

Response to Reviewers

Manuscript title: “Stoichiometric deviation and regulatory mechanisms of AOU-nutrient ratio in the oligotrophic Northwest Pacific Ocean”

Presentation: Reviewer comments are reproduced in black, and the authors’ responses and descriptions of revisions are shown in blue.

Response: Dear Editor and Referees, we sincerely thank you for the careful assessments and constructive comments. These comments substantially improved the methodological transparency, analytical consistency, organization, and balance of the manuscript. We have addressed every comment below and revised the manuscript and Supplementary Materials accordingly.

Because the comments were received at different stages of the interactive review, several related issues were revised iteratively. In preparing this consolidated response, we considered the comments jointly and updated all descriptions of revisions to correspond to the final revised manuscript. The scientific rationale of our earlier online responses remains unchanged; where a later comment refined an earlier modification, the response below reports the integrated final treatment.

Summary of the integrated revisions

1. We now treat $r\text{PreNO}_x$ and $r\text{PrePO}_4$ strictly as parameter-dependent diagnostic residuals. The final calculations use nine mass-balance-constrained parameter combinations, and the principal conclusions remain based on regressions of directly measured AOU and nutrients.
2. We reorganized the analytical framework around three hydrographic groups: upper, intermediate, and deep waters—while retaining the eight named water masses only to establish hydrographic context. These groups are explicitly distinguished from fixed depth intervals and the entire-water-column analysis.
3. Figures 2–5 and 7, their captions, and the associated Results and Discussion were reorganized to present consistent vertical profiles and groupwise comparisons. The upper and intermediate AOU/DIP slope labels in Fig. 5b were corrected to 116.04 and 103.17, respectively.
4. We expanded the descriptions of nutrient and chlorophyll-*a* preservation, and fully documented the laboratory-generated xanthan gum calibration for TEP.

5. We moderated the interpretation of TEP: its absolute concentration is not globally exceptional, and TEP production is presented as one plausible contributor to the upper-water deviations rather than as a sole or dominant driver. We also added a qualified discussion of denitrification within particle microsites.

Response to Referee #1 — Initial Review

Overall assessment

From the observed AOU/nutrient ratios, the deviations from the classical Redfield stoichiometry are actually relatively modest. Nevertheless, the authors provide a detailed mechanistic interpretation of these subtle deviations from multiple perspectives, particularly focusing on TEP production and microbial metabolic strategies. In my opinion, this represents an interesting and worthwhile attempt.

The central hypothesis proposed by the authors is that strong stratification in the TNWP leads to chronic nutrient limitation in surface waters, which promotes the production of carbon-rich TEP by phytoplankton. Subsequently, microbial communities selectively degrade nitrogen- and phosphorus-rich organic components while preferentially regenerating phosphorus, resulting in nutrient regeneration exceeding organic carbon oxidation and ultimately producing AOU/nutrient ratios slightly lower than the classical Redfield values.

From the apparent regression slopes, however, the AOU/nutrient ratios reported here are substantially higher than those previously reported by Zhu et al. in the northern South China Sea (e.g., AOU/DIN = 3.09 in Zhu et al. versus 6.28 in the present study). In my view, these represent two fundamentally different scenarios. The extremely low ratios observed by Zhu et al. were likely dominated by physical processes such as water-mass mixing, whereas the present study represents a typical open-ocean environment where biogeochemical processes are expected to play a much more important role.

TEP itself is a carbon-rich, nitrogen- and phosphorus-poor organic material. From the perspective of remineralization, degradation of TEP consumes oxygen but releases relatively little nitrogen and phosphorus. Consequently, TEP degradation would be expected to increase, rather than decrease, the AOU/nutrient ratios relative to the Redfield expectation. Therefore, the degradation of TEP alone does not appear to support the mechanism proposed in this manuscript.

In contrast, from the perspective of TEP production, the mechanism is much more convincing. TEP production originates from phytoplankton photosynthesis, during which oxygen is produced and carbon is fixed, whereas the associated consumption of nitrogen and phosphorus is substantially lower than predicted by the Redfield ratio because of the high C:N composition of TEP. Under this scenario, AOU decreases whereas nutrient concentrations decrease much less than

expected, potentially leading to relatively low AOU/nutrient ratios. Therefore, it seems reasonable that phytoplankton production of TEP could contribute to reduced AOU/nutrient ratios within the euphotic zone, or approximately the upper 0–300 m where active photosynthesis occurs.

Overall, I consider the attempt to explain the reduced AOU/nutrient ratios in the upper ocean from the perspective of TEP production to be novel and potentially important. My major comments and concerns are as follows:

Response: We thank the reviewer for this thoughtful assessment and for distinguishing the effects of TEP production from those of TEP degradation. In the final manuscript, we retain this distinction, limit the TEP-related interpretation to a plausible upper-water contribution, and discuss the intermediate- and deep-water processes separately. Below, we provide detailed point-by-point responses to all comments.

Major comments

1. Use and interpretation of preformed nutrients

The use of preformed nitrate (PreNO_3) and preformed phosphate (PrePO_4) requires additional caution. The classical concept of preformed nutrients is based on the assumption that organic matter remineralization and nutrient regeneration follow a prescribed stoichiometric relationship, typically the Redfield ratio (or a fixed empirical remineralization stoichiometry). However, the central premise of the present manuscript is precisely that organic matter remineralization in the study region deviates from the classical Redfield stoichiometry. This creates a potential inconsistency between the methodological assumptions and the scientific conclusions.

For this reason, I do not suggest removing the calculations of PreNO_3 and PrePO_4 , as they may still serve as useful diagnostic residuals. However, I recommend avoiding interpreting them as the true "preformed nutrients." Instead, they should be presented as residual indicators derived under a predefined remineralization stoichiometry. Otherwise, there is a risk of circular reasoning: the manuscript uses preformed nutrients calculated under an assumed stoichiometric relationship to demonstrate that the remineralization stoichiometry itself deviates from that assumption. This logical issue should be explicitly acknowledged and discussed.

In addition, the manuscript adopts the modified approach proposed by Letscher and Villareal by introducing different remineralization coefficients for DOM and POM, which is intended to improve the realism of the calculations. This is good. Nevertheless, the parameters used in the

calculations (f_{DOM} , f_{POM} , r_{DOM} , and r_{POM}) remain empirical assumptions rather than values constrained by observations from the study region. In particular, Table 1 lists multiple candidate values of r_{POM} , yet the manuscript does not explain which value was ultimately adopted or the criteria used for selecting among them. Consequently, the calculated PreNO_3 and PrePO_4 are highly dependent on these empirical parameterizations and therefore should not be regarded as independent evidence supporting the proposed mechanism responsible for the low AOU/nutrient ratios. Rather, they should be interpreted as model-dependent diagnostic quantities whose uncertainties deserve further discussion.

Response: We thank the reviewer for identifying this potential circularity. We agree that the original terminology overstated both the physical meaning and the evidential independence of the calculated quantities. In the final manuscript, the former PreNO_3 and PrePO_4 terms are consistently presented as residual preformed total oxidised nitrogen ($r\text{PreNO}_x$) and residual preformed phosphate ($r\text{PrePO}_4$). These are parameter-dependent diagnostic residuals: a nonzero value indicates a departure from the prescribed reference parameterization, but it is not a model-independent estimate of the true preformed nutrient inventory and does not independently demonstrate a particular mechanism.

The main evidence for nonclassical oxygen–nutrient coupling therefore remains the regression relationships calculated directly from measured AOU, DIN, and DIP. The residuals are used only as complementary comparisons with published reference frameworks. We also avoid using locally derived AOU–nutrient slopes as calculation inputs, because doing so would be circular and would reduce the residuals by construction.

Following the reviewer’s subsequent mass-balance comment, the final sensitivity analysis no longer uses the 27 unrestricted combinations described in the first online response. It uses 9 physically constrained combinations: three complementary $f_{\text{DOM}}/f_{\text{POM}}$ pairs (0.4/0.6, 0.5/0.5, and 0.6/0.4), each combined with three r_{POM} values (6.9, 8.75, and 10.6) (Response Table 1/Revised Table S2). For each observation, the 9 results were summarized as minimum–maximum (arithmetic average $\pm$ standard deviation). The arithmetic average was used in the figures and interpretation, whereas the minimum, maximum, and standard deviation describe sensitivity to the prescribed parameter combinations rather than measurement or sampling uncertainty. These summaries have been deposited in the public Figshare data set (Tian, 2026; <https://doi.org/10.6084/m9.figshare.31841125>), allowing direct assessment of parameter

dependence. We also clarify that ensemble averaging does not remove this dependence and that the 9 calculations are not interpreted as independently validated local parameter sets. The detailed nine-member ensemble is shown under Follow-up comment 1 below.

Changes in the final manuscript and Supplement:

Section 2.3 was renamed “Calculation of $rPreNO_x$ and $rPrePO_4$.” Supplementary Method 2 now gives the full equations, explicitly imposes $f_{DOM} + f_{POM} = 1$, and reports the nine parameter combinations. The Introduction, Results, Discussion, Fig. 3, Supplementary Materials, and public dataset were updated consistently.

The revised method is as follows (Revised manuscript, lines 215-222; Supplementary materials, lines 37-64):

“2.3 Calculation of $rPreNO_x$ and $rPrePO_4$

$rPreNO_x$ and $rPrePO_4$ were calculated using the modified formulation of Letscher and Villareal (2018), with parameter sensitivity evaluated following Smyth and Letscher (2023). For each observation, 9 calculations were performed using the prescribed DOM and POM parameter combinations. The arithmetic mean was used as the reported diagnostic value, whereas the minimum, maximum, and standard deviation describe sensitivity to the prescribed parameters. Full equations and parameter values are provided in Supplementary Method 2.”

“**Supplementary Method 2.** Calculation of $rPreNO_x$ and $rPrePO_4$

$rPreNO_x$ was computed as:

$$rPreNO_x = NO_{xmeas} - (f_{DOM} \bullet \frac{AOU}{r_{DOM}} + f_{POM} \bullet \frac{AOU}{r_{POM}}) \quad (2),$$

where measured NO_x (NO_{xmeas}) is defined as the sum of the concentrations of NO_3 -N and NO_2 -N. AOU is given by:

$$AOU = DO_{sat} - DO_{meas} \quad (3),$$

where DO_{sat} is the saturated DO concentration at a given temperature, salinity, and pressure (Garcia and Gordon, 1992), and DO_{meas} is the measured DO concentration. f_{DOM} and f_{POM} represent the fractions of total oxygen consumption attributed to DOM and POM, respectively. r_{DOM} and r_{POM} denote the moles of O_2 consumed per mole of nitrogen regenerated during DOM and POM remineralization. To ensure mass balance of oxygen consumption partitioning, f_{DOM} and

f_{POM} were constrained as: $f_{DOM} + f_{POM} = 1$. Accordingly, three partitioning scenarios were considered ($f_{DOM}/f_{POM} = 0.4/0.6$, $0.5/0.5$, and $0.6/0.4$), and r_{POM} was assigned three values (6.9, 8.75, and 10.6), representing the minimum, Redfield, and maximum estimates reported in the literature (Table S2) (Letscher and Villareal, 2018; Smyth and Letscher, 2023). These combinations produced 9 calculations for each observation. These calculations were used to represent the uncertainty associated with applying empirical parameters to the broader Pacific Ocean. For each observation, the 9 calculations were summarized as minimum–maximum (arithmetic average $\pm$ standard deviation). The arithmetic average was used as the central diagnostic value in the manuscript, whereas the minimum, maximum, and standard deviation describe sensitivity to the prescribed parameter combinations. The complete summaries are available in the public data set (Tian, 2026).

Similarly, $rPrePO_4$ was computed as:

$$rPrePO_4 = PO_{4meas} - \frac{1}{16}(f_{DOM} \bullet \frac{AOU}{r_{DOM}} + f_{POM} \bullet \frac{AOU}{r_{POM}}) \quad (4)$$

Because the remineralization parameters specific to phosphorus could not be independently constrained, the nitrogen regeneration terms derived from DOM and POM were converted to phosphorus equivalents using an N:P ratio of 16:1 (Letscher and Villareal, 2018), solely as a common conversion reference.

Response Table 1/Revised Table S2. Parameters for the calculation of residual $PreNO_x/PrePO_4$ in the Pacific Ocean

Depth (m)/neutral density (γ_n)	f_{DOM}	f_{POM}	r_{POM}	r_{DOM}
0–100 m/ γ_n : 24.2–24.7	0.4, 0.5, 0.6	0.4, 0.5, 0.6	6.9, 8.75, 10.6	18.1
$\gamma_n > 24.7$	0.4, 0.5, 0.6	0.4, 0.5, 0.6	6.9, 8.75, 10.6	18.9

We also revised the Introduction to limit the interpretation of the residuals (Revised manuscript, lines 72-80):

“Because strong mixing can obscure local relationships between AOU and nutrient, residual nutrient diagnostics can be particularly useful in stratified oligotrophic systems, where water masses are more clearly resolved. Residual preformed total oxidised nitrogen ($rPreNO_x$) and

residual preformed phosphate ($r\text{PrePO}_4$) can be used to evaluate whether measured nutrient inventories are consistent with the regeneration predicted from AOU under parameterized dissolved organic matter (DOM) and particulate organic matter (POM) remineralization (Letscher and Villareal, 2018). Although dependent on the chosen parameters, these residuals can identify deviations from the preset relationship.”

