ORGANIC

UNDERGRADUATE EXPERIMENT

The Fischer Esterification of 4-Amino-3-Nitrobenzoic Acid

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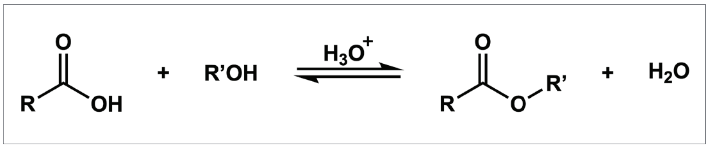
INTRODUCTION

An undeniably important part of undergraduate chemistry courses is the associated laboratory period, which aids in developing a deeper understanding of classroom theory using lab experiments. It is with this hands-on experience that students become exposed to fundamental concepts such as solubility, polarity, extraction/isolation techniques, and characterization using spectroscopic methods (^{1}H and ^{13}C magnetic resonance spectroscopy, infrared spectroscopy, etc.), among other analytical techniques. Specifically, nuclear magnetic resonance (NMR) spectroscopy is a topic introduced in early organic chemistry courses; however, students are commonly exposed to software-generated or idealized spectra, and many undergraduate students still do not have direct access to an NMR spectrometer due in part to their significant upfront and recurring costs.[1] However, with the advent of benchtop NMR, students are able to work directly with an NMR instrument, gathering and interpreting their own spectra within a typical 3-hour laboratory block. Time and time again, NMR has proven itself to be one of the most important analytical tools in chemistry due to its extensive structural elucidation capabilities, and benchtop NMR provides unparalleled access to this technique.

One of the most studied reactions in organic chemistry is the Fischer esterification, which is usually taught in first-year organic chemistry courses.[2] It is a reversible condensation reaction, involving the reaction of a carboxylic acid with an alcohol under acidic conditions to produce an ester and water, outlined in **Scheme 1**.



Scheme 1. *General reaction scheme for the Fischer esterification reaction.*

In this sample experiment, a simple Fischer esterification reaction is performed, based on work published by Schweiker *et al.* in *The Journal of Chemical Education*.[3] The ^{1}H and ^{13}C{^{1}H} NMR spectra are acquired using a 60 MHz benchtop spectrometer, illustrating the benefits of incorporating this technology into undergraduate teaching labs. In addition, this experiment emphasizes the pedagogic goals of liquid-liquid extractions, solubility, polarity and offer a closer look into benchtop NMR analysis.

PROCEDURE

Materials

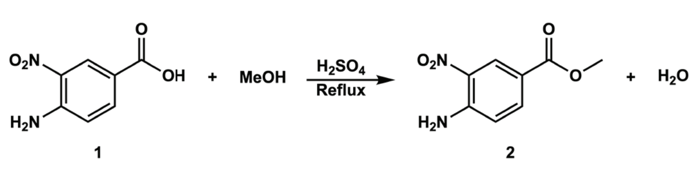
4-Amino-3-nitrobenzoic acid (97%), sulfuric acid (95-98%), anhydrous methanol (99.8%), sodium bicarbonate (≥99.5%), anhydrous ethyl acetate (99.8%), pentane (98%) and DMSO-d_6 (99.9%) were purchased from Sigma Millipore and used without further purification.

Instrumentation

All NMR data was obtained using a Nanalysis 60PRO instrument. The ^{1}H experiments were performed using the following parameters: spectral width, 20 ppm; spectral center, 5 ppm; number of points, 4096; number of scans, 4 for 4-amino-3-nitrobenzoic acid (**1**), 16 for methyl 4-amino-3-nitrobenzoate (**2**); dummy scans, 0; interscan delay, 5 seconds for 4-amino-3-nitrobenzoic acid (**1**), 7 seconds for methyl 4-amino-3-nitrobenzoate (**2**); pulse angle, 90°; receiver gain, auto. The ^{13}C{^{1}H} experiments were performed using the following parameters: spectral width, 220 ppm; spectral center, 100 ppm; number of points, 4096; number of scans, for 4-amino-3-nitrobenzoic acid (**1**), 16384 for methyl 4-amino-3-nitrobenzoate (**2**); dummy scans, 0; interscan delay, 0 seconds; pulse angle, 67.70° for 4-amino-3-nitrobenzoic acid (**1**), 30° for methyl 4-amino-3-nitrobenzoate (**2**); receiver gain, auto. All spectra were manually corrected for phase and baseline distortions using the MestReNova software (v14.1.1).

