Microbes at extreme environments and the Spanish Network of Extremophiles

Juan M. Gonzalez, Enrique J. Gómez,
José A. Romero, Alba M.P. Cuecas

jmgrau@irnase.csic.es

Instituto de Recursos Naturales y Agrobiología de Sevilla (IRNAS),
Consejo Superior de Investigaciones Científicas (CSIC),
Sevilla, Spain

Outline

Introduction

Main

Hot springs microbial communities

Fervidobacterium isolate and genome project

CO-utilizing, thermophilic microorganisms

Soil thermophiles

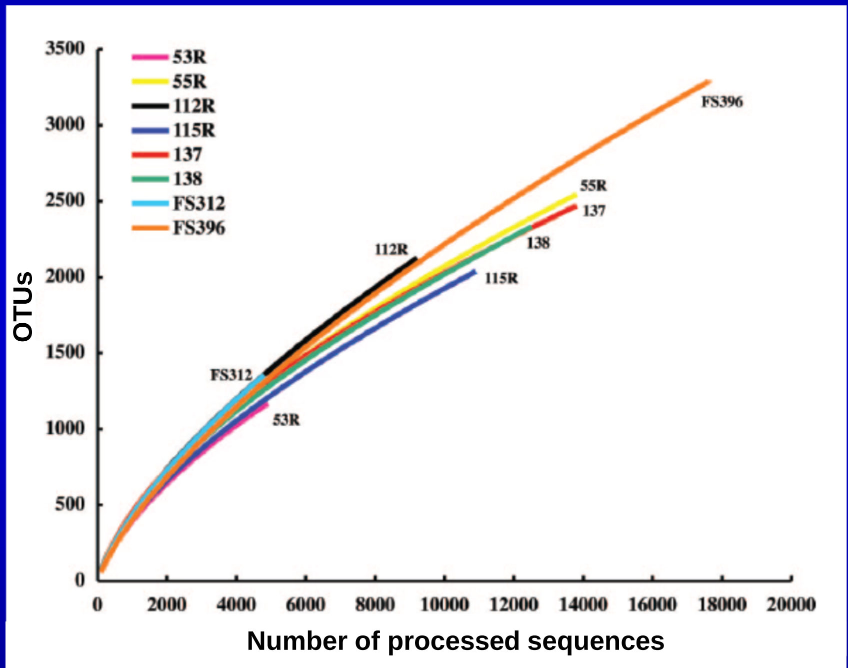
- Isolates
- Communities
- Enzyme activity

Spanish Network of Extremophiles

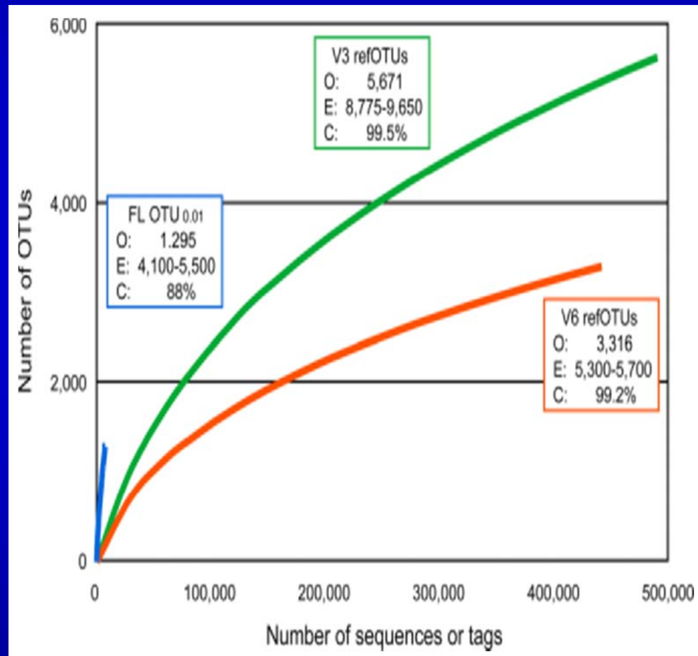
Acknowledgements

Introduction

Microbial diversity



Sogin et al. 2006



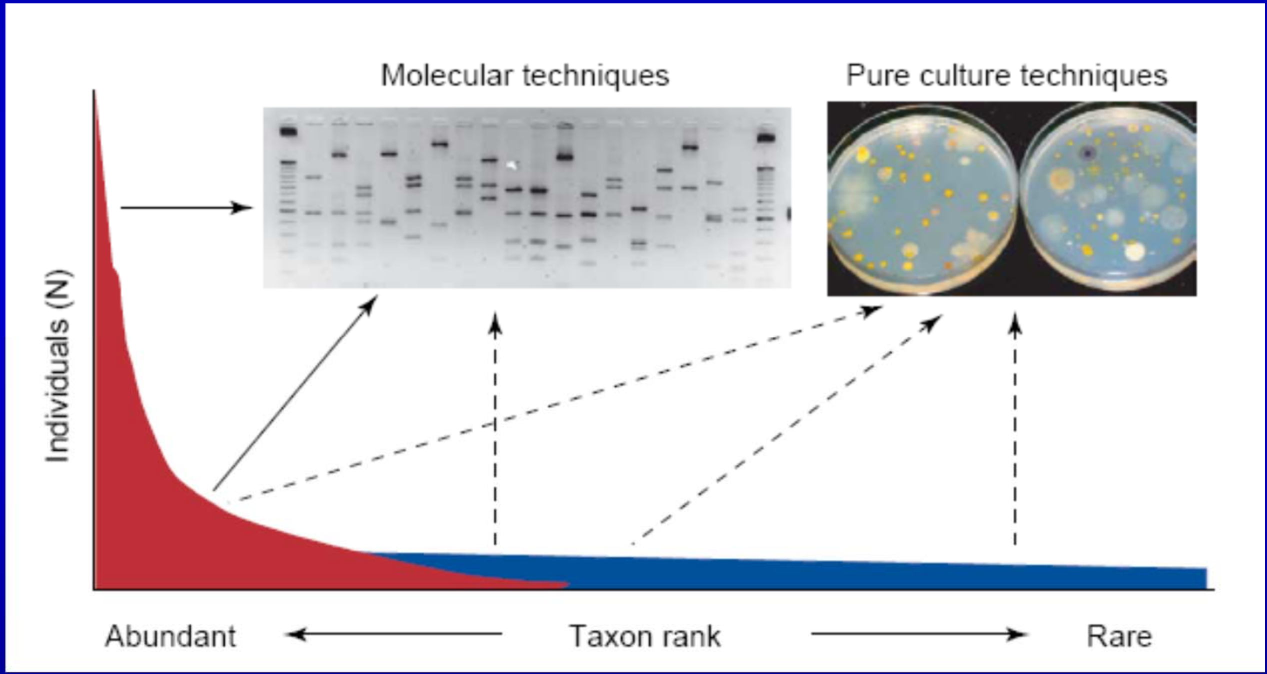
Dethlefsen et al. 2013

How high is microbial diversity?

