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Chelated Cobalt Bis(ethylenediamine) Amino Acid Ester Complexes: Racemization during Peptide Synthesis

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A study of the acylation of the chelated amino acid ester complex $[(en)_2Co(l\text{-Phe-OMe})]^{3+}$ (II) with other amino acids was carried out. The reaction of II with *l*-Phe-OMe and *l*-Phe-O-*t*-Bu in Me_2SO resulted in the formation of 18% of the racemized product, *d*-Phe-*l*-Phe (after the cobalt was removed). The $[(en)_2Co^{III}(peptide)]$ complexes formed undergo further racemization of the amino acid residue bound to Co(III) when they are dissolved in neutral aqueous solution. This large degree of racemization observed makes the use of these *chelated* Co(III) complexes unsuitable in the general synthesis of biologically active peptides. A different strategy where monodentate cobalt(III) complexes act as C-terminal protecting groups has been demonstrated to be useful in peptide synthesis, with peptide formation without racemization.

The use of chelated $[(en)_2Co^{III}(\text{amino acid})]$ complexes in peptides synthesis has been suggested by two groups as early as 1967.[1,2] Recently, advances in synthetic methodologies allowed the ready synthesis of a number of chelated $[(en)_2Co^{III}(\text{amino acid ester})]$ complexes in pure form.[3]

The comparative study of these cobalt(III) active esters with organic active esters is important for evaluating their use in peptide synthesis. However, evaluations of these cobalt active esters by two independent groups were not in agreement.[3,4] Wautier et al. concluded that peptides formed by using cobalt(III) amino acid esters such as $[(tren)Co^{III}(\text{amino acid ester})]$, where tren is 2,2′,2″-triaminotriethylamine, are extensively racemized at the chiral carbon of the amino acid bound to Co(III).[4] Tasker et al., who studied mainly $[(en)_2Co^{III}(\text{amino acid ester})]$ complexes under slightly different conditions, reported that a number of peptides could be synthesized without appreciable racemization.[3]

Our work in the area of peptide synthesis using Co(III) complexes centers around limiting the role of the metal ion to the role of a protecting group. Kinetically inert metal ion protecting groups have many advantages over existing organic protecting groups, including their color, charge, ease of monitoring, purification, and finally ease of removal by mild reducing agents.[5,8]

However, the $[(en)_2Co^{III}(\text{amino acid ester})]$ complexes, as reported,[3] appeared to offer exciting new possibilities for mild and rapid methods of peptide synthesis. In this paper we have investigated the use of the reported $[(en)_2Co^{III}(\text{amino acid ester})]$ complex (amino acid = *l*-phenylalanine = l-Phe) for peptide synthesis and studied the effect of the Co(III) complex on the chirality of the chelated amino acid undergoing acylation.

Experimental Section

Chemicals. The amino acids and esters, *l*-Phe, *l*-Phe-OMe·HCl (Aldrich), Boc-*d*-Phe (Boc = *tert*-butoxycarbonyl) (Peninsula Labs, San Carlos, CA), *l*-Phe-O-*t*-Bu·HCl, *d*-l-Phe, and *l*-Phe-*l*-Phe (Sigma) were used as supplied. Triethylamine (Eastman) was distilled before use. Dimethyl sulfoxide (99.9%, Aldrich) was dried over CaH_2, then distilled under reduced pressure at 70 °C, and stored over 4A molecular sieves. Trimethyl phosphate and methyl trifluoromethanesulfonate (Aldrich) were used as supplied. The SP-Sephadex C25 cation-exchange resin (Pharmacia) was used as supplied.

Methods. HPLC was performed on a Waters Associates liquid chromatography system with an Altech C_{18} Nucleosil column (5 μm, 4.6 mm × 25 cm). HPLC grade methanol (J. T. Baker Co.) and all the HPLC solvents were filtered through 0.2 μm Nylon-66 membrane filters (Rainin).

Proton NMR spectra were recorded on a T-60 spectrometer at room temperature using D_2O or CD_3CN as solvents and 3-(trimethylsilyl)-1-propionic acid as an external standard.

Visible absorption spectra were recorded on a Cary 118 C spectrophotometer.

