Synthetic Models for Bis-Metallo Active Sites. A Porphyrin Capped by a Tetrakis(pyridine) Ligand System

Sir:

Recently, several binuclear metal complexes have been prepared in an attempt to model those enzymes which have two metal ions in their active sites[1] (hemerythrin,[2] superoxide dismutase,[3] type III copper proteins[4,5] (e.g., laccase, hemocyanin)), or those whose function involves an iron protoporphyrin IX residue interacting with an adjacent metal center[6-9] (cytochrome oxidases,[10] hydroxylases (P$_{450}$),[11] and electron-transfer systems[12]). Collman,[6] Ogoshi,[7] Kagan,[8] and their co-workers have prepared "face-to-face" or "strati-bis" metalloporphyrins in which two porphyrin residues are held in juxtaposition by flexible or nonflexible linkages. Alternatively, Chang[9] has "crowned" a porphyrin nucleus with a crown ether which is able to coordinate group 1a or 2a metal ions.

We report the synthesis and some properties of a simple bis-metallo system consisting of a tetrapyridine coordination arrangement joined via nonflexible links to a tetraphenylporphyrin. This arrangement, which is capable of considerable modification, allows the insertion of the same or different metal ions into the two different coordination sites. We are interested in using this type of binding to investigate metal–metal interactions, redox behavior, and rates of electron transfer in systems containing various metal ions held in axial proximity.

Treatment of *meso*-tetra[$\alpha,\alpha,\alpha,\alpha$-(o-aminophenyl)]porphyrin[13] with excess nicotinic anhydride[14] in CH$_2$Cl$_2$/pyridine gives 1 (P–N$_4$), isolated as the purple hydrate[15] in 70% yield

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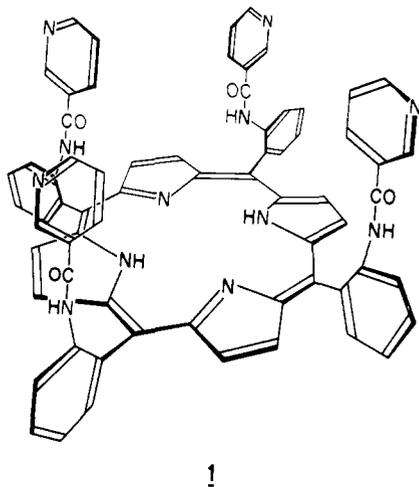
(homogeneous by TLC and HPLC; parent ion 1095; λ_{max} 423, 516, 548, 590, 645 nm in CHCl$_3$; ν_{CO} 1660 cm^{-1}). The ^{1}H NMR spectrum in Me$_2$SO-d_6 agrees with the $\alpha,\alpha,\alpha,\alpha$ con-

Scheme I
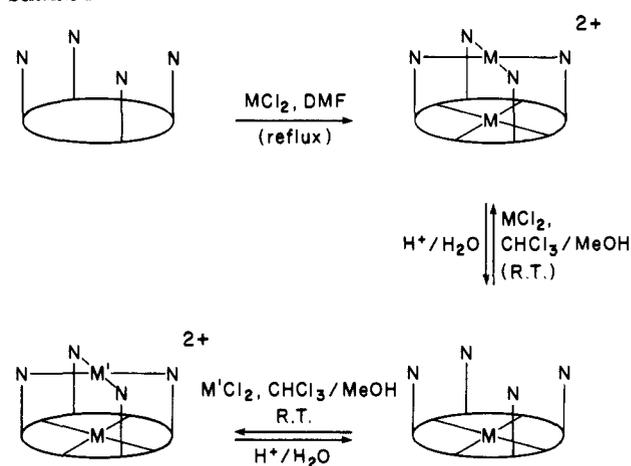
figuration, with pyrrole protons at δ 8.72 (sharp singlet), amide protons at 3.30 (singlet), 2'-, 4'-, 6'-, and 5'-pyridine protons at 9.65 (singlet), 8.08, 7.25 (doublets), and 6.68 (multiplet), respectively (parts per million from Me$_4$Si). The downfield shift of the 2'-H and upfield shifts of the 4'-, 5'- and 6'-H's compared with those of nicotinamide ($\Delta\delta$ + 0.57, −0.66, −0.88, and −0.99, respectively) are consistent with a configuration in which the pyridine N's are oriented outward.[16]