Introduction

Understanding microbial communities

The 'Common' and the 'Rare'



Pedrós-Alió, 2006

A few highly abundant and many low abundant microorganisms

Most microorganisms represent minor fractions of the communities

Introduction

Extremophiles

Extremophiles are those (micro)organisms living under extreme conditions

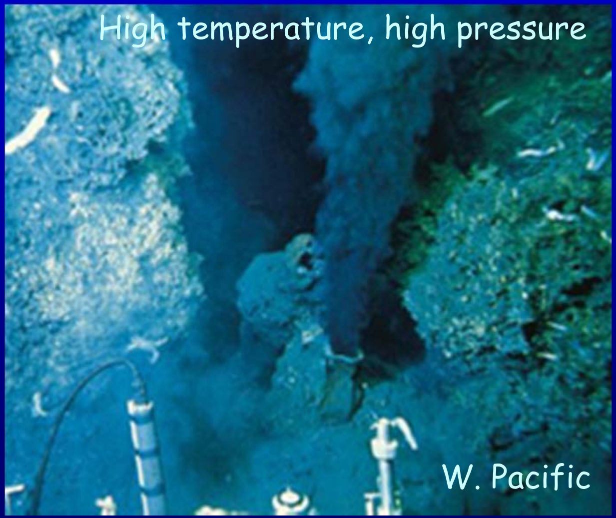
Some examples:

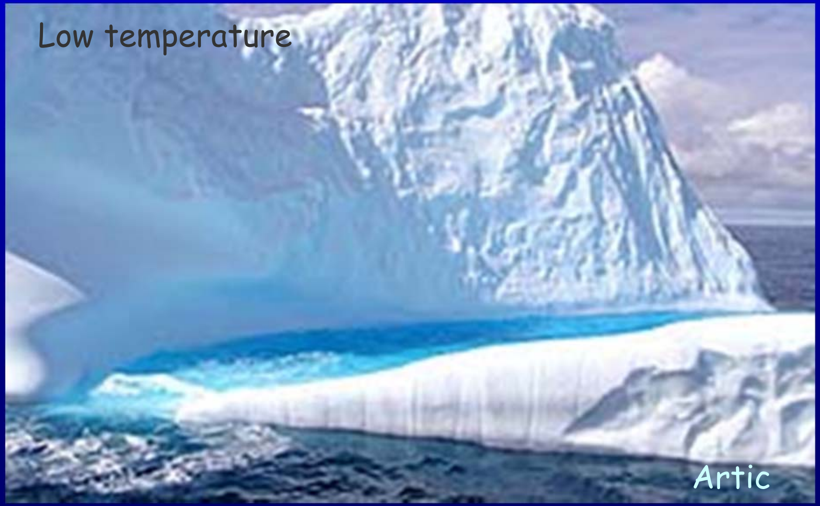
High temperature	up to 113-120°C	Hyperthermophiles, thermophiles
Low temperature	below 0°C	Psychrophiles
Low pH (acid)	<3	Acidophiles
High pH (alkaline)	>9	Alkalophiles
High salt	up to saturation	Halophiles
High pressure	>500 atm	Barophiles
Dehydration	Reduced water availability ($<0.7\ a_w$)	Xerophiles
Other conditions	Example: methanogens	

Introduction

Extremophiles

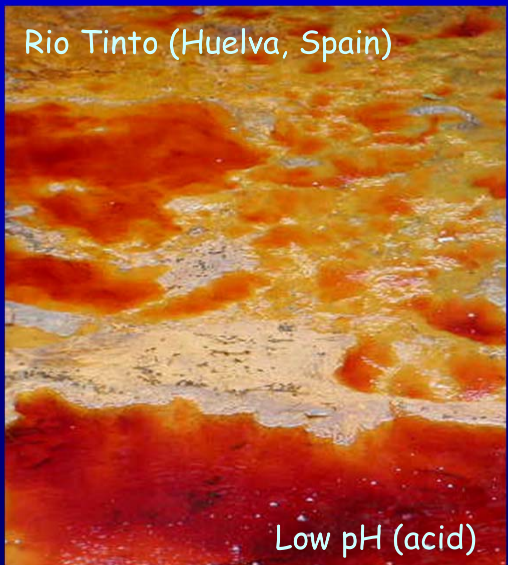
High temperature





Introduction

Extremophiles




Canary Islands (Spain)

Limited water availability

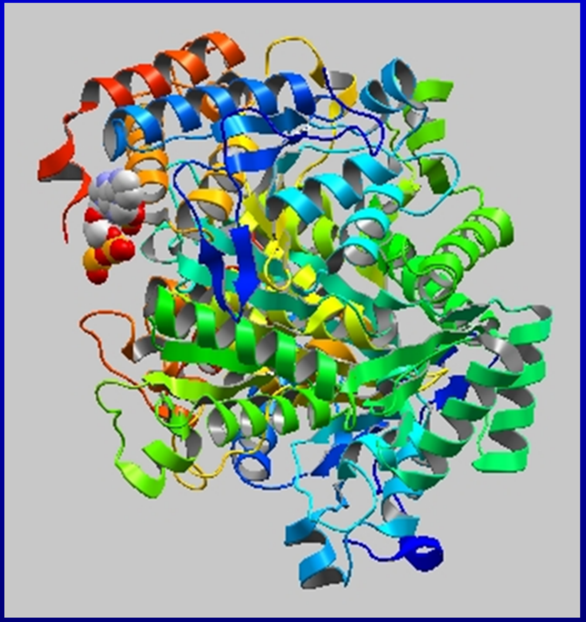

Sahara desert

Introduction

Extremophiles

High molecular stability

Interest in Biotechnology



Hot Springs

Microbial communities at a 50°C temperature gradient

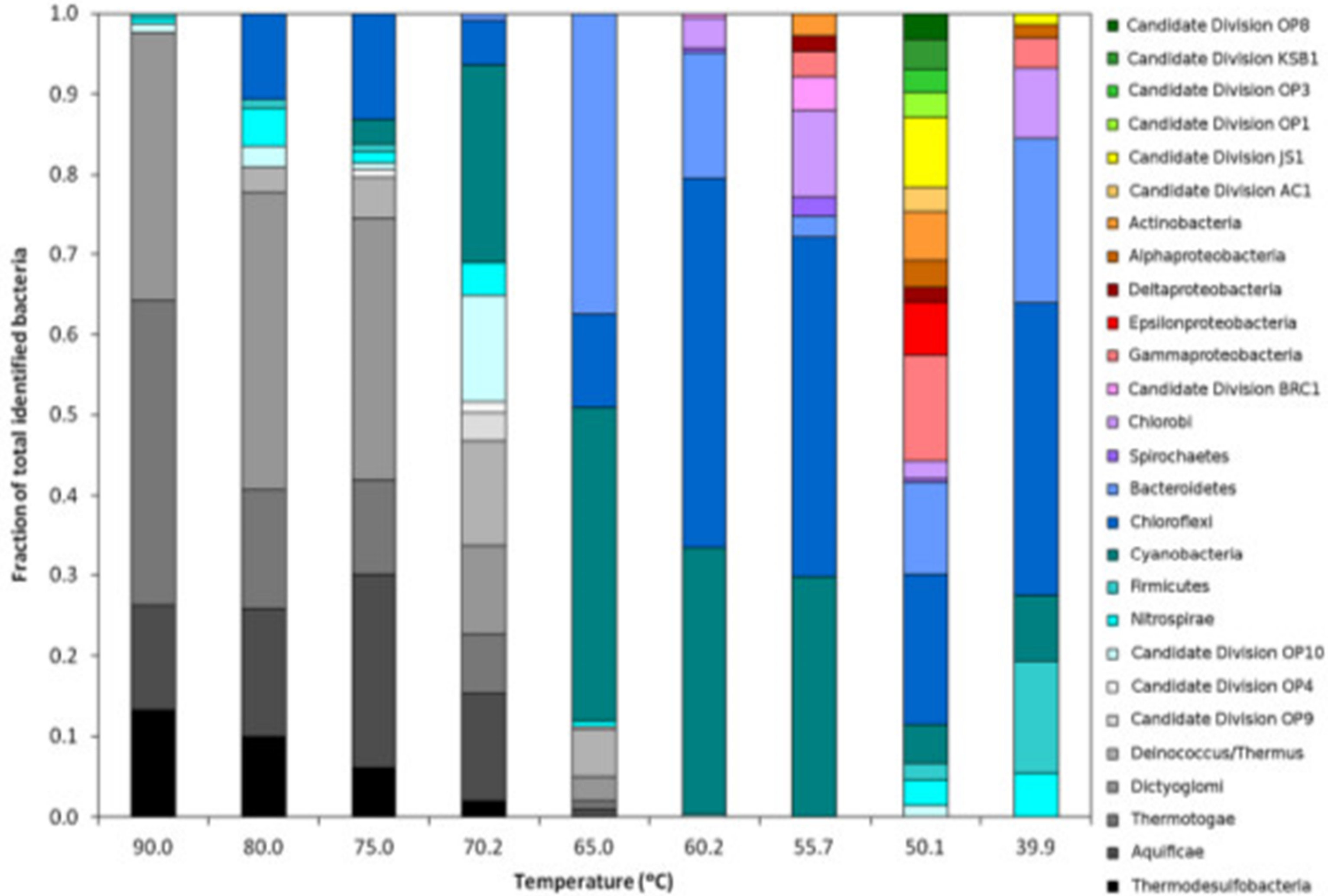


Fig. 2 Relative abundance of major bacterial phyla along a 50 °C temperature gradient at Mae Fang Hot Springs