Racemization Studies. Two independent methods were used to detect any racemization of intermediates during peptide formation. The first method utilized HPLC analysis with a reverse-phase C_{18} column and the second method utilized an amino acid analyzer with a cation-exchange column. In the HPLC method the Altech Nucleosil C_{18} column described above was used. The eluent for the peptides was 81% 0.1 M H_3PO_4 (adjusted to pH 3.27 with K_2HPO_4) and 19% CH_3CN. The flow rate was 0.9 mL/min, and the absorbance was monitored at 254 nm.

In the second method the peptides were hydrolyzed in 6 M HCl, and then amino acid analysis was done by using a Glenco amino acid analyzer with a Durrum DC-4A (0.3 × 300 mm) cation-exchange resin. The eluent for the amino acids was 0.1 M sodium acetate, pH 5.5 (adjusted with acetic acid), 8×10^{-3} M $CuSO_4$, and 1.6×10^{-3} M *l*-proline. The flow rate was 13.0 mL/h. The column pressure was 700–750 psi, and the column temperature was 62 °C. Detection of the amino acids, after derivatization with *o*-phthalaldehyde, was achieved by using a Perkin-Elmer fluorescence detector. Separation of *l*- and *d*-amino acids from the column was done by using the method of Hare and Gil-Av.[9]

Synthesis of Cobalt(III) Complexes. *cis*-$[(en)_2CoCO_3]Cl$ and *cis*-$[(en)_2Co(CF_3SO_3)_2]_2(CF_3SO_3)_2$ were prepared by using literature methods.[10,11]

***cis*-$[(en)_2Co(l\text{-Phe})]I_2$.** The complex *cis*-$[(en)_2Co(CF_3SO_3)_2]CF_3SO_3$ was converted to *cis*-$[(en)_2Co(Me_2SO)_2](CF_3SO_3)_3$ by dissolution in Me_2SO. The amino acid *l*-Phe was added to form *cis*-$[(en)_2Co(l\text{-Phe})]I_2$, by using the procedure of Tasker et al.[3] The visible spectrum has λ_{max} 467 nm (ϵ = 96 M^{-1} cm^{-1}), 346 nm (ϵ = 108 M^{-1} cm^{-1}). ^1H NMR: δ 7.4 (5 H, s, C–H aromatic), 3.2 (2 H, d, CH_2), 2.6 (8 H, br, CH_2).

***cis*-$[(en)_2Co(l\text{-Phe-OMe})](CF_3SO_3)_3$.** This complex was also prepared by using the procedure of Tasker et al.[3]

***cis*-$[(en)_2Co(l\text{-Phe-}l\text{-Phe-OR})]Cl_3$ (R = Me or *t*-Bu).** *cis*-$[(en)_2Co(l\text{-Phe-}l\text{-Phe-OMe})]^{3+}$ complex was prepared by using Tasker's method[3] with a reaction time of 20 min. A detailed description of the preparation is as follows: *l*-Phe-OMe·HCl (0.683 g, 3.17 mmol) and dry triethylamine (264 μL, 1.9 mmol) in Me_2SO (25 mL) were added to $[(en)_2Co(l\text{-Phe-OMe})](CF_3SO_3)_3$ (0.51 g, 0.63 mmol) in Me_2SO (5 mL), and the solution was stirred at room temperature for 20 min. The reaction was quenched by adding the mixture into 250 mL of 1% acetic acid. The mixture was then loaded onto a SP-Sephadex C_{25} cation-exchange column (1.5 × 10 cm, Na^+ form).

After the column was extensively washed with deionized water (500 mL), two orange bands were eluted with sodium chloride. The first band,

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which eluted with 0.125 M NaCl, was identified as $[(en)_2CoPhe)]Cl_2$, and the second band, which eluted with 0.6–0.8 M NaCl, was the cobalt dipeptide product. The second band was concentrated to dryness by rotary evaporation. Methanol was added to the dry solid, and the insoluble sodium chloride was removed by filtration. The methanol filtrate was concentrated to 0.5 mL by rotary evaporation and then was added dropwise to cold anhydrous ether. The resulting orange precipitate was collected on a fine fritted funnel, washed with 30 mL of anhydrous ether, and dried in a vacuum desiccator over $CaCl_2$. Yield: 0.25 g (63%) of *cis*-$[(en)_2Co(l\text{-Phe-}l\text{-Phe-OMe})]Cl_3$. The visible spectrum in water shows λ_{max} 488 nm (ϵ = 97 M^{-1} cm^{-1}), 344 nm (ϵ = 110 M^{-1} cm^{-1}). ^1H NMR: δ 7.2 (s), 7.0 (br, 10 H, C-H aromatic), 3.8 (3 H, s, CH_3), 3.2 (4 H, complex, CH_2), 2.6 (8 H, br, $CH_2(en)$).

