Soil-specific responses of methanotrophic communities in rice rhizosphere to genotype and nitrogen fertilization Sandy Jan Labarosa¹, Katharina Frindte¹, Michael Frei², Claudia Knief¹ ¹Institue of Crop Science and Resource Conservation (INRES), University of Bonn, Germany ²Department of Agronomy and Crop Physiology, University of Giessen, Germany

Methanotrophs

are microbes that metabolize methane (CH_4) , a potent greenhouse gas, as their carbon and energy source.

Why study them?

Methanotrophs play an essential role in the reduction of CH₄ emissions in rice paddies. Their occurence is influenced by diverse factors, which have often been studied one by one but rarely in combination.



Methods



Microcosm experiment 5x2x2 factorial in Randomized Complete Block Design with 5 replications. **Treatment levels** Genotype: Oryza sativa: IR64, Nipponbare, and Kasalath Oryza rufipogon (Rufi) Paddy soil: Philippine and Italian paddy soil Nitrogen fertilisation (N) rate: 0 kg ha^{-1} and 50 kg ha $^{-1}$

Bulk soil

Rhizosphere

Objective

Determine and compare the methanotrophic communities in the rice root and rhizosphere between four Oryza sp. genotypes grown in two different paddy soils and fertilized with or without additional nitrogen fertilizer by *pmoA*-gene targeted amplicon sequencing.

DNA extraction and pmoA gene-targeted amplification and sequencing

Development of feature classifier for pmoA gene, and data analysis

Main findings

Alpha diversity

Philippine paddy soil has more diverse methanotrophic taxa than Italian paddy soil. In Philippine paddy soil, bulk soil has significantly lower diversity compared to root and rhizosphere compartment.

Mixed-model ANOVA n = 171, *P* < .001***

Beta diversity

Philippine and Italian paddy soil are well separated indicating that they harbored different methanotrophic communities. Among the genotypes, Kasalath and Nipponbare had similar methanotrophic communities. The effect of nitrogen fertilisation is only significant in the root compartment of rice grown in the Italian paddy soil.

30 -	PERMANOVA (B ² values)	A	Paddy s	
	$Genotype = 2.2^{ns}$	_	0	Itali
	Cenotype = 2.2		•	Phi
	I N - rate = 0.6		-	

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Italian - Rhizosphere

Philippine - Rhizosphere



Note: Boxplots sharing the same letter are not significantly different at $\alpha = 0.05$

Take home messages

- > Different methanotrophic bacteria dominate the two paddy soils.
- > Community assembly, diversity, and recruitment of methanotrophs by different genotypes in the





roots and rhizosphere depend mainly on the native population present in the soil.

> Relatedness of rice genotypes (i.e. Kasalath to Nipponbare; where Nipponbare is phylogenetically closer to Rufi) does not necessarily reflect the preferential recruitment of dis-/similar methanotrophic communities.

> Effect of nitrogen fertilisation is very small and soil specific.