Synthesis

The synthesis of methyl 4-amino-3-nitrobenzoate (**2**) from 4-amino-3-nitrobenzoic acid (**1**) was adapted from a literature procedure described by Schweiker *et al.* and is outlined in **Scheme 2**.[3]



Scheme 2. *Preparation of methyl 4-amino-3-nitrobenzoate (**2**) from the reaction of 4-amino-3-nitrobenzoic acid (**1**) with methanol under reflux conditions and using concentrated sulfuric acid as a catalyst.*

4-Amino-3-nitrobenzoic acid (307 mg, 1.69 mmol) was dissolved in anhydrous methanol (40 mL). Concentrated sulfuric acid was added dropwise (6 drops) to the reaction mixture and was heated to reflux for 24 hours. It should be noted that the experimental procedure presented by Schweiker *et al.* indicates that the reaction can be done in 1 hour (suitable for an undergraduate laboratory), but for maximum yield, 16 hours is optimal.[3] The reaction was quenched with a saturated solution of sodium bicarbonate (40 mL) and transferred to a separatory funnel. Ethyl acetate (40 mL) was used to extract **2** and and was further washed with the sodium bicarbonate solution. This was repeated until the sodium bicarbonate layer (bottom layer) remained clear (shown in wash 3 of **Figure 1**).

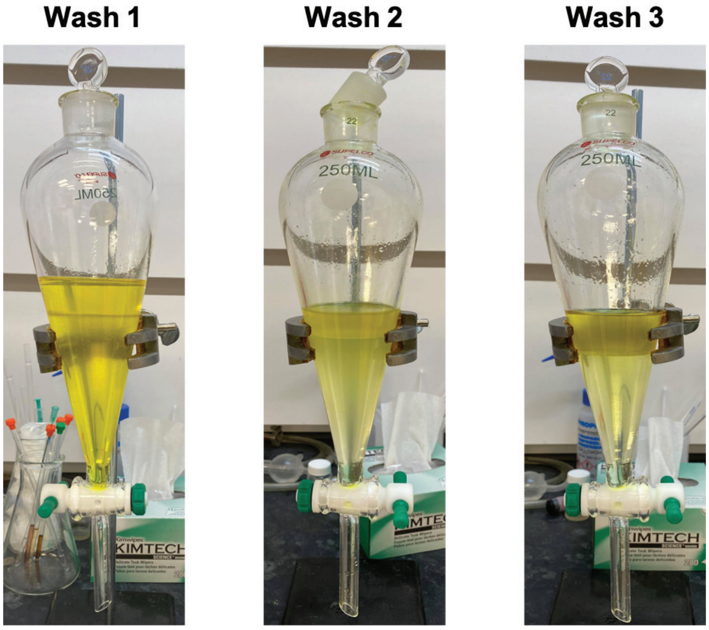


Figure 1. *Ethyl acetate (top layer) washes illustrating the sodium bicarbonate (bottom layer) becoming clear after consecutive washes.*

The ethyl acetate layer was concentrated *in vacuo* using a rotary evaporator. The resulting solid was then redissolved in a minimal amount of ethyl acetate, filtered through a silica plug to remove undesired sodium salts and concentrated *in vacuo* to obtain **2**. The final product was obtained as a cotton-like, bright-yellow solid (**Figure 2**).

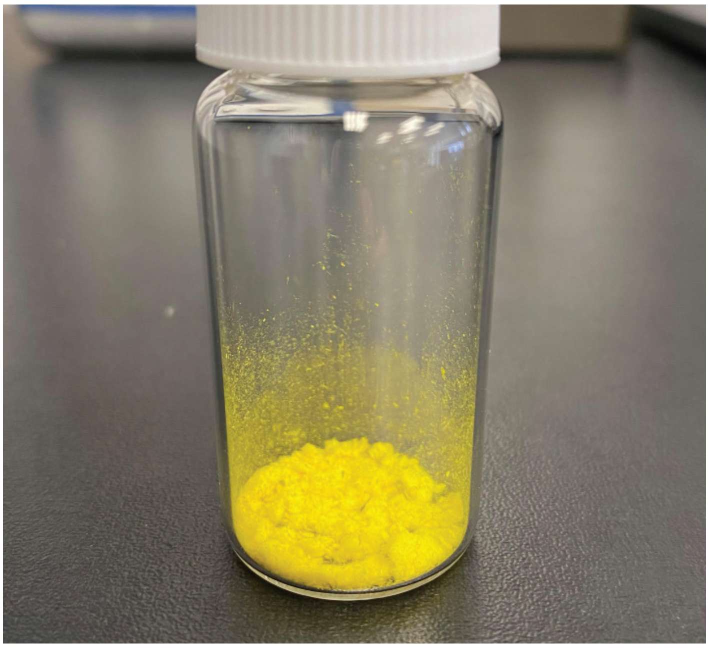


Figure 2. *Isolated product, methyl 4-amino-3-nitrobenzoate (**2**), after wash and purification steps.*

RESULTS AND DISCUSSION

Using ^{1}H and ^{13}C{^{1}H} NMR spectroscopy, both the starting material **1** and product **2** were elucidated, and their spectra were compared. **Figures 3** and **4** depict the ^{1}H (top) and ^{13}C (bottom) spectra for **1** and **2**, respectively.

