Managing Nitrate in Agricultural Soils: Organisms and Process

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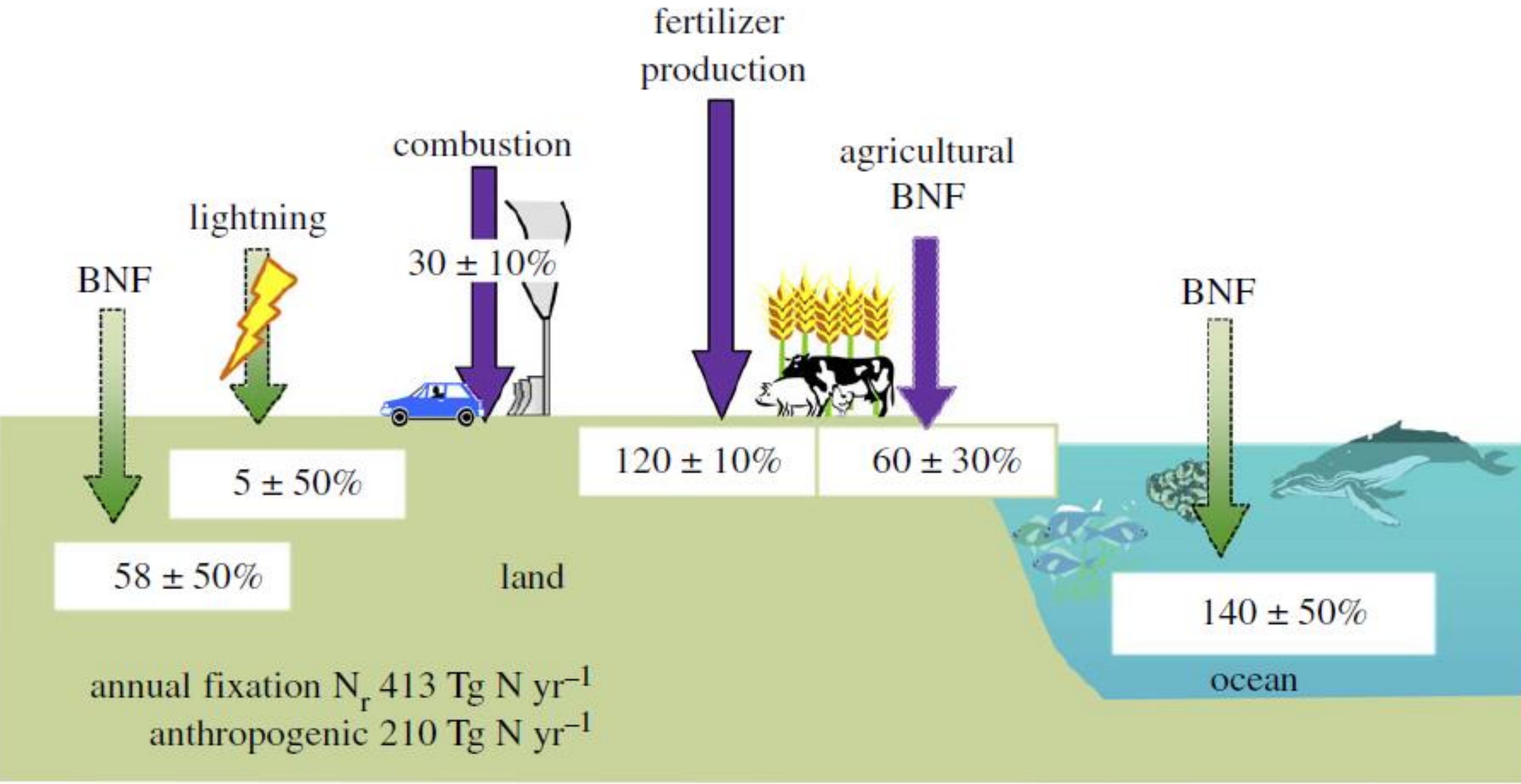
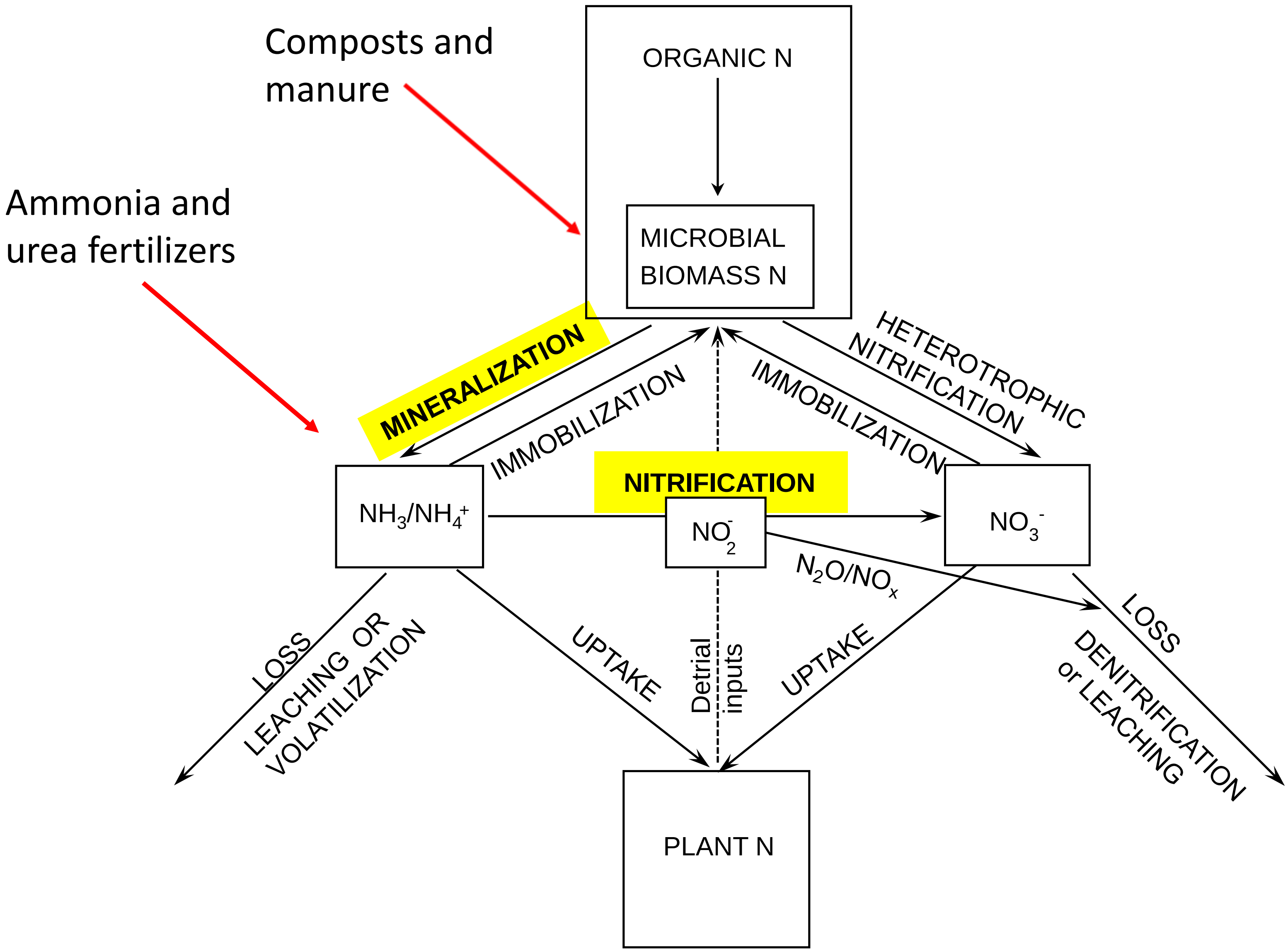
On a global basis reactive N inputs related to human activities now exceeds natural N inputs



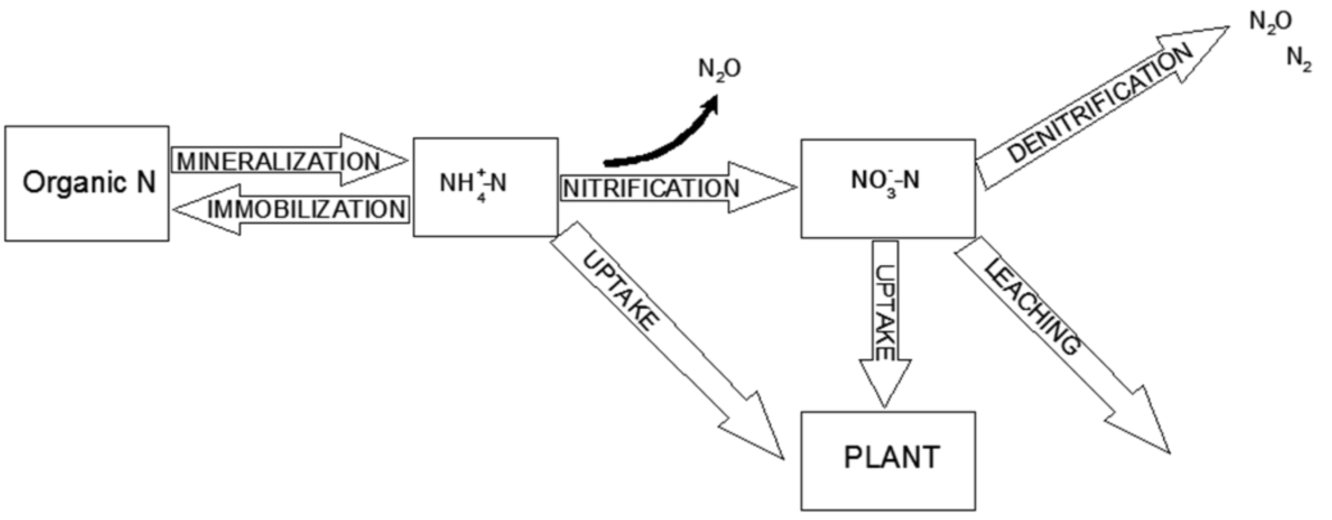
From Fowler, D., M. et al. 2013. The global nitrogen cycle in the twenty-first century. Philosophical Transactions of the Royal Society B-Biological Sciences 368.