Treatment of P–N$_4$ with MCl$_2$ salts (M = Cu, Ni) in boiling DMF[17] results in the formation of the bis-metal complex cations [M$_1^{II}$(P)-M$_2^{II}$(N$_4$)]$^{2+}$, isolated as chloride salts (2a, 2c, Table I).[18,19] The bis-Cu^{II} complex acetate crystallizes as purple needles from CH$_2$Cl$_2$/hexane and a full x-ray study of this compound is in progress. The M$_2^{II}$ metal ion can be readily removed by treatment with dilute aqueous acids to give the [M$_1^{II}$(P)-(N$_4$)] nonelectrolyte (Scheme I) (2b, 2d, Table I)[20] and this process can be reversed by reinsertion of the metal ion at room temperature. Alternatively, a different metal salt can be used to give the mixed bis-metal systems [M$_1^{II}$(P)-M'$_2^{II}$(N$_4$)]$^{2+}$ and [M$_1^{II}$(P)-M'$_2^{III}$(N$_4$)]$^{3+}$; the cobalt(III) complex made in this way (2j, Table I) presumably results from air oxidation of the cobalt(II) species. Such interconversions have been repeated several times without displacement of the metal ion from the porphyrin nucleus, and the full ramifications of this type of metal ion insertion are still being explored. The bis-metal complexes of Table I are soluble in methanol and water-methanol mixtures as well as CHCl$_3$, CH$_2$Cl$_2$, acetone, and similar polar organic solvents, and can be easily separated from the monometallated complexes by TLC on silica gel (CHCl$_3$/MeOH 9:1).

The mono-Fe^{III} acetate complex (2f, Table I) was prepared by the ferrous acetate/acetic acid method,[21,22] (Scheme II) and the bis-metal complexes containing Cu^{II} and Fe^{III} have so far been prepared and characterized as given in the table;

Table I. Physical Properties of the Binuclear Complex Ions [M$_1^{II/III}$(P)–M$_2^{II/III}$(N$_4$)]$^{n+}$

Compd	M$_1$	M$_2$	Color	ESR parameters[a]			Electronic spectra[c]		
				$g_\parallel$	$g_\perp$	$A_\parallel$[b]	S	β	α
2a	Cu	Cu	Red	d	d	d	422	540	570
2b	Cu		Red	2.199	2.058	185	422	540	570
2c	Ni	Ni	Orange	e	e	e	410	523	552
2d	Ni		Orange	e	e	e	412	524	555
2e	Ni	Cu	Orange-red	2.273	2.050	155	414	526	552
2f	Fe^{III} g		Brown-green	2.00	6.06		420	502	571
2g	Fe^{II}		Red-brown	e	e	e	425	533	563
2h	Fe^{III} h	Cu	Brown-green	f	f	f	419	526	574
2i	Fe^{III}	Fe^{III}	Brown-green	f	f	f	417	505	572
2j	Cu	Co^{III}	Orange-red	2.200	2.065	186	421	540	570

