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## ***Interactive comment on “Diversity and distribution of *amoA*-type nitrifying and *nirS*-type denitrifying microbial communities in the Yangtze River Estuary” by Y. Zhang et al.***

**Anonymous Referee #2**

Received and published: 8 January 2014

This study presents an interesting dataset on the abundance and diversity of bacterial and archaeal *amoA* gene, and the dissimilatory nitrite reductase *nirS* gene of denitrifiers along two transects in the Yangtze River Estuary during spring and summer in the case of the transect along a salinity gradient. In addition, during the summer cruise, both the particle-associated and the free-living microbial communities were analyzed. Overall, the paper provides valuable information on the diversity and distribution of nitrifiers and denitrifiers in estuarine ecosystems. Interestingly, the authors find an apparently higher coupling between potential nitrification and denitrification in the particle-associated compared to the free-living fraction, based on the abundance of *amoA* and *nirS* genes. Despite the overall quality and interest of the manuscript I

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have some comments which should be revised and addressed before publication. One aspect greatly hindering the conclusion of whether AOA or AOB may contribute more to nitrification is that abundance, as determined by quantification of amoA gene copy numbers, does not necessarily reflect an actual contribution to nitrification. The same holds for the conclusion of higher nitrification vs denitrification potential, based on gene copy numbers. Several recent studies found no direct correlation between AOA abundance and potential nitrification rates in both marine, estuarine and soil ecosystems. Therefore the author should carefully revise their statements and adequately discuss their results.

Specific comments Page 2, lines 6-10. Please revise the order of words in this sentence. Page 2. Line 15, 21. What do the authors mean by “distinctly”? Throughout the text is not clear if they use this term as a synonym of “significantly”. Page 2, lines 22-24. Please indicate which correlations. Page 8, lines 25-27 and page 9, lines 1-11. The description of the environmental conditions is very poor. It is not clear at which depth the samples for analysis were taken (the authors just mention surface and bottom waters). I suggest including at least a table with all the biogeochemical variables for each sampling point. Page 9, lines 9-11. The authors should clearly explain these patterns. Page 11, lines 19-29. This description of the abundances is hard to follow. Also I do not think table 1 is the best way to represent these data. I think that a graphic presentation would help to compare between samples and periods. Page 12, lines 1-19. There are several fragments here that should go in the discussion rather than in the results section. Page 12, lines 22. The authors should include the error bars in figure 7. Page 13, lines 5-6. I do find figure 8 rather complicate and not much illustrative of what the authors try to show. I suggest just representing nitrification rates vs amo A gene abundance using different symbols for the particulate and the free-living fractions. Also I do not think that the regression equations make any sense. Page 13, lines 7-10. Please revise this sentence, as it is, it makes not much sense. I guess the authors mean something like: “In the latitudinal transect, amoA and nirS genes in the free-living fraction mostly outnumbered those in the particle-associated fraction”. Page

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13, lines 20-24. Again, a higher number of amoA or nirS copies does not mean higher nitrification or denitrification rates. Page 13 lines 24-29. Please revise these sentences for English usage. Page 14, lines 14-16. I would delete this part of the sentence "...and the advantages...outer estuary". In addition, this relationship is rather obvious, the more particles, the more number of gene copies in the particle-associated than in the free-living fraction. I wonder if the authors could provide the total abundance of prokaryotes in each sample and fraction. Page 14, lines 18-21. As mentioned above, I do not think that figure 8 is adequate as a correlation between ratios is rather complicated. I think that the authors should represent rates vs gene copies. Page 14, lines 22-29. The authors could simplify this idea, and provide some more cites. Page 15, line 13-14. As already mentioned, the correlations should be between rates and gene copies, not between P:F ratios. Page 15, lines 15-21. Please revise this sentence, it makes no sense. The authors should better explain the undetectable denitrification rates. Page 15, lines 22-25. The authors could calculate the AOB/AOA ratio and see if the ratio changes in relation to environmental factors (such as ammonium concentration). Page 16, lines 11-29, page 17, lines 1-7. This section should be revised for clarity. Moreover, the authors should explain figure 9 in the results section. I think that the authors could simplify figure 9 just representing total amoA gene abundances vs nirS gene abundances. It has been already stated that B-amoA gene abundance was very low. On the contrary, it would be interesting to simultaneously show such relationship in the particle-associated vs the free-living fraction, in order to show the higher potential coupling between both processes in particles. Page 17, lines 9-29, and page 18, lines 1-23. As for the previous section, figures 10 and 11 should also be explained in the result section. I do not find that figure 11 is necessary; the authors can provide the correlation coefficients in the text. Also figure 11c is redundant, the authors have already discussed that there amoA and nirS genes are more abundant in the particle-associated than in the free-living fraction.

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