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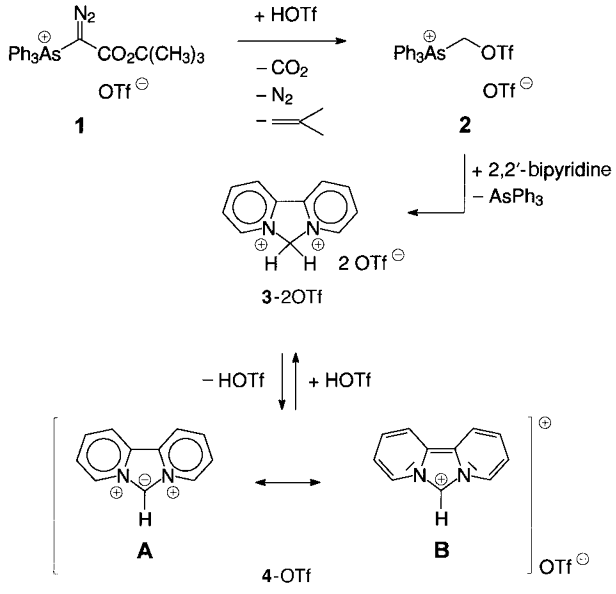
Generation and Trapping Reactions of a Formal 1:1 Complex between Singlet Carbon and 2,2′-Bipyridine[**]

Robert Weiss,* Silvia Reichel, Matthias Handke, and Frank Hampel

Dedicated to Professor George A. Olah
on the occasion of his 70th birthday

2,2′-Bipyridine is a classical chelating ligand for metals as well as nonmetals in different oxidation states and bonding types.[1,2] However, to our knowledge analogous complexes are unknown for all oxidation states of carbon. Here we report a first compound of this kind. In 1994 we described the first S$_N$ reactions at the α-carbon atom of aryliodonio diazo compounds.[3] Using AsPh$_3$ as nucleophile, we obtained the diazo compound **1**, which served as starting material for the new class of compounds.

The reaction of **1** with trifluoromethanesulfonic acid (HOTf, Scheme 1) afforded the arsonium salt **2** in high yield as the result of an acid-induced fragmentation of the *tert*-butyl ester functionality, followed by a proto-dediazonation.[4] Compound **2** is a potent 1,1-bis(electrophile) which reacted with a host of neutral nucleophiles to form symmetrical and unsymmetrical geminal bis(onio)-substituted salts.[5] Accordingly, the reaction of **2** with 2,2′-bipyridine provided the cyclic bis(onio)-substituted salt **3**-2OTf, a bis(azonia) analogue of fluorene. Under the reaction conditions, this C–H-acidic



Scheme 1. Synthesis of **3**-2OTf and its deprotonation to **4**-OTf.

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compound was converted into **4**-OTf with excess 2,2′-bipyridine (Scheme 1) and isolated as **4**-Br after anion exchange (yield: 75 % relative to **2**); this salt crystallizes more readily.[6] Figure 1 shows the result of the X-ray structure analysis of **4**-Br·$\mathrm{H_2O}$.[7]

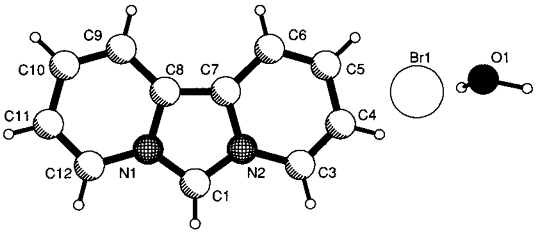
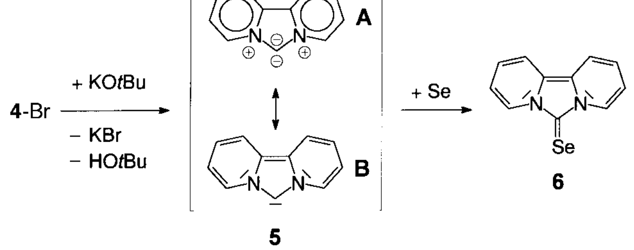


Figure 1. Structure of **4**-Br·$\mathrm{H_2O}$ in the crystal. Important bond lengths [Å] and angle [°]: C1–N1 1.338, N1–C8 1.394, C7–C8 1.387, C8–C9 1,41, C9–C10 1.34, C10–C11 1.41, C11–C12 1.33, C12–N1 1.39; N1-C1-N2 107.6.