[a] Frozen solution, CHCl$_3$, 103 K. [b] Units of 10^{-1} cm^{-1}. [c] CHCl$_3$ solution, nm. [d] Triplet spectrum—see text. [e] Diamagnetic. [f] No ESR signals observed at 103 K. [g] [Fe(P)OAc-(N$_4$)]; [Fe(P)Br-(N$_4$)], λ_{max} 422, 512, 580 nm. [h] [Fe(P)Cl-Cu(N$_4$)]Cl$_2$.
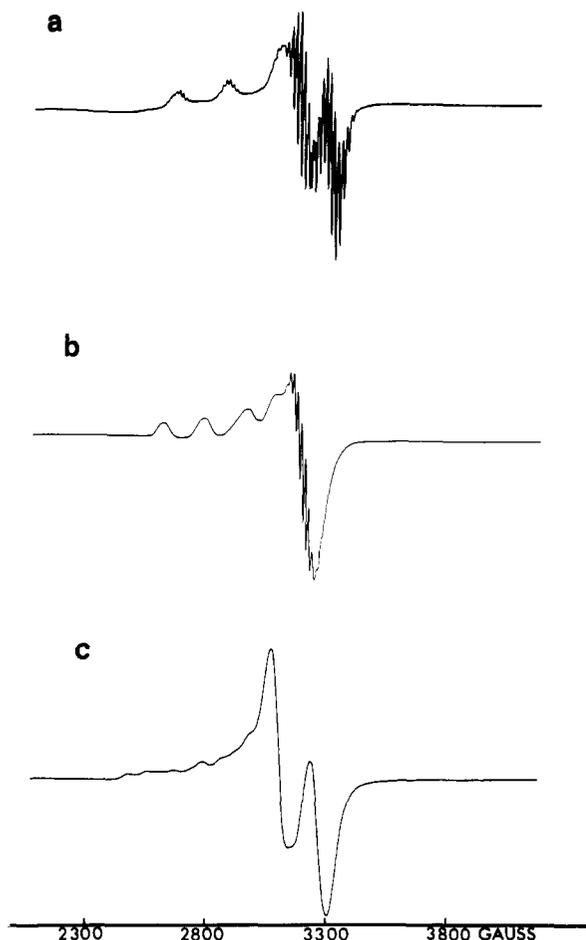
Figure 1. X-band EPR spectra of Cu complexes, 3mM in CHCl₃, frozen solutions, 103 K: (a) $[Cu^{II}(P)-(N_4)]$, **2b**; (b) $[Ni^{II}(P)-Cu^{II}(N_4)]Cl_2$, **2e**; (c) $[Cu^{II}(P)-Cu^{II}(N_4)]Cl_2$, **2a**.

Scheme II
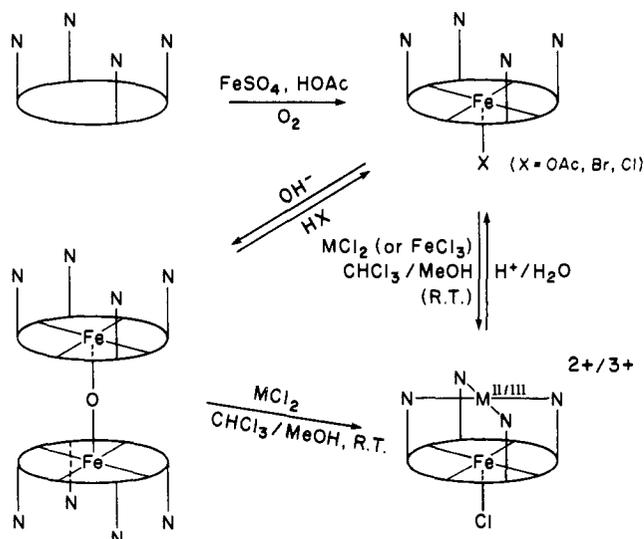
this system also appears capable of considerable extension. Treatment of a solution of the mono-Fe^III derivative in CHCl₃/MeOH with aqueous NaOH results in the μ-oxo dimer $[(N_4)-Fe(P)]_2O$ (λ_{max} 423, 575, 620 nm in CHCl₃/MeOH), which on treatment with MCl₂ salts is easily converted to the mixed bis-metal ion systems (Scheme II).[23] Alternatively, the μ-oxo dimer can readily be reconverted to $[(N_4)-Fe(P)X]$ (X = Cl, Br) by treatment with aqueous HCl or HBr.

