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I. Introduction

In the $^{14}$C AMS technique, the element of interest (carbon) is chemically separated from the original sample and loaded as a solid target (graphite) in the sputter ion source of the tandem accelerator. The procedure to convert the raw sample material into a graphite target suitable for the ion source includes a series of necessary steps (removal of macroscopic contaminants, chemical cleaning procedures, combustion and graphitization). These steps are essential for a reliable AMS measurement.

This protocol outlines the graphite sample preparation procedure at UCI KCCAMS prep-laboratory for organic and carbonate samples. Through this process, the CO$_2$ produced from carbonaceous raw materials is cryogenically purified (separated from non-condensable gases) and reduced to solid graphite.

II. Graphitization reaction

To catalyze the production of small amounts of elemental carbon from CO$_2$, we use the Bosch reaction (Manning and Reid, 1977) that can be summarized as:

$$\text{CO}_2 + 2\text{H}_2 \xrightarrow{550 \text{ - } 650^\circ C} \text{C} + 2\text{H}_2\text{O}$$

Catalyst (Fe)

The reaction takes place as two successive reductions: first to carbon monoxide and then to carbon, which permeates and adheres to the surface of the iron powder (catalyst). More details on graphitization can be found in Vogel et al, 1984.

During the graphitization procedure at the KCCAMS prep-lab, cryogenically purified CO$_2$ is transferred into Pyrex culture tubes (Lloyd et al, 1991) and reduced to graphite, using hydrogen over pre-cleaned (preconditioned or reduced) iron powder (Alfa Aesar iron powder, -325 mesh, reduced, 98%) at 400$^\circ$C for 45 minutes.

III. Graphitization Procedure

The graphitization line (Figure 1) at UCI KCCAMS prep-lab has 12 H$_2$/Fe reactors, allowing us to graphitize 72 organic and/or carbonate samples per day. The vacuum lines are made of glass and stainless steel and are pumped by turbomolecular pumps backed by oil free diaphragm pumps (Santos et al, 2004).
The design was based on sample graphitization lines from CAMS/Lawrence Livermore National Laboratory (LLNL).

In order to begin the graphitization procedure, make sure that you are signed up for the correct day and time on the sign-up sheet located on the upper left of each line. Before beginning, check and sign the “magnesium perchlorate use” card above the line. If the three spaces allotted for signatures are filled then you must change the magnesium perchlorate (section X). However, if there are more spaces available, then sign in your name and proceed to section IV (Preconditioning the Fe catalyst).

![Fig.1: Picture showing the main section of the graphitization line with labeled valves](image)

**Note:** When you use Fe as a catalyst you MUST FIRST PRE-CONDITION the Fe. This procedure takes 1 hour to complete.

1. If there is a card that states “Preconditioned” on the graphitization line then you can skip this step and proceed to Section V.
2. If the graphite-reactor tubes have something in them but there is no sign to clarify what is in the tubes, talk to an AMS assistant.
3. If the graphite-reactor tubes are empty or have samples in them, follow the procedure below.

**IV. Preconditioning the Fe catalyst:**

1. Take 12 baked Pyrex graphite-reactor tubes from the wet cabinet on the island tabletop on line A.
2. In a drawer labeled “graphitization tools” there is a box (each graphite line has its own tools in its corresponding island). From inside the box take out the following (Figure 2): a) vial of Fe powder (catalyst) and b) Fe scoop.

3. WEARING GLOVES, scoop 2 packed spoonfuls of Fe into each tube. Each tube should receive approximately 5 milligrams of iron. Weigh out each scoop and record the weight on the white cards found in the back of the graphitization logbook found in the drawer labeled with the same name on line A. Evenly distribute iron inside the Pyrex tube walls by rolling the tube on a sterilized surface (clean foil) before placing into reactors.

4. Once the tubes have been used for graphitization and the reaction is complete, flush the reactors on the line 3 times with 450-600 torr H\(_2\). First close the system to the pump using valve #1 (Figure 1). Next open the H\(_2\) valves #10 and 11 (Figure 4) to allow H\(_2\) to flow into the reaction line. Then flush with H\(_2\) by carefully opening valve #9. Use valve #11 to adjust the speed at which H\(_2\) enters the line and close valve #9 when the desired pressure has been reached. Evacuate the entire line and wait until the gauge reaches its baseline again (x10\(^{-4}\) torr).

