An Aziridinium Imide Intermediate in the Ene Reaction of *trans*-Cycloheptene and *N*-Methyl-1,2,4-triazoline-3,5-dione

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The ene reaction of *N*-alkyl-1,2,4-triazoline-3,5-diones has generated interest both for its synthetic usefulness[1] and for its interesting mechanism.[2] Isotope-effect investigations have concluded that an aziridinium imide (AI) intermediate is involved in the reaction and further that this intermediate must be involved in a prior equilibrium with other isomers or zwitterions.[3] We report here experimental proof of this mechanism by direct observation of an aziridinium imide intermediate and the rate of conversion of this species to ene products.

An AI intermediate has been previously observed, but because elimination could not occur, it went on to form the four-membered-ring product, a diazetidine.[4] The hypothesis of prior equilibration of these intermediates rests on the observation of trans-isotope effects of labeled 2-butenes[3] and the observation of isomerization in related reactions of *N*-alkyl-1,2,4-triazoline-3,5-diones with 2,4-hexadiene[5] and 1,4-di-*tert*-butoxy-1,3-butadiene[6] isomers. We have recently reported on the production and spectroscopic characterization of *trans*-cycloheptene.[7] Here we examine the ene reaction of *N*-methyl-1,2,4-triazoline-3,5-dione (MTAD) with the reactive double bond of this strained species.

When a 0.25 M solution in dimethyl-d_6 ether of *cis*-cycloheptene with 0.01 M methyl benzoate is photolyzed with broad-band irradiation in a Pyrex NMR tube ($\lambda > 285$ nm) at −135 °C, a 1:5 mixture of the trans and cis isomers is produced.[7] When a cold (−140 °C) solution of 0.9 equiv (relative to *trans*-cycloheptene) of MTAD in dimethyl-d_6 ether is added to the still cold NMR tube, a series of new, rather broad resonances in the ^{1}H NMR spectrum appear at 3.68, 2.98, 2.12, 1.90, and 1.82 ppm (Figure 1). These peaks grow in with a concomitant decrease in the resonances associated with *trans*-cycloheptene. The NMR spectrum of this intermediate suggests a symmetrical structure and is consistent with either a cis-fused diazetidine or an aziridinium imide structure. This intermediate cannot involve a trans-fused cycloheptane ring because this would be expected to adopt an unsymmetrical structure similar that found for *trans*-cycloheptene itself.[7] The symmetric structure is also implied by the ^{13}C spectrum, which shows peaks at 23.16, 25.79, 25.79, 32.45, 56.00, 159.42, and 160.52 ppm.[8] These resonances show additional broadening as the temperature is lowered from −115 to −135 °C. The presence of two carbonyl carbons rules out a cis-fused diazetidine as this intermediate, and ^{13}C ^{1}H-coupled spectra show a 179-Hz coupling constant for the hydrogen attached to the bridgehead carbon at 56.00 ppm. This coupling constant is substantially larger than those observed in diazetidines[5] and implies the presence of the three-membered ring of an aziridinium imide.

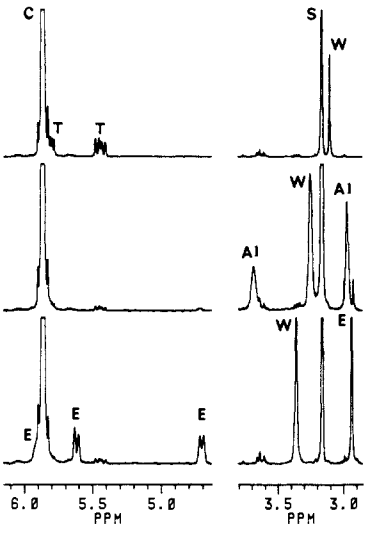
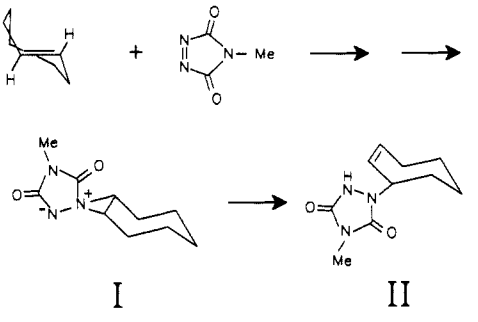