Soil Internal Nitrogen Cycle



Norton and Stark 2011

Why study nitrification and nitrogen mineralization and in agricultural systems?



- N mineralization and nitrification are key N-transformations that largely determine the availability and mobility of N in soils
- Nitrification leads to soil N_2O emissions and nitrate leaching
- Typical N use efficiencies for fertilizers are often less than 50% due to N loss
- Decrease the impacts of N loss and increase N fertilizer use efficiency in agricultural ecosystems, aim to match soil N supply with plant demand

Nitrification is a unique multi-step process

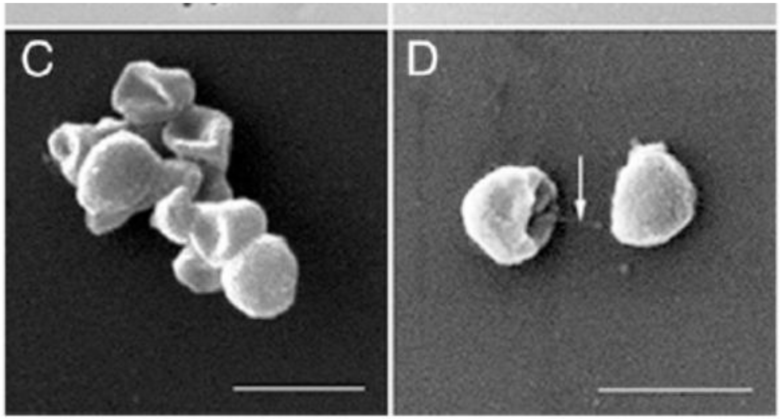
ammonia oxidation to nitrite

$$2NH_4^+ + 3O_2 \rightarrow 2NO_2^- + 2H_2O$$

Enzyme ammonia monooxygenase *amoA* gene

AOA

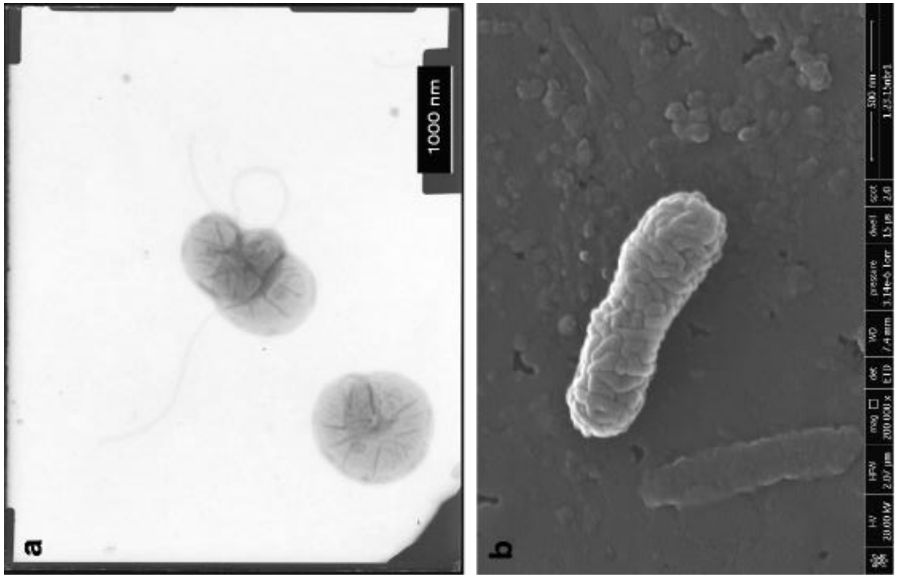
Ammonia oxidizing archaea
Nitrososphaera