5. Fill the line with 1 atm of H\(_2\) before removing the reactor tubes containing the graphite. Close the reactor valves, and then remove the reactor tubes already on the reactor and replace with the tubes containing the newly weighed Fe. This should be done one tube at a time to minimize the exposure of the MgClO\(_4\) to air. If the MgClO\(_4\) card is full (used 3x), then it must be replaced as well. The perchlorate can be found in the labeled drawer on line B. You will use the metal strainer to sieve out any excess and unwanted fine powder. You want to use only the larger chunks of material for the water traps. Use the tubes presently on the line as a guide for replacing with new tubes (level in tube, size of chunks used). Again, these should be done one tube at a time. MAKE SURE THE TUBES ARE SECURELY FASTENED INTO THE REACTORS!
6. Evacuate each reactor slowly and individually. Then open all valves # 1-7 (Figure 1) completely and wait until the vacuum reaches baseline. Evacuate out all reactors by opening the valves slowly (Fig.3a and 3b). Pause when you see the pressure gauge reading (left-hand end of the line - above valve 1) start to rise, open the valves fully when the pressure reading starts to fall.

![Fig. 3a Valve in closed position](image1)
![Fig. 3b Valve in open position](image2)

7. Monitor the vacuum by looking at the pressure gauge.

8. Flush entire line (including bypass line) with H₂ three times. First close the system to the pump using valve #1 (Figure 1). Next open the H₂ valves # 10 and 11 (Figure 4) to allow H₂ to flow into the reaction line. Then flush with H₂ by carefully opening valve #9. Use valve #11 to adjust the speed at which H₂ enters the line and close valve #9 when the desired pressure has been reached. Evacuate the entire line and wait until the gauge reaches its baseline again.

9. Once the gauge reaches baseline, close valve # 1 (Fig. 3a)

10. Now you will add between 700-800 torr of H₂ to each reactor in order to clean the Fe (preconditioning of the Fe). Carefully open valve # 9 again. Close it when the desired pressure is achieved. You can monitor the amount of H₂ added by looking at the pressure readings above the line.

![Fig. 4 Hydrogen cylinder with labeled valves](image3)
11. If you have added between 700-800 torr proceed to step 13. Avoid exceeding these values so that 
  \( \text{H}_2 \) is not wasted.

12. If you have exceeded 800 torr then you must vent some \( \text{H}_2 \). Close valves # 2 and the left bypass 
  valve then open valve #1 to evacuate the isolated section (Figure 1). Open valve #2 and the 
  bypass valve. You will notice that the pressure readings have lowered. If your new reading is 
  700-800 torr, then proceed to step 13. If not, repeat until the proper value is obtained.

13. If you have not exceeded 800 torr, then close all the graphite-reactor valves and the \( \text{H}_2 \) valves 10 
  and 11.

14. Pump the excess \( \text{H}_2 \) that is trapped in the line. DO NOT BE ALARMED IF THERE IS A LOUD 
  SOUND, THIS IS THE EXCESS \( \text{H}_2 \) BEING PUMPED AWAY.

Be careful not to flood the pump with more than 1 atm (760 torr) of \( \text{H}_2 \). Excessive amounts of gas 
may cause the pump to auto shut off or even damage the pump. To check that the pump is still 
operational look for the little green light on the pump controller and check that the pump cooling fan 
is still working. If the light is red, seek assistance.

15. Slip the corresponding heaters onto the reaction tubes and set the timer and temperature controls 
(Figure 5). To set the temperature make sure all the controllers are on (the current temperature 
should be indicated for each reactor) \textbf{FOR PRE-CONDITIONING THE Fe THE TEMP. MUST BE 400°C.} Check the set temperature by pressing the "sel" button next to each pressure 
monitor. This will display the set temperature. If the set temp is not 400°C then use the up and 
down buttons to raise or lower the set temp and then reselect by pressing "sel". Double check 
that the temp set is at 400°C by pressing the "sel" twice.

\begin{center}
\includegraphics[width=0.5\textwidth]{Fig_5.png}
\end{center}

\textit{Fig. 5 Control locations to set temperature and timer}

16. To set the timer, turn the dial to 45 minutes. Go to the computer that controls the monitoring of 
the reactions with graphs (Time vs. Torr). Select the appropriate line/bench. Click “start” and
wait for it to turn green. Then start the timer by slowly flipping the power switch off and then on again.

V. Preparation for graphitization:

Before you begin graphitization you must:

1. Prepare the slush (dry ice + methanol) for the water trap (Figure 1). If you do not know how to prepare the slush go to Section XI.

2. Get 2 dewars of liquid nitrogen (one tall dewar and one short dewar). Liquid nitrogen is provided in the SW corner of the prep lab next to the pressing station. IF YOU HAVE NEVER GOTTEN LIQUID NITROGEN BEFORE, PLEASE ASK FOR ASSISTANCE.

