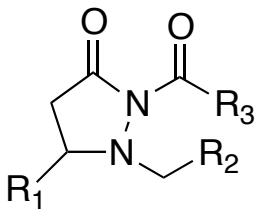
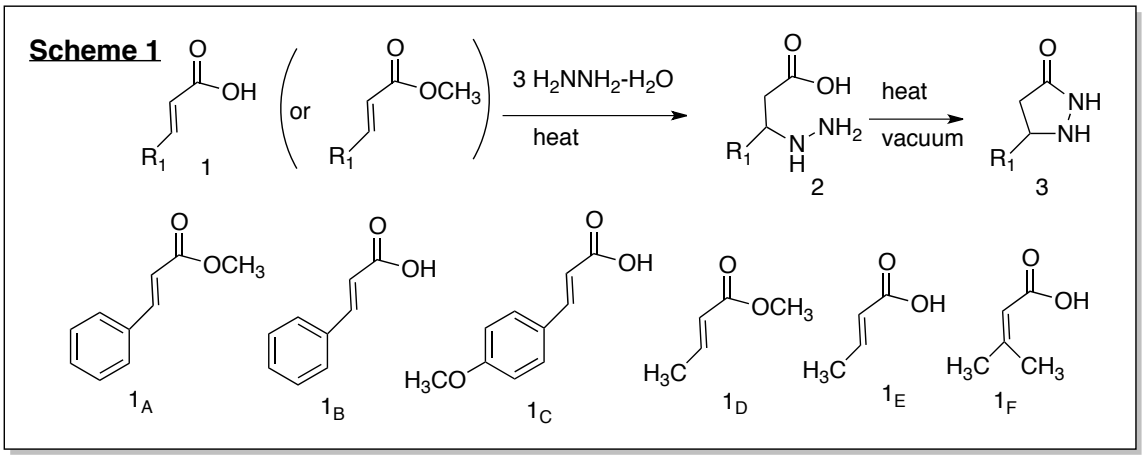
Multistep synthesis of an Acyl Pyrazolidinone

Introduction

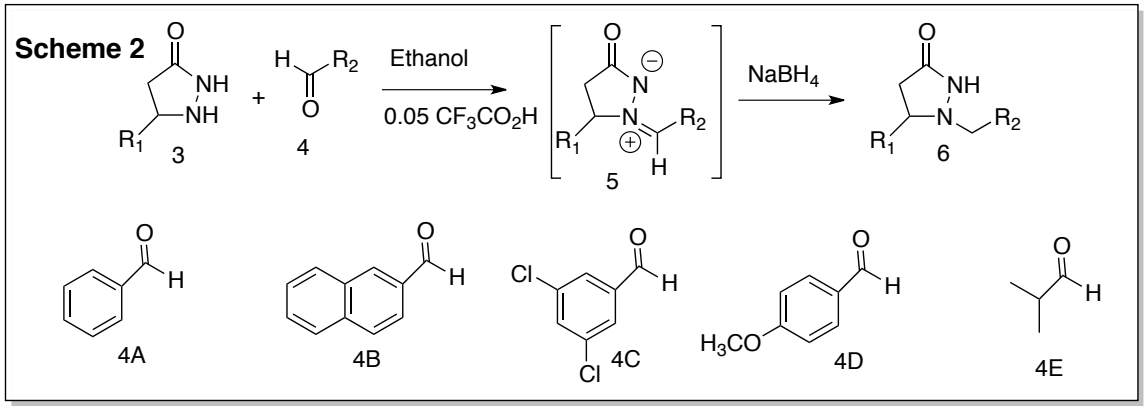
We will begin a multi-step semi-research sequence in which we make a library of different "acyl pyrazolidinones". The project will illustrate a variety of fundamental organic reactions and fundamental procedures. The acyl pyrazolidinones are of interest as potential medicinal candidates, as well as for use in further synthetic reactions. (It's possible that students in next year's research module will use some of the products.)



As shown in the picture, there are three different variable R groups. We will use four different R_1 groups; four different R_2 groups; and four different R_3 groups. This will produce a library of up to 4 x 4 x 4 = 64 different products, many of which have never been synthesized before. This kind of modular preparation of chemical libraries is routine in industry and medicinal chemistry. Subsequent biological testing can provide insights into the relationships between structure and activity. (Which parts of the molecule matter? Where does getting bigger or smaller help? Etc.)



In the first stage, an unsaturated acid or ester will be treated with excess hydrazine hydrate (Scheme 1). The nitrogen will initially add nucleophilically to the alkene, in a mechanism that can be viewed as involving either an enol or an enolate intermediate. This reaction is done in the absence of any solvent other than the hydrazine hydrate itself. Following addition, the product **2** undergoes loss of water and cyclic amide formation under high-temperature and vacuum, again with no solvent. The vacuum is important because it helps to distill off the water, whose hydrogen-bonding otherwise inhibits the ring closure. The product rings **3** are referred to as pyrazolidinones. They are often thick and gummy. Both the intermediate (**2**) following initial heat and the product (**3**) following the vacuum heating will need to be evaluated. NMR, GC-MS, and TLC are three analytical techniques that might be used. In a "real" synthetic lab, a subsequent step (like **2 → 3**) would not be attempted until after there was assurance that the earlier step (**1→2**) had been completed successfully. In our case, we will need to trust that the original conditions are pretty effective, and suffer any losses that come if in fact that conversion isn't so great.