The mono-Fe^II complex (**2g**, Table I) was obtained by dithionite reduction of the Fe^III–X derivative in toluene, benzene,

or chloroform solutions under N₂. Addition of excess (40×) 2-methylimidazole or 1-tritylimidazole gives the typical six-coordinate hemochrome (λ_{max} 426, 534, and 561 and 426, 533, and 563 nm, respectively, $\beta > \alpha$) and these complexes do not reversibly bind O₂ at ~20 °C, being completely oxidized within 15 min. A similar situation eventually occurs with the bis(pyridine) complex (λ_{max} 427, 531, 561 nm), although it is somewhat more stable, being <5% oxidized in 30 min at 20 °C (40× pyridine in benzene). Thus the uncomplexed N₄ "cap" does not prevent coordination of other good donors, nor its irreversible oxidation at ambient temperatures, and in this respect $[Fe^{II}(P)-(N_4)]$ resembles $Fe(\alpha,\alpha,\alpha,\alpha-T_{tos}PP)$[6] more than the "picket-fence" porphyrin $Fe(\alpha,\alpha,\alpha,\alpha-T_{piv}PP)$.[6]

The visible spectra of the bis-metallo systems (**2a, 2c, 2e, 2j,** Table I) are dominated by the mono metalloporphyrin chromophore with little or no change being observed on insertion of the metal into the "cap". However, the ESR properties show strong interactions between the metal centers. Thus, the ESR spectrum of the bis-Cu system (**2a**) (Figure 1c) shows marked Cu···Cu interactions, with the two $S = \frac{1}{2}$ states combining to produce a triplet $S = 1$ state split by the zero-field splitting parameter D.[24] The presence of the $\Delta M_s = \pm 2$ ("half-field") lines characteristic of the $S = 1$ state occurs at $g = 4.25$ in a polycrystalline sample of **2a** at 103 K. The Cu···Cu separation (R) was calculated at 5.9 Å using the expression for interacting dissimilar metal ions,[24]

$$D = -\beta^2[(g_{\parallel})^2 + \frac{1}{2}(g_\perp)^2]/R^3$$

where $g_{\parallel}$ and $g_\perp$ are the averaged values for the separate species $[g_{\parallel} = (g_{\parallel}^1 + g_{\parallel}^2)/2, g_\perp = (g_\perp^1 + g_\perp^2)/2]$ and were obtained from the spectra of $[Cu(P)-(N_4)]$ and $[Ni(P)-Cu(N_4)]^{2+}$ (Figure 1a and 1b), respectively (Table I, **2b, 2e**);[25] D was evaluated from the full-field perpendicular component of Figure 1c (154 G).[27] This inter-metal-ion separation is in excellent agreement with that estimated from Dreiding stereomodels, and one of the features of the amide bridges is their inflexibility to compression and twisting so that it is very likely that the geometry of the bis-metal complexes is rigid and will be maintained in solution. Titration of a CHCl₃/MeOH solution of the mono-Fe^III complex **2f** with CuCl₂ in methanol results in the gradual disappearance of the $g = 6$ signal (Table I, **2f**) and no signals due to Cu²⁺ are seen at $g \sim 2$; the spectrum is featureless after 1 equiv of Cu²⁺ has been added. This demonstrates strong coupling of the $S = \frac{5}{2}$ and $S = \frac{1}{2}$ spin systems in $[Fe(P)Cl-Cu(N_4)]^{2+}$. This behavior can be readily reversed by washing the CHCl₃ solution with dilute mineral acid.

These aspects, the full x-ray characterizations of the bis-copper and mono-Fe^III complexes and the electrochemical behavior of these systems are now being explored.

