SOP 1

Water sampling for the parameters of the oceanic carbon dioxide system

1. Scope and field of application

This SOP describes how to collect discrete samples, from a Niskin or other water sampler, that are suitable for the analysis of the four measurable inorganic carbon parameters: total dissolved inorganic carbon, total alkalinity, pH and CO_2 fugacity.

2. Principle

A sample of sea water is collected in a clean glass container in a manner designed to minimize gas exchange with the atmosphere (note: CO_2 exchange affects the various carbon parameters to differing degrees ranging from the very sensitive CO_2 fugacity, $f(CO_2)$, to alkalinity which is not affected by gas exchange). The sample may be treated with a mercuric chloride solution to prevent biological activity, and then the container is closed to prevent exchange of carbon dioxide or water vapor with the atmosphere.

3. Apparatus

The sample containers are somewhat different depending on which parameter is being collected, but the basic concept is similar for the four possible inorganic carbon samples. In general, one needs a flexible plastic drawing tube, a clean[1] glass sample container with stoppers, a container and dispenser for the mercuric chloride solution (if it is being used) and a sampling log to record when and where each of the samples were collected.

3.1 Drawing tube

Tygon[®] tubing is normally used to transfer the sample from the Niskin to the sample container; however, if dissolved organic carbon samples are being collected from the same Niskins, then it may be necessary to use silicone tubing to prevent contamination from the Tygon[®]. The drawing tube can be pre-treated

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[1] Cleaning sample containers by precombustion in a muffle furnace will remove any organic carbon and associated microorganisms. Some groups soak the bottles in 1 N HCl; however, care must be taken to remove all residual acid during rinsing.

by soaking in clean sea water for at least one day. This minimizes the amount of bubble formation in the tube when drawing a sample.

3.2   Sample container

The sample container depends on the parameter being measured. Typically, the $f(CO_2)$ samples are analyzed directly from the sample container so they are collected in 500 cm^3 volumetric flasks that have been pre-calibrated for a documented volume and sealed with screw caps that have internal plastic conical liners. Samples for pH are also typically analyzed directly from the sample containers. For spectrophotometric pH measurements, the samples are collected directly into 10 cm path-length optical cells and sealed with polytetra-fluoroethylene (Teflon$^{®}$) caps ensuring that there is no headspace. For C_T and A_T, high quality borosilicate glass bottles, such as Schott Duran (l.c.e. 32×10^{-7} K^{-1}), are recommended for both temporary and longer term storage. The bottles should be sealed using greased ground glass stoppers held in place with some form of positive closure, or in some alternate gas-tight fashion[2].

3.3   Mercury dispenser

The $f(CO_2)$ and C_T samples should be poisoned with a mercuric chloride solution at the time of sampling. The A_T samples have historically been poisoned as well, but tests have suggested that poisoning may not be required if open ocean samples are kept in the dark at room temperature and are analyzed within 12 hours. Samples for pH are typically not poisoned because the sample size is relatively small and the samples are usually analyzed very quickly after sampling. Although any appropriately sized Eppendorf pipette can be used to add the mercuric chloride solution, it may be more convenient to use a repipetter that can be mounted near the sample collection area. All equipment should be properly labeled for safety.

4.   Reagents

4.1   Mercuric chloride solution

Samples collected for $f(CO_2)$, C_T, and, in some cases, A_T, should be poisoned with a mercuric chloride solution to stop biological activity from altering the carbon distributions in the sample container before analysis. A typical solution is saturated mercuric chloride in deionized water. However, saturated solutions have been known to clog the pipette in very cold weather, so some investigators use twice the volume of a 50% saturated solution. Standard volumes used for saturated solutions are 0.05–0.02% of the total sample volume.

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[2] Some groups use screw-cap bottles with apparent success, but this method has not been thoroughly tested and should not be used if samples are to be stored for extended periods.

4.2 Stopper grease

C_T and A_T samples are typically collected in borosilicate glass bottles with ground glass stoppers. To form an airtight seal, the stoppers should be greased. Apiezon® L grease has been found to be suitable for this purpose; other greases may also work. Care should be taken not to transfer the grease onto the Niskin bottle as this could interfere with other analyses.

