









This project has received funding from the European Union's Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement No 675546.

### Mosses matter

- Carbon sink
- Major source of N N<sub>2</sub>-fixing bacteria
- Higher temperatures  $\rightarrow$  increase in N<sub>2</sub> fixation
- Shading by higher plants  $\rightarrow$  decrease in N<sub>2</sub> fixation
- Moisture
  - Drier conditions  $\rightarrow$  decrease in N<sub>2</sub> fixation
- Recent meta-analysis: experimental warming has no effect (Salazar et al 2019 Ecology in press)
  - Is it due to a shift in the bacterial community?

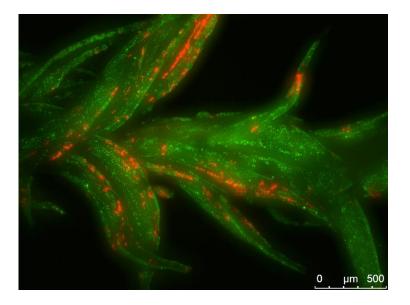


## The bryosphere – the bacterial component

"the combined complex of living and dead moss tissue and associated organisms"

Lindo and Gonzalez 2010 Ecosystems

- Cyanobacteria
- <u>Alphaproteobacteria</u>
- Betaproteobacteria
- Gammaproteobacteria
- Verrucomicrobia
- Plancomycetes
- Bacteroidetes
- Acidobacteria
- Actinobacteria



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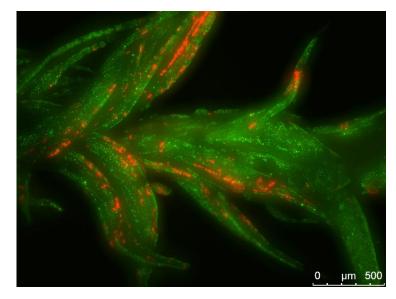


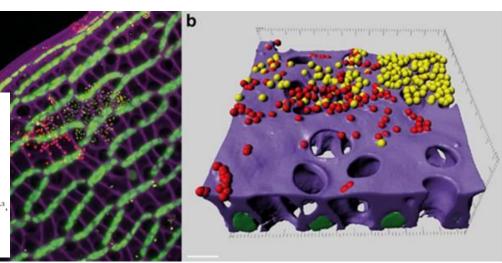
#### **ORIGINAL ARTICLE**

**Sphagnum** mosses harbour highly specific bacterial diversity during their whole lifecycle

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## What is the effect of long-term warming on the bacterial community associated with *Racomitrium lanuginosum*?

"Warming leads to a shift in  $N_2$ -fixing bacterial taxa with consequences for  $N_2$ -fixation rates"

- N<sub>2</sub> fixation rates and *nifH* gene copy numbers
- Community structure
- Changes in relative abundance

• Taxonomic composition

of potential N<sub>2</sub>-fixing taxa



## Experimental design



### Auðkúluheiði, Iceland

- Species-rich dwarf shrub heath
- 10 OTCs
- 10 control plots
- since 1997/1998 (20 years)





Racomitrium lanuginosum

## Sample processing

### N<sub>2</sub>-fixation rates

Acetylene reduction assays

### N<sub>2</sub>-fixation potential

qPCR of *nifH* genes

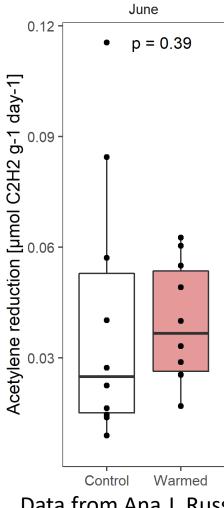
# Taxonomy and relative abundance of potentially N<sub>2</sub>-fixing bacteria

