

Self-Assembly, Guest Capture, and NMR Spectroscopy of a Metal– Organic Cage in Water

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S Supporting Information

ABSTRACT: A green organic—inorganic laboratory experiment has been developed in which students prepare a selfassembling iron cage in D_2O at room temperature. The tetrahedral cage captures a small, neutral molecule such as cyclohexane or tetrahydrofuran. ¹H NMR analysis distinguishes captured and free guests through diagnostic chemical shifts, splitting patterns, diffusion coefficients (using DOSY), and the appearance of captured hydrophobic molecules in D_2O even when free guests are insoluble in water. Students are invited to test their hypotheses about guest binding and to perform control and competition experiments. All of the reagents are commercial, the 23-component self-assembly is complete in 1 week at room temperature (requiring neither workup nor purification), and the product solution is intensely purple. In



their laboratory reports, students effectively related organic chemistry with molecular self-assembly, supramolecular chemistry, and host-guest interactions.

KEYWORDS: Upper-Division Undergraduate, Organic Chemistry, Inorganic Chemistry, Laboratory Instruction, Green Chemistry, Microscale Lab, Molecular Recognition, NMR Spectroscopy, Aqueous Solution Chemistry, Inquiry-Based/Discovery Learning

 ${f S}$ pontaneous self-assembly of molecular complexity in an aqueous environment is a hallmark of biochemical systems and a major focus of supramolecular chemistry research. Discrete self-assembled structures have found applications in catalysis, information transfer, and guest capture.¹ These have great potential for illustrating principles of green chemistry to students and for blurring the boundary of organic/inorganic chemistry. There exist a few experiments for solution-phase self-assembly²⁻⁴ and for host–guest chemistry^{2,5-9} of finite structures, most notably Rebek's "tennis ball" dimer that captures methane.² The present experiment has additional advantages: commercial building blocks for direct assembly, a larger cavity for capturing more varied and interesting guests, a combination of organic and inorganic components, and water as solvent.

Water-soluble metal-organic capsule 1 (Figure 1), first described by the Nitschke group at the University of Cambridge,¹⁰ is capable of efficiently trapping the harmful substances benzene (a carcinogen),¹¹ SF₆ (a potent greenhouse gas),¹² and P₄ (white phosphorus, an air-sensitive chemical warfare agent).¹¹ Their capture is reversible, noncovalent, and selective; toluene, cycloheptane, and *t*-butanol are not competent guests.¹³ The tetrahedral cage spontaneously forms from 22 subcomponents: 12 2-formylpyridines, six sulfonated benzidines, and four iron(II) ions (Scheme 1). The structure is held together by 12 imine bonds and 24



Figure 1. Iron cage **1** with encapsulated P_4 .¹¹ Hydrogen atoms are omitted for clarity. Fe, violet; N, blue; C, gray; O, red; S, yellow; P, orange. Figure generated from CCDC-727817 using Mercury.

nitrogen-to-iron bonds, satisfying octahedral coordination for all four iron(II) atoms.

An advanced undergraduate laboratory experiment is described here that focuses on the aqueous self-assembly and host-guest chemistry of iron cage complex 1. The iron cage may be simply prepared at room temperature directly in D_2O ,



Scheme 1. Self-Assembly of Iron Cage and Guest Capture^a



^{*a*}Counterions and the ligands at the other five edges of the tetrahedron are omitted for clarity.

eliminating the need for heating, product isolation, or any additional solvents. Importantly, all of the reagents used in the preparation of 1 are readily available and inexpensive.

EXPERIMENTAL OVERVIEW

This experiment required 15 min at the end of one laboratory period to set up the reactions. The following laboratory period was dedicated to the acquisition of NMR spectra, the examination of computer models of the cage (using the Cambridge Crystallographic Data Centre, CCDC), and analysis of NMR spectra. Students working individually in laboratory sections of 11 typically acquired three ¹H spectra each (33

total) in about 3.5 h. Smaller sections (or fewer ¹H spectra per student) allowed students to collect COSY and DOSY (diffusion-ordered spectroscopy) spectra themselves. The NMR time requirement for each student the second week was minimized when the instructor provided DOSY and COSY spectra (see Supporting Information for spectra). Students could also work in pairs.

Each student set up three reactions in microcentrifuge tubes: one empty cage control reaction, and two reactions with the cage and one or two guests each. The control reaction contained 2-formylpyridine (80 μ mol), 4,4'-diaminobiphenyl-2,2'-disulfonic acid (40 μ mol), sodium hydroxide (80 μ mol), iron(II) sulfate heptahydrate (27 μ mol), and *t*-butanol (6.7 μ mol) in D₂O (600 μ L). The other two reactions also contained one or two of the following potential guest molecules (chosen by the student): cyclopentane, cyclohexane, tetrahydrofuran, tetrahydropyran, 1,3-dioxolane, and 1,4-dioxane. All three reactions were left at room temperature for a week in a centrifuge tube rack (optimally with magnetic stirring for cyclopentane and cyclohexane, as they are water-insoluble), and then analyzed by ¹H NMR spectroscopy.

