Determination of total dissolved inorganic carbon in freshwaters by reagent-free ion chromatography

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Available online 18 April 2006

Abstract

Studies of inorganic carbon cycle in natural waters provide important information on the biological productivity and buffer capacity. Determination of total inorganic carbon (TIC) and dissolved carbon dioxide gives an indication of the balance between photosynthesis and respiration by biota, both within the water column and sediments, and carbon dioxide transfers from the water column to the atmosphere. There are few methods to measure and distinguish the different forms of inorganic carbon, but all require a measure or an indirect quantification of total inorganic carbon. A direct measurement of TIC in water is made possible by the introduction of electrolytic generated hydroxide eluent in ion chromatography which allows to detect a chromatographic peak for carbonate. The advantage of this method is that all the inorganic forms of carbon are converted in carbonate at eluent pH and can be detected as a single peak by conductivity detection. Repeatability of carbonate peak was evaluated at different levels from 0.02 to 6 mequiv. l $^{-1}$ both in high purity water and in real samples and ranged from 1 to 9%. The calibration curve was not linear and has to be fitted by a quadratic curve. Limit of detection was estimated to be 0.02 mequiv. l $^{-1}$. Accuracy has been estimated by comparing ion chromatography method with total inorganic carbon calculated from alkalinity and pH. The correlation between the two methods was good ($R^2 = 0.978$, $n = 141$). The IC method has been applied to different typologies of surface waters (alpine and subalpine lakes and rivers) characterised by different chemical characteristics (alkalinity from 0.05 to 2 mequiv. l $^{-1}$ and pH from 6.7 to 8.5) and low total organic carbon concentrations. This analytical method allowed to describe the distribution of TIC along the water column of two Italian deep lakes.

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Keywords: Ion chromatography; Total inorganic carbon; Freshwaters

1. Introduction

Studies of inorganic carbon cycle in natural waters provide an important indicator of their biological productivity and buffer capacity. Determination of total dissolved inorganic carbon (TIC), total alkalinity (Talk) and dissolved carbon dioxide gives an indication of the balance between photosynthesis and respiration by biota, both within the water column and sediments, and carbon dioxide transfers from the water column to the atmosphere [1,2]. Research on carbon dynamics indicates that oceans and terrestrial areas are generally sink for carbon dioxide [3]. However it is not clear whether freshwater bodies release or take up carbon dioxide [4]. On the consequence there is a huge interest in developing reliable analytical methods for the accurate determination of inorganic carbon species.

By knowing the concentrations of TIC and pH, it is possible to estimate the equilibrium concentrations in water of all inorganic carbon forms, according to the following equations:

$$\text{H}_2\text{CO}_3 = \text{TIC}_0$$
$$\text{HCO}_3^- = \text{TIC}_1$$
$$\text{CO}_3^{2-} = \text{TIC}_2$$

where

$$\alpha_0 = \left(1 + K_1 + K_1K_2 \frac{[\text{H}^+]^2}{[\text{H}^+]^2}\right)^{-1}$$
$$\alpha_1 = \left(\frac{[\text{H}^+]}{K_1 + 1 + K_2/[\text{H}^+]^2}\right)^{-1}$$

where

$K_1$ is the acid dissociation constant of carbonic acid
$K_2$ is the acid dissociation constant of the bicarbonate
$K_3$ is the acid dissociation constant of the carbonate
$[\text{H}^+]$ is the concentration of hydrogen ions

$\alpha_0$ is the ratio of the equilibrium concentrations of the species
$\alpha_1$ is the ratio of the equilibrium concentrations of the species

$\alpha_0$ and $\alpha_1$ are related to the pH of the water

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doi:10.1016/j.chroma.2006.03.015
TAlk and α methods based on sample acidification, CO2 stripping followed exclusion coupled with conductivity detection[10]. Carbonate of a weak acid the chromatographic technique of choice was ionization has not been widely diffused. Since carbonate is the anion concentration step[11], or by capillary electrophoresis[12], but few non dispersive-IR [9].

by the measure of carbon dioxide by coulometric titration[8] or ionic strength.

alkalinity and pH values, taking into account temperature and concentration of anions from organic acids is negligible TAlk is defined by the following equation:

\[
TAlk = 2 \left[ CO_3^{2-} \right] + \left[ HCO_3^- \right] + \left[ OH^- \right] - \left[ H^+ \right]
\]

This calculation needs the accurate knowledge of thermodynamic constants, the dependence on temperature and ionic strength of the equilibrium constants.