3. Check the pressure of each reactor by slowly rotating the knob on the pressure control box. All reactor pressures displayed on the control box should be within a few torr of each other. If not ask for assistance. Evacuate out H₂ from preconditioning. Evacuate out all reactors to baseline by opening valves # 1-7 and then open all the reactor valves (slowly). Monitor the vacuum by looking at the pressure gauge. Flush the reactors three times with H₂ using the method described in section IV. Then close reactor valves, valves # 2-7, and both bypass valves.

4. For each sample YOU MUST HAVE AN INDEX CARD WITH YOUR ASSIGNED UCIG NUMBER (for information on how to print cards refer to the combustion protocol). Every sample must be entered into the computer and have a corresponding index card. At this point you should write down the date of graphitization, your name or initials and the number of the reactor(s) that each sample will be in. Below is an example of a UCIG card (new info is in blue. The info in green will be added later):

<table>
<thead>
<tr>
<th>22000</th>
<th>Rockwell</th>
<th>Yield:</th>
</tr>
</thead>
<tbody>
<tr>
<td>4/10/2008</td>
<td>IFD-T2-C47</td>
<td>φC GB:</td>
</tr>
<tr>
<td></td>
<td>charcoal</td>
<td>φC EA:</td>
</tr>
<tr>
<td>Pretreatment:</td>
<td>ABA (4/11/08)</td>
<td>NONE:</td>
</tr>
<tr>
<td>Combustion date:</td>
<td></td>
<td>SW: (mg), CuO W: (mg)</td>
</tr>
<tr>
<td>Graphitization date:</td>
<td></td>
<td>Operator:</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Non-Cond.:</td>
</tr>
<tr>
<td>Reactor #:</td>
<td>CO2</td>
<td>H2</td>
</tr>
</tbody>
</table>

Fig. 6 An example of an index card
VI. Graphitization:

VI.1. Using combusted organic samples

1. Wearing safety glasses, score the tube using the glass cutter found in the "graphitization tools" drawer in the plastic box. This prevents unnecessary wear and tear on the bellows.

2. Carefully clean the combustion tube using methanol and a Kimwipe. Be aware that this will erase any label you may have written on the tube. Then use the compressed air can to remove any Kimwipe fibers from the tube.

3. Once the line is pumped down to baseline ($10^{-4}$), and the valves have been toggled to release any trapped gas, toggle close valves # 2, 3, 4, 5, 7, and the bypass valves, then unscrew the bellows. If there is glass in the bellows discard into the glass waste. Do not accidentally discard the end support for the bellows (Figure 7) – either take it out or hold it so it does not go into the glass waste container.

4. Holding the bellows at an angle, place the sample tube into the bellows. DO NOT drop the tube vertically into the bellows as it may cause the scored tube to crack prematurely.

5. Securely fasten the bellows back into the line.

6. Open valve # 2 to evacuate the air in the bellows.

7. Monitor the vacuum by looking at the pressure gauge. If the vacuum does not reach baseline in one minute or so then open valve # 3 to trap excess water that may be in the bellows. Once the gauge reaches baseline close valve # 2.

8. Crack the combustion tube that is in the bellows.

9. If valve # 3 is not open yet, open it to allow the gases to go through the water trap for about 1-3 minutes depending on your sample. During this step, water that is in your sample will be "trapped". So if your sample contains a lot of water you should wait for at least 2-3 minutes.

10. While you are waiting submerge the larger culture tube between valve # 4 and 5 (figure 8) in liquid nitrogen. Make sure the dewar is completely filled to the top and adjust the height of the dewar with the retractable stand (lab jack). The liquid should be covering the bottom inch of the tube.
11. After the water has been trapped, open valve #4 to allow the CO\(_2\) to freeze in the presence of the liquid nitrogen for 2 minutes. During this time, raise the liquid nitrogen level on the culture tube by raising the jack, giving the CO\(_2\) more surface area to freeze on. You should see a white cloudy ring of frozen CO\(_2\) in the tube.

12. Non-condensable gases will not freeze. Open valves #5 and 6. Watch the digital pressure monitor labeled "meas. vol." (MV) and record any non-zero readings on the card and in the computer (i.e. Non-C: 8T). If the MV transducer does not read the non-condensable gas, you must then read the value at the pump pressure gauge. Close valve #1, then open valve #2. This is the value that you will record.

13. Evacuate the non-condensable gases by opening valve #2. Wait until the vacuum reaches baseline. IT MAY TAKE WHILE.

14. Close valves #2, 3, and 4 and then remove the dewar to allow the frozen CO\(_2\) to thaw. You can use the heat gun (adjust the heat gun to a low setting of 6 or 7) or your hand (gently) to accelerate the process (but DO NOT GET THE TUBE TOO HOT or PULL DOWN ON TUBE).

15. Move the dewar to the tube on the measure volume between valve #6 and 7. The CO\(_2\) will be transferred to this tube to quantify the CO\(_2\) amount. Freeze the gas for 10 seconds.