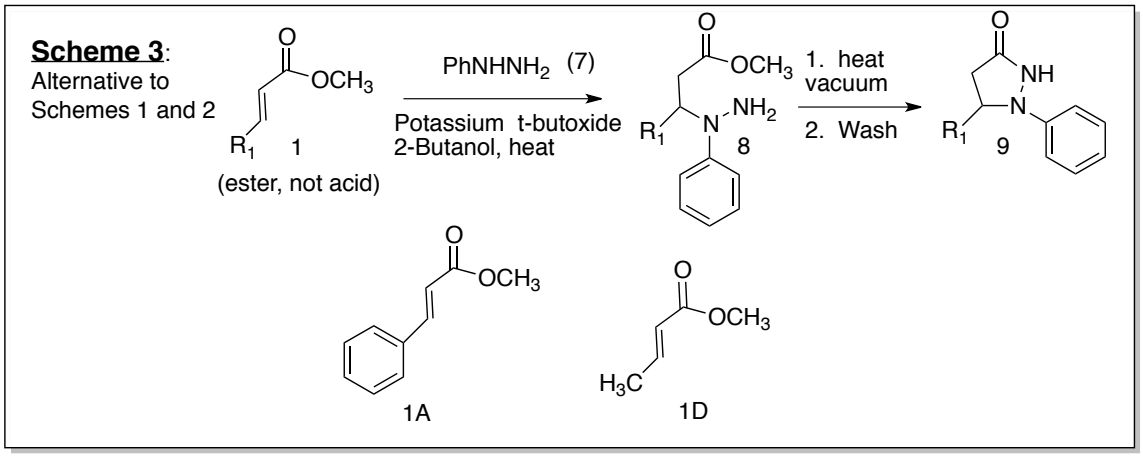


In the second stage (Scheme 2), the bottom nitrogen will exchange a hydrogen for a new carbon substituent, CH_2R_2. This will be done via a reductive amination using an aldehyde. Initially a C=N bond forms, which effectively forces a formal positive charge onto the bottom nitrogen; this in turn makes the top nitrogen so acidic that it probably gets deprotonated in solution to give intermediate **5**. Structure **5** has two formal charges, but is overall neutral. A molecule with this kind of situation is referred to as being an "ylide", comparable to the phosphorus ylides involved in the Wittig reaction. Acid is probably required to catalyze the formation of **5**. (If two people or groups wanted to do side-by-side reactions one with and one without the acid, it would be interesting to compare the two reactions…)

It is noteworthy that the two nitrogens in pyrazolidinones 3 behave very differently. The bottom nitrogen is sp^3-hybridized and is basic/nucleophilic, essentially like an "amine" nitrogen. The top nitrogen is sp^2-hybridized and is non-basic/non-nucleophilic because it is essentially an "amide" nitrogen, and is stabilized by conjugation to the carbonyl.

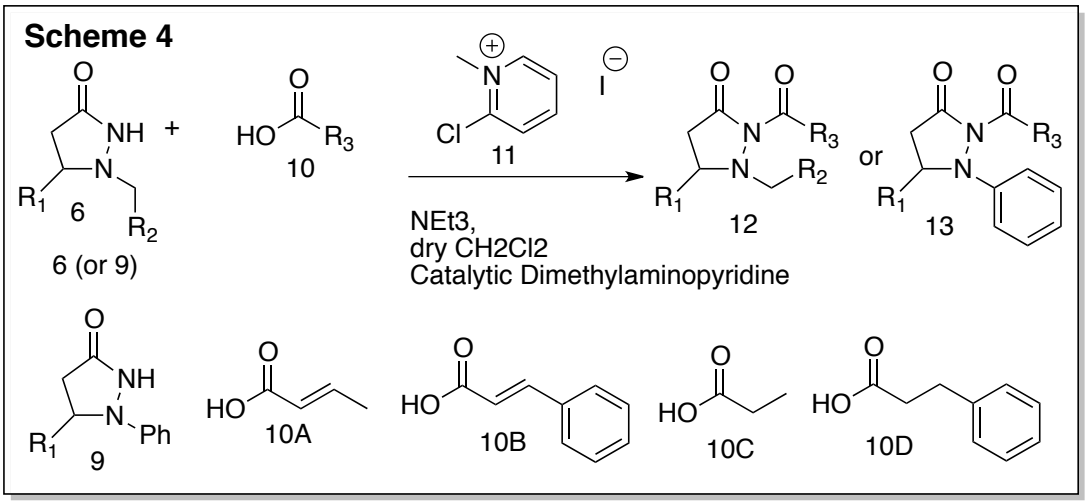
The product **5** can be evaluated by NMR; the hydrogen on the C=N carbon typically appears somewhere in the 8-9 ppm area.