Figure 1. Top: 400-MHz ^{1}H NMR spectrum at −135 °C in dimethyl-d_6 ether of 1:5 *trans*:*cis*-cycloheptene mixture. Trans and cis peaks are labeled with T and C, respectively. Middle: After addition of MTAD solution at −135 °C. Aziridinium imide (I) peaks are labeled with AI. Bottom: After warming to −85 °C for 20 min. Ene product (II) peaks are labeled with E. The peak of variable chemical shift is water (W) while the peak at 3.16 ppm is solvent (S).

Scheme I



There are four possible isomers of an aziridinium imide cis fused to a cycloheptane ring, two pairs of ring-flip-related conformers. While our NMR results cannot distinguish between these, molecular mechanics calculations[9] on the four protonated AIs suggest that the exo, exo isomer (I) pictured in Scheme I is lower in energy by at least 4.5 kcal/mol when compared to the other isomers. While most of the *trans*-cycloheptene has reacted before the tube could be transferred to the NMR spectrometer and a spectrum could be taken, the rate of appearance of the aziridinium imide could just be monitored by taking quick successive spectra at −135 °C. The rate found indicates a maximum activation energy of 9.7 kcal/mol for the production of I. Upon further warming of the solution, additional changes occur. At −85 °C, species I slowly disappears and resonances at 9.36, 5.92, 5.61, 4.70, 2.92 and several between 2.3 and 1.1 ppm in the ^{1}H spectrum (Figure 1) and 24.91, 27.24, 29.20, 30.07, 33.08, 58.90, 133.24, 134.13, 156.55, and 156.77 ppm in the ^{13}C spectrum grow in. This spectrum is that of species II, the ene product of MTAD with *cis*-cycloheptene. This observation offers the first direct proof of the intermediacy of aziridinium imides in the ene reaction of *N*-alkyltriazolidine-3,5-diones with alkenes. The rate of appearance of the ene could be followed and leads to an activation energy of 13.4 ± 0.1 kcal/mol for the breakdown of I.

Isotope effects on the reaction of *N*-phenyl-1,2,4-triazoline-3,5-dione and labeled 2-butenes were interpreted as demanding

(1) (a) Barton, D. H. R.; Shiori, T.; Widdowson *J. Chem. Soc.* C **1971**, 1968. (b) Corey, E. J.; Snider, B. B. *Tetrahedron Lett.* **1973**, 3091. (c) Snider, B. B.; Corcoran, R. J.; Breslow, R. *J. Am. Chem. Soc.* **1975**, *97*, 6580. (d) Eyley, S. C.; Williams, D. H. *J. Chem. Soc., Perkin Trans. 1* **1976**, 731.

(2) (a) Ohashi, S.; Leong, K.; Matyjaszewski, K.; Butler, G. B. *J. Org. Chem.* **1980**, *45*, 3467. (b) Hoye, T. R.; Bottorff, K. J.; Caruso, A. J.; Dellaria, J. F. *J. Org. Chem.* **1980**, *45*, 4287. (c) Seymour, C. A.; Greene, F. D. *J. Am. Chem. Soc.* **1980**, *102*, 6384. (d) Cheng, C. C.; Seymour, C. A.; Petti, M. A.; Greene, F. D.; Blount, J. F. *J. Org. Chem.* **1984**, *49*, 2910.

(3) (a) Orfanopoulos, M.; Foote, C. S.; Smonou, I. *Tetrahedron Lett.* **1987**, *28*, 15. (b) Orfanopoulos, M.; Smonou, I.; Foote, C. S. *J. Am. Chem. Soc.* **1990**, *112*, 3607.

(4) Nelsen, S. F.; Kapp, D. L. *J. Am. Chem. Soc.* **1985**, *107*, 5543.

(5) Jensen, F.; Foote, C. S. *J. Am. Chem. Soc.* **1987**, *109*, 6376.

(6) Clennan, E. L.; Earlywine, A. D. *J. Am. Chem. Soc.* **1987**, *109*, 7104.

(7) Squillacote, M.; Bergman, A.; DeFelippis, J. *Tetrahedron Lett.* **1989**, *30*, 6805.

