

Archaea are species- and habitat-specific plant colonizers unravelled by metagenomics

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Introduction & overall objective

Archaea are primarily known as extremophiles, found in hostile environments like hot springs, black smokers or salines. Due to advanced OMICS-technologies, recent studies further revealed their abundance in the ocean, in the soil and even on humans. But their ecological roles and interactions with their hosts remained mostly unclear. Plant-Archaea associations are poorly understood, however in our recent studies we found high abundances of endophytic Archaea (up to 67.3% of total reads) in olive trees (*Olea europaea* L.), indicating interactive mechanisms of Archaea with their hosts. Our principle objective was to unravel structure and function of plant-associated Archaea along a broad host-spectrum. Therefore we compared 37 different plants of the families *Pinaceae*, *Ericaceae*, *Cyperaceae*, *Poaceae*, *Hylocomiaceae*, *Polytrichaceae*, *Aulacomniaceae*, *Sphagnaceae*, *Typhaceae*, *Brassicaceae*, and *Amaranthaceae* in a combined approach.

Plant-specific colonization

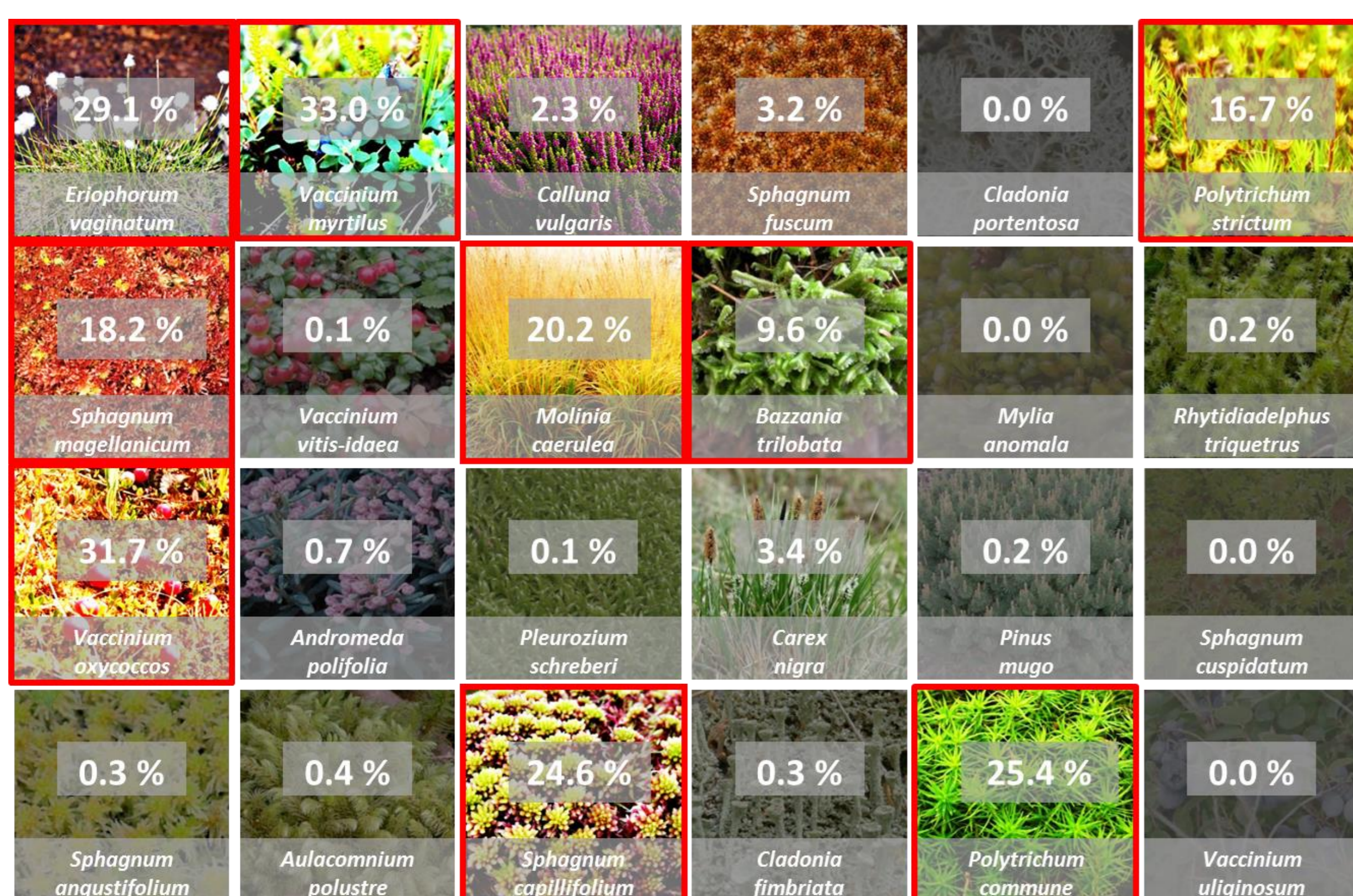


Fig.1: Relative archaeal abundances of alpine bog vegetation, analyzed by 16S rRNA amplicon sequencing; brightness of the picture correlates with the relative abundance of Archaea

- 16S rRNA gene sequencing analysis
- High archaeal proportion up to 33%
- Highest abundances in vegetation forming lignified parts, including Eudicots (*Vaccinium*) and Monocots (*Eriophorum*)