The product **5** will then be reduced with sodium borohydride, which provides a nucleophilic hydride that attacks the electrophilic C=N(+) bond. The borohydride also reacts with the ethanol to produce hydrogen gas and bubbling; thus the $NaBH_4$ reaction needs to be conducted cold, in a container with some space for foaming, and with restrained addition of the sodium borohydride. Because product **6** will be produced in a solvent (ethanol), with lots of sodium and boron as well as other side products around, purification will be required. Many of the side products will hopefully be removed by a series of extractions (separatory funnel), followed perhaps by rotary evaporation, and perhaps also high temperature vacuum distillation. Depending on the quality of the product **6**, purification by automated chromatography ("combiflash") may also be required.



Scheme 3 shows an alternative route to pyrazolidinones **9** which have an N-phenyl substituent, in contrast to pyrazolidinones **6** which have an N-alkyl substituent . In this scheme, one of the phenylhydrazine nitrogens (see **7**) already has a carbon attachment (phenyl). The conjugating phenyl group differentiates the two nitrogens, and renders the substituted nitrogen acidic enough to be deprotonated by the alkoxide base, since the transient nitrogen anion is stabilized by conjugation/resonance. Unfortunately alkyl hydrazines don't work, because alkyl substituents don't facilitate the anion formation. Once the nitrogen anion forms, it adds to the alkene **1** so as to make a transient enolate anion, which in turn gets protonated to form product **8**. Unlike in Scheme 1, shown earlier, esters but not acids are required in Scheme 3. Acids get deprotonated and then don't do anything. Once the addition is completed (**1 →8**), heat and vacuum can again be used to close the ring (**8 → 9**). This is an amide-forming step, and basically corresponds to a downhill Cl-A-vE-N-O reaction. Isolation and purification of product **9** will be needed. With luck, a separatory-funnel type extraction workup followed by some drying and rotary evaporation might be sufficient.

The last stage (Scheme 4) will involve acylation of the top amide nitrogen. The procedure has been invented and developed by MSUM students (most notably Amie Nowacki and Kris Brandvold). A water molecule effectively needs to be eliminated (H from the amide nitrogen in structure **6**, OH from the carboxylic acid **10**). The water oxygen gets absorbed by "Mukayama's Reagent" **11**, and the two H's end up getting absorbed by basic triethylamine. Dimethylaminopyridine may function as an essential catalyst. (It would be interesting to have two people or two groups try to run the reaction with and without the dimethylaminopyridine to see whether it is actually necessary.) Since the function of **11** is to absorb water, the solvent needs to be dry, so that Mukayama's agent acts on reactants **6** and **10** rather than getting destroyed by water in the solvent. This reaction may take several hours or more. So it may be desirable to start it at the end of one lab period, and work it up during the following period.



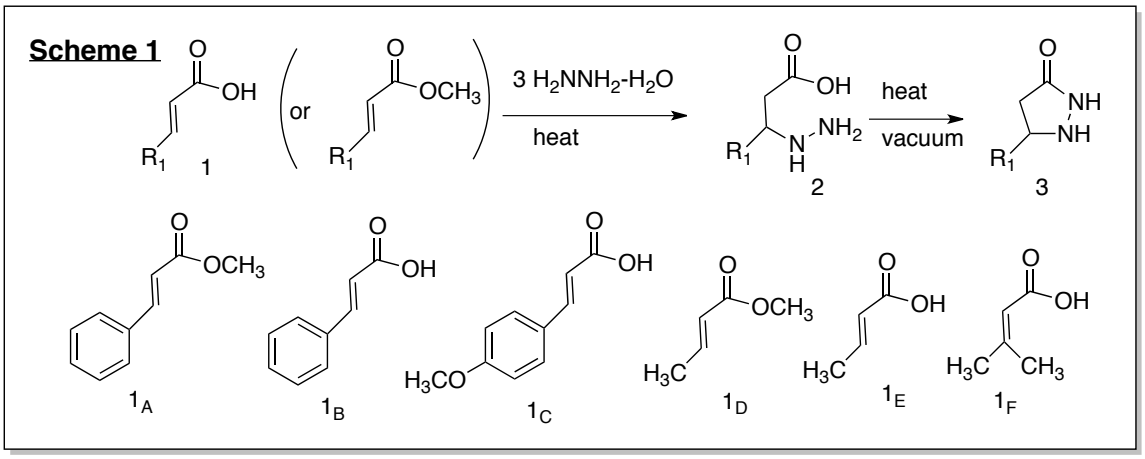
The acyl pyrazolidinone products **12** and **13** will be formed in solvent and accompanied by multiple side products. Purification will involve liquid-liquid extraction (separatory funnel), some high-speed vacuum-accelerated column filtration/chromatography, and rotary evaporation. Automated combiflash chromatography may be needed, depending on how pure or impure a product is. As usual, NMR and GC-MS will be used to monitor the crude and purified samples.

There is a fair chance that products **12** and **13** will be contaminated by a structural isomers in which the acyl group is actually connected to the amide carbonyl oxygen rather than to the amide nitrogen. As of this writing, an easy solution to this has not been identified, nor has a way to get the wrong isomer to convert over to the desired isomer been found. If time permits, some experiments to explore possible solutions may be appropriate.

Overall Format and Initial Assignments: You can work with a partner. Each hood has a number above the top of it. Targets will be assigned based on which hood you are working in.

