



Archaeal Metabolic Profiles at Deep-Sea Hydrothermal Vents in the Mid-Cayman Rise



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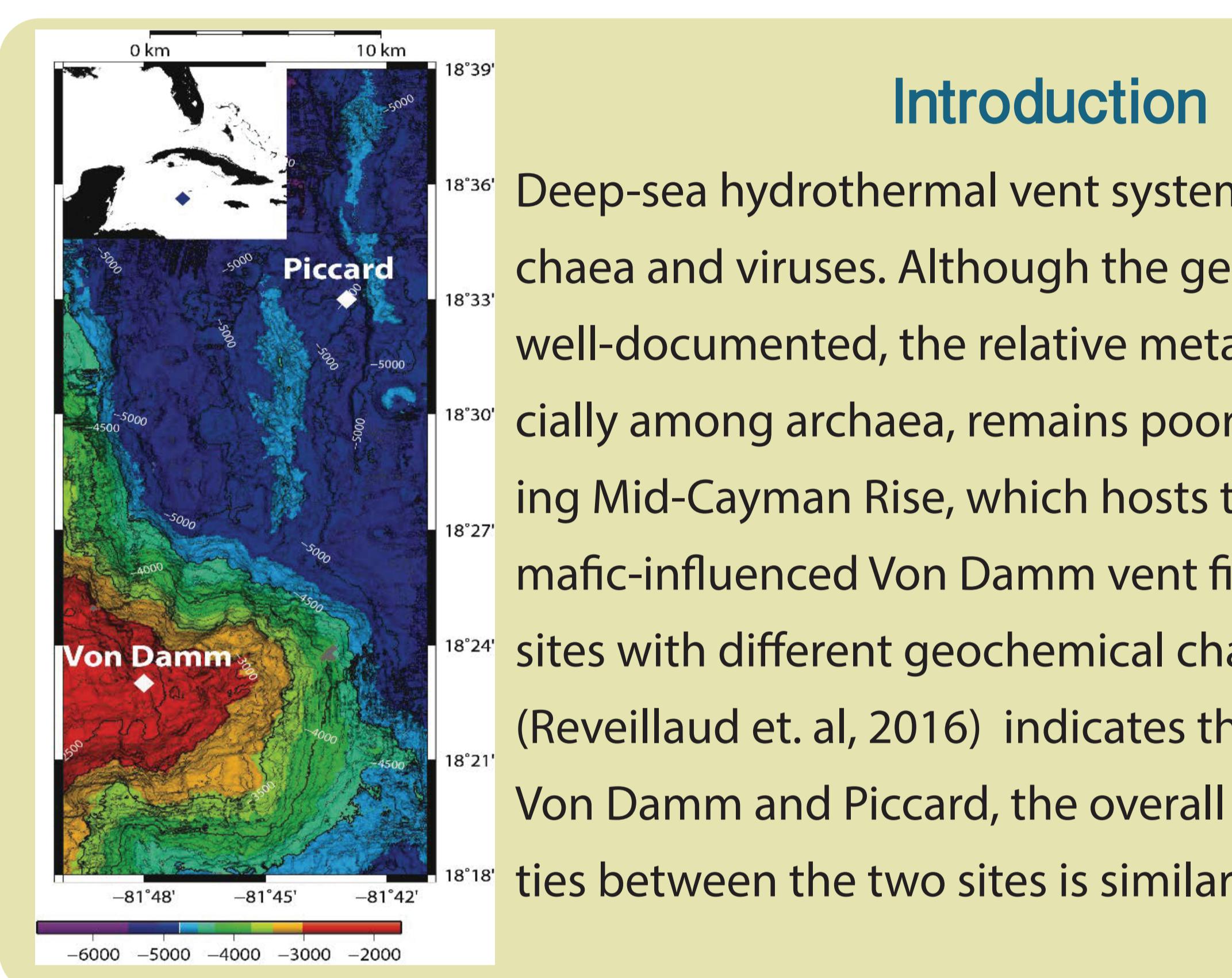
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Motivating Questions

- Which metabolic genes are most highly expressed at the community level?
- Which taxonomic groups (esp. archaea) have the highest rates of overall gene expression?
- What is the metabolic potential (esp. methanogenesis) of each taxonomic group?
- What nutrient cycling relationships define community-level methane metabolism?



