THE DETERMINATION OF TOTAL CARBON DIOXIDE IN WATER

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INTRODUCTION

Compared with oxygen the estimation of carbon dioxide in water presents much greater difficulties, and the methods published until now do not approach in sensitivity the Winkler method for dissolved oxygen by which the latter can be measured to 0.02 mg./l. in unpolluted and 0.2 mg./l. in polluted natural waters (Wheatland & Smith, 1955, Beadle, 1958). This is partly due to the high solubility and rate of diffusion of carbon dioxide in water which makes it difficult to avoid a significant change in concentration when sampling or during the process of estimation. Those methods which depend upon measurement of pH are particularly subject to errors arising from this factor, and the sensitivity of any measurement of pH is in any case a factor limiting accuracy. In polluted natural waters, as in swamps or water in which the respiration of organisms in low oxygen concentrations is being studied, the presence of acids other than carbonic may also introduce errors in methods relying on measurement of pH.

The method described in this paper was designed to measure the concentration of total carbon dioxide in the swamp waters of Uganda and to be used in conjunction with a constant flow respirometer where oxygen consumption and carbon dioxide production by aquatic organisms were measured simultaneously. In order to obtain meaningful results with the respirometer, it was found necessary to determine the total carbon dioxide concentrations of the effluent water samples to a degree of accuracy not readily achieved by any of the published methods (see Table 1 for comparative data). At the same time as these experiments were in progress, attention was directed to the carbon dioxide content of the waters which formed the natural habitat of the experimental material. This method has proved to be suitable, not only for determinations on these waters where the total concentration may be as high as 180 mg./l., but also for rain water where the range may be only 1–2 mg./l.

PRINCIPLE OF THE METHOD

All gases in solution are removed from an acidified sample by the vacuum extraction technique of van Slyke & Neill (1924) and are then transferred without contact with the atmosphere to a standard volume of sodium hydroxide solution with a known electrical resistance at a standard temperature. Within a given range of carbon dioxide content, the change in resistance of the absorbent after equilibration
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with the gas bears a strictly linear relationship to the total carbon dioxide content of the sample. The apparatus has been calibrated by using a series of standard test solutions of sodium carbonate.

The time required for a single determination is 15–20 min.

RANGE AND ACCURACY

The concentration of total carbon dioxide is expressed throughout in terms of mg. per litre but the actual quantity measured is obviously determined by the volume of the sample. The volume of 10 ml. which was eventually adopted is a compromise value of which (a) the upper limit is governed by a 1:10 relation between the sample volume to the volume of the extraction chamber to ensure that most of the dissolved gas will be extracted (van Slyke & Neill, 1924), and (b) the lower limit, below which there are significant errors in measuring the sample.

Thus the method has been used for quantities of total carbon dioxide within the ranges of 0.02–0.5 mg. and 0.5–2.0 mg./10 ml. sample, though it is necessary to use absorbent solutions of different strengths to obtain the maximum accuracy for each range. This is achieved in theory by adjusting the strength of the absorbent to equate with the highest value of carbonate concentration likely to be encountered. In practice, however, the strength of the absorbent is best adjusted to a fairly wide range of carbon dioxide values and we found the 0.005N solution very suitable for concentrations up to 50 mg./l.

A maximum variation of 2Ω was noted for the final resistance values of the absorbent from five replicate determinations on the same solution during a routine calibration of the apparatus for total carbon dioxide values up to 50 mg./l. Many subsequent determinations showed this variation to be a maximum, though it was often less, and a series of critical determinations, performed after more experience with the technique had been gained, showed that even more exact replication was possible and the performance of the bridge then became a limiting factor in the accuracy of the method. The final resistance of the absorbent was constant to 1Ω when replicate determinations were performed on a standard carbonate solution containing 36.0 mg./l. carbon dioxide and on solutions containing amounts as low as 5.2 mg./l. Ordinarily the coefficient of variation is 0.2% for any concentration within the 0–50 mg./l. range, but the percentage error is inversely proportional to the concentration. Greater accuracy could be achieved at the lower concentrations by reducing the strength of the absorbent which would in turn limit the range.

