

Functional ecology of the methanotroph community

An adaptive response to afforestation
of pasture in New Zealand

by Loïc Nazaries

Contents

- | Background
- | Aims
- | Materials and methods
- | Results
- | Conclusions
- | Future works
- | Acknowledgements

Background

- | Methane (CH₄)
 - Potent green-house gas
 - Removed from atmosphere by methanotrophs (type I and type II)
 - Increased oxidation in temperate forests
- | Afforestation improves C sequestration in soils
 - Photosynthesis activity from trees
 - CH₄ sink from methanotroph activity
 - Slow process

Effect of afforestation in New Zealand

- | Stronger CH₄ sinks in NZ
- | Afforestation of pastures with pine trees ^{1, 2, 3}
- | Increased CH₄ sink è C sequestration in soils
 - Combination of changes in abiotic (soil properties) and biotic (methanotrophs) factors
 - Methanotroph community structure
 - type I vs. type II

¹ Singh *et al.*, 2007. *Appl. Environ. Microbiol.* 73, 5153-5161;
Biochemistry 41, 2196-2205;

² Singh *et al.*, 2009. *Soil Biology &*

Biochemistry 39, 1437-1449.
³ Tate *et al.*, 2007. *Soil Biology & Biochemistry* 39, 1437-1449.

Aims of this study

| Meta-analysis

- Compare sites with similar vegetation and response to afforestation

| Impact of long-term afforestation on:

- Methane fluxes
- Diversity and structure of methanotroph community

| Observe evolution of changes in structure of methanotroph population

- Pastures vs. afforested pastures vs. long-established forests

Materials and methods



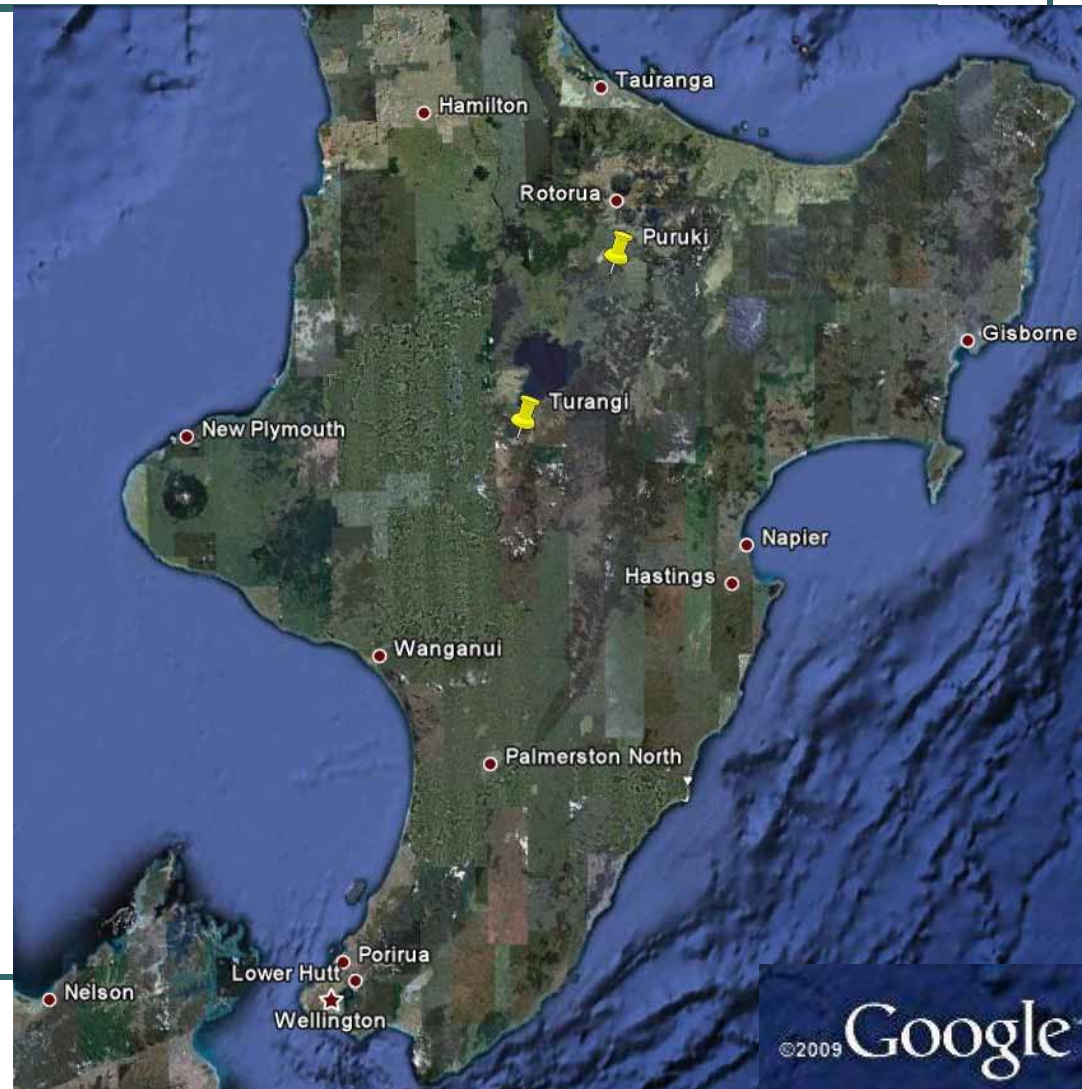
Description of sites

Turangi

- Tongariro National Park
- Manuka-Kanuka tree forest
- Stands aged 35 and 55 years

Puruki

- Near Rotorua
- Indigenous forest (mixed vegetation)



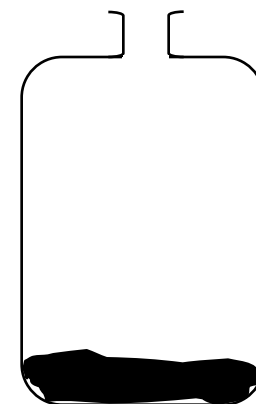
Methane flux measurements

- | Intact cores (10x10 cm) in a closed PVC chamber
- | Measurement of variations in CH₄ concentration in the headspace over time, using gas chromatography (GC)
- | **Indication on whether a soil is a source or a sink of methane**

Microcosm experiment (PLFA-SIP)

- | Injection of 50 ppm of isotopic methane ($^{13}\text{CH}_4$)
- | Incubation in dark for 14 days
- | Extraction of phospholipid fatty acids (PLFAs)
- | **Identification of active methanotroph population**

