MEASUREMENT OF DOC AND DIC

Objectives:
In this lab we will measure the concentrations of dissolved inorganic carbon (DIC) and dissolved organic carbon (DOC) in the water samples that were collected at the beginning of the quarter. In doing the measurements, students will reiterate earlier lessons on (1) how to construct calibration curves; (2) how to determine the error (uncertainty) in calibration curves (linear regressions) and in the sample concentrations predicted from the calibration curves. From the results, we will learn (1) ranges of concentrations of DIC and DOC in area waters; (2) factors influencing concentrations of DIC and DOC; (3) elements of carbonate chemistry including calculation of $pCO_2$.

Principles of Analysis
Measurement of carbon species

Carbon is a ubiquitous and important dissolved component of natural waters. Because $CO_2$ is present in the atmosphere and because carbonate minerals are widespread, there does not exist a natural water sample that totally lacks inorganic carbon species. Concentrations of inorganic carbon in natural waters range from about 10 $\mu$molar in acidic rain and fogwaters to nearly 1 molar in some saline lakes. The oceans, ground waters and lakes in contact with carbonate minerals typically have concentrations of 1-4 millimolar.

There also are few natural waters that are totally lacking in organic carbon. Particulate organic matter may consist of small organisms or remnant parts of larger organisms. Dissolved organic matter may consist of small molecules excreted by living organisms, or it may consist of larger molecules formed during the decay of organic matter. Even deep ground waters have dissolved organic matter as a result of the dissolution of ancient, fossil organic matter in the rocks. Concentrations of organic matter usually are expressed as mg/L of organic carbon. Because we seldom know the exact composition of the organic matter, we cannot convert these units to moles per liter of dissolved organic molecules. We can express the concentrations as moles per liter of organic carbon. Concentrations of dissolved organic carbon (DOC) range from 0.2-1 mg/L ($\approx$20-100 $\mu$mole/L) in ground waters and from 1 to 8 mg/L (100-800 $\mu$mole/L) in most surface waters. Swamps are notorious sources of dissolved organic matter; concentrations of DOC in brown bog waters may reach 150 mg/L ($\approx$12 mmole/L).

The sources and reactivity of inorganic and organic carbon are very different, and it usually is desirable to distinguish between these two fractions of carbon in natural waters. Filtration may be used to separate the dissolved from particulate phases of carbon, but then the chemical differences between inorganic and organic C must be exploited to distinguish these species. $CO_2$ absorbs infra red radiation very strongly, and this is commonly the basis for detection and quantification of inorganic carbon. By acidifying samples and purging with another gas (e.g., $N_2$), one can convert the inorganic carbon species in a water sample to $CO_2(g)$ (this will be explained later in the class lectures) and remove the $CO_2$ from the water into the gas stream. Passage of the gas stream through a beam of infra red light allows one to quantify the amount of $CO_2$ present (proportional to the absorbance or attenuation of the infra red light).
To measure organic carbon, one must convert the organic matter into CO₂. This oxidation may be achieved chemically (with a chemical oxidant such as persulfate), thermally (heating to 550°C), or photochemically (exposure to intense ultraviolet light). The carbon analyzer used in this lab combines chemical and photochemical oxidation. Clearly, to measure organic carbon one must first remove the inorganic carbon present.

**Figure 1.** Schematic illustration of the Total Carbon Analyzer.

In the Total Carbon Analyzer used in this lab (diagrammed above), the sample is injected in the sample inlet. If inorganic carbon (DIC) is to be measured, acid will be added from the syringe pump on the left. The sample will then pass to the membrane module where the CO₂ will diffuse across a membrane to a solution of higher pH. In this solution, the CO₂ reacts to form bicarbonate ions that may be quantified by the increase in conductivity of the solution (see Lab 1). If organic carbon (DOC) is to be measured,
the procedure is the same except that an oxidant (peroxydisulfate or persulfate) is added with the acid. The sample then is exposed to intense UV irradiation in the oxidation reaction cell. The CO₂ generated from the oxidation of the organic matter then is measured in the same fashion as the DIC. The DOC concentration equals the total carbon concentration minus the DIC concentration. For improved accuracy and precision, one may purge the DIC from a sample prior to measurement of DOC.

