unusual conformation with the histidine chelate ring in a λ conformation and a pseudoequatorial C-α substituent. This arrangement has the drawback of producing a rather facile racemization of the coordinated amino acid residue in the presence of base,[23b] and such an effect also occurred here on treatment of $[Cu_2PHI]^{4+}$ or $[Cu_3PHI]^{6+}$ with small amounts of sodium hydroxide, in attempts to generate a hydroxy-bridged structure similar to that existing in the biological clusters shown in Scheme 1. However, the peculiar conformational arrangement at the A sites is probably unavoidable, because it originates from the attachment chain to the tridentate, L-histidine residues, and should be taken into consideration in any synthetic approach towards protein biomimetics involving derivatized histidines as metal ligands. Further studies on the reactivity of the present model complexes derived from PHI are currently in progress in our laboratory.

3. Experimental

3.1. Materials and physical methods

All reagents and solvents from commercial sources were of the highest purity available and were used as received. Acetonitrile (spectral grade) was distilled from potassium permanganate, and dried with sodium carbonate; it was then stored over calcium hydride and distilled prior to use under an

inert atmosphere. Tetrahydrofuran was dried by refluxing and distilling from metallic sodium. Elemental analyses were obtained at the microanalytical laboratory of the chemistry department in Milan. ^{1}H and ^{13}C NMR spectra were recorded on a Bruker AC-200 spectrometer, operating at 200 MHz and 50.3 MHz, respectively. Optical rotations were obtained on a Perkin–Elmer 241 polarimeter at 20°C using a quartz cell of 10 cm path length. Optical spectra were obtained on HP 8452A and 8453 diode array spectrophotometers. EPR spectra were measured in frozen solutions using a Varian E-109 spectrometer operating at X-band frequencies. Integration of the EPR signals was performed using a copper(II)–EDTA standard in the same conditions. FABMS spectra were obtained with a VG 7070 EQ spectrometer and CD spectra were recorded on a Jasco J-500 dichrograph using quartz cells of 0.1–1 cm path length.

3.2. Ligand synthesis

3.2.1. L-N$^\tau$*-Methylhistidine methyl ester dihydrochloride* ***2***

This compound was obtained from L-*N*$^\tau$-methylhistidine dihydrochloride, prepared in accordance with Noordam,[17] by esterification in methanol with gaseous HCl. Anal. calcd for $C_8H_{13}N_3O_2\cdot 2HCl\cdot H_2O$ (274.15): C 35.05, H 6.25, N 15.33; found C 35.12, H 5.99, N 15.42. The free amino acid methyl ester was obtained by bubbling gaseous NH_3 through a solution of the hydrochloride salt in dry, cooled dichloromethane. After filtration over Celite, the solution was evaporated to dryness and dried under vacuum. ^{1}H NMR (200 MHz, $CDCl_3$, 25°C): δ=7.34 (s, 1H, CH), 6.68 (s, 1H, CH), 3.83 (m, 1H, CH), 3.72 (s, 3H, CH_3), 3.62 (s, 3H, CH_3), 3.04 (dd, 1H, ^{2}J=14 Hz, ^{3}J=5 Hz, CH_2), 2.84 (dd, 1H, ^{2}J=14 Hz, ^{3}J=8 Hz, CH_2). Anal. calcd for $C_8H_{13}N_3O_2$ (183.21): C 52.45, H 7.15, N 22.94; found C 52.42, H 7.32, N 22.54.

3.2.2. 4,4′-Dioxo-4,4′-piperazine-1,4-diyl-bisbutanoic acid) ***3***

This compound was prepared in accordance with Asay et al.[16] as white crystals. ^{1}H NMR (200 MHz, D_2O, 25°C): δ=3.39 (t, ^{3}J=10 Hz, 8H, CH_2), 2.55–2.37 (m, 8H, CH_2); MS (FAB): *m*/*z* (%): 286 (100) [M^+]; $C_{12}H_{18}N_2O_6$ (286.28): calcd C 50.35, H 6.34, N 9.79; found C 50.28, H 6.48, N 9.67.