50 ppm
 $^{13}\text{CH}_4$



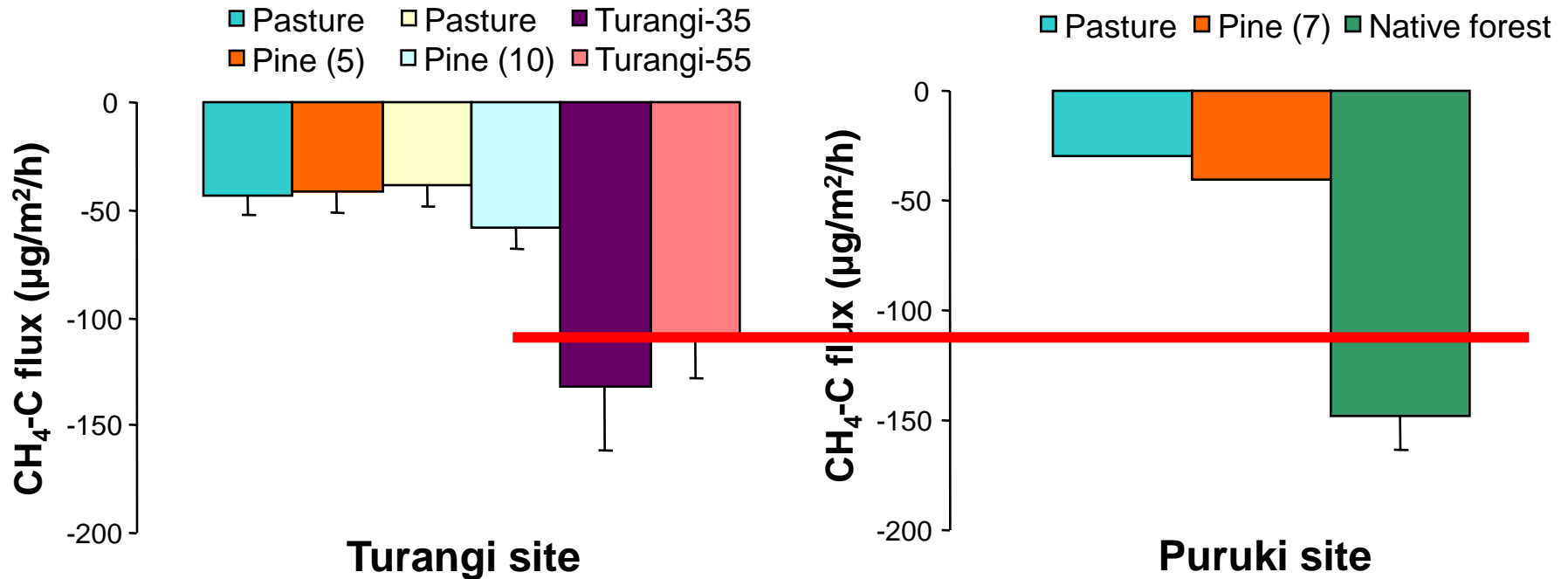
Molecular analysis

- | Terminal-Restriction Fragment Length Polymorphism (T-RFLP)
- | Analysis of three genetic markers:
 - 16S rRNA gene of type I and type II methanotrophs
 - *pmoA* gene
- | Production of unique fragmentation patterns (or T-RFLP profiles)
 - Observation of specific fragments called terminal-restriction fragments (T-RFs)
- | **Information on the structure and diversity of the methanotroph community**