cis-$[(en)_2Co(l\text{-Phe-}l\text{-Phe-O-}t\text{-Bu})]Cl_3$ was prepared by using *l*-Phe-O-*t*-Bu·HCl and following the procedure outlined above. Similar results were obtained by using reaction times of 5 and 20 min. The yield was 0.6 g of product (60%). The visible spectrum in water shows λ_{max} 487 nm (ϵ = 97 M^{-1} cm^{-1}), 344 nm (ϵ = 110 M^{-1} cm^{-1}).

Removal of Cobalt from the Dipeptide Ester Complexes. A portion of *cis*-$[(en)_2Co(l\text{-Phe-}l\text{-Phe-O-}t\text{-Bu})]Cl_3$ was dissolved in 18% acetic acid, and 8 equiv of $NaBH_4$ were added. The solution was stirred vigorously for 5 min, during which time it changed colors from orange to pink to colorless. Finally a black solid appeared mixed with a fluffy white precipitate. This mixture was extracted with ether, and the white solid *l*-Phe-*l*-Phe-O-*t*-Bu dissolved. The combined ether extracts were dried with anhydrous $MgSO_4$, filtered, and concentrated to dryness by rotary evaporation. Yield: 78%.

^1H NMR of the dipeptide ester: δ 7.2 (10 H, s(split), C–H aromatic), 2.9 (4 H, d(br), CH_2), 1.2 (9 H, s, CH_3).

Hydrolysis of the Dipeptide Esters *l*-Phe-*l*-Phe-OR (R = *t*-Bu or Me). *l*-Phe-*l*-Phe-O-*t*-Bu was dissolved in a 1:1 mixture of CH_2Cl_2/HTFA, and the solution was stirred for 3 h at room temperature (to insure complete hydrolysis). The solution was concentrated to dryness by a rotary evaporator. Upon addition of anhydrous ether a white precipitate formed which was filtered and dried in a vacuum desiccator over $CaCl_2$. ^1H NMR: δ 7.2 (10 H, s, C–H aromatic), 3.0 (4 H, two doublets, CH_2).

The dipeptide *l*-Phe-*l*-Phe was also obtained from *l*-Phe-*l*-Phe-OMe directly after cobalt removal from *cis*-$[(en)_2Co(l\text{-Phe-}l\text{-Phe-OMe})]^{3+}$ with $NaBH_4$ in 18% acetic acid/methanol (1:3). The solution was adjusted to pH 11 with LiOH and was stirred for 45 min at room temperature. The completeness of ester hydrolysis was checked on HPLC after aliquots were neutralized with acetic acid.

Synthesis of the Dipeptide *d*-Phe-*l*-Phe. The *d*-Phe-*l*-Phe standard was prepared by using two different methods.

Method I.[6] To a solution of Boc-*d*-Phe (1.5 g, 5.6 mmol) in 4 mL of CH_2Cl_2 at 0 °C was added a solution of hydroxybenzotriazole (HOBT) (0.87 g, 5.6 mmol) in 1.5 mL of DMF at 0 °C. A solution of dicyclohexylcarbodimide (DCC) (1.18 g, 5.6 mmol) in 1.5 mL of CH_2Cl_2 at 0 °C was added to the reaction mixture. The formation of the active ester was continued for 1 h at 0 °C and then for 1 h at room temperature. The solution was then filtered and evaporated to dryness with a rotary evaporator. It was dissolved in DMF (2 mL) and $[l\text{-Phe-Co}(NH_3)_5](CF_3CO_2)_2$ (1.0 g, 1.87 mmol) was added to it. Coupling was initiated by the addition of diisopropylethylamine (0.4 mL, 2.3 mmol). The reaction mixture was stirred for 2 h at room temperature, evaporated to a viscous liquid, and treated with 2 × 15 mL of ether. The aqueous layer containing the cobalt complex was concentrated on a rotary evaporator and loaded on a preparative C_{18} column (37–50μ, 1 × 15 cm). A minor band eluted with the solvent 20% MeOH–H_2O and 0.2% HTFA, pH 3.0 adjusted with NaOH. The major band eluted with the solvent 60% MeOH–H_2O and 0.2% HTFA, pH 3.0 adjusted with NaOH, and was concentrated to dryness by rotary evaporation, dissolved in the minimum volume of absolute methanol, and then precipitated with anhydrous ether. It was filtered, washed with 15 mL of ether, and dried in a vacuum desiccator over $CaCl_2$ (1.30 g, 87% yield).