Introduction

Deep-sea hydrothermal vent systems host a wide diversity of bacteria, archaea and viruses. Although the geochemical conditions at these vents are well-documented, the relative metabolic activity of microbial lineages, especially among archaea, remains poorly characterized. The deep, slow-spreading Mid-Cayman Rise, which hosts the mafic-influenced Piccard and ultra-mafic-influenced Von Damm vent fields, allows for the comparison of vent sites with different geochemical characteristics. Previous metagenomic work (Reveillaud et al., 2016) indicates that despite the distinct geochemistry at Von Damm and Piccard, the overall functional profile of microbial communities between the two sites is similar.

Methods

Sequencing of metagenomes and metatranscriptomes was conducted on an Illumina Hi Seq 1000. Reads were assembled with IDBA (Peng et al., 2010). Mapping was done with Bowtie2 (Langmead and Salzberg, 2012). Genes were called using Prodigal (Hyatt et al., 2010) and annotated using the IMG-JGI pipeline and the KO (Kanehisa and Goto, 2000) and Pfam (Finn et al., 2016) databases. MAG taxonomies were assigned using phlyosift (Darling et al., 2014). We used anvi'o v2.3.2 (Eren et al., 2015) for supervised binning of the metagenome-assembled genomes (MAGs) based on coverage and tetranucleotide frequency. We also used anvi'o v2.3.2 for the pangenomic analysis. Figures were generated using matplotlib (Hunter, 2007) and seaborn.

