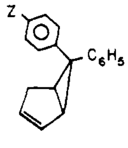
tially vacant p orbital of the carbene.[7] It now appears that inclusion of large substituent-generated steric effects offers an alternative explanation of these observations. That is, when Z is electron withdrawing, the substituted aryl function aligns to maximize the overlap with the electron rich σ-orbital of the carbene. Assuming electrophilic attack upon the π-framework of the olefin, the transition state for endo addition (**9**) is



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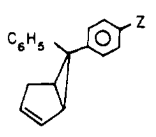

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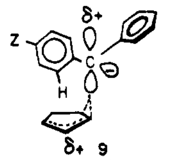

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therefore impeded by the steric repulsion between the olefin and an ortho ring hydrogen. Such congestion will be relieved for modes of addition in which the endo aryl ring is coplanar with the electron deficient carbenic orbital (exo addition for Z = electron withdrawer; endo addition for Z = electron donor).

Examination of the calculated minimum energy reaction profiles for decomposition of para-substituted phenyldiazomethanes is in progress.

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(3) In the present system the diphenyldiazomethanes (**1**) thermolyze in water/1,2-dimethoxyethane to benzhydrols (95–99%) and benzophenone azines (0–5%). The kinetics indicate clearly that **1** decomposes unimolecularly in a rate limiting process to **2** which is then captured by water or **1** to yield the benzhydrol or the benzophenone azine. The details of this investigation will be described separately.

(4) M. J. S. Dewar, R. C. Haddon, W-K. Li, W. Thiel, and P. K. Weiner, *J. Am. Chem. Soc.*, **97**, 4540 (1975), have treated methylene in various electronic configurations by MINDO/3 methods.

(5) QCPE Program No. 279 was modified to double precision for use with the IBM 370/168 facility available at The Ohio State University. The phenylcarbene system was chosen as a theoretical model because it provided a working simplification within the chemical and budget requirements. Throughout the calculations, the aromatic ring was treated as a rigid, regular hexagon with carbon–carbon and carbon–hydrogen bond lengths of 1.400 and 1.080 Å, respectively. All remaining geometrical parameters were optimized as a function of energy.

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Metal Complexes of Poly(α-amino acids). Optical Rotatory Properties of Copper(II) Complexes of Poly-L-lysine, Poly-L-ornithine, and Poly-L-diaminobutyric Acid

Sir:

In the last few years a considerable amount of work has been carried out on metal complexes of poly(α-amino acids).[1] Such compounds can be considered useful models in order to understand the way of action of metalloproteins. It is known that all water soluble poly(α-amino acids), with the only exception of poly-L-proline, bind Cu(II) ions in alkaline aqueous solution, forming complexes of the biuret type.[2] Poly-L-histidine[3] and poly-L-lysine[4,5] bind copper ions even in the pH region below 8 forming other kinds of complexes in which imidazole and amino side-chains, respectively, are involved.

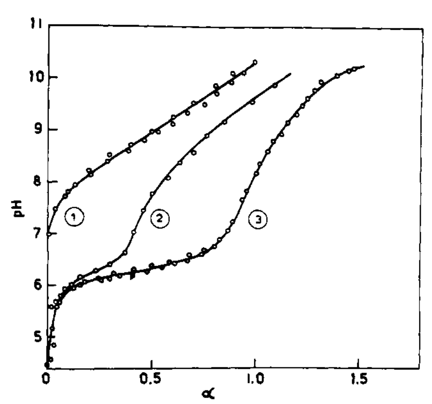


Figure 1. Titration curves of PDBA-Cu(II) aqueous solutions at different Cu(II)/PDBA molar ratios: (1) Cu(II)/PDBA = 0, (2) Cu(II)/PDBA = 0.1, (3) Cu(II)/PDBA = 0.25. In all cases [PDBA] was in the range $(5.2-5.4) \times 10^{-3}$ molar residue.