^1H NMR for $[Boc\text{-}d\text{-Phe-}l\text{-Phe-Co}(NH_3)_5]^{2+}$: δ 7.2 (10 H, s(br), C–H aromatic), 3.8 (3 H, trans-NH_3), 3.4 (12 H, cis-NH_3) 3.0 (4 H, br, CH_2), 1.2 (9 H, s, CH_3).

The Boc-protecting group was removed by dissolving the cobalt complex in 1:1 CH_2Cl_2/HTFA and stirring for 3 h at room temperature, and the product was purified by using cation-exchange chromatography (SP-Sephadex C_{25}) followed by gel filtration (Bio-Gel P2). Yield: 1.0 g, 88%.

^1H NMR for $[d\text{-Phe-}l\text{-Phe-Co}(NH_3)_5]^{3+}$: δ 7.3 (10 H, s(br), C–H aromatic), 3.8 (3 H, trans-NH_3), 3.6 (12 H, cis-NH_3), 3.0 (4 H, d, CH_2).

The cobalt(III) protecting group was removed with $NaBH_4$ as described earlier.

Method II. To the active ester formed (as in Method I) was added 6.0 mL of DMF. To this solution were added solid *l*-Phe-O-*t*-Bu·HCl (1.0, 3.88 mmol) and 0.42 mL of *N*-methylmorpholine to initiate the coupling reaction. The reaction mixture was stirred for 5 h at room temperature. The reaction was quenched with 50 mL of 0.01 M HTFA and extracted with ether. The combined ether extracts were dried with anhydrous $MgSO_4$, filtered, and concentrated to dryness on a rotary evaporator. The Boc and the *tert*-butyl ester groups were removed as described earlier.

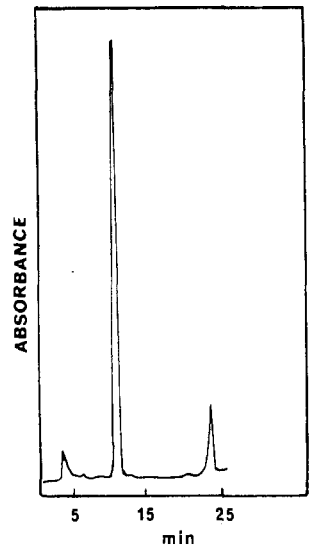
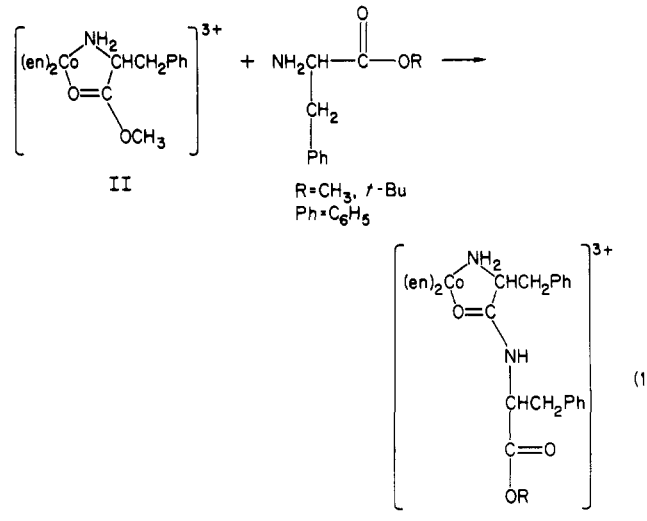


Figure 1. HPLC chromatogram of the peptide diastereomers *l*-Phe-*l*-Phe and *d*-Phe-*l*-Phe, formed during acylation of $[(en)_2Co\text{-}l\text{-Phe-OCH}_3]^{3+}$.

^1H NMR for *d*-Phe-*l*-Phe: δ 7.0 (10 H, s, C–H aromatic), 2.8 (4 H, two doublets, CH_2), 4.05 (1 H, t, CH), 4.15 (1 H, t, CH).