HAZARDS

2-Formylpyridine, 4,4'-diaminobiphenyl-2,2'-disulfonic acid, *t*butanol, tetrahydrofuran, tetrahydropyran, and 1,4-dioxane are inhalation, skin, and eye irritants. 2-Formylpyridine and 1,4dioxane are toxic by inhalation. Sodium hydroxide may cause burns on the skin. Tetrahydrofuran, tetrahydropyran, and 1,4dioxane are flammable and may form explosive peroxides. *t*-Butanol, cyclopentane, cyclohexane, and 1,3-dioxolane are



Figure 2. Representative student ¹H NMR spectrum (*x*-axis) and DOSY spectrum of iron cage **1** with 1,4-dioxane in D_2O . (a) Occupied iron cage, (b) water, (c) free dioxane, (d) captured dioxane, (e) *t*-butanol standard. All eight major peaks at 6–10 ppm are from the occupied cage, with only trace amounts (very small peaks) of unoccupied cage.

flammable. Deuterium oxide is nonhazardous. The hazards of iron cage 1 are unknown, but it should be treated as a skin and eye irritant and disposed of in hazardous aqueous waste. Gloves and protective eyewear should be worn for this experiment.

RESULTS AND DISCUSSION

This experiment has been used for five years in an upperdivision undergraduate organic laboratory course with an average of 11 students per year. Cage formation was very reliable after 1 week at room temperature, and encapsulation was consistently observed for all of the recommended guests. Estimated student yields of the cage ranged from 50 to 97% (by NMR, comparing the imine signal to the *t*-butanol standard). Students were impressed by the intense purple color of the cage solutions and the changes in NMR chemical shifts of captured guests. Students were able to rationalize the observed shielding of captured guests' NMR signals due to the aromatic rings lining the cage interior upon examination of physical and electronic models of the cage. Diffusion-ordered spectroscopy (DOSY), but not COSY, was entirely new to every student.

A student DOSY spectrum for cage 1 with 1,4-dioxane is shown in Figure 2. The x-axis displays the ¹H NMR spectrum. The peaks from the ¹H spectrum are projected along the *y*-axis based on the diffusion coefficient of the species. Larger (or more slowly diffusing) species, such as the assembled cage, appear near the top of the DOSY spectrum. Smaller species, such as water, t-butanol, or free guest molecules, appear closer to the bottom of the DOSY spectrum (they diffuse more rapidly in solution). Captured guest molecules diffuse more slowly than free guest molecules and appear closer to the level of the cage on the y-axis. Additionally, in the case of encapsulated 1,4-dioxane, the cage inhibits dioxane ring flips so that chemical inequivalence is observed between axial and equatorial hydrogens (see Figure 2d). A similar phenomenon for 1,4-dioxane has been previously observed in an organic host at -30 °C¹⁴ or in solution at -107 °C,¹⁵ but never at room temperature.

With respect to green chemistry,¹⁶ the atom economy is excellent. The small scale and use of D_2O as the only solvent minimize potential waste. Moreover, the reagents are all commercial, and the single-step procedure requires no heating, no workup, and no purification. Rarely do students encounter such complex self-assembly in non-biological systems.

CONCLUSIONS

This experiment is suitable for a typical undergraduate advanced organic laboratory course, and it could also be adapted for an inorganic laboratory course with additional emphasis on the iron(II) coordination environment. Formation of the cage and guest encapsulation expose students to imine formation, metal coordination, supramolecular systems (including dynamic covalent chemistry and host–guest chemistry), the hydrophobic effect, molecular visualization software, and advanced NMR techniques.

The main pedagogical goal of this experiment is for students to relate organic chemistry to molecular self-assembly, supramolecular chemistry, and host-guest interactions. Students achieved these goals by demonstrating mastery of the following learning objectives in their laboratory reports:

 characterize a highly symmetric structure using ¹H, COSY, and DOSY NMR

- interpret the shielding of guest ¹H NMR signals by an aromatic host
- visualize CCDC structures of a molecular capsule
- show proficiency with M_xL_y terminology for metal– organic cages
- predict good and bad guests for a specific host structure

Potential extensions of this experiment could include capping a face of the cage with guanidinium ion^{17} or the stepwise displacement of guests by stronger-binding or more abundant guests.¹³

ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available on the ACS Publications website at DOI: 10.1021/acs.jchemed.Sb00714.

Instructions for students and notes for instructors; a full set of student NMR spectra (¹H and COSY for an empty cage; ¹H and DOSY for six guest-containing cages); instructions for acquiring DOSY NMR spectra (PDF, DOCX)

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Notes

The authors declare no competing financial interest.