Tourna et al 2011

AOB

Ammonia oxidizing bacteria
Nitrosospira



Rice, Norton et al 2016

Nitrification is a unique multi-step process

nitrite oxidation

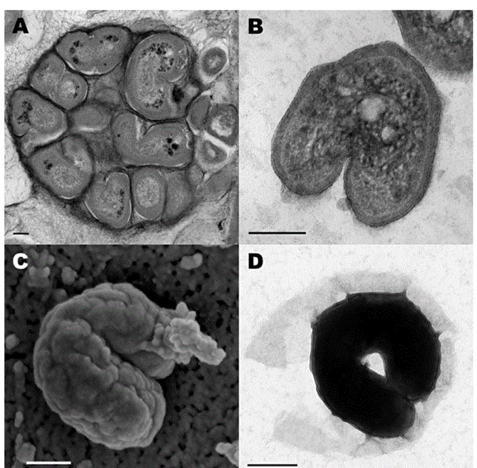
$$2NO_2^- + O_2 \rightarrow 2NO_3^-$$

Enzyme nitrite oxidoreductase encoded by *nxrB gene*

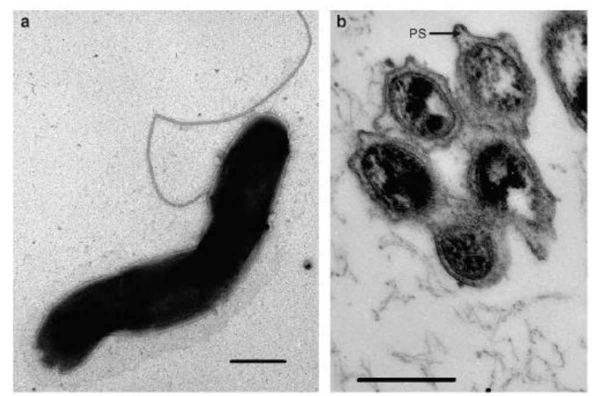
NOB

Nitrite oxidizing bacteria
Nitrospira

Then just to make things interesting
Some *Nitrospira* can do both
Comammox



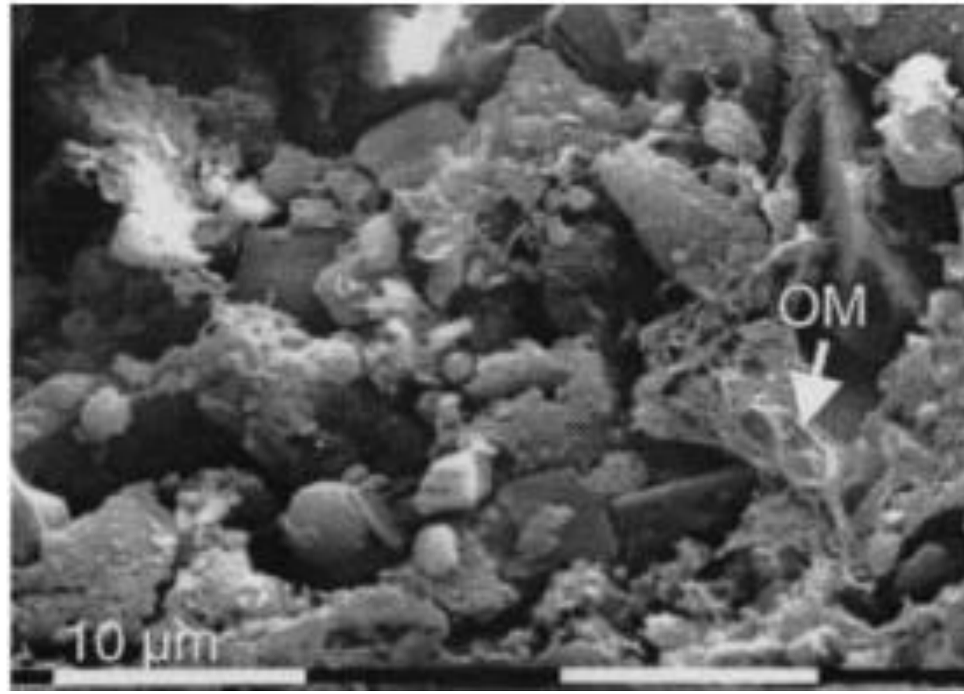
Keuter et al., 2011



Daims et al., 2015

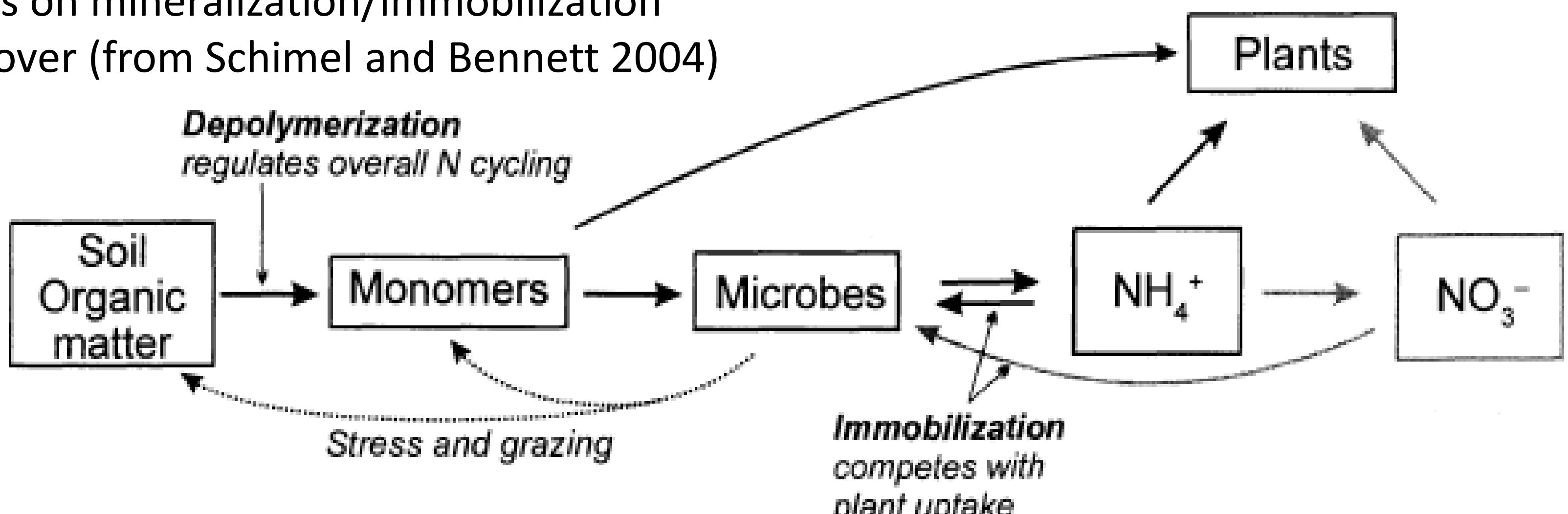
Mineralization

Diverse community of bacteria and fungi and a wide range of enzymes including proteases, chitinases and urease decompose organic matter



Chenu et al. 2001 BFS 34:349

Focus on mineralization/immobilization turnover (from Schimel and Bennett 2004)



What is controlling actual rates of nitrification in agricultural soils?

Environmental conditions

Temperature
Moisture/ Aeration
pH

Substrate supply

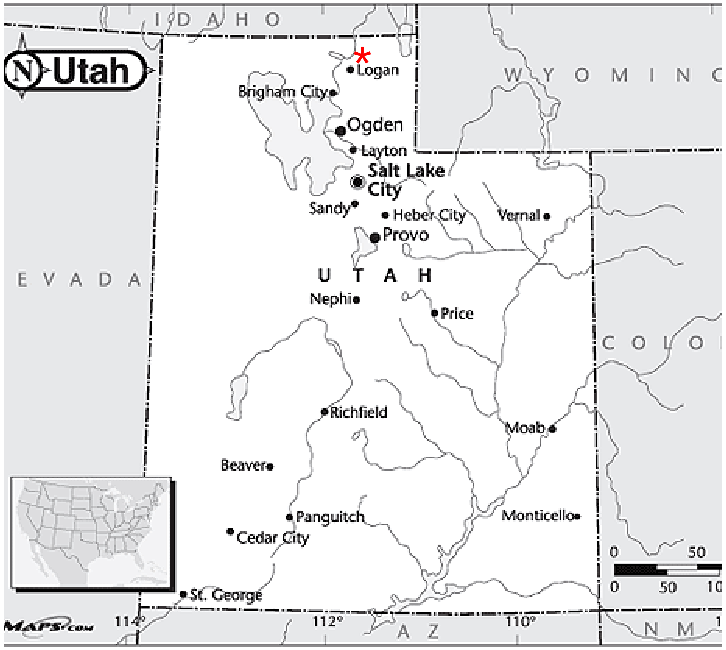
Fertilization
Inorganic nitrogen: pools or turnover
Organic nitrogen pools
Rates of ammonium production - mineralization
Substrate characteristics (C/N and complexity)

Microbial actors

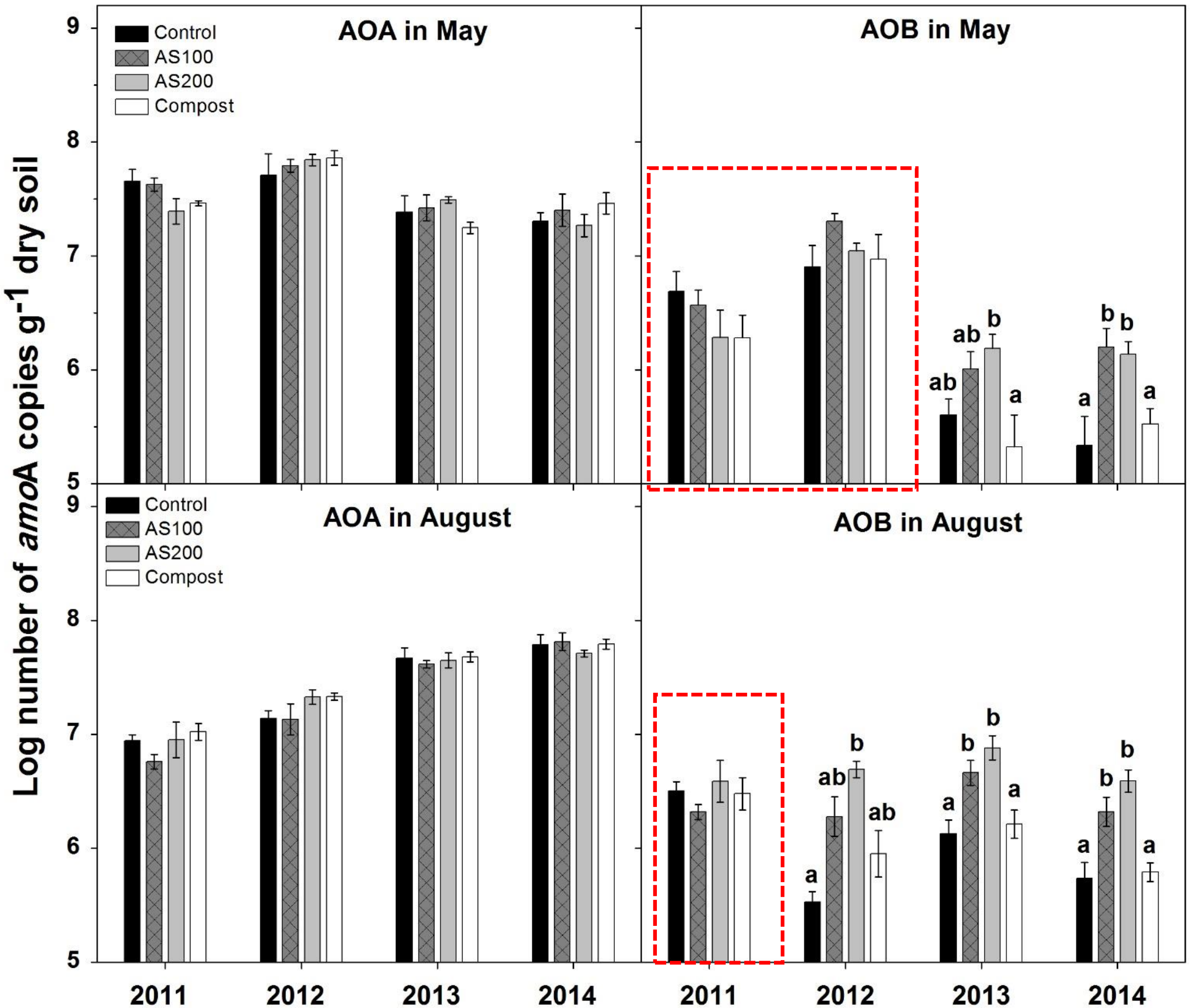
Populations of ammonia oxidizers, nitrite oxidizers
Types of ammonia oxidizers, nitrite oxidizers

Utah State University Site – N Cycle Plots

Strongly calcareous Millville silt loam pH 8.0, irrigated
Field plots planted to corn, started 2011
CRB 4 N source treatments x 4 reps
Control (no additional N)
Ammonium sulfate (AS100 & 200 kg N ha^{-1})
Composted steer manure (200 kg N ha^{-1})