Alternatively the excess partial pressure of CO2 (pCO2) and TIC can be calculated from alkalinity and pH values. This procedure has been applied in studies on freshwater systems[5–7], but the calculation outputs have not been validated by direct measures of total inorganic carbon. In fact improving accuracy of carbon speciation in a water sample depends also on the availability of a reliable direct measure of total inorganic carbon.

In oceanographic campaigns, planned to evaluate the sinking capacity of ocean for CO2, TIC was determined by automated methods based on sample acidification, CO2 stripping followed by the measure of carbon dioxide by coulometric titration[8] or non dispersive-IR [9].

The use of chromatographic methods for carbonate determination has not been widely diffused. Since carbonate is the anion of a weak acid the chromatographic technique of choice was ion exclusion coupled with conductivity detection[10]. Carbonate was also determined in high purity water by IC after a preconcentration step[11], or by capillary electrophoresis[12], but few validation data have been reported.

A direct measurement of TIC is made possible by the introduction of electrolytic generated hydroxide eluent in ion chromatography which allows to detect a chromatographic peak for carbonate [13]. The advantage of this method is that all the inorganic forms of carbon are converted in carbonate in the basic eluent and can be detected as a single peak by conductivity detection. Because samples are filtered before injection, dissolved inorganic carbon is actually measured.

The aim of our work has been to test the feasibility of using ion chromatography as a reliable technique for the determination of TIC. Measurements carried out by IC on samples of Italian lakes and rivers are compared with TIC values calculated from alkalinity and pH values, taking into account temperature and ionic strength.

2. Experimental

2.1. Materials and chemicals

All solutions were prepared by dissolving salts of analytical grade in ultra pure (18 MΩ·cm−1 quality) water produced by a Milli-Q system (Millipore, Bedford, MA, USA). Concentrated standard solutions were prepared from sodium hydrogen carbonate (Merck, Darmstadt, Germany) dried at 105 °C for 2 h and kept in a desiccator. Working standard solutions were daily prepared by dilution of these concentrated solutions with ultra pure water.

2.2. Ion chromatographic determination

Chromatographic analyses were performed on a DX-320 Ion Chromatograph ( Dionex, Sunnyvale, CA, USA) equipped with AS50 Autosampler, GS50 Gradient Pump, LC25 column compartment and ED50 conductivity detector. Separations were carried out using a Dionex IonPac AS17 (250 mm × 4.0 mm) analytical column and an IonPac AG17 (50 mm × 4.0 mm) guard column.

Anions were detected using an electrochemically regenerated Atlas suppressor coupled to conductivity detection. Injection volume was 100 µL. Isocratic 12 mM KOH eluent was electrolytically generated at the flow rate of 1 ml min−1 using an EG40 Eluent Generator equipped with EluGen-OH cartridge and a continuously regenerated anion trap column.

All measurements were carried out at 35 °C and all samples were filtered through 0.45-µm filters. Dionex Chromeleon 6.40 chromatography software controlled data collection and the operation of all components in the system.

2.3. Total alkalinity determination

TAlk was determined by computer guided acidimetric titration using the Gran method [14] with Radiometer ABU91 and TDM900 automatic titrators. In the range 0–5 mequiv. L−1, 30–50 ml of sample were titrated with 0.05N HCl [15].

2.3.1. Calculation of total inorganic carbon

TIC was calculated from TAlk, pH, ionic strength and temperature using the stoichiometric approach of inorganic carbon equilibrium [1].

\[
TIC = \frac{TAlk - K_2}{\alpha_2} + \left[ H^+ \right] \frac{1}{\alpha_1 + 2\alpha_2}
\]

Because of the low TOC concentrations (all samples were below 5.0 mg C L−1 and 95% of the samples below 2.0 mg C L−1), the influence of organic anions on alkalinity was considered negligible. The calculation took into account the dependence of the inorganic carbon (K1, K2) and water (Kw) equilibrium constants from temperature and ionic strength [1,5].
2.4. Total carbon determination by high temperature catalytic oxidation

Total carbon (TC) determinations by high temperature catalytic oxidation (HTCO) were performed using a simultaneous total carbon and total nitrogen analyser (Skalar Formacs TOC/TN Analyzer). The sample (typically 150 μl) was injected into the high-temperature reactor (850 °C) with CeO2 catalyst. Carbon dioxide absorbance is measured at 4.2 μm by a non-dispersive IR detector.