Initial Assignments:
Hoods 1 and 2: Use phenylhydrazine **7** and methyl cinnamate **1A**. (Scheme 3)
Hoods 3 and 4: Use hydrazine hydrate and cinnamic acid **1B**. (Scheme 1)
Hoods 5 and 7: Use hydrazine hydrate and dimethylacrylic acid acid **1F**. (Scheme 1)
Hoods 8 and 9: Use hydrazine hydrate and crotonic acid **1E**. (Scheme 1)
Hood 12: Use hydrazine hydrate and methoxycinnamic acid **1C**. (Scheme 1)
Hoods 13 and 14: Use hydrazine hydrate and methyl cinnamate **1A**. (Scheme 1)

Note: Hoods 1 and 2 may have a longer period pursuing Scheme 3 during the first week, compared to the other hoods 3-14 who will be pursuing Schemes 1 and 2. But hoods 1 and 2 should get a shorter experience the second week. If you want to "trade" your assignment with somebody from hoods 3-14 who will be pursuing Scheme 1 rather during the first week, you can do that.



Scheme 1 Procedure: Formation of Pyrazolidinone Ring Using Hydrazine

Turn your hot-plate heater to setting of 7. Use either a 125-mL ground-glass jointed Erlenmeyer or a 100-mL round-bottomed flask. Add a medium stir-bar. Weigh the combination and record the mass. Clamp the array securely at the bottom neck.

Add 20 mmol of whichever alkene acid (or ester) **1** that you are using. Then add 60 mmol of hydrazine hydrate (50.06 g/mol, d = 1.032 g/ml, 48.5 ml/mol, 0.0485 mL/mmol). Set the flask with the condenser directly on the hot plate and stir for 15 minutes. The condenser should have a gentle water flow.

Be prepared to work fast for what you will want to do after the 15 minutes is complete. During the 15 minutes, prepare the following:

- two long-stemmed pipets and pipet bulbs ready;
- two NMR tubes standing inside an empty Erlenmeyer;
- the other partner's reflux condenser, without any water tubing;
- a vacuum adapter hooked up to the vacuum hose, and plugged into the top of the new reflux condenser;
- either a rubber septum or stopper that will fit your flask.

After the 15 minutes is complete, swing the clamped array off of the hot plate. Re-set the hot-plate to a setting of 6. Get a glove for your hand, and while holding the hot flask with the gloved hand, twist the reflux condenser off with the other hand. Rapidly dip into the mix with your long-stemmed pipet and draw up about half an inch of material, and set the pipet into an NMR tube. The material will probably harden. If it doesn't, try to squirt it into the NMR tube. Whether it hardens or not, just take the pipet bulb off for now and let the NMR tube just sit there with the pipet inside it for later attention.

As soon as the analysis sample has been removed, insert the new water-tubing-free reflux condenser back into your jointed Erlenmeyer, attach the vacuum adapter to the vary top, turn the vacuum on gently (so that it doesn't cause everything to erupt), and swing the mixture back onto the hot plate. Stir/heat/vacuum for ten minutes, then immediately swing the array off of the heat again. Detach the vacuum hose from the adapter, and turn your vacuum off. As before, grip the flask with a glove, remove the reflux condenser while the mixture is still hot, pipet out a quarter inch sample of your mixture, and put it into your second NMR tube. As before, the material will probably harden. If it doesn't, try to squirt it into the NMR tube. Whether it hardens or not, just

take the pipet bulb off for now and let the NMR tube just sit there with the pipet inside it for later attention. Do not put the reflux condenser back onto the flask this time. Insert a rubber stopper or a septum instead. Other than analysis, that flask and sample are now ready for N-alkylation as described in Scheme 2. If you have time, proceed with that, or the first part of that, immediately. If not, store the flask with the septum or stopper into your drawer and save it for next lab period.

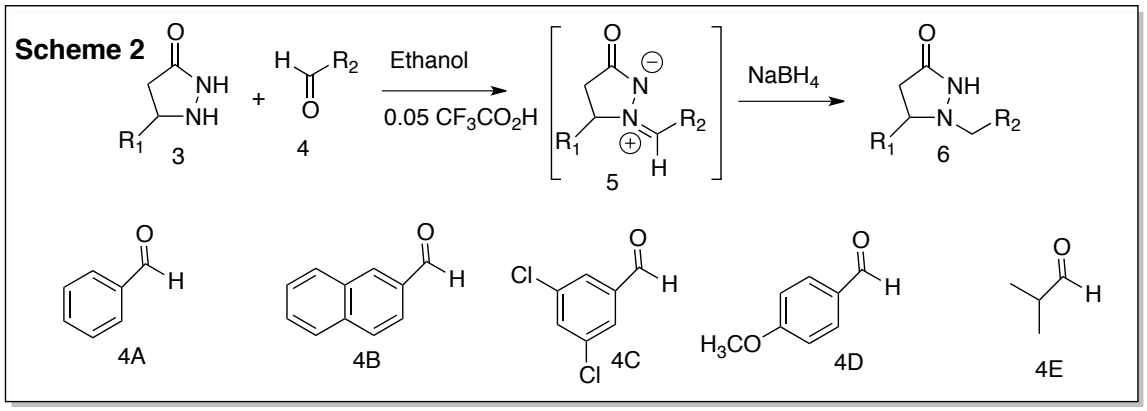
For each of the two NMR's (keep straight which is which), each of which should still have their long-stemmed pipets resting in them, add CDCl3 directly into the pipets. This will help to rinse/dissolve material that will be plugging a pipet or sticking to the sides of the pipet. It may be that the sample will have hardened and will be blocking the pipet. In this case, get the heat gun over and try to focus some heat so as to melt the hardened material. The instructor can help you with this if necessary. Add enough CDCl3 so that the NMR tube is half full, and use the pipet to rinse and flush by sticking the pipet in as far as it can into the NMR tube, drawing some liquid up that it can reach, and shooting it back into the tube, etc. This should ensure that all of your material will have gotten into the NMR tube. Then use the same pipet to remove some of the CDCl3 solution from the NMR tube and transfer it into a GC vial. The NMR tube should be left 1/3 full with solution. Dilute the GC vial to the 0.5 mL mark with either CDCl3 or CH2Cl2.