Results

Racemization Studies of the Cobalt Amino Acid Complexes $[(en)_2Co(l\text{-Phe})]^{3+}$ (I) and $[(en)_2Co(l\text{-Phe-OMe})]^{3+}$ (II). Experiments to determine the racemization induced during the formation of the cobalt amino acid $[(en)_2Co(l\text{-Phe})]^{3+}$ (I) and its esterification to II (eq 1) were done with an amino acid an-



(1)

alyzer. In both cases, the cobalt complexes were dissolved in acidic solution, the Co(III) was removed with $NaBH_4$, and the resulting solution was analyzed on an amino acid analyzer and compared with standards *d*-Phe and *l*-Phe. During the synthesis of $[(en)_2Co(l\text{-Phe})]^{3+}$ (I), <0.1% *d*-Phe was formed. Esterification of I with $CF_3SO_3CH_3$ to form II resulted in the formation of 0.2% *d*-Phe.

Racemization during Peptide Synthesis with $[(en)_2Co(l\text{-Phe-OMe})]^{3+}$ (II). Several experiments were conducted where II was used to make dipeptides by using Tasker's method[3] as shown in eq 1. The products of the reaction were isolated and analyzed for their isomeric content. Two independent methods were used to estimate the racemization of the amino acids undergoing acylation.[7,12]

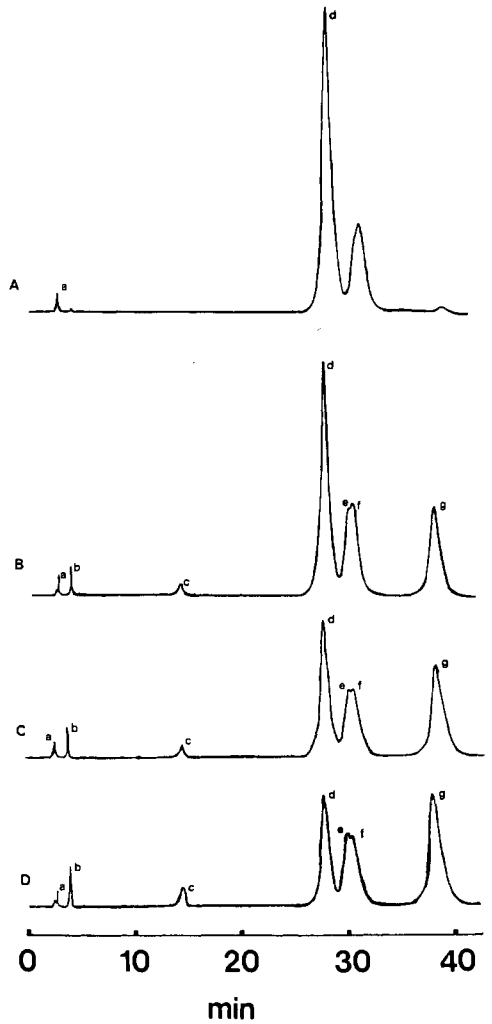


Figure 2. HPLC chromatograms showing racemization of the Co(III) center and the amino acid bound to it: (a) $[(en)_2Co(OH_2)OH]^{2+}$; (b) $[(en)_2Co(l,d\text{-Phe})]^{2+}$; (c) *l*-Phe-$OCH_3$; (d) Λ-$[(en)_2Co(l\text{-Phe-}l\text{-Phe-}OCH_3)]^{3+}$; (e) Λ-$[(en)_2Co(d\text{-Phe-}l\text{-Phe-}OCH_3)]^{3+}$; (f) Δ-$[(en)_2Co(l\text{-Phe-}l\text{-Phe-}OCH_3)]^{3+}$; (g) Δ-$[(en)_2Co(d\text{-Phe-}l\text{-Phe-}OCH_3)]^{3+}$. Chromatograms A–D were obtained at t = 0, 60, 150, and 195 min, respectively.

In the first method, the dipeptide esters and the free dipeptides (formed after cobalt removal) were analyzed directly on HPLC with UV absorption spectroscopy. Standard samples of the dipeptides, *l*-Phe-*l*-Phe and *d*-Phe-*l*-Phe, were used for comparison. Figure 1 shows the dipeptide diastereomers, *l*-Phe-*l*-Phe and *d*-Phe-*l*-Phe, which formed in the reaction of $[(en)_2Co(l\text{-Phe-OMe})]^{3+}$ with *l*-Phe-OR (R = Me, *t*-Bu) after cobalt removal and ester hydrolysis. The results show that 18.3% of the racemized product, *d*-Phe-*l*-Phe, formed during the peptide formation step (in Me_2SO).