16. After the CO\(_2\) is once again completely frozen close valve #6, isolating the section between valves #6 and 7.

17. Remove the dewar. Allow the CO\(_2\) to thaw. You can use your hand to speed things up. Watch the digital pressure monitor labeled "meas. vol." and when the numbers stop going up you know your CO\(_2\) is completely thawed. Write the amount of CO\(_2\) (the digital reading from "meas. vol.") on the index-card (Figure 6 - emphasized in green). Also write the multiplier value: (1) for 1 aliquot sample.
If you are preparing 4 aliquot samples see section VII of this protocol (Graphitizing a standard - 4x standard) on how to transfer this amount of gas to the next 4 reactors.

18. If there is more than 580 torr of CO₂ (for 1 aliquot sample) **you must evacuate** some of the CO₂ by following the procedure below.
   a. To evacuate 1/10 of the total CO₂ open valves # 5 and 6. To evacuate 1/4 of the total CO₂ just open valve # 6. Before vacuuming out any CO₂ calculate how much CO₂ will be lost to ensure you will be left with enough CO₂ (540T gives 1mg carbon). If you are unsure ask for assistance.
   b. Close valve # 6 and use the bypass to vacuum out the excess CO₂ from the measure volume.
   c. Collect the CO₂ to the measure volume using the liquid nitrogen. Recheck amount of remaining CO₂ and repeat if the CO₂ level is still too high. After the correct amount of CO₂ is achieved close the valves to the bypass to prevent pumping samples away.

19. Next the CO₂ must be transferred to the reactor. Move the reactor dial to the correct number and ensure that the reactor is still under vacuum.

20. Place the liquid nitrogen under the reactor that you are going to transfer the CO₂ into. Submerge the reactor tube in liquid nitrogen to the level of the magnesium perchlorate. Do not allow liquid nitrogen to touch the Ultratorr fittings. Be careful not to freeze the o-ring!

21. Check that both bypass valves are closed then open the valve to your reactor, then open valve # 7.

22. Monitor the pressures of the measure volume and reactor. Wait at least 5 seconds after both numbers are 0 to ensure that complete CO₂ transfer has occurred. Close the reactor valve.

23. Vacuum out the main line by opening valves # 2-7 (some may already be open) and both bypass valves, monitoring the millitorr gauge.

24. Allow CO₂ to thaw into the reactor volume. DO NOT USE HEAT GUN. Write the amount of CO₂ displayed on the digital screen labeled "reactor" on the index-card (figure 6). Make sure you are reading the correct reactor each time by making sure the reactor dial is on the correct reactor number.

25. Repeat steps 1-24 for each sample.

**VI.2. Leaching carbonate samples.**

Prior to acidifying a carbonate sample to generate CO₂ for graphitization, the user should leach the sample to get rid of secondary carbonates. This procedure is required for anything that has been in the ground, fresh water, etc. In general it is best to maximize the percent leach preformed while retaining
a sample size as close to 1 mg carbon as possible. There are exceptions to this rule and if you are unsure as to the best course of action, ask for assistance. Leaching is conducted using HCl as shown in the equation below:

$$\text{CaCO}_3 + 2\text{HCl} \rightarrow \text{Ca}^+ + 2\text{Cl}^- + \text{H}_2\text{O} + \text{CO}_2$$

1. Weighing the sample into the Vacutainer:
   a. Place a weigh paper or an aluminum foil wrapped Vacutainer (a vial used for the leaching and subsequent acidification of a carbonate sample) with the septum removed (Santos et al, 2004) onto the scale. The Vacutainer should be labeled with the UCIG number of the sample.
   b. Tare the scale and then weigh out 10mg of a carbonate sample (for 1mg C graphite). When conducting the cleaning procedure (leaching), you have to calculate an extra amount of material to compensate for material that will be consumed by the HCl. If you are leaching using 2ml of 0.01N HCl, you have to add at least more 10% of total amount.

<table>
<thead>
<tr>
<th>Amount of sample (mg)</th>
<th>Strength and volume (mL) of Acid used for:</th>
<th>Amount carbonate removed (mg)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>50% leach 0.1 N HCl</td>
<td>30% leach 0.1 N HCl</td>
</tr>
<tr>
<td>22</td>
<td>2.2</td>
<td>11.0</td>
</tr>
<tr>
<td>21</td>
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<tr>
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</tr>
<tr>
<td>1</td>
<td>0.60</td>
<td>0.2</td>
</tr>
<tr>
<td>&lt;1</td>
<td>Ask</td>
<td>Na</td>
</tr>
</tbody>
</table>

c. Replace the septum after weighing.