Analysis: Submit the two NMR's for proton analysis. (Perhaps we'll run 2-dimensional H-H correlation spectroscopy (COSY) and or 2-dimensional H-C correlation spectroscopy as well to help evaluate the samples. If everything is good, the second sample could be simpler and cleaner to look at. But if that doesn't turn out so clean, it could be helpful to look at the first sample from prior to the vacuum.

Likewise submit the two GC's for GC-MS analysis. These may take longer, since some of the products are fairly big and might not come off very fast. In some of our labs we haven't really analyzed the mass spectra, but if samples aren't very clean (as seems well possible), it might be helpful to look at the mass spectra for the different peaks.

Weigh your flask. The balance closest to the acetone hood will probably not work (the flasks will likely be too heavy), but the other three should. Calculate your mass of product by subtracting the original mass of the flask and the stirbar. Calculate the molar mass of your product, calculate your theoretical yield, and calculate your percent yield. It may well be over 100%, if the vacuum didn't remove all of solvent and side materials. If it is well below 100%, is it possible that a significant amount of sample splashed up into your reflux condenser? If so, you could weigh your reflux condenser and evaluate the amount of yield inside it.

Depending on the time, you might have time to start Scheme 2, or else this might be a good time to quit for week one. If the latter, then stopper your flask and clean everything up.



Assignments:
Hoods 3, 5, 8, and 13: Benzaldehyde **4A**. (Scheme 2)
Hoods 4, 7, 9, and 14: Use napthaldehyde **4B**. (Scheme 2)
Hoods 12: Use methoxybenzaldehyde (anisaldehyde) **4D**. (Scheme 2)

Note: Hoods 1 and 2 will not be doing this reaction, since their nitrogen already has a phenyl group attached. See Scheme 3 procedure.

Scheme 2 Procedure: N-Alkylation Using Aldehyde

If you have time, you may wish to get this started during week one. Alternatively, you could conduct this during the second week.

To the flask with your product **3** from Scheme 1, add 30 mL of anhydrous ethanol. You may wish to heat this on the hot plate (setting of 5?) for a few minutes to try to warm up the ethanol and get the pyrazolidione to dissolve.

If you warmed the solution, cool it back to room temperature or below (with ice bath, perhaps). Once it's cooled to room temperature, add 20 mmol of your aldehyde and 1 mmol of trifluoroacetic acid (77.0 mL/mol). Stir for at least 15 minutes. Aldehyde **4B** might benefit from more time, 30 minutes.

Analysis: Using a long-stem pipet, carefully draw some solution into the skinny part of the pipet to about 1-1.5 inch, and transfer to an NMR tube. This can be diluted with CDCl3 and submitted for a proton NMR a little bit later.

For the next step, cool the solution in an icewater bath, and cautiously add 24 mmol of sodium borohydride (37.83 g/mol) in small portions. The reaction may be exothermic and it will produce hydrogen bubbling, so don't add too fast or things might foam over. Add carefully at first, and then add as fast as conditions allow. Once it's all in, remove the ice bath and then stir for 30 minutes at room temperature. During the 30 minutes, submit your NMR sample for analysis. Check with Dr. Jasperse on which experiment to run. (We want to make sure the proton NMR collects beyond 10 ppm.)

Workup: Add 20 mL of dichloromethane solvent, then cautiously add 20 mL of acqueous bicarbonate. Pour the mixture into a separatory funnel, and rinse the original Erlenmeyer with another 20 mL of aqueous bicarbonate and 20 more mL of dichloromethane and

add this to the separatory funnel. Shake the separatory funnel carefully, with venting. (There may be considerable pressure buildup?)

Prepare a pre-weighed 100-mL roundbottomed flask with a pre-weighed flea stir bar, and add a fritted filter funnel. Connect a vacuum tube, but don't turn it on yet. Add a 1.5-inch pile of sodium sulfate. Drain the lower dichloromethane layer directly onto the sodium sulfate filter. Gradually crack open the vacuum so that it turns on slowly and pulls the solution through without causing excessive foaming or getting material sucked back up into the tube. Add an additional 15 mL of dichloromethane to the separatory funnel, shake, allow it to settle, and again drain the dichloromethane layer through the filtration pack. Repeat this one final time, with an additional 15 mL of dichloromethane.

