



A Gas Chromatographic Method for the Determination of Bicarbonate and Dissolved Gases

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A gas chromatographic method for the rapid determination of aqueous carbon dioxide and its speciation into solvated carbon dioxide and bicarbonate is presented. One-half mL samples are injected through a rubber septum into 20-mL vials that are filled with 9.5 mL of 0.1 N HCl. A one mL portion of the headspace is withdrawn and injected onto a gas chromatograph equipped with a thermal conductivity detector. Using the dimensionless Henry's constant for carbon dioxide and an adaptation of the Henderson-Hasselbalch equation, carbon dioxide in the samples can be categorized among solvated, bicarbonate, and carbonate forms. Natural water samples as well as wastewater from a municipal sewage treatment plant and a swine rearing operation were analyzed by this method and the results compared favorably to those obtained by titration. Samples stored for up to 5 weeks showed no significant changes in carbon dioxide concentrations. In addition, using flame ionization and electron capture detectors, methane and nitrous oxide concentrations in the samples were also measured.

Keywords: alkalinity, bicarbonate, carbon dioxide, dissolved gas, greenhouse gas, methane, nitrous oxide

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INTRODUCTION

Measuring alkalinity and hardness is important in gauging a waters buffering capacity and hence its ability to resist rapid changes in pH due to runoff, rain, and acid spills as well as its potential for scale formation in pipes, pumps, and boilers. Alkalinity is usually reported in bicarbonate equivalents, since calcium and magnesium bicarbonates are usually the most important buffering agents in nature (Boyd et al., 2016). Bicarbonate is also important since it represents the largest pool of carbon dioxide in most waters (Raymond et al., 2008). In karst waters in particular, wherein dissolved carbonate materials cause high concentrations of bicarbonate, the buffering capacity is increased and can have an influence on other water quality parameters and the precipitation of minerals that can impact infrastructure (Ford and Williams, 2007).

The most common means of measuring alkalinity and buffering is by titration of water samples with a weak or strong acid depending on the anticipated buffering capacity of the sample (APHA, AWWA, and WEF, 1998). This requires minimal equipment and gives acceptable results in most cases; however, when a significant portion of the alkalinity is due to other ions such as ammonium, hydroxide, silicates, borates, and phosphates, bicarbonate concentrations may be overestimated. The presence of interference from non-carbonate alkalinity may be assumed based on the shape of the titration curve, however in any case it is recommended that titration be performed with 48 h of sample collection and preferably within 24 h (APHA, AWWA, and WEF, 1998; United States Geological Survey, 2012).

In this paper, we present a rapid and convenient gas chromatographic (GC) method for the determination of bicarbonate buffering in water. It allows for the collection of numerous samples in the field and long term storage for subsequent laboratory analysis. Depending on the capability of the GC instrument other dissolved greenhouse gases may also be analyzed. Since carbon dioxide is measured directly, interference from other contributors to alkalinity is eliminated. We also discuss methods and means by which the accuracy of the technique could be enhanced.

MATERIALS AND METHODS

Sample Collection

Water and wastewater samples were collected from the Barren River in Warren County, KY, the Bowling Green Municipal Utilities wastewater treatment plant in Bowling Green, KY just after large solids screening and from deep pit storage at a local swine producer. For analysis of sample integrity over time, samples were collected from Meadow Creek in Taylor County, KY. The samples were kept in a cooler and transported back to the laboratory where samples were taken for gas chromatographic (GC) analyses.

One-half mL of water was withdrawn from the water samples using a disposable syringe equipped with an 18-gauge needle. The syringe needle was used to pierce the septum on a 20-mL headspace vial that was filled with 9.5 mL of 0.1 N HCl. The samples were then analyzed by gas chromatography as described below.

Samples were analyzed for alkalinity as titratable bicarbonate by the Hydroanalytical Lab at Western Kentucky University within 48 h of receipt using standard method 2320 B (APHA, AWWA, and WEF, 1998).

Gas Chromatographic Analysis

Methane, CO₂, and N₂O (nitrous oxide) were analyzed on a Varian Model CP-3800 (Agilent Technologies, Santa Clara, CA) GC modified for greenhouse gas (GHG) analysis by RSC Group LLC (Katy, TX). The GC was equipped with a model 1041 on-column injector operated at 75°C and 263 kPa which was connected to a 10-port gas-sampling valve and pressure-actuated solenoid valve. One-half mL of vial headspace was injected using a syringe temperature of 35°C and syringe flush time of 30 s with 250 µL of the sample transferred onto a 1.8 m by 1.6 cm o.d. column packed with 80/100 mesh Hay Sep Q (Varian Associates) with a He flow rate of 55 mL min⁻¹ for methane analysis. The column was connected to a thermal conductivity detector (TCD) operated at 120 °C and with a filament temperature of 200°C for CO₂ analysis. From the TCD, the sample went to a flame ionization detector operated under the following conditions: N₂ makeup gas 15 mL min⁻¹, H₂ 30 mL min⁻¹, air 300 mL min⁻¹, temperature 275°C.