(Cuecas et al. 2015)

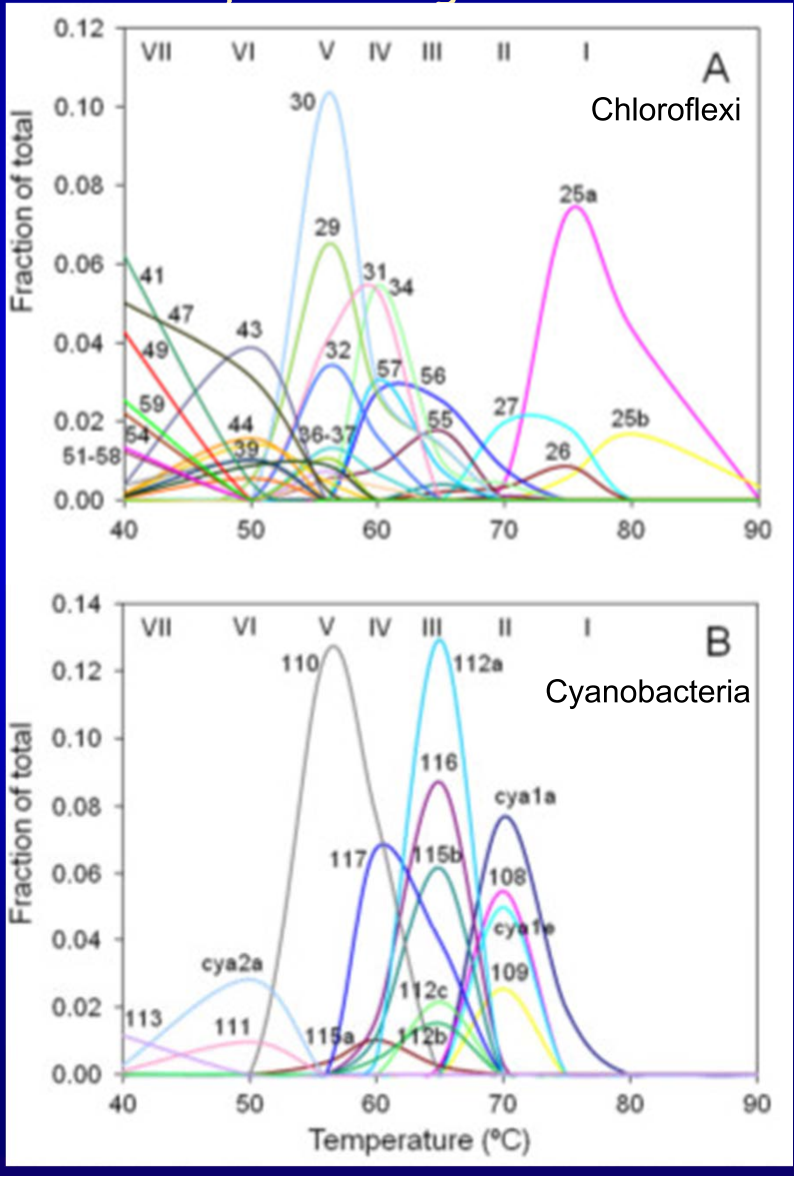
Mae Fang Hot Springs (Northern Thailand)

Hot Springs

Microbial communities at a 50°C temperature gradient

Distribution of major phylotypes within the Cyanobacteria and Chloroflexi

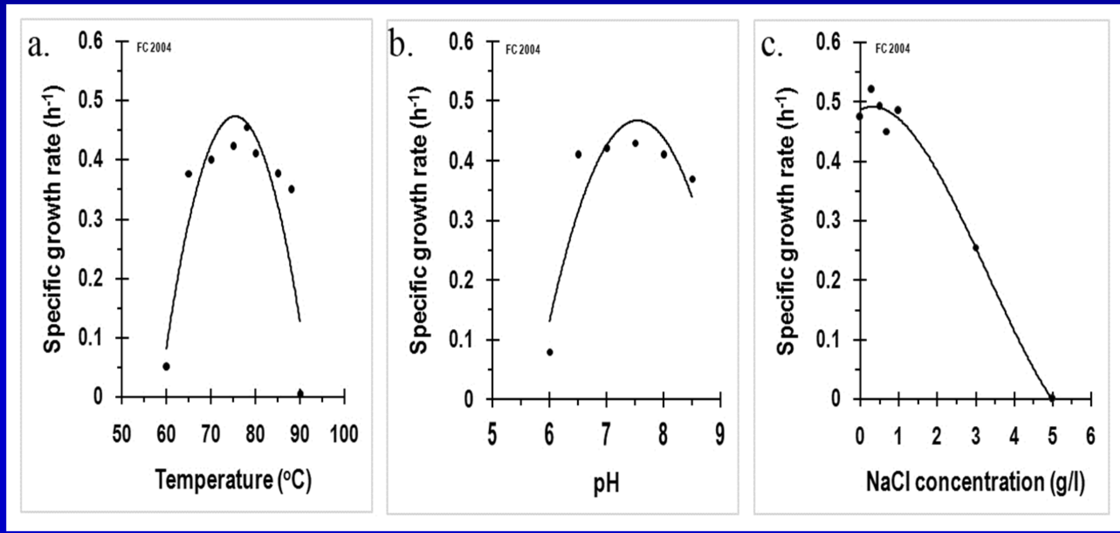
Differences between communities increased with difference in temperature

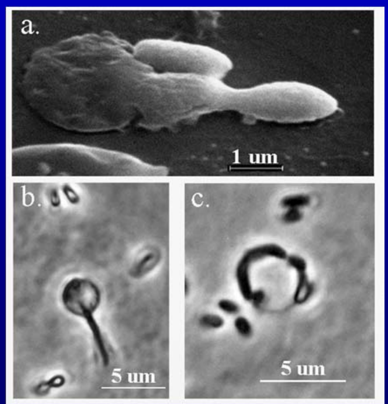


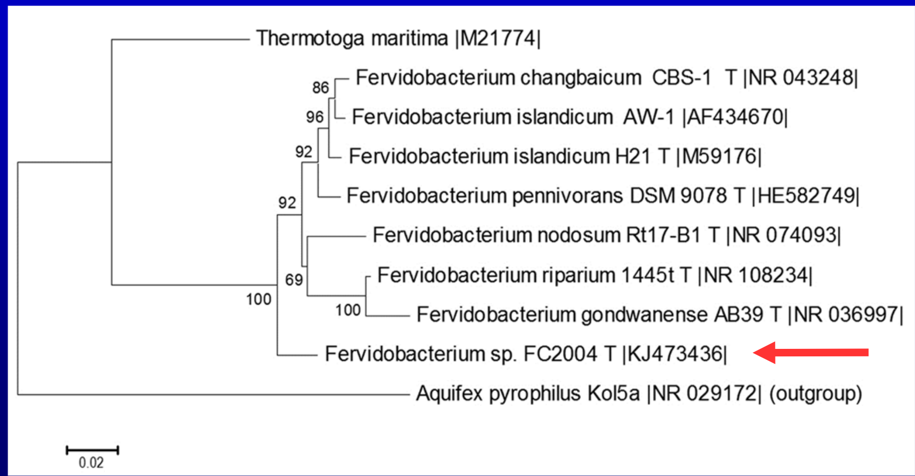
(Cuecas et al. 2015)

