Dissolved organic phosphorus concentrations in the northeast subarctic Pacific Ocean

Abstract—Shipboard determinations of dissolved organic P (DOP) concentrations were made for NE subarctic Pacific Ocean samples with three different oxidation-hydrolysis methods: UV irradiation, acid persulfate digestion, and a rigorous method that combines the UV and persulfate treatments (combination method). The UV technique was 71 ± 9% and the persulfate technique 83 ± 9% efficient relative to the combination method for these coastal and open-ocean surface samples. Combination method results indicate that levels of DOP in the mixed layer of the NE Pacific during August and September ranged from 0.17 to 0.38 µm. These values are comparable to those found for the NW Atlantic in a previous study with similar methodology. A significant inverse correlation ($r^2 = 0.76$) was found between DOP, measured by the combination method, and apparent oxygen utilization. Analysis of a small number of samples by crossflow ultrafiltration showed that 75–85% of DOP in surface seawater was comprised of < 10,000 nominal molecular weight (NMW) material. However, samples taken from 100- and 1,000-m depths were relatively enriched in colloidal (> 10,000 NMW) fractions, which formed 33–100% of the total DOP in these waters.

Reports of high concentrations of dissolved organic N (DON; Suzuki et al. 1985) and dissolved organic C (DOC; Sugimura and Suzuki 1988) in seawater have initiated renewed interest in the determination of dissolved organic matter in the oceans. Using new, high-temperature combustion oxidation methodology, these researchers obtained DOC and DON values for surface seawater that were 2–4 times greater than those measured with UV or persulfate techniques. In a recent study, we sought to ascertain whether dissolved organic P (DOP) concentrations were similarly underestimated by comparing the efficiencies of the UV and persulfate DOP methods with a rigorous two-step oxidation-hydrolysis method that combines the UV and persulfate techniques (Ridal and Moore 1990). It was shown that for a variety of NW Atlantic seawaters, the UV and persulfate techniques gave similar results, each yielding ~90% that of the new two-step method. However, evidence has been presented which suggests that surface seawater DOC values may be 30–50% higher in the North Pacific than in the North Atlantic (Y. Suzuki pers. comm.; Kirchman et al. 1991). We therefore questioned whether measurements of DOP in Pacific waters made by the new two-step oxidation method would be higher than those measured by standard persulfate or UV oxidation procedures. We report here results of such a comparison study, using NE subarctic Pacific seawaters and including a profile taken at Ocean Weather Station Papa.

Sampling was performed aboard the CSS Parizeau (Inst. Ocean Sci., Patricia Bay, B.C.) from 22 August to 7 September 1990 during a line P cruise (Fig. 1). Water samples were taken with acid-cleaned 10-liter Go-Flo bottles. Samples for soluble reactive P (SRP) and total dissolved P (TDP) were pressure-filtered (N2, 0.35 kg cm$^{-2}$) through acid-washed 0.4-µm Nuclepore filters supported on polycarbonate filter holders.
Fig. 1. Sampling stations in the subarctic NE Pacific. Major current patterns of the region are also shown (after Tabata 1974).

Samples were taken with thoroughly cleaned Teflon bottles after 250 ml of sample had passed through the filter. SRP values were determined immediately after sampling, using the molybdenum blue technique as described by Koroleff (1983) and a LKB Ultraspec 4050 spectrophotometer fitted with a 10-cm pathlength cell. Absorbances were determined at 885 nm and found to be linear over the sample concentration range. Analytical precision for shipboard SRP determinations was less than or equal to ±0.01 μM, when taken as 1 SD of triplicate analyses.

Routine nutrient samples were taken from 1.7-liter Niskin bottles and processed onboard ship with a Technicon AutoAnalyzer. Excellent agreement was found for SRP values determined manually and by AutoAnalyzer. Dissolved oxygen was determined onboard ship with a semi-automated Winkler titration method, and temperature and salinity measurements were made from reversing thermometers and a salinometer.

