SOP 6b

Determination of the pH of sea water using the indicator dye *m*-cresol purple

1. Scope and field of application

This procedure describes a method for the spectrophotometric determination of the pH of sea water on the total hydrogen ion concentration pH scale. The total hydrogen ion concentration, $[H^+]$, is expressed as moles per kilogram of sea water.

2. Definition

The total hydrogen ion concentration of sea water includes the contribution of the medium ion sulfate and is defined as

$$\begin{aligned}[H^+] &= [H^+]_F(1 + S_T / K_S) \\ &\approx [H^+]_F + [HSO_4^-] \end{aligned} \quad (1)$$

where $[H^+]_F$ is the *free* concentration of hydrogen ion in sea water, S_T is the total sulfate concentration $([HSO_4^-] + [SO_4^{2-}])$ and K_S is the acid dissociation constant for HSO_4^-. The pH is then defined as the negative of the base 10 logarithm of the hydrogen ion concentration:

$$\mathrm{pH} = -\log_{10}\left(\frac{[H^+]}{\text{mol kg-soln}^{-1}}\right). \quad (2)$$

3. Principle

The values of pH are determined by adding an indicator dye to sea water. For the sulfonephthalein indicators such as *m*-cresol purple, the reaction of interest at sea water pH is the second dissociation

$$HI^-(aq) = H^+(aq) + I^{2-}(aq) \quad (3)$$

where I represents the indicator dye, which is present at a low level in a sea water sample. The total hydrogen ion concentration of the sample can then be determined:

$$\mathrm{pH} = \mathrm{p}K(\mathrm{HI}^-) + \log_{10}\frac{[\mathrm{I}^{2-}]}{[\mathrm{HI}^-]}. \quad (4)$$

The principle of this approach uses the fact that the different forms of the indicator have substantially different absorption spectra. Thus the information contained in the composite spectrum can be used to estimate $[I^{2-}]/[HI^-]$.

At an individual wavelength, λ, the measured absorbance in a cell with a path length, l, is given by the Beer–Lambert law as

$$\frac{A_\lambda}{l} = \varepsilon_\lambda(\mathrm{HI}^-)[\mathrm{HI}^-] + \varepsilon_\lambda(\mathrm{I}^{2-})[\mathrm{I}^{2-}] + B_\lambda + e \quad (5)$$

where B_λ corresponds to the background absorbance of the sample and e is an error term due to instrumental noise. Provided that the values of the extinction coefficients: $\varepsilon_\lambda(HI^-)$ and $\varepsilon_\lambda(HI^{2-})$ have been measured as a function of wavelength, absorbance measurements made at two or more wavelengths can be used to estimate the ratio $[I^{2-}]/[HI^-]$.

In the case that only two wavelengths are used, and provided that the background can be eliminated effectively by a subtractive procedure, (5) can be rearranged to give (assuming no instrumental error)

$$\frac{[\mathrm{I}^{2-}]}{[\mathrm{HI}^-]} = \frac{A_1 / A_2 - \varepsilon_1(\mathrm{HI}^-)/\varepsilon_2(\mathrm{HI}^-)}{\varepsilon_1(\mathrm{I}^{2-})/\varepsilon_2(\mathrm{HI}^-) - (A_1 / A_2)\varepsilon_2(\mathrm{I}^{2-})/\varepsilon_2(\mathrm{HI}^-)} \quad (6)$$

where the numbers 1 and 2 refer to the wavelengths chosen. For the best sensitivity, the wavelengths corresponding to the absorbance maxima of the base (I^{2-}) and acid (HI^-) forms, respectively, are used. The various terms ε are the extinction coefficients of the specified species at wavelengths 1 and 2, respectively.

4. Apparatus

4.1 Flexible drawing tube

Approximately 40 cm long, sized to fit snugly over cell port. Silicone rubber is suitable for this (see Footnote 1 in SOP 1).

4.2 Spectrophotometric cells

These should be made of optical glass with a 10 cm path-length, two ports and polytetrafluoroethylene (Teflon®) stoppers. A sufficient number of cells are needed to collect all the samples that will be analyzed from a particular cast (see section 6).

4.3 Micropipette

A micropipette is used to add the dye to the cell. It should be of ~ 0.1 cm^3 capacity with a narrow Teflon® tube attached to act as a nozzle.

4.4 High-quality spectrophotometer

For work of the highest sensitivity and precision, a double-beam spectrophotometer is desirable. However, good results can be obtained with a high-quality single-beam instrument.

4.5 Temperature-control system for spectrophotometer cell

Commercially manufactured, thermostated spectrophotometer compartments that can accommodate 10 cm cells are rarely available and one will probably have to be custom-made. The temperature should be regulated to within 0.1°C.