2. Turn on the heating block to low on setting 7-8, monitor until the thermometer is at 70-80°.
3. Add the appropriate amount of HCl with a pipette to the Vacutainer and sample. Be sure to use the correct concentration as well.

4. Place sample onto heat block, with the septum removed. Place the Al foil tent over the vials and wait at least 25 minutes.

5. Upon completion rinse twice with MilliQ water, pipette excess water with an ultra-fine disposable pipette and dry on the heating block. DRY UNDER AN ALUMINIUN FOIL "TENT" TO SPEED UP THE PROCESS.

VI.3. Acidifying Carbonate Samples:

1. Go to the four port carbonate line (on the other side of line A) and close the valve to be used.
2. Remove the tube fitting and replace it with a needle fitting (Fittings, needles, and the injection syringe are found in the “Carbonate sample prep” drawer to the left). Lightly grease the needle with the high vacuum grease found in the drawer labeled “Carbonate line tools.”
3. To evacuate each Vacutainer/sample, place one drop of MQ water on the septum and slowly press the needle through the water and septum (Figure 9). Open the valve to the rest of the line and evacuate the container.

![Fig. 9 Vacutainer evacuating on the carbonate line](image)

4. Monitor the vacuum by looking at the pressure gauge. Once the gauge reaches baseline close the valve above the tube.

5. To avoid sucking the water into the vacuum pump while removing the Vacutainer tube, carefully pull the Vacutainer half way down the needle, then blow off the water with the air duster and completely remove the tube. Repeat for all tubes.
Be aware that the needles rapidly become blunt and it will get increasingly harder to push the needle through the Vacutainer septa. After you have used a needle 5 times, replace it. Discard the old needle into the used needle container (on top of the bench island).

6. Bring evacuated tubes to the hood and gather the syringe, needles, and a beaker (in the drawer labeled “Carbonate Sample Prep”) and phosphoric acid (underneath fume hood in the cabinet to the far right) on a tray. Line the tray with a large Kimwipe.

7. Attach the needle to the syringe.

8. Wearing gloves and safety goggles fill beaker with about 0.8mL of phosphoric acid per sample that needs to be acidified.

9. Remove the plunger from the syringe and fill the syringe to 5mL with phosphoric acid.

10. Replace the plunger and expel any air bubbles (Figure 10). Turn the syringe completely upside down and expel bubble.

11. Take your sample tube and puncture the GREASED needle through the septum (Figure 11). To help prevent leaks, put a little drop of acid onto the septum and push the needle in through it.

12. Expel 0.8mL of acid into the tube by counting down the hash marks on the syringe. Leave the drop of acid on the top of the septum to help prevent leaks. Place the tube on the heating block (Figure 12) for 20-40 min. or until the liquid is clear and not bubbling.

Note: If using calcite as a blank, you will want to do this sample first, as it takes more time to acidify than regular carbonate samples.
13. Repeat until all samples are done. Replace the needle every 5 samples.
14. Discard needle in the used needle container and discard excess acid in acid/base waste. Wash beaker and syringe with ample amounts of hot water and then rinse with MQ water. Dry with the hot air gun and put back in the drawer.
15. Make sure all valves with needle fittings are closed. When acidification is complete remove the needle fittings and replace the original tube fittings. Open the valves to evacuate.
16. At the graphitization line, close valves #2-7. Replace the bellows with the needle adapter.
17. Using an empty Vacutainer, check the vacuum on the graphitization line with the needle fitting. If a good vacuum is not reached, ask for assistance.
18. Take a sample from the heat block. Using a kimwipe, carefully remove the acid drop from the top of the septum. Replace with a drop of MQ. Grease the needle on the line. Insert the sample septum halfway up the needle on the adapter (through the drop of water) being very careful not to fully penetrate the septum. If that were to happen, your sample would mix with the air in the needle headspace and would become unsuitable for $^{14}$C measurements.
19. Open valve # 2 and allow the atmospheric gases in the needle to evacuate.
20. When the gauge reactor reaches baseline close valve #2, then push the needle completely through the septum. Continue procedure by going to section VI.1 (Using combusted organic samples - steps 9 to 24).
21. To remove the vacutainer after the sample is in the reactor tube, close valves # 2 and 3. Then pull the vacutainer half-way down the needle. (YOU MAY WANT TO DO THIS STEP WHEN PUMPING AWAY THE NON-CONDENSABLE GAS TO REACH BASELINE MORE QUICKLY.) Blow off the water drop using the air duster. Completely remove the vacutainer and replace with a dummy vacutainer or the next sample.
VII. Graphitizing a standard (4x standard) sample.

