Keira Johnson Pre-Lab

Lab 2

**Purpose**: To (1)determine the number of, and identify, the organic compounds in each analgesics and (2) study how solvent polarity effects Rf.

**Procedure Outline**

* **Part 1: analysis** 
  + Unknown sample preparation
    - Make a 1% solution of the 2 unknowns that are assigned. To do so, take a small amount of the crushed sample and put it in the 1-dram vial. Add a few drops of ethanol and label this with the unknown value
    - Take the known substances, aspirin, acetaminophen, and caffein and label these vials. Add half-a full dropper of the known solutions to these vials
    - Grab 4/5 TLC plates, 1 ruler, a pencil, a capillary spotter, and a paper towel
    - Fill the 25 mL Erlenmeyer flask halfway with acetone
  + Preparing and spotting the plate
    - The powdered side of the plate is spotted, NOT shiny
    - Very light, draw a pencil line about a half centimeter from an end of the plate, do NOT scratch the plate too deep. Make 3 spots along the line representing where the samples will be spotted.
    - Label and spot accordingly:
      * Plate 1 = aspirin, acetaminophen, and unknown 1
      * Plate 2 = the same as 1, except use unknown 2 instead of 1
      * Plate 3 = the same 2 knowns as well as caffeine
      * To perform a spot, dip a micropipette into the sample and make the spot as small as possible. Do NOT hold spotter to the plate, but just tap and release. Do this several times to concentrate the sample. Check the spots under the UV light
      * \*do NOT look into the UV light directly
      * If the spot is too large after checking with the UV light, prepare a new plate, if it is too small, spot again, no need for a new plate
      * Rinse the spotter with acetone between every use. Dip the spotter in acetone, then lightly on the paper towel. Repeat 4-5 times to ensure a through rinse
      * Use a micropipette to obtain a sample of the known solution. Dip the pipetter in the solution to fill.
  + Development
    - The chamber is a 150 mL beaker with a 5.5 cm filter paper inside. Cover the chamber with aluminum foil
    - Place 2 mL of ethyl acetate in the chamber. (fill the bulb of a Pasteur pipet)
    - Place the TLC plate with tweezers into the beaker with the spotted side at the bottom. Replace foil. Make sure to keep the beaker covered with foil at all time besides when inserting/removing the plate.
    - \*\*\*MAKE SURE:
      * The solvent is below the level of spots
      * The plate has to vertically stand in the beaker. The gel can not touch the filter paper, and the solvent can not run to the top of the plate
      * The correct amount of material is spotted
      * Do NOT touch the beaker while the plate is running
    - After the solvent is almost at the top of the plate, remove it and mark the position with a pencil before the solvent evaporates. \*\*\*\* must always allow the solvent that is developing to run ALMOST to the top of plate
    - Dry the plate in the hood, development should take a few minutes. if the plate comes out badly, redo the plate
  + Visualization
    - Hold the plate closely to the UV light to see the spots. The observed spots should then be outlined with pencil
    - Place the plate in a jar with iodine crystals for a few minutes. Keep it lidded. Circle any new spots observed
    - \*\*\*The UV method must be done before the iodine method or else compound may react with iodine, possibly changing results
    - After these steps are done, take a photo of the plate. It should be taken while under UV light. IN the picture make sure to include the ruler next to the pkate to use for scale
    - Used plates will go o the solid waste container
    - Calculate the Rf values for the known compounds for ALL components of the unknowns. Then, identify the components in the unknowns. More than one may be in the unknown. \*\*\*if a spot if purposely overloaded, this can now help determine a component that is present in small amounts
* PART 2 : solvent effect on Rf values
  + Anthracene, benzil, and triphenylmethanol will be developed in 2 different solvents
  + Prepare 2 plates spotted with each
  + Develop one plate with ethyl acetate
  + Develop the other plate with a mixture of 95% hexane and 5% t-butyl methyl
  + Run one of the plates, then discard the solvent in liquid waste. Dry the beaker. Now aff the second solvent and run the other plate.
  + Only look at the plates with the UV light. Circle with pencil any spots that come up
  + Take picture of these plates under UV light. Label clearly. Put the TLC plates in solid organic waste container
  + Calcluate Rf values and discuss effect of solvent on Rf
* \*\*\* DO NOT keep the TLC plates. The silica will flake
* SAFETY
  + Do NOT stare into UV light
* WASTE
  + Dispose of glass in “GLASS ONLY” boxes
  + Used capillaries should be placed in glass waste or in the designated dishes
  + Dispose of the developing solvents and solutions in “Organic Liquid Waste”
  + Pour most of the solvent as possible in waste container and put the beaker back in designated drawer
* WHEN LEAVING
  + Return all supplies to designated spots
  + Turn off all electrical equipment
  + Clean work area
  + Close fume hood sash