In the revised Results, their distributions are reported objectively (Revised manuscript, lines 269-274):

“The ensemble mean $r\text{PreNO}_x$ and $r\text{PrePO}_4$ values were positive in the upper waters and generally increased with depth, reaching their highest values in the deep waters (Figs. 3d, e). Although the absolute magnitudes of the residuals varied among the 9 parameterizations, the main vertical pattern remained consistent across the parameter ensemble. The corresponding ranges and summary statistics are available in the public data set (Tian, 2026).”

In addition, we realized that that ensemble averaging does not eliminate the dependence of the residuals on the transferred empirical parameters based on the reviewers’ comments. Therefore, we have revised their role in the interpretation of our results. The low AOU/DIN and AOU/DIP slopes are derived directly from the measured AOU and nutrient concentrations and constitute the observational basis for the reported deviations in oxygen–nutrient coupling. The $r\text{PreNO}_3$ and $r\text{PrePO}_4$ distributions are used as complementary diagnostics of departures from the prescribed DOM and POM remineralization parameterizations, rather than as independent evidence for the TEP-related or microbial mechanisms proposed in the Discussion. In contrast, they are used together with the directly observed AOU–DIN and AOU–DIP relationships, TEP distributions, and microbial observations to interpret the processes associated with the stoichiometric deviations (Revised manuscript, lines 347-383):

*“The low AOU/DIN and AOU/DIP slopes in the upper and intermediate waters indicate greater nutrient accumulation per unit increase in AOU than expected from the classical reference ratios. Limited vertical nutrient supply maintains persistently oligotrophic conditions in the upper water column (Chl-*a* in the deep chlorophyll maximum: $0.19\text{--}0.31\ \mu\text{g L}^{-1}$). Under nutrient limitation, phytoplankton may allocate a relatively large proportion of photosynthetically fixed carbon to extracellular polysaccharides and TEP with high C:N ratios rather than to nutrient-rich cellular biomass (Smyth and Letscher, 2023; Curran et al., 2025). The observed TEP-C/POC ratios in the upper waters (mean of $43.81 \pm 14.62\%$) indicate that estimated TEP-associated*

carbon constituted a substantial component of the local POC pool (Fig. 3i, Table S1). Positive ensemble average $r\text{PreNO}_x$ and $r\text{PrePO}_4$ residuals occurred together with elevated TEP concentrations in the upper water column (Figs. 3d, e, and i). This indicates that measured nutrient concentrations were higher than predicted from AOU under the prescribed DOM and POM remineralization parameters, resulting in a lower AOU/nutrient ratio in the upper waters. Although standing TEP concentrations do not directly quantify production, the substantial contribution of TEP-associated carbon to the local POC pool and its co-occurrence with low AOU/nutrient ratios support carbon allocation to TEP as one plausible contributor to the observed deviations. Future studies should examine whether similarly low AOU/nutrient ratios occur in other ocean regions with comparably high TEP-C/POC ratios to assess the spatial generality of this association. Furthermore, evidence indicates that heterotrophic microorganisms preferentially consume components with low C:N, such as proteins, in organic matter during aerobic respiration (Hwang et al., 2006; Tian et al., 2025). Therefore, in the intermediate waters, preferential degradation of relatively nutrient rich and labile organic components may enhance nutrient regeneration relative to bulk organic carbon oxidation, thereby contributing to the low AOU/nutrient slopes (Fig. 5a, b). In the deep waters, the absence of significant AOU/nutrients relationships precludes the calculation of meaningful remineralization ratios. In this depth horizon, AOU remains relatively stable whereas nutrient concentrations continue to increase, suggesting that their distributions are governed primarily by the cumulative effects of water-mass aging, lateral transport, and diverse biogeochemical transformations operating on different timescales. Among these, nitrification mediated by ammonia-oxidizing archaea such as *Nitrosopumilus* may consume oxygen without concomitant phosphorus regeneration (Martens-Habbena et al., 2009), while DOP hydrolysis associated with *Alteromonas* may mobilize phosphate independently of bulk organic matter oxidation (Saavedra et al., 2025). Collectively, these processes likely contribute to the observed decoupling between AOU and nutrients in the deep ocean.”

2. Distinguishing whole-water-column observations from layer-specific mechanisms

According to the authors' own results, statistically significant AOU–nutrient relationships are observed only in the upper (0–300 m) and intermediate (300–1000 m) water masses, whereas no significant relationships exist below 1000 m ($p > 0.05$). I therefore recommend that the authors more clearly distinguish between the statistical patterns derived from the entire water column and the mechanisms operating within individual water masses.

If the proposed TEP-driven mechanism is intended to explain the low AOU/nutrient ratios throughout the entire water column, additional evidence is needed to demonstrate that its influence extends from the surface into the mesopelagic and deeper waters. Conversely, if the mechanism is primarily intended to explain the upper ocean, the corresponding statements in the Abstract, Discussion, and Conclusions should be moderated accordingly. In particular, a mechanism fundamentally linked to surface phytoplankton production should not be directly extrapolated to the deep ocean, especially where no statistically significant AOU–nutrient relationships are observed.

Furthermore, it should be noted that the production of TEP and the degradation of TEP are two distinct biogeochemical processes that are expected to have different, and potentially opposite, effects on AOU/nutrient stoichiometry. Current understanding suggests that TEP is produced predominantly within the euphotic zone by phytoplankton, whereas TEP observed in mesopelagic waters mainly represents exported material undergoing degradation rather than in situ production. The manuscript would therefore benefit from explicitly distinguishing between surface TEP production and subsurface TEP remineralization when discussing the mechanisms responsible for the observed AOU/nutrient ratios. This distinction is particularly important because TEP production and TEP degradation may influence AOU/nutrient stoichiometry in different directions.

Response: We sincerely thank the reviewer for this constructive comment. Based on the reviewers' suggestions, we differentiated the statistical relationships between AOU and nutrients and the mechanisms explaining these relationships according to water masses at different depths, rather than extrapolating to the entire water column. Specifically:

In the upper waters, where significant AOU to nutrient relationships and elevated TEP concentrations were observed, we now restrict the proposed mechanism to phytoplankton production of carbon rich TEP under chronic nutrient limitation. This process can decouple carbon fixation and oxygen production from nitrogen and phosphorus assimilation and thereby contribute to the low AOU to nutrient slopes in the upper ocean.

In the intermediate waters, we distinguish the transformation of exported organic matter from TEP production in the euphotic zone. Based on evidence that microorganisms preferentially degrade relatively labile substrates with lower C:N ratios (e.g., proteins) (Hwang et al., 2006; Tian et al., 2025), we propose that selective remineralization enhances nutrient release relative to oxygen consumption and contributes to the low AOU to nutrient slopes in these waters.

In deep waters, no significant AOU to nutrient relationships were observed. We therefore no longer extend the surface TEP mechanism to the deep waters. Instead, the deep water decoupling is discussed in relation to water mass aging, transport, and additional microbial and geochemical processes that affect oxygen consumption and nutrient accumulation.

Changes in the final manuscript and Supplement:

The Abstract, Discussion, Conclusions, and conceptual model in Fig. 7 were reorganized around these vertically distinct processes. Figure 5 now separates upper, intermediate, and deep hydrographic groups and labels the entire-water-column regressions separately.

Revised text in the Abstract (Revised manuscript, lines 14-33):

“In oligotrophic oceans, the stoichiometric ratios of apparent oxygen utilization (AOU) to nutrients often deviate from the classical Redfield ratio, yet the mechanisms remain poorly constrained. Contrary to the commonly held view that these ratios are typically elevated, we found that the mean ratios of AOU to dissolved inorganic nitrogen (DIN) and AOU to dissolved inorganic phosphorus (DIP) over the upper 2000 m of the oligotrophic Northwest Pacific are only 6.28 and 86.79, respectively, both below the classical values of 8.6 and 138. Strong stratification indicates that physical mixing alone cannot explain these integrated patterns, while depth-resolved regressions further reveal distinct controls among water layers. In upper waters, nutrient limitation promotes phytoplankton to produce TEPs with high C:N ratios, decoupling carbon fixation from nutrient assimilation, a process in which *Pelagibacter* may play a key role in recycling small organic molecules. In intermediate waters, preferential degradation of low C:N compounds within sinking organic matter enhances nutrient regeneration relative to oxygen consumption. In deep water, AOU/nutrient relationships are not statistically significant, indicating that oxygen consumption and nutrient accumulation become increasingly decoupled; this decoupling is likely influenced by *Alteromonas* activity in polymer degradation and phosphorus mobilization, alongside archaeal nitrification. These findings suggest that biogeochemical models should account for such biological feedbacks to improve predictions of ocean carbon export and nutrient cycling under future climate scenarios.”

Revised text in the Discussion (Revised manuscript, lines 341-383):

“To examine how these biogeochemical processes vary across water column, we calculated AOU/nutrient regression slopes separately for the upper, intermediate, and deep waters (Fig. 5).

The AOU/DIN value is lowest in the upper waters (6.87), increases in the intermediate waters (7.22), and becomes statistically insignificant in the deep waters ($p > 0.05$) (Fig. 5a). The AOU/DIP slopes is higher in the upper waters (116.04), decreasing in the intermediate waters (103.17), and non-significant in the deep waters (Fig. 5b). The low AOU/DIN and AOU/DIP slopes in the upper and intermediate waters indicate greater nutrient accumulation per unit increase in AOU than expected from the classical reference ratios. Limited vertical nutrient supply maintains persistently oligotrophic conditions in the upper water column (Chl-a in the deep chlorophyll maximum: $0.19\text{--}0.31\ \mu\text{g L}^{-1}$). Under nutrient limitation, phytoplankton may allocate a relatively large proportion of photosynthetically fixed carbon to extracellular polysaccharides and TEP with high C:N ratios rather than to nutrient-rich cellular biomass (Smyth and Letscher, 2023; Curran et al., 2025). The observed TEP-C/POC ratios in the upper waters (mean of $43.81 \pm 14.62\%$) indicate that estimated TEP-associated carbon constituted a substantial component of the local POC pool (Fig. 3i, Table S1). Positive ensemble average $r\text{PreNO}_x$ and $r\text{PrePO}_4$ residuals occurred together with elevated TEP concentrations in the upper water column (Figs. 3d, e, and i). This indicates that measured nutrient concentrations were higher than predicted from AOU under the prescribed DOM and POM remineralization parameters, resulting in a lower AOU/nutrient ratio in the upper waters. Although standing TEP concentrations do not directly quantify production, the substantial contribution of TEP-associated carbon to the local POC pool and its co-occurrence with low AOU/nutrient ratios support carbon allocation to TEP as one plausible contributor to the observed deviations. Future studies should examine whether similarly low AOU/nutrient ratios occur in other ocean regions with comparably high TEP-C/POC ratios to assess the spatial generality of this association. Furthermore, evidence indicates that heterotrophic microorganisms preferentially consume components with low C:N, such as proteins, in organic matter during aerobic respiration (Hwang et al., 2006; Tian et al., 2025). Therefore, in the intermediate waters, preferential degradation of relatively nutrient rich and labile organic components may enhance nutrient regeneration relative to bulk organic carbon oxidation, thereby contributing to the low AOU/nutrient slopes (Fig. 5a, b). In the deep waters, the absence of significant AOU/nutrients relationships precludes the calculation of meaningful remineralization ratios. In this depth horizon, AOU remains relatively stable whereas nutrient concentrations continue to increase, suggesting that their distributions are governed primarily by the cumulative effects of water-mass aging, lateral transport, and diverse biogeochemical transformations operating on different timescales. Among these, nitrification mediated by ammonia-oxidizing archaea such as *Nitrosopumilus* may

consume oxygen without concomitant phosphorus regeneration (Martens-Habben et al., 2009), while DOP hydrolysis associated with Alteromonas may mobilize phosphate independently of bulk organic matter oxidation (Saavedra et al., 2025). Collectively, these processes likely contribute to the observed decoupling between AOU and nutrients in the deep ocean.”

References:

- Hwang, J., Druffel, E. R. M., Eglinton, T. I., Repeta, D. J.: Source(s) and cycling of the nonhydrolyzable organic fraction of oceanic particles, *Geochim. Cosmochim. Acta.* 70(20), 5162-5168, <https://doi.org/10.1016/j.gca.2006.07.020>, 2006.
- Tian, D., Li, X., Song, J., Ma, J., Yuan, H., Duan, L.: Biogeochemical layering and transformation of particulate organic carbon in the Tropical Northwestern Pacific Ocean inferred from $\delta^{13}\text{C}$, *Ocean Sci.*, 21(4), 1627-1639, <https://doi.org/10.5194/os-21-1627-2025>, 2025.