3. Results and discussion

The aim of this work was to test the possibility of determining TIC by IC on a routinary basis during a 6 month monitoring of different rivers and lakes in Northern Italy. In the monitoring campaign we used the IC method already validated for the determination of main anions (chloride, nitrate and sulphate), which was based on the separation on an Ion Pac AS17 column with a 12 mM KOH eluent, without a specific optimisation for carbonate elution. In fact these chromatographic conditions allowed to detect the carbonate peak sufficiently resolved from adjacent peaks (Fig. 1).

Before starting with the measurements we collected data on the quality parameters of the carbonate determination in order to validate this method.

3.1. Calibration curve

A calibration curve was built to draw the relationship between carbonate amount (expressed in mM) and chromatographic peak area from 0.02 to 6 mM. Standard solutions were prepared from sodium hydrogen carbonate: the use of sodium carbonate as starting salt for preparing standard solutions must be avoided because atmospheric carbon dioxide can dissolve into basic carbonate solutions. Peak areas of standards have been subtracted by mean blank value measured in ultrapure water used for the standard preparation. Since carbonic acid is a weak acid, the calibration curve was not linear and was fitted by a 2nd order polynomial curve with the following equation:

\[ y = -0.47x^2 + 3.0x + 0.082 \ (R^2 = 0.992) \]

Looking at the peak shape, we can observe that the peak asymmetry increases as carbonate concentration increases (Fig. 2) and the marked tailing should have a negative effect on the accuracy of area integration and on the resolution with the neighbouring peaks. The distortion of the peak shape can be attributed to the interconversion of carbonate into hydrogencarbonate in the column, due to a local pH variation of the eluent [17]. This chromatographic behaviour may induce a systematic error at concentrations higher than 1 mM, but this problem can be overcome by reducing injection volume that in our standardised procedure was 100 μl.

3.2. Repeatability and detection limit

Repeatabilities of carbonate determination by IC, estimated from RSD values of consecutive injections (n ≥ 6) in a concentration range from 0.02 to 6.0 mM both in high purity water and in real samples, ranged from 1 to 9%. Intra-day repeatabilities of IC method was of the same order of inter-day repeatabilities of alkalinity determination by the titrimetric method estimated from control charts obtained in our laboratories (1–6% in the 0.1–6 mequiv. l⁻¹ range) [15].

Limit of detection (LOD) and limit of quantification (LOQ) were estimated as the mean blank value plus, respectively, 3- or 10-fold the standard deviation of the blank (n = 8) giving a LOD of 0.02 mM and a LOQ of 0.03 mM. The blank was determined by injection of ultrapure water samples in equilibrium with atmospheric CO₂.

3.3. Comparison between calculated TIC and IC measured TIC

The routine determinations carried out in our monitoring program on Italian lakes and rivers included titrimetric alkalinity determination by Gran method. Knowing TAlk, pH, ionic strength in each sample, total inorganic carbon content could be calculated by the equilibrium equations of CO₂ species in water. This calculated TIC (TICcalc) has been compared with IC measured TIC (TICIC) in the same set of sample. The bias due to the presence of dissolved CO₂, which is
Table 1