APPARATUS

The apparatus consists of the following main parts:

(a) Vacuum extraction unit (Fig. 1).
(b) Gas absorption unit for use with (a) (Fig. 2).
(c) Conductivity cell (Fig. 3).
(d) Sensitive a.c. bridge for resistance determinations of the absorbent.
Table I. Comparative data on methods for the determination of carbon dioxide in water

<table>
<thead>
<tr>
<th>Type of method</th>
<th>No.</th>
<th>Author and reference</th>
<th>Free or total CO₂</th>
<th>Sample size</th>
<th>Accuracy</th>
<th>Suitable range of concentration</th>
<th>Time required for one determination</th>
<th>Main sources of error</th>
</tr>
</thead>
<tbody>
<tr>
<td>Evolution of gas followed by absorption in standard alkali</td>
<td>1</td>
<td>Conway (1950)</td>
<td>Total</td>
<td>0.2 ml</td>
<td>16 ml.</td>
<td>0.5 % coefficient of variation</td>
<td>No. 1 unit down to 0.2 mg. No. 2 unit down to 0.04 mg.</td>
<td>50-60 min.</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>Partridge &amp; Schroeder (1932)</td>
<td>Total</td>
<td>100 ml</td>
<td>p.p.m.</td>
<td></td>
<td>Tested up to 87 p.p.m.</td>
<td>Presence of volatile acids other than CO₂; titration errors</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>Larsen (1949)</td>
<td>Total (Normally solid) up to 70 mg.</td>
<td>Known</td>
<td>Max. error</td>
<td>8.8 - 0.2 44.0 - 2.5 87.0 - 2.0</td>
<td>0.2-2.0 mg.</td>
<td>30 min.</td>
</tr>
<tr>
<td>Titration methods</td>
<td>4</td>
<td>McKinney &amp; Amorosi (1944)</td>
<td>Total</td>
<td>50 ml</td>
<td>p.p.m.</td>
<td></td>
<td>Tested up to 516.5 p.p.m. 20-25 min. less for a series</td>
<td>Exchange of free CO₂ with atmosphere; salt effect on indicators; presence of acids other than CO₂</td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>Moore (1939)</td>
<td>Free</td>
<td>25 ml</td>
<td>p.p.m.</td>
<td></td>
<td></td>
<td>Inaccurate pH measurement; loss of free CO₂ to atmosphere; presence of acids other than CO₂</td>
</tr>
<tr>
<td>Modified Winkler</td>
<td>6</td>
<td>Review by Partridge &amp; Schroeder (1932)</td>
<td>Total Variable</td>
<td>Low in presence of sulphate or hydroxide</td>
<td>Tested up to 86 mg./L</td>
<td></td>
<td>Exchange of free CO₂ with atmosphere; presence of sulphate or hydroxide; indicator adsorbed onto carbonate particles</td>
<td></td>
</tr>
<tr>
<td>Method</td>
<td>Source</td>
<td>Total Volume</td>
<td>Volume Tested</td>
<td>Measurement</td>
<td>Error Range</td>
<td>Time</td>
<td>Notes</td>
<td></td>
</tr>
<tr>
<td>--------------------------------------------</td>
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<td>-----------------------------------------------------------------------</td>
<td></td>
</tr>
<tr>
<td>Phenolphthalein-methyl orange method</td>
<td>Review by Partridge &amp; Schroeder (1932)</td>
<td>Total</td>
<td>0.5 ml.</td>
<td>Variable</td>
<td>Low in presence of hydroxide and other interfering subs.</td>
<td>Tested up to 86 mg/l.</td>
<td>—</td>
<td>Exchange of free CO₂ with atmosphere; interfering substances e.g. weak acids; hydroxide, sulphate</td>
</tr>
<tr>
<td>Vacuum extraction followed by:</td>
<td></td>
<td></td>
<td></td>
<td>Known</td>
<td>Max. error</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(a) Manometric measurement</td>
<td>van Slyke &amp; Neill (1924)</td>
<td>Total</td>
<td>0.5 ml.</td>
<td>Variable</td>
<td>6.0 ± 1.8 7</td>
<td>15-20 min.</td>
<td>Reading errors of the manometer; variable rate at which gas is reabsorbed by the sample</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>1.0 ml.</td>
<td>Known</td>
<td>46.1 ± 10.9</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>2.0 ml.</td>
<td>Max. error</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(b) Manometric measurement (modified van Slyke)</td>
<td>Oesting (1934)</td>
<td>Total</td>
<td>10 ml.</td>
<td>Known</td>
<td>45.36 ± 0.5</td>
<td>15-20 min.</td>
<td>Similar to (a)</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Max. error</td>
<td>45.98 ± 1.0</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>47.04 ± 0.1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(c) Volumetric measurement (modified van Slyke)</td>
<td>McLendon (1917)</td>
<td>Total</td>
<td>10 ml.</td>
<td>Known</td>
<td>137.4 ± 1.5</td>
<td>Tested c.</td>
<td>Variable rate of reabsorption in relation to salinity of sample; corrections necessary for temperature and barometric pressure</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Max. error</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(d) Conductivity measurement</td>
<td>Our method</td>
<td>Total</td>
<td>10 ml.</td>
<td>Known</td>
<td>0.3% coeff. var. or ±0.15 mg/l, up to 50 mg/l, total concentration</td>
<td>Tested from 4 to 50 and 50 to 200 mg/l.</td>
<td>15-20 min.</td>
<td>Handling of samples; error of sample measurement; variable rate of reabsorption; incomplete transfer of gas</td>
</tr>
</tbody>
</table>
(a) The gas extraction unit and sample pipette (Fig. 1)