Fervidobacterium thailandensis

Isolate from Mae Fang Hot springs (N Thailand)







Fervidobacterium thailandensis

Genome comparison for 4 *Fervidobacterium* genomes

	F.thailandensis	*F.nodosum*	*F.pennivorans*	*F.islandicum*
CDS	1846	1829	2012	1961
Length (bp)	1978803	1948941	2166381	2359755
Contigs	1	1	1	12
GC%	45.84	34.99	38.88	40.74
rRNA operons	2	2	2	

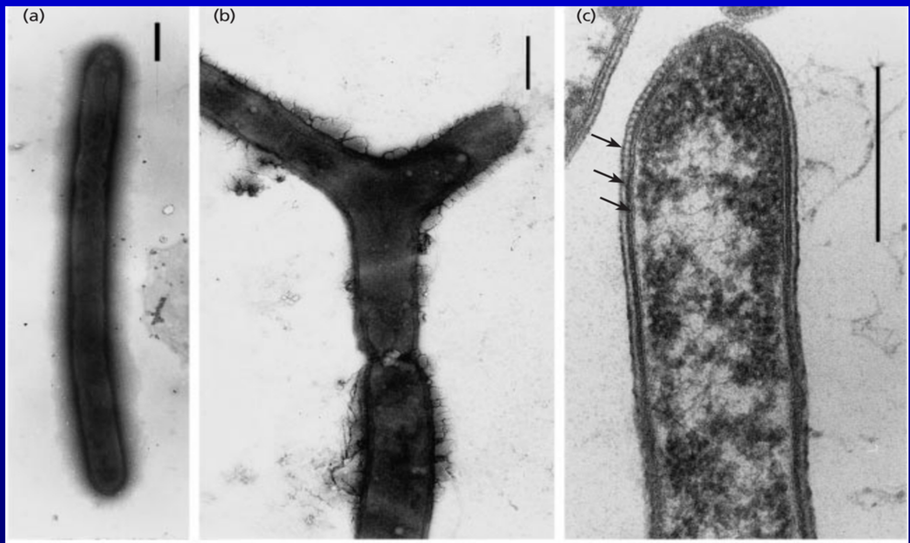
CO-utilizing, thermophilic microorganisms

Caldanaerobacter subterraneus species

Thermophilic fermentative bacteria growing on carbohydrates producing acetate, alanine, H_2 and CO_2.

They are able to utilize CO (hydrogenogenic microorganisms)

Optimum growth temperature: 70°C (range 50°C-80°C)



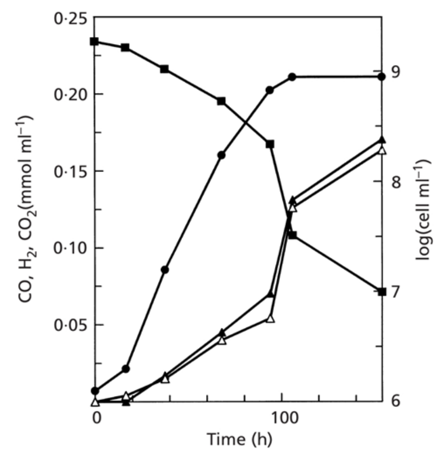


Fig. 2. Growth of strain JM^T at 70 °C in Medium 1 supplemented with 0·2 g yeast extract l^{-1} under a CO atmosphere (●), CO consumption (■), H_2 production (▲) and CO_2 production (△).

Caldanaerobacter subterraneus pacificus (former *Carboxydobrachium pacificum*) (Sokolova et al. 2001)

CO-utilizing, thermophilic microorganisms

Caldanaerobacter subterraneus genomes

C. s. pacificus

C. s. tengcongensis (Bao et al. 2002)

C. s. yonseiensis (Lee et al. 2013)

Table 1 Overview of *C. subterraneus* genomes

	C. subterraneus subsp. *pacificus*	*C. subterraneus* subsp. *tengcongensis*	*C. subterraneus* subsp. *yonseiensis*
Genome size (Mb)	2.39	2.69	2.7
Genome GC content (%)	37.7	37.8	37.7
Number of contigs	135	1 (complete chromosome)	102
CDS	2511	2588	2711
Operon	871	1291	880
Hypothetical proteins[a]	962 (38.31 %)	855 (33.04 %)	836 (30.84 %)
Average gene length	819	905	834
rRNA	11	12	18
rRNA average GC content (%)	59.8	59.81	59.3
tRNA	49	56	59
tRNA average GC content (%)	60.26	60.12	59.98
Number of horizontally transferred CDSs[b]	173 (6.88 %)	121 (4.67 %)	127 (4.68 %)
Origin	Pacific Ocean hot vents	Terrestrial hot spring	Geothermal hot stream
Reference	This study	[12]	[14]

[a]Percentage of hypothetical proteins of all genome proteins is in parentheses

[b]Detected by GOHTAM. In parentheses is the percentage of horizontally transferred CDSs of all CDSs present in the genome

CO-utilizing, thermophilic microorganisms

Caldanaerobacter subterraneus and Horizontal gene transfer (HGT)