Several 6-liter samples were gravity filtered from Go-Flo bottles through acid-cleaned 0.45-μm Gelman Mini Capsule Versapor polycarbonate filters into a 20-liter acid-cleaned PVC carboy and then treated by a Millipore Pellicon crossflow ultrafiltration system fitted with a 10,000 nominal molecular weight (NMW) filter cassette as described elsewhere (Moran and Moore 1989), except that a peristaltic pump was used in lieu of the more cumbersome air-driven pump-compressor apparatus. Concentration factors for the >10,000 NMW (colloidal) material ranged from 10 to 15. The units of NMW are used here because the ultrafilter size cutoff is not precise, and the filter may retain macromolecules ≥10,000 NMW (Millipore Corp.).

Once subsamples had been extracted for SRP analysis, the remaining sample was acidified with sulfuric acid to pH 3 and refrigerated to stabilize the samples before TDP analysis (Ridal and Moore 1990). A replicate set of samples from Station Papa was stored frozen in acid/persulfate-cleaned 500-ml polypropylene bottles and analyzed ashore to compare TDP recoveries obtained with different UV apparatuses (see below).

TDP determinations followed Ridal and Moore (1990) with the modifications outlined below. DOP was taken as the difference between TDP and SRP values, which may represent upper limits for the values of truly organically bound P analyzed by each method, as minor amounts of inorganic P compounds may be present in unpolluted surface seawater (Solórzano and Strickland 1968; Armstrong and Tibbetts 1968). These compounds are not reactive to the molybdate reagent but can be hydrolyzed to phosphate after treatment with the TDP methods.

The shipboard UV oxidation apparatus was provided by J. Thompson. It was fitted with a 550-W Hg lamp (Hanovia) that had been used for 25 h before experimental use. The lamp sat in a quartz cooling well through which a continuous flow of seawater (0.45-μm filtered) was maintained. Quartz tubes (15 × 1.80-cm i.d., 2.18-cm o.d.) contained 35-ml samples, and the centers of the tubes were 4 cm from the walls of the UV lamp. Before sample analysis, all quartz tubes were checked for possible release of contaminating substances from the walls of the tubes during UV irradiation (including excessive leaching of silica) by irradiating distilled-deionized Millipore Super-Q water samples.
for 6 h. No detectable contamination was observed. Surface coastal and open ocean seawater samples were used to determine the irradiation time necessary for maximum recovery (Ridal and Moore 1990). Asymptotic values were achieved after ~6 h of exposure; however, all samples were irradiated for 9 h to ensure maximum release of DOP by the UV method. Sample temperatures measured immediately after this UV irradiation period were 65°-70°C.

Due to the UV apparatus design, not all samples could be processed at the same time. Surface samples were processed immediately after collection, and all samples were processed within 32 h. Replicate samples from station P26 samples, stored frozen, were analyzed with the UV apparatus described previously (Ridal and Moore 1990) which was equipped with a new 1,200-W Hg lamp (Hanovia). \( \text{H}_2\text{O}_2 \) (75 µl) was added to the samples before irradiation. All UV runs included a blank of Super-Q as a check against inadvertent contamination of the UV tubes, acid reagent, or \( \text{H}_2\text{O}_2 \) solution. The analytical precision (1 SD) for triplicate TDP determinations by both UV methods was less than or equal to ±0.01 µM.

The acid persulfate method (Menzel and Corwin 1965; Ridal and Moore 1990) was modified for 25-ml sample volumes contained in Teflon reaction flasks. The Teflon flasks (60 ml) were cleaned before analysis by leaching for several days with 4.5 M \( \text{H}_2\text{SO}_4 \), followed by applying the persulfate method with Super-Q water contained in the flasks. Analytical precision (1 SD) for shipboard TDP analyses by both the single-step, acid persulfate oxidation and the two-step, UV + persulfate (combination) method was ±0.015 µM.