(8) The accidental equivalence of the two peaks at 25.79 ppm was revealed in the ^{13}C ^{1}H-coupled spectrum, which showed that a methylene and the *N*-methyl carbon have this resonance frequency. When toluene-d_8 is added to the cooled solution, these peaks separate into two resonances at 25.1 and 25.6 ppm.

(9) (a) Allinger, N. L.; Hindman, D.; Helmut, H. *J. Am. Chem. Soc.* **1977**, *99*, 3282. (b) The PCMODEL/MMX program that was used is available from Serena Software, Bloomington, IN.

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an interconversion of AI intermediates presumably through a zwitterionic species.[3] Our results also support this conclusion since we do not see any trans-fused intermediates and only one of the four possible cis-fused AI intermediates. It is possible that a trans-fused AI intermediate never forms and that an initially formed zwitterionic intermediate collapses down to the cis-fused AI. It is unlikely, however, that the collapse of the zwitterion would produce only one of the possible AIs, and it is more probable that equilibration among the possible AI intermediates is occurring to produce the isomer with the least steric strain, I. This equilibration may also explain the broadening in the resonances of I.

Registry No. I, 126156-51-2; I (conjugate acid), 126652-65-1; II, 126156-52-3; *N*-methyl-1,2,4-triazoline-3,5-dione, 13274-43-6; *trans*-cycloheptene, 45509-99-7.

Radiometallacarboranes as Tumor Imaging Reagents

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Monoclonal antibodies (Mab), when conjugated with bifunctional chelation reagents containing a radiometal, have provided sensitive and accurate imaging agents for the detection of cancer and other diseases.[1] The bifunctional chelates presently in use are generally of the aminocarboxylate family and subject to catabolism with release of metal ion in vivo. We have now designed, synthesized, and evaluated a functionalized cluster containing a radiotransition metal (venus flytrap cluster, VFC) which makes use of an inorganic ligand set, incorporates exceedingly strong cluster bonding based upon a bridged *commo*-bis(dicarbollide) structure,[2] and can be prepared in the aqueous media[3] commonly used to supply radiometal salts. The species reported here presages the existence of a large family of functionalized metallacarborane clusters which may serve as biologically inviolable radiotransition-metal carriers for the antibody-mediated γ-imaging or β-therapy of tumors.

Scheme I presents the synthesis of the diastereomeric VFC precursors and their conversion to the corresponding isomers of the very stable Co^{3+}-VFC-COOH (*dl*- and *meso*-**4**). The synthesis of the diastereomers of **2** from *closo*-1,8-$C_2B_9H_{11}$[4] and the anion of 4-carbomethoxypyrazole was suggested by the well-known nucleophilic opening of this carborane by reaction with electron-pair donors to produce *nido*-10-substituted-7,9-$C_2B_9H_{11}$ species[4] having a weakly acidic bridge hydrogen atom between BH vertices 10 and 11. Conversion of **2** to the formal 7,9-bis(dicarbollide) intermediates, **3**, and thence to **4** involves the loss of both of the B-H-B bridge protons present in **2** and hydrolysis of the carbomethoxyl function in the presence of Co^{3+} and aqueous KOH at pH 13 (15 min at 100 °C). Acidification and extraction with diethyl ether affords a mixture of *dl*- and *meso*-**4** in up to 50% combined yields. The scope of this reaction sequence has been investigated by using precursors derived from unsubstituted pyrazole anion combined with Ni^{3+} and Cu^{3+} as well as Co^{3+} present in the VFC products. The structures of the chromatographically separated *meso*-Cu^{3+}, *meso*-Ni^{3+}, and *dl*-Co^{3+} have been determined crystallographically and the structures of the Co^{3+} isomers correlated with their ^1H and ^{11}B FT NMR spectra. These results will be reported elsewhere.