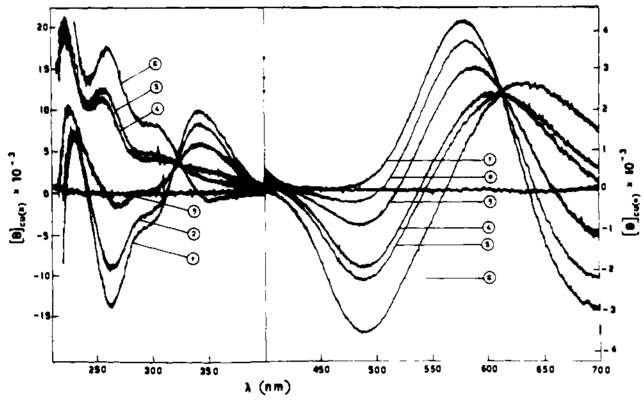


Figure 2. CD spectra of PDBA-Cu(II) aqueous solutions at different pH's. [PDBA] = 4.8×10^{-3} molar residue [Cu(II)] = 1.2×10^{-3} M: (1) pH 8.29, (2) pH 9.31, (3) pH 10.18, (4) pH 11.67, (5) pH 12.09, (6) pH >13. The ellipticity values are given per bound Cu(II).

In view of the importance of copper complexes of peptides and poly(α-amino acids) as models for copper proteins, and of the effects of stereospecific catalysis, we have reexamined the problem of complex formation of Cu(II) with poly-L-lysine (PLL), and the study has been extended to poly-L-ornithine (PLO) and poly(L-diaminobutyric acid) (PDBA). The specific purpose of our work was to ascertain the relationships between complex structure and conformation of the polypeptide backbone.

As an example the potentiometric titration curve of PDBA in absence and in presence of Cu(II) ions is shown in Figure 1. When cupric ions are present, two distinct buffer regions are observed, corresponding to the formation of two kinds of complexes. Type I complex is formed at pH <8, while type II complex is stable at very alkaline pH's. Inspection of the potentiometric titration data of PDBA with the Bjerrum method reveals that four protons are displaced per mole of bound Cu^{2+} at pH $\simeq$7.5. This result is consistent with that previously reported by Matano et al.[4,5] on a type I complex of PLL, in which four amino nitrogens have been suggested to occupy square-planar coordination positions of Cu^{2+}. Type II complexes are formed at strongly basic pH values, and the hypothesis has been made that *amido* nitrogens are involved in complex formation.[4,5]

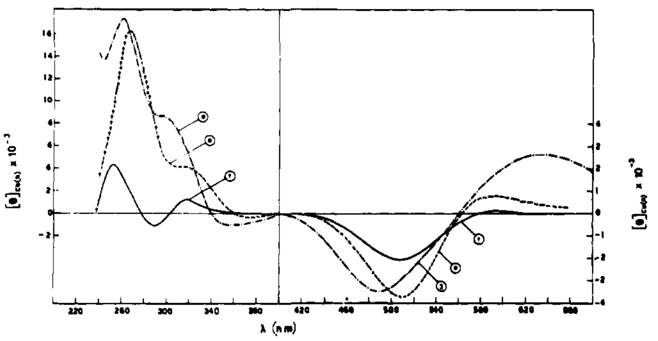


Figure 3. CD spectra of Cu(II) complexes of PLL, PLO, and PDBA in water: (1) [PLL] = 5.2×10^{-3} molar residue, [Cu(II)] = 1.3×10^{-3} M, pH 10 (the complex precipitates at higher pH); (2) [PLO] = 4.2×10^{-3} molar residue, [Cu(II)] = 1.16×10^{-3} M, pH >13; (3) [PDBA] = 4.8 $\times 10^{-3}$ molar residue, [Cu(II)] = 1.2×10^{-3} M, pH >13. The ellipticity values are given per bound Cu(II).