The waters sampled are within the Alaskan gyre system which is bounded by the easterly flowing Subarctic Current to the south, and the westerly flowing Alaskan Current to the north (Fig. 1). In this region, a 100-120-m layer of low-salinity water caps the permanent halocline, and upwelling across the halocline is limited to a few tens of meters a year (Tabata 1974). Chl a levels are uniformly low throughout the year and phytoplankton stocks are dominated by <10-µm-sized plankton (Booth 1988). High inorganic nutrient concentrations are observed throughout the year (surface \( \text{NO}_3^- \) levels are always >5 µM; Anderson et al. 1969), which has been explained by grazer limitation of phytoplankton (Frost 1987), Fe limitation (Martin et al. 1989), and grazer-enhanced recycling and preferential \( [\text{NH}_4]^+ \) uptake (Wheeler and Kokkinakis 1990).

Oceanographic data shown in Figs. 2 and 3 are measurements of samples taken at two open-ocean stations (P20 and P26 Papa) in the subarctic NE Pacific. Stations P20 and P26 had mixed-layer depths of ~30 m with the high nutrient concentrations typical of subarctic Pacific waters (Anderson et al. 1969; Martin et al. 1989). Seasonal thermoclines and well-developed subsurface \( \text{O}_2 \) maxima also were observed in the upper 125 m at these stations. Chl a max values were 0.6 µg liter\(^{-1}\) for P20 and 0.3 at P26, the latter being a typical value for phytoplankton standing stock at Station Papa in August (Booth 1988). At the coastal-slope station (P4, 1,300-m depth), the mixed layer was ~10 m, and in this layer nutrients were moderately depleted (0.45 µM \( \text{PO}_4^-; 0.8 \text{µM NO}_3^-; 3.0 \text{µM Si} \)), and Chl a levels were 0.8 µg liter\(^{-1}\) and uniform to 20 m.

DOP data are presented in Table 1 for surface and some subsurface depths as measured by UV, persulfate, and combination methods. The combination method measured more DOP than the other methods for the great majority of samples. The largest differences between methods are generally found for samples taken from above 100 m; samples taken from below this depth show better agreement between methods. This trend suggests production of chemically resistant P compounds in the upper layer by the biota, and degradation by microorganisms on time scales shorter than those of subsurface mixing processes. Chemically resistant P compounds might include the phosphonates, which are known products of marine biota (Cembella et al. 1984), and have been identified in marine sediments by \( ^{31}\text{P} \) NMR (Ingall et al. 1990). Phosphonates were found to be resistant to perchlorate oxidation, and a common algal P metabolite, 2-aminoethylphosphonate, required a very long UV irradiation time.
Fig. 2. Hydrographic and nutrient data at Station P20. TDP values measured with the combination method are shown for comparison with the SRP data.

(18 h) to achieve near-quantitative release of the organically bound P (Cembella et al. 1986). Aminophosphonates are susceptible to breakdown by marine microbes, although degradation processes are inhibited by the presence of easily degraded orthophosphate-containing compounds, such as phosphate monoesters (Rosenberg and La Nauze 1967). These compounds may therefore be degraded slowly on biological time scales (days to weeks), but are broken down on time scales shorter than subsurface mixing processes (tens of years).

Figure 4 presents regression analysis graphs derived from all samples analyzed by the three different methods. The slope of the graph is a measure of the relative efficiency of the UV or persulfate method when compared with the combination method. The slope of the line for the UV method was 0.707±0.090 (95% C.I.), suggesting an approximate 71% efficiency relative to the combination method; the persulfate graph slope of 0.832±0.086 suggests an 83% efficiency for DOP recovery by that method.

When these efficiencies were compared with those found from the earlier study with NW Atlantic waters (UV, 90±6%; persulfate, 87±8%), it appeared that the UV method used for this study performed poorly compared with the combination and persulfate methods. To test if poor recovery was a function of UV apparatus, we froze replicate samples from P26 and analyzed them at Dalhousie with the 1,200-W Hg lamp UV method (Ridal and Moore 1990). Regression analysis of these UV DOP values against the combination method values resulted in a slope of 0.635, in good agreement with the slope of 0.650 derived from shipboard measurements. This comparison study suggests that the shipboard method was operating efficiently; therefore the relatively low DOP recovery obtained for subarctic Pacific Ocean samples may be evi-
Fig. 3. As Fig. 2, but for Station P26.