16S rRNA amplicon sequencing



## No difference in N<sub>2</sub>-fixation rates





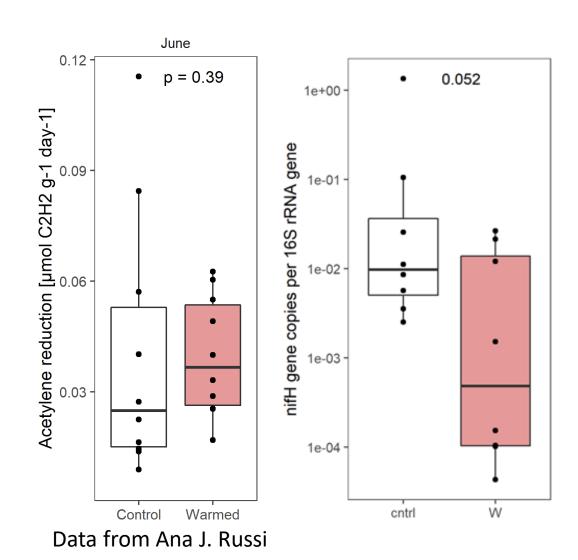
Data from Ana J. Russi

Introduction Hypothesis Methods Results Conclusio

## No difference in $N_2$ -fixation rates, but $N_2$ -fixation potential tends to be lower under warming

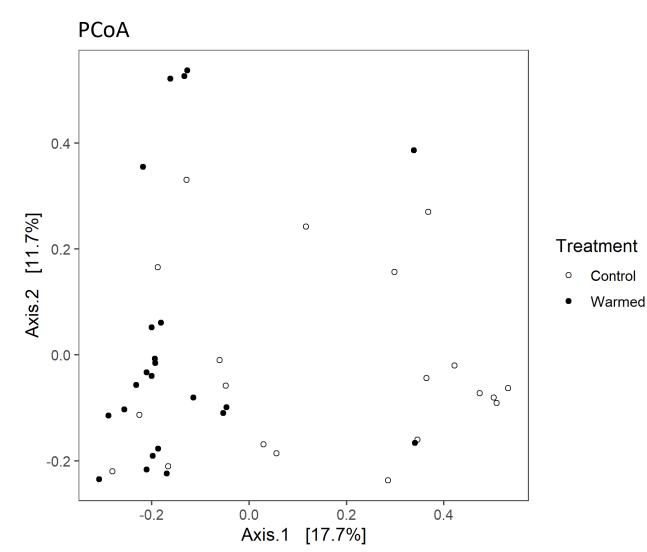
- Proportion of N<sub>2</sub>-fixing taxa (*nifH* copy per 16S rRNA gene copy) tends to be lower under warming
- Potential for N<sub>2</sub>-fixation as part of the total bacterial community is lower under warming





## Warming slightly affects N<sub>2</sub>-fixing community composition

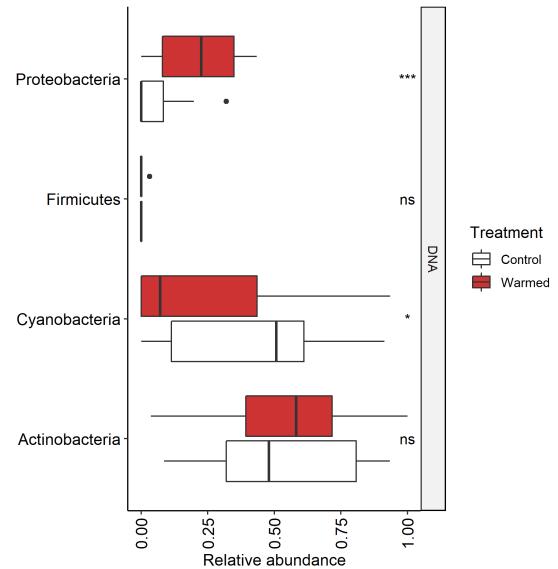
Potential  $N_2$ -fixers Permanova ~ Treatment P = 0.001 and  $R^2$  = 0.08



## Changes in relative abundance of taxa on phylum level

### Potential N<sub>2</sub>-fixers

- Cyanobacteria ↓
- Proteobacteria 个
  - Probably not adding significant amounts



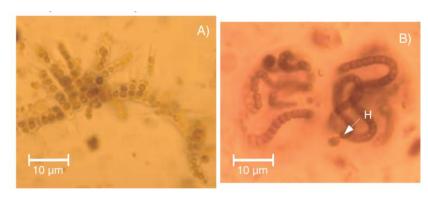
## Cyanobacterial indicator taxa

• Cyanobacterial genera:

Stigonema warming

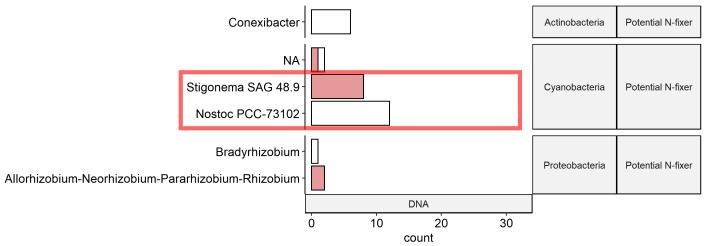
Nostoc control

- Stigonema most transcriptionally efficient N<sub>2</sub>-fixer in boreal forest feathermosses (Warshan et al. 2016)
- Might explain why nifH gene abundances decrease while N<sub>2</sub>-fixation rates stay similar



Houle et al (2006) Identification of two genera of N2-fixing cyanobacteria growing on three feater moss species in boreal forests of Quebec, Canada

#### Potential N<sub>2</sub>-fixing indicator taxa



### Conclusion

N<sub>2</sub> fixation rates did not change, due to shifts in the bacterial community

Other work: lichen bacterial community under warming, moss bacterial community during primary succession



## Thanks for your attention!











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