Community-Level Expression

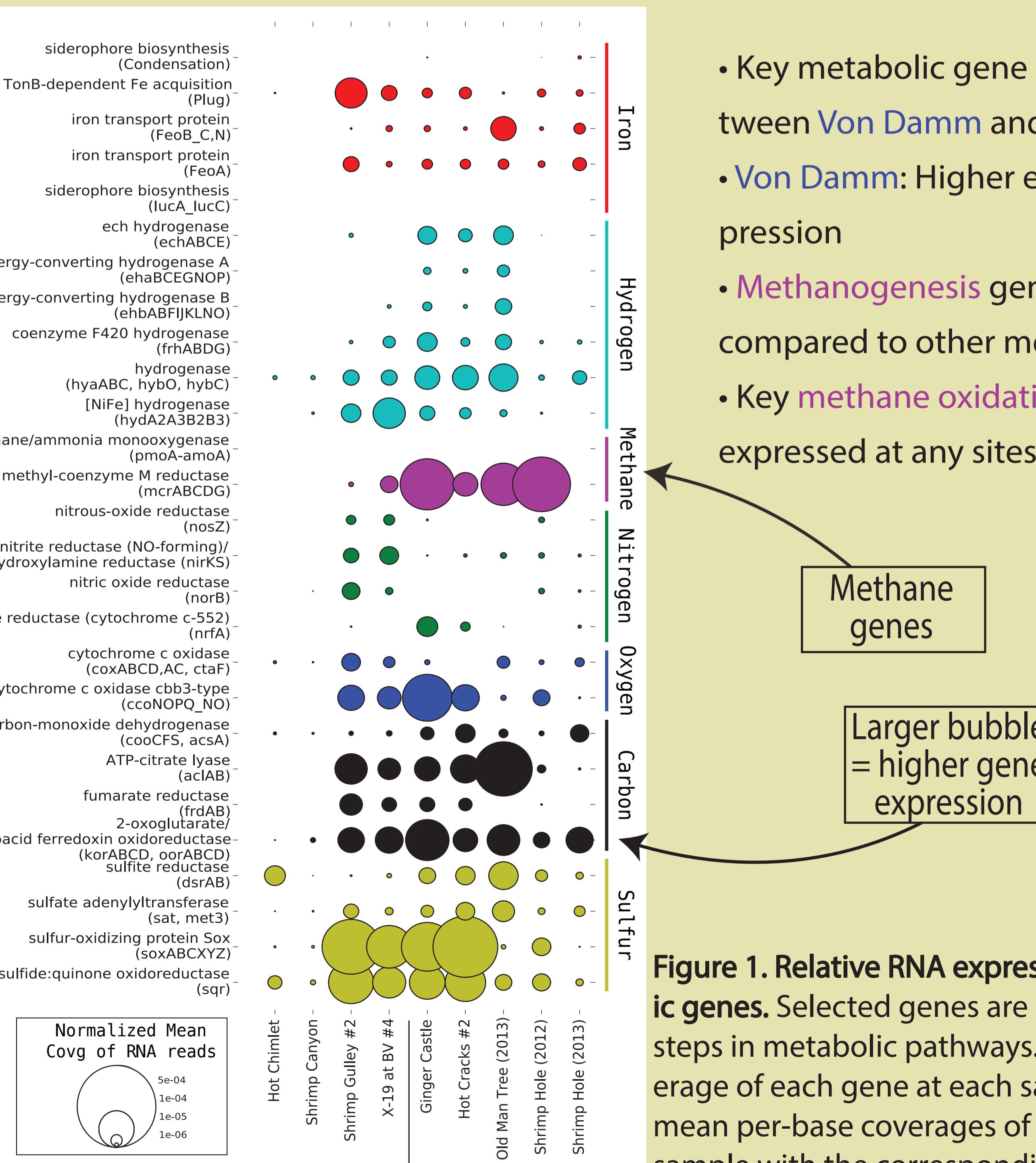


Fig. 1

- Key metabolic gene expression similar between Von Damm and Piccard
- Von Damm: Higher evenness of gene expression
- Methanogenesis gene (mcr) highly active compared to other metabolic groups
- Key methane oxidation gene (pmoA) not expressed at any sites

Figure 1. Relative RNA expression of select metabolic genes. Selected genes are representative of major steps in metabolic pathways. We calculated the coverage of each gene at each sample by summing the mean per-base coverages of each contig in the sample with the corresponding annotation. Coverage values were normalized by dividing by the total number of RNA reads present in the sample.