A noticeable feature in the ^{1}H NMR spectra of **4**-Br and **4**-OTf is the large ^{1}H – ^{13}C coupling constant (232.59 Hz) for the C–H bond in the five-membered ring. This coupling constant almost corresponds to sp hybridization of the C atom in this bond (the exact value is sp$^{1.15}$).[8] A PM3 calculation, combined with an NBO analysis,[9] provided a hybridization of sp$^{1.23}$. This unprecedented result can also be reached qualitatively by applying Bent's rules:[10] The carbon atom bridging the six-membered rings in **4** ("bridging C atom") has two strongly electronegative substituents (the N centers), which should induce a higher p character in the C hybrid orbitals associated with them. This automatically leads to a higher s proportion in the remaining σ orbital of the bridging C atom with a trigonal-planar environment.

With an excess of HOTf in THF, **4**-Br was quantitatively converted into the dicationic salt **3**-2 OTf, the first σ complex of an S_E reaction at an imidazolium system to be isolated. The ^{1}H – ^{13}C coupling constant of 170.65 Hz for the geminal C–H bonds corresponds to sp$^{1.93}$ hybridization in these bonds, which is highly unusual for a saturated C atom.[8]

Salt **4**-Br was deprotonated to the neutral species **5** in a fast and quantitative reaction with KO*t*Bu in THF at $-30\,^{\circ}$C (Scheme 2). Under these reaction conditions, **5** was stable for several hours. The ^{13}C NMR spectrum of the reaction mixture shows five signals in the range of $\delta = 114-129$ and a sixth signal at $\delta = 196.41$ for the bridging C atom. This value is shifted upfield only slightly with respect to the corresponding signals for Arduengo carbenes[11] and, according to research by Chen et al. on dimerized Arduengo carbenes,[12] cannot be ascribed to a dimer of **5**. Compound **5** can be trapped in high yields by a number of electrophiles.[13] For example, the reaction with selenium led to the chalcogenone **6**, which was characterized by X-ray structure analysis (Figure 2).[14]



Scheme 2. Subsequent reactions of **4**-Br.

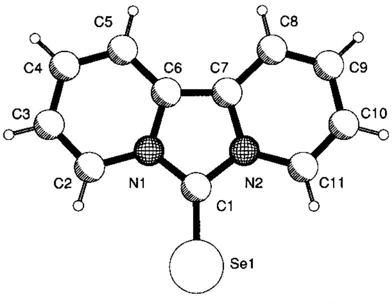
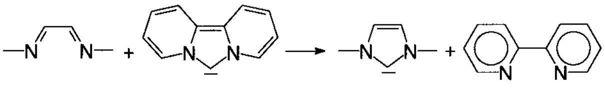


Figure 2. Structure of **6** in the crystal. Important bond lengths [Å] and angle [°]: Se1–C1 1.826, C1–N1 1.364, C1–N2 1.378, N1–C6 1.39, C6–C7 1.377, N1–C2 1.39, C2–C3 1.34, C3–C4 1.41, C4–C5 1.34, C5–C6 1.41; N1-C1-N2 103.8.

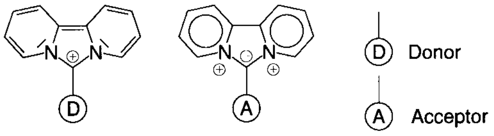
Compound **6** is characterized by a large degree of localization of the double bonds of the hydrocarbon moiety and partial delocalization in the selenourea structural element. The C(Se)–N bonds are longer and the C–Se bond is shorter in **6** than in an open-chain selenourea (each by about 0.04 Å).[16] This indicates that the hybridization of the bridging C atom corresponds to that in **4**-Br.