Revised text in the Conclusions (Revised manuscript, lines 519-537):

“The oligotrophic tropical Northwest Pacific is characterized by pronounced vertical stratification, which retains nutrients at depth and sustains chronic nutrient limitation in the upper ocean. In surface waters, phytoplankton respond to nutrient stress by producing carbon rich transparent exopolymer particles, altering the balance among carbon fixation, oxygen production, and nutrient assimilation. In intermediate waters, selective transformation of labile and nutrient rich components promotes nitrogen and phosphorus regeneration while carbon rich polysaccharides persist longer. These depth dependent changes are accompanied by shifts in microbial function, with Pelagibacter contributing to the recycling of small organic molecules in upper waters and Alteromonas participating in polymer degradation and potentially in phosphorus regeneration through alkaline phosphatase activity at greater depths. In deep water, the absence of significant AOU to nutrient relationships indicates that nutrient accumulation is increasingly influenced by the aging of waters, additional microbial and geochemical processes. The low integrated AOU/DIN and AOU/DIP slopes of 6.28 and 86.79 therefore reflect the combined effects of distinct processes operating among waters. These findings show that vertical changes in organic matter production, export, and microbial transformation can substantially modify nutrient regeneration stoichiometry and should be considered when representing carbon and nutrient cycling in oligotrophic ocean models.”

3. Strength of the evidence linking TEP to the observed AOU/nutrient ratios

Another issue concerns the interpretation of the TEP observations. Based on the reported concentrations, the TEP levels observed in this study are generally within the low to moderately low range compared with those reported from the global ocean. Even within the oligotrophic western tropical North Pacific, the measured concentrations appear to be lower than those reported in previous studies, and they are substantially lower than those typically observed in productive coastal regions, frontal systems, or the surface Southern Ocean. Therefore, the statement that TEP-related process is responsible for the observed low AOU/nutrient ratios should be made with greater caution. In other words, the manuscript currently assumes that TEP abundance is the principal driver of the stoichiometric deviation, yet the observational evidence demonstrates only the presence of TEP rather than an exceptional accumulation relative to other oceanic regions. Therefore, the authors should better justify why relatively modest TEP concentrations are sufficient to produce measurable deviations in AOU/nutrient stoichiometry. Alternatively, the discussion could place greater emphasis on the quality, composition, or turnover of TEP, rather than its absolute concentration, as these characteristics may be more directly linked to remineralization stoichiometry than concentration alone.

Response: We thank the reviewer for this important comment. Although the TEP concentrations in the upper 0–300 m of our study area, ranging from 7.53 to 34.22 $\mu\text{g Xeq L}^{-1}$ with a mean of $22.52 \pm 8.54 \mu\text{g Xeq L}^{-1}$, were within the low range of published oceanic values. However, the relative contribution of TEP to the small particulate carbon pool was substantial. TEP-C accounted for 15.45–71.75% of POC, with a mean contribution of $43.81 \pm 14.62\%$, which was generally higher than those observed in most productive coastal, shelf, and open ocean environments. Thus, the relevant feature of our observations is not exceptional TEP accumulation in absolute terms, but the relatively large contribution of TEP to POC. To provide a more balanced assessment, we have added a new supplementary table (Table 2/Revised Table S1) comparing TEP concentrations and TEP-C/POC ratios from this study with those reported for oligotrophic open-ocean waters, productive coastal systems, frontal regions, and the surface Southern Ocean. All TEP-C/POC values were standardized using a conversion factor of 0.51 based on Engel and Passow (2001).

Accordingly, we have revised the description of TEP absolute abundance in the Introduction (Revised manuscript, lines 98-100):

“Although their absolute concentrations are often low in oligotrophic oceans, TEP can constitute a substantial fraction of the small POC pool and can be abundant relative to the low phytoplankton biomass (Kodama et al., 2014).”

And we have revised the Results to include a description of the TEP-C/POC ratio (Revised manuscript, lines 269-274, 290-300):

“The ensemble mean rPreNO_x and rPrePO₄ values were positive in the upper waters and generally increased with depth, reaching their highest values in the deep waters (Figs. 3d, e). Although the absolute magnitudes of the residuals varied among the 9 parameterizations, the main vertical pattern remained consistent across the parameter ensemble. The corresponding ranges and summary statistics are available in the public data set (Tian, 2026).”

“...POC and TEP exhibit a distribution pattern that is nearly opposite to that of nutrients and DIC (Figs. 3g, h, i). TEP concentrations in the upper water ranged from 7.53 to 34.22 $\mu\text{g Xeq L}^{-1}$ (mean $22.52 \pm 8.54 \mu\text{g Xeq L}^{-1}$), exceeding those in deeper waters. These concentrations were within the low range reported for open ocean environments (Table S1). After conversion to carbon equivalents using a factor of 0.51 (Engel and Passow, 2001), the TEP-C/POC ratio ranged from 15.45% to 71.75%, with a mean of $43.81 \pm 14.62\%$. The mean ratio was commonly higher than those observed in eutrophic oceanic regions (Response Table 2/Revised Table S1). POC concentrations decrease rapidly in the intermediate and deep waters, while the AOU continuously increased (Figs. 3f, g). In contrast, TEP concentrations decrease more slowly and remain within a measurable range even in the deep waters (Fig. 3i).”

Reference:

Engel, A., Passow, U.: Carbon and nitrogen content of transparent exopolymer particles (TEP) in relation to their Alcian Blue adsorption, Mar. Ecol. Prog. Ser., 219, 1-10, <https://doi.org/10.3354/meps219001>, 2001.

Response Table 2/Revised Table S1. The review of the surface POC concentrations, TEP concentrations and TEP-C/POC ratios in the literature.

Region	Depth	POC ($\mu\text{mol L}^{-1}$)	TEP ($\mu\text{g Xeq L}^{-1}$)	^b TEP-C/POC (%)	Reference
^a Tropical Northwest Pacific	Upper Water	1.13–3.22 (2.27 ± 0.53)	7.53–34.22 (22.52 ± 8.54)	15.45–71.75 (43.81 $\pm$ 14.62)	This study
Fram Strait	5–150 m	^c 23.10	5–517 (75 ± 78)	13.8 ± 6.1	Engel et al. (2017)
Northern South China Sea	0–200 m	0.20–4.38	0.6–78.6	5.6–55.5 (27.4 ± 1.3)	Ge et al. (2022)
^a South China Sea	0–220 m	^c 1.70	14–47 (30 ± 8)	32.0–173 (75.0 ± 37.0)	Guo et al. (2022)
^a Western tropical Pacific	0–220 m	^c 2.37	12–54 (34 ± 10)	45.0–114 (61.0 ± 40.0)	Guo et al. (2022)
Jaran Bay	Subsurface	9.15–100.07 (33.23 $\pm$ 15.53)	26.5–1695.4 (215.9 ± 172.2)	2.4–67.3 (21.7 ± 11.4)	Lee et al. (2020)
East China Sea coast	Surface	6.99–76.08 (30.20)	11.25–509.58 (214.90)	30.2	Wen et al. (2022)
North of the South Orkney Islands	Surface	13.8 ± 2.6	144.4 ± 21.7	45.9 ± 5.1	Zamanillo et al. (2019a)
South of the South Orkney Islands	Surface	5.5 ± 1.2	48.1 ± 6.5	40.8 ± 10.2	Zamanillo et al. (2019a)
Northwest of South Georgia	Surface	18.0 ± 4.4	125.5 ± 21.1	30.6 ± 5.1	Zamanillo et al. (2019a)
West of Anvers Island	Surface	19.3 ± 3.5	111.6 ± 13.0	25.5 ± 5.1	Zamanillo et al. (2019a)
^a Open Atlantic Ocean	Surface	1.7–7.1 (4.2 ± 1.9)	18.3–131.7 (59.8 ± 27.4)	34.0–103 (66.0 ± 19.0)	Zamanillo et al. (2019b)
Southwestern Atlantic Shelf	Surface	6.8–44.3 (16.6 ± 15.8)	98.6–427.2 (255.7 ± 130.4)	28.0–110 (73.0 ± 36.0)	Zamanillo et al. (2019b)

^aOligotrophic ocean

^bA factor of 0.51 was used to convert TEP ($\mu\text{g Xeq L}^{-1}$) to TEP-C ($\mu\text{g C L}^{-1}$)

^cCalculated from the reported mean TEP concentration and TEP-C/POC ratio

References:

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We have also restructured the Discussion to address the causes of deviations in AOU/nutrient stoichiometry separately for different depth layers, thereby avoiding any implication that TEP abundance alone is the primary driver (See the reply to major comment 2 for details).

Minor or specific comments

1. The reported analytical precision of the POC measurements deserves further clarification. In general, the instrumental precision of an elemental analyzer for carbon determination is typically on the order of approximately 1% under routine laboratory conditions. For real samples, however, the overall analytical precision is usually lower because additional uncertainties are introduced during sample collection, filtration, acidification (or acid fumigation), drying, and other pretreatment procedures. Therefore, the reported analytical precision of 0.8‰ appears to be exceptionally high—nearly two orders of magnitude better than the analytical precision commonly achieved in routine laboratory measurements of marine suspended particulate organic carbon. I am therefore curious about how this level of precision was achieved. Does this value refer to the repeatability of the elemental analyzer itself, the precision of repeated analyses of laboratory standards, or the overall analytical precision including sample pretreatment? Alternatively, could this simply be a typographical error? I recommend that the authors clarify how this value was determined and what it specifically represents.

Response: We thank the reviewer for identifying this issue. After reexamining the analytical procedure and the cited methods, we found that the reported value of $\pm 0.8\text{‰}$ was inappropriately expressed and did not represent the overall precision of POC determination. Because full process replicate field samples were unavailable, the overall method precision, including uncertainties associated with filtration, acid treatment, drying, and instrumental measurement, could not be independently quantified. We have therefore removed the $\pm 0.8\text{‰}$ value. The revised Methods are as follows (Revised manuscript, lines 180-185):

“The pretreated samples were analyzed for POC concentration using an elemental analyzer (Thermo Fisher Scientific Flash EA 1112, United States). Carbon quantification was calibrated using standard reference materials, including USGS64 (C % = 31.97 %, Indiana University), USGS40 (C % = 40.8 %, U.S. Geological Survey), and Urea #2a (C % = 20 %, Indiana University), which were also used to monitor instrument performance (Ma et al., 2021b).”

2. The description of the TEP analytical method requires clarification. The current protocol omits several key steps of the standard Alcian Blue assay (e.g., staining and washing), making it unclear whether the standard method was actually followed or whether these procedures were inadvertently omitted from the manuscript. In addition, the authors should clarify whether the xanthan gum calibration curve was established under their own experimental conditions or directly adopted from Bittar et al. (2018), as calibration curves are generally expected to be generated within each laboratory to ensure quantitative reliability.

Response: We thank the reviewer for pointing out these important issues regarding the TEP analytical method. The staining and rinsing steps were performed during sample collection and were described in Sect. 2.1 (original manuscript, lines 139-144): *“TEP samples were collected by filtering 1–4 L of seawater through polycarbonate filters (0.45 μm pore size, Whatman) under a vacuum of 170 mmHg. The filtered membrane was stained with 0.5–1 mL of a 400 mg L⁻¹ Alcian Blue solution (Sigma-Aldrich, USA) for 5–10 s, rinsed with 5 mL of MilliQ water, and stored at -20 °C until measurement (three replicate samples were collected at each depth).”* Because the sampling procedure and the laboratory analysis were presented in separate subsections, the complete method may not have been immediately evident. In addition, the pH of the Alcian Blue solution was omitted from the original manuscript. We have now added that the staining solution was adjusted to pH 2.5 with glacial acetic acid. This information has now been added to the revised Methods (Revised manuscript, lines 146-149):

“The retained particles were stained with 0.5–1 mL of a 400 mg L⁻¹ Alcian Blue solution (Sigma-Aldrich, USA) for 5–10 s, prepared in Milli Q water and acidified to pH 2.5 with glacial acetic acid. The filters were then rinsed with 5 mL of MilliQ water, and stored at -20 °C until measurement (three replicate samples were collected at each depth).”