This section illustrates a variation of the graphitization method (section VI.1. Using combusted organic samples). It allows 4 aliquots to be shared in one tube (e.g. 4 graphite samples). This is being applied at UCI KCCAMS prep lab to prepare standards (OX-I's, OX-II's, ANU's, etc).

Also see the UCI KCCAMS combustion protocol to learn how to accommodate 4 mgC samples in the combustion tube.

1. If you are preparing 4 aliquot samples, you will have to isolate CO$_2$ gas between valves #5 and 7, instead of valves #6 and 7, as is explained in step 15 (section VI.1. Using combusted organic samples). The combined volume between the valves #5 and 7 is enough to accommodate 4 times 1 mgC sample. Write the amount of CO$_2$ (the digital pressure reading from "meas. vol.") on the index-card (Figure 6 – emphasized in green). Also write the multiplier value: (4) for 4 aliquot sample.

2. Calculate and record the “yield” on the appropriate place on the index card. If this number is greatly below the percent carbon of the material, seek assistance. (The % yield for oxalic acid standards should be in the range of 18.5-20.5%)
   \[
   \text{% Yield} = \frac{(\text{Measure Volume Pressure})(\text{Multiplier})}{(540\text{Torr})(\text{mg of sample combusted})} \times 100
   \]

3. Standards must be split into 4 different reactors. This step requires sharing the amount of CO$_2$ gas that is trapped between valves #5 and 7.

To ensure almost equal amounts of CO$_2$ in each reactor, Santos systematically evaluated the splitting processes and recommends this splitting order:

1 2 3 3 2 4

a. The gas is expanded between valve #5 and 7. Now close valve #6. Transfer the amount of gas trapped between valves #6 and 7 to the first reactor available.
b. Again expand the gas between valves #5 and 7. By opening valve #6 (wait 30 seconds).
c. Close valve #6 and transfer the CO$_2$ trapped between valves #6 and 7 to the second reactor available.
d. Repeat (b) and transfer the CO$_2$ trapped between valves #6 and 7 to the third reactor available.
e. Repeat (b) and transfer the CO$_2$ trapped between valves #6 and 7 to the third reactor available.

f. Close valve #6 and transfer the CO$_2$ trapped between valves #6 and 7 to the second reactor available.

g. The final amount of gas (when you are on the last number in the splitting order) trapped between valves #5 and 7 can be completely transferred to the fourth reactor.

By using this transferring method none of the CO$_2$ gas will be wasted. Waiting the allotted time allows the CO$_2$ gas to expand evenly throughout the line volume thereby reducing fractionation.

4. During CO$_2$ transfer to the reactor, move the liquid nitrogen to the reactor that you are going to transfer the CO$_2$ into. Place the dewar under the magnesium perchlorate tube and raise the liquid nitrogen just enough to cover the magnesium perchlorate.

5. Check that both bypass valves are closed. Open the individual valve of the particular reactor, then valve #7 and transfer the gas.

6. Monitor the pressures of the measure volume and reactor. Wait at least 5 seconds after both numbers are 0 to ensure that complete CO$_2$ transfer has occurred. Close the reactor valve.

7. After completing the splitting order, vacuum out the line by opening valves #2-7 (some may already be open), monitoring the millitorr gauge.

8. Allow CO$_2$ to thaw on each individual reactor. **DO NOT USE HEAT GUN ON THE MAGNESIUM PERCHLORATE.** Write the amount of CO$_2$ (displayed on the digital screen labeled "reactor") on the index-card (Figure 6 - CO2 torr- emphasized in green). Make sure you are reading the correct reactor.

9. Repeat the entire section VII for each standard (4X standards).

**VIII. Hydrogen addition:**

Twice the amount of hydrogen (compared to CO$_2$) must be added to each reactor (Vogel et al, 1984, Santos et al, 2004). Since each reactor has a different amount of CO$_2$ each reactor must be dealt with separately. At this point the valves to the reactor tubes should remain closed until the H$_2$ is ready to be added.

1. Before adding H$_2$ close valves #3 and 4. Open all other valves (excluding the reactor valves).

2. Flush the entire line three times with H$_2$ (Figure 4) and as described in IV. Step #4.
3. Calculate the amount of H₂ needed for each reactor. Write this value on the index card. Now you will add the appropriate amount of H₂ to each reactor. Begin with the reactors that require the most or the least amount of H₂ addition. Working in order of increasing or decreasing amounts of H₂, saves time and H₂.

4. Regulate the amount of H₂ entering the line by using valve #9 and 11. Monitor the amount of H₂ added by looking at the pressure reading for the measure volume. Once the appropriate amount of hydrogen is reached, close vale #9.