This is basically similar to the apparatus of van Slyke & Neill, but there are some modifications. The size of the sample (10 ml.) is larger than that normally used, and therefore the dimensions of the extraction chamber have been increased accordingly to about 130 ml. The latter consists of a stout round-bottomed flask connected to the manometer and taps $T_4$ and $T_5$ by means of a ball-and-socket joint. The sample in the extractor chamber is stirred by a magnet ($F$) sealed into stout Pyrex tubing and actuated by a second rotating horseshoe magnet ($M$) placed near it.

Fig. 1. General layout of gas extraction apparatus with magnetic stirrer. The rotating magnet $M$ shown to the side of the extraction chamber is more conveniently placed behind it. The absorption unit is shown in position. $A$ is about 10 cm. long and 2 mm. bore. $T_1$, $T_2$, $T_3$ and $T_4$ are van Slyke stop cocks. For explanation see text.
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The sample is introduced into the extraction chamber through the inlet tube $A$ and is contained in that part of the extraction chamber above the upper graduation mark $B$. The lower graduation mark $C$ permitted a standard quantity of orthophosphoric acid to be drawn into the pipette $A$ and 2 ml. were adequate to displace the sample from $A$. Tap $T_9$ should be of the standard high-quality van Slyke type with a properly aligned plug, so as to allow complete transference of the gas (see 'Manipulation').

(b) Gas absorption unit (Fig. 2)

This consists essentially of a 20 ml. all glass syringe connected to a 3-way tap ($T_3$), of the same type and quality as $T_3$, with a piece of 1 mm. bore capillary tubing. The unit is connected to the extraction unit by a B 10 standard cone, the tip of which should be ground on the inside to reduce the thickness of the glass and thus to ensure that the gas is not trapped between the mercury and glass when the transfer is made (Fig. 2). The single way tap ($T_4$) provides an additional outlet which is used both when filling the unit with sodium hydroxide solution and when transferring it to the conductivity cell after equilibration with the gas.

(c) Conductivity cell (Fig. 3)

This is of standard design and the dimensions are not critical (Jones & Bollinger, 1931). For maximum sensitivity, however, the plates must not be too large or too close together. Our home-made cell has circular platinum foil plates cut out with a no. 2 cork borer and placed 22 mm. apart. These plates, which are finally blackened (Jones & Bollinger, 1935), are sealed rigidly on to the ends of the side arms. Platinum wires connect the plates through mercury and platinum connexions to the bridge.
The cell was closed, excluding air bubbles, with a B10 stopper to prevent contamination of the absorbent by atmospheric carbon dioxide. The constancy of the cell was checked with a standard sodium chloride solution.

\[(d)\] A.C. bridge for resistance determinations

All resistance determinations were made at 30°C when the cell and contents had equilibrated to this temperature in a water-bath. The temperature was constant to ±0.01°C.

The a.c. bridge was of standard design, comprising non-inductively wound ratio arms and variable resistance to 10,000Ω, Mullard oscillator (1000 c.p.s.) and headphones, and incorporated variable condensers (0.005 mF) symmetrically placed in the circuit opposite the cell. These latter were adjusted to compensate for the capacitance of the cell, the value of which varies with the resistance of the solution. By this means it is possible to obtain a sharp end point.