4.6 System to warm samples to measurement temperature

Although it is possible to warm up the cells containing samples in Ziploc® bags in a thermostat bath, this is inconvenient. It is much better to build a custom-made thermostated compartment that can hold approximately 12 cells at once without getting them wet.

4.7 Thermostat bath (± 0.05°C)

A thermostat bath is used to regulate the temperature of the cell compartment and the temperature of the system described in section 4.6.

5. Reagents

5.1 Solution of *m*-cresol purple

A concentrated (at least 2 mmol dm^{-3}) dye solution of known pH adjusted to be in the range 7.9 ± 0.1 pH units—chosen to match pH measurements from an oceanic profile—is required; this implies that for *m*-cresol purple $A_1/A_2 \approx 1.6$.[1]

6. Sampling

Draw the sample—using the drawing tube—directly from the Niskin bottle (or other water sampler) into the optical cell. After flushing with several hundred cm^3 of sea water—a flushing time of 15–20 seconds—seal the cell with the Teflon® caps ensuring that there is no headspace. Since the pH samples must be analyzed immediately, there is no long-term storage or preservation protocol. However, while awaiting analysis, store the samples in the dark at room temperature.

[1] The absorbance ratio of a concentrated dye solution can be measured using a cell with a short path length (0.5 mm).

7. Procedure

7.1 Warm sample cell to 25.0°C (± 0.1°C)

This is done by placing a number of cells in a thermostated compartment (see section 4.6) for a few hours.

7.2 Measure absorbances for the cell + sea water

Clean and dry the exterior of the cell; place the cell in the thermostated sample compartment of the spectrophotometer. Measure and record the absorbances at three wavelengths: a non-absorbing wavelength (730 nm for *m*-cresol purple) and at the wavelengths corresponding to the absorption maxima of the base (I^{2-}) and acid (HI^{-}) forms of the dye respectively (578 and 434 nm).

7.3 Inject dye into cell

Remove one of the cell caps, add approximately 0.05–0.1 cm^3 of concentrated dye (~2 mmol dm^{-3}) to the sample, replace the cap and shake the cell to mix the sea water and dye. The amount of dye required is that which will produce absorbance values of between 0.4 and 1.0 at each of the two absorbance peaks.

7.4 Measure absorbances of cell + sea water + dye

Return the cell to the spectrophotometer and again measure the absorbances at the three wavelengths used in section 7.2. Cells should be positioned to maintain consistent alignment(s) between baseline and indicator absorbance measurements.

8. Calculation and expression of results

8.1 Correction of measured absorbances

At each of the three wavelengths, subtract the absorbances measured for the background measurement (without dye) from the corresponding absorbances measured for the system containing dye.

In addition, the absorbance measured at a non-absorbing wavelength is used to monitor and correct for any baseline shift due to error in repositioning the cell, instrumental shifts, *etc.*[2]. This assumes that the magnitude of any observed baseline shift is identical across the visible spectrum. To do this, subtract the measured shift from the background-corrected absorbances at wavelengths 1 and 2 to obtain the final corrected absorbance value at each wavelength.

These final absorbance values, corrected for background absorbances *and* any observed baseline shifts, are used to calculate A_1/A_2, the absorbance ratio which describes the extent of protonation of the dye.

[2] The difference between the baseline absorbance (sea water only) and the absorbance of the sample + dye at 730 nm should be no greater than ± 0.001; if this value is exceeded, the cell should be removed and the optical windows cleaned before the absorbances are measured again.

8.2 Calculation of the pH of the sea water + dye

The pH of the sea water and dye in the cell is computed from

$$\mathrm{pH} = \mathrm{p}K_2 + \log_{10}\left(\frac{A_1 / A_2 - \varepsilon_1(\mathrm{HI}^-)/\varepsilon_2(\mathrm{HI}^-)}{\varepsilon_1(\mathrm{I}^{2-})/\varepsilon_2(\mathrm{HI}^-) - (A_1 / A_2)\varepsilon_2(\mathrm{I}^{2-})/\varepsilon_2(\mathrm{HI}^-)} \right) \qquad (7)$$

where pK_2 is the acid dissociation constant for the species HI^- (expressed on the total hydrogen ion concentration scale in mol kg-soln^{-1}), and A_1 and A_2 are the corrected absorbances measured at the wavelengths corresponding to the absorbance maxima of the base and acid forms, respectively. The various extinction coefficient terms ε correspond to values measured for the specified species at wavelengths 1 and 2, respectively (Table 1).

Table 1 Extinction coefficient ratios for *m*-cresol purple.

$\varepsilon_1(\mathrm{HI}^-)/\varepsilon_2(\mathrm{HI}^-)$	0.00691
$\varepsilon_1(\mathrm{I}^{2-})/\varepsilon_2(\mathrm{HI}^-)$	2.2220
$\varepsilon_2(\mathrm{I}^{2-})/\varepsilon_2(\mathrm{HI}^-)$	0.1331

$\lambda_1 = 578$ nm; $\lambda_2 = 434$ nm.