**Chemical effects of dissolved organic and inorganic carbon**

Carbonate species (primarily HCO₃⁻) frequently are the dominant anion in fresh waters. Hence they are a major contributor to the conductivity of fresh waters. As the major anion, carbonate species also regulate the solubility of several toxic trace metals and, in some cases, of the essential nutrient phosphate. The carbonate is derived from either the dissolution of carbonate minerals (calcite, dolomite), the activity of microorganisms, or the weathering of rocks by CO₂. The cations that accompany the carbonate anions can indicate the source of the carbonate. Calcium and magnesium, derived both from dissolution of carbonates and from weathering of silicate rocks, have a major economic impact on water use. These cations with their accompanying carbonate anions comprise the "hardness" of a water. Considerable money is invested in "softening" waters (removing the hardness). Precipitation of calcium carbonate in hard waters also has major economic impacts in the form of plugging of heaters, heat exchangers, and pipes. Carbonate species also are the major buffer for natural waters; the ability of natural waters to resist acidification (the definition of alkalinity) depends on the concentration of carbonate species. On the other hand, high concentrations of dissolved CO₂ can decrease the pH of natural waters. In other labs you determine the influence of inorganic vs. organic carbon on alkalinity, and on the solubility and complexation of metals.

How can carbonate species both cause and resist the acidification of water? Again, this is related to the source of the carbonate species. If the carbonate species are derived originally from CO₂ (either due to atmospheric CO₂ dissolving in water in response to uptake by photosynthesis or due to release of CO₂ by aerobically-respiring bacteria), they will have a net acidifying effect. Any process that adds CO₂ to water causes a lowering of pH. This decrease in pH is caused by the following reactions:

1. \[ \text{CO}_2(g) \rightleftharpoons \text{CO}_2(aq) \]
2. \[ \text{CO}_2(aq) + \text{H}_2\text{O} \rightleftharpoons \text{H}_2\text{CO}_3 \]
3. \[ \text{H}_2\text{CO}_3 \rightleftharpoons \text{H}^+ + \text{HCO}_3^- \]
4. \[ \text{HCO}_3^- \rightleftharpoons \text{H}^+ + \text{CO}_3^{2-} \]

The net result of these reactions is an increase in concentration of hydrogen ions or an acidification. The carbonation of soft drinks also acidifies the drinks by these reactions.

If, on the other hand, the carbonate species are derived from weathering (dissolution) of carbonate rocks (limestone, dolomite) or silicate rocks or if the carbonate comes from anaerobic respiration of bacteria (e.g., sulfate reduction), then the carbonate species will contribute to *alkalinity*, the ability of waters to resist acidification. In these cases, reactions 3 and 4 above occur in reverse, and a net uptake of H⁺ occurs. Addition of acid to a water with a high concentration of HCO₃⁻ or CO₃²⁻ will drive reactions 3 and 4 further to the left, and much of the acid will be neutralized.
The major chemical effects of dissolved organic carbon are to either acidify or buffer waters, and to complex trace metals and thereby facilitate their transport. Dissolved organic carbon consists of a very complex mixture of organic molecules; we cannot even characterize the structure of many of these molecules. Many of the molecules have carboxylic functional groups (RCOO') that act as weak acids or bases. You will learn much more about this topic later in the course. Similarly to carbonate species, if the DOC enters a water in the acid form (RCOOH) as it does in peat bogs, then it can have a net acidifying effect. If it is already neutralized and enters the water as a base (RCOO'), then it can contribute to the alkalinity. You can determine whether the DOC in your samples functions to acidify them or contributes to their alkalinity in the titration lab. The wide variety of functional groups in DOC result in DOC having the ability to bind to or complex with numerous trace metals. Again, this is a topic that we will study in detail later in the course. When the metals are bound to the DOC they are not toxic, they are more soluble, and they may be transported long distances with the DOC. Fish in brown water streams do not suffer from aluminum toxicity even in streams receiving acid rain. In a subsequent lab you will examine the effects of the DOC in your samples on the binding of Fe and Cu.

Procedure

The water samples should be removed from the refrigerator at the beginning of the lab period to allow them to come to room temperature. The chemicals for standards will transferred (in Al weighing dishes or beakers) from the oven at 60 °C to the desiccator to cool. At this point the TA will take you on a tour of the analytical lab and explain the instruments that are to be used for the analyses. Each group (3-4 students) will be responsible for making standards for either DIC or DOC and preparing their three samples. The standards must span the range of concentrations expected in all of the samples and must include a standard blank. The TA will analyze the samples after the lab period and have the results ready for you on the following day. From the raw results, each group will be responsible for calculating the concentrations and the uncertainties for the substance (DIC or DOC) for which they made standards.