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REFERENCES

(1) Chakrabarty, R.; Mukherjee, P. S.; Stang, P. J. Supramolecular Coordination: Self-Assembly of Finite Two- and Three-Dimensional Ensembles. *Chem. Rev.* **2011**, *111* (11), 6810–6918.

(2) Hof, F.; Palmer, L. C.; Rebek, J., Jr. Synthesis and Self-Assembly of the "Tennis Ball" Dimer and Subsequent Encapsulation of Methane. An Advanced Organic Chemistry Laboratory Experiment. *J. Chem. Educ.* **2001**, 78 (11), 1519–1521.

(3) Pentecost, C. D.; Tangchaivang, N.; Cantrill, S. J.; Chichak, K. S.; Peters, A. J.; Stoddart, J. F. Making Molecular Borromean Rings. A Gram-Scale Synthetic Procedure for the Undergraduate Organic Lab. *J. Chem. Educ.* **2007**, *84* (5), 855–859.

(4) Fernández, A.; López-Torres, M.; Fernández, J. J.; Vázquez-García, D.; Vila, J. M. A One-Pot Self-Assembly Reaction To Prepare a Supramolecular Palladium(II) Cyclometalated Complex: An Undergraduate Organometallic Laboratory Experiment. *J. Chem. Educ.* **2012**, 89 (1), 156–158.

(5) Tardajos, G.; González-Gaitano, G. Chemical Equilibrium in Supramolecular Systems as Studied by NMR Spectrometry. *J. Chem. Educ.* **2004**, *81* (2), 270–274. (6) Haldar, B.; Mallick, A.; Chattopadhyay, N. Supramolecular Inclusion in Cyclodextrins: A Pictorial Spectroscopic Demonstration. *J. Chem. Educ.* **2008**, *85* (3), 429–432.

(7) Umali, A. P.; Anslyn, E. V.; Wright, A. T.; Blieden, C. R.; Smith, C. K.; Tian, T.; Truong, J. A.; Crumm, C. E.; Garcia, J. E.; Lee, S.; Mosier, M.; Nguyen, C. P. Analysis of Citric Acid in Beverages: Use of an Indicator Displacement Assay. *J. Chem. Educ.* **2010**, *87* (8), 832–835.

(8) Mendicuti, F.; González-Álvarez, M. J. Supramolecular Chemistry: Induced Circular Dichroism to Study Host-Guest Geometry. J. Chem. Educ. 2010, 87 (9), 965–968.

(9) Mendes, D. C.; Ramamurthy, V.; Da Silva, J. P. Identification of Guest-Host Inclusion Complexes in the Gas Phase by Electrospray Ionization–Mass Spectrometry. *J. Chem. Educ.* **2015**, *92* (6), 1091–1094.

(10) Mal, P.; Schultz, D.; Beyeh, K.; Rissanen, K.; Nitschke, J. R. An Unlockable-Relockable Iron Cage by Subcomponent Self-Assembly. *Angew. Chem., Int. Ed.* **2008**, *47* (43), 8297–8301.

(11) Mal, P.; Breiner, B.; Rissanen, K.; Nitschke, J. R. White Phosphorus Is Air-Stable Within a Self-Assembled Tetrahedral Capsule. *Science* **2009**, *324* (5935), 1697–1699.

(12) Riddell, I. A.; Smulders, M. M. J.; Clegg, J. K.; Nitschke, J. R. Encapsulation, Storage and Controlled Release of Sulfur Hexafluoride From a Metal-Organic Capsule. *Chem. Commun.* **2011**, 47 (1), 457–459.

(13) Smulders, M. M. J.; Zarra, S.; Nitschke, J. R. Quantitative Understanding of Guest Binding Enables the Design of Complex Host-Guest Behavior. J. Am. Chem. Soc. 2013, 135 (18), 7039–7046. (14) Chapman, R. G.; Sherman, J. C. Restricted Motion of Guests Confined in Carceplexes and Capsules. J. Org. Chem. 2000, 65 (2),

513–516.
(15) Jensen, F. R.; Neese, R. A. Ring Inversion Barrier of Dioxane. J.

Am. Chem. Soc. 1971, 93 (23), 6329–6330. (16) Anastas, P. T.; Warner, J. C. Green Chemistry: Theory and

(16) Anastas, P. 1.; Warner, J. C. Green Chemistry: Theory and Practice; Oxford University Press: New York, 1998.

(17) Zarra, S.; Smulders, M. M. J.; Lefebvre, Q.; Clegg, J. K.; Nitschke, J. R. Guanidinium Binding Modulates Guest Exchange within an $[M_4L_6]$ Capsule. *Angew. Chem., Int. Ed.* **2012**, *51* (28), 6882–6885.