In the second method, the amino acids resulting from peptide hydrolysis (in 6 M HCl) were analyzed with ion-exchange chromatography. When this method was used, 16.8% of the racemized amino acid, *d*-Phe, was found.

Racemization of *cis*-$[(en)_2Co(Phe\text{-}l\text{-}Phe\text{-}OMe)]Cl_3$ (III). When *cis*-$[(en)_2Co(Phe\text{-}l\text{-}Phe\text{-}OMe)]Cl_3$ is dissolved in aqueous solution (pH 6), several changes occur on the time scale of hours at room temperature. The *l*-Phe bound to cobalt undergoes further racemization, and the Co(III) center also undergoes racemization (resulting in the formation of a number of diastereomers). In addition to these changes, loss of Phe-OMe was observed, a process known to occur in similar complexes.[13] This hydrolysis resulted in an increase in pH from pH 6 to pH 7.5 over a period of 4 h. These changes are shown qualitatively in Figure 2 with the HPLC chromatograms of the reaction at pH 6 over several hours. At time zero, the *cis*-$[(en)_2Co(Phe\text{-}l\text{-}Phe\text{-}OMe)]^{3+}$ already has $\Lambda l,l$, $\Delta l,l$ and $\Lambda d,l$ isomers chelated to cobalt(III). As the reaction proceeds, the $\Lambda l,l$ and the $\Delta l,l$ isomers decrease, while the $\Lambda d,l$ and the $\Delta d,l$ isomers increase. Over a period of several hours, peptide hydrolysis occurs with the release of *l*-Phe-OMe and the chelated $[(en)_2Co(Phe)]^{3+}$ (appearance of bands b and c, respectively). Assignment of the Λ and Δ cobalt isomers was made by following the work of Tasker et al.[3] on the corresponding Λ,Δ-$[(en)_2Co(Phe)]^{3+}$; on this basis band d is assigned to $\Lambda l,l$ and f to $\Delta l,l$. As the peptide undergoes racemization, isomers $\Lambda d,l$ (e) and $\Delta d,l$ (g) are formed; isomer $\Delta d,l$ (g) increased significantly as the pH of the solution increased.

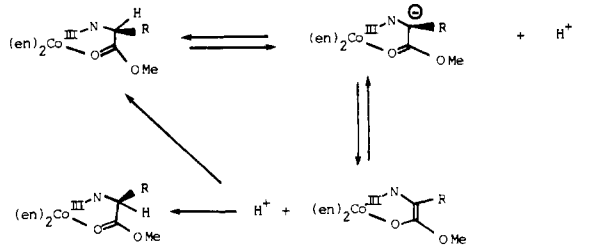


Figure 3. Racemization pathway of $[(en)_2Co(l\text{-Phe-}OCH_3)]^{3+}$ in basic media via enolization.

Discussion

The work described here is an investigation of the utility of $[(en)_2Co^{III}(amino\ acid\ ester)]$ complexes in peptide synthesis. The amino acid *l*-phenylalanine (*l*-Phe) was chosen for detailed study because peptides derived from it can be easily monitored by UV absorption. Also, phenylalanine is an amino acid not especially prone to racemization, such as the more reactive trifunctional amino acids.[14]

The formation of the $[(en)_2Co^{III}(amino\ acid)]$ (I) and $[(en)_2Co^{III}(amino\ acid\ ester)]$ (II) complexes is not accompanied by any significant racemization (eq 1). These results on the absence of racemization during formation of the cobalt amino acid complexes are in agreement with those of Tasker et al.[3] However, extensive racemization was observed during the rapid aminolysis of $[(en)_2Co^{III}(Phe\text{-}OMe)]$ to form the $[(en)_2Co^{III}(Phe\text{-}Phe\text{-}OMe)]$ in Me_2SO in the presence of triethylamine. The results of this work show that 18% *d*-Phe is formed during this aminolysis reaction. These results on racemization during peptide formation are very different from those reported by Tasker et al, where only minimal racemization (0.2–0.4%) was observed.[3]

The extent of racemization during peptide formation in this work was verified by two independent methods: (1) direct analysis of the dipeptide diastereomers formed and (2) amino acid analysis of the *d*- and *l*-amino acids after acid hydrolysis. Control experiments showed that cobalt removal from $[(en)_2Co^{III}(l\text{-Phe})]$ and $[(en)_2Co^{III}(l\text{-Phe-OMe})]$ did not induce any measurable racemization.