Regarding the calibration curve, we appreciate the reviewer's concern. Our original wording may have inadvertently implied that we directly adopted the calibration curve from Bittar et al. (2018). In fact, the updated procedure described by Bittar et al. (2018) was used only as a methodological reference for establishing the xanthan gum calibration curve, and the calibration curve was independently generated in our laboratory under our own experimental conditions. To avoid any ambiguity, we have revised the TEP analysis subsection to explicitly state that the xanthan gum calibration curve was prepared specifically for this study following the protocol of Bittar et al. (2018), with the absorbance converted to xanthan gum equivalents (Xeq) weight using this

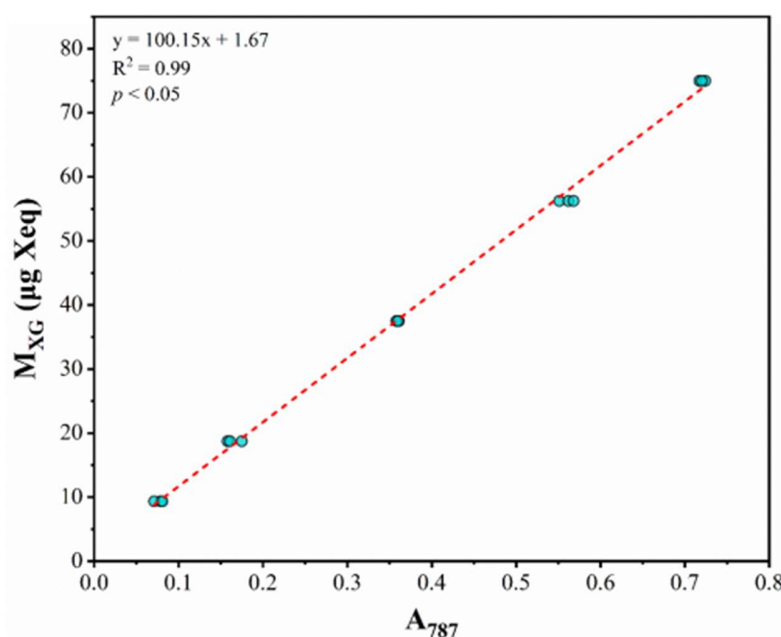
laboratory-generated calibration curve (Response Figure 1/Revised Figure S1). The relevant calibration curve and supplementary method description are detailed in the supplementary materials (Supplementary material, lines 19-35):

“Supplementary Method 1. Preparation of the xanthan gum calibration curve

A fresh xanthan gum stock solution (75 mg L⁻¹) was diluted to obtain five standards. Aliquots of 0.125, 0.250, 0.500, 0.750, and 1.000 mL of the stock solution were pipetted into clean 5 mL polypropylene tubes, and the final volume of each standard was adjusted to 1 mL with ultrapure water. This produced standards containing 9.37, 18.75, 37.50, 56.25, and 75.00 µg xanthan gum, respectively, with three replicates at each level. Each standard was mixed with 0.5 mL of Alcian Blue solution (400 mg L⁻¹, pH 2.5) for 5–10 s and filtered through a 0.45 µm polycarbonate membrane (Whatman) under a vacuum below 175 mmHg. Procedural blanks were processed in parallel. The filters were extracted with 6 mL of 80% sulfuric acid for 10 h, and absorbance was measured at 787 nm. A linear regression of xanthan gum mass against absorbance after blank correction yielded:

$$M_{XG} = 100.15A_{787} + 1.67 \quad (1)$$

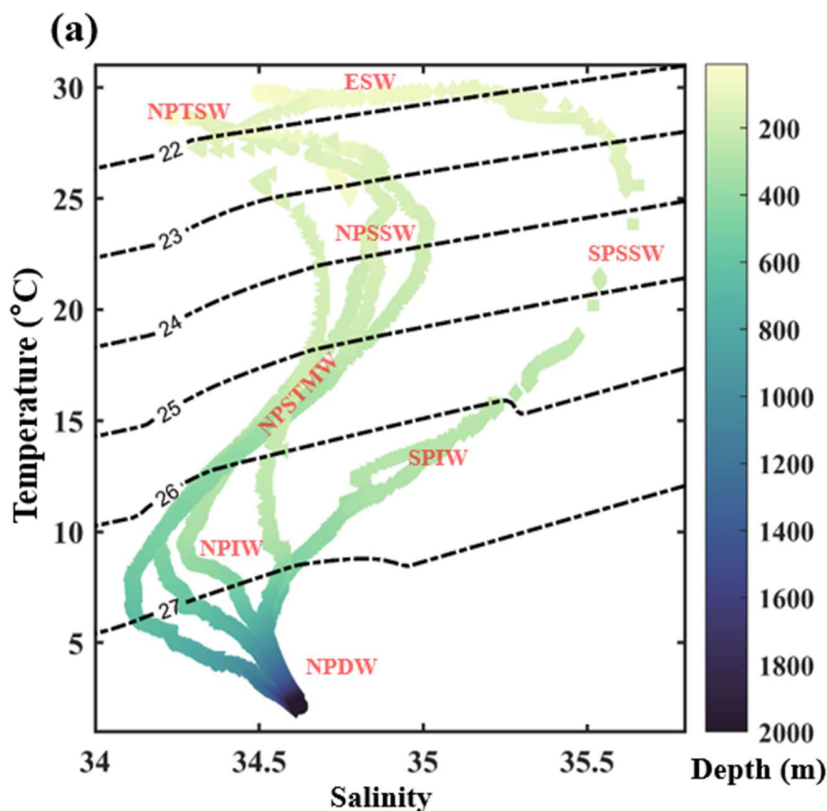
where M_{XG} is the xanthan gum equivalent mass in µg and A_{787} is the absorbance at 787 nm ($R^2=0.99$; Fig. S1). TEP concentrations were calculated by dividing the xanthan gum equivalent mass by the filtered seawater volume and are reported as µg Xeq L⁻¹.



Response Figure 1/Revised Figure S1. Xanthan gum calibration curve for TEP quantification.”

3. What do the red arrows mean in fig 2a?

Response: We thank the reviewer for pointing out this ambiguity. In the original Fig. 2a, the red arrows with horizontal lines were intended to indicate the boundaries distinguishing different water masses. However, we recognize that this representation was not sufficiently clear and could cause confusion. To improve readability, we have revised the figure by removing the arrows, as the T-S diagram itself, with its characteristic clustering of data points, already allows for clear identification of each water mass without additional markings. We believe this modification makes the figure cleaner and easier for readers to interpret (Response Figure 2/Revised Figure 2a).



Response Figure 2/Revised Figure 2a. Temperature-salinity diagram of six stations.

Response to Referee #1 — Follow-up Review

Overall assessment

The authors have carefully considered my comments and have made substantial and generally appropriate revisions to the manuscript. In particular, the revised manuscript now distinguishes the

mechanisms operating in the upper, intermediate, and deep waters, clarifies the diagnostic rather than independent evidential role of residual preformed nutrients, and provides additional methodological information and literature comparisons for TEP. These revisions have substantially improved the clarity and balance of the manuscript.

I consider the manuscript suitable for publication after the following minor revisions.

Response: We thank the reviewer for the positive assessment and for the two focused suggestions. Both points were incorporated into the integrated final revision.

1. The f_{DOM} and f_{POM} issue

f_{DOM} , f_{POM} are defined as the fractions of total oxygen consumption attributable to DOM and POM remineralization, respectively. Therefore, I think these two fractions should satisfy the mass-balance constraint: $f_{DOM} + f_{POM} = 1$.

If this is the case and I am right, then the current sensitivity analysis varies f_{DOM} and f_{POM} independently among 0.4, 0.5, and 0.6, producing 27 parameter combinations. Some of these combinations assign less or more than 100% of the total oxygen consumption to DOM and POM together and are therefore not physically consistent.

I recommend recalculating $rPreNO_3$ and $rPrePO_4$ under the above constraint. The corresponding means, ranges, and standard deviations should be updated in the manuscript, supplementary material, figures, and public dataset as necessary. The authors should also confirm whether the principal vertical patterns and interpretations remain unchanged after this recalculation.

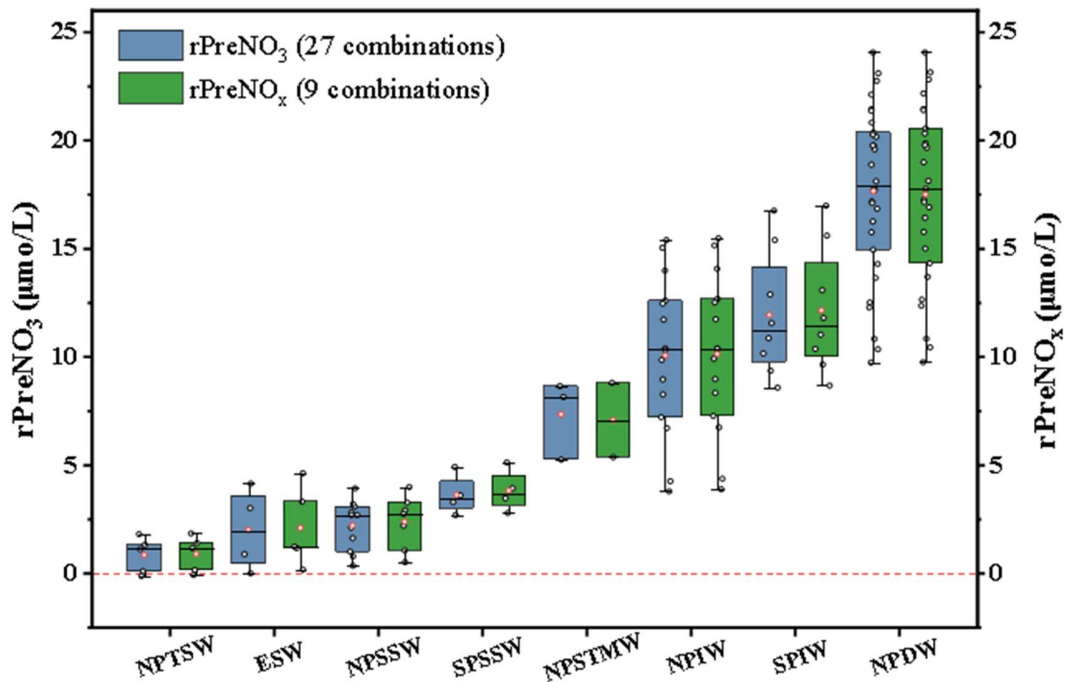
Response: We thank the reviewer for pointing out this important issue. Following the reviewer's suggestion, we recalculated $rPreNO_3$ and $rPrePO_4$ using nine physically constrained parameter combinations, including three DOM–POM partitioning scenarios ($f_{DOM}/f_{POM} = 0.4/0.6$, $0.5/0.5$, and $0.6/0.4$) combined with three r_{POM} values (6.9, 8.75, and 10.6) (Response Table 1/Revised Table S2). The constraint of $f_{DOM} + f_{POM} = 1$ has now been explicitly stated in the calculation method (Supplementary material, lines 47-58).

“To ensure mass balance of oxygen consumption partitioning, f_{DOM} and f_{POM} were constrained as: $f_{DOM} + f_{POM} = 1$. Accordingly, three partitioning scenarios were considered ($f_{DOM}/f_{POM} = 0.4/0.6$, $0.5/0.5$, and $0.6/0.4$), and r_{POM} was assigned three values (6.9, 8.75, and 10.6), representing the minimum, Redfield, and maximum estimates reported in the literature (Table S2) (Letscher and Villareal, 2018; Smyth and Letscher, 2023). These combinations

produced 9 calculations for each observation. These calculations were used to represent the uncertainty associated with applying empirical parameters to the broader Pacific Ocean. For each observation, the 9 calculations were summarized as minimum–maximum (arithmetic average $\pm$ standard deviation). The arithmetic average was used as the central diagnostic value in the manuscript, whereas the minimum, maximum, and standard deviation describe sensitivity to the prescribed parameter combinations. The complete summaries are available in the public data set (Tian, 2026).”

The recalculated ensemble mean values of $r\text{PreNO}_3$ and $r\text{PrePO}_4$ showed no differences from the previous calculations. This is because the original parameter space was centered around $f_{\text{DOM}} = 0.5$ and $f_{\text{POM}} = 0.5$, and thus the mean remineralization coefficients remained unchanged after applying the mass balance constraint. However, the standard deviations and ranges were reduced because the revised calculation excluded parameter combinations that did not satisfy the mass balance constraint (See the attached Table. 1 for details).

We also revised the definition of the measured nitrate term in the calculation. Specifically, $\text{NO}_{3\text{meas}}$ has been changed to $\text{NO}_{x\text{meas}}$, representing the sum of nitrate and nitrite concentration, because the calculation considers the total inorganic nitrogen regenerated during remineralization. Nitrite concentrations were very low and therefore had negligible effects on the calculated $r\text{PreNO}_x$ values (Response Figure 3).



Response Figure 3. Comparison of $r\text{PreNO}_3$ and $r\text{PreNO}_x$ estimates across different water masses. $r\text{PreNO}_3$ was calculated using the original 27 combinations, whereas $r\text{PreNO}_x$ was recalculated using the 9 mass-balance constrained combinations and $\text{NO}_{x\text{meas}}$ (nitrate + nitrite).

Importantly, the recalculated results confirmed that the depth distributions, relationships with AOU and nutrients, and associated interpretations were consistent with the previous results, indicating that the main conclusions are robust to the revised mass balance constraint.

We have updated the manuscript, Supplementary Materials, figures, and public dataset accordingly to reflect the revised mass-balance constrained calculations.

2. Interpretation of TEP abundance

The authors have appropriately acknowledged that the absolute TEP concentrations observed in this study fall within the relatively low range reported for oceanic environments. The newly added TEP-C/POC comparison is useful because it shows that TEP-associated carbon represents a substantial fraction of the relatively small POC pool in the study region.

Nevertheless, Table S1 also shows that the TEP concentrations and TEP-C/POC ratios reported here are not among the highest values observed globally. In particular, comparable or higher TEP-C/POC ratios have been reported from some other sites (Table S1). Therefore, statements such as the TEP-C/POC ratio being “generally higher than those observed in most productive coastal, shelf, and surface open-ocean environments” should be moderated.

I personally think a more accurate interpretation would be that the absolute TEP concentration is not exceptionally high on a global scale, but that TEP constitutes a substantial component of the local POC pool under the oligotrophic conditions of the study region. This relative importance makes TEP production a plausible contributor to the observed upper-ocean AOU/nutrient deviations, although it does not necessarily demonstrate that TEP is the sole or dominant driver. Eventually, further study is needed to reveal or confirm this point--particularly to check whether similar abnormal AOU/nutrient ratios are present in those regions with similarly high TEP/POC ratios. This call for further study should also be included in the manuscript.

Response: We thank the reviewer for the careful suggestion regarding the interpretation of TEP abundance. Our original intention in the Results section was to convey that the observed TEP-C/POC ratios were higher than those reported for most eutrophic oceanic regions (i.e., sites without

“*” in Response Table 2/Revised Table S1). However, we recognize that our original phrasing may overstated the global significance of the TEP-C/POC ratios. Therefore, we have revised the Results section to a more objective and precise statement (Revised manuscript, lines 294-297):

“After conversion to carbon equivalents using a factor of 0.51 (Engel and Passow, 2001), the TEP-C/POC ratio ranged from 15.45% to 71.75%, with a mean of $43.81 \pm 14.62\%$. The mean ratio was commonly higher than those observed in eutrophic oceanic regions (Table S1).”

In the Discussion section, we have clarified that while absolute TEP concentrations are not globally exceptional, TEP-C/POC ratios indicate that TEP-associated carbon constitutes a substantial component of the local POC pool under oligotrophic conditions. We have also acknowledged that the co-occurrence of elevated TEP and low AOU/nutrient ratios supports TEP production as a plausible contributor, but not necessarily the sole or dominant driver, and we have added the recommended call for future studies to examine whether similar AOU/nutrient anomalies occur in other regions with comparably high TEP-C/POC ratios (Revised manuscript, lines 354-367).

“The observed TEP-C/POC ratios in the upper waters (mean of $43.81 \pm 14.62\%$) indicate that estimated TEP-associated carbon constituted a substantial component of the local POC pool (Fig. 3i, Table S1). Positive ensemble average $rPreNO_x$ and $rPrePO_4$ residuals occurred together with elevated TEP concentrations in the upper water column (Figs. 3d, e, and i). This indicates that measured nutrient concentrations were higher than predicted from AOU under the prescribed DOM and POM remineralization parameters, resulting in a lower AOU/nutrient ratio in the upper waters. Although standing TEP concentrations do not directly quantify production, the substantial contribution of TEP-associated carbon to the local POC pool and its co-occurrence with low AOU/nutrient ratios support carbon allocation to TEP as one plausible contributor to the observed deviations. Future studies should examine whether similarly low AOU/nutrient ratios occur in other ocean regions with comparably high TEP-C/POC ratios to assess the spatial generality of this association.”

Response to Referee #2

Overall assessment

The manuscript by Tian et al. studies the low AOU to nutrient (DIN and DIP) ratios in the oligotrophic tropical Northwest Pacific by combining hydrographic, biogeochemical and microbial community analysis. They found consistently low AOU/DIN and AOU/DIP ratios compared to Redfield ratios, together with high TEP concentrations in the upper water column and depth-dependent shifts. The results indicate that variations in TEP abundance and the functional composition of microbial communities are closely associated with nutrient regeneration in their working area. Therefore, the manuscript is of interest as it improves the understanding of how microbial processes and organic matter regulate nutrient regeneration and carbon cycling in the ocean.

The dataset is of high quality, and the main findings are robust and supported by the analyses presented. However, major revisions are necessary to better explain certain methodological aspects and to improve the organization and terminology of the results section.

Response: We sincerely thank the reviewer for the thoughtful and constructive assessment. The comments helped us clarify the analytical hierarchy, improve methodological transparency, reorganize the figures around vertical profiles, and delimit the mechanistic interpretation more carefully. The final integrated revisions are described below.

General remarks

1. The calculation of residual PreNO_3 and PrePO_4 relies on fixed DOM/POM remineralization stoichiometries adopted from previous studies, while the main conclusion of the manuscript is that Redfield-type stoichiometric relationships break down in the study region. The authors should discuss the consistency of applying fixed remineralization ratios under these conditions and clarify the uncertainty associated with this assumption.

Response: We appreciate this important comment. The anomalous residuals derived from the reference remineralization ratios indeed motivated us to question the applicability of Redfield-type relationships in this region; however, we do not treat these residuals as independent lines of evidence. Our primary conclusions are instead drawn from regressions based directly on measured AOU and nutrient concentrations. While fixed stoichiometric ratios were employed in the calculations, we systematically evaluated 9 parameter combinations to assess the sensitivity of our

results (Response Table 1), with the resulting values and ranges fully reported in the figures and accompanying public datasets. In the revised manuscript, we have made it clearer that rPreNOx and rPrePO4 are diagnostic residuals that depend on the chosen parameters, and that their spread primarily reflects parameter sensitivity rather than measurement uncertainty.

Changes in the final manuscript and Supplement:

Section 2.3, Supplementary Method 2, Fig. 3, and the associated Results and Discussion were revised consistently. Supplementary Method 2 explicitly imposes $f_{\text{DOM}} + f_{\text{POM}} = 1$, lists the three paired fraction scenarios and three r_{POM} values, and states that the mean, range, and standard deviation describe sensitivity to the prescribed parameters. The 16:1 conversion used for rPrePO4 is identified solely as a common reference.

For the revised methodology, please refer to our reply to Reviewer 1's Major Comment 1.

2. Throughout the manuscript, I was occasionally confused about the level at which the reported ratios are calculated and discussed. In some sections, the text refers to ratios for specific water masses, whereas in others it refers to the three predefined depth intervals (0-300 m, 300-1000 m, and >1000 m). However, the terminology is not always used consistently, and at times the depth intervals appear to be discussed as if they were distinct water masses. This makes it difficult to distinguish whether a given result relates to a hydrographically defined water mass, a depth-based grouping, or the entire water column. I encourage the authors to carefully review the manuscript and use consistent terminology throughout. In particular, the distinction between water masses and depth intervals should be made explicit at all times. These represent fundamentally different ways of grouping the data, yet the distinction between them is not always clear in the current text, which can make the results difficult to interpret.

More broadly, I am not fully convinced that the current presentation of the results is the most effective way to communicate the findings. Much of the Results section is structured around analyses of the individually identified water masses, yet the Discussion predominantly focuses on the three depth intervals. This raises the question of what additional insight is gained from the detailed water-mass-specific analyses if the subsequent interpretation is largely based on the depth-based grouping. The rationale and added value of the water-mass analysis therefore deserve further clarification. As an alternative, I would suggest retaining the T-S diagram to define and visualize the identified water masses, but reconsidering the presentation of Figures 2 and 3. Rather than

organizing these results by individual water masses, depth-profile figures of the relevant parameters might provide a clearer overview of vertical gradients and stratification while remaining more closely aligned with the depth-interval framework that is ultimately emphasized in the Discussion. Such an approach could also help avoid some of the terminology issues arising from the current mixture of water-mass-based and depth-based analyses.

Response: We thank the reviewer for identifying this ambiguity and for suggesting a presentation that is more closely aligned with the interpretive framework. We have therefore addressed them together through a manuscript-wide reorganization of the grouping terminology, data presentation, and associated interpretation.

In the revised manuscript, the eight named water masses are retained to establish the hydrographic setting and define the identity of each observation. All subsequent grouped data summaries, regression analyses, and interpretations are organized using three hydrographic groups: upper waters (NPTSW, ESW, NPSSW, SPSSW, and NPSTMW; sampled depths 5-400 m), intermediate waters (NPIW and SPIW; sampled depths 300-1000 m), and deep waters (NPDW; sampled depths 1000-2000 m). Group membership is assigned from the identified water mass, supported by the T-S properties and vertical position of the samples, rather than from depth alone.

These three hydrographic groups are therefore not identical to the fixed depth intervals of 0-300 m, 300-1000 m, and >1000 m. In particular, the upper hydrographic group includes some samples collected at 300-400 m because they were identified as NPSTMW. We now reserve the term “water mass” for the eight named hydrographic entities, use “upper waters” “intermediate waters” and “deep waters” for the three aggregated hydrographic groups, use “fixed depth interval” only for the sensitivity analysis, and use “entire water column” only when all observations are analyzed together.

To determine whether the choice of grouping affected the results, we repeated all regressions using the fixed depth intervals 0-300 m, 300-1000 m, and >1000 m and compared those slopes with the slopes obtained from the three hydrographic groups. Paired row-bootstrap comparisons with 4,999 resamples retained the correspondence between the same observations classified under the two schemes, and the resulting p values were adjusted using the Benjamini-Hochberg procedure. No slope difference was significant after correction for AOU-DIN, AOU-DIP, AOU-SiO₃-Si, AOU-DIC, or DIN-DIP in any of the three groups (Response Table 3; BH adjusted $p = 0.08$ – 1.00).

This agreement shows that the central conclusions are not an artifact of either grouping scheme. We finally use the hydrographic groups as the primary framework because they preserve information from the identified water masses and provide a physically meaningful basis for grouping the observations

Response Table 3. Comparison of hydrographic-group and fixed-depth slopes using all observations.

Response– predictor	Vertical Layer	Water- mass group slope	Fixed- depth slope	BH- adjusted p	Different at 0.05
AOU–DIN	Upper	6.87	6.74	0.89	No
	Intermediate	7.22	7.58	0.89	No
	Deep	-0.90	-1.43	0.89	No
AOU–DIP	Upper	116.04	107.14	0.70	No
	Intermediate	103.17	109.45	0.70	No
	Deep	-1.86	-6.81	0.70	No
AOU–SiO ₃ - Si	Upper	3.70	5.50	0.24	No
	Intermediate	1.47	1.49	0.91	No
	Deep	0.00	-0.02	0.91	No
AOU–DIC	Upper	0.37	0.66	0.08	No
	Intermediate	0.86	0.73	0.19	No
	Deep	0.05	0.05	1.00	No
DIN– DIP	Upper	17.89	16.26	0.61	No
	Intermediate	11.08	11.03	0.95	No
	Deep	7.79	8.36	0.95	No

Changes in the final manuscript and Supplement:

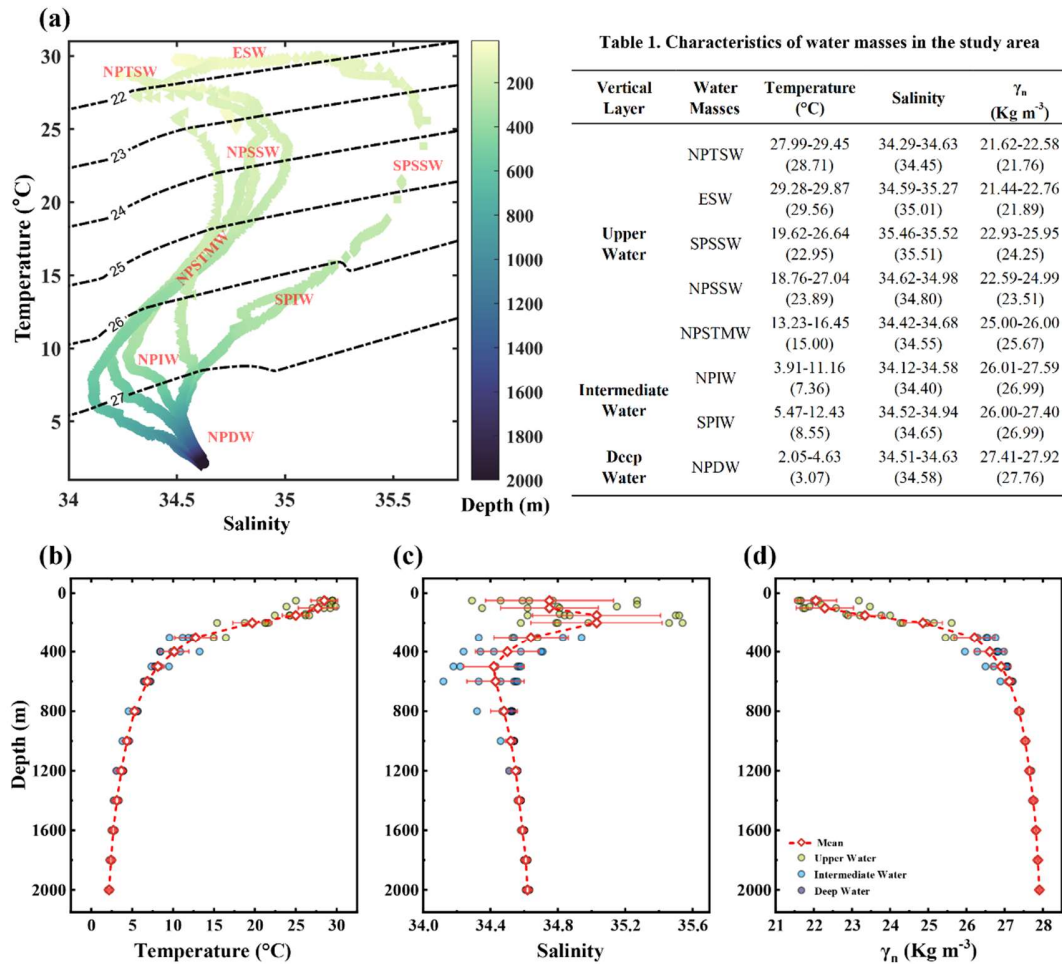
Figures 2 and 3 were converted to hydrographic and biogeochemical depth-profile presentations; Fig. 4 now summarizes microbial genera for the three hydrographic groups; Fig. 5 presents all regressions consistently for those groups and labels entire-water-column fits separately; and Fig. 7 uses the same three-group framework. The Results and Discussion now follow this organization throughout. The individually named water masses provide hydrographic context rather than a parallel biogeochemical analysis.

The revised results are as follows (Revised manuscript, Lines 233-319):

“3. Results

3.1 Hydrological characteristics of the study area

Temperature-salinity (T-S) diagram and the vertical distribution of temperature, salinity, and γ_n indicate pronounced stratification in the water column. Eight distinct water masses were identified and subsequently categorized into upper, intermediate, and deep waters based on their hydrographic properties and vertical positions (Fig. 2a, Table 1) (Sun et al., 2008). γ_n increases with depth from approximately 21.6 to 27.7 kg m⁻³ (Fig. 2d), reflecting the transition from low-density tropical upper waters to high-density deep waters.



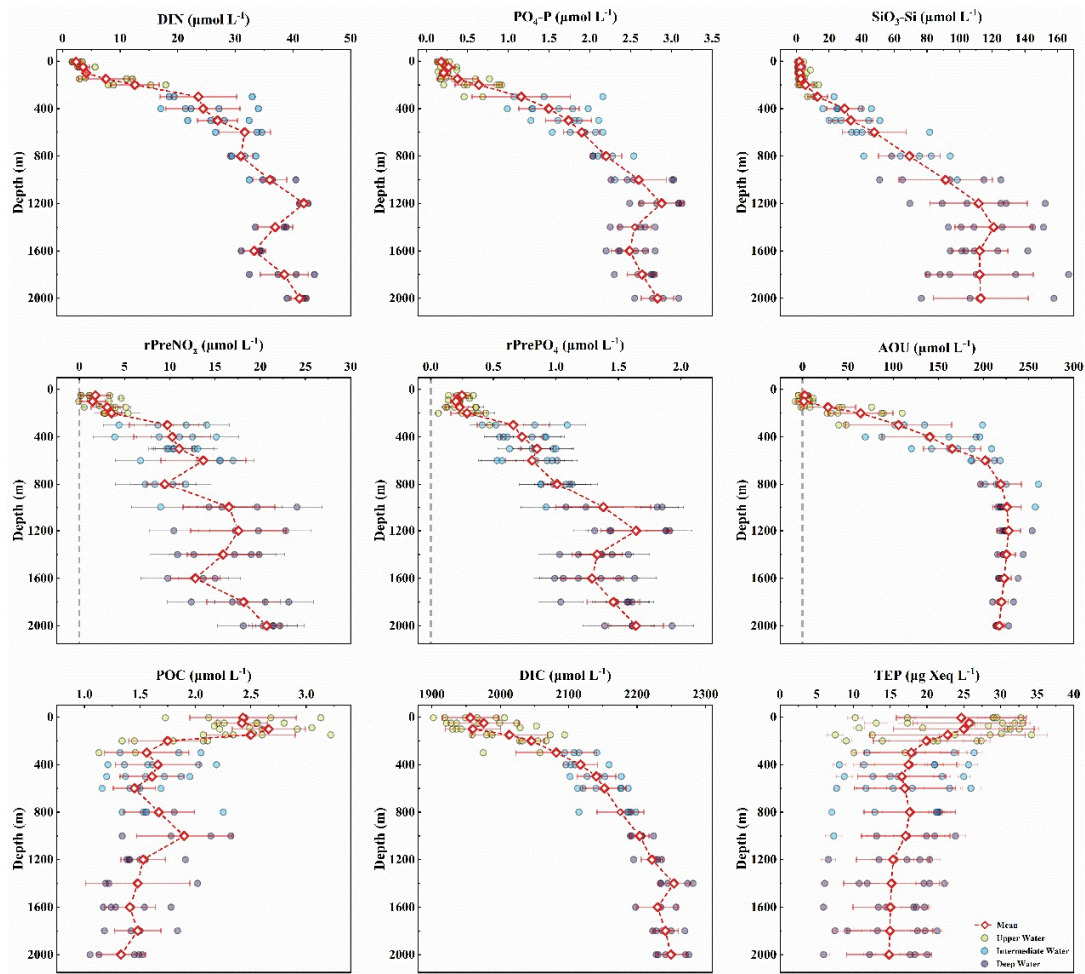
Response Figure 4/Revised Figure 2. Hydrographic characteristics of the water column.
(a) Temperature-salinity diagram of six stations, and the vertical profiles of (b) temperature,
(c) salinity, (d) neutral density (γ_n). Red diamonds connected by dashed lines and their

horizontal error bars denote the mean $\pm$ standard deviation of all samples collected at each nominal sampling depth.

In the upper waters, tropical surface waters and subsurface waters dominate. The North Pacific Tropical Surface Water (NPTSW) and Equatorial Surface Water (ESW) lie at the warmest end of the T-S diagram, with temperatures ranging from approximately 28–30 °C, characteristics of the WPWP. Below the mixed layer, the North Pacific Subsurface Water (NPSSW) and South Pacific Subsurface Water (SPSSW) exhibit pronounced salinity maxima along the 23–25 kg m⁻³ isopycnals. Both water masses have temperatures lower than the overlying surface waters (approximately 19–27 °C). The salinity of the SPSSW is the highest in the upper ocean (mean 35.51), while the NPSSW is slightly fresher (mean 34.80) (Fig. 2a, Table 1). The North Pacific Subtropical Mode Water (NPSTMW) constituted the lower part of the upper water, with stable physicochemical properties. The intermediate waters comprised the North Pacific Intermediate Water (NPIW) and South Pacific Intermediate Water (SPIW). The NPIW is characterized by low salinity (mean 34.12), exhibiting a salinity minimum in the T-S diagram (Fig. 2a). The SPIW has slightly higher temperature and salinity relative to NPIW. Its γ_n value largely overlaps with those of the NPIW, suggesting substantial mixing between the two within the study area. The deep water was represented by North Pacific Deep Water (NPDW), which had low temperatures, relatively uniform salinity, and high density (Figs. 2b-d, Table 1).

3.2 Vertical distribution of chemical parameters across water column

DIN, phosphate, and silicate concentrations were low in the upper waters and increased markedly with depth, reaching their maxima in the deep waters (Figs. 3a–c). The ensemble mean $r\text{PreNO}_x$ and $r\text{PrePO}_4$ values were positive in the upper waters and generally increased with depth, reaching their highest values in the deep waters (Figs. 3d, e). Although the absolute magnitudes of the residuals varied among the 9 parameterizations, the main vertical pattern remained consistent across the parameter ensemble. The corresponding ranges and summary statistics are available in the public data set (Tian, 2026).



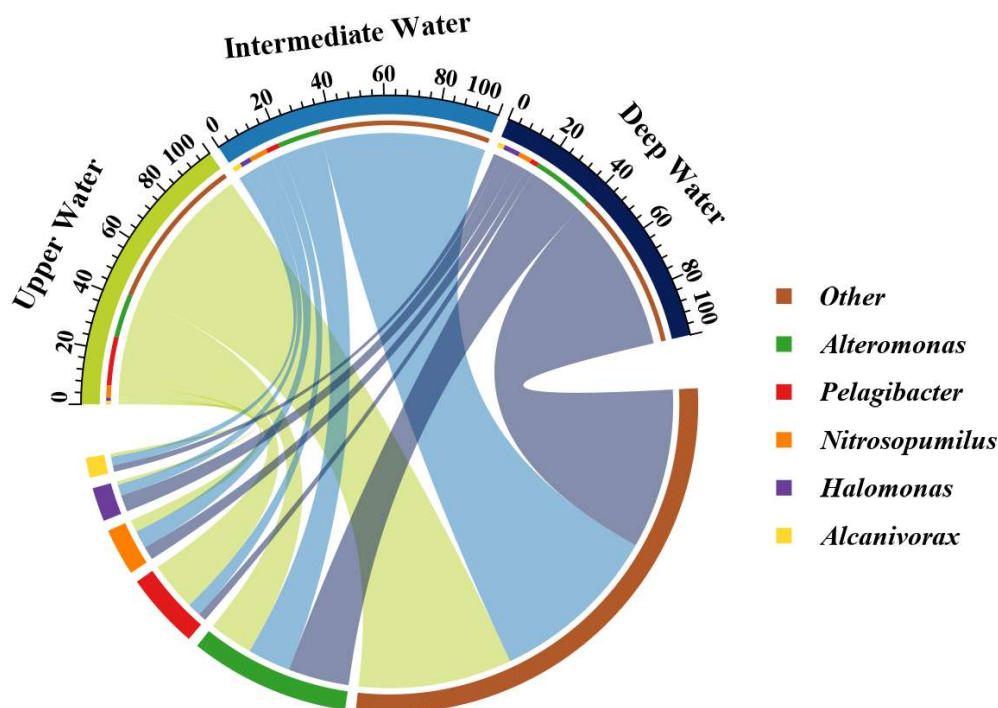
Response Figure 5/Revised Figure 3. Vertical distributions of chemical and particulate parameters in the water column. Profiles show (a) dissolved inorganic nitrogen (DIN), (b) phosphate ($PO_4\text{-P}$, as dissolved inorganic phosphorus, DIP), (c) silicate ($SiO_3\text{-Si}$, as dissolved inorganic silicon, DSi), (d) residual preformed NO_x (rPre NO_x), (e) residual preformed phosphate (rPre PO_4), (f) apparent oxygen utilization (AOU), (g) particulate organic carbon (POC), (h) dissolved inorganic carbon (DIC), (i) transparent exopolymer particle (TEP). For rPre NO_x and rPre PO_4 , each point and its horizontal error bar represent the mean $\pm$ standard deviation derived from the 9 parameter combinations for an individual sample. For TEP, each point and its horizontal error bar represent the mean $\pm$ standard deviation of triplicate measurements. Red diamonds connected by dashed lines and their horizontal error bars denote the mean $\pm$ standard deviation of all samples collected at each nominal sampling depth.

AOU and DIC showed broadly parallel vertical distributions, with low upper water values and marked increases into the intermediate and deep waters (Figs. 3f, h). In contrast, POC and

TEP exhibit a distribution pattern that is nearly opposite to that of nutrients and DIC (Figs. 3g, h, i). TEP concentrations in the upper water ranged from 7.53 to 34.22 $\mu\text{g Xeq L}^{-1}$ (mean $22.52 \pm 8.54 \mu\text{g Xeq L}^{-1}$), exceeding those in deeper waters. These concentrations were within the low range reported for open ocean environments (Table S1). After conversion to carbon equivalents using a factor of 0.51 (Engel and Passow, 2001), the TEP-C/POC ratio ranged from 15.45% to 71.75%, with a mean of $43.81 \pm 14.62\%$. The mean ratio was commonly higher than those observed in eutrophic oceanic regions (Table S1). POC concentrations decrease rapidly in the intermediate and deep waters, while the AOU continuously increased (Figs. 3f, g). In contrast, TEP concentrations decrease more slowly and remain within a measurable range even in the deep waters (Fig. 3i).

3.3 Microbial community structure

Among the five identified dominant genera, Pelagibacter, an oligotrophic bacterial genus, and Alteromonas, an opportunistic heterotrophic bacterial genus, had the highest relative abundances, but displayed different vertical patterns (Fig. 4). In the upper waters, Pelagibacter was the most abundant highlighted genus, accounting for 18.4 % of the community. Alteromonas represented a comparable proportion of 16.6 %. Nitrosopumilus, a chemoautotrophic ammonia-oxidizing archaeal genus, accounted for 4.6 %, whereas Halomonas and Alcanivorax were minor components, accounting for 1.3 % and 1.2 %, respectively. The intermediate waters showed a different community composition, the relative abundance of Pelagibacter decreased to 5.0 %, whereas that of Alteromonas remained relatively high at 16.2 %. The relative abundance of Nitrosopumilus increased to 6.8 %, and those of Halomonas and Alcanivorax increased to 4.2 % and 3.3 %, respectively. In the deep waters, Alteromonas increased to 22.9 % and became the most abundant identified genus, whereas Pelagibacter declined further to 3.0 %. Halomonas reached its highest relative abundance of 6.3 %, while Nitrosopumilus and Alcanivorax accounted for 5.0 % and 2.5 %, respectively.

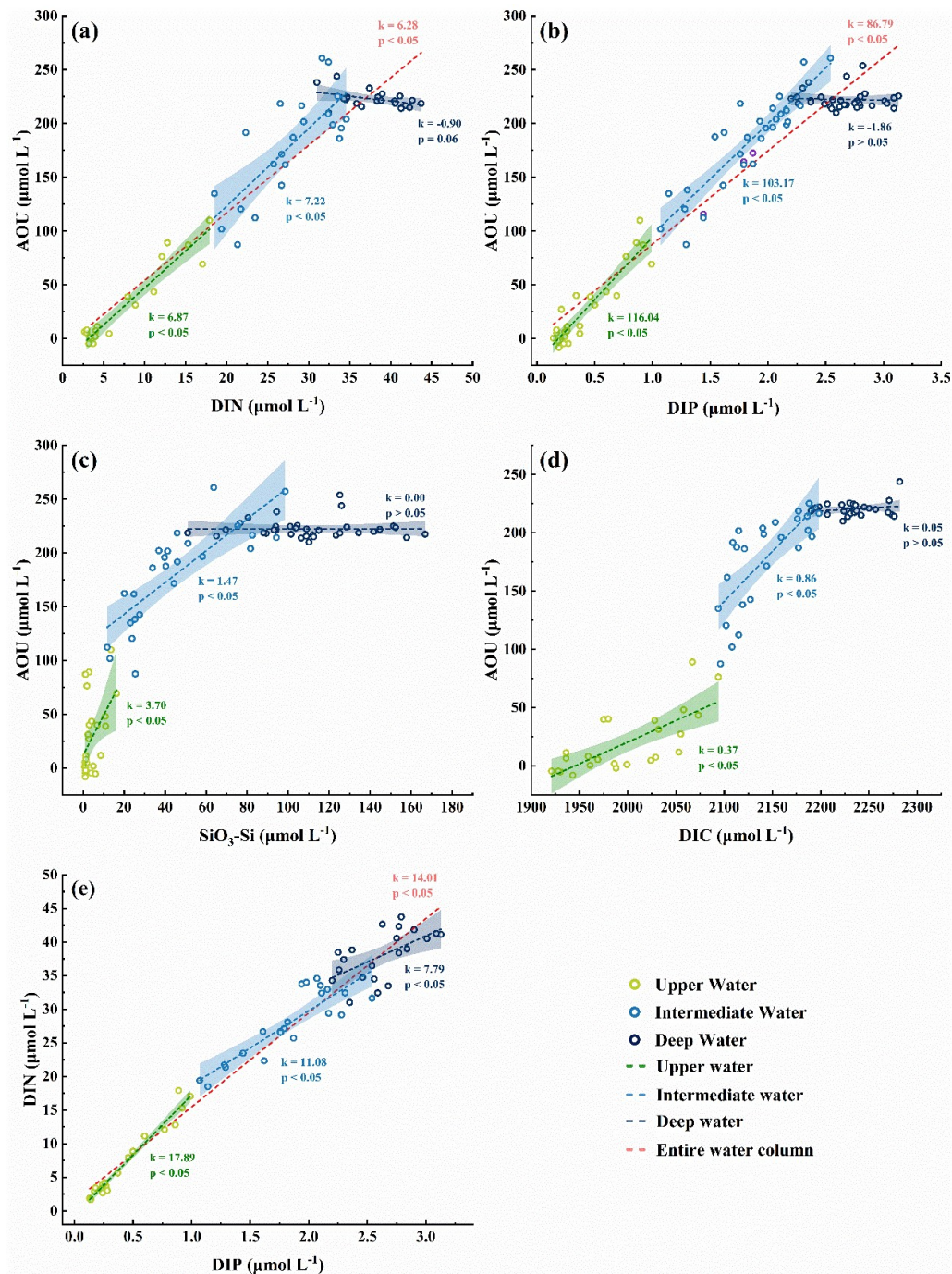


Response Figure 6/Revised Figure 4. Relative abundance (%) of major microbial genera across the upper, intermediate, and deep waters.”

Corresponding revisions were also made throughout the Discussion. In the discussion associated with Figure 5 (Response Figure 7), all AOU–nutrient and DIN–DIP relationships are now interpreted consistently for the upper, intermediate, and deep hydrographic groups defined by water mass membership, whereas slopes calculated for the entire water column are clearly identified and discussed separately (Revised manuscript, Lines 341-389):

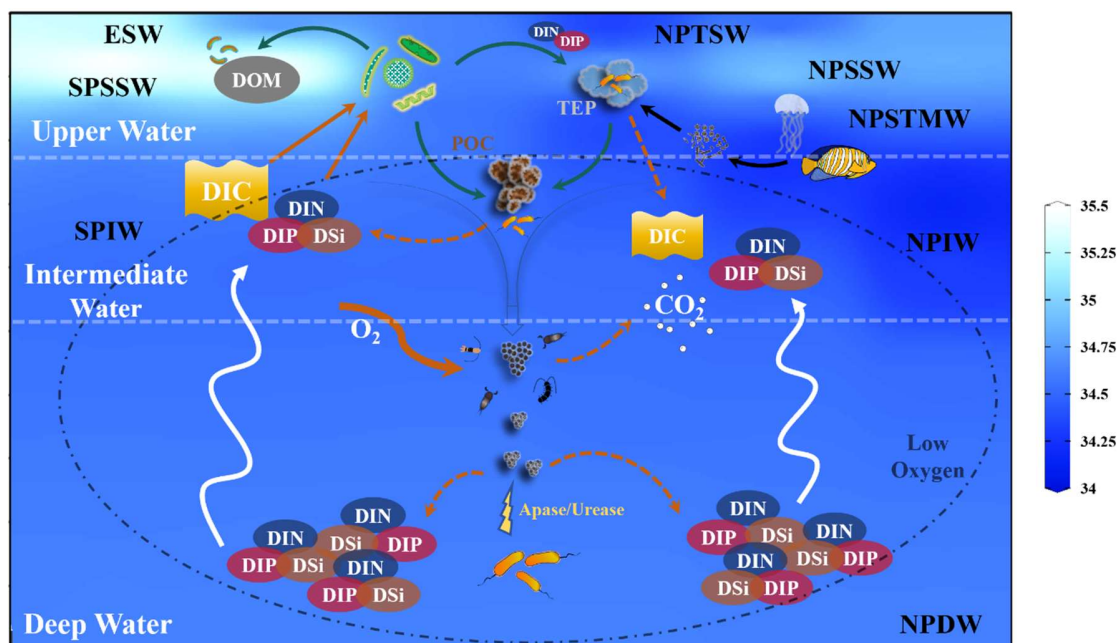
“To examine how these biogeochemical processes vary across water column, we calculated AOU/nutrient regression slopes separately for the upper, intermediate, and deep waters (Fig. 5). The AOU/DIN value is lowest in the upper waters (6.87), increases in the intermediate waters (7.22), and becomes statistically insignificant in the deep waters ($p > 0.05$) (Fig. 5a). The AOU/DIP slopes is higher in the upper waters (116.04), decreasing in the intermediate waters (103.17), and non-significant in the deep waters (Fig. 5b). The low AOU/DIN and AOU/DIP slopes in the upper and intermediate waters indicate greater nutrient accumulation per unit increase in AOU than expected from the classical reference ratios. Limited vertical nutrient supply maintains persistently oligotrophic conditions in the upper water column (Chl-a in the deep chlorophyll

maximum: $0.19\text{--}0.31\ \mu\text{g L}^{-1}$). Under nutrient limitation, phytoplankton may allocate a relatively large proportion of photosynthetically fixed carbon to extracellular polysaccharides and TEP with high C:N ratios rather than to nutrient-rich cellular biomass (Smyth and Letscher, 2023; Curran et al., 2025). The observed TEP-C/POC ratios in the upper waters (mean of $43.81 \pm 14.62\%$) indicate that estimated TEP-associated carbon constituted a substantial component of the local POC pool (Fig. 3i, Table S1). Positive ensemble average $r\text{PreNO}_x$ and $r\text{PrePO}_4$ residuals occurred together with elevated TEP concentrations in the upper water column (Figs. 3d, e, and i). This indicates that measured nutrient concentrations were higher than predicted from AOU under the prescribed DOM and POM remineralization parameters, resulting in a lower AOU/nutrient ratio in the upper waters. Although standing TEP concentrations do not directly quantify production, the substantial contribution of TEP-associated carbon to the local POC pool and its co-occurrence with low AOU/nutrient ratios support carbon allocation to TEP as one plausible contributor to the observed deviations. Future studies should examine whether similarly low AOU/nutrient ratios occur in other ocean regions with comparably high TEP-C/POC ratios to assess the spatial generality of this association. Furthermore, evidence indicates that heterotrophic microorganisms preferentially consume components with low C:N, such as proteins, in organic matter during aerobic respiration (Hwang et al., 2006; Tian et al., 2025). Therefore, in the intermediate waters, preferential degradation of relatively nutrient rich and labile organic components may enhance nutrient regeneration relative to bulk organic carbon oxidation, thereby contributing to the low AOU/nutrient slopes (Fig. 5a, b). In the deep waters, the absence of significant AOU/nutrients relationships precludes the calculation of meaningful remineralization ratios. In this depth horizon, AOU remains relatively stable whereas nutrient concentrations continue to increase, suggesting that their distributions are governed primarily by the cumulative effects of water-mass aging, lateral transport, and diverse biogeochemical transformations operating on different timescales. Among these, nitrification mediated by ammonia-oxidizing archaea such as *Nitrosopumilus* may consume oxygen without concomitant phosphorus regeneration (Martens-Habbena et al., 2009), while DOP hydrolysis associated with *Alteromonas* may mobilize phosphate independently of bulk organic matter oxidation (Saavedra et al., 2025). Collectively, these processes likely contribute to the observed decoupling between AOU and nutrients in the deep ocean.



Response Figure 7/Revised Figure 5. The correlation between apparent oxygen utilization (AOU) and (a) dissolved inorganic nitrogen (DIN), (b) dissolved inorganic phosphorus (DIP), (c) silicate ($\text{SiO}_3\text{-Si}$, as dissolved inorganic silicon, DSi), (d) dissolved inorganic carbon (DIC), and (e) relationship between DIN and DIP. The shaded areas represent the 95% confidence intervals of the fitted regression lines.”

Original Figure 7 was also reorganized using the same three hydrographic groups (Response Figure 8/Revised Figure 7) . The eight named water masses are retained only to provide hydrographic context within the appropriate groups :



Response Figure 8/Revised Figure 7. Schematic diagram of coupled physical-biogeochemical-microbial controls on carbon and nutrient cycling in the study area with a salinity background.

Specific remarks

1. L131: Why were nutrients samples preserved with chloroform? Chloroform is generally not recommended as a preservation method for dissolved nutrient samples due to its volatility and potential impacts on nutrient concentrations, particularly phosphate, nitrate, nitrite, and ammonia, during storage. Previous studies have reported possible nutrient alterations associated with chloroform preservation, and alternative approaches such as filtration followed by freezing are more commonly applied in marine biogeochemical studies (see Kim et al., 2025 and references therein).

Response: We thank the reviewer for raising this important concern. Because immediate shipboard analysis was unavailable, the nutrient samples were filtered before being amended with chloroform at approximately 1:500 (v/v) to suppress residual biological activity and were subsequently stored at -20°C until laboratory analysis. Kim et al. (2025) also noted that careful

filtration is required when chloroform is used to minimize the release of intracellular nutrients, particularly phosphorus, from microalgae during storage. Thus, the removal of particulate material and microalgal cells reduced this potential source of nutrient alteration. In addition, a closely related procedure was reported by Ma et al. (2023), who filtered seawater through GF/F filters, added chloroform, and stored the samples at -20°C for nitrate and phosphate analyses. We have revised the methods section to include more detailed information on nutrient sampling and have cited relevant supporting literature (Revised manuscript, Lines 131-137):

“...the seawater was filtered through pre-combusted and acid-washed GF/F glass fiber filters (0.7 μm pore size, Whatman; 450°C for 4 h, 0.5 M HCl for 24 h), followed by the collection of nutrient and DIC samples. Samples for nutrient determination were aliquoted into 250 mL high-density polyethylene bottles, amended with chloroform (approximately 1:500, v/v) to suppress biological activity during delayed analysis, and stored frozen at -20°C before measurement (Ma et al., 2023).”

References:

- Kim, M. S., Choi, M. S., Rhee, T. S.: Comparative assessment of preservation methods for major nutrients in polar seawater, Mar. Chem., 272, 104546, <https://doi.org/10.1016/j.marchem.2025.104546>, 2025.
- Ma, J., Li, X., Song, J., Wang, Q., Wen, L., Xu, K., et al.: Relationship and stratification of multiple marine ecological indicators: A case study in the M2 seamount area of the Western Pacific Ocean, Ecol. Indic., 146, <https://doi.org/10.1016/j.ecolind.2022.109804>, 2023.

2. L160: How much time elapsed between sample collection and chlorophyll extraction?

Response: We thank the reviewer for requesting this clarification. Chlorophyll-a was measured as a cruise-wide shared parameter. After consulting the technical staff and rechecking the sampling records, we found that the storage temperature was incorrectly reported in the original manuscript. Following filtration, the filters were stored at -80°C onboard, rather than at -20°C , and remained frozen until extraction in the shore-based laboratory.

Because the stations were sampled sequentially, the interval between sample collection and extraction varied among stations. The longest interval was approximately 50 days for station E142-11, which was sampled earliest. Kratzer et al. (2022) recommended -80°C storage for chlorophyll-a filters and documented laboratory protocols involving GF/F filters stored at this temperature for

periods of up to three months. Thus, this study supports the storage temperature and provides a relevant reference range for the storage duration. We have corrected the storage temperature in the revised Methods (Revised manuscript, Lines 150-154).

“Samples for chlorophyll-a (Chl-a) determination were collected from five depths between 5 and 200 m. First, 2 L of seawater samples were filtered through a 200 μ m mesh to remove zooplankton and then filtered onto a 0.7 μ m GF/F glass fiber membrane (Whatman; 450 °C for 4 h, 0.5 M HCl for 24 h). The filters were then stored at -80 °C until analysis in the shore-based laboratory.”

References:

Kratzer, S., Harvey, E. T., Canuti, E.: International Intercomparison of In Situ Chlorophyll-a Measurements for Data Quality Assurance of the Swedish Monitoring Program, *Frontiers in Remote Sensing*, 3, <https://doi.org/10.3389/frsen.2022.866712>, 2022.

