

The Development of Seawater Standards for Dissolved Nutrients

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Introduction

Mass balance equations have shown that the input of the major biogenic nutrients (nitrogen, phosphorous and silicon) to the ocean play an important role in regulating global oceanic production (McElroy, 1983). Most of these nutrients are terrestrial in origin, finding their way to the oceans via atmospheric and riverine pathways. Once in the marine system nutrients are utilised by biological organisms for primary production either by advection of nutrient rich water from depth or from local regeneration resulting from cell breakdown.

In the open ocean the utilisation of nutrients by organisms in the surface waters usually results one or more of those nutrients becoming limiting to its growth. However coastal and estuarine waters are increasingly subject to anthropogenic input stress whereby regular input of nutrients as 'run-off' from agricultural land results in artificially prolonged algal growth. In either case the need for high quality nutrient measurement is driven by economic factors which may result from climate change, depleted fisheries, eutrophication and imbalanced aquatic ecosystems.

Nutrients are measured using a variety of wet chemistry techniques which generate a colour reaction measurable with a colorimeter or spectrophotometer. The basis of the technique is to add reagent (or reagents) to the seawater sample, allow a colour to develop and then to measure the intensity of the colour against blanks and standards. Manual methods usually allow the colour to develop fully before measurement whereas most automated methods (eg segmented flow analysis, flow injection analysis) provide partial colour development with time controls. Concentrations of nutrients are measured in optical cells (static or flowthrough), using a spectrophotometer tuned to defined wavelengths.

Quality assurance is an essential part of any chemical analysis. The measurement of nutrients in seawater has posed a number of difficulties particularly in terms of matrix interference and calibration. Ocean Scientific International, who produce the IAPSO seawater standards for Practical Salinity, have been working for some years to develop seawater calibration standards for dissolved nutrients. This article describes that development and the current availability of reference materials and standards.

Certified Reference Materials

The International Council for Exploration of the Seas (ICES) has expressed an urgent need for the development of certified reference seawater for dissolved nutrients. Unfortunately, this expression took place almost a decade ago at the meeting of the IOC-IAEA-UNEP Group of Experts on Standards and Reference Materials (GESREM) and no such material has appeared to date.

A certified reference material (CRM) is a material or substance, one or more properties of which, are sufficiently well established and certified to be used for the calibration of an apparatus. In many laboratories, a CRM is analysed, as a sample, to determine the accuracy of analysis as part of a quality assurance scheme. In the case of nutrients, the ideal CRM would be a natural seawater which has been analysed, by more than one method, for dissolved nutrients, with confidence limits provided for the determinands.

One of the main problems encountered in the production of a CRM, has been the stability of the product. Biological and chemical activity can lead to changes in the nutrient concentrations in short periods of time. Various forms of chemical preservatives have been used which include organic solvents, formalin, potassium cyanide, sodium fluoride, various acids and mercuric chloride (see Kirkwood, 1992).

The use of additives is generally undesirable as it changes the matrix of the reference. In addition, mercuric chloride, the most commonly used poison, is known to degrade the efficiency of the cadmium reduction columns on some nitrate analysers. There are also problems associated with the handling and transport of solutions containing such toxic elements. As a result, some workers have investigated the use of heat as a means of preserving the nutrient concentrations in seawater (Aminot, 1991). Uncertified reference materials have been produced for dissolved nutrients in seawater by the Quasimeme Project (see Topping 1997). These materials which comprise seawaters, spiked with nutrient salts, are provided as part of a laboratory performance study. The Quasimeme Project preserves mixed nutrients in seawater by autoclaving the samples.

Although reference materials are important in quality assurance, the preparation of working standards is a fundamental component of any chemical analysis. Ocean Scientific International Ltd. (OSIL) studied the feasibility of producing commercially available working standards for nutrients in seawater. A number of products were already available from other sources, but none offered natural seawater matrix at salinity 35.

