

Exp't 812

Bakers' Yeast Reduction of α -Diketones

Adapted by Laura Nigbor (Penn State Univ.) from a micro-scale experiment developed by Pascale Besse; Jean Bolte; and Henri Veschambre, published in *Journal of Chemical Education*.

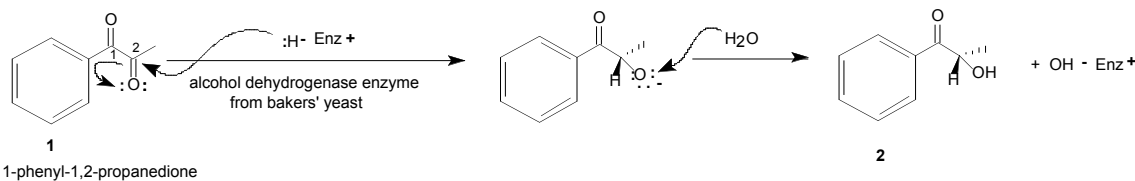
Introduction:

Lithium aluminum hydride and sodium borohydride are effective reducing agents. Treatment of a ketone with either agent will reduce the carbonyl group to yield an alcohol. However, these reducing agents do not react to form a chiral alcohol because the hydride can attack both sides of the planar carbonyl group yielding a mixture of both enantiomers.⁴ In order to achieve a chiral alcohol, a chiral reducing agent must be used.

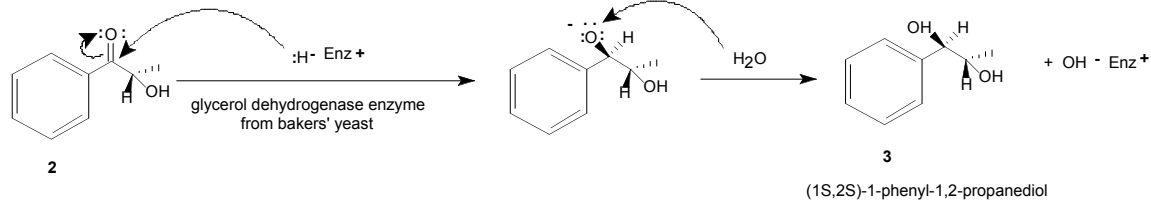
Chiral reducing agents such as the enzymes that result from the fermentation of bakers' yeast are effective in reducing ketones such as 1-phenyl-1,2-propanedione, **1**, to chiral alcohols such as (1R,2S)-1-phenyl-1,2-propanediol, **3**. The first enzyme to react is of the alcohol dehydrogenase type. This chiral enzyme attacks carbon 2 (seen in scheme 1) creating an (S)-configuration at that carbon. To reduce carbon 1, a second chiral enzyme from bakers' yeast, glycerol dehydrogenase type, is used. This enzyme has the opposite configuration of the first enzyme which leads to the resulting (1R,2S)-diol.²

The reaction of **1** with bakers' yeast to yield **3** follows the mechanism below. In this experiment, (1R,2S)-1-phenyl-1,2-propanediol forms in 65-70% yield.² Ideally, the optical purity would be tested by polarimetry to verify that the diol formed is indeed 1R,2S. In some labs this is not possible so ¹H NMR is used to verify that the correct product was formed, as ¹H NMR cannot give information about stereochemistry.

Scheme 1



Scheme 2



Pre-laboratory Exercise: (Include in Introduction Section)

1. Use Prelog's Rule to explain why the hydride in Scheme 1 attacks the carbonyl at the site shown.²

Cautions:

- Vent the separatory funnel often during extractions.
- The reaction of organic product with MgSO_4 is exothermic. Add MgSO_4 slowly and stir until the reaction is complete.
- Diethyl ether is flammable, extractions should be done in the hood.

Synthesis:

Using a 500 mL Erlenmeyer flask, suspend 20 g of freeze-dried baker's yeast in 250 mL of water. Add 0.227 mL 1-phenyl-1,2-propanedione to the flask and using a magnetic stir bar, react at room temperature for 1 h.

Isolation and Purification:

Filter mixture on Celite 545 after 1 h of stirring. Rinse flask with 25 mL water and 25 mL ether. Saturate the filtrate with NaCl (about 70 g) and extract three times with 30 mL ether. If an emulsion occurs during the extraction, remove the emulsion and add MgSO_4 . The organic layer is dried on solid MgSO_4 . The ether solvent is removed by rotary evaporation and the resulting dark yellow product is purified by column chromatography. Fill the column with 1.5 g silica gel using dichloromethane as the eluent. Dissolve the diol in minimal dichloromethane and obtain a first fraction with 20 mL dichloromethane. The second fraction is obtained with 3 mL ether and the last two with 15 mL ether each. Allow the solvent to evaporate overnight.

Cleaning-Up:

The light brown solid remaining in the Hirsch funnel and the solid MgSO_4 should be dried in the hood then disposed of in the trash bin. The aqueous layer from the extraction can be washed down the drain with water.

Analysis:

A TLC plate should be run on both the starting material, **1**, and the crude product, **3** (before column chromatography) using a 50/50 mixture of cyclohexane/ether as the eluent. Carefully calculate and mark R_f values for both samples. Use TLC plates to identify which fractions contain the diol. Take a ^1H NMR of the crude product. The purified product is to be analyzed by ^1H and ^{13}C NMR, GC-MS, and IR. The percent yield of both the crude and purified products should be calculated.

Final Report:

Compare the calculated yield with the expected yield given in the introduction discuss causes for any differences. Attach TLC plates with R_f calculations shown clearly and interpret ^1H NMR data. Include answers to the following questions in the Discussion section of the final report.

1. Explain why the reaction is run at room temperature.
2. To purify the product, column chromatography was used. Explain why the first fraction was obtained using dichloromethane while the second and third fractions were collected with ether.²

References:

1. Bakers' Yeast Mediated Enantioselective Reduction of Phenothiazine Based Prochiral Ketones (accessed December 2002)
<http://www.bio.unibuc.ro/biochemistry/Enzimology/Vol6No4-2001/040406-2001.pdf>
2. Besse, Pascale; Bolte, Jean; Veschambre, Henri, Bakers' Yeast Reduction of α -Diketones. *J. Chem. Ed.* **1995**, *72*, 277-278.
3. Chemfinder <http://chemfinder.camsoft.com> (accessed November 2002).
4. McMurry, J. *Organic Chemistry*, 5th ed.; Books/Cole New York, 2000.
5. 2001 Chemistry Nobel Prize (accessed December 2002)
<http://www.ias.ac.in/resonance/feb2002/pdf/feb2002p55-65.pdf>