Fig. 4. Least-squares regression analysis of the DOP results obtained with the UV and persulfate methods plotted against the combination method results. Dashed lines are linear regression fits to the data.
Table 1. Shipboard DOP determinations with three different methods for samples from a coastal-slope station (P4) and two open-ocean stations (P20 and P26).

<table>
<thead>
<tr>
<th>Sta.</th>
<th>Depth (m)</th>
<th>UV (±0.02)*</th>
<th>Persulfate (±0.025)*</th>
<th>Combination (±0.025)*</th>
</tr>
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<tbody>
<tr>
<td>P4</td>
<td>2</td>
<td>0.29 0.33</td>
<td>0.38</td>
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<tr>
<td></td>
<td>10</td>
<td>0.24 0.33</td>
<td>0.31</td>
<td></td>
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<tr>
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<td>10</td>
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<td>0.22</td>
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<td></td>
<td>50</td>
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<td>100</td>
<td>0.06 0.12</td>
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<td>200</td>
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<td>400</td>
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<td>800</td>
<td>0.07 0.08</td>
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<td>0.00 0.17</td>
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<td>3,800</td>
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</tr>
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<td>10</td>
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* Uncertainty for DOP determinations taken as the sum of TDP and SRP precisions.

Acknowledgment for different reactivities to UV of DOP from differing oceanic regions. Other workers have noted that the UV method can result in variable recoveries of DOP for seawater taken from different marine environments (Solorzano and Strickland 1968; Armstrong and Tibbetts 1968).

Because studies of DOP in marine environments are often limited to the upper water column (Jackson and Williams 1985; Orrett and Karl 1987), a statistical ANOVA was performed for the P20 and P26 data from the upper 100 m (n = 8), pooled on the basis that these values were measured from similar oceanic environments. Although the analysis resulted in rejection (P = 0.147) at the 0.05 level of significance of the hypothesis that a difference between the combination and persulfate data exists, the hypothesis was accepted (P = 0.0199) at the 0.05 level of significance for an analysis comparing the UV and combination method results.

Although statistically higher DOP values were measured by the combination method compared with those found by the UV technique for upper ocean NE Pacific water samples, the DOP levels measured by the combination method are still within the range of DOP values (0.1–0.4 μM) previously reported for Pacific Ocean surface samples measured with standard (Jackson and Williams 1985; Orrett and Karl 1987; Walsh 1989) or specialized TDP procedures (Solorzano and Strickland 1968; Cembella et al. 1986; Karl and Tien 1992). A similar range of DOP values was measured with the combination method for surface seawater samples from the NW Atlantic (Ridal and Moore 1990), suggesting that C : P ratios of dissolved organic matter in surface waters are high (375–1,000 : 1) when calculated with DOC values measured by high-temperature combustion oxidation methods for Atlantic (Y. Suzuki pers. comm.) and Pacific (Druffel et al. 1989; Suzuki et al. 1992) surface waters. Such high C: P ratios are consistent with high bioavailability and rapid turnover of DOP compounds in marine environments (Jackson and Williams 1985; Orrett and Karl 1987).