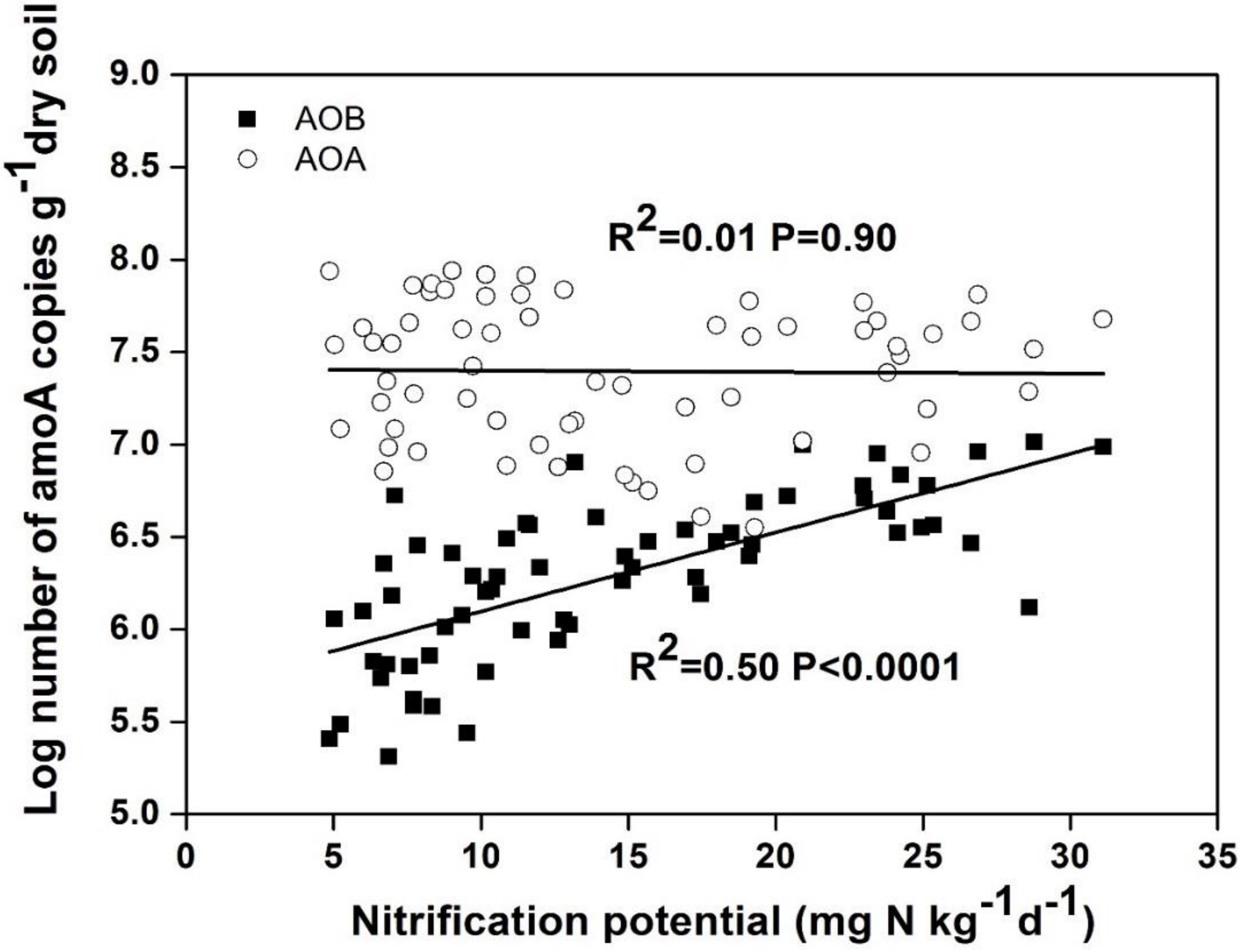
Abundance of ammonia oxidizers



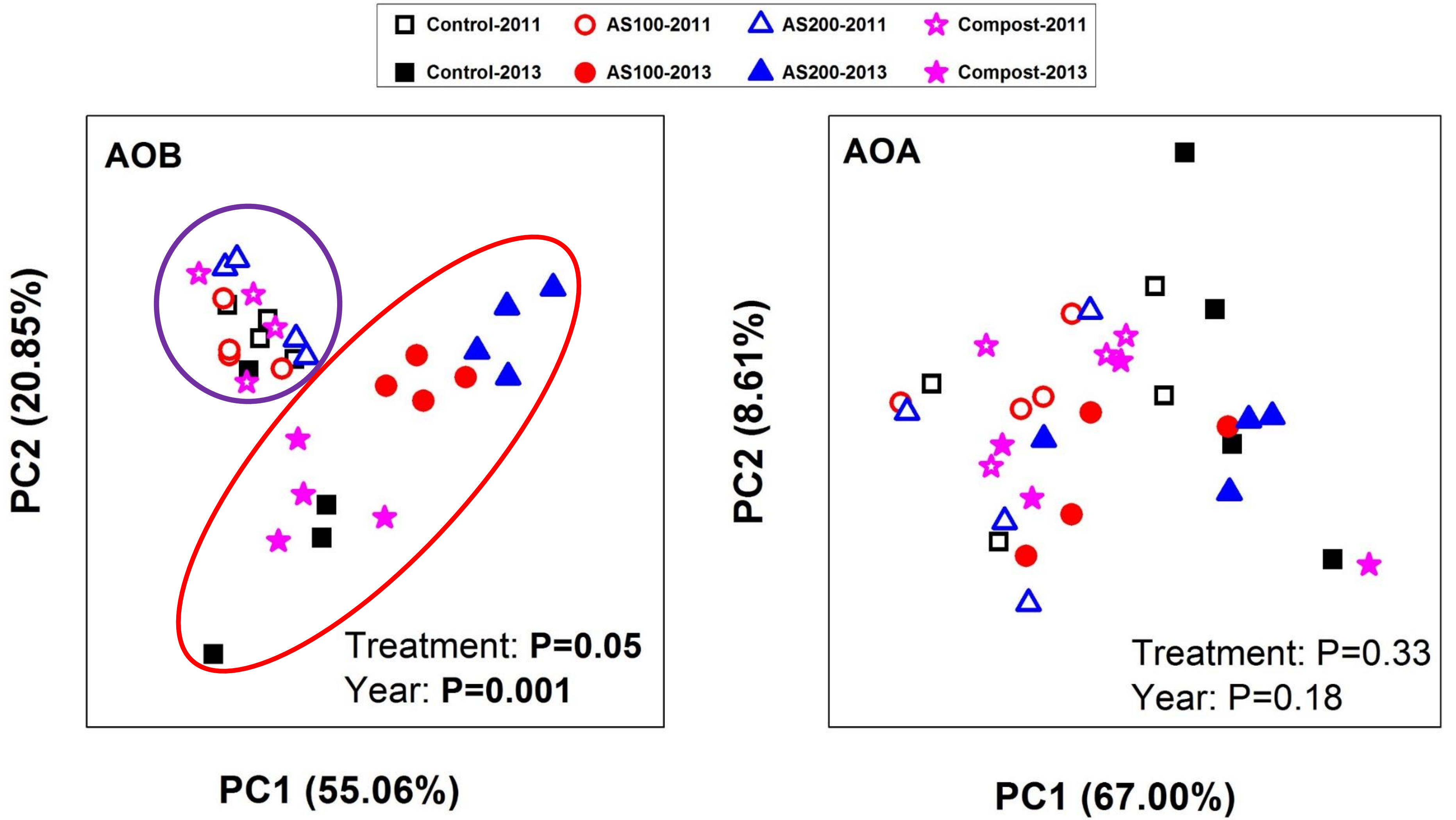
- AOA numbers higher than AOB but unchanged by treatment
- AOB numbers affected by treatment and year, significant after 2 fertilizations
- AOB increase with repeated treatments of AS fertilizers

Ouyang, Y., J.M. Norton, J.M. Stark, J.R. Reeve and M.Y. Habteselassie. 2016. Ammonia-oxidizing bacteria are more responsive than archaea to nitrogen source in an agricultural soil. Soil Biol. Biochem. 96: 4-15.doi:http://dx.doi.org/10.1016/j.soilbio.2016.01.012