Habitat-specific colonization

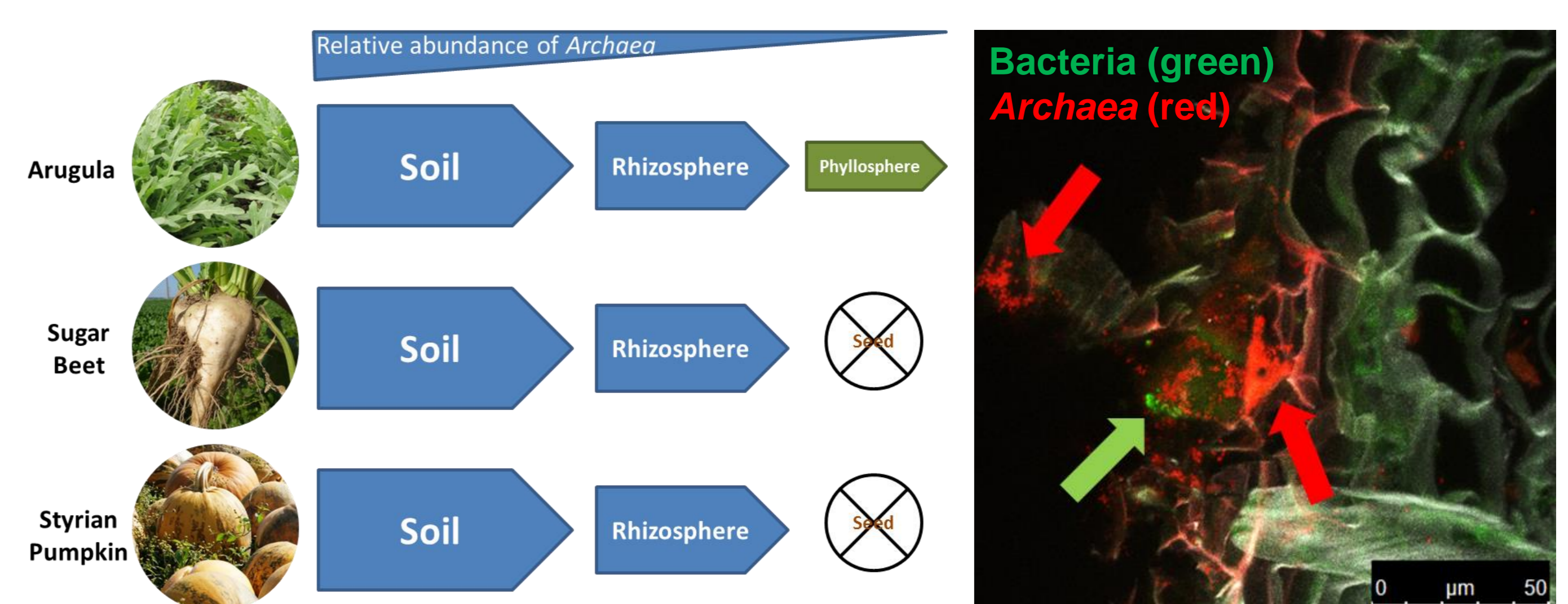


Fig.2: Schematic illustration of archaeal abundances of the habitats soil, rhizosphere, phyllosphere and seed of arugula (*Eruca sativa*), sugar beet (*Beta vulgaris*) and styrian pumpkin (*Cucurbita pepo* var. *styriaca*); analyzed by metagenome, 16S amplicon and 16S amplicon analysis, respectively; size of the arrows indicate the relative abundance of Archaea

Fig.3: Fluorescence in situ hybridization (FISH) and confocal laser scanning microscopy (CLSM) images of rotten root of arugula

- Highest archaeal abundance generally found in soil
- No Archaea were found in seeds
- Soil potentially acts as primary source of Archaea for the colonization of the plant
- Indications for Archaea accumulating on nutrient rich plant compartments (Fig.3.)