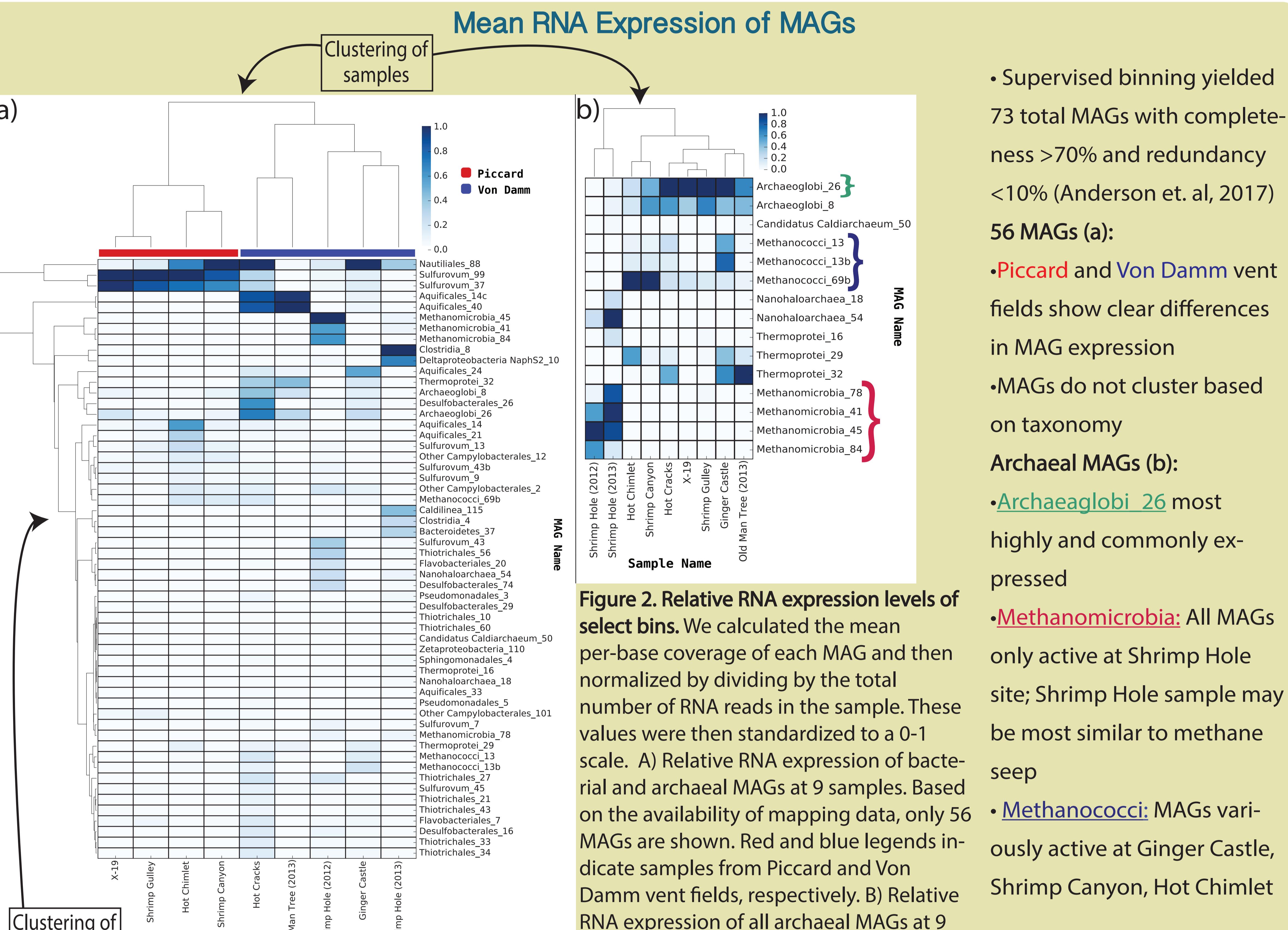


Fig. 2

Figure 2. Relative RNA expression levels of select bins. We calculated the mean per-base coverage of each MAG and then normalized by dividing by the total number of RNA reads in the sample. These values were then standardized to a 0-1 scale. A) Relative RNA expression of bacterial and archaeal MAGs at 9 samples. Based on the availability of mapping data, only 56 MAGs are shown. Red and blue legends indicate samples from Piccard and Von Damm vent fields, respectively. B) Relative RNA expression of all archaeal MAGs at 9 samples. MAGs highlighted in the text are indicated with a bracket of the corresponding color.

Pangenomics and Metabolic Potential

Group I: Core to methane metabolism. MAGs contain majority of genes for 1+ methanogenesis pathways

- Methanomicrobia:** FULL acetate \Rightarrow methane, partial CO₂ \Rightarrow methane. mcr present.
- Methanococci:** partial CO₂ \Rightarrow methane, partial acetate \Rightarrow methane. mcr present.
- Archaeoglobi:** partial CO₂ \Rightarrow methane, partial acetate \Rightarrow methane. mcr not present.

Group II: Accessory to methane metabolism. MAGs contain 1-3 genes required for 1+ pathways

- Clostridia (Bacteria):** partial trimethylamine \Rightarrow methane (only MAG contributing to this pathway)

Group III: MAGs contain genes for first steps of acetate \Rightarrow methane pathway (acs, ack, or pta)

Group IV: MAGs do not contain genes sufficient for a step in any methanogenesis pathway

- Possible contamination in Aquificales_24, Desulfobacterales_74

Figure 3. Presence-absence table of key methanogenesis protein clusters in 43 MAGs. Genes were selected to represent all steps of the CO₂ \Rightarrow methane, acetate \Rightarrow methane, and trimethylamine \Rightarrow methane pathways. Some protein clusters have annotations to 1 functionally similar genes. MAGs included in the table 1. had >1 methanogenesis gene or 2. shared a taxonomic classification with a MAG that had >1 methanogenesis gene. Taxonomic groups highlighted in the text are indicated with brackets of the corresponding color.