The two-step deprotonation of dication **3** leads to a far-reaching electronic reorganization. Whereas **3** contains two aromatic rings, **4** (in **4**-Br·$\mathrm{H_2O}$, see Figure 1) is characterized by resonance structure **4B** (see Scheme 1) with a reduced bipyridine subsystem. Further deprotonation of **4** should then lead to an even more strongly localized π system, which according to ab initio calculations (3-21G*)[17] is better represented by the resonance structure **5B** (typical of arduengo carbenes) than by the bis(ylide) form **5A** (see Scheme 2). The latter is probably relevant for the description of excited states and gives an indication of the structural relationship of this class of compounds with "true" bis(ylides) such as carbodiphosphoranes. These were recognized early on as formal complexes of an excited singlet C atom with two molecules of triorganophosphane as stabilizing donor ligands.[18] By analogy, **5** can be seen as a chelate complex of such a C atom and 2,2′-bipyridine. Model calculations (3-21G*) for C1-transfer reactions with suitable reaction partners show that this view, beyond formal considerations, is supported by thermodynamics. Therefore, **5** should react according to the isodesmic Equation (1) with *N,N′*-dimethyl-1,4-diazabutadienein a highly exothermic reaction ($\Delta H_f = -51.06\,\mathrm{kcal\,mol^{-1}}$) to the corresponding stable imidazol-2-ylidene and 2,2′-bipyridine. This should render **5** much more suitable for C1 transfer than Arduengo carbenes. Kinetic factors should be decisive for the experimental realization.



(1)

When an electrophile is coordinated to the nucleophile **5**, two electronic limiting cases can be distinguished for the resulting adducts: If the substituent at the bridging C atom shows donor character, resonance structure **7** dominates (e.g. selone **6**). If, on the other hand, the substituent is an acceptor, the adduct is characterized by resonance structure **8** with complementary polarization. We have already experimentally verified this structural classification.[19] Introducing carbene **5** into organic and inorganic substrates thus creates a type of ligand which optimally adapts to the electronic needs of the substrate. The two-step bipyridinium redox system of **5**, which is immanent in the system, can flexibly fulfill the electronic demands on the bridging C atom with its reduced (situation in **7**) and oxidized forms (situation in **8**) as a built-in umpolung reagent. Corresponding adducts of Arduengo carbenes do not show a comparable electronic flexibility.



7 **8**

Experimental Section

All reactions were carried out under a nitrogen atmosphere with anhydrous, N_2-saturated solvents in Schlenk vessels. Correct elemental analyses (C,H,N) are available.

2: Diazo compound **1** (4.560 g, 7.65 mmol) was dissolved in CH_2Cl_2 (20 mL), and HOTf (0.71 mL, 8.0 mmol) added. After gas formation had ceased the mixture was stirred for 36 h at room temperature; the color became brown-red in the process. The solution was then concentrated under vacuum to about 5 mL. The white product was precipitated with Et_2O (150 mL), isolated by filtration, washed with Et_2O (25 mL), and dried under vacuum for 2 h; yield: 4.303 g (91 %). ^{1}H NMR (400 MHz, $CDCl_3$): $\delta = 6.52$ (s, 2H; CH_2), 7.73 (m, 12H; C_6H_5), 7.82 (m, 3H; C_6H_5); ^{13}C NMR (100 MHz, $CDCl_3$): $\delta = 68.51$ (CH_2), 118.13 (C1), 118.16 (q, ^1J(C,F) = 320 Hz; $F_3CSO_3CH_2$), 120.47 (q, ^1J(C,F) = 320 Hz; $F_3CSO_3^-$), 131.24 (C3/C5), 133.05 (C2/C6), 135.10 (C4); ^{19}F NMR (470 MHz, $CDCl_3$, C_6F_6): $\delta = -73.07$ ($F_3CSO_3CH_2$), -78.62 ($F_3CSO_3^-$).

4-Br: A solution of **2** (1.208 g, 1.95 mmol) in CH_3CN (60 mL) was treated with 2,2′-bipyridine (0.671 g, 4.30 mmol) and heated at reflux for 5 d. After removal of the solvent under vacuum the residue was stirred for 2 h in Et_2O (50 mL), filtered, washed with Et_2O (15 mL), and dried for 2 h under vacuum. The solid was dissolved in CH_2Cl_2 (50 mL), and a solution of Bu_4NBr (1.290 g, 4.00 mmol) in CH_2Cl_2 (10 mL) added. After 2 h the mixture was filtered to remove insoluble components. A yellow-brown solid was then precipitated with Et_2O (100 mL), isolated by filtration, washed with Et_2O (20 mL), and dried under vacuum for 2 h. Crystallization from EtOH afforded dark yellow **4**-Br; yield: 0.365 g (75 %). Crystals suitable for X-ray structure analysis formed in a supersaturated solution in EtOH at ambient temperature. ^{1}H NMR (400 MHz, [D$_6$]DMSO): $\delta = 7.56$ (m, 4H), 8.69 (m, 2H), 9.07 (m, 2H), 10.40 (s, 1H); ^{13}C NMR (100 MHz, [D$_6$]DMSO): $\delta = 116.01$ (bridging C atom), 118.76, 120.38, 121.71, 122.65, 123.34.

