INORGANIC

SAMPLE EXPERIMENT

Diastereotopicity by NMR Spectral Analysis of *O,O'*- and *N,N'*- Acetals

INTRODUCTION

Nuclear magnetic resonance (NMR) spectroscopy remains one of the most valuable analytical methods at a chemist's disposal due to its powerful capabilities for structural elucidation and its inherently quantitative nature. By understanding the fundamental concepts of NMR spectroscopy (coupling, chemical shift and integration), scientists can take advantage of this technique to gain insight into the different atoms present in a molecule, such as how they are connected and their relative spatial proximities, information which many other common analytical techniques cannot provide. For example, mass spectrometry can afford crucial information on the mass of molecular ions and the presence of characteristic fragmentation patterns that can help piece together a molecular structure. Unfortunately, it does not provide much insight into a molecule's bonding or spatial arrangements. Comparatively, with NMR, two identical functional groups within the same molecule can give rise to distinct chemical shifts due to the difference in surrounding atoms, making the molecule constitutionally heterotopic or stereoheterotopic.[1] As shown in **Figure 1**, in the case of a constitutionally heterotopic molecule, the difference stems from the connectivity of the atoms (constitutional isomers), whereas a stereoheterotopic molecule differs in the atoms' spatial arrangement, which can then be further classified into enantiomers and diastereomers. For students, constitutional heterotopic nuclei can be easily differentiated visually, either on paper or using molecular model kits. However, stereoheterotopic nuclei (*i.e.*, diastereomers) are much more difficult to visualize and identify.[2]

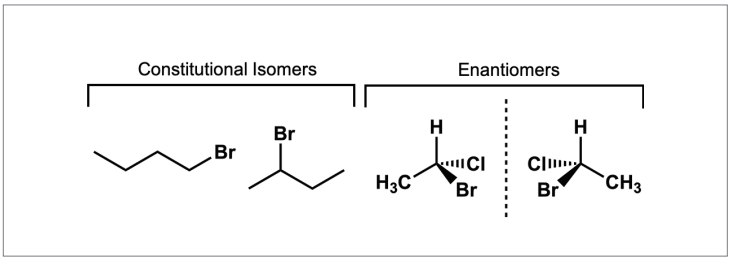

Figure 1. Examples of constitutional isomers (left) and stereoheterotopic isomers (right).

Following a modified procedure by Saba *et al.*,[2] two achiral molecules, 1-(diisopropoxymethyl)-4-nitrobenzene (*O,O'*-acetal) and 1,3-diisopropyl-2-(4-nitrophenyl) imidazolidine (*N,N'*-acetal) (**Figure 2**), are synthesized and studied via ^{1}H and ^{13}C NMR spectroscopy to reveal how the presence of a stereocenter is not a precondition for diastereotopicity.[2] Rather, diastereomers can arise from a prochiral center, where a prochiral center is a stereocenter that is not present in the initial molecule, but that can be generated from a simple substitution.[2] As shown in **Figure 3**, substitution of a methyl in the isopropyl group on either of the molecules leads to the formation of a stereocenter, which can then be named by the descriptors pro-rectus (pro-*R*) or pro-sinister (pro-*S*).

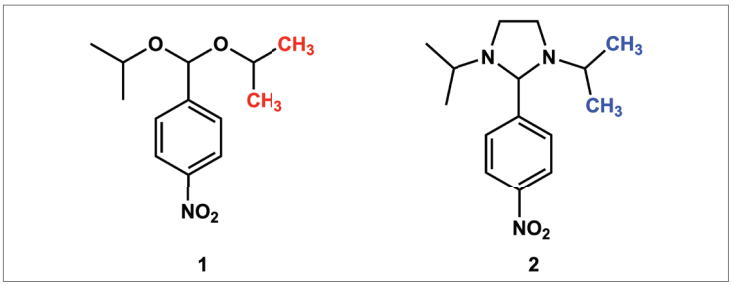

Figure 2. Molecular structures of 1-(diisopropoxymethyl)-4-nitrobenzene (O,O'-acetal, 1) and 1,3-diisopropyl-2-(4-nitrophenyl) imidazolidine (N,N'-acetal, 2). The diastereotopic methyl groups that will be focused on are highlighted in each molecule.