References and Notes

(1) For general reviews, see G. L. Eichhorn, Ed., "Inorganic Biochemistry", Vols. 1 and 2, Elsevier, New York, N.Y., 1973.
(2) J. M. Pratt in "Techniques and Topics in Bioinorganic Chemistry", C. A. McAuliffe, Ed., MacMillan, London, 1975, p 109; A. E. Martell in "Metal Ions in Biological Systems", Vol. 2, H. Sigel, Ed., Marcel Dekker, New York, N.Y., 1973, p 207.
(3) J. A. Fee and R. G. Briggs, *Biochim. Biophys. Acta*, **400**, 439 (1975); G. Kolks, C. R. Frihart, H. N. Rabinowitz, and S. J. Lippard, *J. Am. Chem. Soc.*, **98**, 5720 (1976).
(4) J. A. Fee, *Struct. Bonding (Berlin)*, **23**, 1 (1975).
(5) J. E. Bulkowski, P. L. Burk, M. F. Ludmann, and J. A. Osborn, *J. Chem. Soc., Chem. Commun.*, 498 (1977); W. D. McFadyen and R. Robson, *J. Coord. Chem.*, **5**, 49 (1976); A. W. Addison, *Inorg. Nucl. Chem. Lett.*, **12**, 899 (1976); J. S. de Courcy, T. N. Waters, and N. F. Curtis, *J. Chem. Soc., Chem. Commun.*, 572 (1977).
(6) J. P. Collman, C. M. Elliot, T. R. Halbert, and B. S. Tovrog, *Proc. Natl. Acad. Sci. U.S.A.*, **74**, 18 (1977).
(7) H. Ogoshi, H. Sugimoto, and Z. Yoshida, *Tetrahedron Lett.*, 169 (1977).
(8) N. E. Kagan, D. Mauzerall, and R. B. Merrifield, *J. Am. Chem. Soc.*, **99**, 5484 (1977).
(9) C. K. Chang, *J. Am. Chem. Soc.*, **99**, 2819 (1977).
(10) G. Palmer, G. T. Babcock, and L. E. Vickery, *Proc. Natl. Acad. Sci. U.S.A.*, **73**, 2206 (1977), D. C. Wharton in "Metal Ions in Biological Systems", Vol.

3, H. Sigel, Ed., Marcel Dekker, New York, N.Y., 1974, p 157.

(11) H. A. O. Hill, A. Röder, and R. J. P. Williams, *Struct. Bonding (Berlin)*, **8**, 123; G. S. Boyd and R. M. S. Smellie, Ed., "Biological Hydroxylation Mechanisms", Academic Press, New York, N.Y., 1972; I. C. Gunsalus, J. R. Meeks, J. D. Lipscomb, P. Debrunner, and E. Münck, "Molecular Mechanisms of Oxygen Activation", O. Hayaishi, Ed., Academic Press, New York, N.Y., 1973, Chapter 14.

(12) Reference 1, Chapters 19, 29; H. Sigel, Ed., "Metal Ions in Biological Systems", Vol. 6, Marcel Dekker, New York, N.Y., 1976.

(13) J. P. Collman, R. R. Gagne, C. A. Reed, T. R. Halbert, G. Lang, and W. T. Robinson, *J. Am. Chem. Soc.*, **97**, 1427 (1975).

(14) H. Rinderknecht and M. Gutenstein, "Organic Syntheses", Collect. Vol. V, Wiley, New York, N.Y., 1973, p 822.

(15) Anal. Calcd for $C_{68}H_{48}N_{12}O_4\cdot H_2O$: C, 73.36; H, 4.35; N, 15.10. Found: C, 73.27; H, 4.35; N, 14.96.

(16) The 2'-H is held in the deshielding region of the porphyrin macrocycle, while the 4'-, 5'-, and 6'-H's are held in the shielding region. See H. Scheer and J. J. Katz in "Porphyrins and Metalloporphyrins", K. M. Smith Ed., Elsevier, New York, N.Y., 1975, p 399.

(17) A. D. Adler, F. R. Longo, F. Kampas, and J. Kim, *J. Inorg. Nucl. Chem.*, **32**, 2443 (1970).