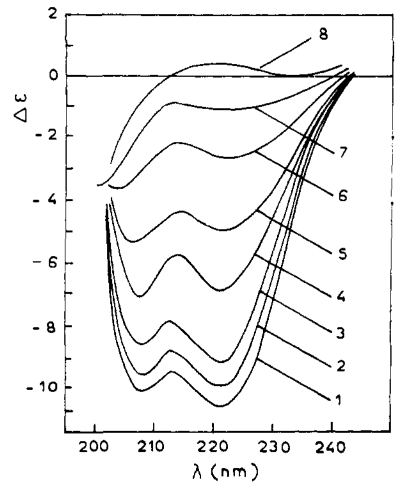
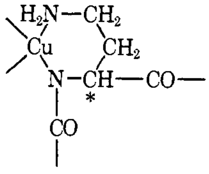


Figure 4. CD spectra of the Cu(II) complex of PLL in water at pH 11.2 and at different Cu(II)/PLL ratios in the region 250-200 nm: (1) Cu(II)/PLL = 0, (2) Cu(II)/PLL = 0.017, (3) Cu(II)/PLL = 0.033, (4) Cu(II)/PLL = 0.057, (5) Cu(II)/PLL = 0.082, (6) Cu(II)/PLL = 0.106, (7) Cu (II)/PLL = 0.130, (8) Cu(II)/PLL = 0.161. In all cases [PLL] was 5.4×10^{-3} molar residue.

In order to obtain structural information on both types of copper complexes of the three examined poly(α-amino acids), the visible and UV absorption spectra, and the CD properties as well, have been investigated in the pH range 5–14. Considering first type I low pH complexes, we have found that the PLL and PLO copper complexes exhibit almost identical absorption characteristics, with a broad band at 650 nm, in the region of Cu(II) d-d transitions. On the other hand the visible absorption maximum of the PDBA complex is blue shifted down to 580 nm. In general a blue shift is observed when amide nitrogens replace amino nitrogens in the planar positions of the coordination sphere of Cu^{2+}.[6,7] Our results therefore suggest that, differently from the case of PLL and PLO, in type I complex of PDBA at least one deprotonated amide nitrogen coordinates in one of the square-planar positions of Cu^{2+}. This is not an unusual result since deprotonated amide nitrogens have been recognized as one of the most important binding sites in oligopeptides, and in many cases it has been found that deprotonation of peptide nitrogens occurs at pH around 7.

Additional support to the hypothesis that a peptide bond participates in the coordination of Cu(II) in the complex of PDBA at low pH stems from CD measurements at various pH values (Figure 2). While type I complex of PLL and PLO possess vanishingly small optical rotation, both in the visible and in the UV absorption regions, the corresponding complex of PDBA exhibits intense dichroic absorption both in the regions of d-d transitions and of charge-transfer transitions. Two major bands opposite in sign are observed in the visible: a negative one at 700 nm and a positive one at 580 nm. Additional bands in the UV are observed at 340 nm (positive), 300 nm (negative), and 265 nm (very strong and negative) which can be reasonably assigned to charge-transfer transitions. Inspection of the CD properties in the peptide absorption region reveals that in type I complexes the polymeric backbone is always in the random coil conformation, and that in no case is such a conformation altered by the binding of Cu^{2+} ions. The strong optical activity observed in the case of type I complex of PDBA must be therefore attributable to an asymmetric induction from the chiral center of single amino acid residues. If side-chain amino groups only were involved in the complex formation, as in the case of PLL and PLO, very small optical rotation should be expected, owing to the large distance of the asymmetric α-carbon atom from the coordination sphere of Cu(II). When at least one peptide nitrogen is coordinated at one of the square positions, the asymmetric α-carbon atom is much closer, and asymmetric induction is therefore expected in the absorption regions of the complex. A possible explanation of why amide groups are coordinated to Cu(II) only in PDBA in the low pH complex could rest on the fact that a side-chain amino group and an adjacent peptide nitrogen can form a stable hexatomic chelate ring involving two square-planar coordination positions (I), the remaining positions being occupied by amino nitrogens. Such a possibility clearly does not exist for type I copper complexes of PLL and PLO, in which only unstable hepta- or octaatomic chelate rings are possible, and therefore coordination with amino nitrogens is preferred.



