From: https://www.thevespiary.org/rhodium/Rhodium/chemistry/expt21_1corr.htm

Revised Procedure: Expt. 21.1 Multistep Synthesis of Benzocaine Analogs

Detailed Procedures Incorporating the Changes:

Part A: Miniscale Synthesis of *p*-Acetotoluidide

Prepare a solution of sodium acetate by combining 2.15 g of sodium acetate trihydrate ($CH_3CO_2Na \cdot 3\ H_2O$) and 5-6 mL of water in a 10-mL Erlenmeyer flask. Swirl vigorously to dissolve. Set the sodium acetate solution aside for later use.

In a 125-mL Erlenmeyer flask fitted with a stir bar, dissolve 1.61 g of p-toluidine in 40 mL of water. With stirring, add 1.3 mL of concentrated HCl. Stir for 2 minutes. With stirring, add 2.1 mL of acetic anhydride and immediately add the sodium acetate solution. Stir vigorously to mix the reagents. Cool the solution in an ice bath and continue to stir vigorously while the product crystallizes. Isolate the product by vacuum filtration, washing the crystals with several small portions of ice-cold water. Let the crystals air dry or place in a warm drying oven. Weigh the product. Save at least 25 mg of the product for characterization and spectral analysis. The remainder may be used without purification in the next step.

Part B: Miniscale Synthesis of *p*-Acetamidobenzoic Acid

This procedure is designed for use of 1 g of p-methylacetanilide. To a 250-mL Erlenmeyer flask fitted with a stir bar, add 1 g of dry *p*-methylacetanilide (or all of the remaining solid from part A), 2.6 g of magnesium sulfate heptahydrate ($MgSO_4 \cdot 7\ H_2O$) and 64 mL of water. Heat to 85oC on a steam bath or water bath. While vigorously stirring the solution of *p*-methylacetanilide, slowly add via pipet a hot solution of potassium permanganate (previously prepared by dissolving 2.6 g $KMnO_4$ in 14 mL of boiling water). The addition should take approximately 30 minutes. It is important to add the permanganate solution slowly and uniformly to avoid local build up of the oxidant. After all of the oxidant has been added, add 2 mL of ethanol, stir vigorously, and bring to a boil. Check to make certain that no purple color remains, then filter over a pad of Celite using suction filtration, washing with water to dissolve any adsorbed product. Transfer the clear solution to an Erlenmeyer flask. Cool the filtrate in an ice bath and acidify with 20% sulfuric acid until the pH is 3-4. Collect the product using vacuum filtration, rinsing the crystals with small amounts of ice-cold water. Dry the crystals as much as possible by continuing suction. The product does not need to be completely dry for the next step. Save at least 25 mg of the product and let it dry thoroughly for characterization and spectral analysis.

Part C: Miniscale Synthesis of *p*-Aminobenzoic Acid

Add 1.0 g of *p*-acetamidobenzoic acid and 5 mL of 6M HCl to a 10-mL round-bottom flask containing a flea bar. Attach a reflux condenser to the round-bottom flask and reflux gently, with stirring, for 30 minutes. After the heating time is over, let cool to room temperature. Transfer the contents of the flask to a 50-mL Erlenmeyer flask, rinsing with 2.5 mL of cold water. Add the rinses to the flask. Add concentrated (15 M) ammonia dropwise until the pH is between 7 and 8. Do not go beyond pH 8. During the addition, precipitates will form and redissolve. Estimate the volume of the solution: then

add 1 mL of glacial acetic acid for every 30 mL of solution to induce crystallization. Stir vigorously and cool the solution in an ice bath. It may be necessary to add more glacial acetic acid, to add a seed crystal or to scratch the inside surface of the flask with a glass rod. Suction filter the product and let air dry until the next lab period. Weigh the dry product. Save 25 mg of product for analysis and use the remainder in Part D.

Part D: Microscale Esterification of *p*-Aminobenzoic Acid

The instructor will assign each student or pair of students a specific primary alcohol to be used to esterify *p*-aminobenzoic acid.

To a 10-mL round bottom flask fitted with a stir bar, add 0.50 g of dry *p*-aminobenzoic acid and 3.8 mL of the assigned alcohol. While stirring, add 0.38 mL of concentrated H_2SO_4 dropwise. The precipitate that forms upon the addition of sulfuric acid should dissolve when the solution is heated. Reflux, with stirring, for 1 hour. Cool to room temperature, then transfer the solution into a centrifuge tube. Neutralize cautiously with dropwise addition of 10% Na_2CO_3 until the pH is approximately 8. (Gas evolution will be vigorous.) Extract with two 3-mL portions of methylene chloride. Wash the combined methylene chloride layers with two 8-mL portions of water. Dry the methylene chloride solution over anhydrous sodium sulfate. Gravity filter into a clean Erlenmeyer flask containing a boiling stone. Evaporate the methylene chloride under the hood on low heat. Recrystallize the whitish residue using as a solvent pair, the assigned alcohol and water. Suction filter the product, and let air dry. Weigh the white crystals.

Testing the Efficiency of Sunscreens:

Sarratia marcescens is a bacterium that is red when grown at room temperature. This procedure will examine the effectiveness of sunscreens and sunglasses in absorbing UV radiation. Work in pairs or small groups, as directed by the instructor. Each group should obtain 4 nutrient agar plates, 4 sterile swabs, a sample of Serratia marcescens in nutrient broth, a cardboard cutout, clear plastic wrap, and mineral oil. Wear gloves and goggles at all times!

Label the nutrient agar plates as A, B, C, and D. Swab all four plates with the broth culture. Be sure to cover the whole plate.

Plate A: Plate A will serve as the control. Set it aside until ready to incubate all of the plates.

Plate B: Place the cardboard cutout on top of the plate. Place the plate in a UV chamber, about 12" from the source. Expose the plate to 260 nm UV light for 3 minutes. Remove the cardboard cutout and replace the lid.

Plate C: Prepare a nujol mull of the benzocaine analog. Place a small piece of clear plastic wrap over the cutout and smear a thick layer of the mull on top. Remove the lid and place the plastic wrap and cutout on the plate. Expose the plate to 260 nm UV light for 3 minutes. Then remove the

cardboard cutout and replace the lid.

Plate D: Place a pair of sunglasses on top of Plate D. Expose to 260 nm UV light for 3 minutes. Remove the sunglasses and replace the lid.

Invert the plates and incubate at room temperature for 24 hours. At the end of this time, examine the plates for growth. Pool the results. Based upon the areas of growth, analyze the effectiveness of the benzocaine analogs as sunscreens.