*3.2.3. 4,4′-Dioxo-4,4′-piperazine-1,4-diyl-bisbutanoic acid-bis[1-*S*-methoxycarbonyl-2-(1-methyl-1*H*-imidazol-4-yl)]ethanamide* ***4***

Piperazine derivative **3** (0.94 g, 3.27 mmol) was added under nitrogen at 0°C, to a solution of HBTU (2.5 g, 6.55 mmol) in dry CH_3CN (5 mL). The suspension was stirred for 15 min and then a solution of free L-*N*$^\tau$-methylhistidine methyl ester (1.2 g, 6.55 mmol) in dry CH_3CN (3 mL) was added and the mixture was stirred at room temperature for 20 h. The reaction was followed by silica gel TLC, using ethyl acetate:isopropanol:ammonium hydroxide (5:4:1, v/v/v) as eluent. After evaporation of the solvent at reduced pressure, the crude material was chromatographed on a neutral alumina column, using a gradient of CH_2Cl_2–CH_3OH. The fractions containing the product were combined and the solvent was removed under vacuum, affording compound **4** (1.45 g, yield 72%) as a yellow oil. $[\alpha]_D^{20}$=+4.59 (c=0.0148 in methanol). ^{1}H NMR (200 MHz, $CDCl_3$, 25°C): δ=7.38 (s, 2H, CH), 7.31 (d, 2H, ^{1}J=8 Hz, NH), 6.70 (s, 2H, CH), 4.76 (m, 2H, CH), 3.70 (s, 6H, CH_3), 3.65 (s, 6H, CH_3), 3.67–3.47 (m, 8H, CH_2), 3.03 (m, 4H, CH_2), 2.60 (m, 8H, CH_2). ^{13}C NMR (50.3 MHz, $CDCl_3$, 25°C): δ=28.1, 28.8, 29.0, 30.8, 33.3, 33.4, 38.4, 39.7, 41.4, 44.8, 45.9, 52.1, 52.3, 52.9, 57.1, 118.1, 118.7, 135.9, 137.2, 137.5, 161.9, 170.4, 171.1, 171.7, 171.8. MS (CI): *m*/*z* (%): 617 (100) [M^++1]. Anal. calcd for $C_{28}H_{40}N_8O_8$ (616.67): C 54.54, H 6.54, N 18.17; found C 54.53, H 6.14, N 17.92.

*3.2.4. 1,4-Bis[4-(*N*-(1-hydroxy-3-(1-methyl-1*H*-imidazol-4-yl))-2-*S*-propyl)amino-butyl]piperazine* ***5***

A solution of $BH_3 \cdot Me_2S$ (15 mL, 162 mmol) in dry THF (10 mL) was added dropwise to a suspension of **4** (1 g, 1.62 mmol) in boiling, dry THF (10 mL) under nitrogen. Then, the solvent and Me_2S were slowly distilled for a further 5 h while fresh, dry THF (80 mL) was added. The solvents were removed under vacuum and the solid residue was taken up with a saturated solution of HCl in methanol. The suspension was refluxed under stirring for 1 h until the solid was completely dissolved. After cooling, the solution was evaporated and the residue was saturated with gaseous NH_3 in dry, cooled (0°C) dichloromethane. The resulting precipitate was filtered off and the filtrate was concentrated. The precipitate was submitted twice to this treatment and the filtrates, after evaporation of the solvent, afforded compound **5** (0.76 g, 1.51 mmol), which was used in the subsequent reaction without further purification. $[\alpha]_D^{20}$=+4.70 (c=0.0127 in methanol). ^{1}H NMR (200 MHz, $CDCl_3$, 25°C): δ=7.45 (s, 2H, CH), 6.67 (s, 2H, CH), 3.65 (s, 6H, CH_3), 3.73–3.42 (m, 4H, CH), 2.94 (m, 2H, CH), 2.77–2.60 (m, 4H, CH_2), 2.66 (m, 4H, CH_2), 2.45 (m, 8H, CH_2), 2.33 (m, 4H, CH_2), 1.50 (m, 8H, CH_2). ^{13}C NMR (50.3 MHz, $CDCl_3$, 25°C): δ=24.4, 28.0, 30.2, 33.2, 46.7, 53.0, 58.3, 58.4, 62.6, 117.7, 137.1, 139.1. MS (FAB): *m*/*z* (%): 504 (100) [M^+]. Anal. calcd for $C_{26}H_{48}N_8O_2$ (504.72): 61.87, H 9.59, N 22.20; found C 61.76, H 9.50, N 22.20.