3-2 OTf: Salt **4**-Br (0.295 g, 1.18 mmol) was suspended in (THF) 20 mL, and HOTf (0.23 mL, 2.61 mmol) added at -20 °C. After 18 h the mixture was filtered. The white solid isolated was washed with THF (10 mL) and Et_2O (10 mL), and was dried under vacuum for 2 h; yield: 0.497 g (90 %). ^{1}H NMR (400 MHz, CD_3CN): $\delta = 7.41$ (s, 2H; CH_2), 8.41 (m, 2H), 8.97 (m, 4H), 9.35 (m, 2H); ^{13}C NMR (100 MHz, CD_3CN): $\delta = 77.65$ (CH_2), 120.72 (q, ^1J(C,F) = 320 Hz; CF_3), 125.28, 131.35, 142.95, 144.21, 150.34.

5: KO*t*Bu (0.065 g, 0.58 mmol) was added to a suspension of **4**-Br (0.097 g, 0.39 mmol) in THF (3 mL) at -30 °C. After 30 min the inorganic components (KBr, excess KO*t*Bu) were removed by filteration over Celite at -60 °C. The orange filtrate was examined with NMR spectroscopy. ^{13}C NMR (100 MHz, THF, -30 °C, [D$_6$]acetone): $\delta = 114.12$, 117.44, 117.64, 121.83, 128.72, 196.41.

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[14] Crystal structure data for **6**: $C_{11}H_8N_2Se$, $M_r = 247.15$, triclinic, space group $P1$, a = 7.163(2), b = 9.054(2), c = 9.070(2) Å, $\alpha = 93.46(3)$, $\beta = 112.74(3)$, $\gamma = 112.69(3)°$, $Z = 2$, $V = 485.1$ Å^3, $\rho_{calcd} = 1.692$ g cm^{-3}; yellow plates, crystal dimensions: $0.25 \times 0.10 \times 0.10$ mm^3. The data were collected on a Nonius Mach3 single-crystal diffractometer with monochromated (graphite) $Mo_{K\alpha}$ radiation ($\lambda = 0.71073$) at T = 293(2) K and a scan range of $3.09° < \theta < 26.29°$. The absorption correction was carried out with ψ scans. Of 2105 reflections measured, 1975 were independent and 1198 with $I > 2\sigma(I)$. The structure was solved with direct methods (SHELXS 86). The 127 parameters were refined on F^2 with all data according to the least-squares method using SHELXL 93 (G. M. Sheldrick, Göttingen, **1993**). The hydrogen atoms were fixed in idealized positions with a riding model; R values of refinement: $R1 = 0.0376$ for $I > 2\sigma(I)$ and $wR2 = 0.1110$ for all data; max./min. residual electron density: 0.318/ -0.392 e Å^{-3}.[15]

[15] Crystallographic data (excluding structure factors) for the structures reported in this paper have been deposited with the Cambridge Crystallographic Data Centre as supplementary publication no. CCDC-100542. Copies of the data can be obtained free of charge on application to CCDC, 12 Union Road, Cambridge CB2 1EZ, UK (fax: int. code + (44) 1223-336033; e-mail: deposit@ccdc.cam.ac.uk).

[16] *N,N'*-Dimethylselenourea: C–Se 1.866, C(Se)–N 1.32 Å: H. M. K. K. Pathirana, T. J. Weiss, J. H. Reibeneis, R. A. Zingaro, E. A. Meyers, *Z. Kristallogr.* **1994**, *209*, 697.
[17] With the length of the [0] bridge between the six-membered rings, calculated with ab initio methods, as an indicator: **3**: 1.461, **4**: 1.379, **5**: 1.353 Å.
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[19] We will report on compounds of this type separately.