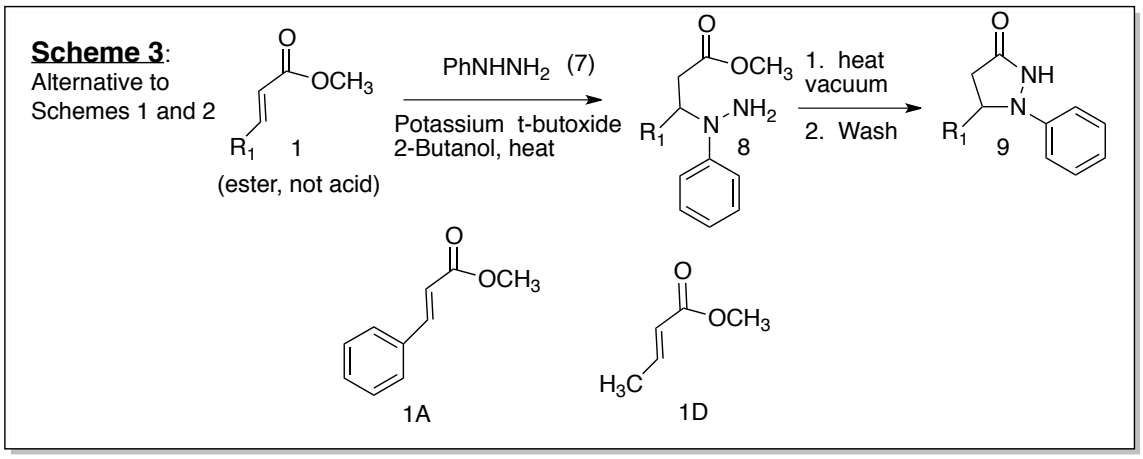
Concentrate this solution. Rotary evaporation can be used, or if the line is too long you can try to just heat-distill off the dichloromethane in your hood. Once concentrated, add another 10 mL of dichloromethane and reconcentrate it again. (It's hard to get rid of all the ethanol, and much residual ethanol will interfere with the final reaction.) Finally, connect the vacuum adaptor directly to your roundbottom, and heat the residue under vacuum on your hot plate, maybe with a setting of 5 or 6, for 10 minutes.

Try to use a long-stem pipet to draw out about one centimeter worth of warm material for an NMR sample. Fill to half-tube depth with CDCl3, then take some out and transfer into a GC vial. Submit for both GC-MS and for NMR.

Analysis: Submit the two NMR's for proton analysis. The one was for the sample taken prior to sodium borohydride, the other following the workup. [Perhaps we'll run 2-dimensional H-H correlation spectroscopy (COSY) and or 2-dimensional H-C correlation spectroscopy as well to help evaluate the samples.] If everything is good, the second sample could be simpler and cleaner to look at. But if that doesn't turn out so clean, it could be helpful to look at the first sample. Key questions will be whether the intermediate formed well; that would involve a signal that shows up somewhere in the 8-9.5 area of the spectrum. For the final product, a question will be how much ethanol and/or residual aldehyde and/or aldehyde-that-got-reduced-to-alcohol are present.

Weigh your flask and subtract the mass of the flask and stir bar to evaluate your mass of product. Figure out your % yield based on mass. (The actual yield may be less, since it's not certain that all of the mass is desired product.)

Critical: Start the next reaction as described in Scheme 4. Before week two is completed, it is essential that you get the final reaction started, see Scheme 4. This reaction takes a couple of hours plus requires workup. So it is essential that it gets set up before the third lab period. It should get set up during the second lab period, or else sometime during the week in between, at least a day before the final lab period.



Scheme 3 Procedure: Formation of Pyrazolidinone Ring Using Phenylhydrazine (7)

Turn your hot-plate heater to setting of 7. Use either a 125-mL ground-glass jointed Erlenmeyer or a 100-mL round-bottomed flask. Add a medium stir-bar. Weigh the combination and record the mass. Clamp the array securely at the bottom neck.

Add 20 mmol of methyl cinnamate **1A** (162.2 g/mol). Add 24 mmol of potassium t-butoxide (112.2 g/mol). This material is water sensitive, so close the container really fast after weighing it out, so that it's still healthy for later users. Add 30 mL of anhydrous 2-butanol. (This should be of high quality, preferably HPLC-grade.) Prepare a reflux condenser with tubing for gentle water flow, with a septum on top, and with a syringe needle poking through the septum to enable pressure equalization. (You normally don't want to heat up a closed container, or vapor pressure will want to make it perhaps explode!) Add 20 mmol of phenylhydrazine **7** (0.0954 mL/mol). Attach the reflux condenser, make sure a gentle water flow is running, and heat the solution on the hot plate. Reflux the mixture for two hours. If time is problematic, a reduction in time could be possible, although it may compromise the product purity or yield to some extent. Plan ahead so that you are ready for the wrap-up steps that follow.

After the time is done, you want to remove a small sample for NMR and GC-MS analysis. Remove the reflux condenser. (If it sticks, you may need to get a glove so that you can grip the hot flask while you twist the condenser off.) Use a long-stemmed pipet and draw up about one inch of material, and transfer the little sample into an NMR tube. Leave the pipet in the barrel of the NMR tube for now, and worry about it in a few minutes.