5. Procedure

5.1 Introduction

Collection of water at sea from the Niskin bottle (or other sampler) must be done soon after opening the sampler and before much other water has been removed from it. This is necessary to minimize exchange of CO_2 with the air space in the sampler which affects all carbon parameters except A_T. Other gas samples (*e.g.*, He, CFCs, O_2) have faster exchange rates than CO_2 and are usually sampled before carbon, but it is desirable that the carbon samples be collected before the Niskin bottle is half empty and within 10 minutes of it being first opened. A typical sampling order for carbon is $f(CO_2)$, pH, C_T, then A_T.

5.2 Filling procedure

Rinse the sample bottle — If the bottle is not already clean, rinse it twice with 30–50 cm³ of sample to remove any traces of a previous sample.

Fill the sample bottle — Fill the bottle smoothly from the bottom using a drawing tube which extends from the Niskin drain to the bottom of the glass sample bottle. For $f(CO_2)$, pH, and C_T, it is critical to remove any bubbles from the draw tube before filling. Overflow the water by at least a half, and preferably by a full, bottle volume[3].

Adjust the headspace — A headspace of 1% of the bottle volume is left to allow for water expansion (see Annexe to this procedure), *i.e.*, 2.5 cm³ for a 250 cm³ bottle. This can be achieved by pinching off the draw tube before removing it from the sample bottle or removing excess water using a plastic pipette with a bulb. pH samples should not have a headspace.

Add mercuric chloride — Mercuric chloride is added to poison the sample; the recommended minimum amount is about 0.02% by volume of a saturated aqueous solution. Thus to poison a 250 cm³ sample requires 0.05 cm³ (50 μl) of saturated mercuric chloride (or 0.10 cm³ of a 50% saturated solution). Maximum amount is 0.1% by volume of a saturated aqueous solution, or a smaller percentage than measurement precision of C_T and $f(CO_2)$.

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[3] The amount of overflow water can be estimated by measuring how long it takes to fill a sample bottle, and allowing the water to flow for a period of 1.5 times that.

Close the bottle — Seal the bottle carefully to ensure that it remains gas-tight. If it is to be sealed using a greased ground glass stopper[4], first wipe the excess water from the ground glass in the bottle neck, then insert the stopper completely, and finally twist the stopper to squeeze the air out of the grease to make a good seal.

Finally, secure the lid — Use a rubber band or other positive closure, then invert the bottle several times to disperse the mercuric chloride solution thoroughly.

5.3   Sample storage

The samples should be stored in a cool, dark, location (preferably refrigerated but not frozen) until use.

5.4   Sample documentation

The following information must be recorded in the sampling logbook at the time of sampling:
- Time and date when taken;
- Full name of person who took sample;
- Location: an unambiguous designation of the station, cast, and bottle number from which the sample was taken;
- Container designation: a number or alphanumeric symbol unique to the sample container;
- Comments: additional information such as conditions when sampling, problems with sample collection, *etc.*

6.   Quality assurance

Some duplicate sampling is recommended, both from the same sampler (*e.g.*, Niskin bottle) and, if possible, from two samplers tripped together at the same depth, to assess the quality of the sampling procedures.

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[4] The recommended procedure for re-greasing (or greasing) a stopper is as follows: (a) wipe the stopper with a tissue to remove as much grease as possible and (b) grease the stopper with 4 strips of grease, each strip extending two thirds of the way from the top towards the bottom of the ground portion of the stopper. This provides a path for air to escape when the stopper is inserted into the neck of the bottle.

Annexe

How large a headspace should be left in a sample bottle?

The volume of the headspace is chosen so as to leave room for expansion of the sea water on warming, while being sufficiently small to minimize the amount of gas exchange between the headspace and the bulk of the sample. The closure system must be adequate to retain the pressure exerted by the expansion.

The apparent change in the volume of a fixed mass of sea water can be calculated by allowing for the change in the density of the sea water and the expansion of the glass container. The total change over the temperature range 0–40°C is about 1%. (The effect of expansion on the volume of the borosilicate glass bottle is only 0.04% over this range.) One third of this expansion occurs on heating the sea water from 0 to 20°C, the remaining two thirds on heating it from 20 to 40°C.

