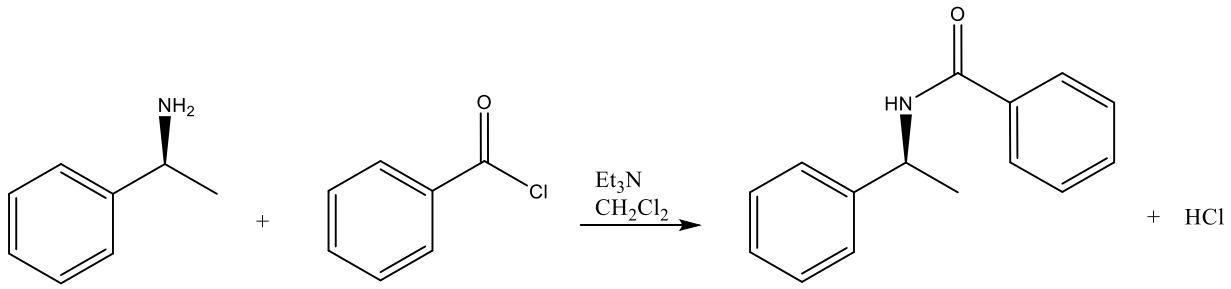
Preparation of *N*-(1-phenylethyl)benzamide and Chiral HPLC Analysis

Reading

Klein (3rd ed.): 20.8; Pavia (5th): Techniques: 11.10 (especially important), 21

Introduction

A sample of the enantiomerically enriched (S)-α-phenylethylamine produced in an earlier experiment will be converted into the corresponding benzamide by reacting the amine with benzoyl chloride (an acid chloride). The crude benzamide will be purified using a mixed solvent crystallization technique. The purified amide will be analyzed on a chiral HPLC column. The benzamide derived from the (S)-amine will separate from the minor (R)-benzamide on the special column used for this HPLC procedure.



Enantiomers generally cannot be separated by ordinary chromatography techniques (for example on a C-18 modified silica gel column typically used in HPLC). In the case of a chiral column, the stationary phase is either comprised of a single enantiomer of a chiral compound, or consists of silica gel modified with a chiral ligand. The two enantiomers in the analyte solution have slightly different interactions with the chiral stationary phase causing one enantiomer to experience a longer retention time than the other isomer. If difference in retention times is sufficient, baseline resolution of the isomers can be achieved. The column that will be employed in this experiment uses structurally modified cellulose as the stationary phase. Cellulose is a long-chain polymer of chiral D-glucose molecules (cellulose is found in nature in the walls of plant cells). As it is composed of only the D-isomer of glucose, cellulose can serve as a chromatographic resolving agent. Unfortunately, the enantiomers of unmodified α-phenylethylamine do not readily separate on this column, but the benzamide derivatives do. By converting the amine to the benzamide, the enantiomers can effectively be separated. Integration of the chromatogram will provide another means to assess the optical purity of the resolved amine.

Preparation

Your prelab summary should include the usual components (**Name, Date, Title, Purpose, Reaction Equation, Reagent Table, and Outline).** The reagent table should include stoichiometric data and the calculated percent yield of the product. Be sure to include density values for the reagents (all of which are liquids) and the melting point of the product.

Procedure

Reaction

Using an autopipette, transfer 0.27 mL of your retained α-phenylethylamine to a pre-weighed 25 mL round bottom flask (microscale joint). Weigh the flask with the amine to find the actual mass of the reactant added. Add 2 mL of

methylene chloride and 2 mL of triethylamine to the flask. Clamp the flask to a ring stand, add a magnetic stirring bar, and attach a Claisen adapter. To the center joint of the Claisen head attach a rubber septum—to the sidearm attach a drying tube filled with anhydrous calcium chloride. Using an autopipette, dispense 0.25 mL of benzoyl chloride into a 3 mL conical vial (caution—dispense the benzoyl chloride in the fume hood)—add 0.5 mL of methylene chloride to the vial. Cap the vial and gently shake to dissolve the benzoyl chloride. Draw the benzoyl chloride solution into a 1 mL syringe with an attached needle. Insert the needle into the septum of the Claisen adapter. Add the benzoyl chloride solution slowly over a 15 min period while stirring the reaction mixture. After adding the benzoyl chloride, continue to stir the mixture for an additional 45 minutes. The reaction mixture should become cloudy during this time.

Workup

At the end of the reaction period, add 2 mL of 2 M NaOH solution and stir the mixture for several minutes. Transfer the mixture to a 60 mL separatory funnel, rinse the round bottom flask with about 2 mL of methylene chloride and transfer the rinse solution to the funnel. Remove the bottom methylene chloride layer and set aside. Drain the upper aqueous layer from the funnel. Return the methylene chloride solution to the separatory funnel and wash (i.e., extract) with 2 mL of 2 M HCl solution. Again, drain the lower organic layer from the funnel, followed by removing the aqueous phase. Return the methylene chloride solution to the funnel one last time and wash with 2 mL of DI water. Drain the bottom organic layer into a clean, dry 50 mL Erlenmeyer flask. Add small quantities of anhydrous sodium sulfate until the solid ceases to clump and free-flowing crystals are observed. Allow the organic solution to dry over the sodium sulfate for 10 minutes. Carefully transfer the solution to a clean, dry 25 mL round bottom flask (large joint) leaving the solid drying agent behind. Remove the methylene chloride on the rotary evaporator.

Crystallization of the Crude Product

Scrape as much of the solid from the round bottom flask as possible onto a piece of weighing paper. Using the paper as a chute, transfer the solid to a 25 mL Erlenmeyer flask. Place two 50 mL Erlenmeyer flasks, one containing about 20 mL of ethanol and a second containing about 20 mL of DI water on a hotplate—heat the solvents to boiling on a medium setting. Add hot ethanol to the crude benzamide until it just dissolves. Be sure to heat the mixture and swirl in between additions of ethanol. Once the solid has dissolved, gradually add hot water to the mother liquor until a slight cloudiness is observed. At this point, add hot ethanol dropwise until the solution is again clear. Remove the solution from the heat and allow to cool slowly to room temperature during which time crystallization should commence. Continue to cool the suspension in an ice bath. Collect the crystal by vacuum filtration in a Hirsh or Buchner funnel. Wash the crystals with ice-cold water. Allow the crystals to dry on the vacuum apparatus for several minutes. Transfer the crystals to a pre-weighed, 50 mL beaker and dry in the oven for 30 min. Reweigh the beaker and solid to obtain the mass of the purified product.

Analysis of Product

Measure the melting point of the benzamide and record the infrared spectrum using the KBr pellet sampling method.

HPLC Sample Preparation

Dissolve approximately 5 mg of the purified benzamide in about 1.5 mL of HPLC grade methanol. Transfer the solution to an auto-sampler vial while passing it through a 0.2 μm syringe tip filter (the instructor will review the use of the filter). Cap the vial and place in the auto-sampler rack.

Completing the Experiment

In addition to the melting point and IR spectrum, collect the HPLC chromatogram of the product (integrate just the two major peaks which should be the peaks for the two isomeric benzamides). Calculate the %ee of the mixture using the HPLC data.