If you're in a time situation, you could actually leave things as they are, labeled and with the clamp such that it's not in the way, and continue next week. If so, you should at least submit your NMR sample. Dilute your NMR tube to half full, then remove some of the NMR solution and transfer it into a GC Vial, still leaving the NMR tube 1/3 full. Submit the NMR sample for proton (and perhaps some 2-D experiments). Dilute the GC vial to at least 0.5 mL with either dichloromethane or more CDCl3, and submit that for GC-MS analysis. If you do continue with the following paragraphs, though, you could save this NMR/GC preparation for a few minutes and do it during a pause time.

If you do have time, it would be helpful to spend another ten-fifteen minutes to concentrate your reaction mix to dryness using vacuum and heat. Add a dry reflux condenser (no hoses or water line) from the partner's drawer. Connect the vacuum hose to the top of reflux condenser using a vacuum adaptor. Gently turn the vacuum open (so that it doesn't erupt and go crazy). Once the bubbling isn't excessive, swing the array back over the hot plate, and try to boil the mixture dry over the heat. Once most of the 2-butanol is gone (hopefully within five minutes or less?), pull out the reflux condenser and have the vacuum adapter directly connected to the

flask for an additional five minutes. Swing everything off the balance, detach the vacuum hose, turn off the vacuum, remove the adapter, and put a rubber stopper or septum into your flask. Now you will be good till next week to work up your product.

During this time you could be preparing the NMR and GC-MS sample, as described above.

Note: The reflux time obviously creates a time issue. If you notify the instructor, you could perhaps leave for a while during the reflux period. Another option would be to have it all set up during lab, then come in for a time period outside of your normal lab period to turn it on and turn it off.

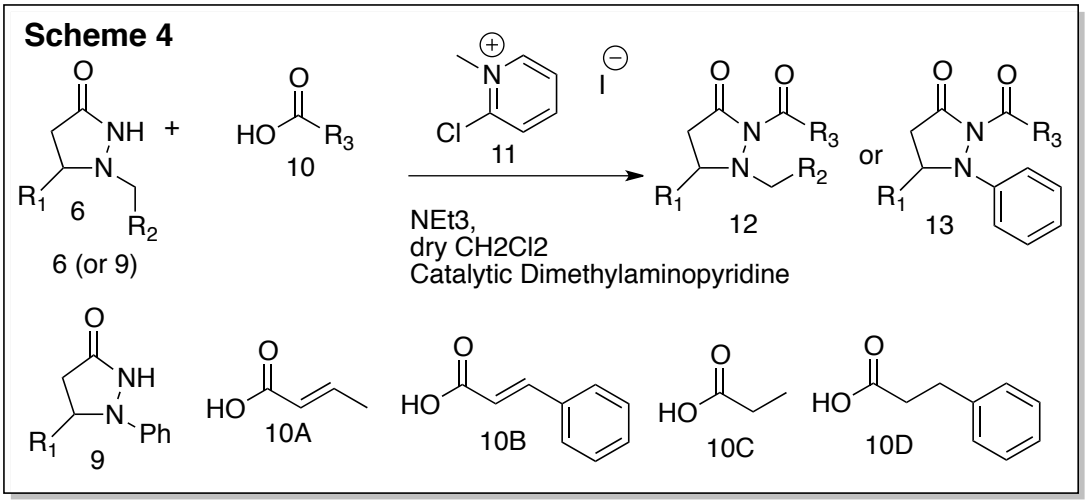
For students using Scheme 3 with N-phenyl pyrazolidinones: Week two workup.

Add 40 mL of dichloromethane solvent, then add 24 mmol of solid ammonium chloride (53.5 g/mol), and 40 mL of water. Try to break things up and stir for either 15 minutes or until the solids pretty much dissolve. Pour the solution into a separatory funnel. Use an additional 10 mL of dichloromethane and 10 mL of water to rins the flask.

Prepare a pre-weighed 100-mL roundbottomed flask, and add a fritted filter funnel. Connect a vacuum tube, but don't turn it on yet. Add a 1.5-inch pile of sodium sulfate. Drain the lower dichloromethane layer directly onto the sodium sulfate filter. Gradually crack open the vacuum gradually so that it turns on slowly and pulls the solution through. Add an additional 15 mL of dichloromethane to the separatory funnel, shake, allow it to settle, and again drain the dichloromethane layer through the filtration pack. Repeat this one final time, with an additional 15 mL of dichloromethane.

Concentrate this solution. Rotary evaporation can be used. Once concentrated, add another 10 mL of dichloromethane and reconcentrate it again. (It's hard to get rid of all the 2-butanol, and much residual 2-butranol will interfere with the final reaction.) Finally, connect the vacuum adaptor directly to your roundbottom, and heat the residue under vacuum on your hot plate, maybe a setting of 5 or 6, for 10 minutes.

Make up an NMR sample in CDCl3 and a GC sample and submit. A key question will be how pure this is going into your final reaction (Scheme 4). However, it will be essential to begin the Scheme 4 reaction and have the actual reaction completed before the final week. So during week two, once you've worked up and concentrated your N-phenyl pyrazolidinone product, you should get the final reaction started so that the final week will be free for workup and possible purification.