Results

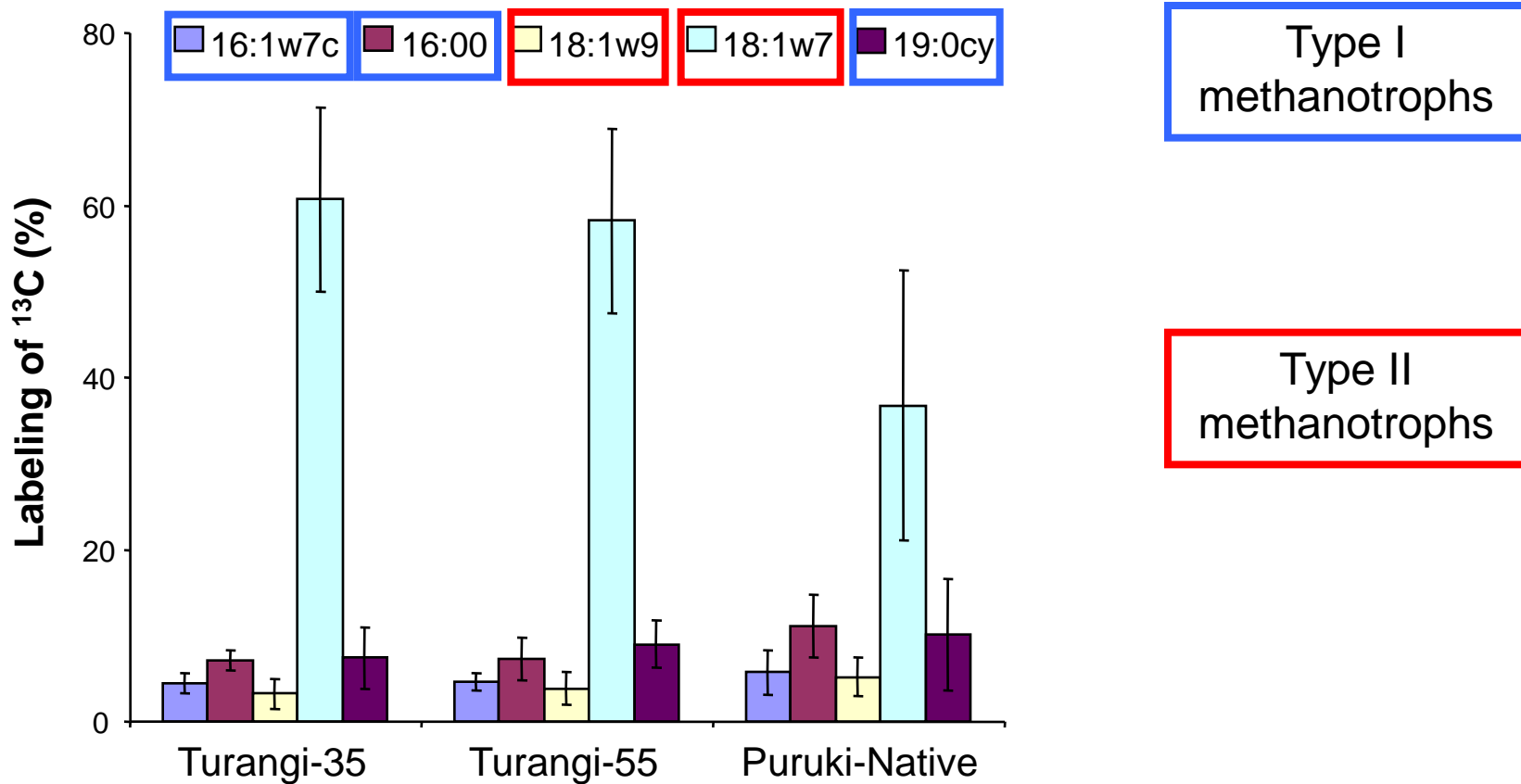


Methane fluxes and afforestation

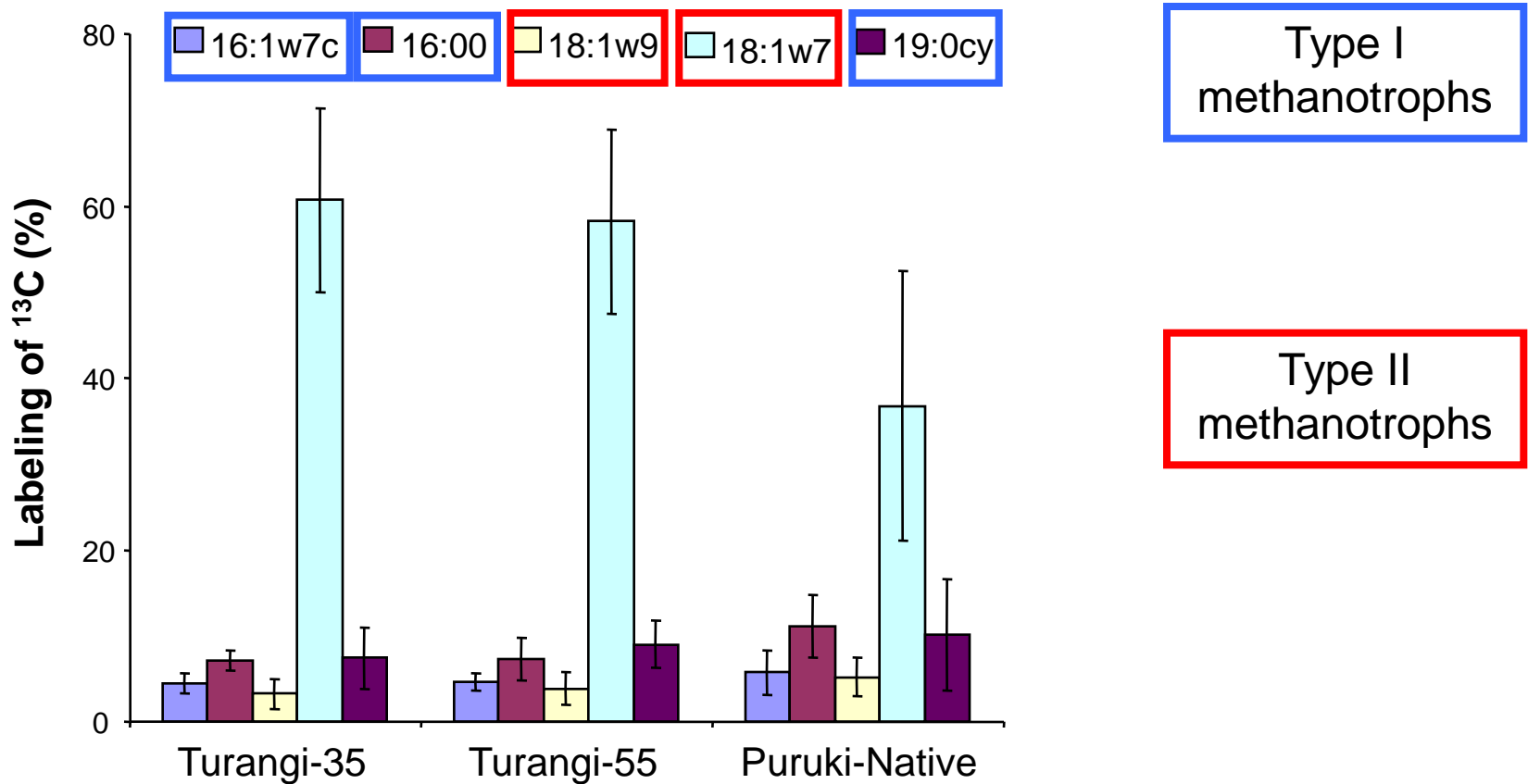


Higher oxidation rates in old and native forests

Linking community with function (PLFA-SIP)



Linking community with function (PLFA-SIP)



Type II methanotrophs most active at oxidising atmospheric CH₄

Methanotroph community structure (T-RFLP) – T-RF presence

Site	<i>pmoA</i>
Turangi	
Pasture ²	T-RF 33/129 T-RF 245
Pine forest ²	T-RF 33/129; T-RF 245
Turangi-35	T-RF 33/129
Turangi-55	T-RF 33/129

Methylocapsa spp.
(type II methanotrophs)

Methylococcus capsulatus
(type I methanotrophs)

Methanotroph community structure (T-RFLP) – T-RF presence

Site	<i>pmoA</i>
Turangi	
Pasture ²	T-RF 33/129 T-RF 245
Pine forest ²	T-RF 33/129; T-RF 245
Turangi-35	T-RF 33/129
Turangi-55	T-RF 33/129
Puruki	
Pasture ³	T-RF 129 T-RF 245
Pine forest ³	T-RF 129; T-RF 245
Native forest	T-RF 33/129

Methylocapsa spp.
(type II methanotrophs)