*3.2.5. 1,4-Bis[4-(*N*-(1-hydroxy-3-(1-methyl-3-(1-methyl-1*H*-imidazol-4-yl)))-2-*S*-propyl)-*N*-(1-methyl-1*H*-imidazol-2-ylmethyl)aminobutyl]piperazine* ***6***

Compound **5** (0.425 g, 0.84 mmol), 1-methyl-1*H*-imidazole-2-carbaldehyde **8** (0.37 g, 3.37 mmol) and acetic acid (0.19 mL, 3.37 mmol) were stirred under nitrogen in dry THF (15 mL) at room temperature for 20 min. $NaBH(OAc)_3$ (0.892 g, 2.52 mmol) was added, the reaction was stirred for 15 h, and then an additional amount of compound **8** (0.21 g, 1.68 mmol), acetic acid (0.096 mL, 1.7 mmol), and $NaBH(OAc)_3$ (0.53 g, 1.7 mmol) were added. The reaction was left under stirring for a further 24 h. At the end of this time 4 N HCl (5 mL) was added and the solution was concentrated under vacuum. The residue was suspended in dry, cooled (0°C) dichloromethane saturated with gaseous NH_3. The solid precipitated was separated and treated again with cooled, NH_3 saturated dichloromethane. After filtration, the combined filtrates were evaporated under vacuum and the yellow oil **6** was purified by flash silica gel column chromatography, eluting with a mixture of Et_2O:*i*PrOH (5:4, v/v) and adding an increasing amount of concentrated NH_3 from 1% to 15%. The fractions containing the product afforded, after the evaporation of the solvent, compound **6** (0.345 g, yield 60%) as a yellow oil. $[\alpha]_D^{20}$=−10.06 (c=0.0154 in methanol). ^{1}H NMR (200 MHz, $CDCl_3$, 25°C): δ=7.29 (s, 2H, CH), 6.87 (d, 2H, J=1 Hz, CH), 6.79 (d, 2H, J=1 Hz, CH), 6.65 (s, 2H, CH), 3.88 (d, 2H, ^{2}J=16 Hz, *H*–C–H), 3.73 (d, 2H, ^{2}J=16 Hz, H–C–*H*), 3.62 (s, 6H, CH_3), 3.60 (s, 6H, CH_3), 3.67–3.40 (m, 4H, CH_2), 3.14–2.96 (m, 2H, CH), 2.81 (dd, 2H, ^{2}J=4 Hz, ^{3}J=14 Hz, *H*–C–H), 2.73–2.44 (m, 14 H, CH_2), 2.35–2.16 (m, 4H, CH_2), 1.53–1.15 (m, 8H, CH_2). ^{13}C NMR (50.3 MHz, $CDCl_3$, 25°C): δ=23.4, 25.1, 26.3, 32.7, 33.2, 46.8, 49.8, 51.9, 57.6, 62.2, 62.8, 117.1, 121.2, 126.7, 136.8, 139.9, 146.0. MS (FAB): *m*/*z* (%): 692 (100) [M^+]. Anal. calcd for $C_{36}H_{60}N_{12}O_2$ (692.95): C 62.40, H 8.73, N 24.26; found C 62.62, H 8.61, N 24.02.

*3.2.6. 1,4-Bis[4-(*N*-(1-acetoxy-3-(1-methyl-1*H*-imidazol-4-yl))-2-*S*-propyl)-*N*-(1-methyl-1*H*-imidazol-2-ylmethyl)aminobutyl]piperazine* ***7*** *(PHI)*