5. Move the liquid nitrogen dewar to the appropriate reactor. Raise the lab jack so that just the tip of the magnesium perchlorate is in the liquid nitrogen. It is best to have the dewar completely topped off. BE CAREFUL! DO NOT SUBMERGE THE ENTIRE TUBE INTO THE LIQUID NITROGEN. Watch the reactor pressure (make sure the reactor dial is measuring the correct reactor). When it reaches 0, your CO₂ is now frozen.

6. VERY QUICKLY, open and close the reactor valve, allowing the H₂ to enter the reactor. Carefully watch the reactor gauge and note the highest value. This value is recorded on the index card as the “H₂ Real” value. Remove the liquid nitrogen dewar.

7. Move the reactor dial to the next reactor and repeat steps 4-6 until all reactors have H₂.

8. When all the reactors have H₂, allow the gases to thaw. Write the TOTAL amount of gas (displayed on the digital screen labeled "reactor") on the index-card (Figure 6 - CO₂+H₂ Total-emphasized in green). Make sure you are reading the correct reactor.

9. Put heaters onto each graphite-reactor tube. CHECK THAT THE HEATERS DO NOT COME TOO CLOSE TO THE ULTRA-TORR FITTING. THERE SHOULD BE 1/8 INCH – 1/4 INCH CLEARANCE (Figure 13). If the heater is too close, ask for assistance.

10. Set the timer and temperature controls (Figure 5). To set the temperature make sure the controls are on (the current temperature should light up). FOR GRAPHITIZATION THE
TEMP. MUST BE 550°C. Check the set temperature by pressing the "sel" button next to each temperature monitor. This will display the set temperature.

a. If the set temp is 550°C proceed to part c.
b. If the set temp is not 550°C then use the up and down buttons to raise or lower the set temp and then reselect by pressing "sel".
c. Double check that the temp set is at 550°C by pressing "sel" twice.

11. To set the timer, turn the dial to 3 hours.

12. At the computer next to line B, start monitoring the graphitization line by clicking the “Start” button on the correct line (Use the tabs to toggle between line A, B, and C). Let the computer monitor the reactor pressures for 30 seconds before starting the heaters on the line. If pressures remain steady go to step 13. If any reactors show a noticeable increase/decrease in pressure, the reactor may be leaking. Ask for assistance.

13. Start the timer by slowly flipping the “On/Off” switch off and then on again. (When using the Alfa Aesar -325 mesh iron this is plenty of time to complete graphitization, however, if a different iron (blend or mesh) is used then the timing may vary.)

14. After you have begun graphitization use the following procedure to clean the line:
   a. Empty the bellows of broken glass.
   b. Open valve #3 and take off the water trap to let the water vapor evacuate. It is best to have the water trap isolated to the pump at this point. Then you will not allow the water vapor into the entire line and should pump down quicker. You can use the heat gun or your hand to speed up the process.
   c. Pump all excess gas from the entire line (except reactors).
   d. Put away capped slush dewar and any excess liquid nitrogen into the big white cooler.

15. Take index-cards to the computer and record all numbers into the Sample Master list in the sample list folder of the Irvine 6. Place index-cards back on the line.

16. Use the computer by Line B to monitor the graphitization. Individual reactors may be turned off when the reaction is no longer proceeding and pressure becomes steady.

IX. Post-Graphitization Procedures (collect your graphite)

1. Take heaters off of the reactors and hang them on the bar. Ensure the leads all hang the same way to avoid tangles (Figure 14).
2. Obtain caps for your sample tubes from the drawer labeled "graphitization tools".
3. Record the residual pressure for each sample on the index cards (Figure 6).
4. Evacuate the residual H₂ from each reactor individually. Open the first valve partway to allow slow release of the H₂. When the gauge is at baseline you may completely open the valve. Close the valve and repeat this with all 12 reactors.
5. Once all 12 reactors have been individually evacuated, open all 12 valves.
6. Flush the entire line with fresh H₂ (3 times).
7. Close valve #1 and fill the entire line with H₂ until the pressure reaches approximately 1 atm (maximum)(~780 T). **DO NOT EXCEED 1 ATM OR THE PYREX TUBE MAY FLY OFF THE REACTORS, CAUSING INJURY OR SAMPLE LOSS.**
8. One reactor at a time, take off the graphite sample, then replace with a tube containing new Fe catalyst. Have baked Pyrex tubes with new catalyst ready to put into the reactors immediately after you take each sample off. The magnesium perchlorate is very hydroscopic and upon contact with the air will trap water (Santos et al, 2004).
9. Apply a cap to the graphite sample tube and label with a sharpie the UCIG # assigned to the sample, put on a new tube with Fe catalyst.
10. Repeat this for all samples and then pre-condition the line for the next user.
11. When the line is not in use leave it under vacuum (including the bypass line).
12. Take index -cards to the computer and record the residual values into the Sample Master list.
13. Place the graphite samples in a box with the client’s name on it and attach the index cards. Put the box in the wet cabinet by the pressing station for further AMS measurements.
X. Changing the magnesium perchlorate