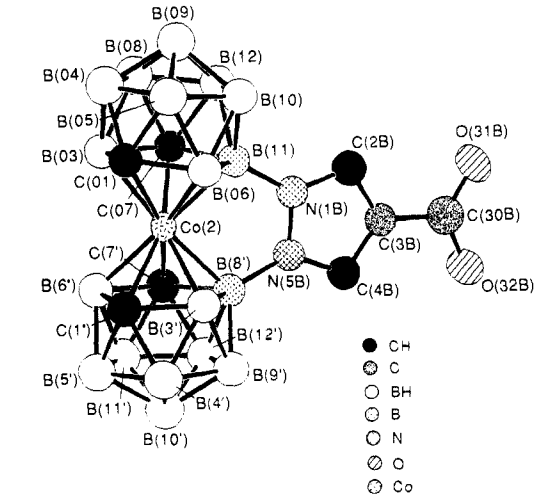
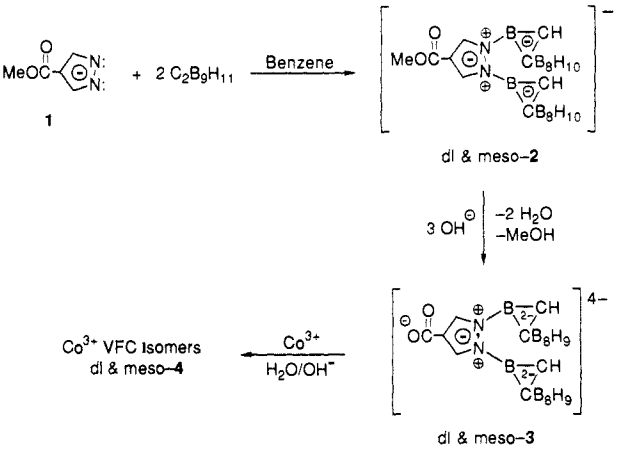


Figure 1. ORTEP representation of *meso*-**4** with hydrogen atoms removed for clarity, showing the numbering scheme. Interatomic distances from Co, in Å: C(01), 2.085 (9); C(07), 2.061 (8); C(1'), 2.077 (9); C(7'), 2.056 (9); B(03), 2.088 (11); B(06), 2.079 (11); B(11), 2.034 (10); B(3'), 2.103 (11); B(6'), 2.072 (11); B(8'), 2.039 (10).

Scheme I



The structure of *meso*-**4** has been determined crystallographically (Figure 1), and the results have been used to correlate the ^1H and ^{11}B FT NMR spectra and HPLC retention times of the meso and *dl* isomers. Both isomers of **4** exhibit the characteristic stability of the *commo*-7,8- and -7,9-bis(dicarbollide) complexes of Co^{3+}.

Conversion of the mixture of isomeric **4** species to their active *N*-hydroxysulfosuccinimide esters was carried out by using 1-ethyl-3-[3-(dimethylamino)propyl]carbodiimide in acetonitrile, and the diastereomeric product mixture, **5**, was purified by reverse-phase HPLC using a C_8 column (62% yield). High conjugation yields were achieved with **5** and model lysine-containing peptides. Reaction of **5** which contained radioactive ^{57}Co ($t_{1/2}$ = 271 days, γ-emission) with the anticarcinoembryonic antigen Mab T84.66[5] produced a conjugate which carried, on the average,

(1) (a) Meares, C. F.; Wensel, T. G. *Acc. Chem. Res.* **1984**, *17*, 202. (b) Brechbiel, M. W.; Gansow, O. A.; Atcher, R. W.; Schlom, J.; Esteban, J.; Simpson, D. E.; Colcher, D. *Inorg. Chem.* **1986**, *25*, 2772.

(2) Shelly, K.; Knobler, C. B.; Hawthorne, M. F. *New J. Chem.* **1988**, *12*, 317.

(3) Hawthorne, M. F.; Young, D. C.; Andrews, T. D.; Howe, D. V.; Pilling, R. L.; Pitts, A. D.; Reintjes, M.; Warren, L. F.; Wegner, P. A. *J. Am. Chem. Soc.* **1968**, *90*, 879.

(4) Tebbe, F. N.; Garrett, P. M.; Hawthorne, M. F. *J. Am. Chem. Soc.* **1968**, *90*, 869.

(5) Wagener, C.; Yang, Y. H.; Crawford, F. G.; Shively, J. E. *J. Immunol.* **1983**, *130*, 2308.

0002-7863/90/1512-5365$02.50/0