Pergamon

Tetrahedron Letters, Vol. 35, No. 6, pp. 825-828, 1994
Elsevier Science Ltd
Printed in Great Britain
0040-4039/94 $6.00+0.00

0040-4039(93)E0371-P

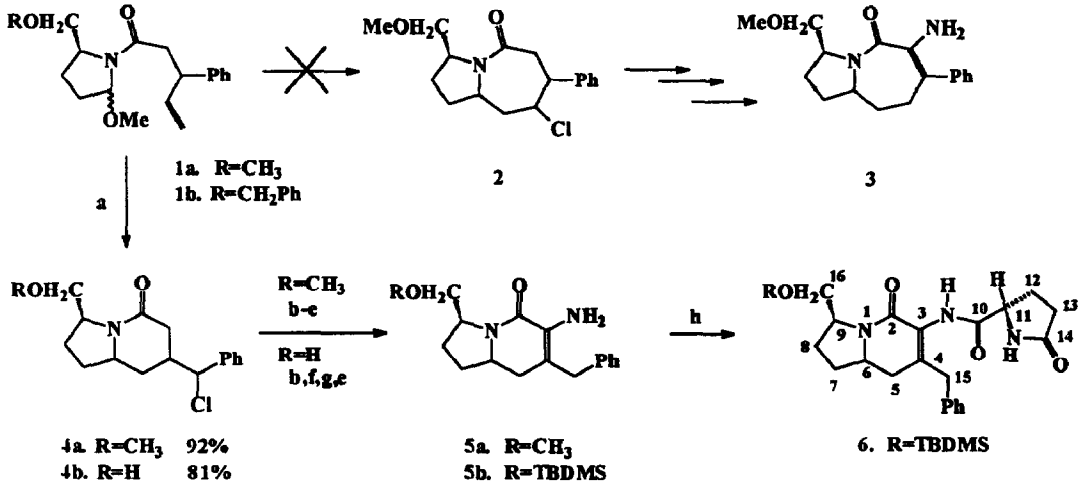
THE USE OF HMQC-TOCSY EXPERIMENTS FOR ELUCIDATING THE STRUCTURES OF BICYCLIC LACTAMS: UNCOVERING A SURPRISE REARRANGEMENT IN THE SYNTHESIS OF A KEY PRO-PHE BUILDING BLOCK.

Kevin D. Moeller,* Cathleen E. Hanau, and Andrè d'Avignon
Department of Chemistry, Washington University, St. Louis, Missouri 63130

Abstract: HMQC-TOCSY experiments were used to unequivocally assign the ring skeletons of several bicyclic lactams. This work demonstrated the power of these techniques for establishing the complete carbon connectivity of peptide building blocks with closely overlapping protons. In addition, it has led to the discovery of a surprise rearrangement reaction and allowed for the correction of a previously misassigned Pro-Phe building block ring skeleton.

Recently, we reported that the titanium tetrachloride initiated cyclization of the α-methoxyalkyl amide **1a** (R = CH_3) led to the formation of the bicyclic seven-membered ring lactam **2** (Scheme 1).[1]

Scheme 1[2]



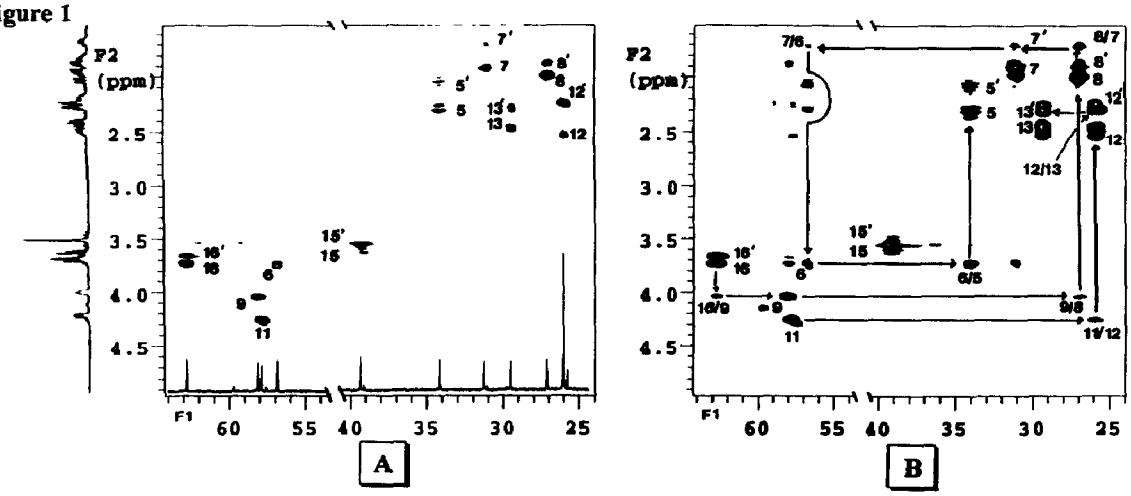
Reagents: **a.** $TiCl_4$, CH_2Cl_2, -78°C to room temperature; **b.** H_2, 5% Pd/C, NaOMe, MeOH, 88% (R=CH_3) and 89% (R=H); **c.** i. LDA, THF, -50 to -40°C, ii. (+)-(2R,8aS)-(camphorsulfonyl)oxaziridine (Davis' reagent), 78%; **d.** i. NCS, DMS, CH_2Cl_2, -30 to -20°C, ii. Et_3N 76%; **e.** NH_3, MeOH, 69% (R=Me) and 75% (R=TBDMS/ the bridgehead isomers could be separated here), **f.** TBDMSCl, imidazole, DMF, 81%; **g.** i. LDA, THF, -78°C, ii. O_2, 60%, **h.** pyroglutamic acid, EDC, HOBt, *N*-ethylmorphiline, 25% (unoptimized, R=TBDMS).