An interesting feature of high-temperature combustion DOC data from the NW Pacific (Sugimura and Suzuki 1988; Suzuki et al. 1992) is their high negative correlation with apparent oxygen utilization (AOU). The NE Pacific exhibits a large dissolved oxygen gradient and an extremely shallow O2 minimum, and we therefore questioned whether a similar negative correlation would be found between DOP and AOU in vertical profiles from this region. Figure 5 shows graphs of AOU vs. DOP measured by the combination method for the two open-ocean stations. Both sets of DOP results show rapid decreases with depth down to the O2 minimum, consistent with the increases in AOU. DOP values below the O2 minimum show marginal increases with decreasing AOU. A significant correlation was found from statistical analysis of all AOU and DOP data \( r^2 = 0.76; \text{AOU (μM)} = -0.00052 (\text{μM DOP}) + 0.167 \). Because comparisons between AOU and DOP are hampered by the relative imprecision of the DOP data, particularly for deep-water samples, a more precise method for measuring DOP and a greater number of deep-water samples are required to clarify this relationship.
A limitation of the DOP methodology used here is that the resulting DOP fraction is operationally defined, providing little information concerning the bioavailability of the measured P compounds (Cembella et al. 1984; Orrett and Karl 1987). However, an interesting result, shown in Fig. 5, is that DOP levels decrease to below detection limits for depths near the $O_2$ minimum. Although certainly part of the DOP measured by the combination method represents a reservoir of slowly degrading organophosphorus compounds, our observation that DOP values can fall to below the detection limit supports the idea that, within the time scales of oceanic mixing, all DOP measured by the combination method is available to the microbiological community. In the light of the measurement of a significant pool of very old DOC (Williams and Druffel 1987), it would appear that this refractory DOM (dissolved organic matter), having survived a number of oceanic mixing cycles, is depleted in P-containing compounds.

Suzuki et al. (1985) and Sugimura and Suzuki (1988) used size exclusion chromatography to show that the dominant molecular weight range for the tropical Pacific surface DOM analyzed by their instruments was $>4,000$ daltons, comprising $\sim75\%$ of the total. We used crossflow ultrafiltration to investigate whether a similar distribution of molecular weight would be found in the subarctic NE Pacific. Figure 6 shows the data from molecular weight separation obtained with our ultrafiltration technique. In each case, the sums of the low and high molecular weight fractions equaled the DOP values obtained from direct measurements within experimental errors, indicating that losses to the ultrafilter were insignificant. The DOP analyzed by ultrafiltration was mainly $<10,000$ NMW material ($75$–$85\%$), in agreement with the DOP fractionation
results found for the NW Atlantic. Although the portion of colloidal DOP was small (15–25%) in surface samples, subsurface samples became proportionately more rich in colloidal P, with 33% of the 100-m and 50–100% of the 1,000-m DOP found in the colloidal fraction. A similar trend was observed for crossflow ultrafiltered samples from the NW Atlantic (Ridal and Moore 1990). These observations are consistent with preferential biological uptake of the low molecular weight DOP fraction, containing compounds such as phosphate monoesters which are labile to enzymatic hydrolysis, compared with the colloidal fraction, thought to contain components such as phosphoproteins, DNA, and other polynucleotides that require extensive exoenzymatic breakdown prior to heterotrophic uptake (Cembella et al. 1984).

There is an increasing awareness that dissolved organic matter plays important roles in biological and chemical oceanic systems (Hedges 1987). It is therefore important to critically examine the sizes of various organic compartments. Using a rigorous chemical oxidation-hydrolysis procedure, we find that mixed-layer DOP values ranged from 0.17 to 0.38 μM for NE Pacific waters. The standard persulfate method detected 83% and the UV method 71% of the DOP measured with the combination method, compared with respective values of 87 and 90% for Atlantic waters (Ridal and Moore 1990). These results suggest therefore that historical oceanic analyses have not grossly underestimated DOP concentrations. A similar conclusion was reached by Karl et al. (1992) based on the agreement between TDP results from UV and high-temperature oxidation analyses of central Pacific waters and on their occurrence in the Redfield proportion with TDN values measured by high-temperature combustion oxidation. Our DOP results display an inverse correlation between DOP and AOU for samples taken above the oxygen minimum, with DOP levels falling to near or below detection limits at the oxygen minimum, suggesting that the bulk of measured DOP compounds are ultimately available to the marine microbiological community.

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References


Cembella, A. D., N. J. Antia, and P. J. Harrison. 1984. The utilization of inorganic and organic phosphorus compounds as nutrients by eukaryotic

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Notes

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