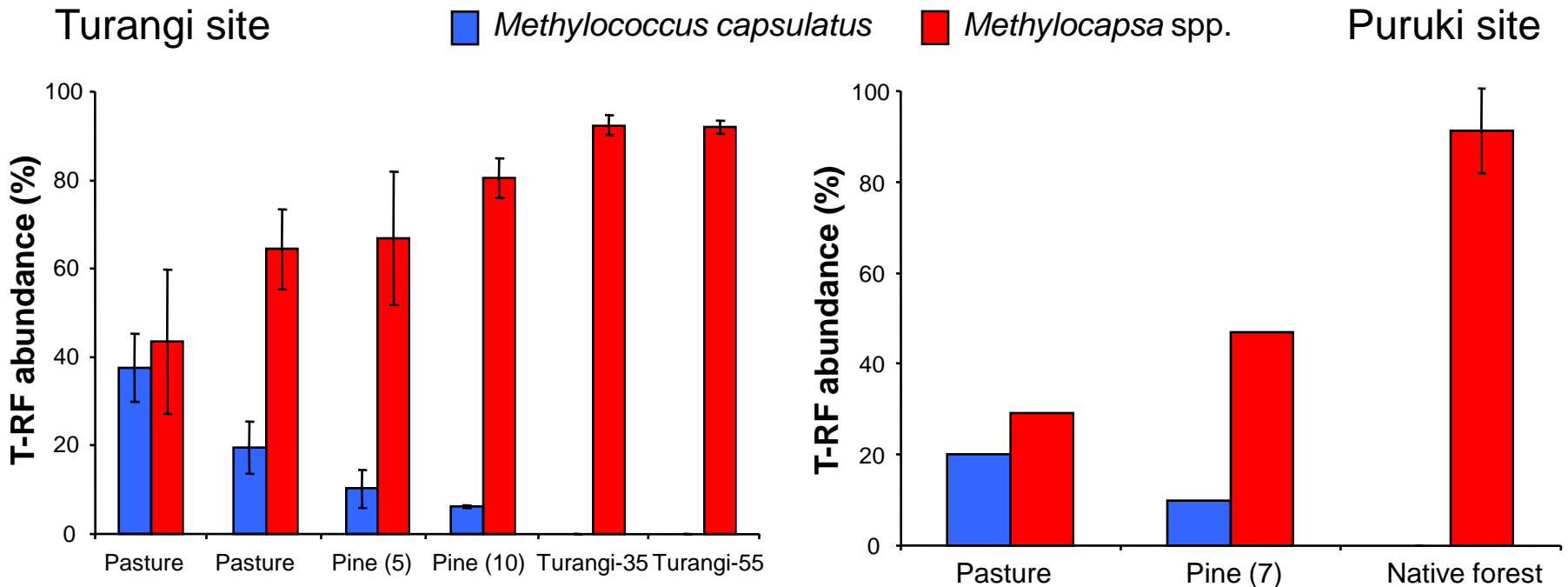
Methylococcus capsulatus
(type I methanotrophs)

Disappearance of *Methylococcus capsulatus* (type I methanotroph) in old and native forests

² Singh *et al.*, (2009);

³ Tate *et al.*, (2007)

Methanotroph community structure (T-RFLP) – T-RF abundance



Pasture à young forest à old/native forest
***M. capsulatus* (type I) è *Methylocapsa* sp. (type II)**

Conclusions

- | Rapid effect of afforestation of pastures (10-35 years)
 - Methane fluxes
 - Methanotroph activity and community structure
- | Establishment of a stable and active population of type II methanotrophs in forests
 - Independent of vegetation
- | No differences between the old forests (Turangi) and the native forest (Puruki)