(18) Anal. Calcd for $C_{68}H_{44}N_{12}O_4Cu_2Cl_2\cdot 1.5CHCl_3$ (**2a**): C, 56.77; H, 3.12; N, 11.43; Cu, 8.64. Found: C, 56.61; H, 3.41; N, 10.84; Cu, 8.22. Anal. Calcd for $C_{68}H_{44}N_{12}O_4Ni_2Cl_2\cdot H_2O$ (**2C**): C, 62.85; H, 3.49; N, 12.93; Cl, 5.46. Found: C, 62.73; H, 3.19; N, 12.58; Cl, 5.45. The solvent molecules in these and related compounds proved difficult to remove; heating **2a** at 200 °C (0.001 mm) for 3 h failed to remove all the $CHCl_3$. Anal. Calcd for $C_{68}H_{44}N_{12}O_4Cu_2Cl_2\cdot 0.25CHCl_3$: C, 62.05; H, 3.37; N, 12.71. Found: C, 62.22, H, 3.60; N, 12.11. Similar difficulty in removing solvated molecules from the "picket-fence" porphyrins has been noted; see ref 13.

(19) Complex formation by the pyridine ligands in 1 involves rotation of the plane of the pyridine rings through 180°. This is seen in the upfield shift of the 2'-H and the downfield shift of the 4'-, 5'-, 6'-H's ($\Delta\delta$ −1.95, 0.03, 0.33, 0.35 ppm, respectively, compared with those of nicotinamide) in the ^{1}H NMR spectrum of **2c** (Me_2SO-d_8).

(20) Anal. Calcd for $C_{68}H_{44}N_{12}O_4Cu\cdot CHCl_3$ (**2b**): C, 64.94; H, 3.55; N, 13.17. Found: C, 65.01; H, 3.89. N, 13.02. M$^+$ is 1156.

(21) O. Warburg and E. Negelein, *Biochem. Z.*, **244**, 9 (1932).

(22) Anal. Calcd for $C_{68}H_{44}N_{12}O_4Fe\cdot OAc\cdot H_2O$: C, 68.57; H, 4.03; N, 13.71. Found: C, 68.65; H, 3.97; N, 13.56. This compound crystallizes from CH_2Cl_2/hexane as black cubes and in the triclinic space group *P*1; a full x-ray investigation is in progress.

(23) Isosbestic points were maintained during the titration of [(N_4)–Fe(P)]$_2$O with $CuCl_2$ solution in MeOH (512, 541 nm); an isosbestic point was also observed during the titration of [Fe(TPP)]$_2$O with $CuCl_2$ solution in MeOH (550 mm) to give Fe(TPP)Cl.

(24) For a full description of the ESR characteristics of dimeric transition metal ion complexes, see T. D. Smith and J. R. Pilbrow, *Coord. Chem. Rev.*, **13**, 173 (1974), and references therein.

(25) The ESR spectrum of [Cu(P)–(N$_4$)] (Figure 1a) closely resembles that for Cu(TPP)^{26} with Cu hyperfine and N superhyperfine splittings in both the perpendicular and parallel regions; that for [Ni(P)–Cu(N$_4$)]Cl$_2$ (Figure 1b) is typical of Cu^{II} in a pseudo-square-planar field with resolution of the parallel, but not the perpendicular, components and some N superhyperfine interaction in the perpendicular region.

(26) P. T. Manoharan and M. T. Rogers, "Electron Spin Resonance of Metal Complexes", Plenum Press, New York, N.Y., 1969, p 143.

(27) The estimate for *D* of 154 G and the calculation of *R* of 5.9 Å assumes axial symmetry and ignores pseudodipolar contributions^{28} in [Cu(P)–Cu(N$_4$)]$^{2+}$.

(28) N. D. Chasteen and R. L. Belford, *Inorg. Chem.*, **9**, 169 (1970).