Potential rates proportional to AOB abundance



Ammonia oxidizing bacteria community changes



Evaluate the relative contribution of AOA and AOB

Use a differential inhibitor: 1-Octyne

AOB are inhibited by octyne **AOA are resistant to octyne**

Fresh soil

NH_4^+

Nitrification potential

Octyne-resistant nitrification potential
AOA

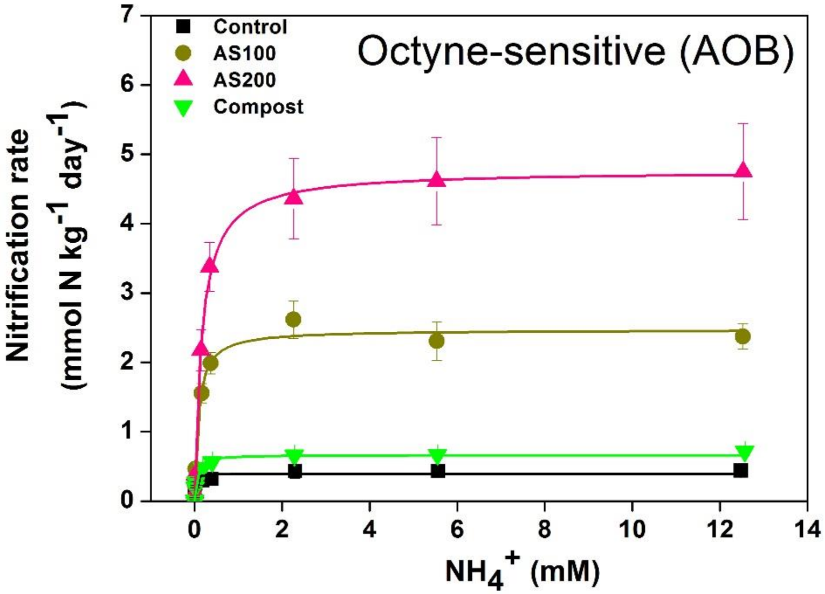
By difference

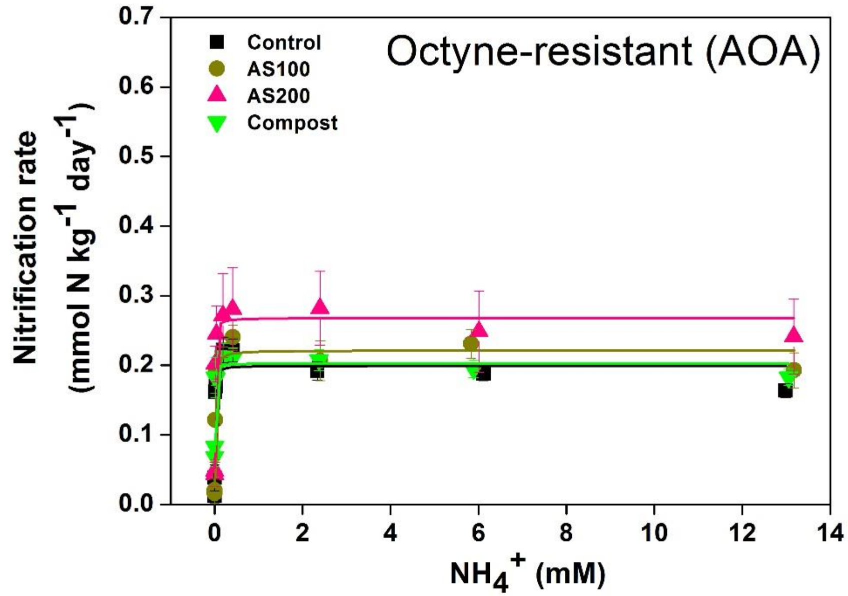
Octyne-sensitive nitrification potential
AOB

(method based on Taylor et al 2013)

AOB and AOA activity responds differently to ammonium substrate

Note scale 10X expanded

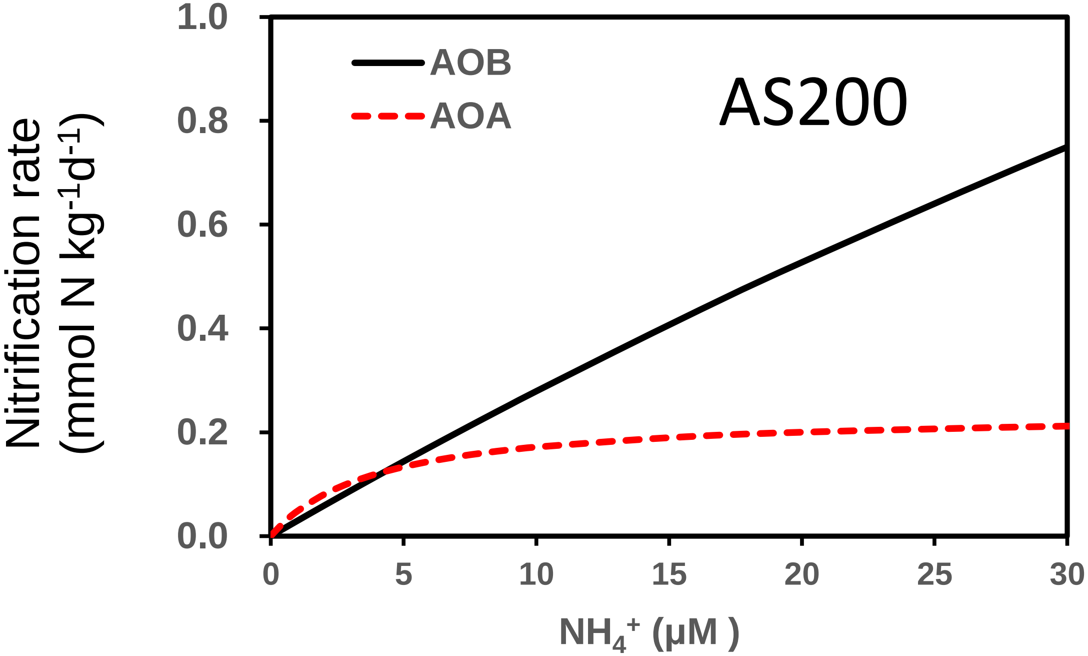




- AOB respond to additional NH_4^+
- AS fertilizer history affects response
- AOA capacity is saturated at low NH_4^+ concentration

Ouyang, Y., J.M. Norton and J.M. Stark. 2017. Ammonium availability and temperature control contributions of ammonia oxidizing bacteria and archaea to nitrification in an agricultural soil. Soil Biol. Biochem. 113: 161-172. doi:https://doi.org/10.1016/j.soilbio.2017.06.010.