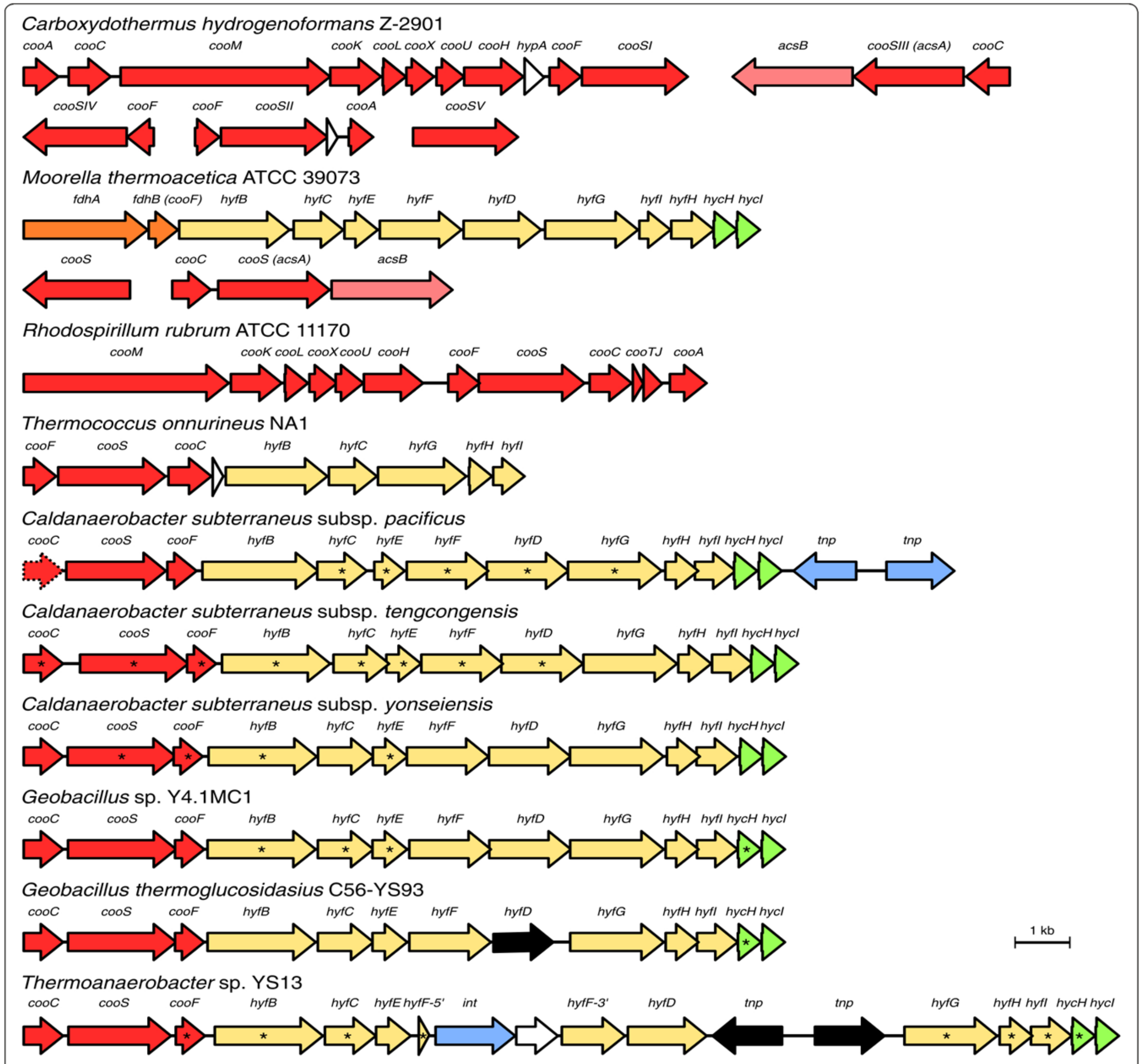


Fig. 3 Comparison of the CODH-hydrogenase gene cluster organization between *C. subterraneus* and other species. *Arrows* represent genes and their respective direction of transcription. *Asterisks* indicate significant prediction of HGT in *C. subterraneus* and *Geobacillus thermoglucosidans* strains

CO-utilizing, thermophilic microorganisms

Comparative phylogenetic tree of 16s rRNA and CooS genes

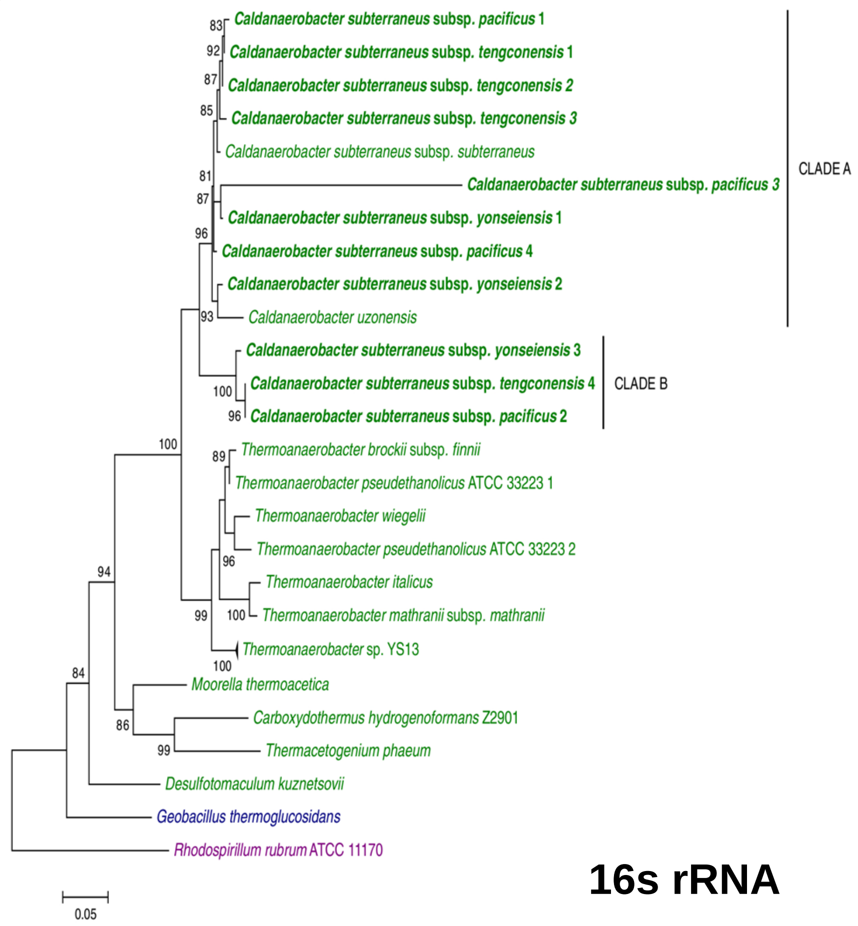


Fig. 1 Evolutive history of *Caldanaerobacter subterraneus* subspecies. The 16S rRNA tree was constructed using the maximum-likelihood method. aLRT values greater than 70 % are shown next to the branches. The tree is drawn to scale, with branch lengths in the same units as those of the evolutionary distances used to infer the phylogenetic tree. *Caldanaerobacter subterraneus* subspecies are in **bold**. Other Clostridiales species are in *green*, Bacillales species in *blue*. *R. rubrum* was used as outgroup (in *purple*)

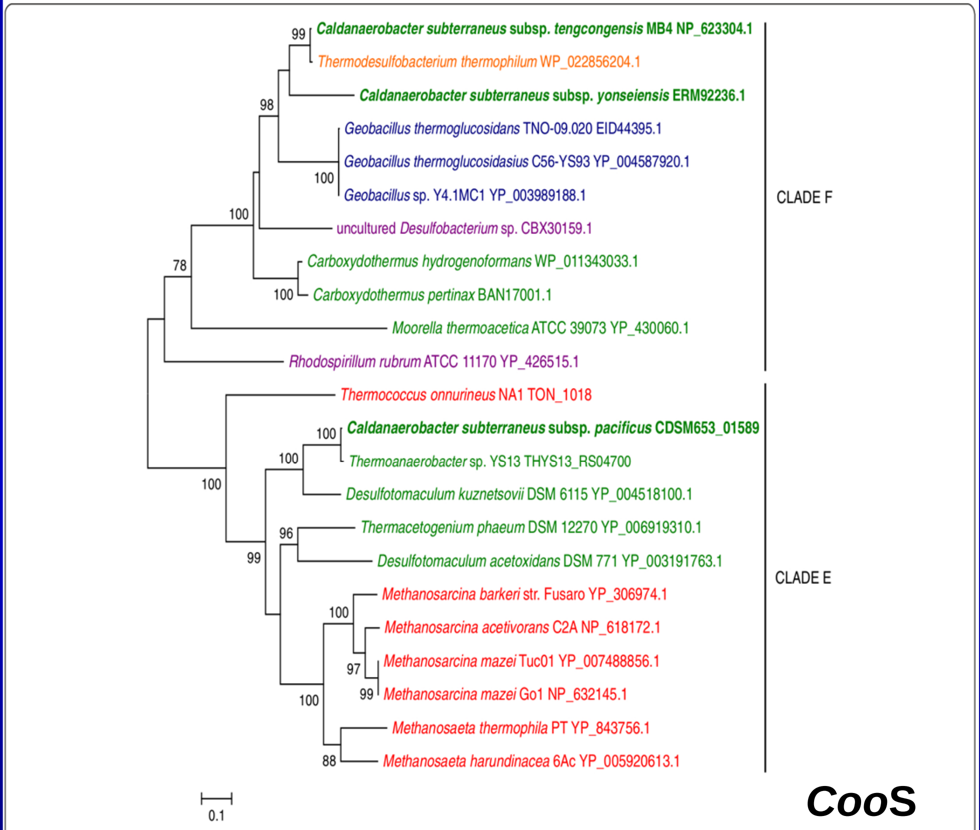


Fig. 4 Evolutive history of CODH (CooS) from *C. subterraneus* subspecies. Details are as shown in Fig. 1, unless specified otherwise. *Thermodesulfobacterium* is in *orange*, and Archaea are in *red*. Accession number or locus tag are adjacent the species name. This tree is mid-point rooted. Classification of clades as in Techtmann et al. [29]