The pressure in the headspace of a container heated from a temperature t_1 to t_2 can be estimated, allowing for the following:
- the expansion of the sea water in the bottle,
- the change in solubility of gases such as N_2, O_2, Ar,
- the thermal expansion of the gas phase,
- the change in the vapor pressure of H_2O in the gas phase.
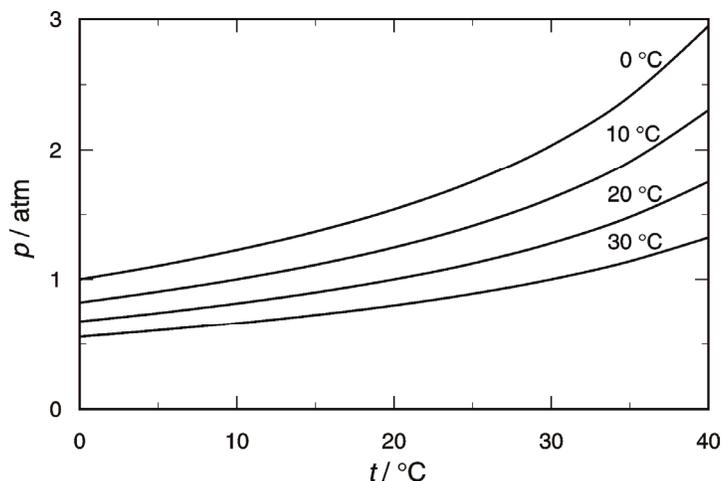
Fig. 1 Pressure in headspace with $r = 0.01$ as a function of temperature for various initial temperatures. In calculating this, it was assumed that the gases—N_2, O_2, Ar, H_2O—were initially at solubility equilibrium with sea water at the starting temperature, that they behaved ideally and that the initial pressure in the bottle at the indicated temperature was 1 atm.

Defining the initial headspace ratio,

$$r = \frac{V(\text{headspace})}{V(\text{sea water})} \,, \tag{1}$$

allows the calculation of the approximate pressure in the headspace of a closed container as a function of temperature. Clearly, if cold samples (< 10°C) are likely to be heated above 30°C, there is a risk of them leaking if the headspace ratio is significantly less than 1%. There is, however, an additional factor to be taken into account when determining the optimal headspace size: gas exchange with the headspace. The change in total dissolved inorganic carbon (ΔC_T) resulting from this gas exchange can be derived from mass balance considerations:

$$\Delta C_T = \frac{\left(\dfrac{p(CO_2)V}{T}\right)_{\text{initial}} - \left(\dfrac{p(CO_2)V}{T}\right)_{\text{final}}}{R \cdot m(\text{sample})} \tag{2}$$

where $p(CO_2)$ is the partial pressure of CO_2 in a headspace of volume V and at a temperature T corresponding to the initial condition (when the bottle is closed) and the final condition (when the bottle is analyzed), R is the gas constant and m(sample) is the mass of the sample.

The volume of the headspace decreases as the contents heat up (due to the expansion of the sea water) partially compensating for the decrease in the solubility of the various gases, thus the greatest loss of CO_2 will occur if the sea water has a high $p(CO_2)$ but does not warm up significantly in the container. Even then, provided that the headspace ratio is less than 0.01, ΔC_T will be less than 0.5 μmol kg^{-1}.

Gain or loss of CO_2 gas is not significant when collecting discrete samples for alkalinity measurement; however, if $p(CO_2)$ is to be measured, the so-called "buffer factor" comes into play and the resultant relative error in $p(CO_2)$ is approximately 10 times that in C_T, *i.e.*, for a change of –0.5 μmol kg^{-1} in C_T, the change in $p(CO_2)$ is about –0.25%. This corresponds to a change in pH of about +0.001.

A headspace of 1% is thus optimal for the collection of CO_2 samples, provided that they will not be exposed to temperature changes of 30°C or more. If this cannot be assured, it is preferable to allow a larger headspace and to estimate the appropriate correction.