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Interactive comment on “Ecosystem-specific selection of microbial ammonia oxidizers in an acid soil” by M. Saiful Alam et al.

Anonymous Referee #1

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General comment:

To reveal niche specialization of ammonia-oxidizing archaea and bacteria (AOA and AOB, respectively) in acidic soils, the authors studied the distribution and community composition of both groups along a moisture gradient from a paddy soil to an upland maize soil.

Although the authors did good work, there are a number of weaknesses in the manuscript. In the first place, the authors have analyzed the communities of AOA and AOB with different methods, which are DGGE and clone library construction, respectively. It is very likely that both methods screen the respective communities to a different depth. In the second place, the authors have shown that in the creation of paddy fields from upland soil by flooding these soils for more than hundred years, the soil carbon

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content and the pH increased. Nevertheless, the authors hang their entire argument for the change in communities on the restricted availability of oxygen in flooded soils (that they have not measured), while it is known that both organic nitrogen (as part of the soil carbon) and pH are selective factors in relation to niche specialization between AOA and AOB.

Hence, I believe that the present manuscript is not acceptable for publication and should be rejected.

Specific comments:

Lines 2-3 What about the availability of other nutrients such as phosphate, carbon dioxide and iron. I can imagine that they vary also in acidic soils. The authors may better say “the major energy-generating compounds, i.e. ammonia and oxygen”

Line 10 Insert “a” before “slight decline”

Line 23 Replace “as” by “is”

Lines 28-29 Can this still be stated with the knowledge on the global importance of the Anammox bacteria?

Lines 32-46 Order of the sentences not clear. Say first something on the genomics and metabolism and later something on the mutual distribution and community composition of the AOA and AOB in (acidic) soils

Line 33 Bacteria and Archaea not italic

Lines 48-49 Do natural environments include anthropogenically disturbed ecosystems? Please describe what is meant with natural.

Line 57 Contrasting to what? The former sentence?

Line 75 Insert “the” before “natural environment”

Line 76 Change order to “different long-term fertilizations”

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Line 79 Insert “from” before “maize field”

Line 79 Change “are originated” to “originate”

Line 81 Insert “the” before “soil”

Line 81 Replace “ecological” by “environmental”

Lines 122-124 Samples from the upland soil were collected from 0-20 cm depth and in April, whereas samples from the paddy soil were collected from 0-15 cm depth and in November? Why these differences and could they have affected the outcome of the analyses?

Lines 176-178 What could have been the consequences of comparing AOA and AOB communities by to different methods, i.e. DGGE and cone libraries, respectively? Both methods will yield a different insight in the community composition.

Lines 200-202 Why that specific for the clone library construction and not for the DDGE?

Line 239 and following

This paragraph is apparently meant to compare the paddy and the upland soil in general. Saying then something on the effect of N amendments makes it confusing as it seems the next paragraph is meant to describe the effects of fertilization sec. Hence, I would suggest to omit the sentence on the N amendments starting in line 246. The same holds for lines 252 – 254.

Line 243 Omit “particularly” as the whole statement deals already with paddy soils.

Lines 249-250 Are such differences really significant?

Lines 288-289 In contrast to the text, Figure 2 presents triplicates.

Line 292 Figure 2 shows only the DGGE bands in CK, P and K. Why not N? The latter could have been more interesting in relation to the function of the AOA.

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Lines 294-295 Could one conclude from the numbers of DGGE bands that the paddy soils are more diverse? With DGGE only the dominant genes are shown, while a large part of the diversity can still be hidden. Maybe, one could say that the paddy soil showed more evenness with respect to the bands of *amoA* genes.

Lines 301-304 I miss the quantitative data that would underpin statements about dominance or changes in community composition. A phylogenetic tree is hardly the instrument to do so.

Lines 308-330 The text is only based on visual inspection of the DGGE patterns. I wonder why the authors did not apply statistical methods like the ANOSIM method to compare communities of AOA in the different treatments in the two soils.

Lines 331-338 It would be helpful to include a table with the numbers found in each OTU in each treatment in each soil type.

Lines 341-343 Please rephrase, as the present sentence is hard to follow.

Lines 346-348 The authors stress the role of oxygen in the selection of AOA and AOB in flooded soils. However, as we can see from Table 1, also the amount of carbon and the pH changes. Both can also be a selective factor towards the selection of different ammonia oxidizers, especially when the carbon also comprises organic nitrogen.

Line 351 Insert “the” before “field”

Line 353 Insert “act” after “oxygen and ammonia”

Line 354 “experiments” instead of “experiment”

Line 357 Replace “are originated” by “originate”

Lines 360-361 Transfer “communities” to “structures”, i.e. “community structure of obligate aerobic AOA and AOB”

Line 361 Omit the first “AOA” as ratio is sufficient here.

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Lines 362-363 Again, is it only oxygen that selects the distinct communities?

Line 371-372 Why would oligotrophic lakes be microaerophilic?

Line 385 Change “maintenance and emergence” in “emergence and maintenance”

Line 411 Change “growth parameters” to “kinetic parameters” as the experiments had been done in continuous culture at fixed growth parameters.

Line 414 Insert “that” before “AOB communities”

Lines 414-415 Omit “deprivation” as this is already indicated by “severe starvation”

Lines 415-416 Change in “the relationship between oxygen status and AOA and AOB in natural habitats remains poorly understood”

Lines 430-433 Rephrase, as the sentence is hard to understand.

Lines 433-453 Is this conclusion not based on the use of different methods that have been applied to analyze the communities of AOA and AOB?

Line 448 Again, only oxygen?

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