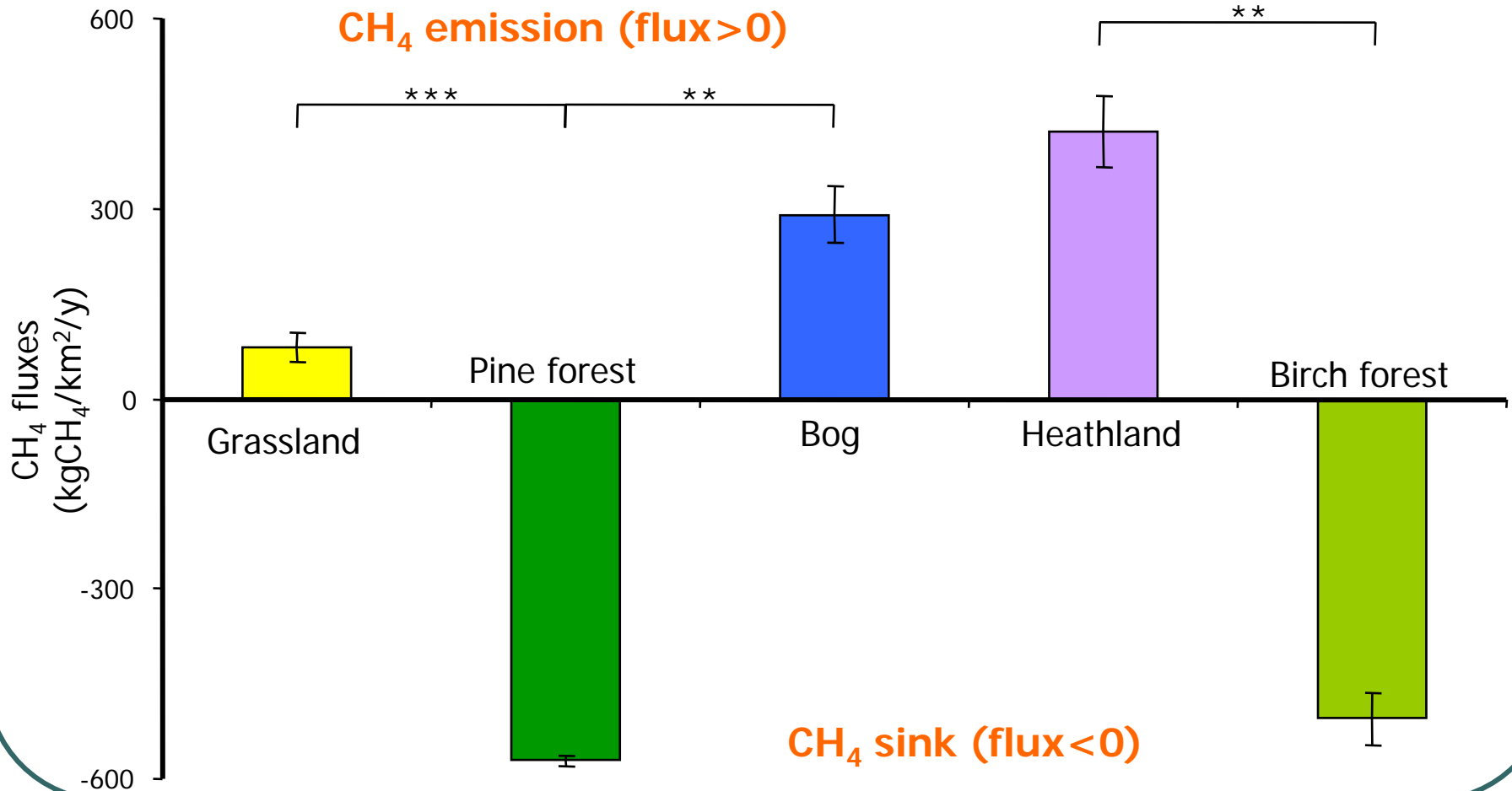
On-going works

- | Submission for publication
- | Work on Scottish soils
 - Study the impact of land-use change on the CH₄ fluxes and shifts in methanotroph community structure

CH₄ fluxes in Scotland

- **Effect of afforestation on methane fluxes**
 - Compare methane fluxes from sites with differing land uses
 - Grassland or bog → pine forest
 - Heathland → birch forest

Effect of land-use change on methane fluxes



Acknowledgements

| PhD supervisors

- Dr Brajesh K. Singh and Prof. Pete Millard, MLURI
- Prof. Colin Murrell, University of Warwick
- Dr Elizabeth M. Baggs, University of Aberdeen

| At Landcare Research, New Zealand:

- Dr Kevin Tate, Dr Jagrati Singh, Dr Des Ross

The End

Thank you!!





Questions?