Further evidence for the extensive racemization of $[(en)_2Co^{III}(amino\ acid\ ester)]$ complexes can be found in the recent work of Wautier et al.[4] Using $[(en)_2Co^{III}(Gly\text{-}OMe)]$ at pH 9.8, Wautier et al. demonstrated that deuterium exchange at the methine carbon proceeds with a half-life of 19 s. This deuterium exchange was estimated to be 1.9×10^6 times greater for $[(en)_2Co^{III}(Gly\text{-}OMe)]$ than that for the analogous $[(en)_2Co^{III}Gly]$ complex.[4] Esterification of the chelated cobalt amino acid complex activates the C=O group and allows it to undergo rapid aminolysis, but also makes the methine carbon a significantly stronger acid, which undergoes proton exchange more readily.

The magnitude of the difference in the rate of proton exchange between the $[(en)_2Co^{III}(amino\ acid)]$ and the $[(en)_2Co^{III}(amino\ acid\ ester)]$ (ca. a factor of 10^6) is surprising. One can understand

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this difference by examining the carbanions formed at the methine carbon for both the amino acid and the amino acid ester complexes. Enolization of the deprotonated amino acid ester (Figure 3) may result in the stabilization of the negative charge on the oxygen directly bound to the tripositive Co(III) center. Such stabilization is absent for the $[(en)_2Co^{III}(amino\ acid)]$ complex, which already possesses a negative charge on the carboxylate oxygen.

The extensive racemization observed in the chelated Co(III) amino acid ester complexes during peptide formation makes these reagents unsuitable for the synthesis of biologically active peptides. It may be possible in the future to modify the ligands around the cobalt or to find conditions for aminolysis without racemization; however, for the two systems investigated thus far, the $[(en)_2Co^{III}(amino\ acid\ ester)]$ and the $[(tren)Co^{III}(amino\ acid\ ester)]$ complexes, extensive racemization is observed during peptide formation. Another drawback reported earlier in the use of these chelated cobalt(III) complexes for peptide synthesis is the sensitivity of these complexes to steric effects, thus requiring long reaction times for aminolysis of amino acids with bulky side chains.

Recent results from our laboratory have shown that more practical uses of cobalt complexes for peptide synthesis can be achieved, when the role of the cobalt complex is limited to that of a blocking group.[5-8] In this strategy, one can take advantage of the kinetic inertness of Co(III) centers, which allows them to withstand the conditions required for stepwise peptide synthesis[15] (ca. strong acids) and yet be easily removed from the synthesized peptides by mild reducing conditions.

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Registry No. *cis*-$[(en)_2Co(l\text{-Phe})]I_2$, 18717-13-0; *cis*-$[(en)_2Co(CF_3SO_3)_2]CF_3SO_3$, 75522-52-0; *cis*-$[(en)_2Co(Me_2SO)_2](CF_3SO_3)_3$, 99546-59-5; *cis*-$[(en)_2Co(l\text{-Phe-OMe})](CF_3SO_3)_3$, 80585-84-8; *cis*-$[(en)_2Co(l\text{-Phe-}l\text{-Phe-OMe})]Cl_3$, 99546-60-8; *cis*-$[(en)_2Co(l\text{-Phe-}l\text{-Phe-O}t\text{-Bu})]Cl_3$, 99546-61-9; *l*-Phe-*l*-OMe, 13082-29-6; *l*-Phe-*l*-Phe-O-*t*-Bu, 47555-30-6; *l*-Phe-*l*-Phe, 2577-40-4; Boc-*d*-Phe, 18942-49-9; HOBT, 2592-95-2; $[l\text{-Phe-Co}(NH_3)_5](CF_3CO_2)_2$, 81751-79-3; $[Boc\text{-}d\text{-Phe-}l\text{-Phe-Co}(NH_3)_5]^{2+}$, 99546-62-0; $[d\text{-Phe-}l\text{-Phe-Co}(NH_3)_5]^{2+}$, 99603-01-7; *l*-Phe, 63-91-2; Boc-*d*-Phe HOBT ester, 99532-43-1; *l*-Phe-O-*t*-Bu·HCl, 15100-75-1; *d*-Phe-*l*-Phe, 2577-22-2; Λ-$[(en)_2Co(l\text{-Phe-}l\text{-Phe-OCH}_3)]^{3+}$, 99603-02-8; Λ-$[(en)_2Co(d\text{-Phe-}l\text{-Phe-OCH}_3)]^{3+}$, 99603-03-9; Δ-$[(en)_2Co(l\text{-Phe-}l\text{-Phe-OCH}_3)]^{3+}$, 99603-53-9; Δ-$[(en)_2Co(d\text{-Phe-}l\text{-Phe-OCH}_3)]^{3+}$, 99559-65-6.