Compound **2** was then converted into what we assigned as the peptide building block **3**. Both of these compounds were obtained as an inseparable mixture of isomers at the bridgehead position; a minor issue

since we eventually needed both isomers for biological testing. The 1-aza-2-oxobicyclo[5.3.0]decane ring skeleton was assigned by comparison of the spectral data to previous cyclization reactions that were known to form seven-membered ring lactams.[3] *However*, after repeating the synthesis with a more readily cleavable TBDMS group in place of the methoxy group,[4] separating the bridgehead isomers, and coupling the building block to pyroglutamic acid, *it became apparent that there were problems with the assignment of the building block as having a seven-membered ring lactam moiety.* The molecule appeared to contain a benzylic methylene that was split into an A,B pattern (see Figure 1A). This led to the initial assignment of **6** as a six-membered ring lactam.

Evidence for this structure was obtained with an HMQC-TOCSY experiment.[5] The HMQC (A) and HMQC-TOCSY (B) contour plots for the upfield region of one of the two bridgehead isomers of **6** are illustrated in Figure 1.[6]

Figure 1



The HMQC plot (A) allowed for assignment of the one-bond C-H correlations. The cross peaks (contours) in the plot were labelled in a fashion consistent with the numbering scheme illustrated for compound **6**. Proton resonances associated with methylene carbons were readily identified since each of the non-equivalent protons yielded a contour with the same carbon. These methylene protons were labeled on the contour plot with primed and non-primed numbers (see for example contours 16' and 16). The contours observed in the HMQC plot also appeared in the HMQC-TOCSY plot (B) and were labelled in an identical fashion. In the HMQC-TOCSY plot, additional correlations appeared that corresponded to relayed heteronuclear magnetization. These correlations indicated which contours from the HMQC plot were coupled to each other, and were labeled with a slash (/) between the numbers representing the two coupled spin systems. For example, the cross peak labeled 16/9 indicated that one of the proton resonances on carbon 16 was coupled to the proton resonance on carbon 9. The net effect of the HMQC-TOCSY experiment was to spread the proton-proton coupling data over the ^{13}C chemical shifts. This simplified the spectrum by reducing the degree of overlap between proton resonances and allowed for the complete assignment of all resonances in the coupled spin system. Using this data, it was possible to trace out the connectivity of the spin system and "walk" around the ring skeleton. The arrows in the HMQC-TOCSY plot indicate the sequential coupling pathway and the mode of assignment for compound **6**. Note how clearly this spectrum showed that compound

6 did not contain a seven-membered ring lactam. As the coupled spin system is traced starting from contour 16, contour 5 becomes a dead end. The methylene protons on carbon 5 are only coupled to the methine proton at carbon 6, an arrangement that is inconsistent with structure **3**. It was also clear that the benzylic carbon was a methylene and was not bound to a neighboring carbon with an attached proton, supporting the assignment of the structure as **4b**.

The presence of the 1-aza-2-oxobicyclo[4.3.0]nonane ring skeleton in **6** was a surprise and indicated that a rearrangement of the original carbon chain had occurred somewhere in the synthesis. Conventional proton and carbon NMR data of **4b** and the intermediates leading to **6** did not indicate where this rearrangement had taken place. Due to the mixture of diastereomers generated during the cyclization step and the significant overlap in the proton resonances, the spectra were simply too complex to distinguish between potential six- and seven-membered ring lactam containing molecules. HMQC-TOCSY experiments again proved pivotal in solving this dilemma. In this case, HMQC-TOCSY data allowed for the assignment of the proton and carbon connectivities for each of the four isomers obtained from the cyclization of **1b**. This was most impressive because the four isomers could not be separated and the assignments had to be done on the mixture! Since every carbon in the ring skeleton (except the carbonyl) was bound to a proton, the *complete* connectivity of the entire ring skeleton could be established for each isomer. All four isomers were found to possess the 1-aza-2-oxobicyclo[4.3.0]nonane ring skeleton (**4b**). Clearly, the rearrangement reaction had occurred during the cyclization step. A similar analysis on the dechlorinated product from **4a** indicated that the cyclization of **1a** had also led to the rearrangement reaction.