**ABSORBENT**

For the range of total carbon dioxide concentration usually required (0.50 mg./l.), the most convenient absorbent was found to be 0.005 N (approx.) sodium hydroxide solution. This was prepared by diluting a clear saturated solution of the alkali with relatively carbon-dioxide-free distilled water.\* An approximately normal solution was first prepared by diluting 55 ml. to 1 l. and 50 ml. of this stock solution were then diluted to 10 l. with carbon-dioxide-free distilled water prepared by aeration for several hours. The final strength of the solution is about 0.005 N, with a carbonate content of 1 mg./l., and the resistance measured by our cell was approximately 2730Ω. With a little practice it is quite possible to prepare successive stocks of absorbent which will have practically the same resistance value. The minor variations between different batches of the absorbent, which affect the accuracy of the results, can be adjusted by the method given below without the frequent necessity of re-calibrating the apparatus.

The absorbent was protected from contamination by atmospheric carbon dioxide by a caustic soda trap and mercury-to-glass seals. In spite of these precautions, however, the resistance of the solution increased by about 3Ω per day.\† Since this drift will cause significant errors if corrections are not applied, the resistance of the absorbent was measured before each series of determinations and the increase over the value of the original calibration was noted. This increase was then subtracted from the final values obtained for each determination and fixed base-line was thus retained. The correctness of this method was tested by varying the resistance of the absorbent by adding water or sodium hydroxide solution and subsequently performing a series of determinations on two standard solutions.

\* Prepared by aerating conductivity water for several hours. Complete removal of the carbon dioxide by this means is not possible and water treated in this way contained 0.8 mg./l. (Calculated value is 0.3 mg./l. at 24°C and 660 mm. barometric pressure.)

\† The carbonate concentration was constant over a period of 14 days, but the resistance increased by 41Ω, presumably due to a reaction between the alkali and glass container.
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carbonate solutions with a total carbon dioxide content of 29.0 and 22.8 mg./l. Table 2 shows that the determinations agree within the limits of the method, irrespective of the variations in resistance of the absorbent solution.

The strength of the absorbent solution was increased when the samples contained more than 50 mg./l. and a 0.02 N solution was found to be satisfactory for values up to 200 mg./l. The resistance of this solution was 730Ω in our cell and the resistance determination then becomes a factor limiting the accuracy of the method. By using a fractional Ω box, replicate determinations on solutions containing 180 mg./l. total carbon dioxide differed by no more than 1 mg./l. The errors incurred in handling samples such as this which contain a very high concentration of free carbon dioxide may, in fact, mask the errors of measurement.

Table 2. Carbon dioxide content of two standard carbonate solutions determined by using absorbent solutions of differing strengths
(Strength expressed as resistance measured at 30 °C.)

<table>
<thead>
<tr>
<th>CO₂ content (mg./l.)</th>
<th>CO₂ content (mg./l.)</th>
</tr>
</thead>
<tbody>
<tr>
<td>test solution (prepared)</td>
<td>Initial</td>
</tr>
<tr>
<td></td>
<td>R₅₀ absorbent</td>
</tr>
<tr>
<td>A 29.0</td>
<td>2724</td>
</tr>
<tr>
<td>29.0</td>
<td>2767</td>
</tr>
<tr>
<td>29.0</td>
<td>2762</td>
</tr>
<tr>
<td>B 22.8</td>
<td>2797</td>
</tr>
<tr>
<td>22.8</td>
<td>2790</td>
</tr>
<tr>
<td>22.8</td>
<td>2782</td>
</tr>
<tr>
<td>22.8</td>
<td>2772</td>
</tr>
<tr>
<td>22.8</td>
<td>2764</td>
</tr>
</tbody>
</table>

When there is no previous indication of the range of carbon dioxide concentration in a series of samples, it is advisable to make a preliminary measurement using a relatively strong solution of sodium hydroxide (e.g. 0.02 N). If there is less than 50 mg./l. carbon dioxide and greater accuracy is required, the weaker (0.005 N) solution can then be used.

During preliminary experiments barium hydroxide solution was used as the absorbent, but it was rejected on finding that the carbonate precipitate caused erroneous resistance values, presumably by settling on the plates of the cell. Thomas (1933) does not appear to have experienced this trouble, but we found that stable readings could be obtained only when the precipitate was filtered off under carbon-dioxide-free conditions. This was abandoned for the greater convenience of sodium hydroxide.