(Sant'Anna et al. 2015)

Thermophiles in temperate environments

Temperatures of 40°C and above occur in soils

Thermophiles (growth 40°C-80°C) are present in temperate soils (Marchant et al. 2002)

Role of these thermophiles and capability to grow/survive

Potential role in S cycle:

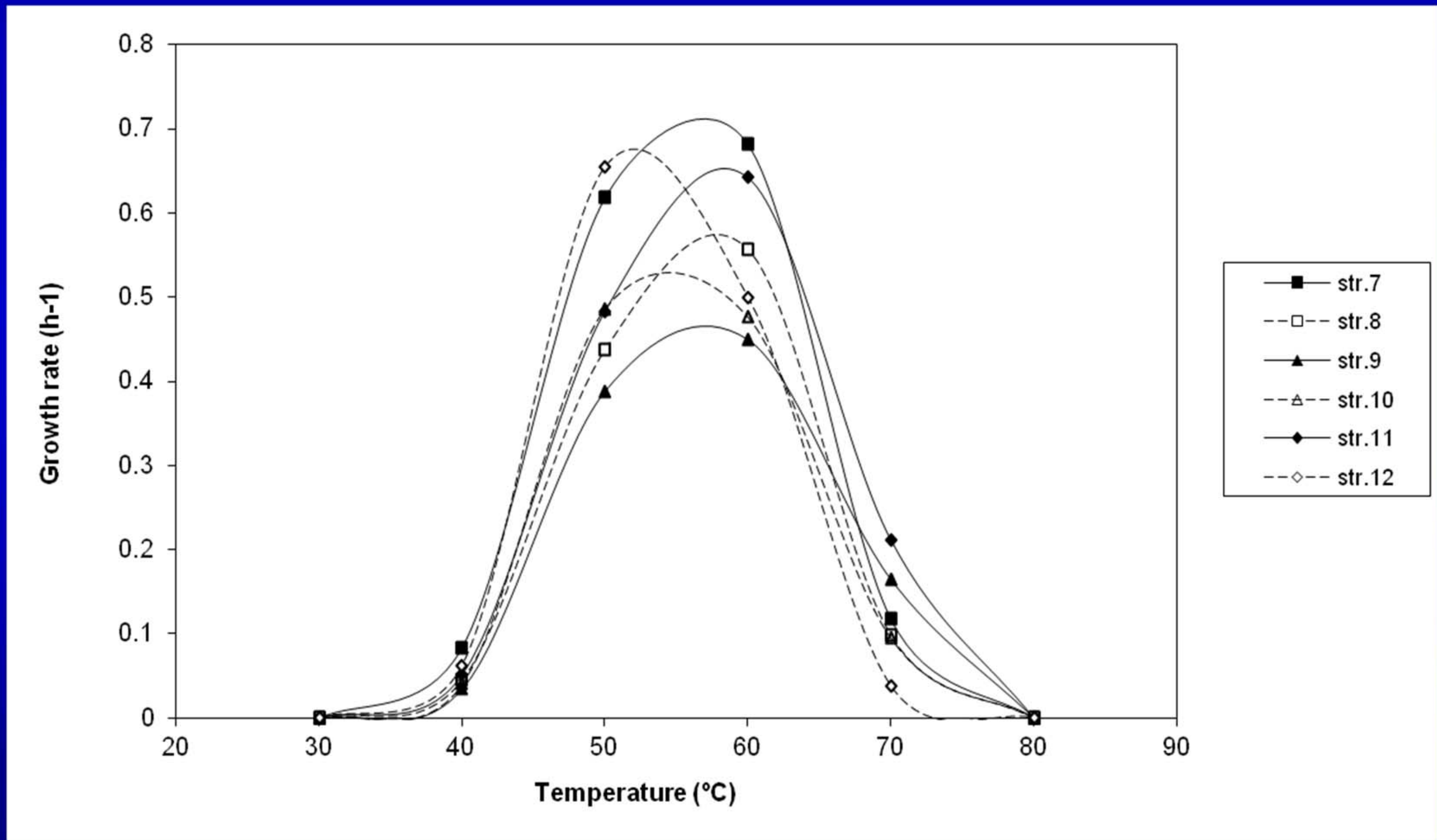
In soils, most S is in organic compounds (>90%)

Poor S mineralization by soil microorganisms at 20°C (Ghani et al. 1993)

Ramiro Alloza and Enrique Arranz (U. Zaragoza)

Thermophiles in temperate environments

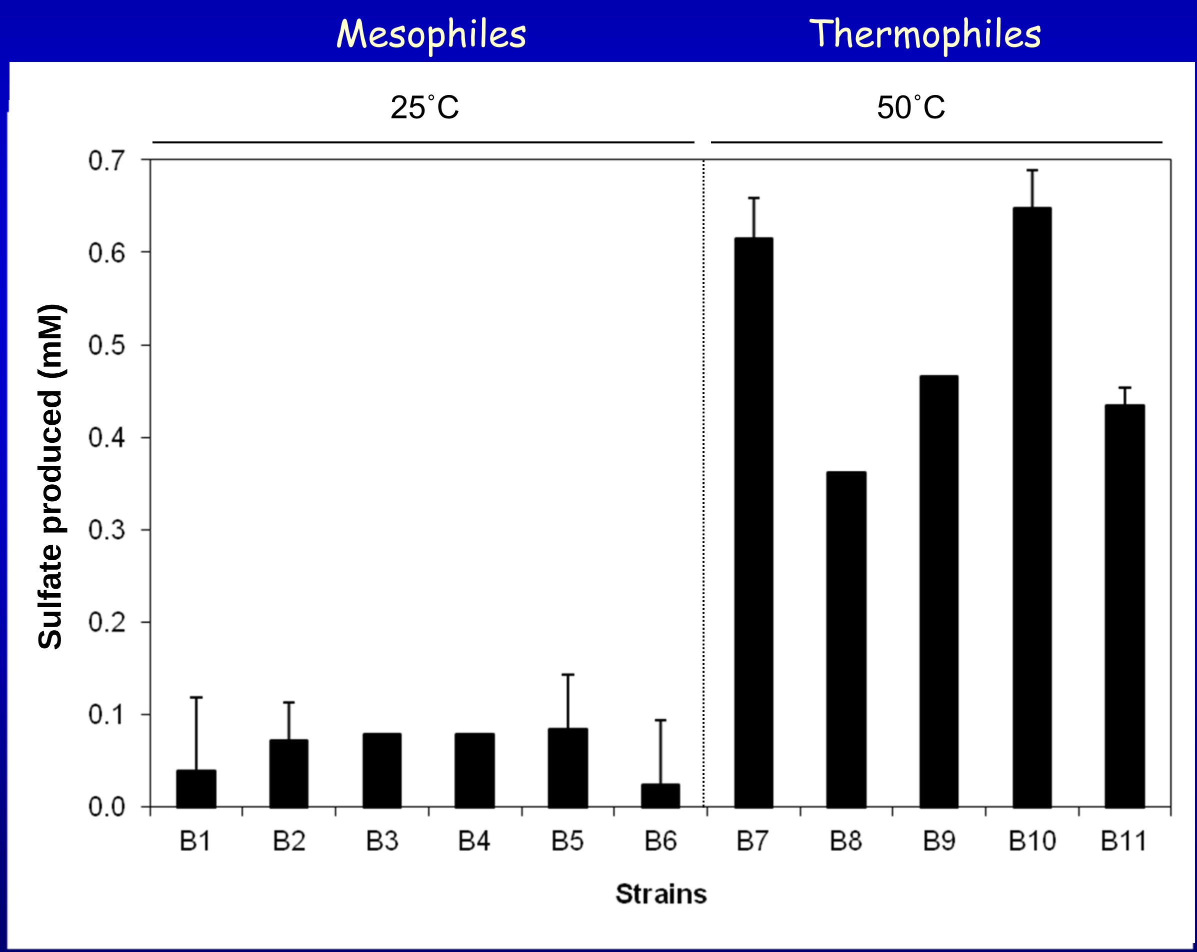
Effect of temperature on the growth rate of thermophilic isolates from soil