Apparently, the six-membered ring lactam product (**4b**) arose from cyclization to form the expected seven-membered ring carbocation **7** followed by subsequent migration of the C_3-C_4 bond. Molecular models indicated that the C_3-C_4 bond was perfectly aligned with the carbocation for this rearrangement, and hence migration of C_3 favored stereoelectronically over migration of the phenyl ring (illustrated for one of the isomers in Figure 2). The net result of this migration was the formation of a second secondary carbocation

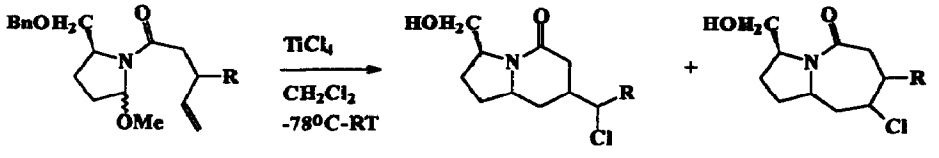
Figure 2

7 **8**

(**8**) that was then trapped by chloride. Initially, the driving force for this migration was thought to be the formation of the benzylic carbocation in **8**. However, when the *N*-α-methoxyamide having a methyl group (**9a**) in place of the phenyl group was synthesized and cyclized, six-membered ring lactam products (**10a**) were again obtained (Scheme 2). None of the seven-membered ring lactam product **11a** was observed. Clearly, relief of the strain associated with the seven-membered ring was sufficient to drive the rearrangement without any added benefits from increased delocalization of the cation. As in earlier examples,[3] the rearrangement could be stopped by eliminating the substituent on the side chain altogether. The cyclization of **9b** led to only the seven-membered ring product **11b**.

In summary, we have found that the use of HMQC-TOCSY experiments can be extremely powerful tools for accurately assigning bicyclic lactam ring skeletons. In the present study, these experiments allowed for the correction of a previously misassigned structure, and the discovery of an unexpected rearrangement

Scheme 2



9a. R=CH$_3$ **10a. 82%** **11a. --**
9b. R=H **10b. --** **11b. 82%**

reaction. We are currently using HMQC-TOCSY experiments to elaborate the complete proton and carbon connectivity of all new ring skeletons. Work aimed at utilizing building block **5b** for the construction of conformationally restricted peptide mimetics, as well as, reinvestigating the synthesis of building block 3 is currently underway and will be reported in due course.

Acknowledgments: We thank the National Institutes of Health (P01 GM24483-12), the National Science Foundation (CHE-9023698), and Washington University for their generous support of our programs. Cathleen Hanau thanks Monsanto for a graduate student fellowship. We also gratefully acknowledge the Washington University High Resolution NMR Facility, partially supported by NIH grants RR02004, RR05018, and RR07155, and the Washington University Mass Spectrometry Resource Center, partially supported by NIHRR00954, for their assistance.

References and Notes

1. Moeller, K. D.; Rothfus, S. L. *Tetrahedron Lett.* **1992**, *33*, 2913.
2. The syntheses of amides **1a**, **1b**, **9a**, and **9b** will be reported later. Unpublished results with Cathleen E. Hanau and Wenhao Li.
3. Moeller, K. D.; Rothfus, S. L.; Wong, P. L. *Tetrahedron* **1991**, *47*, 583. In the simpler model systems (no substituents), the seven- and six-membered lactam ring products were readily distinguishable by conventional NMR techniques.
4. Unpublished results with Wenhao Li.
5. For descriptions of the use of HMQC-TOCSY and HMBC experiments see a. Martin, G. E.; Crouch, R. C. *J. of Nat. Prod.* **1991**, *54*, 1, b. Spitzer, T. D.; Crouch R. C.; Martin, G. E.; Sharaf, M. H. M.; Schiff, Jr., P. L.; Tackie, A. N.; Boye, G. L. *J. Heterocyclic Chem.* **1991**, *28*, 2065, c. Spitzer, T. D.; Crouch, R. C.; Martin, G. E. *J. Heterocyclic Chem.* **1992**, *29*, 265, d. Castle, L. W.; Johnston, Jr., M. D.; Camoutsis, C. L.; Castle, R. N. *J. Heterocyclic Chem.* **1992**, *29*, 1805.
6. The contour plots shown were recorded for a 15 mg sample of compound **6** dissolved in 0.6 mL of $CDCl_3$. The spectra were obtained on a VXR-500 (Varian Associates, Palo Alto, CA) equipped with a reverse-detection probe (Nalorac, Martinez, CA) by acquiring in the hypercomplex mode, 2048 x 248 x 2 points, with spectral windows of 4658 Hz in t_2 (^{1}H shift axis) and 17825 Hz in t_1 (^{13}C shift axis). A delay time of 1.2 s was employed between each of four (HMQC) or sixty-four (HMQC-TOCSY) transients per evaluation increment. ^{13}C Waltz decoupling was used during acquisition with a power sufficient to decouple over 19,000 Hz. Typical 90^0 pulse lengths were 8 us (^{1}H) and 20 us (^{13}C). For HMQC-TOCSY, a spin lock period is maintained for 15 ms at a field strength of 11 Kz. The data were zero-filled to 2048 x 1024 x 2 and subjected to Gaussian apodization in both dimensions prior to double Fourier transformation.

(Received in USA 22 October 1993; revised 17 November 1993 accepted 18 November 1993)