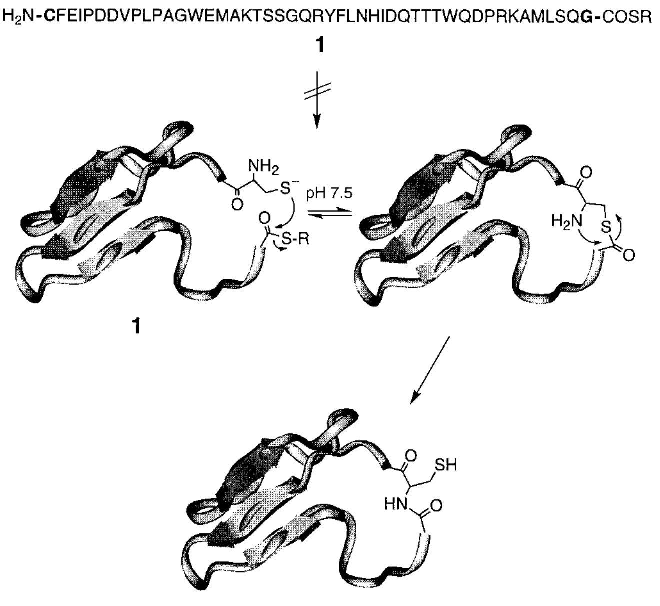
Chemical Synthesis of a Circular Protein Domain: Evidence for Folding-Assisted Cyclization[**]

Julio A. Camarero, Joanna Pavel, and Tom W. Muir[*]

The chemical-ligation approach to the condensation of peptide fragments offers a unique route for generating proteins possessing both natural[1] and unnatural topology.[2] Of the various architectures possible, one of the most fascinating from the perspective of both protein engineering and protein folding is the circular topology.[3] Although head-to-tail cyclization is commonly used to constrain the structure of small peptides,[4] relatively little is known about the affect of this modification on the structure and function of a protein. Indeed, the bulk of our current knowledge is based on the pioneering work of Creighton and co-workers on a cyclized version of bovine pancreatic trypsin inhibitor (BPTI) prepared by random chemical cross-linking.[3] Here we demonstrate that a backbone-cyclized protein domain, prepared by an intramolecular version[5] of Kent's native chemical ligation,[6] is able to spontaneously fold into a functional, native-like conformation. In addition, we provide evidence that the rate of cyclization is related to the folded state of the protein.

Our initial studies on protein cyclization focussed on the WW domain from the human Yes kinase-associated protein (YAP).[7] This protein domain possesses a globular structure composed of a three-stranded antiparallel β sheet which positions the flexible N and C termini in close proximity.[8] Given that native chemical ligation is tolerant to the presence of chemical denaturants,[6] it was anticipated that insights into the effect of this juxtaposition on the chemical reaction rate would be forthcoming.

The strategy used to synthesize the circular WW domain is shown in Scheme 1. Key to our approach was the generation of the linear polypeptide precursor **1**, which contains both groups necessary for native chemical ligation: an N-terminal cysteine residue and a C-terminal thioester group.[9] Cyclization of fully unprotected **1** to give the circular WW domain **2** was remarkably clean and extremely rapid (Figure 1). The ligation product was characterized as **2** by a combination of



2

Scheme 1. Synthesis of the circular WW protein domain **2**.

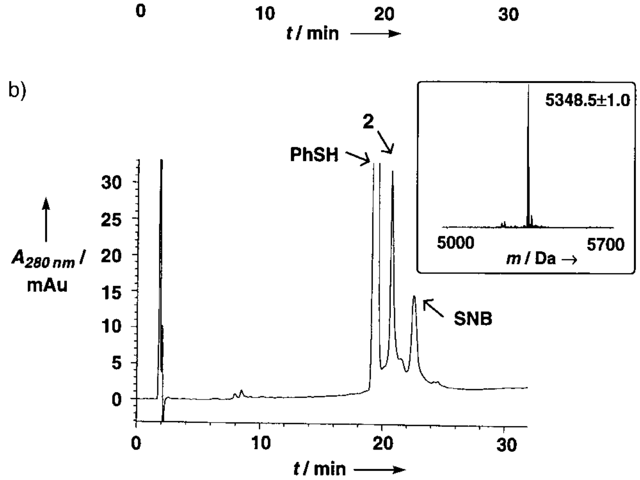


Figure 1. a) Analytical HPLC chromatogram of the purified linear precursor **1**. b) The crude ligation mixture after 2 min of reaction on the same scale as in a). mAu = 10^{-3} of full scale absorbance units. SNB = *S*-(5-sulfenyl-2-nitrobenzoic acid), the by-product. Inset: ES-MS of purified **2**.

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