Compound **6** (0.285 g, 0.411 mmol) was stirred at room temperature for 6 h in distilled acetic anhydride (3 mL) in the presence of 4-dimethylaminopyridine (DMAP) (0.03 g). The solution was then concentrated under vacuum at 50°C and the residue was purified by flash silica gel chromatography, eluting with a mixture of Et_2O:*i*PrOH (5:4, v/v) and adding an increasing amount of concentrated NH_3 from 1% to 15%. The fractions containing the product afforded, after evaporation of the solvent, compound **7** (0.280

g, yield 87%) as a pale-yellow oil. $[\alpha]_D^{20}$=+1.12 (c=0.014 in methanol). ^{1}H NMR (200 MHz, $CDCl_3$, 25°C): δ=7.29 (s, 2H, CH), 6.88 (s, 2H, CH), 6.79 (s, 2H, CH), 6.60 (s, 2H, CH), 4.18–4.00 (m, 4H, CH_2)), 3.84 (m, 4H, CH_2), 3.61 (s, 6H, CH_3), 3.44–3.25 (m, 2H, CH), 2.85 (dd, 2H, ^{2}J=6 Hz, ^{3}J=14 Hz, *H*–C–H), 2.67–2.30 (m, 14H, CH_2), 2.30–2.13 (m, 4H, CH_2), 2.00 (s, 6H, CH_3), 1.50–1.07 (m, 8H, CH_2). ^{13}C NMR (50.3 MHz, $CDCl_3$, 25°C): δ=20.9, 24.0, 26.1, 26.5, 32.5, 33.1, 48.9, 49.9, 52.6, 58.0, 59.5, 64.0, 117.2, 121.4, 126.7, 137.0, 139.8, 145.7, 170.8. UV–vis (acetonitrile): λ_{max} (ε)=298 (1200), 220 sh nm (16 800). CD (acetonitrile): λ_{max} (Δε, M^{-1} cm^{-1})=280 (+0.10). MS (CI): *m*/*z* (%): 778 (100) [M^++1]. Anal. calcd for $C_{40}H_{64}N_{12}O_4$ (777.03): C 61.83, H 8.30, N 21.63; found C 61.51, H 7.94, N 21.35.

*3.2.7. 1-Methyl-1*H*-imidazole-2-carbaldehyde **8***

This compound was obtained by slight modification of the procedure of Chen et al.[18] 2-(Hydroxymethyl)-1-methylimidazole (2.51 g, 0.022 mol) was dissolved in $CHCl_3$ (50 mL) under stirring at room temperature and activated MnO_2 (9.75 g, 0.11 mol) was added to the solution. The mixture was stirred for 22 h and then filtered over Celite. The filtrate was evaporated to dryness and the oil residue was sublimated under vacuum at 60°C, yielding a while crystalline solid (0.61 g, yield 25%) which was stored at 4°C. ^{1}H NMR (200 MHz, $CDCl_3$, 25°C): δ=9.80 (s, 1H, CHO), 7.26 (s, 1H, CH), 7.11 (s, 1H, CH), 4.02 (s, 3H, CH_3). IR (Nujol): 1694 cm^{-1} (C=O). Anal. calcd for $C_5H_6N_2O$ (110.12): C 54.54, H 5.49, N 25.44; found C 54.16, H 5.28, N 25.21.

3.2.8. L-N^{α}*-(1-Methyl-1*H*-imidazol-2-ylmethyl)*-N^{τ}*-methylhistidine methyl ester **1** (IH)*

To a solution of L-*N*$^{\tau}$-methylhistidine methyl ester dihydrochloride **2** (0.503 g, 1.83 mmol) in dry methanol (10 mL) was added dropwise a methanolic solution of NaOH 1 M (3.66 mL) and 1-methyl-1*H*-imidazole-2-carbaldehyde **8** (0.202 g, 1.83 mmol) dissolved in dry methanol (10 mL). Palladium–charcoal (10% palladium content) (1 g) was added and the mixture was hydrogenated at atmospheric pressure with stirring for 20 h. The catalyst was removed by filtration over Celite, and the filtrate was evaporated to a small volume (5 mL). Then, dry dichloromethane was added in order to precipitate the inorganic salts, which were removed by centrifugation, and the solution was evaporated to dryness under vacuum. Spectral analysis of the yellow oil thus obtained demonstrated the lack of the methyl ester group and, therefore, the product was esterified again by dissolving the oil in anhydrous methanol (10 mL), and bubbling into this cooled methanol solution gaseous HCl for 1 h. The solution was refluxed for 1 h and after cooling, concentration to a small volume under vacuum yielded a white precipitate, which was crystallized from methanol (0.52 g, 62%). A sample, extracted in H_2O/Na_2CO_3 and dichloromethane, was submitted to NMR. ^{1}H NMR (200 MHz, $CDCl_3$, 25°C): δ=7.45 (s, 1H, CH), 7.00 (s, 1H, CH), 6.85 (s, 1H, CH), 6.72 (s, 1H, CH), 3.98 (d, 1H, ^{2}J=14 Hz, CH_2), 3.87 (d, 1H, ^{2}J=14 Hz, CH_2), 3.64–3.42 (m, 1H, CH), 3.02 (dd, 1H, ^{2}J=14 Hz, ^{3}J=5 Hz, CH_2), 2.88 (dd, 1H, ^{2}J=14 Hz, ^{3}J=7 Hz, CH_2) Anal. calcd for $C_{13}H_{19}N_5O_2\cdot 3HCl\cdot 4H_2O$ (458.77): C 34.04, H 6.59, N 15.27; found C 34.12, H 6.42, N 15.27.