MANIPULATION

Cleaning procedure

(a) Extraction unit. Traces of sample remaining from the previous determination can be removed by the following procedure:

1. Close control cock (T₅) and open T₃ to J.
2. Raise mercury reservoir to station 1.
3. Open T₅ slowly until the mercury just enters J. Close T₅.
Remove liquid from the walls of \( J \) and the surface of the mercury by means of a small plug of cotton-wool twisted round a spill.

(b) Absorption unit. Mercury is removed from the bore of \( T_2 \) and the whole unit is washed with conductivity water, rinsed with acetone and dried with a jet of compressed air.

**Assembly**

The absorption unit is filled with the solution by closing \( T_2 \) and flooding the syringe by means of a plastic tube connecting \( T_1 \) to the aspirator. The plunger is then inserted for a short distance while the liquid is flowing and \( T_1 \) is then closed. The volume of the absorbent is then adjusted to a standard but undetermined value (20 ml. approx.) by means of a ‘stop’ on the plunger with \( T_1 \) in the open position. The absorbent will not flow into the capillary connexion between \( T_2 \) and the syringe. This avoids the risks of contamination when the transfer of gas is made from the extraction chamber to the unit.

With the foregoing technique it is possible to transfer the absorbent from the aspirator to the unit without contamination by atmospheric carbon dioxide.

The cone at the base of the unit is greased slightly and inserted firmly into \( J \). Springs are attached. With \( T_2 \) open to \( Z \) the whole of the space below \( T_2 \) is filled with mercury by manipulation of \( T_2, T_3, \) and \( T_b \), with the mercury reservoir at station 1. The sample inlet tube (A) is filled to the tip by reversal of \( T_3 \), when the mercury flow is controlled by \( T_b \).

The sample is now introduced, acidified, the gases removed by vacuum extraction and these are transferred to the absorption unit. The details of the manipulation are given below.

**Introduction of sample and acid**

1. With control cocks \( T_b \) and \( T_2 \) closed and \( T_3 \) open to \( A \)—place the mercury reservoir at station 2.

2. Insert \( A \) into sample bottle (30 ml. capacity) so that the tip reaches about half way down.

3. Partially open control cock \( T_5 \) gently and draw in the sample until the mercury meniscus reaches the graduation mark \( B \) (10 ml.).

4. Close \( T_5 \)—remove bottle and wipe off excess sample from \( A \).

5. Place \( A \) into a small bottle of undiluted orthophosphoric acid* and open \( T_b \). Draw in 2 ml. to acidify and displace the sample from the capillary and bore of the cock. Close \( T_3 \) quickly to avoid mixing of the sample with the acid in \( A \). The pH of the mixture is about 1·2.

Operations 2 and 3 must be performed with no delay once the sample has been opened, especially when it contains free carbon dioxide.

* The error due to interfering substances in the orthophosphoric acid is very small, being equivalent to about +0·05 mg./l. carbon dioxide, but the same amount of undiluted lactic (used as a 1N solution in the usual van Slyke procedure) can cause errors up to +3·5 mg./l., presumably due to the presence of a volatile acid.
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Extraction procedure

1. Lower the mercury reservoir to station 3 and, when the liquid meniscus level has reached the region of B, open $T_3$ to $J$ and clear it of mercury.

2. Close $T_5$ when the mercury and sample reach a fixed point ($D$) at the base of the extraction chamber (Fig. 1). Extract for 5 min. with continuous stirring.

Transfer of gas

1. Raise the mercury reservoir to station 1, and partially open the control cock ($T_6$) so that the mercury and sample rise, rapidly but without turbulence. Control the rate at which the sample rises by manipulation of $T_6$ and decrease the rate as soon as the liquid meniscus enters the narrow tube below $T_8$. Close $T_3$ quickly when liquid is seen just about to enter the bore of the cock. The gas is now confined in $J$ under negative pressure.

2. Open $T_3$ to $A$ and clear the pipette of liquid.

3. Partially reverse $T_3$ and allow mercury to enter $J$ steadily. A small volume of liquid may also enter but will be dispersed rapidly over the relatively dry walls of $J$. The surface of the mercury will eventually appear to be dry.

4. Partially open $T_2$ to the syringe and transfer the gas. The mercury will finally fill the bore of the cock, thus clearing the gas, but will not pass into the capillary tube above. This operation is made easier if the upper extremity of the bore faces the operator. Close $T_2$.