1. DIC analysis

   Chemicals: NaHCO₃, Ascarite, N₂

   The TA will place the NaHCO₃ in the oven at 60°C to dry. Obtain 1 L of Milli-Q water in a large Erlenmeyer flask and begin to purge with N₂ that is passed through an ascarite trap to remove CO₂. After 45 minutes, place the bicarbonate in a desiccator to cool. Then make a 250 mM stock solution in a 100 mL volumetric flask using the deaerated water. From the stock solution make standards in 50-mL volumetric flasks of 0, 5.0, and 25 mM also using the deaerated water. From the 25 mM solution make standards (50-mL) of 0.5 and 1 mM, and from the 5 mM standard make a standard of 0.1 mM concentration. Transfer the standards to the analyzer vials. Cap the vials firmly with septa. Label all vials clearly with what is in them and with your group identifier. Clearly record in your lab notebook how all standards were made. To determine the detection limit for the instrument, the least concentrated standard must be analyzed in triplicate. To assess the precision of the sample DIC measurements, a sample also should be analyzed in triplicate. Transfer the samples to the analyzer vials, label the vials and cap them with
septum covers. Deliver all vials to the TA’s. Rinse all glassware three times with tap water followed by three times with milli-Q water.

2. DOC analysis
   Chemicals: Potassium biphthalate, phosphoric acid (1 M).
Place the biphthalate in an aluminum weighing dish in the oven at 60°C for 45 minutes. Remove to the desiccator to cool. Make a stock solution of 100 mg/L C in a 100-mL volumetric flask. Using 50-mL volumetric flasks dilute the stock solution to make standards of 0, 0.4, 1.2, 2, 4, and 20 mg/L C. Note that you will have to use the 20 mg/L standard to make the 0.4 and 1.2 mg/L standards. Transfer the standards to the analyzer vials, add two drops of acid to each vial, and label the vials with the standard concentration and your group identifier. Transfer the samples (2) to the analyzer vials, label the vials and cap them with septum covers. To determine the detection limit for the instrument, the least concentrated standard must be analyzed in triplicate. To assess the precision of the sample DOC measurements, a sample also should be analyzed in triplicate. Deliver all vials to the TA’s. Rinse all glassware three times with tap water followed by three times with Milli-Q water.

Data Analysis for Following Lab Session
For the week following this lab, you should prepare the following:
1. The detection limit of the analyte for which your group prepared standards;
2. The precision (standard error) of the standards prepared by your group;
3. The concentrations of DIC and DOC in your samples;
4. The uncertainty in the predicted DIC or DOC concentrations in the samples.

Your lab report will include the following additional data analyses:
5. The standard deviation for replicate measurements of your sample; (Not in 2008)
6. An assessment of the sample contamination that occurred and a correction for such contamination if it occurred.
7. A graph of the standard curve for the analysis for which your group prepared standards;
   We will discuss the results of all of the groups and compare the sample concentrations. Look over the questions that you are to address in the lab report, and be prepared to ask the TA to clarify any items that are not clear to you.

Lab Report
   In the introduction you should briefly discuss the significance of DIC and DOC in your samples. In the results section, you should present the items listed above in a clear fashion. Mention any significant discrepancies between your results and those of other groups. Tabulate the concentrations of DIC and DOC in your samples. You also should calculate the concentrations of the carbonate species (CO$_3^{2-}$, HCO$_3^-$, and H$_2$CO$_3^*$). In the Discussion, you should address the following issues:
1. What is the range of DIC and DOC among the samples? Is this range consistent with what is stated in the handout? Are the values consistent with the characteristics of the sources? Comment on any surprising values.
2. Is there any relationship between the pH of the samples and the DIC or DOC concentrations? Why or why not?

3. Is there any relationship between conductivity and either DIC or DOC? Why or why not?

4. Is there any relationship between the concentrations of DIC and DOC in the samples? Why or why not?

5. How do your calculated concentrations of HCO$_3^-$ (based on DIC) compare with the measurements of alkalinity? Is there now a balance between cation and anion concentrations in your samples?

Materials needed:
Chemicals: NaHCO$_3$, Ascarite, N$_2$
   Potassium biphthalate, H$_3$PO$_4$ (1 M),
per group:
   7 50-mL volumetric flasks
   1 100-mL volumetric flask
   ≈ 1 aluminum weighing tray
   weighing paper
   spatula
   milli-Q water
   marking tape
   marker
Miscellaneous:
   frits or pasteur pipets and tubing for sparging
   droppers
   pipets - glass or eppendorf as appropriate
   analytical balance(s)
   drying oven (60°C)
   desiccator(s)
   tube for ascarite, cotton wadding
Data records

Group

Analyte (DIC or DOC)

Chemical used for Stock Solution

Mass used for stock solution

Volume of stock solution prepared

Concentration of Stock Solution

<table>
<thead>
<tr>
<th>Nominal concentration</th>
<th>Vol. of Stock used</th>
<th>Volumetric flask size (mL)</th>
<th>Actual Conc. of Std.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blank</td>
<td>0</td>
<td></td>
<td>0</td>
</tr>
</tbody>
</table>

Samples prepared:


