Dear Reviewer,

We appreciate your constructive suggestions that have led to an improvement of the manuscript. We have fully addressed these comments during the revision. To assist your assessment of our revised manuscripts, we have provided point-to-point response (**blue in color**) to each of the comments by reviewers below. The location of the change in the revised manuscript is highlighted in our response.

Sincerely yours,

Dr. Hongbin LIU (Corresponding author, Email address: liuhb@ust.hk)

Responses to review 1:

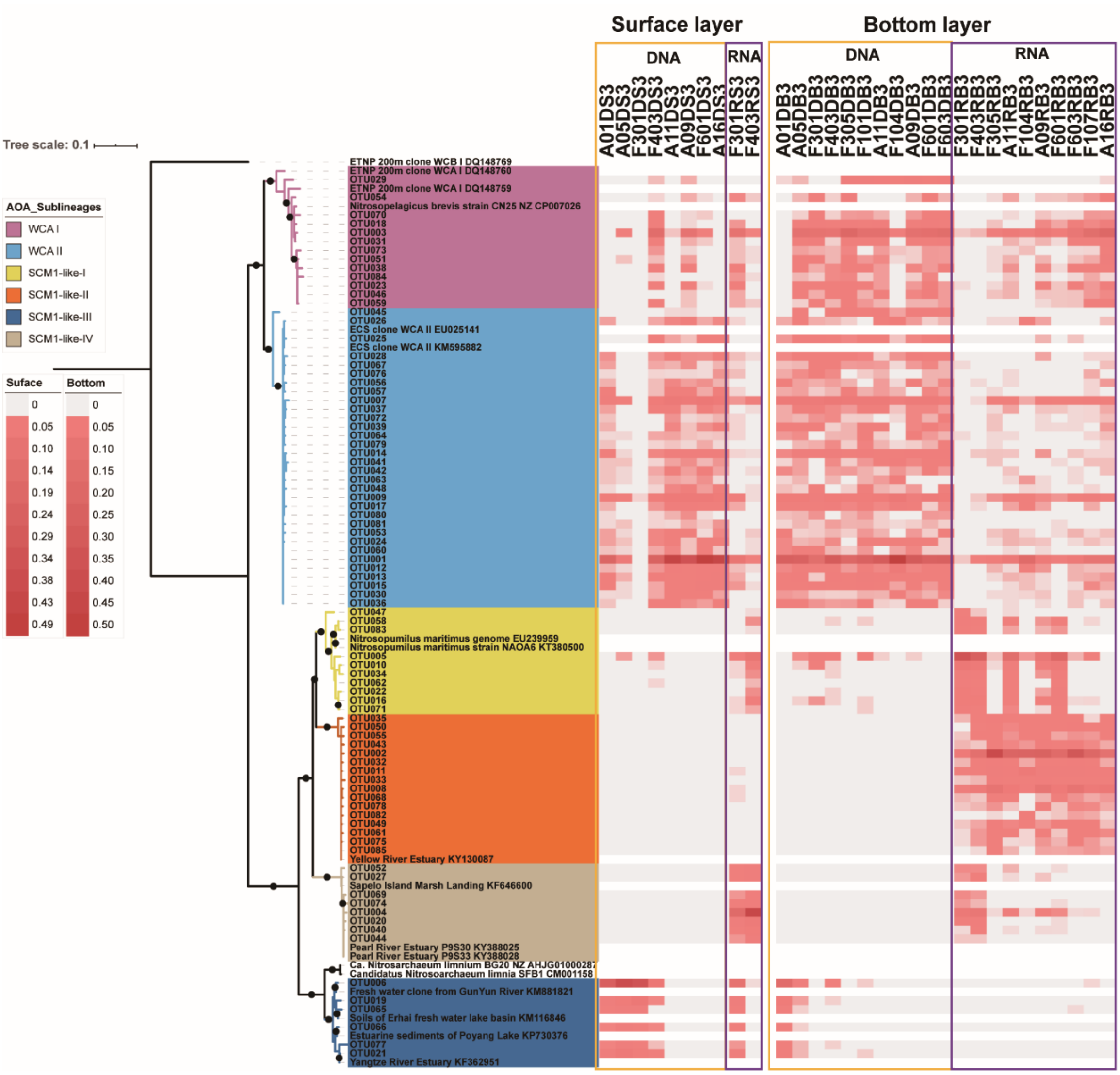
general comments

This study by *Lu et al.* provides valuable new insights into the distribution of ammonia-oxidizing archaea (AOA) sublineages and AOA versus ammonia-oxidizing bacteria in the subtropical Pearl River estuary. The study shows a difference in the composition of AOA sublineages at the DNA and RNA level and correlation of nitrification rates with the relative abundance of only one AOA sublineage suggesting a niche partitioning between different AOA sublineages. Furthermore, the authors present data on the contribution of nitrification to oxygen consumption.

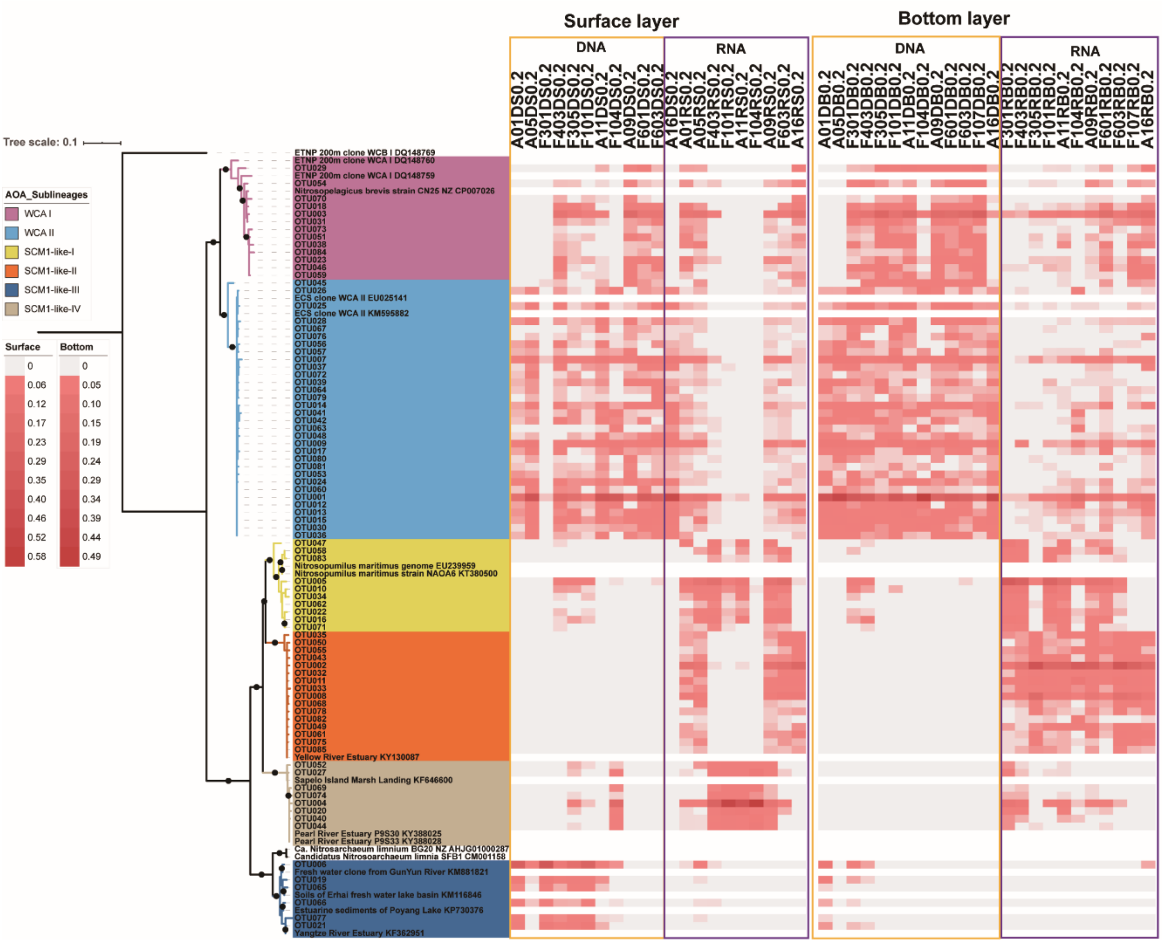
Response: *We thank the reviewer for the accurate summary of our study.*

Parts of the data set are only superficially mentioned in the manuscript (e.g. fig 8) although they contain valuable information. Especially the comparison between particle attached vs free-living AOA community composition deserves more attention.

Response: *While comparing the particle-attached and free-living communities, we did not observe significant difference correspondingly (ANOSIM: r=-0.02177, P=0.797, permutation=999). In contrast, we observed large variation of community along the steep environmental gradient in Pearl River estuary at both DNA and RNA levels (ANOSIM: r=0.7142, P=0.001, permutation=999). Here, we provide two heatmap plots for your reference by splitting Figure 6 (new figure 6 & new figure 7 below): New figure 6: Phylogenetic tree and relative abundance (heatmap)of particle-attached AOA. New figure 7: Phylogenetic tree and relative abundance (heatmap)of free-living AOA. Here, the revised figure 6 and new figure 7 show no significant difference. Therefore, we mainly focused on biogeography of different AOA sublineages and the disagreement between DNA and RNA communities.* Page 28-29 Line 643-651



(Revised) Figure 6. Maximum likelihood phylogenetic tree of top 85 OTUs based on amoA gene sequences using T92+G+I model with 1000 bootstrap. The associated heat map is generated based on the relative abundance of top OTUs in the particle-attached samples. Samples are listed from left to right along the ascending salinity gradient.



(Newly added) ***Figure 7. Maximum likelihood phylogenetic tree of top 85 OTUs based on amoA gene sequences using T92+G+I model with 1000 bootstrap. The associated heat map is generated based on the relative abundance of top OTUs in the free-living samples. Samples are listed from left to right along the ascending salinity gradient.***

NOD/CRs ratios are a central focus of this manuscript. At the same time the NOD rates are part of different manuscript. In order to see the clear separation of focus and content, the other manuscript should be made accessible to the reviewers. This probably would also help to get important information on the method of NOD determination that are missing from this manuscript (e.g. how many time points were taken per rate measurement?).

Response: *We have elaborated the method of rates measurement (showed below) in the revised manuscript. We did not conduct rates measurement with multiple time points. The estimation of NOD is based on stoichiometric equation (*$NH_3 + 1.5\ O_2 \rightarrow NO_2^- + H_2O + H^+$*”). This study (using qPCR, Ion-torrent sequencing, rates measurement, environmental data) provided a comprehensive view of two group of ammonia oxidizers and more importantly, new insight on distinct distribution patterns of AOA sublineages at DNA and RNA level in the estuarine* *environment in 2017 summer cruise. The other study, using two sets of dark ammonia assimilation rates and nitrification rates from 2015 and 2017 cruises, mainly focus on source and sink of riverine ammonium. We think these two studies contain different and separated contents since they only shared a small part of nitrification rates data in 2017 cruise. Here, we provide the title and abstract of Chen L's work for your reference.*

"Title: Title: ***Dark ammonium transformations in the Pearl River Estuary during summer***

Abstract

Growing human activities in recent decades have collectively resulted in large amounts of nutrients export into coastal oceans. As the most reactive nitrogen species, ammonium (NH_4^+) plays the critical role in biogeochemical cycles in estuaries and the coastal ocean. In the highly polluted Pearl River Estuary (PRE), NH_4^+ predominates to be the energy source for nitrification, and to be the material source for bacteria and phytoplankton to grow. Both above processes are affected by light, yet in opposite ways. Nevertheless, rare studies paid attention to dual NH_4^+ transformation processes specifically during dark conditions. By using nitrogen isotope tracer technique, we quantitatively and simultaneously differentiated two distinctive NH_4^+ consumption pathways, i.e., NH_4^+ oxidation (AOD) and assimilation (AAD) rates, specially under dark conditions along the PRE during the 2015 and 2017 summer cruises when biological activities were the highest. We found the NH_4^+ transformations display a bilayer structure with AAD>AOD in almost all the surface waters and vice versa in all bottom waters, suggesting bacteria and phytoplankton (mainly bacteria) control NH_4^+ consumption in surface during the night while nitrifiers are the major NH_4^+ consumer in the bottom waters. Through redundancy analysis, we found that both processes are mainly driven by NH_4^+ in the PRE during summer."

Here is the elaborated method of the rates measurement in the revised manuscript: "Community respiration rates (CR) were measured in triplicate in 60ml BOD bottles without headspace through the dissolved oxygen variance before and after 24 h dark incubation submerged in seawater continuously pumped from sea surface. Nitrification were measured by incubating $^{15}NH_4^+$ amended (less than 10 % of ambient concentration) seawater in duplicated 200 ml HDPE bottles in dark for 6-12 h, with temperature controlled by running seawater. After incubation, filtrate (0.2 μm-syringe-filtered) was collected and stored in -20 ℃ for *downstream* $^{15}NO_x^-$ *(*$^{15}NO_3^-$ + $^{15}NO_2^-$*) analysis (Sigman et al. 2001).*

The nitrification rates were calculated using the following equation:

$$AO_b = \frac{(R_t NO_x^- \times [NO_x^-]_t) - (R_{t0} NO_x^- \times [NO_x^-]_{t0})}{t-t0} \times \frac{[^{14}NH_4^+] + [^{15}NH_4^+]}{[^{15}NH_4^+]} \qquad (1)$$

In equation 1, AO_b *is the bulk nitrification rate.* $R_{t0}\, NO_x^-$ *and* $R_t\, NO_x^-$ *are the ratios (%) of* ^{15}N *in the* NO_x^- *pool measured at the initial (*t_0*) and termination (t) of the incubation.* $[NO_x^-]_{t0}$ *and* $[NO_x^-]_t$ *are the concentration of* NO_x^- *at the initial and termination of the incubation, respectively.* $[^{14}NH_4^+]$ *is the ambient* NH_4^+ *concentration.* $[^{15}NH_4^+]$ *is the final ammonium concentration after addition of the stable isotope tracer (*$^{15}NH_4^+$*). The* NO_x^- *was completely converted to* N_2O *by a single strain of denitrifying bacteria (Pseudomonas aureofaciens, ATCC#13985) which lack* N_2O*-reductase activity (Sigman et al. 2001). The converted* N_2O *was further analyzed using IRMS (Isotope Ration Mass Spectrometer, Thermo Scientific Delta V Plus) to calculate the isotopic composition of* NO_x^- *(Sigman et al. 2001; Casciotti et al. 2002; Knapp et al. 2005).We analyzed the correlation between nitrification rates and AOA sublineages. Equation 2 was generally considered as the oxidation of ammonia to nitrite. Inferred from the nitrification rates, we estimated the nitrification oxygen demand (NOD) based on equations 2. Inferred from the nitrification rates, we estimated the nitrification oxygen demand (NOD) based on equation 2. We used NOD/CR ratio (percentage) to evaluate potential the contribution of nitrification to total oxygen consumption in the field.*

$NH_3 + 1.5O_2 \rightarrow NO_2^- + H_2O + H^+$ *(2)"* Page 5 Line 92-111

A lot of emphasis is put on the relative importance of NOD in CR. It is stressed various times throughout the manuscript that NOD is high and at times amounts to more than 200%. However, at these stations NOD is not significantly higher compared to other stations, instead CR rates are VERY low. A critical discussion of the CR rates is absent and should be added to the discussion section. How can the observed patchiness of CR rates be explained?

Furthermore, this raises the question of how well constrained the CR data are. Are they based on two data points per rate measurement? How many replicates have been performed? No standard deviation is reported for NOD or CR. I ask the authors to add this information to the respective tables in the supplementary information and would like them include the number of replicates performed in the material and method section. According to the material and method section, triplicates were performed for the qPCR data. However, standard deviations are also missing in the respective data tables in the supplementary information. I ask the authors to add this.

Response: *We have added the standard deviation information in Table S2, S3, S4. We also added information in the methodology section that we performed triplicate in community respiration rates measurement. Nitrification rates were measured in duplicates. Both rates were measured only at the end of incubation and we did not perform multi-time-point measurements. We have to admit that the high contribution ratios may be introduced by the underestimation of community respiration rates at low oxygen condition (Sampou and Kemp 1994). Nevertheless, the NOD/CR ratio in our study is to show the potential effect of active nitrification on oxygen consumption in the estuarine system. As the community respiration rates were inhibited but the nitrification rates were not limited at the DO concentrations observed in our survey, it is suggested that nitrification could potentially contribute a large proportion of oxygen consumption under low DO concentration. We have added discussion on community respiration rates in Section 4.1.* Page 11 305-308, 315-317

Please see the attached and revised version of Table S2, S3 and S4 at the bottom of this file.

For the calculation of the inferred nitrification oxygen demand, the authors use improperly balanced equations. This strongly influences the outcome: e.g. for ammonia oxidation, when using

NH3 +1.5 O2 →NO2- + H2O + H+

instead of equation (1), the oxygen demand changes by 33%. During carbon fixation, some electrons are used to reduce CO2 and not oxygen. However, the assumption that for every NH3 molecule 1.98 HCO3 gets fixed is hardly realistic. Furthermore, the authors assume 1:1 coupling between ammonia oxidation and nitrite oxidation. However, no data on the abundance of nitrite oxidizers is provided and the rate measurements provided do not distinguish between nitrite or nitrate production. I suggest that the estimate of oxygen demand should focus on the first step of nitrification only or at least a paragraph needs to be added to the discussion section. The grammar and language need to be revised. There are too many issues throughout the manuscript to list here, which at times makes it hard to follow the authors line of thought.

Response: *We have removed the equation 2 and 3 in the manuscript and changed our NOD calculation based on equation "*$NH_3 + 1.5\ O_2 \rightarrow NO_2^- + H_2O + H^+$*" (which is now equation 2 in the revised manuscript). The nitrification rates measurement in this study were performed by adding* ^{15}N *labeled ammonium before dark incubation, then collected the filtrate containing* $^{15}NO_x^-$*. The* $^{14/15}$*Nitrite and* $^{14/15}$*Nitrate were converted to* N_2O *by denitrifer method (Sigman et al, 2001). We have elaborated the method of the nitrification rates measurement in the revised manuscript in section 2.2. We now assume each molecule of ammonia consumes 1.5 molecule of oxygen. The NOD and NOD/CR were recalculated based on equation 2 and listed in the revised version of Table S3, description in Section 3.2 and Section 4.1.* Page 2 Line 25; Page 8-9 Line 203-215; Page 12 Line 313-317 *We have improved the manuscript by reducing the grammar and syntax as well as following the important suggestions from the reviewer. We hope that the current version is much clearer.*

specific comments

l. 63 they would not have overlooked them, but rather underestimated their activity and relative contribution to ammonia oxidation.

Response: *We have changed "overlooked" into "underestimated the importance of some active groups in the natural environment"* Page 4 Line 62

ll. 86-87 microbial instead of bacterial.

Response: *We have changed "bacterial" into "microbial"* Page 4 Line 86

l. 96 clarify "running seawater"

Response: *We have changed it into "Community respiration rates (CR) were measured in triplicate in 60ml BOD bottles without headspace through the dissolved oxygen variance before and after 24 h dark incubation submerged in seawater continuously pumped from sea surface".* Page 5 Line 93-94

l. 158 please provide an overview over the 76 samples (which stations and depths are they from) and refer to table S5. The 2523 reads per file does not match the data reported in table S5. The sample categories provided in table S5 need further explanations.

Response: *We subsampled the sequencing reads based on the number of the sample that contains minimum number of reads before OTU clustering. We added abbreviations for sample categories under the Table S5. The sampling depth information have been added to Table S2.*

Here is revised Table S5:

(Revised) Table S5. Basic sample information of sequencing samples and corresponding Shannon index, Margalef richness.

Station	Lon (E °)	Lat (W °)	Sample Cat.	Sequence No.	Shannon index	Margalef richness
			A01RS0.2	4469	4.26	42.06
			A01DB0.2	25484	3.70	39.66
A01	113.65	22.74	A01DB3	33527	3.73	37.25
			A01DS0.2	28147	3.64	37.09
			A01DS3	30179	3.68	39.3
			A05RS0.2	10504	4.21	43.33
			A05DB0.2	32747	3.25	33.3
A05	113.77	22.46	A05DB3	28121	4.00	40.49
			A05DS0.2	27297	3.33	35.85
			A05DS3	20389	3.42	33.75
			A09RB0.2	21803	3.78	39.07
			A09RB3	16585	3.87	41.38
			A09RS0.2	12693	4.14	43.61
A09	113.80	22.21	A09DB0.2	21927	4.04	37.99
			A09DB3	21343	3.71	33.55
			A09DS0.2	10794	4.07	29.95
			A09DS3	25603	3.53	37.12
			A11RB0.2	29345	4.12	43.19
			A11RB3	26206	3.78	39.4
			A11RS0.2	4080	3.26	28.6
A11	113.84	22.09	A11DB0.2	24215	3.82	37.84
			A11DB3	22422	3.72	36.47
			A11DS0.2	20568	3.62	38.78
			A11DS3	29216	3.18	34.89
			A16RB0.2	20644	4.12	40.51
			A16RB3	24676	4.01	41.43
A16	114.05	21.66	A16RS0.2	16931	3.88	39.06
			A16DB0.2	30526	3.31	35.74
			A16DS0.2	31112	3.02	31.63

			A16DS3	28739	3.25	35.5
F101	113.12	21.82	F101RB0.2	20949	3.67	38.37
			F101RS0.2	2523	2.61	23.22
			F101DB0.2	20840	3.61	30.87
			F101DB3	15602	3.96	36.95
			F101DS0.2	8348	3.90	35.38
F104	113.25	21.56	F104RB0.2	33200	3.60	32.74
			F104RB3	16037	3.69	31.77
			F104RS0.2	33670	2.22	17.82
			F104DB0.2	30782	2.84	28.32
			F104DB3	30769	2.69	26.59
			F104DS0.2	6990	3.01	30.22
F107	113.42	21.27	F107RB0.2	21167	3.89	40.88
			F107RB3	5633	3.89	38.1
			F107DB0.2	20909	3.90	35.52
F301	113.55	21.99	F301RB0.2	17778	3.76	34.19
			F301RB3	16657	3.48	34.53
			F301RS3	5653	4.03	37.6
			F301DB0.2	22088	3.82	38.42
			F301DB3	3436	4.19	31.49
			F301DS0.2	7823	3.40	27.44
			F301DS3	20310	3.51	26.54
F305	113.63	21.83	F305RB0.2	27580	3.35	36.05
			F305RB3	27095	3.20	33.45
			F305DB0.2	18856	3.96	33.86
			F305DB3	21410	3.78	35.12
			F305DS0.2	7007	4.20	42.21
F403	113.74	22.08	F403RB0.2	10000	3.86	37.69
			F403RB3	8858	3.69	38.31
			F403RS0.2	4431	3.57	31.38
			F403RS3	4166	3.04	28.24
			F403DB0.2	21959	3.91	40.19
			F403DB3	21744	3.85	38.99
			F403DS0.2	19571	4.26	43.7
			F403DS3	20370	3.83	36.83
F601	114.03	22.14	F601RB0.2	27041	4.12	43.22
			F601RB3	22320	3.75	38.81
			F601DB0.2	18421	3.82	34.78
			F601DB3	20092	3.80	33.59
			F601DS0.2	23411	3.70	37.44
			F601DS3	15932	2.94	33.22
F603	114.09	22.04	F603RB0.2	30619	3.55	37.54
			F603RB3	9410	3.55	38.81
			F603RS0.2	5859	3.90	39.93

F603DB0.2	16912	3.96	40.71
F603DB3	19693	3.81	35.48
F603DS0.2	18314	3.78	36.1

* Sample categories: Station ID + D/R (DNA/RNA) + S/B (Surface/Bottom) + 3/0.2 (Particle attached (>3 μm)/Free-living (3-0.2 μm)).

l. 162 Ion torrent is known for introducing homopolymers. Filtering reads with >8 homopolymers is quite a weak setting considering your aim of "performing fine-scale phylogenetic classification". Please comment.

Response: *The quality control standards resulted that the mean length of homopolymers is 3. The length of the maxhomopolymer in the top OTU sequences we used for phylogenetic analysis in our study is 4, so we think the quality control had excluded error from homopolymers introduced by the Ion torrent.*

ll. 170ff. What is the sampling depth of the samples you classified as "bottom".

Response: *The sampling depth information was added to the revised Table S2.*

l. 330 substrate requirement: do the authors mean substrate concentration?

Response: *Yes, we mean substrate concentration. We have added "concentration".* Page12 Line 339

l. 355 "questionable" How so? Such a statement needs to be accompanied with an explanation.

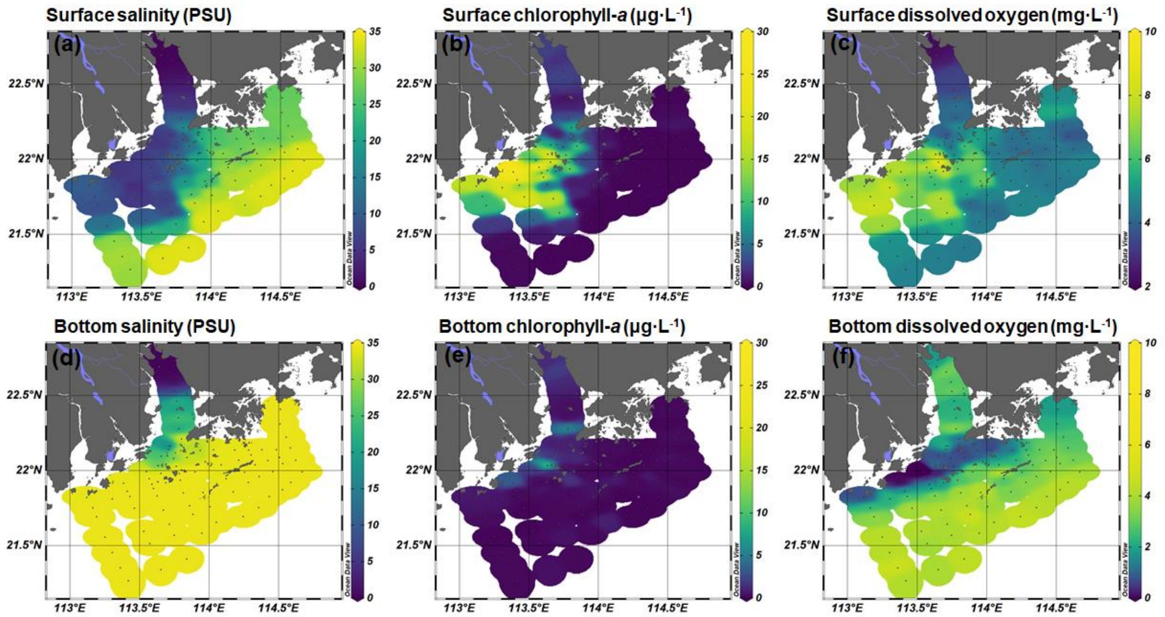
Response: *In line 361 to 363, the low-salinity adapted cluster were proposed by Mosier and Francis in 2008, however, a later study by Molin in 2009 observed these phylotypes in salt marsh with high salinity, which led to the low-salinity adaptation cluster questionable. This was summarized by Bernhard and Bollmann 2010. We think we had the explanation.*

Section 4.1 repeats results in great detail that are already described in the result section. Consider condensing this section.

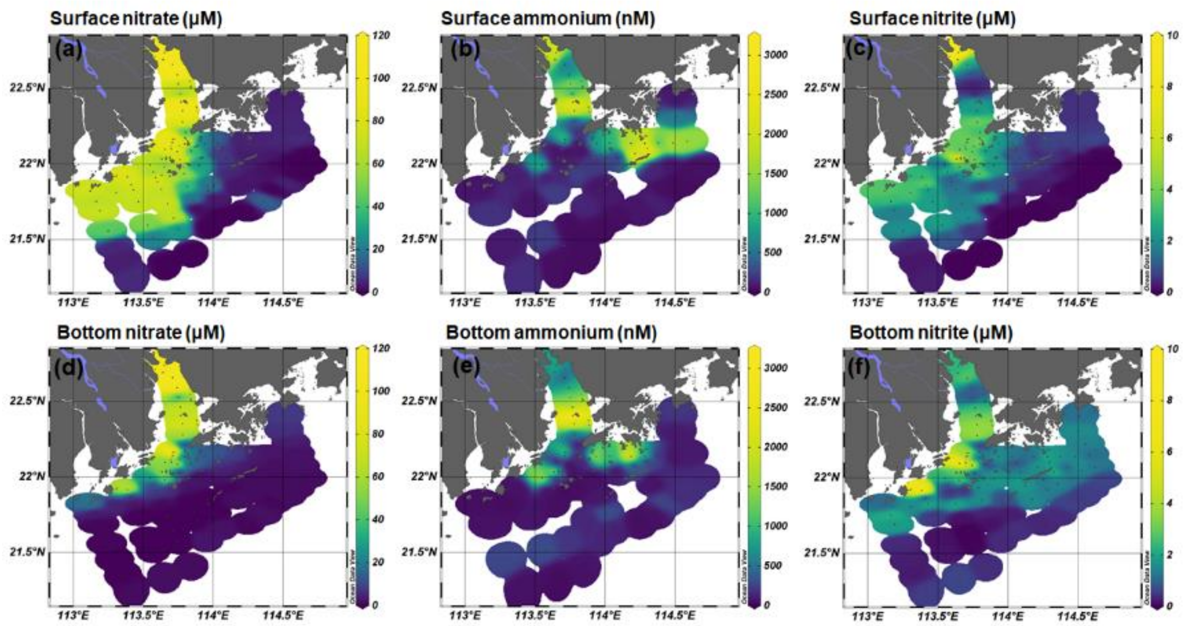
Response: *We have removed the repeated results***.** Page 11 Line 299-300

Fig. 2: figure 2 consists of a selection of graphs to show the most interesting pattern among the environmental parameters measured. This is alright, but the rest of the graphs needs to be provided as well (e.g. supplementary info). For example, surface nitrate concentrations and bottom nitrite concentrations are shown, but bottom nitrate concentrations and bottom salinity are missing.

Response: *We have moved all nutrient plots to the supplementary materials. The current version of figure 2 showed below contains the spatial pattern of salinity, chlorophyll-a and DO concentration at both surface and bottom layer. The nutrient plots of nitrate, nitrite and ammonia were moved to supplementary in Figure S3.* Page 24 Line 627-631; Supplementary Figure S3



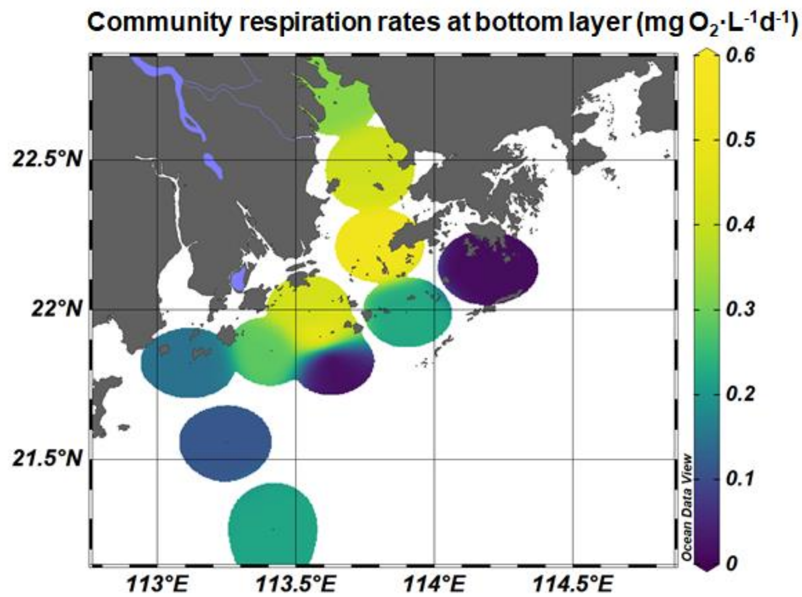
(Revised) Figure 2. Spatial distribution of (a & d) salinity, (b & e) chlorophyll-a, and (c & f) dissolved oxygen concentration at both surface and bottom layer during the 2017 summer cruise in Pearl River estuary. These figures were generated using Ocean Data View v. 5.0.0 (http://odv.awi.de).



(Newly added) Figure S3. Spatial distribution of (a & d) nitrate, (b & e) ammonium, and (c & f) nitrite concentration at both surface and bottom layer during the 2017 summer cruise in Pearl River estuary. These figures were generated using Ocean Data View v. 5.0.0 (http://odv.awi.de).

Fig. 3c: Data are only plotted for a fraction of the stations compared to 3a and b. Why is a part of the data missing?

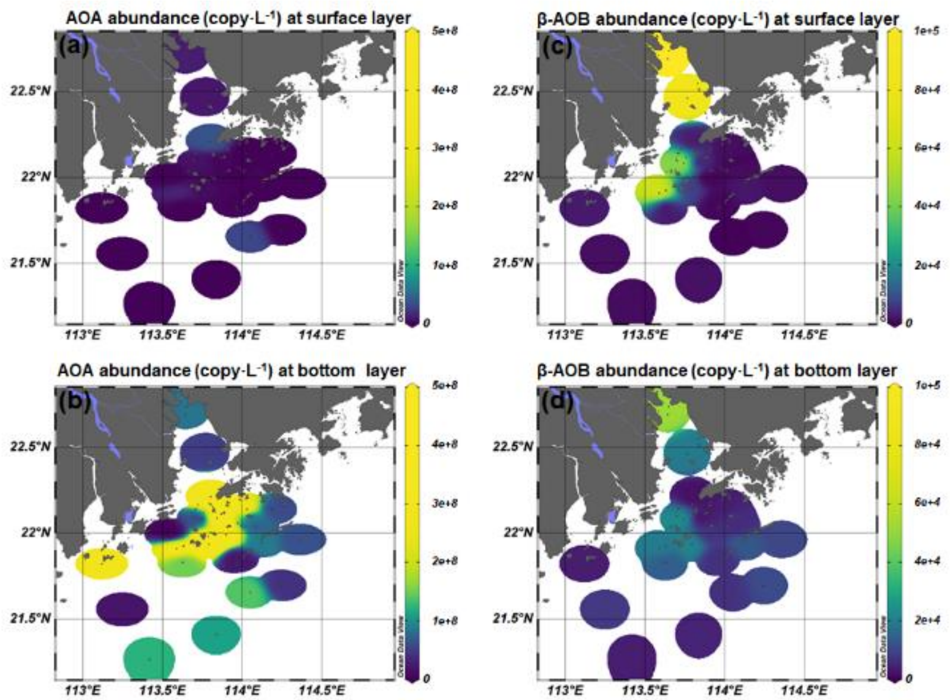
Response: *The comparisons were only performed for stations where community respiration rates were measured. We did not conduct the measurements of community respiration rates at many stations as we did for the nitrification rates. The spatial distribution of community respiration rates at the bottom layer was newly added as Figure S4 in supplementary. The citations of these figures were revised accordingly.*



(Newly added) Figure S4. Spatial distribution of community respiration rates at the bottom layer (mg O_2·$L^{-1}d^{-1}$).

Fig. 4: please provide the scale in the same number format for AOA and AOB. In order to compare abundances between surface layer and bottom layer please use the same range for the scale for 4a and c and b and d respectively.

Response: *We have changed the number format and used same scale range for corresponding figures in Figure 4. (new version is attached below and Figure 4 in the main text had been replaced with this new version).* Page 26 Line 636-638



(Revised) Figure 4. Spatial distribution of AOA and β-AOB abundance at the surface and the bottom layer at DNA level.

Fig. 9: you include the temperature in the Spearman correlation in this table. Therefore, you should also provide the temperature data. Maybe add them to table S2.

Response: *We have added "Temperature" in table S2.* Supplementary information Table S2

Fig.9 and l. 391: How did you quantify heterotrophic bacteria? With the cell quantification method, you reported in the material and method section heterotrophic microbes cannot be distinguished from autotrophic non-phototrophic microbial cells (such as the nitrifiers that this study focuses on).

Response: *We admit that flow cytometry method cannot distinguish the autotrophic non-phototrophic microbial cells. We have changed the term in to "non-phototrophic prokaryotic cells" with abbreviation "NPC" in the figure legend in Figure 9.* Page 30 Line663; Page11 Line279; Page 14 Line 401-402

technical corrections

As pointed out above, there are too many issues throughout the manuscript to address here. Some selected comments:

l. 42 "Based on the" instead of "as revealed by"

Response: *We have revised "as revealed by" to "Based on the"* Page 3 Line 41

l. 47 The WCA, WCB, and SCM1-like groups correspond...

Response: *We have revised accordingly.* Page 3 Line 46

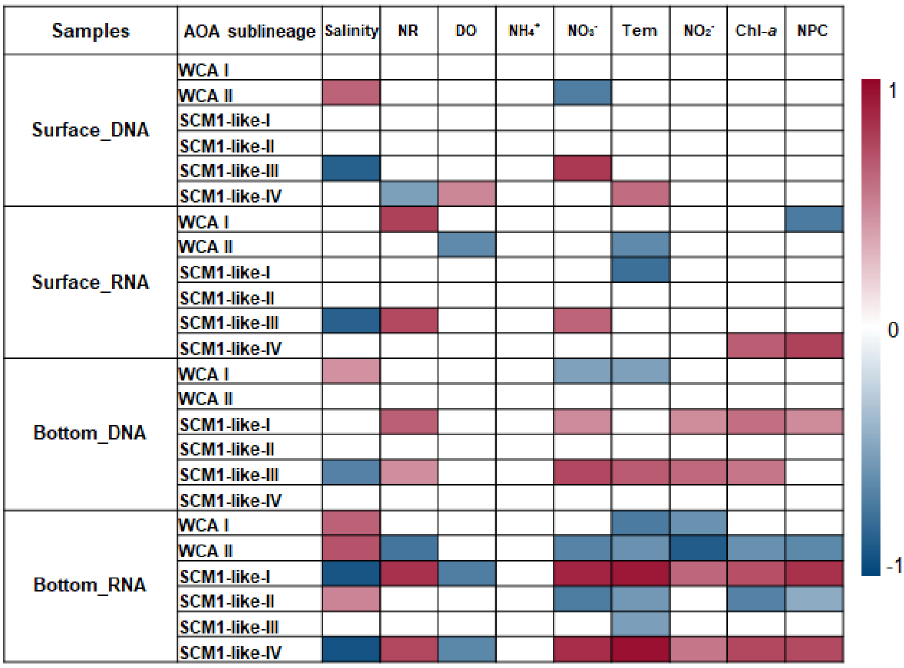
l. 102 introduce the abbreviation CR in line 93

Response: *We have added abbreviation "CR" in line 93.* Page 5 Line 93

Fig. 9: this is a table not a figure. Typos in the first column: Surface.

Response: *Sorry for the typo. We have corrected it. We considered this heatmap as a figure.*

Page 30 Line 663-667. *It is now figure 10.*



(Revised) Figure 10. Spearman correlation between AOA sublineages (relative abundance at DNA and RNA levels) and environmental factors in the surface and bottom layers of the water column in the Pearl River estuary during summer 2017. Only the significant correlations ($P<0.05$) are displayed (NR-nitrification rates; DO-dissolved oxygen; Tem-Temperature; NPC-non-phototrophic prokaryotic cells).

Reference

Casciotti, K. L., D. M. Sigman, M. G. Hastings, J. K. Bohlke, and Hilkert, A. : Measurement of the oxygen isotopic composition of nitrate in seawater and freshwater using the denitrifier method., Anal. Chem., 74, 4905–4912, https://doi.org/10.1021/ac020113w, 2002.

Knapp, A. N., D. M. Sigman, and Lipschultz, F. : N isotopic composition of dissolved organic nitrogen and nitrate at the Bermuda Atlantic time-series study site, Global Biogeochem. Cycle, 19, https://doi.org/10.1029/2004gb002320, 2005.

Sigman, D. M., K. L. Casciotti, M. Andreani, C. Barford, M. Galanter, and Bohlke, J. K. : A bacterial method for the nitrogen isotopic analysis of nitrate in seawater and freshwater, Anal. Chem., 73, 4145–4153, https://doi.org/10.1021/ac010088e, 2001.

(Revised) Table S2. Quantitative PCR results at DNA level of both AOA and β-AOB in 23 stations

Station	Lon (E °)	Lat (W °)	Layer	Salinity (PSU)	DO (mg·L⁻¹)	Temperature (℃)	Ammonium (nmol·L⁻¹)	Nitrification rate (nmol·L⁻¹·h⁻¹)	AOA-PA (Copy·L⁻¹)	AOA-FL (Copy·L⁻¹)	AOB-PA (Copy·L⁻¹)	AOB-FL (Copy·L⁻¹)
F107	113.42	21.27	S-1m	32.30	4.53	29.07	155.70	0.21	1.54E+04 ± 1.35E+03	7.93E+04 ± 4.04E+03	1.81E+02 ±3.02E+01	8.05E+02 ±1.04E+02
			B-41m	34.51	4.09	22.77	48.64	0.96	3.31E+04 ±7.10E+03	1.22E+08 ±3.06E+06	7.77E+02 ±1.57E+02	3.03E+03 ±2.97E+02
F104	113.25	21.56	S-1m	16.69	6.80	31.01	ND	0.14	2.92E+04 ±8.54E+02	1.27E+05 ±1.27E+04	4.90E+02 ±1.11E+02	7.56E+02 ±1.60E+02
			B-28m	34.45	4.26	24.06	ND	0.33	1.09E+06 ±6.11E+04	1.76E+07 ±3.61E+05	5.17E+03 ±7.73E+02	2.83E+03 ±6.77E+02
F101	113.12	21.82	S-1m	10.20	6.38	29.29	67.03	1.18	4.20E+04 ±5.67E+03	1.19E+06 ±3.79E+04	1.11E+02 ±4.40E+01	2.57E+03 ±1.87E+02
			B-9m	33.73	0.54	24.18	34.78	36.62	2.61E+07 ±2.00E+05	3.95E+08 ±4.51E+06	1.67E+03 ±3.30E+02	2.00E+03 ±3.71E+02
F309	113.84	21.41	S-1m	33.91	4.47	29.74	32.41	ND	1.24E+03 ±6.11E+01	2.67E+05 ±1.08E+04	1.31E+02 ±4.05E+01	1.35E+03 ±4.02E+02
			B-43m	34.51	4.21	22.36	56.68	0.40	1.31E+05 ±2.48E+04	1.10E+08 ±4.61E+06	2.57E+03 ±7.72E+02	2.02E+03 ±4.51E+02
F305	113.63	21.83	S-1m	9.04	7.08	30.52	233.66	1.84	4.83E+04 ±9.26E+02	3.21E+05 ±2.04E+04	4.77E+02 ±5.88E+01	8.42E+02 ±1.01E+02
			B-26m	34.43	3.47	23.80	44.11	1.28	7.27E+07 ±2.47E+06	7.42E+07 ±4.36E+06	1.08E+04 ±9.10E+02	2.80E+03 ±2.97E+02
F303	113.59	21.91	S-1m	7.54	6.82	30.14	104.01	0.48	7.55E+06 ±2.29E+05	6.09E+06 ±1.17E+05	2.89E+04 ±1.95E+03	3.42E+04 ±3.47E+02
			B-18m	34.45	1.44	23.40	42.73	36.37	1.40E+08 ±1.25E+07	1.62E+08 ±3.61E+06	1.65E+04 ±3.31E+03	3.16E+03 ±5.28E+02
F301	113.55	21.99	S-1m	6.70	7.67	29.12	865.79	5.20	5.80E+04 ±2.19E+03	3.29E+04 ±3.53E+03	ND	ND
			B-6m	23.17	2.10	27.25	1423.19	41.94	5.04E+03 ±1.72E+03	3.54E+05 ±3.49E+04	ND	ND
F405	113.79	21.94	S-1m	12.29	6.53	29.05	250.81	1.48	2.48E+05 ±8.02E+03	2.65E+06 ±3.61E+04	9.73E+02 ±3.05E+02	6.54E+03 ±1.14E+03
			B-22m	34.43	2.61	23.65	34.19	1.04	5.88E+07 ±2.47E+06	4.39E+08 ±1.24E+07	1.10E+04 ±2.10E+03	1.08E+04 ±1.94E+03
F403	113.74	22.08	S-1m	7.56	4.11	28.85	24.08	3.07	2.02E+06 ±4.77E+04	3.63E+06 ±1.86E+05	9.57E+03 ±1.94E+03	3.62E+04 ±6.24E+02
			B-8m	22.46	1.31	26.19	24.16	9.91	1.42E+07 ±7.22E+05	3.11E+07 ±1.73E+05	7.75E+03 ±7.65E+02	1.59E+04 ±1.23E+03
A16	114.05	21.66	S-1m	33.67	4.73	29.77	35.32	ND	1.70E+07 ±6.61E+04	1.33E+07 ±6.36E+05	ND	ND
			B-45m	34.52	4.21	22.01	111.37	0.65	3.90E+07 ±2.03E+06	9.95E+07 ±1.32E+06	6.91E+03 ±9.79E+02	2.12E+01 ±7.46E+00

Station	Lon (E °)	Lat (W °)	Layer	Salinity (PSU)	DO (mg·L^{-1})	Temperature (℃)	Ammonium (nmol·L^{-1})	Nitrification rate (nmol·L^{-1}·h^{-1})	AOA-PA (Copy·L^{-1})	AOA-FL (Copy·L^{-1})	AOB-PA (Copy·L^{-1})	AOB-FL (Copy·L^{-1})
A14	113.96	21.85	S-1m	24.15	5.26	29.98	69.85	0.44	1.20E+05	1.16E+06	ND	4.77E+02
									±5.63E+03	±4.58E+04		±8.29E+01
			B-25m	34.39	4.00	24.21	355.19	0.06	5.12E+06	1.50E+07	4.68E+03	1.85E+03
									±1.12E+05	±1.73E+05	±4.56E+02	±2.95E+02
A12	113.90	21.99	S-1m	19.56	6.68	29.82	278.65	0.80	9.21E+05	2.73E+05	1.80E+02	2.25E+01
									±3.39E+04	±2.98E+04	±5.64E+01	±9.03E+00
			B-22m	34.41	2.62	26.63	56.18	1.13	6.00E+07	2.61E+08	3.69E+03	3.37E+03
									±3.05E+06	±6.08E+06	±7.40E+02	±5.25E+02
A11	113.84	22.09	S-1m	13.88	6.37	28.72	47.10	1.13	1.24E+06	6.56E+05	2.69E+01	2.83E+03
									±2.30E+04	±4.11E+04	±4.30E+00	±2.58E+01
			B-13m	32.15	0.97	24.56	120.77	2.64	1.02E+08	2.58E+08	1.49E+03	6.81E+02
									±4.86E+06	±1.42E+07	±6.58E+01	±3.59E+01
A09	113.80	22.21	S-1m	17.52	5.39	27.93	161.39	2.58	1.36E+06	3.50E+07	2.56E+02	2.60E+03
									±7.81E+04	±8.62E+05	±2.95E+01	±1.97E+01
			B-21m	33.36	1.15	24.18	91.45	22.43	4.73E+07	3.85E+08	1.10E+03	8.10E+02
									±2.54E+06	±9.50E+06	±2.55E+02	±1.56E+02
A05	113.77	22.46	S-1m	2.28	3.27	28.68	865.84	1.90	5.07E+06	3.77E+06	6.03E+04	3.52E+04
									±2.33E+05	±5.77E+04	±7.06E+03	±1.39E+03
			B-10m	14.96	2.45	26.79	1673.87	35.10	2.04E+07	2.93E+07	1.92E+04	8.13E+01
									±1.92E+05	±3.61E+05	±5.36E+02	±5.26E+00
A01	113.65	22.74	S-1m	0.11	2.00	28.44	2043.89	94.78	9.76E+06	1.74E+06	8.79E+04	1.92E+04
									±5.80E+05	±4.56E+05	±2.43E+03	±1.42E+03
			B-11m	0.11	1.93	27.46	786.73	17.32	5.08E+07	3.26E+07	4.18E+04	1.04E+04
									±4.06E+06	±5.56E+06	±3.50E+03	±9.35E+02
F607	114.24	21.69	S-1m	32.74	4.88	28.74	61.84	ND	2.08E+03	6.07E+04	3.70E+01	5.30E+02
									±3.57E+02	±3.75E+03	±7.50E+00	±1.88E+02
			B-45m	34.49	4.51	22.52	483.80	1.33	3.32E+05	4.07E+07	7.57E+03	2.97E+03
									±9.85E+03	±4.93E+05	±5.13E+02	±4.89E+02
F605	114.12	21.95	S-1m	30.11	4.64	28.10	ND	1.91	4.98E+03	1.29E+06	1.11E+02	2.07E+03
									±1.16E+03	±6.16E+04	±3.14E+01	±1.56E+02
			B-35m	34.39	2.75	23.90	ND	7.08	1.53E+07	7.23E+07	8.69E+03	4.27E+03
									±3.31E+06	±3.15E+06	±2.22E+03	±2.48E+02
F603	114.09	22.04	S-1m	29.09	4.46	28.30	358.38	1.68	1.78E+03	1.44E+06	5.56E+01	8.82E+02
									±4.75E+02	±4.94E+05	±1.38E+01	±4.80E+01
			B-27m	34.40	2.42	23.74	79.18	2.97	1.13E+07	6.04E+07	2.65E+03	3.12E+03
									±8.58E+05	±2.25E+06	±9.33E+02	±5.23E+02
F602	114.06	22.10	S-1m	27.08	4.86	28.96	ND	0.33	6.10E+03	4.69E+05	6.18E+01	2.17E+02
									±2.52E+03	±1.54E+05	±1.19E+01	±8.47E+01
			B-22	34.27	1.56	23.79	ND	4.36	2.68E+06	6.48E+07	4.47E+03	2.32E+03
									±8.65E+05	±2.35E+06	±1.21E+03	±6.52E+02
F601	114.03	22.14	S-1m	25.32	5.09	28.38	983.39	16.09	3.58E+04	7.92E+04	4.85E+01	1.29E+03

Station	Lon (E °)	Lat (W °)	Layer	Salinity (PSU)	DO (mg·L^{-1})	Temperature (°C)	Ammonium (nmol·L^{-1})	Nitrification rate (nmol·L^{-1}·h^{-1})	AOA-PA (Copy·L^{-1})	AOA-FL (Copy·L^{-1})	AOB-PA (Copy·L^{-1})	AOB-FL (Copy·L^{-1})
									±1.26E+03	±1.26E+04	±2.16E+01	±1.18E+02
			B-19m	32.98	0.53	24.49	372.06	7.22	1.68E+06 ±3.91E+05	3.04E+08 ±4.51E+06	1.03E+03 ±1.03E+02	2.22E+03 ±1.10E+03
F701	114.18	22.14	S-1m	26.57	4.63	28.54	1682.83	0.51	1.33E+03 ±5.22E+02	4.86E+05 ±6.24E+04	ND	ND
			B-22m	34.16	1.18	23.88	1993.45	19.13	7.90E+05 ±3.50E+04	5.41E+07 ±9.33E+06	ND	ND
F804	114.36	21.96	S-1m	31.78	4.47	28.70	121.59	0.05	2.43E+03 ±8.98E+02	7.00E+05 ±1.88E+04	1.14E+02 ±9.51E+01	1.14E+03 ±1.81E+02
			B-29m	34.47	3.46	22.91	55.20	2.86	1.47E+07 ±1.69E+06	4.71E+07 ±2.78E+06	6.91E+03 ±3.15E+02	3.16E+03 ±2.24E+03

* S-Surface; B-Bottom; PA-Particle attached (> 3 μm); FL-Free-living (3-0.2 μm); ND-Under detection limit.

(Revised) Table S3. Nitrification, community respiration rates and corresponding oxygen demand.

Station	Layer	Nitrification rate ($nmol \cdot L^{-1} \cdot h^{-1}$)	Nitrification oxygen Demand ($mg\ O_2 \cdot L^{-1} \cdot d^{-1}$)	Community respiration rate ($mg\ O_2 \cdot L^{-1} \cdot d^{-1}$)	NOD/CR%
F101	S	1.1770±0.0447	0.0014	1.4400±0.3024	0.094
F101	B	36.6152±0.1790	0.0422	0.1499±0.0021	28.137
F104	S	0.1443±0.0055	0.0002	1.6813±0.2433	0.010
F104	B	0.3277±0.0433	0.0004	0.1146±0.1568	0.330
F107	S	0.2057±0.0121	0.0002	0.2264±0.0722	0.105
F107	B	0.9596±0.0609	0.0011	0.2191±0.1756	0.505
F301	S	5.1961±0.0285	0.0060	1.1372±0.1240	0.526
F301	B	41.9434±0.4959	0.0483	0.4283±0.1175	11.282
F303	S	0.4847±0.0033	0.0006	1.0797±0.1843	0.052
F303	B	36.3678±1.0384	0.0419	0.5141±0.1635	8.150
F305	S	1.8411±0.2199	0.0021	0.6203±0.1090	0.342
F305	B	1.2795±0.3351	0.0015	0.0023±0.0017	64.894
F701	S	0.5144±0.1081	0.0006	0.9343±0.1157	0.063
F701	B	19.1291±1.0963	0.0220	0.0121±0.1519	181.913
A14	S	0.4443±0.058	0.0005	1.0191±0.1596	0.050
A14	B	0.0609±0.0059	0.0001	0.8222±0.2808	0.009
A12	S	0.8040±0.0692	0.0009	0.9928±0.4831	0.093
A12	B	1.1319±0.0479	0.0013	0.2256±0.0743	0.578
A09	S	2.5768±0.1457	0.0030	1.3144±0.2086	0.251
A09	B	22.4347±0.6230	0.0258	0.6340±0.1077	4.525
A05	S	1.9032±0.186	0.0022	0.2582±0.0848	0.849
A05	B	35.0975±2.5993	0.0404	0.4280±0.0347	9.446
A01	S	94.7793±12.3754	0.1092	0.6128±0.1521	17.819
A01	B	17.3175±0.3106	0.0199	0.3231±0.1861	6.175

* S-Surface; B-Bottom.

(Revised) Table S4. Quantitative PCR results of cDNA (template for RNA level) of AOA and β-AOB in 13 stations

Station	AOA-PA (copy·L^{-1})	AOA-FL (copy·L^{-1})	AOB-PA (copy·L^{-1})	AOB-FL (copy·L^{-1})
A01	3.10E+03 ±1.12E+01	3.08E+03 ±7.11E+02	ND	ND
A01	ND	1.16E+03 ±7.70E+02	ND	ND
A05	8.24E+02 ±4.30E+02	1.02E+04 ±1.84E+03	ND	ND
A05	1.30E+03 ±8.48E+02	6.03E+02 ±3.48E+02	ND	ND
A09	ND	1.18E+05 ±1.06E+04	ND	ND
A09	1.77E+03 ±1.76E+03	1.47E+06 ±1.07E+05	ND	ND
A11	ND	2.56E+03 ±8.36E+02	ND	ND
A11	3.61E+04 ±3.64E+03	1.14E+05 ±1.30E+04	ND	ND
A16	ND	ND	ND	ND
A16	2.62E+04 ±6.64E+03	ND	ND	ND
F101	ND	1.82E+03 ±5.00E+02	ND	ND
F101	7.43E+03 ±1.46E+03	1.87E+04 ±2.70E+03	ND	ND
F104	ND	1.43E+03 ±4.38E+02	ND	ND
F104	1.21E+03 ±7.13E+01	8.26E+03 ±8.37E+02	ND	ND
F107	ND	ND	ND	ND
F107	ND	1.74E+06 ±5.89E+03	ND	ND
F301	2.99E+03 ±1.07E+03	ND	ND	ND
F301	5.09E+03 ±1.15E+02	1.85E+05 ±1.73E+04	ND	ND
F305	ND	8.07E+02 ±5.65E+02	ND	ND
F305	1.05E+04 ±1.44E+03	9.98E+03 ±1.62E+03	ND	ND
F403	6.46E+03	1.18E+05	ND	ND

	±1.26E+03	±1.78E+04		
F403	3.30E+03 ±1.14E+03	1.17E+05 ±9.54E+03	ND	ND
F601	ND	ND	ND	ND
F601	4.28E+03 ±5.20E+02	3.21E+06 ±1.67E+05	ND	ND
F603	ND	3.72E+03 ±3.08E+02	ND	ND
F603	1.03E+03 ±7.51E+01	2.50E+05 ±3.04E+04	ND	ND

* S-Surface; B-Bottom; PA-Particle attached (>3 µm); FL-Free-living (3-0.2 µm); ND-Under detection limit.