Archaea-plant functional model

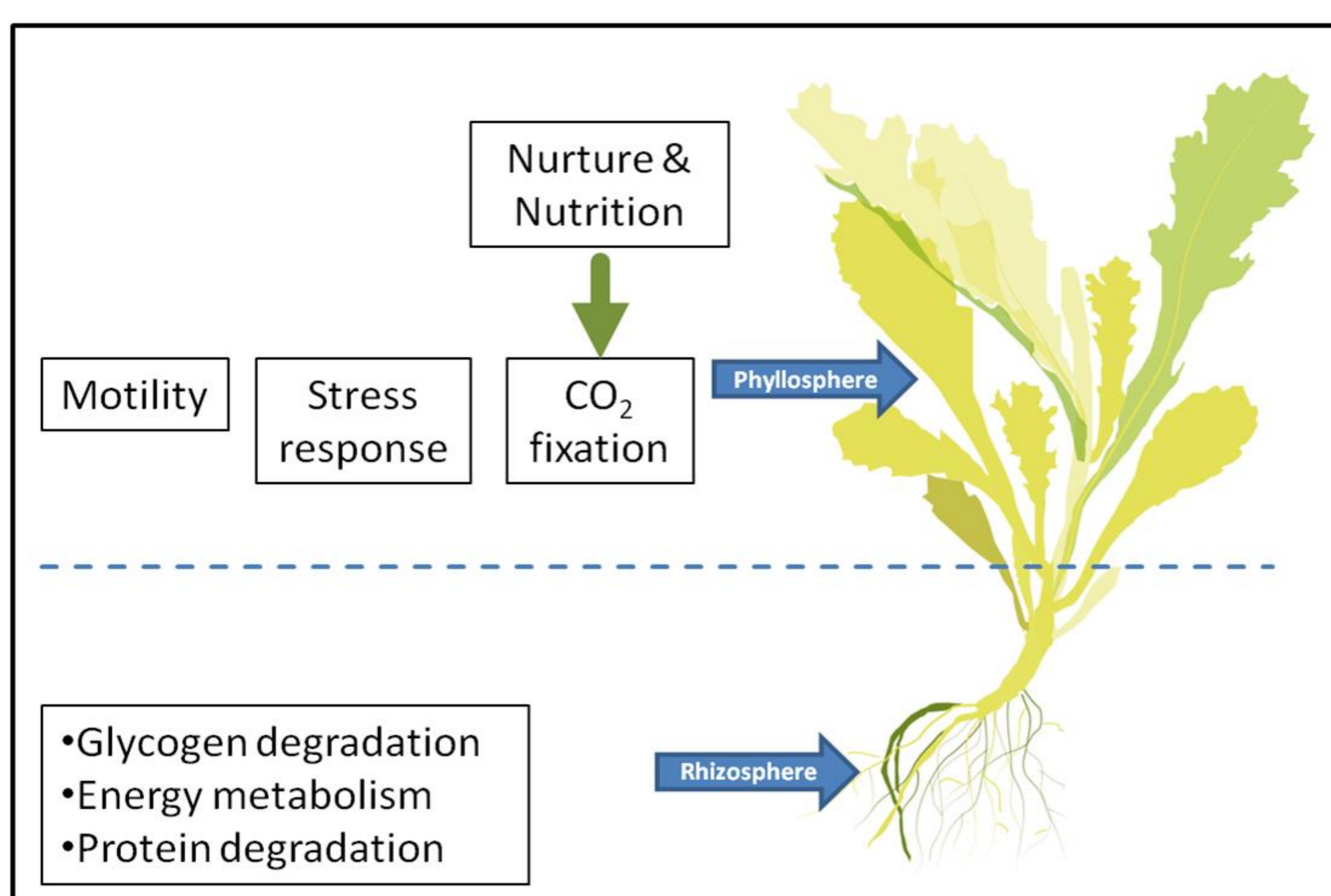
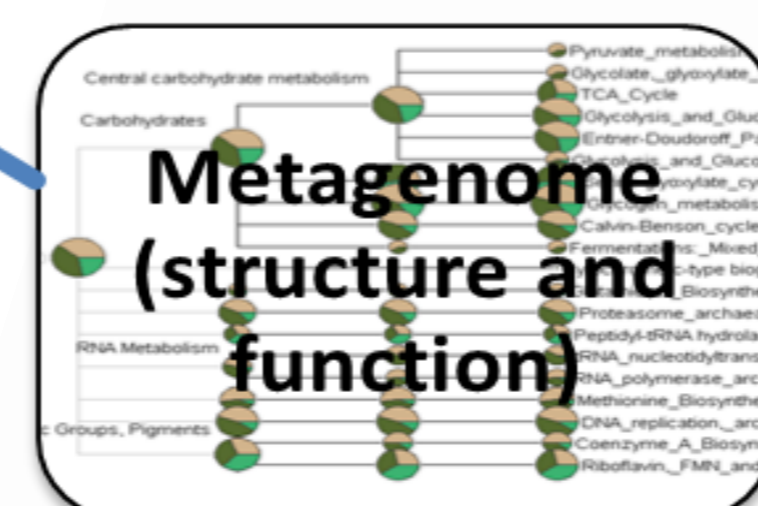
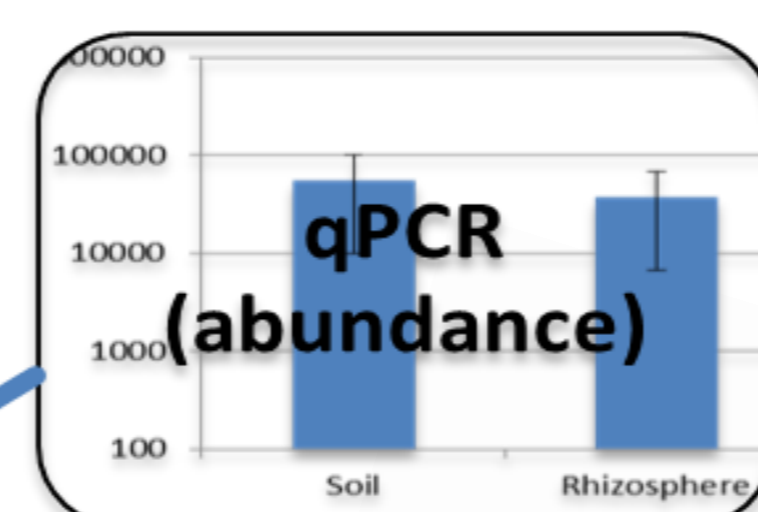


Fig.4: Plant-specific functions of Archaea on arugula comprising rhizosphere and phyllosphere; obtained from metagenome analysis (MG-Rast)

Main archaeal plant-specific functions related to:

- Phyllosphere - Motility, stress response and C-fixation
- Rhizosphere - Glycogen degradation, energy metabolism and protein degradation

Methods



Amplicon sequencing (structure and networks)