3.3. Synthesis of copper(II) complexes

3.3.1. [Cu_2PHI][ClO_4]$_4\cdot 7H_2O$

To a solution of the ligand L **7** (0.021 mmol) in CH_3OH (10 mL) was added copper(II) perchlorate hexahydrate (0.042 mmol) and the resulting green solution was stirred at room temperature for about 30 min. The solution was then evaporated to a small volume (2 mL) and the residue was treated with diethyl ether (1 mL). The light blue-green precipitate was separated by filtration and dried under vacuum (95%).

Anal. calcd for $C_{40}H_{64}N_{12}O_{20}Cu_2Cl_4 \cdot 7H_2O$ (1428.03): C 33.64, H 5.51, N 11.77; found C 33.69, H 5.29, N 11.60.

3.3.2. $[Cu_3PHI][ClO_4]_6 \cdot 7H_2O$

To a solution of the ligand L **7** (0.021 mmol) in CH_3OH (10 mL) was added copper(II) perchlorate hexahydrate (0.063 mmol) and the solution was stirred at room temperature for 40 min. The solution was evaporated to a small volume (3 mL) and the residue was treated with diethyl ether (2 mL). The light blue precipitate was separated from the solution by filtration and dried under vacuum (97%). Anal. calcd for $C_{40}H_{64}N_{12}O_{28}Cu_3Cl_6 \cdot 7H_2O$ (1690.47): C 28.42, H 4.65, N 9.94; found C 28.50, H 4.70, N 9.88.

3.3.3. $[CuIH(Cl)][ClO_4]$

To a solution of the ligand IH (0.04 g, 0.087 mmol) in CH_3OH (10 mL) was added copper(II) perchlorate hexahydrate (0.032 g, 0.087 mmol) and the solution was stirred at room temperature for 1 h. The solution was evaporated to a small volume (3–4 mL) and the residue was treated with cold diethyl ether (10 mL). The light blue precipitate was separated from the solution by filtration and dried under vacuum (57%). Anal. calcd for $C_{13}H_{19}N_5O_6CuCl_2$ (475.78): C 32.82, H 4.03, N 14.72; found C 32.81, H 3.85, N 14.87.

4. Caution!

Perchlorate complexes with organic ligands are potentially explosive and should be handled with great care. Only small amounts of material should be prepared. We did not have problems working with small amounts of the perchlorate complexes described in this paper.

Acknowledgements

This work was supported by the Italian M.U.R.S.T. The authors would like to thank the Italian C.N.R. for allowing us to use its instrumentation facilities.