Our CD results on complex I of PDBA in the d-d transition region are qualitatively similar to those reported by Levitzki et al.[3] on the copper complex of [L-His]$_n$ formed at pH 5. Also in that case the complex exhibits a CD pattern with two oppositely signed bands, and the hypothesis was made that three imidazole nitrogens and one deprotonated amide nitrogen are bound at the planar positions of the Cu(II) coordination sphere.

Discussing now the CD results obtained on type II complexes formed at high pH, we observe again that the behavior of PDBA is different from that of the other two polymers especially in the metal d-d transitions region (Figure 3). There is a strong positive band at 635 nm followed by a negative one of almost identical rotatory strength a 490 nm. Below 400 nm the CD pattern is almost the mirror image of that of complex I, and this reflects the different way of binding and therefore the different structure of the two kinds of complexes.

Interestingly, the entire series of CD spectra of PDBA in the presence of Cu(II) ions, recorded from pH 8 to pH >13 shows two clear isodichroic points at 322 and 613 nm. This fact seems to suggest the presence of a two component equilibrium system, with additive contributions from each component to the total optical activity. Substitution of amino nitrogens with *amido* nitrogens in the square-planar positions likely occurs. Coordination of amido nitrogens at the apical positions, as suggested by other authors for PLL[4,5] is rather improbable. Neither studies on the crystal structure nor any of the solution studies on copper complexes of oligopeptides gave evidence for coordination of amido nitrogens at the apical position.[7,8] Comparison of the optical rotatory properties suggests that the structure of the three polymeric type II complexes are different from each another. The complexes of PLL and PLO exhibit almost identical CD patterns in the visible, characterized by a strong negative band at 510 nm and by a weaker positive band at $\simeq$585 nm. In the charge-transfer transitions region below 400 nm the CD pattern of the PLO complex is similar to that of the PDBA complex, but substantially different from that of the PLL complex. Such variation of the CD properties among type II complexes of the three polymers possibly reflects a different number of amido nitrogens coordinated to Cu(II) at the planar positions and/or different apical interactions of side-chain amino groups in the three cases. Again the CD spectrum of the biuret type complex of PDBA is very similar to the corresponding complex of $[\text{L-His}]_n$ formed at pH 14. As in the latter case it is possible that side-chain amino groups of PDBA occupy apical positions in the coordination sphere of Cu(II).

We have also proved that the α-helical structure of the polypeptide backbone is not compatible with the formation of type II copper complexes of PLL and PLO. If a solution of PLL at pH $\simeq$11.2, (where the polypeptide conformation is 100% α-helix) is titrated with cupric ions, one observes a decrease of helical content which parallels the extent of complex formation (Figure 4). The disruption of the helical structure appears to be noncooperative in character, being linearly proportional to the amount of added metal ions. We can therefore reasonably conclude that cupric ions are coordinated to peptide sequences not in the helical form, and previously proposed models based on coordination to the polypeptide chain in the α-helical form are inconsistent.

More detailed results including potentiometric infrared and CD investigations will be available in due course and will be reported elsewhere.

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Cation Solvation: the Conversion of Contact Ion Pairs to Solvent Separated Ion Pairs

Sir:

Since the proposal by Winstein[1] of two types of ion pairs, many techniques have been developed to differentiate between them,[2] with the distribution depending on solvent donicity, cation size, anion, and temperature. It has also been found that in some systems two or more types of contact ion pairs can be observed,[3] whereas in other systems two or more types of solvent separated ion pairs can exist.[4] Focusing on the contact pairs, it is reasonable to imagine contact pairs with various extents of cation and/or anion solvation (solvated contact pairs). The question then arises as to how many solvent molecules are necessary to convert a contact ion pair to a solvent separated ion pair.