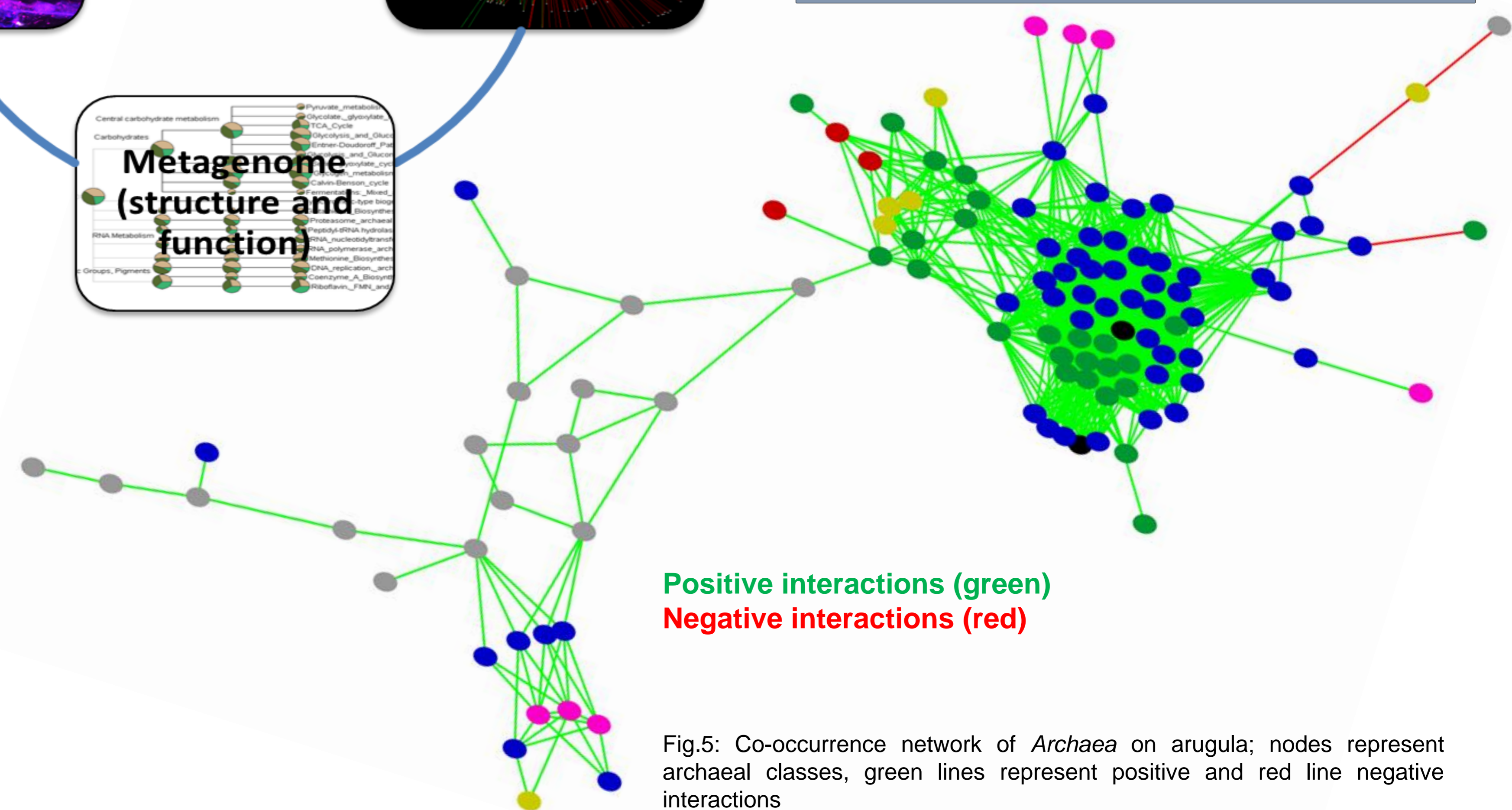


Fig.5: Co-occurrence network of Archaea on arugula; nodes represent archaeal classes, green lines represent positive and red line negative interactions

- Stringent archaeal network
- Joint occurrence prevails over random distribution
- Positive interactions were predominant

Conclusions

- Soil is the preferred archaeal habitat and acts as a source of colonization
- Indications that Archaea have plant-specific and habitat-specific colonization patterns
- Archaea show functional specification on plants (Stress response; Motility; N and C cycling)