Represented genera: *Geobacillus, Ureibacillus, Brevibacillus, Bacillus*

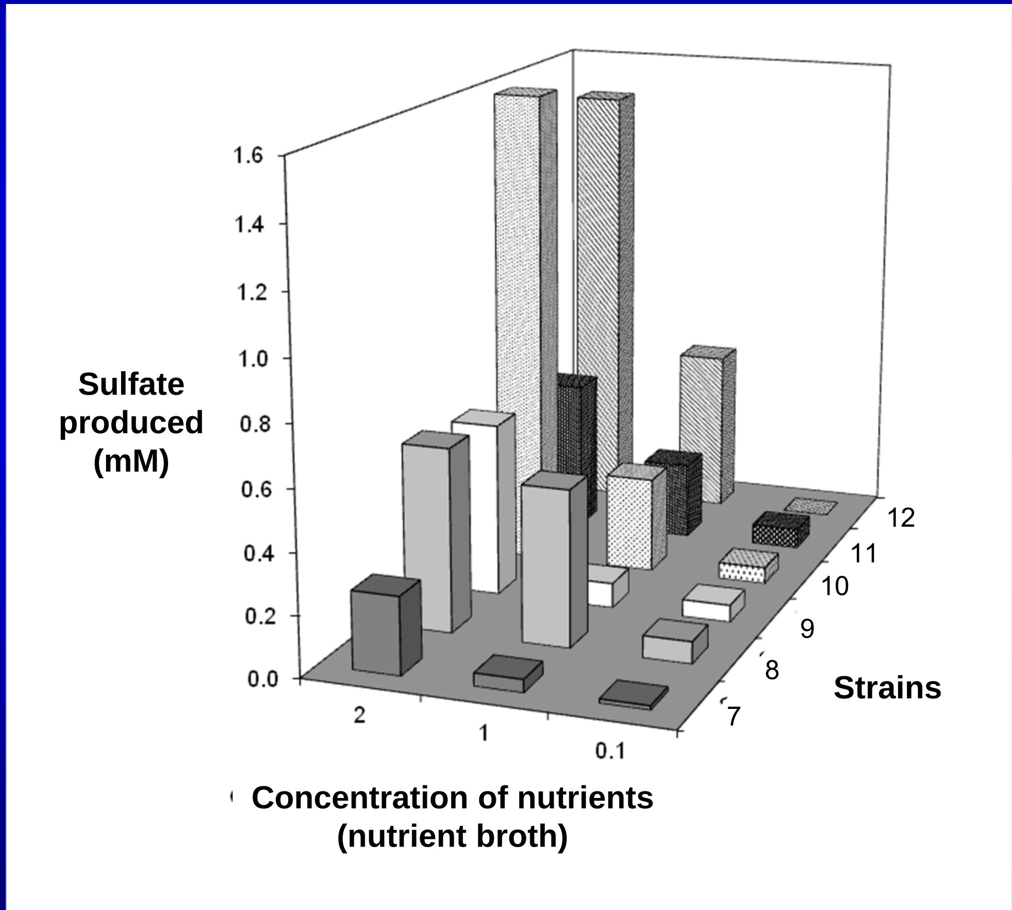
Thermophiles in temperate environments

Production of sulfate by the mesophilic and thermophilic isolates



Thermophiles in temperate environments

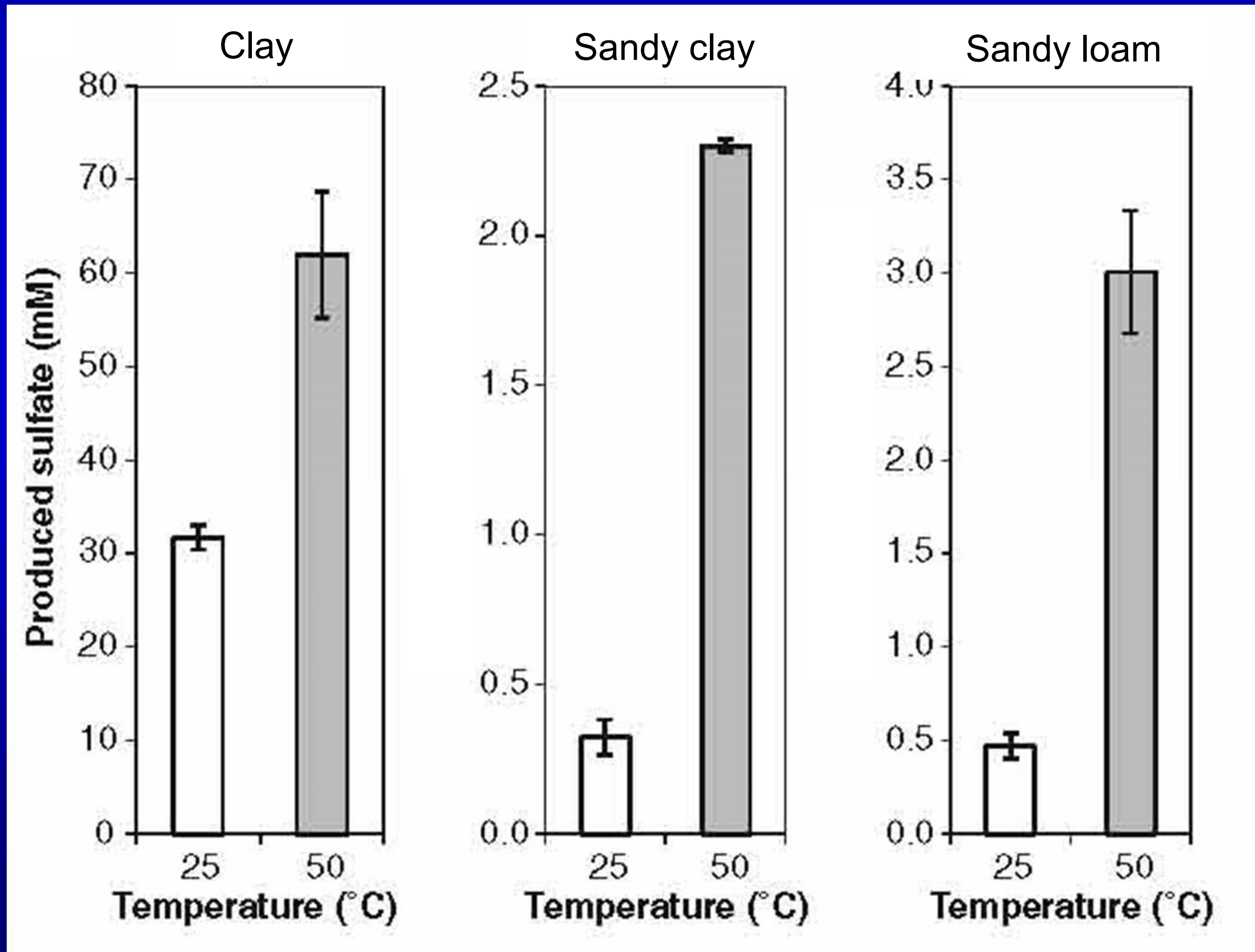
Production of sulfate by thermophilic isolates at different organic loads