References

1. (a) Solomon, E. I.; Sundaram, U. M.; Machonkin, T. E. *Chem. Rev*. **1996**, *96*, 2563; (b) Solomon, E. I.; Baldwin, M. J.; Lowery, M. D. *Chem. Rev*. **1992**, *92*, 521.
2. (a) Reinhammar, B. In *Copper Proteins and Copper Enzymes*, Vol. 3; Lontie R., Ed.; CRC Press: Boca Raton, FL, 1984; p. 1; (b) Thurston, C. F. *Microbiology* **1994**, *140*, 19; (c) Marzullo, L.; Cannio, R.; Giardina, P.; Santini, M. T.; Sannia, G. *J. Biol. Chem*. **1995**, *270*, 3823; (d) Bollag, J. M. *Met. Ions Biol. Syst*. **1992**, *28*, 205.
3. (a) Kroneck, P. M. H.; Armstromg, F. A.; Merckle, H.; Marchesini, A. *Adv. Chem. Ser*. **1982**, *200*, 223; (b) Chichiriccò, G.; Cerù, P.; M. D'Alessandro, A.; Oratore, A.; Avigliano, L. *Plant Sci*. **1989**, *64*, 61; (c) Mondovì, B.; Avigliano, L. In *Copper Proteins and Copper Enzymes*, Vol. 3; Lontie, R., Ed.; CRC Press: Boca Raton, FL, 1984; p. 101.
4. (a) Ryden, L. In *Copper Proteins and Copper Enzymes*, Vol. 3; Lontie, R., Ed.; CRC Press: Boca Raton, FL, 1984; p. 37; (b) Messerschmidt, A.; Huber, R. *Eur. J. Biochem*. **1990**, *187*, 341; (c) Musci, G.; Carbonaro, M.; Adriani, A.; Lania, A.; Galtieri, A.; Calabrese, L. *Biochem. Biophys.* **1990**, *279*, 8.
5. (a) Messerschmidt, A.; Rossi, A.; Ladenstein, R.; Huber, R.; Bolognesi, M.; Gatti, G.; Marchesini, A.; Petruzzelli, R.; Finazzi-Agrò, A. *J. Biol. Chem*. **1989**, *206*, 513; (b) Messerschmidt, A.; Ladenstein, R.; Huber, R.; Bolognesi, M.; Avigliano, L.; Petruzzelli, R.; Rossi, A.; Finazzi-Agrò, A. *J. Mol. Biol*. **1992**, *224*, 179; (c) Messerschmidt, A.; Luecke, H.; Huber, H. *J. Mol. Biol*. **1993**, *230*, 997; (d) Messerschmidt, A. In *Bioinorganic Chemistry of Copper* Karlin, K. D.; Tyeklar Z., Eds; Chapman & Hall: New York, 1996; p. 471; (e) Messerschmidt, A. *Adv. Inorg. Chem*. **1994**, *40*, 121.