Chemical characteristics of the sample set

<table>
<thead>
<tr>
<th>Variables</th>
<th>Unit</th>
<th>Min.</th>
<th>5 percentile</th>
<th>50 percentile</th>
<th>95 percentile</th>
<th>Max.</th>
<th>Mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH</td>
<td></td>
<td>6.66</td>
<td>6.86</td>
<td>7.28</td>
<td>7.78</td>
<td>8.47</td>
<td>7.30</td>
</tr>
<tr>
<td>Conductivity (20 °C)</td>
<td>µS cm⁻¹</td>
<td>19.3</td>
<td>29.1</td>
<td>63.4</td>
<td>216.3</td>
<td>314.9</td>
<td>93.9</td>
</tr>
<tr>
<td>Total Alkalinity</td>
<td>mequiv. l⁻¹</td>
<td>0.050</td>
<td>0.12</td>
<td>0.28</td>
<td>1.27</td>
<td>2.11</td>
<td>0.45</td>
</tr>
<tr>
<td>Sulphate</td>
<td>mg l⁻¹</td>
<td>0.7</td>
<td>0.9</td>
<td>2.7</td>
<td>13.0</td>
<td>35.6</td>
<td>4.5</td>
</tr>
<tr>
<td>Nitrate</td>
<td>mg N l⁻¹</td>
<td>0.02</td>
<td>0.04</td>
<td>1.10</td>
<td>3.05</td>
<td>650.0</td>
<td>12.7</td>
</tr>
<tr>
<td>Ammonium</td>
<td>mg N l⁻¹</td>
<td>0.005</td>
<td>0.006</td>
<td>0.01</td>
<td>0.12</td>
<td>3.90</td>
<td>0.09</td>
</tr>
<tr>
<td>Calcium</td>
<td>mg l⁻¹</td>
<td>3.2</td>
<td>3.9</td>
<td>7.1</td>
<td>34.0</td>
<td>52.5</td>
<td>11.3</td>
</tr>
<tr>
<td>Magnesium</td>
<td>mg l⁻¹</td>
<td>0.1</td>
<td>0.3</td>
<td>1.6</td>
<td>8.5</td>
<td>12.7</td>
<td>2.3</td>
</tr>
<tr>
<td>Sodium</td>
<td>mg l⁻¹</td>
<td>0.2</td>
<td>0.4</td>
<td>2.9</td>
<td>7.0</td>
<td>38.0</td>
<td>4.0</td>
</tr>
<tr>
<td>Potassium</td>
<td>mg l⁻¹</td>
<td>0.2</td>
<td>0.4</td>
<td>1.2</td>
<td>2.2</td>
<td>3.8</td>
<td>1.2</td>
</tr>
<tr>
<td>Silica</td>
<td>mg Si l⁻¹</td>
<td>0.1</td>
<td>0.3</td>
<td>3.1</td>
<td>6.8</td>
<td>8.0</td>
<td>3.6</td>
</tr>
</tbody>
</table>

Fig. 3. Correlation between total inorganic carbon measured by IC (TIC IC) and total inorganic carbon calculated from alkalinity (TIC CALC). Dotted lines are 95% prediction limits, broken lines are 95% confidence limits and the continuous line is linear fit. Expanded view shows good fit also for low TIC values.

measured only by IC and not by titrimetry, can be ignored because at the typical pHs of surface freshwater samples (6.5–8.5) dissolved CO₂ concentration is negligible. This evidence is no more true if deep layers of water bodies, where CO₂ partial pressure is higher, are analysed, as shown in Section 3.5.

The IC method has been applied to surface waters (rivers, alpine lakes and surface layers of deep subalpine lakes) with different chemical characteristics (alkalinity from 0.05 to 2 mM and pH from 6.7 to 8.5) (Table 1).

Data are well correlated (R² = 0.978), and the regression line was y = 1.11 (±0.01) x − 0.032 (±0.008). Both angular coefficient and intercept are statistically significant (p < 0.0001) suggesting that there is a small but significant systematic difference between calculated and measured (Fig. 3). This systematic deviation can be attributed also to the uncertainties in the thermodynamic constants used in the calculation model.

Percent differences, calculated as

\[ \text{%Difference} = \frac{\text{TIC}_{\text{calculated}} - \text{TIC}_{\text{measured}}}{\text{TIC}_{\text{measured}}} \times 100 \]

are shown in Fig. 4 as a function of TIC concentration. Relative differences are equally distributed around 0%, with a light sys-
tematic effect of underestimation for samples at concentrations higher than 1 mM, as already explained in Section 3.1. Eighty-three percent of the whole data set is comprised between ±20%: outside these limits there are only the lowest concentration samples and a group of samples collected from the same river (Ticino river, tributary of Lake Maggiore) which are also the only samples outside the prediction band (95% of confidence) in Fig. 3. This river is characterised by the highest sulphate concentrations (48–104 mg l\(^{-1}\)) in our data set. This evidence suggests that in this case the underestimation of TIC by IC can be attributed to the interference of sulphate peak on the correct integration of carbonate peak at high sulphate concentration (≥50 mg l\(^{-1}\)).

3.4. Comparison with TIC measured by a TOC analyser

A limited set of samples, which included also two samples deliberately collected from Ticino river, has been analysed also by a TOC analyser, which provided an indirect measure of TIC, obtained as the difference between total carbon (TC), measured by direct combustion of the whole sample, and TOC, measured after sample acidification. Results showed that TIC measured by combustion technique (TICTC-TOC) is in perfect agreement with alkalinity data, while ion chromatography underestimated TIC concentrations in samples from Ticino river, confirming results shown in Section 3.3 for the same river but on different samples (Table 2).

3.5. An example of application of IC method in lake limnology

The knowledge of both alkalinity and total inorganic carbon allows to estimate the partial pressure of carbon dioxide in surface waters. Measure of this variable along the lake water column helps to understand the balance between photosynthesis and respiration by biota and carbon dioxide transfers from the water column to the atmosphere.