5. Lower the reservoir to station 2 and open $T_2$ to the outside.

Resistance determination

1. Detach the absorption unit and shake thoroughly to ensure complete absorption of the gas and to homogenize the contents.

2. Clamp the unit on its side with $T_1$ facing downwards and flood out the conductivity cell by forcing the absorbent out steadily through $T_1$ and a plastic tube connexion, by means of the plunger.

3. Stopper the cell quickly (dry stopper) and determine the resistance of the solution after equilibration in a water-bath at 30°C, indicated by two constant readings—about 5 min.

CALIBRATION

A series of twelve standard sodium carbonate solutions with carbon dioxide contents ranging from 9·6 to 124·0 mg./l., was prepared from the A.R. solid (preheated at 270°C. overnight) and relatively carbon-dioxide-free distilled water prepared by aeration for several hours. The total carbon dioxide content of this distilled water was 0·8 mg./l. and this amount is included in the total value for each standard solution.
the sample and the resistance of the sodium hydroxide solution for values up to 52 mg./l. carbon dioxide. The slope changes when the total carbon dioxide content of the sample exceeds this value. A possible explanation is that the two slopes reflect the two equilibrium systems, (1) $\text{-OH}^-/\text{-CO}_3^-$ and (2) $\text{-CO}_3^-/\text{-HCO}_3^-$. The carbon dioxide concentrations produced in our respirometer did not exceed 50 mg./l. and thus the lower portion of the curve could be used.

Large-scale graphs can be constructed from measurements on standard sodium carbonate solutions to cover any desired range using the appropriate sodium hydroxide solution. In practice, however, we have found that 0.005N-sodium hydroxide for carbon dioxide concentrations up to 50 mg./l. and 0.02N for those up to 200 mg./l. were suitable for our particular purposes in respirometry and the study of natural swamp waters. The errors introduced through subsequent small variations of the resistance of the absorbent can be corrected without re-calibration, providing that the conductivity cell constant does not change (see ‘Absorbent’). The initial calibration described above has established the linear relationship

![Graph showing the relation between total carbon dioxide content of standard carbonate solutions and the final resistance of the absorbent (0.005N sodium hydroxide solution) at 30$^\circ$C.](image-url)
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between resistance of the absorbent and the carbon dioxide content of the sample. Three standard carbonate solutions are therefore in fact sufficient to construct the graph.

SOURCES OF ERROR

There are three main sources of error: (A) those incurred by incorrect handling of the samples and absorbent, (B) manipulative errors of the actual determination, and (C) interfering substances.

(A) Errors in handling of samples and absorbent

(1) Samples

Significant errors can be introduced if samples and the absorbent are handled in such a way as to permit exchange of carbon dioxide with the atmosphere. This is particularly liable to occur where weakly buffered samples contain a relatively high concentration of the 'free' gas. Such solutions should be stored under water in glass bottles with well-fitted glass stoppers. The transfer of samples from one type of receiver to another should always be done by siphoning slowly with the tube placed near the bottom of each. The receiving vessel should be flooded out with a large excess of liquid.

(2) Absorbent

(i) The weak solution of sodium hydroxide is easily contaminated by atmospheric carbon dioxide, and therefore the absorption unit must be filled by the flooding technique outlined in 'Manipulation'. The well-fitting plunger of these syringes excludes atmospheric carbon dioxide, at least during the period required to complete a determination.

(ii) Similar considerations apply to the transference of the absorbent from the unit to the conductivity cell. The 20 ml. capacity of the unit is adequate to flood out the cell previously filled with distilled water. Errors were introduced when the volume of the absorption unit was reduced to 10 ml.

(iii) The stock absorbent should be agitated before use and it is advisable to run 10–20 ml. to waste before a determination is started, particularly if it has been standing unused for several days.

(iv) The carbon dioxide is absorbed rapidly by the solution in the unit but the contents must be equilibrated. 'Dead spots' near the taps may be cleared by manipulation of the gas bubbles.

(B) Errors in manipulation

(1) Sample volume

Significant variation may be caused by the presence of residual sample from previous determinations lining the walls of the pipette and related parts, as a thin film. If the glassware is relatively free from grease and the sample is cleared slowly, then liquid is rarely trapped between the mercury and the glass.
(2) Reabsorption of gas after extraction

The manometric method of van Slyke & Neill (1924) (van Slyke & Sendroy, 1927) uses tables of factors which compensate for the reabsorption of carbon dioxide by the samples during the period after extraction when the mercury is adjusted to the datum line. Accurate results therefore depend on careful timing and adjustment of the mercury level. We have found that a significant error due to this factor is eliminated, if the operations of transferring the gas to J and later to the absorbent are done within about 1½ min. An error of about —0-6 mg./l. was introduced when the time was extended to 4 min.