The Need for Nutrient Standards

The need for seawater nutrient standards has intensified in recent years, with international programmes such as WOCE, requiring high-quality nutrient data to be collected at sea. Most shipboard analyses are carried out using automated chemistry systems which measure nutrients colorimetrically. The kinetics, and often the ultimate colour intensities of the chemistry of these techniques, are generally affected by the presence of seawater matrix salts. Elimination of these salt effects requires that working calibration solutions are prepared in natural low nutrient seawater (LNS) rather than in demineralised water. The WOCE protocol produced by Gordon et al (1992) stated a preference for the production of working standards in natural low nutrient seawater (LNS), but recognised that artificial seawater may suffice when LNS was unavailable. In continuous-flow analysis, LNS can also be used as a refractive-index blank. This is to correct for a false positive absorbance signal, generated by the refractive effects within the flow cell when saline samples are run with a demineralised water baseline and inter-sample wash.

Low Nutrient Seawater

The first requirement of a standard for nutrients in seawater, is the seawater matrix itself. It must be significantly lower in nutrients than the required standard concentration. This is particularly difficult for open ocean measurements where analyte levels can be lower than 1µMolar as a result of the surface-water depletion by plankton. Low Nutrient Seawater (LNS) can be prepared naturally or artificially. OSIL regularly collects relatively large quantities of seawater from the mid-Atlantic Ocean for operation of the IAPSO Standard Seawater Service and investigated methods for depleting the nutrient content. Chemical removal, using co-precipitation techniques with iron and aluminium compounds, were found to be effective in terms of nutrient depletion, but the subsequent removal of the precipitate proved difficult and costly. Biological depletion using an inoculum of plankton to create an artificial 'bloom' in the bulk seawater was occasionally successful but was unreliable due to the difficulty in controlling the conditions for growth. As a result, OSIL has now standardised on a practice of measuring each batch of bulk seawater for dissolved nutrients and reserving the very low measuring batches for bottling. This LNS has the advantage of being naturally depleted which, combined with our processing/bottling regime, gives rise to a very stable product. The LNS fulfils all the needs of a seawater matrix for the preparation of standards, carrier solution and RI blank.

Standards Preparation

Initial attempts to produce 'mixed' nutrient standards in LNS gave rise to the problems of preservation. Although we were able to stabilise the product using mercuric chloride, this was considered undesirable, by many users, for the reasons described earlier. Another problem, which arose, was defining the most useful working concentration of the nutrients. It became obvious that it would be commercially uneconomical to produce the wide range of standards necessary to meet all user needs.

The problems described above led to the production of a range of marine nutrient standard kits (MNSK). These comprise concentrates of single nutrients in deionised water, together with bottles of Low Nutrient Seawater (LNS) – (Table 1).

Table 1. Marine Nutrient Standard Kits

<u>(μM)</u>	<u>Code</u>	<u>Analyte</u>	<u>Matrix</u>	<u>Concentration</u>
	NSSPO	Phosphate	Deionised Water	100
	NSSNI	Nitrite	“	100
	NSSNA	Nitrate	“	1000
	NSSSI	Silicate	“	1000
	LNS	All nutrients	Ocean Water	<1 μM

S = 35

The user dilutes the concentrate with the LNS to provide working standards. This method overcomes problems of preservative as the concentrates of single nutrients in deionised water and the LNS, are stable for long periods. However, once diluted, the working standards should be used immediately after preparation. The concentrate method also allows the users to define their own working concentration range, thereby overcoming the need for a wide range of products. Although a confidence is given for the nutrient levels in the concentrates and the LNS, the final working standard concentration is dependent on the dilution made by the user.

Performance Evaluation

Although our standards may be used as working calibration standards, many laboratories make up their own standards using nutrient salts and incorporate our standards as quality assurance (QA) samples. As a result of user requests, we now offer Performance Evaluation (PE) samples for dissolved nutrients and salinity. These PE samples are supplied, ‘unknown’, to analysts who include them in a normal analysis run. After analysis, the measured value for each PE sample is forwarded to our laboratory where a certificate is issued showing the true value and the analytical error.

The need for a certified reference material still exists for nutrients in seawater. However data quality for marine nutrients can be improved by the careful selection of standards, carrier solutions and performance evaluation samples. Further information may be obtained by contacting OSIL (paul.ridout@oceanscientific.co.uk).

References

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