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Studies of Technetium Complexes. 9.† Use of the Tetrachloronitridotechnetate(VI) Anion for the Preparation of Nitrido Complexes of Technetium. Crystal Structure of Bis(8-quinolinethiolato)nitridotechnetium(V)

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The $Tc^{VI}NCl_4^-$ anion is shown to be a useful intermediate for the preparation of Tc≡N complexes. Reaction of $AsPh_4[TcNCl_4]$ with LiBr in acetone gives $AsPh_4[TcNBr_4]$, and reaction of $TcNCl_4^-$ with CsCl gives $Cs_2[TcNCl_5]$. The $TcNCl_4^-$ anion undergoes reduction on reaction with PPh_3, KNCS, $Na[S_2CNEt_2]$, and 8-quinolinethiol (C_9H_6NSH) to give the Tc^{V}≡N complexes $[TcNCl_2(PPh_3)_2]$, $[NEt_4]_2[TcN(NCS)_4(CH_3CN)]$, $[TcN(S_2CNEt_2)_2]$, and $[TcN(C_9H_6NS)_2]$, respectively. A single-crystal X-ray structure determination of bis(8-quinolinethiolato)nitridotechnetium(V), $[TcN(C_9H_6NS)_2]$, is reported. Crystals are monoclinic, space group $C2/c$, with a = 15.92 (1) Å, b = 7.347 (6) Å, c = 15.33 (2) Å, β = 110.89 (8)°, and Z = 4. Full-matrix least-squares refinement gave R = 0.029 for 1618 independent reflections. The coordination geometry of technetium is distorted square pyramidal with the apical Tc≡N bond of length 1.623 (4) Å. Tc-S and Tc-N(quinoline) bond distances are 2.3559 (7) and 2.135 (2) Å, respectively.

Introduction

Technetium(V) complexes containing the $Tc{=}O^{3+}$ core are well-known, and many of these have been prepared by ligand substitution reactions of $TcOX_4^-$ (X = Cl, Br).[1] The nitrido ligand (N^{3-}) is isoelectronic with the oxo ligand (O^{2-}) and is a powerful π-electron donor that tends to stabilize metals in high oxidation states. Mononuclear nitrido complexes appear to be formed most readily by the elements molybdenum, ruthenium, rhenium, and osmium.[2,3] The technetium(V) nitrido complexes $[TcN(S_2CNEt_2)_2]$[4] and $[TcNCl_2(PPh_3)_2]$[5] have been prepared by the reaction of TcO_4^- with hydrazine hydrochloride in the presence of the diethyldithiocarbamate anion and triphenylphosphine, respectively. The preparation of Tc^{V}≡N nitrido complexes by substitution reactions has, to date, been based on $[TcNCl_2(PPh_3)_2]$ as starting material.[5-8]

Recently, we have reported the preparation of the N-*n*-Bu_4^+ and $AsPh_4^+$ salts of the tetrachloro- or tetrabromonitridotechnetate(VI) anion ($TcNX_4^-$ (X = Cl, Br)) by the reaction of TcO_4^- with NaN_3 in the presence of concentrated HCl or concentrated HBr, respectively.[9] We now report some reactions of the $TcNCl_4^-$ anion that show that it is the agent of choice for the preparation of Tc≡N complexes. The X-ray crystal structure determination of bis(8-quinolinethiolato)nitridotechnetium(V) $[TcN(C_9H_6NS)_2]$ is reported. This is the first example of a metal nitrido complex containing the 8-quinolinethiolato (thiooxine) ligand and is pertinent to nuclear medicine since 8-quinolinol complexes of technetium-99m show promise as brain-uptake or as blood-labeling agents.[10]

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