Attempts to answer this question have been complicated because of the nature of the usual electrolyte systems. In a pure solvent, the concentration of the solvating agent cannot be effectively varied, and one typically observes contact, solvent separated, or an equilibrium mixture of the two or more types of ion pairs.[2,5] But it is not possible to determine the extent of solvation of the ions. Mixed solvents could be used to overcome this problem.[2,6] However, they are generally unsatisfactory because of the solubility characteristics of the usual electrolytes, which result in the solvent and the selected donor competing for coordination sites on the ions.

The system sodium tetraethylaluminate ($NaAlEt_4$)-benzene-donor (D) is a particularly unique system for this type of study. $NaAlEt_4$ is soluble in benzene, a nonsolvating solvent. Thus controlled amounts of a solvating agent can be added to a benzene solution of $NaAlEt_4$, thereby permitting one to determine the ion pair type as a function of the D/Na^+ ratio. We wish to report here ratio studies using this system with a number of monodentate and bidentate solvating agents having Gutmann donor numbers ranging from 19.2 to 38.8.[7]

It is possible to distinguish between contact and solvent separated ion pairs in the $NaAlEt_4$–benzene–D system by examining the spin–spin interactions between ^{27}Al and ^1H as reflected in the methylene group proton resonance patterns.[8] In samples containing anions with cubic symmetry, the methylene absorptions appear as well-defined nine-line patterns representing the fortuitous overlapping of resonances resulting from essentially equal spin–spin interactions of methylene protons with both the neighboring methyl protons and the aluminum nucleus. Conversely, in samples containing highly distorted anions, the $^{27}Al-^1H$ spin interactions are not observed in the ^1H NMR spectra. Rather, the resonance collapses to a 1,3,3,1 quartet resulting from the remaining $^1H-^1H$ spin interactions. The two extremes, a nine-line pattern representing the anionic T_d symmetry and a quartet corresponding to the highly distorted anion are logically related to solvent separated and contact ion pairs, respectively.[9]

The preparation of $NaAlEt_4$, solvent, and sample preparation are described elsewhere.[10] The solvating agents were distilled over CaH_2 and handled as described earlier.[10] NMR spectra were obtained on Varian A-60A and HA100 spectrometers. Solutions of these systems tend to form two phases in benzene at certain ratios of donor/Na^+ with the salt and the solvating agent being predominantly in the lower phase. Some of the spectra shown are of the lower phase. Consequently, the concentrations of the salt and the solvating agent will show considerable variation, but since both remain in the lower phase, the D/Na^+ ratio is as listed. Except for a possible effect due to viscosity, the spectra are, in general, independent of this behavior. In those instances where a single phase is formed, the salt concentration is 0.26 M.

In Figure 1, a comparison of the methylene resonances is shown for $NaAlEt_4$ in benzene using diethyl ether (Et_2O), teterahydrofuran (THF), and hexamethylphosphoramide (HMPA) as solvating agents. For the 1:1 ratios of D/Na^+, the expected quartet characteristic of contact ion pairs is observed. However, using HMPA as the donor, a 4:1 ratio of HMPA/Na^+ gives a ^1H NMR spectrum characteristic of solvent separated ion pairs. In contrast, a 4:1 ratio of THF/Na^+ gives a ^1H NMR spectrum indicative of a preponderance of contact ion pairs. Finally, if we consider the limit where the donor is used as the solvent, it is seen that the methylene resonance in HMPA is essentially the same as it is for the 4:1 ratio of D/Na^+. When THF is used as the solvent, the nine-line pattern characteristic of solvent separated ion pairs is also observed. However, the resolution is poorer than that observed with