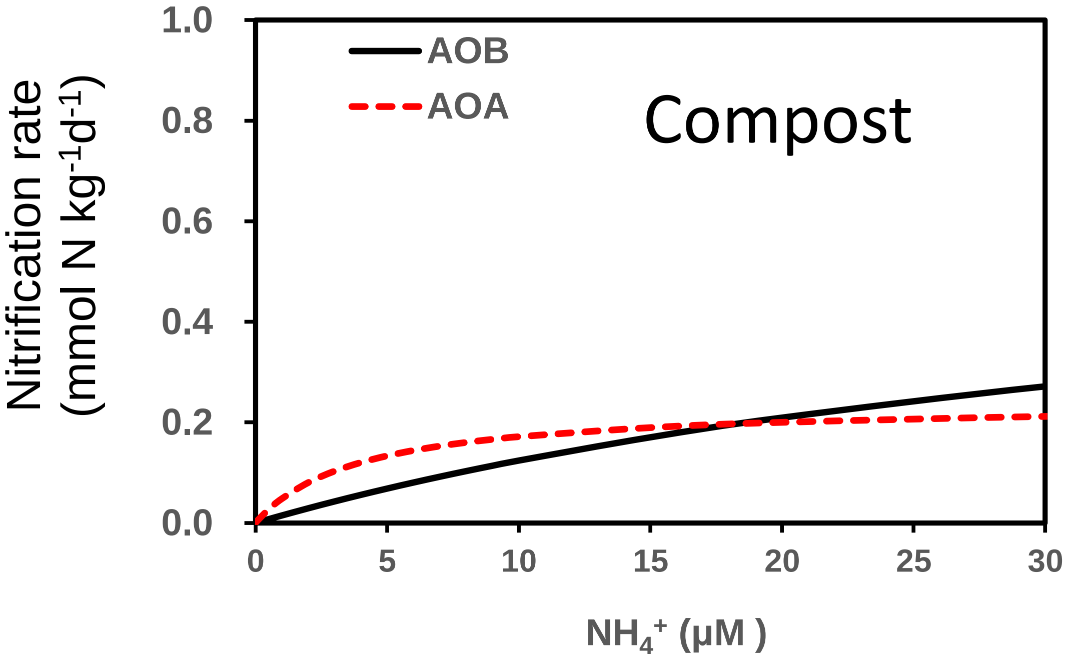
Nitrification kinetic models



$$V=V_{max}*S/(K_m+S)$$

AOB: V_{max}= 4.8 mmol N kg^{-1} d^{-1};
k_m=161 µM

AOA: V_{max}= 0.3 mmol N kg^{-1} d^{-1};
k_m=3.6 µM



AOB: V_{max}= 0.7 mmol N kg^{-1} d^{-1};
k_m=44 µM

AOA: V_{max}= 0.2 mmol N kg^{-1} d^{-1};
k_m=2.9 µM

Ouyang, Y., et al. 2017.
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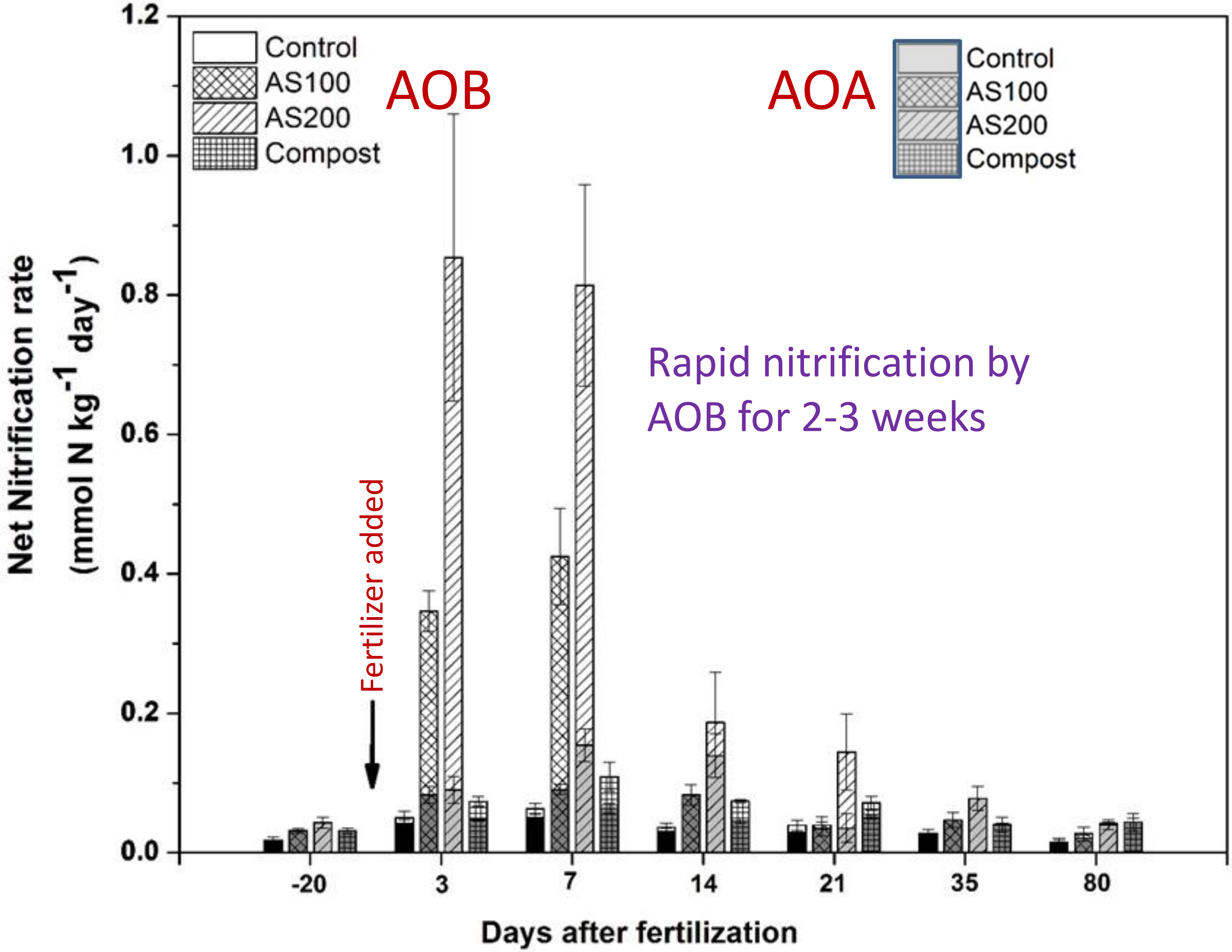
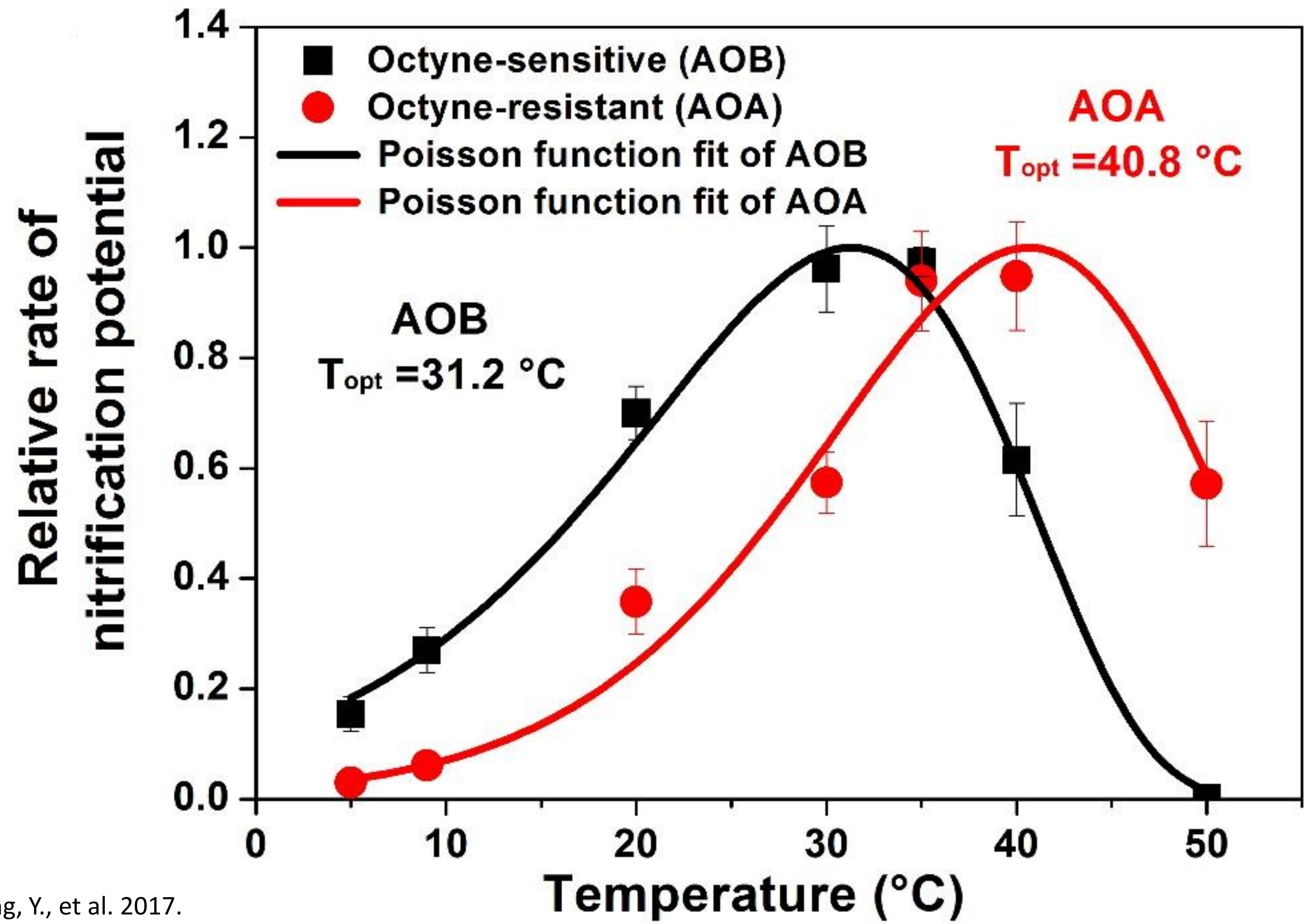

AOB
AOA
Control
AS100
AS200
Compost
Control
AS100
AS200
Compost
Rapid nitrification by AOB for 2-3 weeks
Fertilizer added
Net Nitrification rate (mmol N kg^{-1} day^{-1})
1.2
1.0
0.8
0.6
0.4
0.2
0.0
-20
3
7
14
21
35
80
Days after fertilization