Assignments:
Hoods 1, 3, 4, 5: Butenoic (crotonic) acid **10A**. (Scheme 4)
Hoods 2, 13, 14: Use propanoic (propionic) acid **10C**. (Scheme 4)
Hoods 7, 8: Use 3-phenylpropanoic (hydrocinnamic) acid **10D**. (Scheme 4)
Hoods 9, 12: Use 3-phenylpropenoic (cinnamic) acid **10B**. (Scheme 4)

Scheme 4 Procedure: N-Acylation of Pyrazolidinone Ring Using Carboxylic Acids and Mukayama's Reagent (11)

Determine the maximum number of mmol of pyrazolidione **6** or **9** that you might have. Since you know what your R_1 and R_2 groups are, you can figure out the molar mass for your pyrazolidinones. Since you pre-weighed your flask and stir bar, you should know what mass is from the actual chemicals. Thus you ought to be able to calculate your maximum number of mmol of pyrazolidione. Note: This may not actually be correct, since some of the mass may be contaminants or solvents that were not completely removed by the separatory funnel operations and vacuum concentrations that you have used. You originally started with 20 mmol, so if everything was perfect thus far, the most you should possibly be able to have is 20 mmol-worth of material. If you have more than that, you probably need to do more vacuum heating, or else check with instructor. Hopefully you'll have less than 20 mmol-worth of mass, but hopefully more than 10-12 mmol worth of mass.

Add 25 mL of dry anhydrous CH_2Cl_2, and add a septum to exclude air. Based on how many mmol of pyrazolidinone you might have, add 1.2 equivalents of your carboxylic acid **10**. Then add 1 mmol of dimethylaminopyridine (122.2 g/mol). Weigh out and add 1.4 equivalents of 2-chloro-1-methylpyridinium iodide (Mukayama's Reagent, **11**). Immediately reclose the bottle from which you took reagent **11**, since it is moisture sensitive. If you leave it uncovered, it will go bad and everybody who uses it after you will have problems. Record all of your observations. (Is the mixture homogeneous or heterogeneous, etc.) Last, syringe in 2.8 equivalents of triethylamine (139 mL/mol). This will get the reaction started.

Time: As of this writing, I think the reaction takes several hours. The most convenient way is to set it up at the end of your second week, and let it run until the third week. Alternatively if you were able to come in at least a day before your third lab period, and have it run overnight, that should be OK. (Another possibility might be if somebody tries it with reflux; maybe it will be done within an hour or so under reflux?)

Workup: Pour the contents of the flask into a separatory funnel. To the original flask add 20 mL or ether, 10 mL of dichloromethane, and 60 mL of 1-M HCl/water. Shake and swirl,

and add this rinse solution into the separatory funnel. Shake cautiously, with venting, then allow the mixture to settle. The bottom layer will probably be the organic layer, but if you aren't sure, add some extra water to see if the top layer gets bigger.

To a clean, pre-weighted 250-mL ground-glass-jointed Erlenmeyer (or round-bottomed flask), add a pre-weighed flea stir bar, and attach the filter flask (the one with the 6-inch column above the white fritted filter, with a ground-glass joint on the bottom, and with a vacuum connector on the side.) Pour in a 1-inch pile of silica gel; this will function to try to absorb contaminants. Then add a 1-inch pile of sodium sulfate on that; this will function to try to absorb water.

Carefully drain some of the organic layer onto the filter column. Carefully/gradually open up the vacuum so that liquid gets pulled through without boiling all out and getting sucked into the vacuum tube. Pour much of the organic layer from the separatory funnel, but if you're getting close the edge or if there is solid material or emulsion at the interface between the organic and water layers, leave that behind.

Do two additional dichloromethane extractions from your separatory funnel, each time with about 15 mL of dichloromethane. Finally pour 20 mL of ether through the filter column to try to make sure no desired product is left stuck on the silica.

Concentrate the solution, either on the rotary evaporator or else in your hood on a hot plate. (If you do that, use a boiling stick). Once most of the solution is gone, apply vacuum while stirring and heat on your hot plate at a setting of five or six for ten minutes or so to remove the last residual solvents.

Analysis: Weigh the material, so that you can calculate your final mass and yield. Calculate the % yield for both the final step, but also for the overall process. (If every step of the synthesis had worked perfectly, you'd end up with 20 mmol of product. So 20 mmol is your theoretical number of moles.)

Take a portion out, prepare an NMR sample and a GC-MS sample, and submit. You may want to take the sample out right away while the mixture is still hot; in case your product is a thick oil, the heat might make it easier to transfer to an NMR tube.

There is a fair chance that products **12** and **13** will be contaminated by a structural isomers in which the acyl group is actually connected to the amide carbonyl oxygen rather than to the amide nitrogen. As of this writing, an easy solution to this has not been identified, nor has a way to get the wrong isomer to convert over to the desired isomer been found. If time permits, some experiments to explore possible solutions may be appropriate.

It's also possible that there will simply be an accumulation of side products by this point in the sequence. There may be residual materials from the Mukayama Reagent **11**, or there may have been high-molecular-weight alcohol that was formed by sodium borohydride reduction of aldehydes **4** during Scheme 2. These alcohols may still be present in their original form, or may have been converted to ester during the Scheme 4 reaction. For these reasons, it may be appropriate if time permits to purify the final pyrazolidinones **12** and **13** via automated chromatography (combiflash).