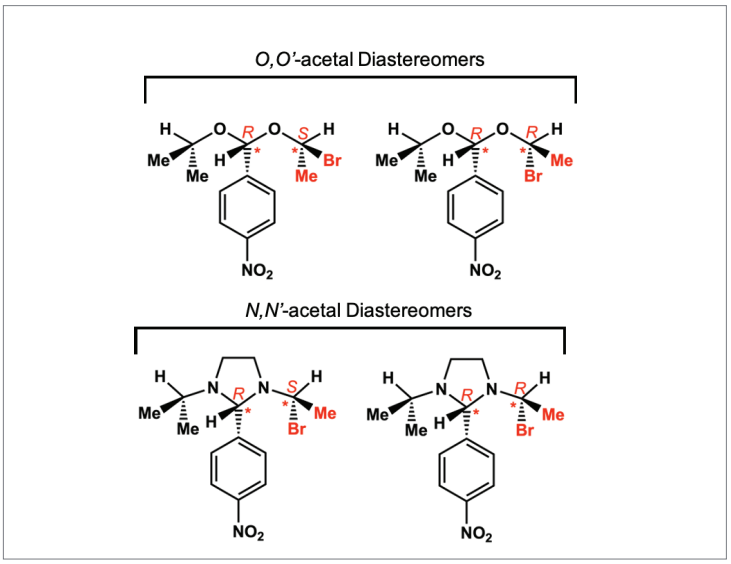

Figure 3. Diastereomers of O,O'-acetal and N,N'-acetal (Top/Bottom) formed by substitution of methyl groups with a bromine atom. Upon substitution, two stereocenters are formed and have been labeled R for pro-R or S for pro-S.

Despite being able to distinguish between diastereotopic fragments with NMR spectroscopy, this technique is unfortunately not always readily available to students during their undergraduate degree, making it difficult for them to gain hands-on experience for this topic in a lab setting. This is due to the fact that traditional high-field instruments are quite costly (sizeable initial capital investment, expensive cryogens, require routine maintenance and knowledgeable full-time staff), necessitate extensive training to operate, and are often difficult to access due to its abundant use for research. However, with the advent of benchtop NMR spectrometers, students can acquire their own spectra with little training and at a fraction of the cost compared to high-field NMR spectrometers.[3] In this sample experiment, benchtop NMR spectroscopy is used to analyze diastereotopic moieties in achiral molecules, by studying *O,O'*-acetal **1** and *N,N'*-acetal **2**.

PROCEDURE

Materials

Triisopropylorthoformate (97%), 4-nitrobenzaldehyde (98%), tetrabutylammonium tribromide (98%), anhydrous isopropanol (99.5%), *N,N'*-diisopropylethylenediamine (99%) and chloroform-*d* (99.8%) were purchased from Sigma Millipore and used without further purification.

Synthesis

O,O'-acetal **1** and *N,N'*-acetal **2** were prepared according to a procedure published by Saba *et al.*[2] *O,O'*-acetal **1**: Triisopropylorthoformate (3.51 mL, 15.8 mmol), 4-nitrobenzaldehyde (2.38 g, 15.8 mmol), tetrabutylammonium tribromide (158 mg, 0.327 mmol) and anhydrous isopropanol (15 mL) were added to a round bottom flask and stirred at room temperature for 75 min. Then, a standard aqueous workup was performed to obtain **1**. *N,N'*-acetal **2**: This reaction was performed neat. *N,N'*-diisopropylethylenediamine (4.3 mL, 24 mmol) and 4-nitrobenzaldehyde (3.0 g, 20 mmol) were added to a round bottom flask and heated to 140 °C in a sand bath over a period of 30-40 min. Then, a standard aqueous workup was performed to obtain **2**.