AOB and AOA distinct temperature optimum
AOB favor cooler AOA favor warmer



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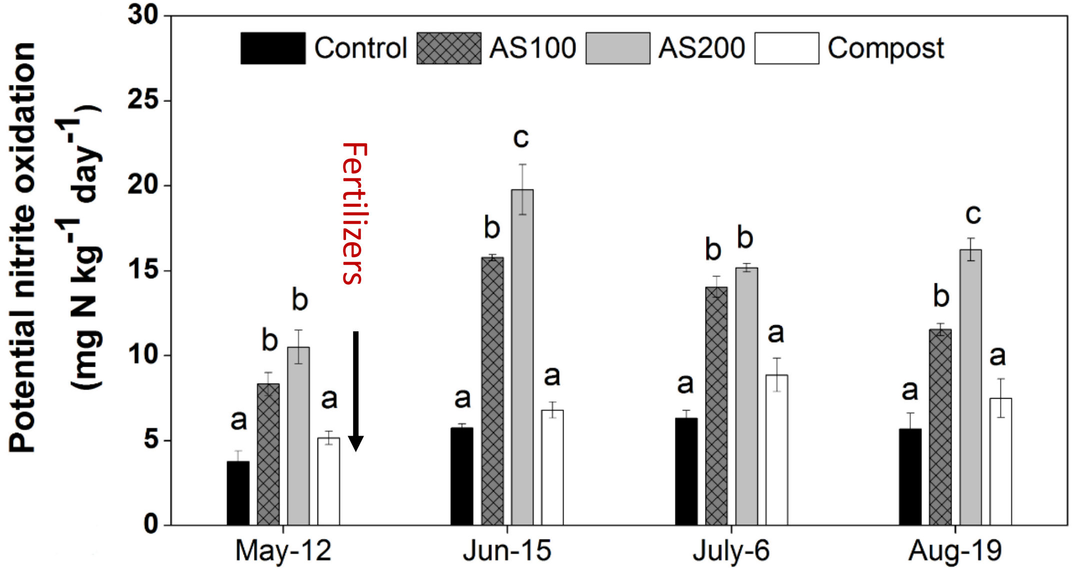
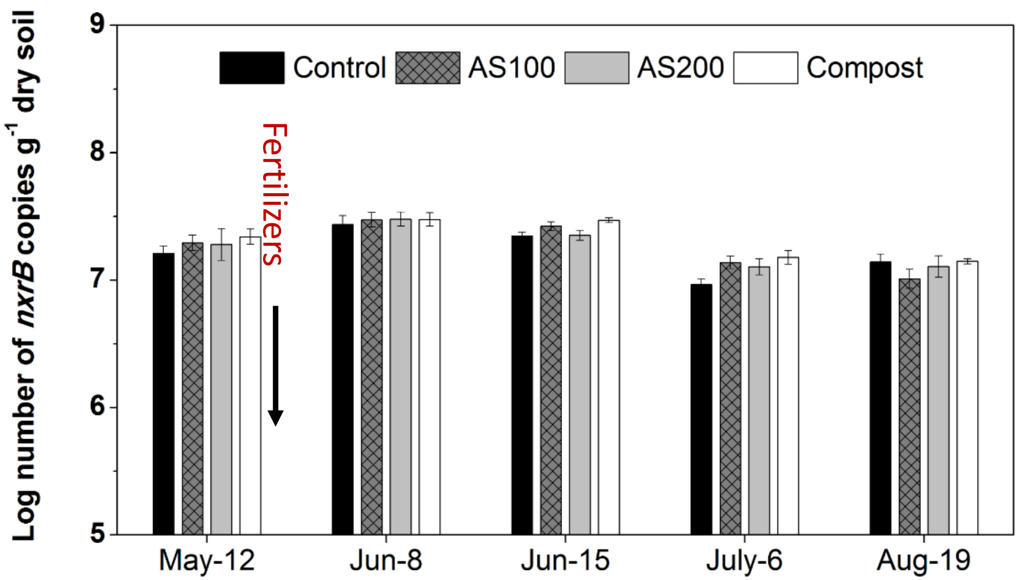
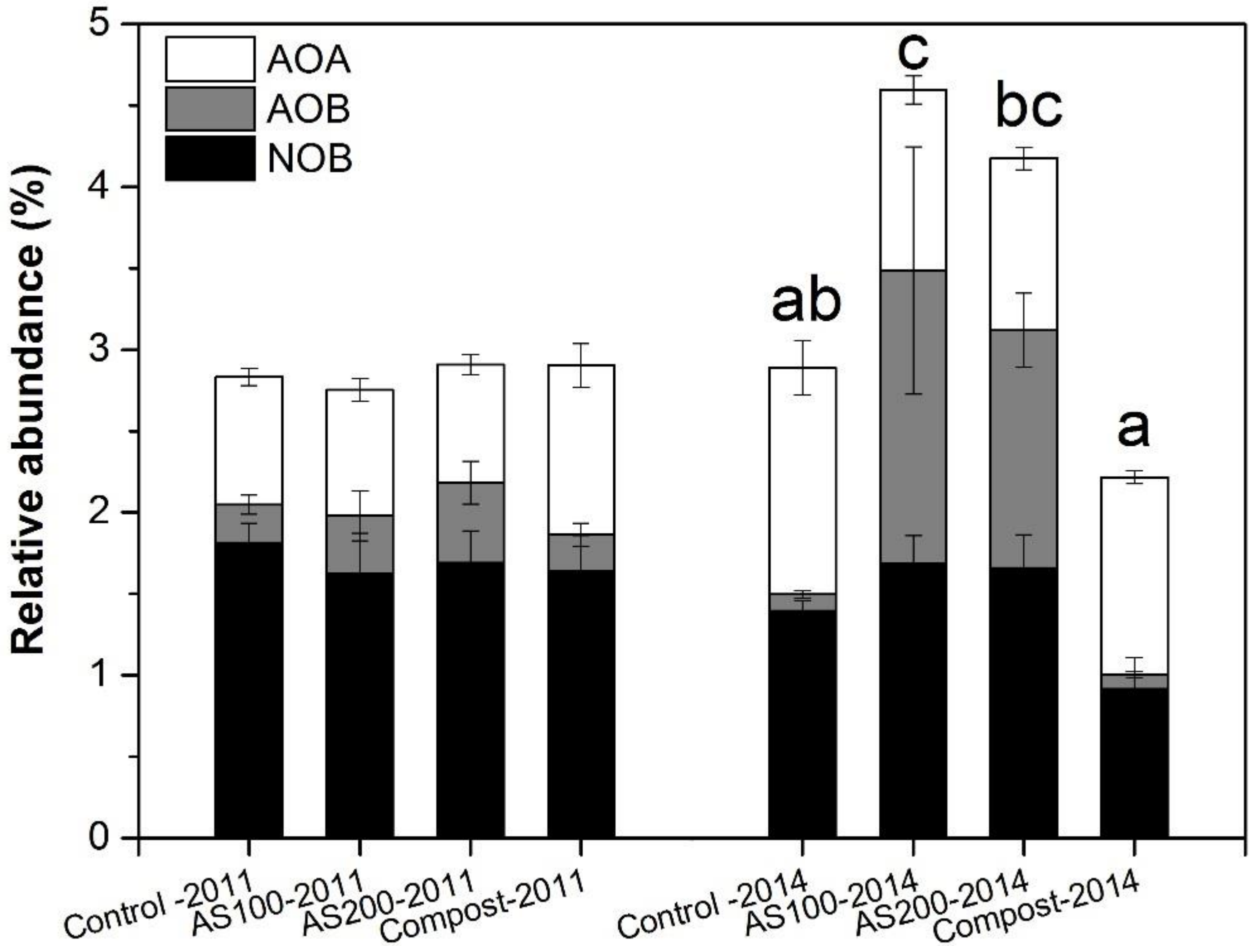
Nitrite oxidizers activity but not abundance changes with ammonium fertilizer