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Received October 20, 1977

Photoinduced Coupling Reaction of 5-Bromouridine to Tryptophan Derivatives[1]

Sir:

The replacement of thymine in DNA by 5-bromouracil sensitizes bacterial and mammalian cells to the lethal effects of UV light.[2] The photochemical mechanism responsible for this sensitizing effect has been studied extensively, and at least three possible mechanisms have been suggested: (1) self-coupling of two 5-bromouracil residues with formation of 5–5'-diuracilyl linkages;[3] (2) induction of single-strand breaks in DNA;[2b,4] (3) enhancement in the rate of production of DNA–protein cross-links in cells.[5] Recently, DNA substituted with bromouracil has been reported to undergo photoinduced cross-linking to RNA polymerase[6] and to *lac* repressor.[6,7] In spite of the importance of the cross-linking of DNA containing 5-bromouracil to proteins,[8] very little is known about the nature
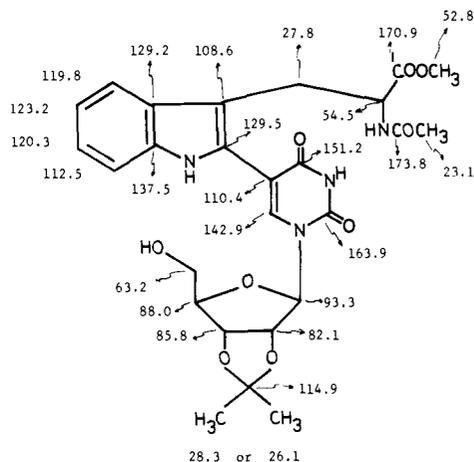
Figure 1. Structure of **3** as determined by ^{13}C NMR in Me_2CO-d_6. Chemical shifts are in parts per million from Me_4Si.

of the amino acid–nucleic acid adducts. Sulfhydryl compounds such as cysteine and glutathione have been reported to undergo photoaddition with 5-bromouracil.[9] We now wish to report that N^b-acetyltryptophan methyl ester, a model for tryptophan in a protein, undergoes a photoreaction with 5-bromouridine or 5-bromo-1,3-dimethyluracil to give the corresponding coupled product in a highly regiospecific fashion.[10] Neither *N*-acetyltyrosine methyl ester nor *N*-acetylhistidine methyl ester undergoes such a coupling reaction.

Acetone-sensitized irradiation[12] of 2',3',*O*-isopropylidene-5-bromouridine (**1**, 1.4 mM) in acetone–acetonitrile (1:3) in the presence of N^b-acetyltryptophan methyl ester (**2**, 3.5 mM) produced a single photoproduct. No other products, except the unreacted starting materials **1** and **2**, were detected on TLC. Separation by column chromatography on silica gel yielded **3**, mp 158–162 °C dec, in 70% yield. Spectral properties,[13] including the ^{13}C NMR spectrum[14] (Figure 1), are in accordance with the assigned structure.

Under similar conditions, acetone-sensitized irradiation[12] of 5-bromo-1,3-dimethyluracil (**4**, 1.5 mM) and **2** (3.5 mM) in acetonitrile gave rise to the coupled product **5**[15] (67%) as the sole product.[16] Quantum yield for the formation of **5** is 0.018.[17] In control runs, irradiation of a solution of **4** and **2** in acetonitrile in the absence of acetone did not produce **5**, and both starting materials were recovered unchanged. Direct irradiation of **4** (2.0 mM) and **2** (4.6 mM) in acetonitrile with 254-nm light resulted in the formation of the debrominated product 1,3-dimethyluracil (**6**, 75%) as the major product, together with minor amounts of **5** (15%).[19] Addition of 1,3-pentadiene to the system inhibited the formation of the coupled product **5**, but had no significant effect on the formation of **6**. The bromouracil derivative **4** undergoes regiospecific coupling reaction with various indolic compounds. For example, ace-