RESULTS AND DISCUSSION

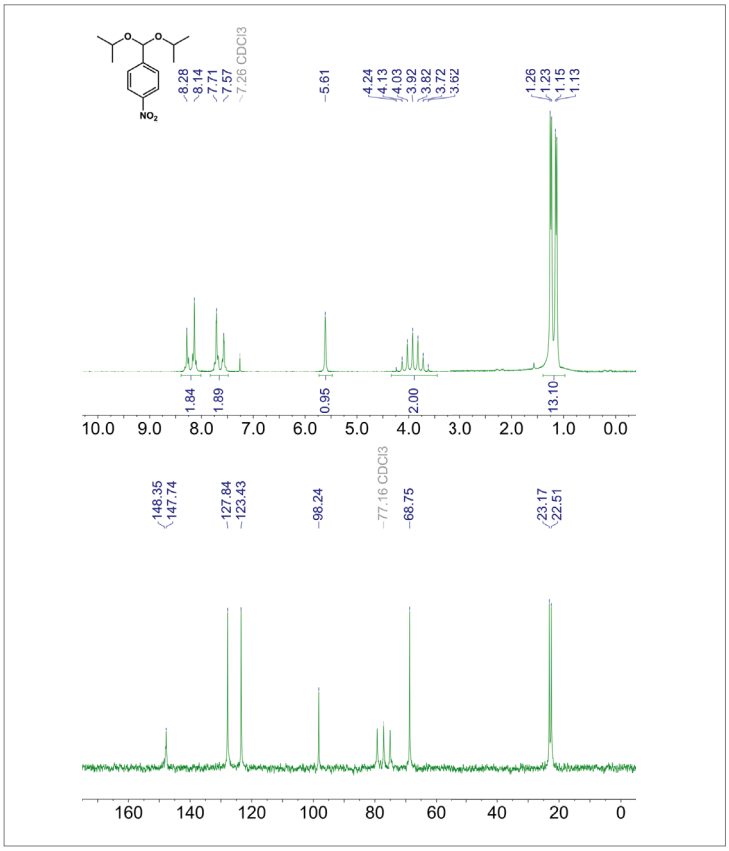

Figure 4: ^{1}H (top) and ^{13}C (bottom) NMR spectra of O,O'-acetal 1 acquired using the benchtop NMR 60 MHz spectrometer.