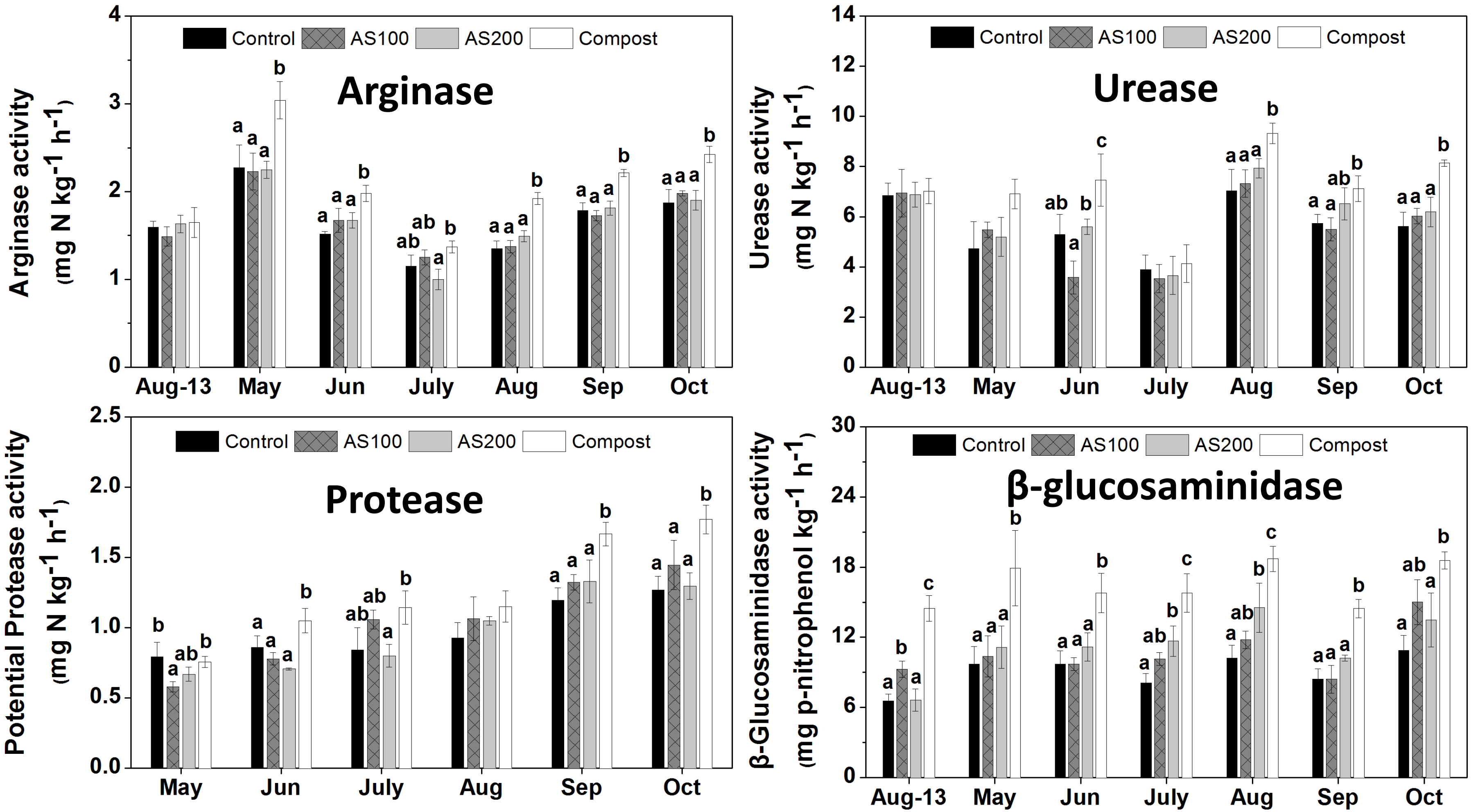


Changes in nitrifying community composition

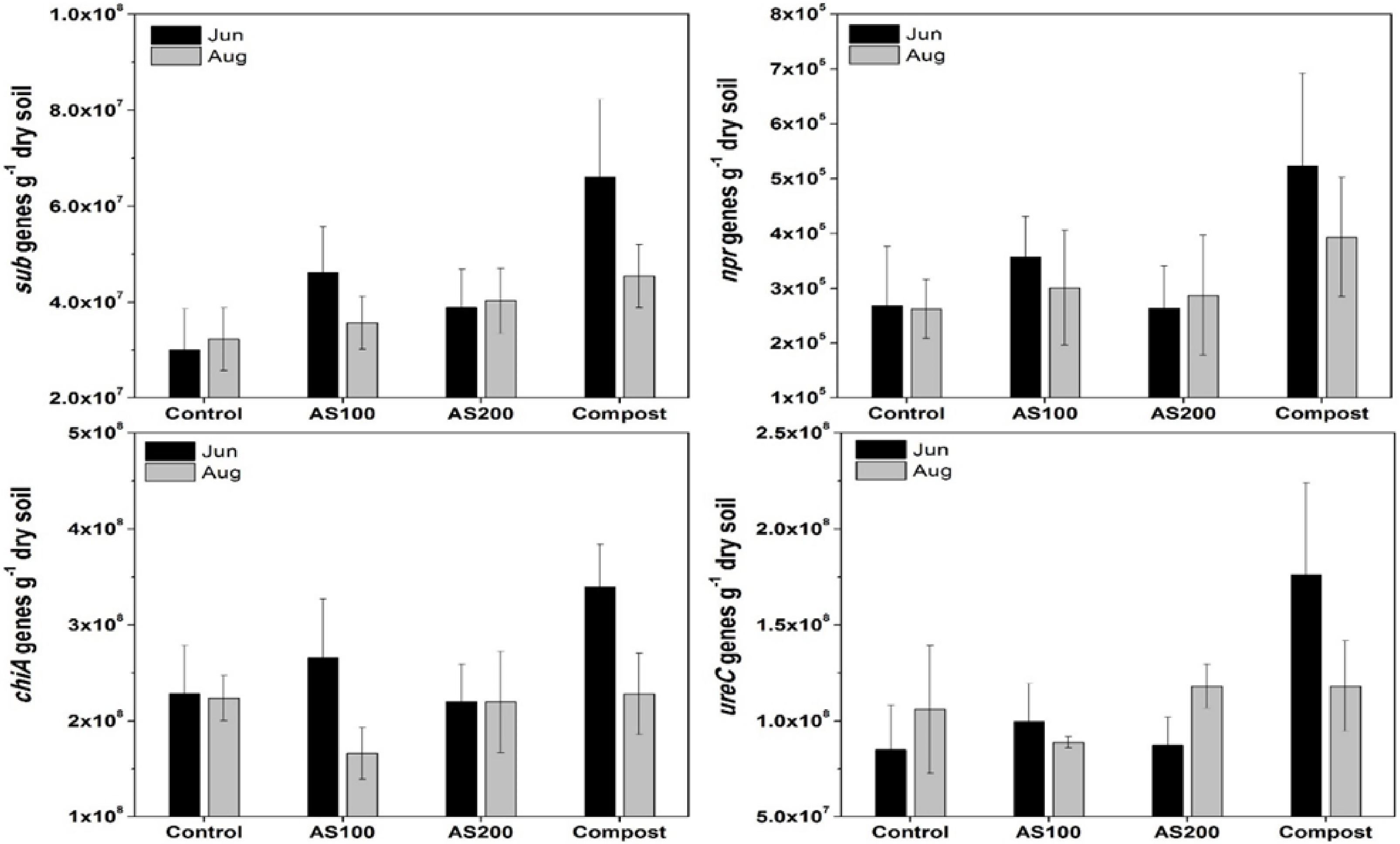


The nitrifying community shifted by four years of repeated application of ammonium fertilizers

N mineralization enzyme activities increase with compost



N mineralization gene targets not significant



Solution – sequence everything

Soil metagenomics

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Take home messages

- Ammonia-oxidizing bacteria were more responsive than archaea to ammonium fertilizers
- Ammonia-oxidizing bacterial abundance and community structure changed by ammonium fertilizers over multiple seasons
- Ammonium availability and temperature control the relative contribution of archaea and bacteria to nitrification
- Control the activity of ammonia oxidizing bacteria immediately after the application of mineral N fertilizer
  >> improve N use efficiency
- Understanding the response of nitrifiers to substrate and temperature allows the effect of microbial community to be included in models of nitrification

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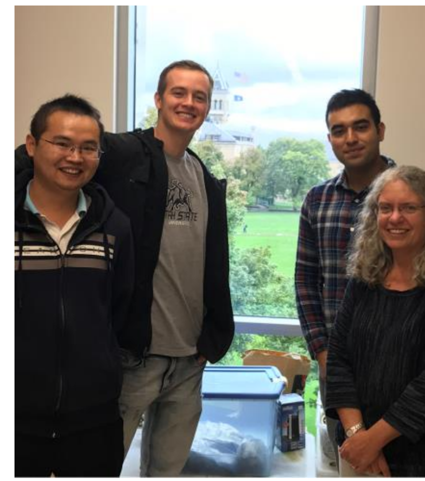

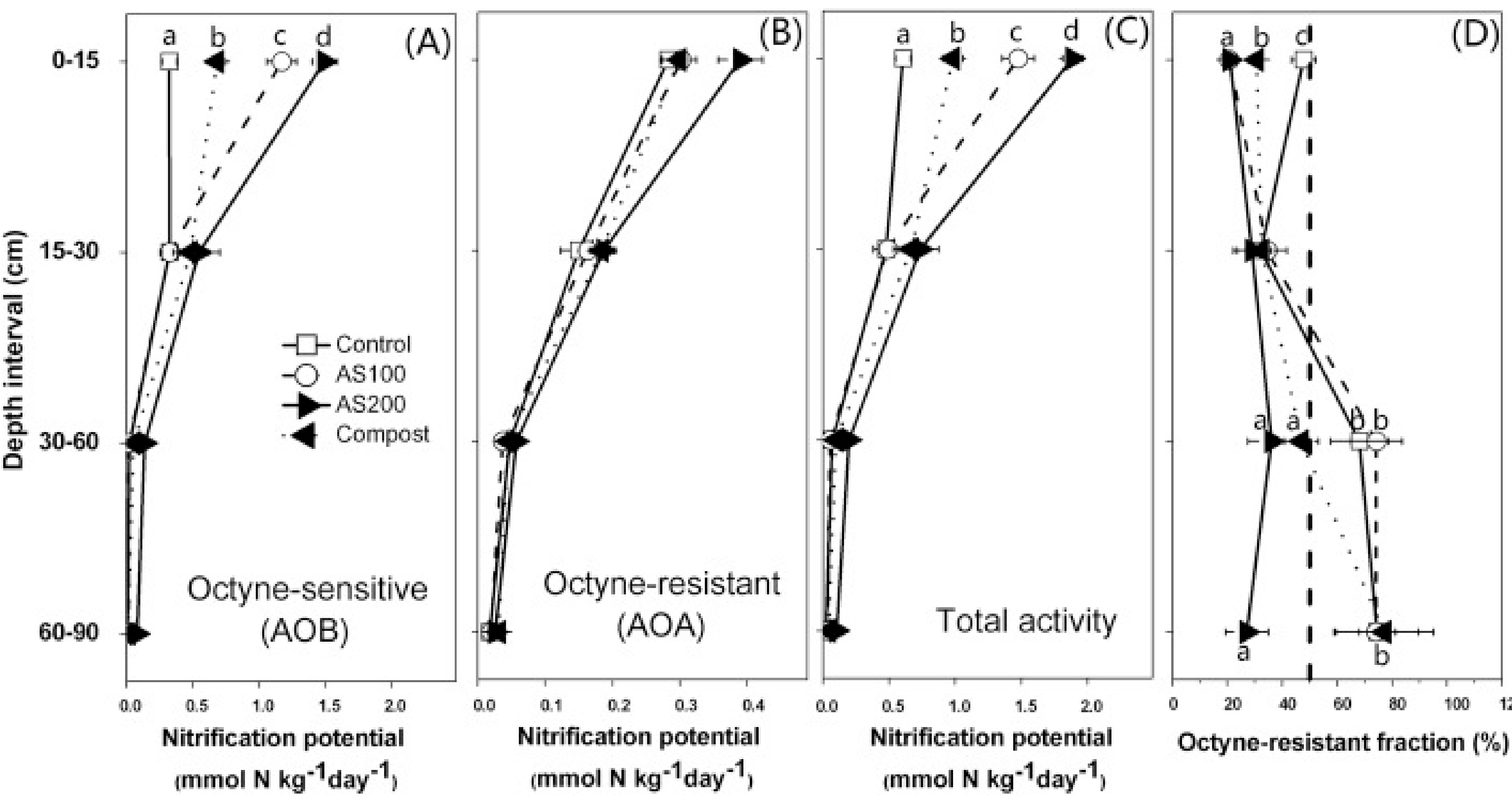

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