As an example of the usefulness of the determination of TIC by IC, we compared TIC and alkalinity concentrations measured along the water column of two deep sub-alpine lakes sited in Northern Italy (Lakes Mergozzo and Maggiore) (Fig. 5). In the case of these low alkalinity lakes, alkalinity is constant along the water column, while TIC showed significant variations. Till 10 m, the euphotic zone, the photosynthetic activity consumes carbon dioxide and the TIC concentrations tends to be equivalent to alkalinity which is equivalent to dissolved bicarbonate. At water depth <10 m respiration prevails and a net accumulation of dissolved CO\(_2\) can be measured by the difference between the two measurements.

![Fig. 5. Behaviour of TIC and alkalinity values vs. depth along the water columns of two Italian deep lakes: (a) Lake Mergozzo; (b) Lake Maggiore.](image)

3.6. Preliminary tests with a different stationary phase: Ion Pac AS19

In order to overcome the loss in accuracy verified at higher concentrations for IonPac AS 17 column, we tested an alternative stationary phase which can provide more efficiency and resolution for carbonate peak.

The column tested was the 2-mm IonPac AS19, which is a high capacity column optimised to separate anions and oxyhalides using hydroxide eluent (Fig. 6). This column allowed to improve peak efficiency and symmetry, providing a repeatability of 4.9% at a carbonate concentration of 0.075 mM and an estimated LOD of 0.014 mM by using an injection volume of 10 μl.

As in the case of AS17 column, the calibration is quadratic, showing a loss in response at higher concentrations which is related to a loss in efficiency and asymmetry of the carbonate peak.

We tested accuracy by correlation with certified bicarbonate concentrations of samples used in recent intercalibration tests on surface waters macroconstituents. They are mainly synthetic

<table>
<thead>
<tr>
<th>Sample</th>
<th>pH</th>
<th>(X_{\text{IC}}) (μS cm(^{-1}))</th>
<th>TIC(_{\text{IC}}) (mM)</th>
<th>TIC(_{\text{calc}}) (mM)</th>
<th>TAlk (mequiv l(^{-1}))</th>
<th>TIC (TC-TOC) (mM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bottled water</td>
<td>7.45</td>
<td>66.5</td>
<td>0.45</td>
<td>0.54</td>
<td>0.46</td>
<td>0.46</td>
</tr>
<tr>
<td>River Cannobino</td>
<td>7.16</td>
<td>37.4</td>
<td>0.19</td>
<td>0.22</td>
<td>0.19</td>
<td>0.23</td>
</tr>
<tr>
<td>River Ticino emissary</td>
<td>7.97</td>
<td>148.6</td>
<td>0.67</td>
<td>0.86</td>
<td>0.84</td>
<td>0.85</td>
</tr>
<tr>
<td>River Ticino tributary</td>
<td>7.70</td>
<td>194.8</td>
<td>0.57</td>
<td>0.79</td>
<td>0.76</td>
<td>0.77</td>
</tr>
</tbody>
</table>

Table 2

Summary of TIC results for representative samples obtained with different techniques
samples, which contain inorganic carbon only in bicarbonate form. The correlation \((n=10)\) is good \((R^2=0.992)\) with an angular coefficient very close to the unity and an intercept not significantly different \((p>0.01)\) from 0.

4. Conclusions

Ion chromatography showed to be a reliable technique for the determination of TIC in surface water samples during a 6 month sampling campaign. Correlation with TIC calculated from alkalinity and pH measurements was good over the whole explored range \((0.05-2\,\text{mM})\) but the most accurate results have been obtained in the lower range \((0.05\,\text{to}\,0.5\,\text{mM})\) due to loss of efficiency of carbonate peak at higher concentrations. Reduction of injection volume, which was 100\,\mu\text{l} in our experiments, is the most obvious solution to improve accuracy in the highest range of concentrations.

The good correlation with alkalinity data allows to use IC method also as a check or an alternative method for alkalinity in routine monitoring of surface waters without an additional analysis; in fact the TIC value could be obtained from the same chromatographic run used for the determination of the other main anions, using a much smaller volume than the titrimetric procedure. Furthermore, it can be easily implemented in an automated analyser for in situ applications.

If sampling have been carried out in order to avoid the equilibration with atmospheric CO\(_2\), IC method can be used as a method of detection of excess or deficiency of carbon dioxide partial pressures in natural waters, opening a new interesting field of research in limnology.

References