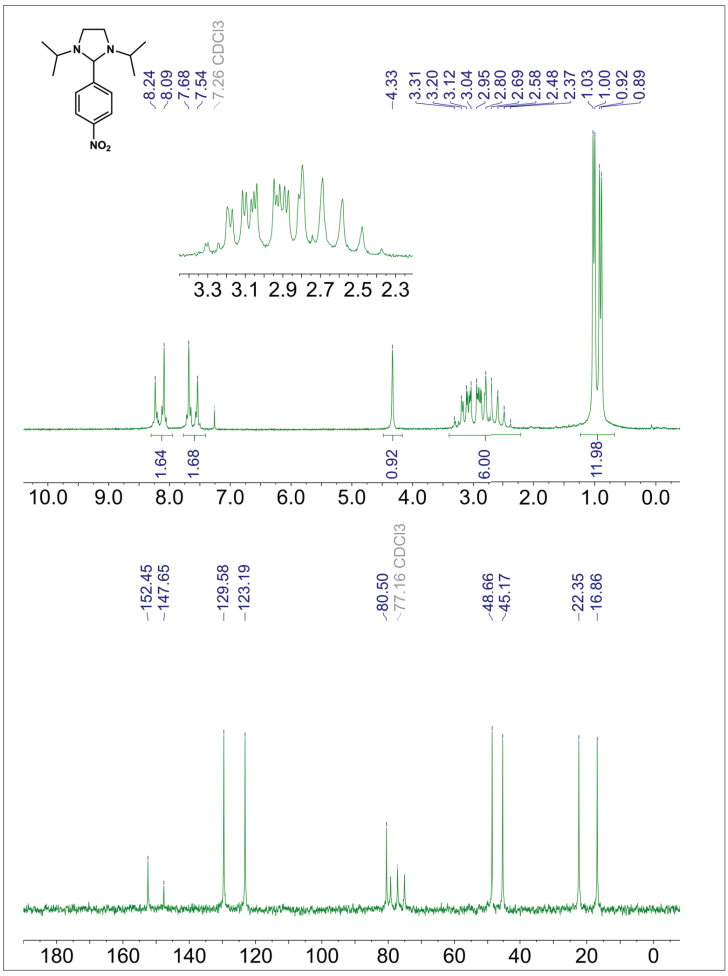

Figure 5: ^{1}H (top) and ^{13}C (bottom) NMR spectra of N,N'-acetal 2 with a zoomed-in view of the isopropyl C-H fragment, acquired using the benchtop NMR 60 MHz spectrometer.

As shown in **Figure 4**, the ^{1}H NMR spectrum of *O,O'*-acetal (**1**) shows two overlapping doublets centered at 1.18 ppm and 1.21 ppm, a septet centered at 3.93 ppm and a singlet at 5.61 ppm. These signals stem from the methyl groups of the isopropyl moiety and the C-H moiety between the two oxygen atoms, respectively.

Two distinct methyl groups are also reflected in the ^{13}C NMR spectrum where two separate methyl resonances are observed at 22.51 ppm and 23.17 ppm. Similarly, in the ^{1}H NMR spectrum of the *N,N'*-acetal (**Figure 5**), there are two overlapping doublets centered at 0.98 ppm and 0.95 ppm, two multiplets superimposed on top of each other in the range of 3.31 ppm to 2.37 ppm, and finally a singlet at 4.33 ppm. The doublets are formed by the methyl groups of the isopropyl groups, the overlapping signal consists of a septet of the C-H fragment of the isopropyl group along with the CH_2 backbone of the imidazolidine moiety and the singlet is formed by the C-H group between the two nitrogen atoms in the imidazolidine group. In parallel to the *O,O'*-acetal, the ^{13}C NMR spectra of *N,N'*-acetal in **Figure 5** also shows two separate methyl resonances at 16.86 ppm and 22.35 ppm.

CONCLUSION

Using benchtop NMR spectroscopy, students are able to gather a ^{1}H NMR spectra within seconds and can then begin analyzing their findings. In this experiment, the ^{1}H and ^{13}C spectra of two achiral molecules, *O,O'*-acetal (**1**) and *N,N'*-acetal (**2**), were studied to afford students with firsthand experience of diastereotopicity. Despite these two molecules lacking stereocenters, anisochronous methyl peaks arise from the isopropyl moiety, which led to the conclusion that achiral molecules can display distinct NMR signals based on the presence of a prochiral center. By using the benchtop NMR 60 MHz, students are able to experience this phenomenon in the lab, for a fraction of the time and cost of a high-field NMR instrument.

References

[1] Eliel, E.L. *J. Chem. Educ.* **1980**, *57*, 52-55.
[2] Saba, S.; Corozo-Morales, A. J. *Chem. Educ.* **2019**, *96*, 354-359.
[3] Riegel, S.D.; Leskowitz, G.M. *Trends Anal. Chem.* **2016**, *83*, 27-38.

> “The Chemistry Department at the University of Florida started using benchtop NMR here in our laboratories in 2013. Prior to that we were using our departmental research instruments but we weren’t able to get our students any hands experience with the NMR. When we heard benchtop NMR was available we were very excited to be able to provide that to our students!”
>
> **— Dr. Tammy Davidson**, Senior Lecturer,
> Department of Chemistry, University of Florida

Bay 1, 4600 – 5 Street NE
Calgary, Alberta, Canada
T2E 7C3

Tel: +1.403.769.9499

nanalysis.com

sales@nanalysis.com