It might be expected that the film of liquid left on the walls of J during the extraction procedure (stage 1) would reabsorb some carbon dioxide when the pressure is increased, but (a) the pH of this residual liquid is very low (1-2), being derived from a previous acidified sample, and (b) the volume is extremely small, and (c) the gas remains in contact only for a few seconds. The results from standard solutions show that there is in fact no significant reabsorption at this stage.

(3) The introduction of mercury into the absorption chamber

This will rarely occur if T₂ is only partially opened during the transfer of gas. The mercury will be contaminated with the contents of the extraction chamber and will thus cause faulty resistance values.

(C) Interfering substances

Non-volatile substances such as phosphate and hydroxide which interfere with titration methods do not affect the accuracy of this method but volatile acidic substances such as hydrogen sulphide will be transferred with the carbon dioxide and be taken up by the absorbent. The ‘apparent’ total carbon dioxide would then be in excess of the real value.

Our swamp water samples have so far contained no significant amount of sulphide, but if such were ever the case it could perhaps be removed by the insertion of a crystal of soluble mercury or cadmium salt into the original sample on collection. The sulphide precipitate must then settle before a sample is taken off into the apparatus. Methane, which is always present in swamp waters, does not of course interfere with the method.

DISCUSSION

Table 1 reviews the various methods used for the determination of total carbon dioxide and one method (Moore, 1939) for ‘free’ (dissolved) carbon dioxide. They vary greatly with respect to the volume of sample required, the range of concentration for which they were designed, and the accuracy. Since our method has been designed primarily for use with natural waters where the amount of sample is rarely limited, the elegant method of Conway (1950) will not be considered as it was designed for use with small volumes of sample containing relatively high concentrations of carbon dioxide.
Determination of total carbon dioxide in water

Titration methods (Table 1; 4, 5, 6 and 7) offer certain advantages in that little specialized equipment is required, but they rarely achieve a desirable degree of accuracy due to the exchange of carbon dioxide with the atmosphere, particularly when the sample is weakly buffered and interfering substances such as hydroxide, phosphate and acids other than carbon dioxide are present. The method of McKinney & Amorosi (1944) corrects for the presence of most interfering substances other than volatile acids, but the titration is performed in open vessels with the consequent loss of the carbon dioxide.

Our method differs from others which use the vacuum extraction technique in that the carbon dioxide content of the sample is related to the resistance changes of a standard solution of sodium hydroxide after equilibration with the extracted gas. Thus the temperature and barometric corrections needed for the volumetric type of van Slyke apparatus (McClendon, 1917) are not required. In addition, the volumetric and manometric methods both require corrections to compensate for the amount of gas which remains unextracted in the sample and for that which is reabsorbed by the sample during the interval between completing the extraction and noting the pressure or volume.

Corrections for both these factors are not required in our method. In the first place the proportion of unextracted gas remaining in the liquid is a constant (van Slyke & Neill, 1924) provided that the volume of the gas phase determined by the mercury level and that of the sample and acid are both constant. Secondly, significant reabsorption did not occur in our experiments when the extracted gas was transferred to the absorbent as rapidly as possible. This is presumably due to the transfer being made to 12 cm of Hg after which the final stages of transfer to the absorbent were made with the gas in contact with relatively dry mercury. This method differs, therefore, from the method of van Slyke where the extracted gas in contact with the sample is often subjected to considerable positive pressure resulting in reabsorption of about 1.4-1.7% of the carbon dioxide.

Since our calibration procedure gives a good linear relationship between the carbon dioxide content of standard solutions and the final resistance of the absorbent, it is justifiable to neglect the corrections for the unextracted portion of the gas and the subsequent reabsorption, provided that the standard procedure is always followed.

APPLICATION OF THE METHOD

Determination of (1) total, (2) carbonate CO\textsubscript{2} and (3) dissolved and bicarbonate CO\textsubscript{2} in natural waters.