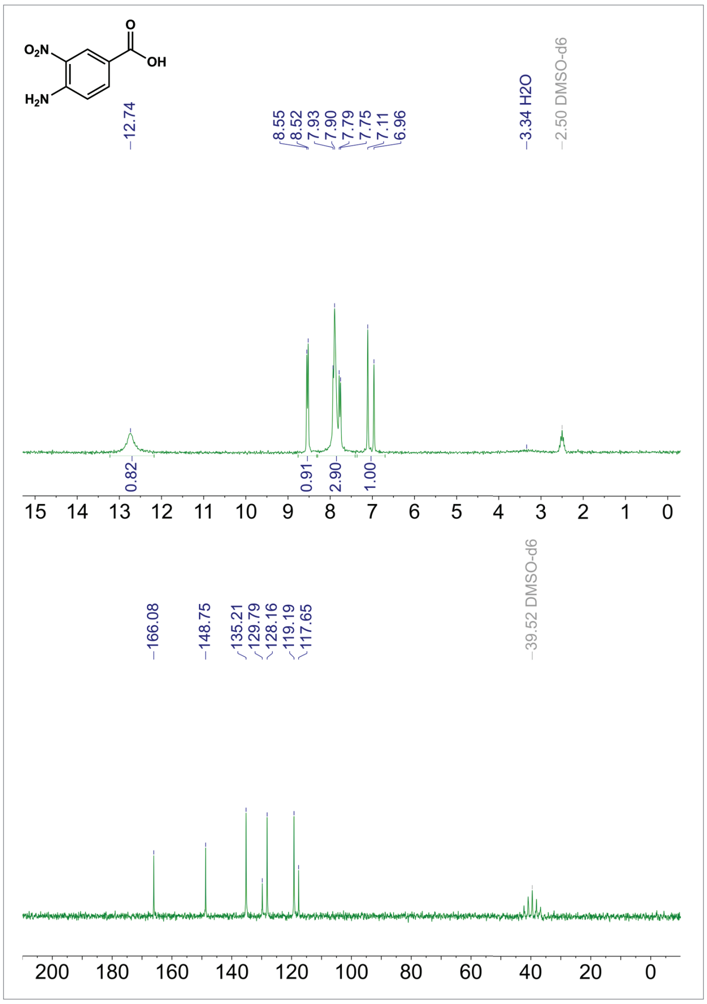


Figure 3. *^{1}H (top, 60 MHz) and ^{13}C{^{1}H}(bottom,15.1 MHz) NMR spectra of 4-amino-3-nitrobenzoic acid (**1**) in DMSO-d_6.*