3. L211/L223: Why were general stoichiometric ratios applied rather than the water mass-specific stoichiometric relationships identified in the study region? Since the manuscript itself demonstrates that the observed AOU-to-nutrient ratios deviate from the expected Redfield stoichiometry, it is unclear why a stoichiometric framework that does not adequately represent the investigated water masses is used for subsequent analyses. The authors should discuss how this choice may affect the calculated ratios, the interpretation of nutrient regeneration processes, and the overall conclusions of the study.

Response: We appreciate this important comment. The published stoichiometric ratios were deliberately used as external reference parameterizations rather than as locally validated remineralization ratios. The relationships determined for individual water masses in this study were derived from the same measured AOU and nutrient data used to evaluate stoichiometric deviation. Using these locally derived slopes as input parameters in the residual calculations would therefore be circular and would tend to reduce the residuals by construction.

We agree that the selected reference ratios affect the absolute values of $r\text{PreNO}_x$ and $r\text{PrePO}_4$. To quantify this dependence, we evaluated 9 mass balance constrained parameter combinations. Although the residual magnitudes varied among these parameterizations, their principal vertical patterns remained consistent. The resulting values and ranges are reported in the figures and accompanying public data set.

Accordingly, $r\text{PreNO}_x$ and $r\text{PrePO}_4$ are now described explicitly as parameter dependent diagnostic residuals rather than quantitative estimates of true preformed nutrient inventories or independent evidence for specific regeneration ratios. Our interpretation of nutrient regeneration and stoichiometric deviation is based primarily on regressions using directly measured AOU and nutrient concentrations, while the residuals provide only complementary comparisons with published reference frameworks. Thus, the parameter choice affects the magnitude of the diagnostic residuals but does not determine the principal conclusions of the study. We have clarified this rationale and its associated uncertainty in the revised Methods and Discussion.

Revised Methods (Revised manuscript, Lines 216-221):

“ $r\text{PreNO}_x$ and $r\text{PrePO}_4$ were calculated using the modified formulation of Letscher and Villareal (2018), with parameter sensitivity evaluated following Smyth and Letscher (2023). For each observation, 9 calculations were performed using the prescribed DOM and POM parameter combinations. The arithmetic mean was used as the reported diagnostic value, whereas the minimum, maximum, and standard deviation describe sensitivity to the prescribed parameters.”

Revised Discussion (Revised manuscript, Lines 354-361):

“The observed TEP-C/POC ratios in the upper waters (mean of $43.81 \pm 14.62\%$) indicate that estimated TEP-associated carbon constituted a substantial component of the local POC pool (Fig. 3i, Table S1). Positive ensemble average $r\text{PreNO}_x$ and $r\text{PrePO}_4$ residuals occurred together with elevated TEP concentrations in the upper water column (Figs. 3d, e, and i). This indicates that measured nutrient concentrations were higher than predicted from AOU under the prescribed DOM and POM remineralization parameters, resulting in a lower AOU/nutrient ratio in the upper waters.”

4. L212: Since $\text{NO}_{3\text{meas}}$ is defined as the sum of $\text{NO}_3\text{-N}$ and $\text{NO}_2\text{-N}$ concentrations, please refer to it as NO_x .

Response: We thank the reviewer for identifying this imprecise nomenclature. We agree and now define $\text{NO}_x = \text{NO}_3^- + \text{NO}_2^-$ and use $r\text{PreNO}_x$ consistently throughout the manuscript and Supplement. This is a nomenclature correction and does not change the calculated values.

5. L224-226: To what extent is this assumption valid if one of the main conclusions of the paper is that the Redfield ratio does not apply?

Response: We appreciate this question. The N:P ratio of 16:1 was used solely as a common reference for converting nitrogen based coefficients to phosphorus equivalents, rather than as a locally validated stoichiometric model. rPrePO₄ was also evaluated using nine parameter combinations, thereby assessing its sensitivity to the prescribed remineralization parameters, although its absolute values remain conditional on the 16:1 conversion. The conclusion regarding nonclassical oxygen–nutrient coupling is based on regressions of measured AOU against measured DIN and DIP; the residuals provide only parameter dependent diagnostic context. This limitation has now been stated in the revised Methods (Supplementary materials, Lines 61-64).

“Because the remineralization parameters specific to phosphorus could not be independently constrained, the nitrogen regeneration terms derived from DOM and POM were converted to phosphorus equivalents using an N:P ratio of 16:1 (Letscher and Villareal, 2018), solely as a common conversion reference.”

6. L369: In what depth is the mixed layer depth? Shouldn't the uppermost part of the water column, which is mixed by wind and wave-driven turbulences, be excluded from the analysis?

Response: We thank the reviewer for this valuable point. We agree that the robustness of the upper water column result to mixed-layer observations should be assessed. We computed station-specific mixed-layer depths (MLDs) following the Thompson (1976) temperature-threshold approach for each of the six sampling stations. The calculated MLDs for the six stations are as follows: EQ-6 (65 m), E142-3 (102 m), E142-7 (68 m), E142-11 (42 m), E142-13 (45 m), and E142-19 (109 m). Twelve of 95 observations were at or above MLD, all in the upper waters. Excluding them changed the five upper-group slopes only modestly. Paired row-bootstrap comparisons with 4,999 resamples found no significant slope differences after Benjamini–Hochberg correction (adjusted $p = 0.12$ – 1.00 , Response Table 4). We therefore retained all observations in the primary analysis. This choice also ensured an adequate sample size for the group specific regressions.

Response Table 4. Sensitivity of upper water slopes to exclusion of observations at or above station-specific mixed-layer depth.

Response– predictor	All slope	Exclude- MLD slope	BH- adjusted p	Different at 0.05
AOU–DIN	6.87	6.59	0.29	No
AOU–DIP	116.04	111.40	0.12	No
AOU–SiO ₃ -Si	3.70	3.46	1.00	No
AOU–DIC	0.37	0.43	0.63	No
DIN–DIP	17.89	17.39	0.18	No

Reference:

Thompson, R. O. R. Y.: Climatological Numerical Models of the Surface Mixed Layer of the Ocean, *J. Phys. Oceanogr.*, 6, 496–503, 1976.

7. L368: Why was this analysis not also performed separately for the individual water masses? Earlier in the manuscript, the analyses are conducted on a water-mass-specific basis, whereas here results are presented as a bulk signal across multiple depth layers. What is the rationale for changing the approach at this stage?

Response: We appreciate the opportunity to clarify this analytical choice. Separate analyses of individual water masses were not conducted because several water masses contained too few samples or covered insufficient AOU and nutrient ranges to produce stable and meaningful regressions. In the revised manuscript, all relevant analyses have been standardized to the upper, intermediate, and deep water groups. Hydrographically related water masses with similar vertical distributions were therefore combined into the three groups. Accordingly, the revised results no longer represent an unexplained bulk signal across multiple layers but provide consistent comparisons among the upper, intermediate, and deep waters throughout the manuscript. Further details on the grouping rationale and the corresponding revisions are provided in our responses to “*General remark 2*”

8. L371: Is this really a single water mass? Figure 5 suggests that the upper 300 m contains more than one water mass. Please be careful with the terminology and ensure that the distinction between water masses and depth intervals is made consistently throughout the manuscript.

Response: We agree that the upper 300 m does not represent a single water mass. The original use of “upper water mass” was therefore inaccurate and has been corrected throughout the manuscript. We now distinguish explicitly between individual named water masses and the broader hydrographic groups used for analysis. The upper waters comprise NPTSW, ESW, NPSSW, SPSSW, and NPSTMW; the intermediate waters comprise NPIW and SPIW; and the deep waters are represented primarily by NPDW.

These three categories are referred to consistently as upper, intermediate, and deep waters rather than individual water masses. Water mass assignment is based on temperature, salinity, and neutral density characteristics, whereas the approximate depth intervals describe only the predominant vertical distributions of these groups. We have checked and revised the terminology throughout the text, figures, tables, and captions to maintain this distinction. Further details are provided in our responses to “*General remark 2*”.

9. L430: What about denitrification in anoxic microsites close to particles (e.g., Bianchi et al., 2018; Wan et al., 2023)?

Response: We thank the reviewer for raising this important possibility. We agree that oxygenated bulk water does not exclude denitrification within anoxic particle microsites. We have therefore revised the Discussion to distinguish this process from classical water column denitrification (Revised manuscript, Lines 422-431):

“Denitrification may nevertheless occur within particles suspended in oxygenated water when respiratory oxygen demand exceeds oxygen supply, particularly in large particles with high respiratory activity (Ploug, 2001; Ploug et al., 2008). The importance of this process depends on particle abundance and size distribution (Bianchi et al., 2018), and has been shown to increase with particle size and concentration in oxygenated coastal waters with high particle loads (Wan et al., 2023). In our study, POC and TEP concentrations were within the lower reported ranges (Table S1), indicating relatively low particle loads. Although these bulk measurements cannot exclude denitrification within individual particles, they suggest that this process was unlikely to dominate the observed DIN:DIP pattern.”

Because particle size, internal oxygen conditions, and denitrification rates were not measured directly, localized denitrification within individual particles cannot be excluded or quantified. This

possibility and the associated uncertainty are now explicitly acknowledged in the revised manuscript.

References:

Bianchi, D., Weber, T. S., Kiko, R., Deutsch, C.: Global niche of marine anaerobic metabolisms expanded by particle microenvironments, *Nat. Geosci.*, 11(4), 263-268, <https://doi.org/10.1038/s41561-018-0081-0>, 2018.

Ploug, H.: Small-scale oxygen fluxes and remineralization in sinking aggregates, *Limnol. Oceanogr.*, 46(7), 1624-1631, <https://doi.org/10.4319/lo.2001.46.7.1624>, 2001.

Ploug, H., Iversen, M. H., Fischer, G.: Ballast, sinking velocity, and apparent diffusivity within marine snow and zooplankton fecal pellets: Implications for substrate turnover by attached bacteria, *Limnol. Oceanogr.*, 53(5), 1878-1886, <https://doi.org/10.4319/lo.2008.53.5.1878>, 2008.

Wan, X. S., Sheng, H.-X., Liu, L., Shen, H., Tang, W., Zou, W., et al.: Particle-associated denitrification is the primary source of N₂O in oxic coastal waters, *Nat. Commun.*, 14(1), 8280, <https://doi.org/10.1038/s41467-023-43997-3>, 2023.

10. Figure 3: I'm missing some very basic depth profiles plots of key water mass properties (also temperature, salinity, AOU, DIN, P, TEP, ...). Such figures would help readers better visualize the stratification and support the separation of the water column into the three depth intervals (0-300 m, 300-1000 m, and >1000 m

Response: We appreciate this helpful suggestion and agree that depth profiles provide a clearer visualization of water column stratification. Accordingly, we comprehensively revised the relevant figures throughout the manuscript. Specifically:

Figure 2 (Response Figure 4). The T-S diagram is retained to identify and visualize the eight named water masses. Temperature, salinity, and neutral density are now shown as depth profiles, with observations distinguished by the three hydrographic groups and depth-specific means shown separately. This retains the added hydrographic information while aligning the physical presentation with the subsequent analyses.

Figure 3 (Response Figure 5). The former water-mass-based presentation has been replaced by depth profiles of DIN, DIP, DSi, rPreNO_x, rPrePO₄, AOU, POC, DIC, and TEP. Individual

observations are colored by hydrographic group, and red diamonds with dashed lines show the mean and standard deviation at each nominal sampling depth.

Figure 4 (Response Figure 6). Microbial relative abundances are now aggregated and displayed for the upper, intermediate, and deep hydrographic groups rather than for each of the eight named water masses.

Figure 5 (Response Figure 7). All regression fits and slopes are now presented for the same three hydrographic groups, while fits calculated for the entire water column are shown and labeled separately. In Figure 5b, we found the slope labels for the upper and intermediate hydrographic groups were inadvertently reversed in the original figure. This error has now been corrected: the AOU/DIP slopes are 116.04 for the upper hydrographic group and 103.17 for the intermediate hydrographic group. The corresponding descriptions in the Discussion have also been corrected (Revised manuscript, Lines 345-347): “*The AOU/DIP slopes is higher in the upper waters (116.04), decreasing in the intermediate waters (103.17), and non-significant in the deep waters (Fig. 5b).*”. Correcting the labels changes the relative ordering of the upper- and intermediate-group slopes, but it does not alter the central conclusion that both slopes are below the classical reference value of 138.

Figure 7 (Response Figure 8). The conceptual schematic has been reorganized around the upper, intermediate, and deep hydrographic groups. The individual water mass names are retained only as hydrographic context within the appropriate group, rather than being depicted as fixed depth intervals.

Together, these revisions make the role of the water mass identification explicit, align the figures with the analytical framework used in the Discussion, and distinguish group-specific slopes from slopes calculated for the entire water column.

We also revised the Results, figure captions, and terminology throughout the manuscript to distinguish consistently between individually identified water masses and the three vertically aggregated water groups. These revisions are described in detail in our responses to “*General remark 2*”.

We again thank the editor and both referees for the detailed and constructive reviews. The integrated revisions have strengthened the physical definition of the analytical groups, clarified the limitations of the residual diagnostics, improved methodological transparency, and moderated the

mechanistic interpretation. We hope that the final revised manuscript and Supplementary Materials now address all concerns and are suitable for publication in *Biogeosciences*.