The equilibrium constant K_2 is a function of salinity and temperature and has been determined by careful laboratory measurements[3]. For *m*-cresol purple,

$$\mathrm{p}K_2 = \frac{1245.69}{(T / \mathrm{K})} + 3.8275 + 0.00211(35 - S) \qquad (8)$$

where $293 \le T/\mathrm{K} \le 303$ and $30 \le S \le 37$.

8.3 Correction for pH change resulting from addition of the dye

The addition of indicator dye to the sea water sample will perturb the pH (another acid–base system has been added!). Although care is taken to minimize this (by adjusting the dye solution pH), it is desirable to correct for the addition of dye to obtain the best pH measurements.

Although, in principle, the pH perturbation could be calculated from a knowledge of the equilibrium chemistry of the sample and the dye, it is simpler to evaluate the magnitude of the correction empirically. A pair of additions of dye is made to each of a series of sea water samples with different pHs, and the change in the measured ratio (A_1/A_2) with the second addition of indicator solution is determined as a function of the measured value (A_1/A_2) determined after the first addition of dye using a least-squares procedure (SOP 23):

$$\frac{\Delta(A_1 / A_2)}{V} = a + b(A_1 / A_2) \qquad (9)$$

[3] Although DelValls and Dickson (1998) have suggested that this pK_2 may be in error because of an error in calibrating TRIS buffer, it seems that there may be a compensating error that largely mitigates the proposed correction. The pK_2 given here is that from Clayton and Byrne (1993).

where V is the volume of dye added at each addition. The final, corrected, absorbance ratio is

$$(A_1 / A_2)_{\text{corr}} = (A_1 / A_2) - V[a + b(A_1 / A_2)]. \quad (10)$$

8.4 Example calculation

$$\begin{aligned} t &= 25°\text{C}, \\ S &= 35, \\ \text{p}K_2 &= 8.0056, \end{aligned}$$

and for indicator stock solution with $A_1/A_2 = 1.6$,

$$. \frac{\Delta(A_1 / A_2)}{V} = 0.125 - 0.147(A_1 / A_2) ..$$

Measured absorbances:

Sea water: $A_{434} = 0.02433$; $A_{578} = 0.01936$; $A_{730} = 0.08365$
Dye + sea water: $A_{434} = 0.45123$; $A_{578} = 0.84574$; $A_{730} = 0.08298$

After addition of dye,

$$A_1 / A_2 = \frac{0.84574 - 0.01936 - (0.08298 - 0.08365)}{0.45123 - 0.02433 - (0.08298 - 0.08365)} = 1.93430.$$

Corrected to zero dye addition ($V = 0.08$ cm^3),

$$\begin{aligned} (A_1 / A_2)_{\text{corr}} &= 1.93430 - 0.08[0.125 - 0.147(1.93430)] \\ &= 1.94705 \end{aligned} \quad (11)$$

and thus

$$\text{pH} = 8.0056 + \log_{10}\left(\frac{1.94705 - 0.00691}{2.2220 - 1.94705 \times 0.1331}\right) = 8.0005.$$

9. Quality assurance

9.1 For general principles of analytical quality control see Chapter 3

9.2 Specific applications of analytical quality control

9.2.1 Spectrophotometer performance

The spectrophotometric performance of the instrument used can be confirmed using reference materials that are available from the U.S. National Institute for Standards and Technology (NIST). SRM 2034 is a holmium oxide solution in a sealed cuvette that allows the wavelength accuracy of the spectrophotometer to be determined; SRM 930d is a set of absorbance filters that allows the

absorbance measurement accuracy to be verified. Property control charts of these measurements should be maintained, and the spectrophotometer adjusted if it goes out of tolerance. (Nevertheless, the procedure detailed here is fairly insensitive to minor changes in spectrophotometer performance.)

A more important concern is that the spectrometer must have a high stability. This can be confirmed by making a series of repeated measurements on a system of constant absorbance (*e.g.*, SRM 930d or a thermostated buffer solution containing indicator dye) and computing the standard deviation at the wavelengths of interest.

9.2.2 Precision

A precision of better than 0.001 pH units (1 SD) is possible with care—particularly in regard to the sample handling. The results of duplicate analyses should be plotted on a range control chart (SOP 22).

9.2.3 Bias

The bias of spectrophotometric pH measurements depends on the accuracy with which the various extinction coefficient ratios were determined, and on the accuracy of the values assigned to the values of pK_2. A significant advantage of spectrophotometric measurements is that, if more accurate information becomes available for these parameters at a later time, the pH results obtained can be adjusted without any degradation in precision provided that the original data are retained. At present, the likely bias is estimated to be less than 0.005 pH units.

10. Bibliography

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