Another portion of the sample was split by means of the gas switching valve to another 10 port gas sampling solenoid valve, and a portion of this sample was transferred to a 1.8 m by 1.6 cm o.d. Silicosteel column also packed with 80/100 mesh Hay Sep Q with a N₂ carrier flow rate of 55 mL min⁻¹ for N₂O analysis. This

column was connected to an electron capture detector (ECD) operated at 300°C.

The GC column oven was operated with an initial temperature of 50°C for 4 min. and then programmed at 50°C min⁻¹ to 100°C and held at this temperature for 1 min. Quantification was performed by five point calibrations from 96,160 to 1,000 µg L⁻¹ for CO₂ and 203,000 to 10,165 µg L⁻¹ for CH₄ in N₂ and one point calibration for N₂O of 2.0 µg L⁻¹ in N₂.

Dissolved GHG and other forms of CO₂ (HCO₃⁻, CO₃²⁻) were measured by collecting 0.5 mL water samples using a disposable syringe with 18-gauge needle. The syringe needle was used to pierce the septum on a 20-mL headspace vial that was filled with 9.5 mL of 0.1 N HCl. Gases were analyzed as described below.

Total CO₂ (solvated CO₂, HCO₃⁻, and CO₃²⁻) concentrations in water were calculated as:

$$\text{Total CO}_2, \text{ mM} = 20 * \frac{(0.8 * \text{Conc} + \text{Conc})}{1,000 \text{ ug mg}^{-1}} * \frac{1}{44.01 \text{ mg mmol}^{-1}} \quad (1)$$

Where 20 equals a multiplication factor to account for the dilution of the sample within the vial, 0.8 equals the dimensionless Henry's constant (K_H) for CO₂ and Conc equals the CO₂ concentration in the gas vial in µg L⁻¹. The sum of the HCO₃⁻ and CO₃²⁻ concentrations were determined by adaptation of the Henderson-Hasselbalch equation (Po and Senozan, 2001):

$$\Sigma \text{HCO}_3^-, \text{CO}_3^{2-}, \text{ mM} = \frac{[\text{Total CO}_2, \text{ mM}]}{1 + 10^{(\text{pH}-6.35)}} * 10^{(\text{pH}-6.35)} \quad (2)$$

Where pH equals the pH of the solution and 6.35 equals the *p*K_{a1} for H₂O + CO₂ ↔ HCO₃⁻ + OH⁻. Carbonate was calculated by the formula:

$$\text{CO}_3^{2-}, \text{ mM} = \frac{[\text{Total CO}_2, \text{ mM}]}{1 + 10^{(\text{pH}-10.33)}} * 10^{(\text{pH}-10.33)} \quad (3)$$

TABLE 1 | Alkalinity as HCO₃⁻ equivalents by gas chromatography and titration.

	Gas chromatography	Titration
	HCO ₃ ⁻ (millimolar) ^a	
BARREN RIVER		
Mean	3.45	1.91
Standard deviation	0.20	0.03
Relative standard deviation	5.8%	1.4%
MUNICIPAL SEWAGE		
Mean	6.23	4.53
Standard deviation	0.33	0.03
Relative standard deviation	5.2%	0.8%
DEEP PIT SWINE WASTE		
Mean	243	252
Standard deviation	13.1	1.22
Relative standard deviation	5.4%	0.48%

^aData represent the mean of six determinations.

with the variables the same as in (2) and substituting pk_{a2} of 10.33 for $\text{HCO}_3^- \leftrightarrow \text{CO}_3^{2-} + \text{H}^+$. Bicarbonate concentrations were calculated by subtracting CO_3^{2-} concentrations from those calculated in Equation (3) from the concentrations calculated in Equation (2). Solvated CO_2 concentrations were calculated by subtracting the values calculated from Equation (2) from those calculated in Equations (1) and (3). In our treatment of CO_2 , H_2CO_3 concentration was included along with that of hydrated CO_2 , since ascribing the partitioning between the species was beyond our means and also because the hydration constant (k_h) for CO_2 is low (1.2×10^{-3}), so that H_2CO_3 constitutes less than one percent of the two species combined (Archer, 1999).

Aqueous CH_4 and N_2O (dissolved and suspended gaseous) concentrations in the wastewater were calculated by Equation (1) using dimensionless Henry's constants of 27.02 and 1.1, respectively and molar masses of $16.04 \text{ mg mmol}^{-1}$ and $44.01 \text{ mg mmol}^{-1}$, respectively (Howard and Meylan, 1997; Jacinthe and Groffman, 2001).

RESULTS AND DISCUSSION

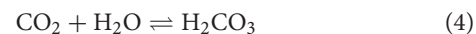
Table 1 presents a comparison of the results of alkalinity (as HCO_3^-) tests by GC and titration. In the river and municipal wastewater samples, analysis by GC estimated bicarbonate concentrations to be 81 and 38% higher respectively than did titration. For samples taken from a swine waste storage pit, however, GC results were 3.6% lower than those obtained by titration, possibly due to interferences with alkalinity measurements. In all cases, nonetheless, standard deviations obtained using titration were lower than those obtained by GC. Both analytical methods had acceptable relative standard deviations (RSD) for analytes in the 1,000 parts per million or less range (Taverniers et al., 2004).