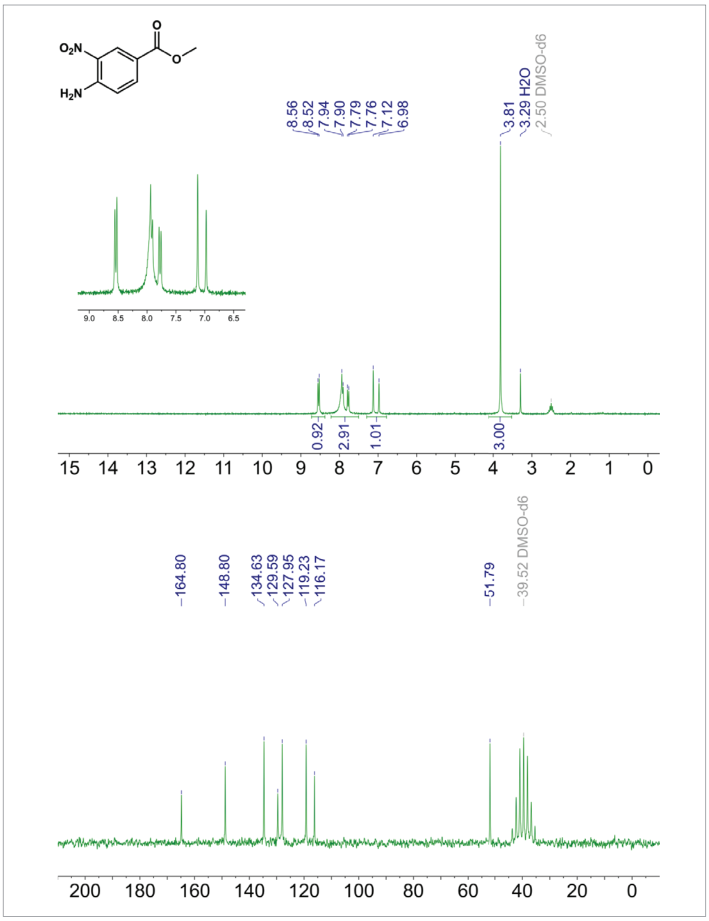


Figure 4. *^{1}H (top, 60 MHz) and ^{13}C{^{1}H}(bottom, 15.1 MHz) NMR spectra of methyl 4-amino-3-nitrobenzoate (**2**) in DMSO-d_6.*

Upon comparison of the ^{1}H spectra of the starting material and product, we observe the loss of a broad peak at 12.74 ppm from **Figure 3** (top) and the appearance of a singlet at 3.81 ppm in **Figure 4** (top). The loss of the broad peak corresponds to the OH group in the carboxylic acid functionality, whereas the singlet peak represents the methoxy methyl group. Aside from these two groups, the spectra for the starting material and product look very similar, which is expected, as they are structurally almost identical. With the help of the other peaks in the ^{1}H spectra, the structures can be easily elucidated. The doublet centered at 7.04 ppm in **Figure 3** (7.05 in **Figure 4**) relates to the *meta* proton of the aryl ring, as it is the most shielded due to its proximity to the electron donating amine group, in addition to the observed splitting caused by coupling to its neighboring proton environment. The broad peak at 7.90 ppm in **Figure 3** (7.94 in **Figure 4**) corresponds to the amine group in the molecule. The proton signal centered at 7.84 ppm refers to the *ortho* proton furthest from the nitro group, which should theoretically be a doublet of doublets due to coupling with its neighboring proton as well as long-range coupling with the other *ortho* proton, which is hidden due to overlapping with the amine signal. Finally, the most deshielded doublet at 8.54 ppm in both ^{1}H spectra refer to the other ortho proton, which is situated between two strong electron withdrawing groups (nitro and ester groups) and is split through long-range coupling.

For the ^{13}C spectra, we once again see a defining characteristic evident in the product, but not present in the starting material. In the spectrum of the product **2**, we see the methoxy carbon resonance appear at 51.79 ppm, which is not present in the starting material **1**. Aside from the methoxy carbon peak in the ^{13}C spectrum of **2**, there are 7 total peaks: 6 between 116 ppm and 149 ppm, relating to the aromatic peaks and 1 at 166.08 ppm for **1** (164.80 ppm for **2**), corresponding to the carbonyl resonance.

CONCLUSION

Using benchtop NMR spectroscopy, students are able to directly work with an NMR spectrometer in their labs and obtain ^{1}H and ^{13}C data within minutes after collecting their product. In this sample experiment, a Fischer esterification reaction was completed to yield the product, methyl 4-amino-3-nitrobenzoate (**2**). The ^{1}H and ^{13}C spectra of this product were obtained, analyzed and compared with the starting material, 4-amino-3-nitrobenzoic acid. Upon analysis, it was confirmed that the final product was successfully synthesized due to the appearance of the methoxy protons and carbons in the ^{1}H and ^{13}C spectra of methyl 4-amino-3-nitrobenzoate (**2**), respectively, that did not appear in that of the 4-amino-3-nitrobenzoic acid (**1**) starting material. As NMR spectroscopy is one of the most powerful structural elucidation tools available to chemists, this experiment demonstrates the potential of benchtop NMR instruments as tools for hands-on learning in an undergraduate laboratory.

References

[1] Yearty, K.L.; Sharp, J.T.; Meehan, E.K.; Wallace, D.R.; Jackson, D.M.; Morrison, R.W. *J. Chem. Educ.* **2017**, *94*, 932-935.

[2] The following references are examples of Fischer esterification reactions performed in undergraduate laboratories: (a) Reilly, M.K.; King, R.P.; Wagner, A.J.; King, S.M. *J. Chem. Educ.* **2014**, *91*, 1706-1709. (b) Brown, D.P.; Durutlic, H.; Juste, D. *J. Chem. Educ.* **2004**, *81*, 1016-1017. (c) Steele, J.H.; Bozor, M.X.; Boyce, G.R. *J. Chem. Educ.* **2020**, *97*, 4127-4132.

[3] Kam, C.M.T.; Levonis, S.M.; Schweiker, S. *J. Chem. Educ.* **2020**, *97*, 1997-2000.