1. Get 12 baked Pyrex tubes from the wet cabinet.
2. Obtain a piece of foil, the magnesium perchlorate, sieve, and magnesium perchlorate spatula (in drawer labeled magnesium perchlorate supplies on island for line B). You will also need the perchlorate waste jar that is by line A.
3. Remove the black tape on the container lid. Using gloves, sieve a small amount of the magnesium perchlorate from the jar through the large mesh sifter and into the waste jar. Close jar immediately when done and place black tape back onto jar (if the tape appears to be old replace with new tape from the magnesium perchlorate drawer).
4. Using the spatula or the Pyrex tube, gather the magnesium perchlorate into the Pyrex tube until 1/8 full. It is important to avoid collecting any powdery magnesium perchlorate.
5. Replace tubes onto the line
6. Dispose of the excess magnesium perchlorate, including what was in the old tubes, into the magnesium perchlorate waste container. Throw away the old tubes into the glass waste.

XI. How to make a slush

1. If there is no dry ice in the lab, go to the School of Physical Science supply store and buy 8 pounds of dry ice.
2. Use the ice shaver to grind the ice (Figure 15)
   a. Lift the top of the shaver and add dry ice to the ice shaver (use leathern gloves). Make sure there is a plastic container below the machine to catch the shaved ice.
   b. Replace lid and switch the motor "on". Press down the lid lever to push dry ice through the machine and into the plastic receptacle, and then turn off the motor.
3. Take a dewar containing methanol (1/3 full) and place into a tray.
4. Slowly add shaved dry ice to the methanol being careful not to add too much, at first, which will result in a loss of methanol by "boiling" out of the dewar.
5. Add dry ice until the mixture becomes the consistency of a stiff slushy. Stir with the spatula provided.
6. Place slush on the water trap located between valve # 3 and 4 (Figure 1).
XII. References


XIII. Overview

1. If graphitizing carbonates, weigh out samples and place them in a vacutainer.
2. Leach samples, rinse them twice with MilliQ water, and dry on heat block under a tent.
3. Evacuate air out of vacutainers, and add 0.8 mL Phosphoric Acid with syringe and needle.
4. Place samples on heating block for 20-40 minutes until no more bubbles are formed.
5. Prepare slushes, make sure the line is ready for graphitization, and replace bellows with needle.
6. Slide the vacutainer partially up the needle so that it does not puncture all the way through the septum, evacuate needle.
7. Close valve #2, transfer to cold trap to remove water. Skip ahead to step #11.
8. If graphitizing organics, make sure bellows is attached to line.
9. Clean combustion tube and blow dry, then score with glass cutter and place gently in bellows.
10. Evacuate bellows, then close valve #2, crack tube, and send to cold trap.
11. For both carbonates and organics, place liquid nitrogen on the tube between valves #4 and 5.
12. Send CO2 from cold trap to culture tube in liquid nitrogen.
13. After sample is frozen, open valves to the meas. vol. and check for non-condensables. If the gauge does not read them, check at the pump.
14. Record non-condensables on index card, and evacuate from line.
15. Transfer CO2 to meas. vol., let thaw, and record volume and multiplier on index card.
16. If there is more than 580 torr, manipulate the sample to remove excess CO2.
17. Transfer the CO2 to the desired reactor, making sure the bypass valves are closed. Close the reactor valve.
18. Repeat steps 5-7, 11-17 for carbonates or 8-17 for organics until all 12 reactors are full or you are out of samples.
19. If preparing a four aliquot sample, follow steps 8-15, but open valve #6 and record measure volume and multiplier on the sample index card.
20. Close valve #6, then open valve #7 and the first reactor valve to transfer sample, making sure the bypass valves are closed.
21. Close valve #7. Open valve #6. Allow the gas to expand. Repeat step 20, except transfer sample to the second reactor. Continue this process by filling the reactors in this order: 1, 2, 3, 3, 2
22. Open valve #6 and #7, and the final reactor to send the remaining sample to the last reactor.
23. Record volumes in reactors, and multiply values by 2 to get the required amount of H2.
24. Flush the line with H2 3x. In descending or ascending order, freeze down CO2 in reactors and add required H2. Record on sample index card.
25. After all samples have thawed, record final pressure in the sample index cards.

26. Place heaters on reactor tubes, allowing ¼ inch gap between the heater and end of the tube.

27. Make sure all reactors are at 550°C, start the graphitization monitor next to line B, and cycle the power switch off and on to start the graphitization reaction.

28. Input all information on sample index cards into the master list at Irvine 6.