Samples were collected in 200 ml. glass-stoppered bottles, with a minimum of disturbance from just below the surface of a stagnant pool in a papyrus swamp. Fig. 5 shows the sampling technique. The water is drawn through a series of bottles by suction from a hand-pump designed for use with a Hearson vacuum-embedding oven. To avoid reducing the pressure to the point at which dissolved gases are extracted and appear as bubbles, the drain vessel must be placed at as low a level...
as possible and pumping must not be too vigorous. The bottles are immediately stoppered, submerged in water and the total carbon dioxide determination is carried out within 30 min. after collection.

The dissolved and bicarbonate CO$_2$ was removed by vigorous aeration of the sample and the residual carbonate CO$_2$ was then determined. The dissolved and bicarbonate CO$_2$ was obtained by difference. Parallel determinations of the 'free' carbon dioxide (dissolved) concentrations were carried out by the method of Moore (1939) which involves the measurement of pH by glass electrode and of alkalinity by titration with standard acid (see Table 3). The samples were taken from the same swamp on three successive dates and show the progressive effect of flooding during a rainy period after drought. It will be noted that the initial concentration of dissolved and bicarbonate CO$_2$ was extremely high and that Moore's method gave a higher value for dissolved carbon dioxide. We would attribute this to the

![Fig. 5. Method for obtaining samples from a swamp pool. A, cork float with tube and glass wool filter. Bottles for: (1) total CO$_2$; (2) carbonate CO$_2$; (3) oxygen; (4) drainage.](image)

Table 3. Carbon dioxide content of water samples from a papyrus swamp, determined by our method and Moore's method (Moore, 1939)

<table>
<thead>
<tr>
<th>Date</th>
<th>Our method</th>
<th>Moore's method</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(A) mg./l. dissolved and bicarbonate CO$_2$ (by difference)</td>
<td>(B) mg./l. 'free' CO$_2$ (dissolved)</td>
</tr>
<tr>
<td></td>
<td>mg./l. total CO$_2$</td>
<td>mg./l. carbonate CO$_2$</td>
</tr>
<tr>
<td>1 24. vii. 59</td>
<td>180</td>
<td>32</td>
</tr>
<tr>
<td>2 21. viii. 59</td>
<td>139</td>
<td>33</td>
</tr>
<tr>
<td>3 27. viii. 59</td>
<td>71</td>
<td>36</td>
</tr>
</tbody>
</table>
Determination of total carbon dioxide in water

presence of non-volatile acids other than carbonic. In one lakeside swamp we found 9.5 mg./l. dissolved and bicarbonate CO₂ by this and 60 mg./l. dissolved CO₂ ('free') by Moore's method. It must, however, be realized that an error of 0.05 in determining the pH is equivalent to 10–15 mg./l. 'free' carbon dioxide with water of this order of alkalinity and pH.

The figures in Table 3 are only quoted to illustrate the use of the method, and cannot be taken as accurately representing the two fractions of these particular samples. It will be seen that the apparent carbonate CO₂ rises slightly in spite of the lower alkalinity values for samples 2 and 3. This was no doubt due to shortening the time of aeration from about 22 hr. (sample 1) to 3–5 hr. for samples 2 and 3. It is clear that the dissolved and bicarbonate CO₂ would be more effectively removed by boiling.

SUMMARY

1. The method described involves extraction of the dissolved gas from an acidified sample in a modified van Slyke apparatus, and measurement of the electrical resistance of a standard sodium hydroxide solution before and after equilibration with the extracted gas.

2. The original sample must be not less than about 30 ml. for which a 10 ml. sample is drawn into the extraction apparatus.

3. There is a linear relation between the resistance finally measured and the total carbon dioxide content of the original sample up to a maximum determined by the strength of the standard hydroxide solution, which is thus adjusted to the desired range of carbon dioxide concentration, e.g. 0.005N-NaOH for 0–50 mg./l. and 0.02N for 0–200 mg./l. CO₂.

4. The method is easily sensitive to 0.3 mg./l. in the range 0–50 mg./l. total carbon dioxide and about 1 mg./l. in the range 0–200 mg./l. With a rigidly standardized procedure increased accuracy was obtained so that replicate determinations on standard solutions in the 0–50 mg./l. range did not differ by more than 0.16 mg./l.

5. Precautions for handling samples containing carbon dioxide in solution prior to determination have been suggested.

6. About 15–20 min. are required for each determination and to Mrs D. E.

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REFERENCES