6. Zaitseva, I.; Zaitsev, V.; Card, G.; Moshkov, K.; Bax, B.; Ralph, A.; Lindley, P. *J. Biol. Inorg. Chem*. **1996**, *1*, 15.
7. Malkin, R.; Malmström, B. G. *Adv. Enzymol*. **1970**, *33*, 170.
8. Nguyen, H.-H. T.; Nakagawa, K. H.; Hedman, B.; Elliott, S. J.; Lidstrom, M. E.; Hodgson, K. O.; Chan, S. I. *J. Am. Chem. Soc*. **1996**, *118*, 12766.
9. (a) Carlin, K. D.; Gan, Q.-F; Farooq, A.; Liu, S.; Zubieta, J. *Inorg. Chem*. **1990**, *29*, 2085; (b) Adams, H.; Bailey, N. A.; Dwyer, M. J. S.; Fenton, D. E.; Hellier, P. C.; Hempstead, P. D.; Latour, J. M. *J. Chem. Soc., Dalton Trans*. **1993**, 1207; (c) Meenakumari, S.; Tiwary, S. K.; Chakravarty, A. R. *Inorg. Chem*. **1994**, *33*, 2085; (d) Frey, S. T.; Sun, H. H. J.; Murth, N. N.; Karlin, K. D. *Inorg. Chim. Acta* **1996**, *242*, 399; (e) Singh, K.; Long, J. R.; Stavropoulos, P. *Inorg. Chem*. **1998**, *37*, 1073.
10. Monzani, E.; Casella, L.; Zoppellaro, G.; Gullotti, M.; Pagliarin, R.; Bonomo, R. P.; Tabbì, G.; Nardin, G.; Randaccio, L. *Inorg. Chim. Acta* **1998**, *282/2*, 180.
11. (a) Sorrell, T. N.; Borovik, A. S. *J. Am. Chem. Soc*. **1987**, *109*, 4255; (b) Sorrell, T. N.; Vankai, V. A.; Garrity, M. L. *Inorg. Chem*. **1991**, *30*, 207; (c) Sorrell, T. N.; Garrity, M. L. *ibid*. **1991**, *30*, 210; (d) Oberhausen, K. J.; Richardson, J. F.; Buchanan, R. M.; Mc Cusker, J. K.; Hendrickson, D. N.; Latour, J. M. *ibid*. **1991**, *30*, 1357; (e) Tolman, W. B.; Rardin, R. L.; Lippard, S. J. *J. Am. Chem. Soc*. **1989**, *111*, 4532; (f) Lynch, W. E.; Kurtz Jr., D. M.; Wang, S.; Scott, R. A. *ibid*. **1994**, *116*, 11030; (g) Sorrell, T. N.; Allen, W. E.; White, P. S. *Inorg. Chem*. **1995**, *34*, 952; (h) Koolhaas, G. J. A. A.; Driessen, W. L.; Reedijk, J.; van der Plas, J. L.; de Graaff, R. A. G.; Gatteschi, D.; Kooijman, H.; Spek, A. L. *ibid*. **1996**, *35*, 1509.
12. Koning, W.; Geiger, R. *Chem. Ber*. **1970**, *103*, 788.
13. Castro, B.; Dormoy, J. R.; Dourtoglou, B.; Evin, G.; Selve, C.; Ziegler, J. C. *Synthesis* **1976**, 751.
14. Fujita, E. *Pure and Appl. Chem*. **1981**, A.*53*, 1141.
15. Dourtoglou, V.; Gross, B. *Synthesis* **1983**, 572.
16. Asay, R. E.; Bradshaw, J. S.; Nielsen, S. F.; Thompson, M. D.; Snow, J. W.; Masihdas, D. R. K.; Izatt, R. M.; Christensen, J. J. *J. Heterocycl. Chem*. **1977**, *14*, 85.
17. Noordam, A.; Maat, L.; Beyerman, H. C. *Recueil J. of the Royal Netherlands Chem. Soc*. **1978**, *97*, 293.
18. Chen, S.; Richardson, J. F.; Buchanan, R. M. *Inorg. Chem*. **1994**, *33*, 2376.
19. (a) Fawcett, T. G.; Bernaducci, E. E.; Krogh-Jespersen, K.; Schugar, H. J. *J. Am. Chem. Soc*. **1980**, *102*, 2598; (b) Bernaducci, E. E.; Schwindinger, W. F.; Hughey IV, J. L.; Krogh-Jespersen, K.; Schugar, H. J. *ibid*. **1981**, *103*, 1686.
20. (a) Casella, L.; Gullotti, M. *J. Inorg. Biochem*. **1983**, *18*, 19; (b) Casella, L.; Gullotti, M. *Inorg. Chem*. **1983**, *22*, 242; (c) Casella, L.; Gullotti, M. *ibid*. **1985**, *24*, 84.
21. (a) Amundsen, A. R.; Whelan, J.; Bosnich, B. *J. Am. Chem. Soc*. **1977**, *99*, 6730; (b) Downes, J. M.; Whelan, J.; Bosnich, B. *Inorg. Chem*. **1981**, *20*, 1081; (c) Kennedy, B. P.; Lever, A. B. P. *J. Am. Chem. Soc*. **1973**, *95*, 6907.
22. Hataway, B. J. In *Comprehensive Coordination Chemistry*, Vol. 5; Wilkinson, G., Ed.; Pergamon Press: New York, 1987; p. 533.
23. (a) Casella, L.; Gullotti, M. *Inorg. Chem*. **1981**, *20*, 1306; (b) Casella, L.; Gullotti, M.; Pacchioni, G. *J. Am. Chem. Soc*. **1982**, *104*, 2386.
24. Casella, L.; Gullotti, M.; Pallanza, G.; Buga, M. *Inorg. Chem*. **1991**, *30*, 221.
25. (a) Wayland, B. B.; Kapur, V. K. *Inorg. Chem*. **1974**, *13*, 2517; (b) Kogane, T.; Hirota, R.; Abe, K.; Hirota, M. *J. Chem. Soc., Perkin Trans. 2* **1981**, 652.