We were limited by our equipment to using 20 mL headspace vials for analysis. Given the difficulty associated with measuring 0.5 mL water solids reproducibly, we feel sure that RSD could be improved by using vials and, hence, larger water aliquots. In addition, our autosampler was not equipped with a vial heater. Henry's constants vary with temperature so that constant vial temperatures could have resulted in lower RSD (DeWulf et al., 1995). Henry's constants also vary with ionic strength but given that samples were diluted 20-fold in the headspace vials, we feel this contributes very little to analysis variation.

Vial size also affects results, in that injecting a portion of the headspace shifts the equilibrium between the gases partitioned

in the gas and liquid phases. Injecting a smaller portion of the headspace onto the GC either by smaller sized injections or using larger vials could lead to more accurate results.

The complete analyses for dissolved gases in the environmental samples as determined by GC are presented as **Table 2**. In addition to bicarbonate, carbonate and solvated carbon dioxide ($s\text{CO}_2$) concentrations were calculated as well for dissolved methane and nitrous oxide. Due to hydrogen bonding, solvated carbon dioxide is surrounded by a "shell" of water molecules. Solvated carbon dioxide reacts with water to form carbonic acid:



While carbonic acid dissociates to a proton and bicarbonate:



The hydration constant (k_h) for $s\text{CO}_2$ to form H_2CO_3 in water is low (1.2×10^{-3} in salt water), so that H_2CO_3 concentrations were considered negligible and were not calculated (Soli and Byrne, 2002).

Solvated methane in the river samples were only about 2 mM (0.3 mg L^{-1}). As expected, they were much higher in municipal

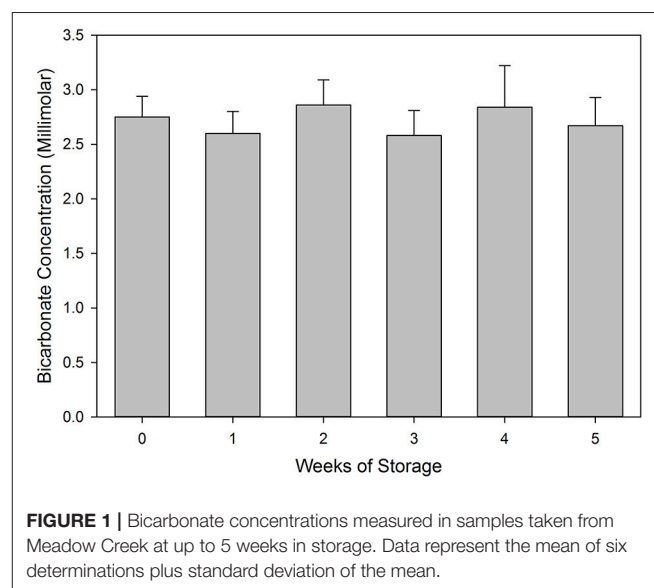


FIGURE 1 | Bicarbonate concentrations measured in samples taken from Meadow Creek at up to 5 weeks in storage. Data represent the mean of six determinations plus standard deviation of the mean.

TABLE 2 | Bicarbonate, carbonate and dissolved gas analyses.

Sample collection site	Measured analyte ^a				
	HCO_3^- (mM)	CO_3^{2-} (mM)	$s\text{CO}_2$ (mM)	CH_4 (mM)	N_2O (nM)
Barren river	3.45 ± 0.20	0.012 ± 0.001	0.11 ± 0.01	0.02 ± 0.003	0.17 ± 0.10
Municipal sewage	6.23 ± 0.33	0.010 ± 0.001	0.40 ± 0.02	0.43 ± 0.02	0.15 ± 0.03
Deep pit swine waste	243 ± 13.1	1.11 ± 0.06	5.59 ± 0.30	11.6 ± 0.79	0.04 ± 0.05

^aData represent the mean of six determinations \pm standard deviation of the mean.

water (0.43 mM) and especially swine waste (11.6 mM). In contrast, nitrous oxide concentrations were greatest in river and municipal sewage while it was virtually undetectable in swine waste. This was not unexpected given that nitrous oxide is not formed in anaerobic conditions in the absence of exogenous electron acceptors that sustain anaerobic respiration (Jetten et al., 1999).

No significant changes in measured bicarbonate concentrations were seen in samples over 5 weeks of storage (Figure 1). This is an advantage to titration techniques in that analysis is recommended within 48 h of sample collection and preferably with 24 h. In addition, the samples must be kept cool to minimize loss of CO₂ or growth of microorganisms that might affect results (APHA, AWWA, and WEF, 1998).

In conclusion, this method allows for the rapid measurement of dissolved gases and carbonate species in water. If measurement of only carbon dioxide and carbonates is desired, this method may be used with an inexpensive GC equipped

with only a TCD or FID equipped with a methanizer accessory.

AUTHOR CONTRIBUTIONS

All authors listed have made a substantial, direct and intellectual contribution to the work, and approved it for publication.

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