Production of sulfate depends on the consumption of organic compounds

Thermophiles in temperate environments

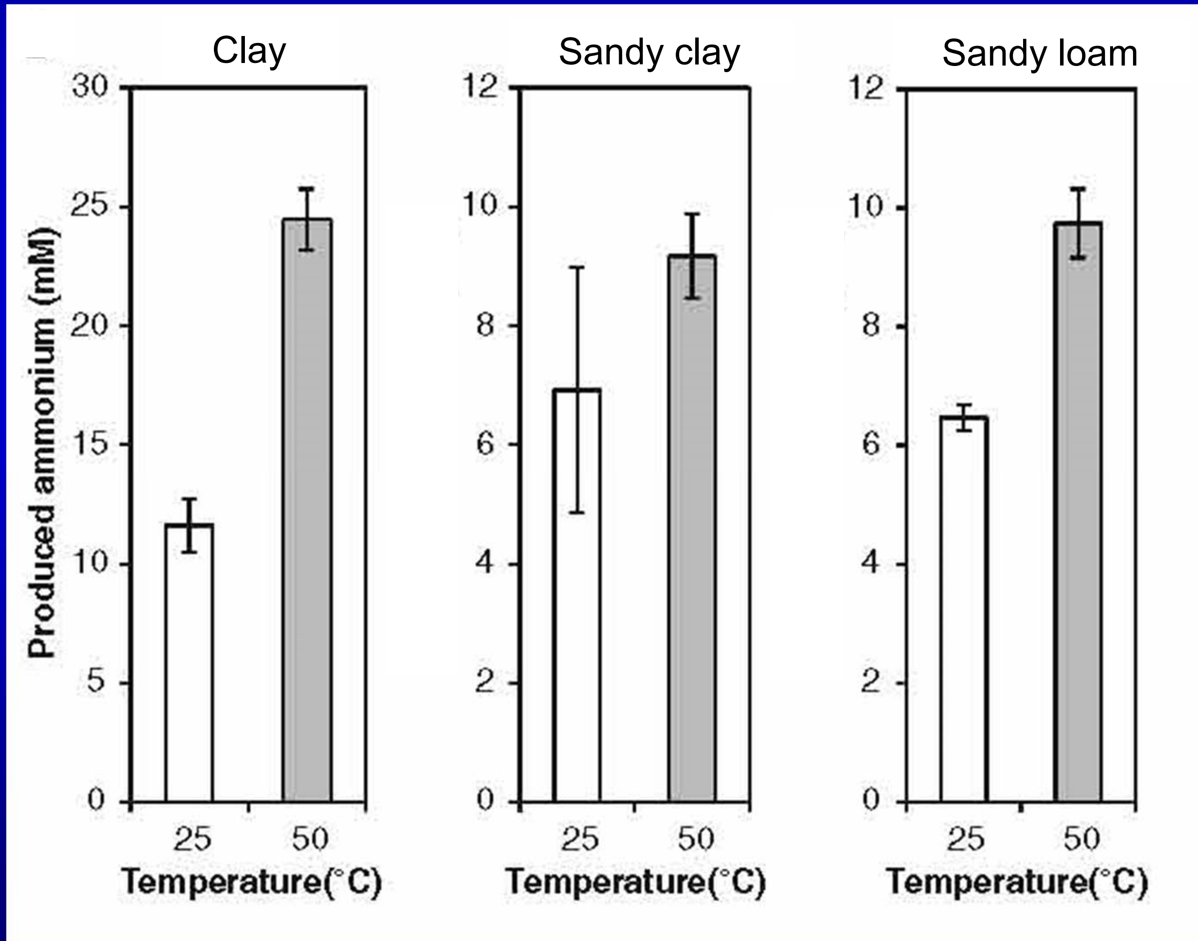
Is sulfate also produced by natural bacterial communities?



(Portillo et al. 2012)

Thermophiles in temperate environments

These thermophilic communities released sulfate and ammonium



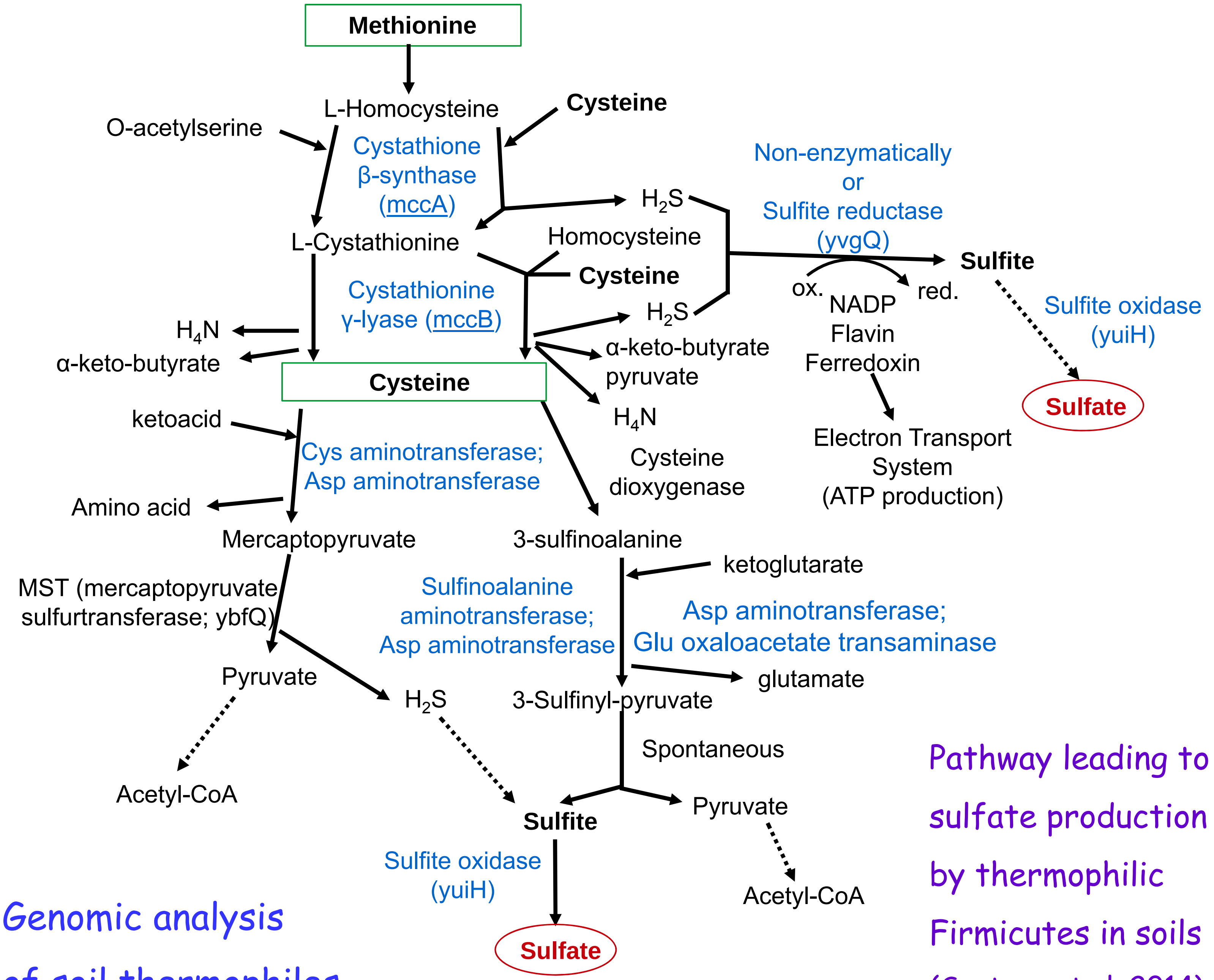
(Portillo et al. 2012)

Thermophiles in temperate environments

- These thermophiles consume organic matter releasing sulfate and ammonium
- They can be important for S and N cycling and the fertilization of soils
- Plant growth is stimulated by this group of thermophiles

Santana et al. 2013

Thermophiles in temperate environments



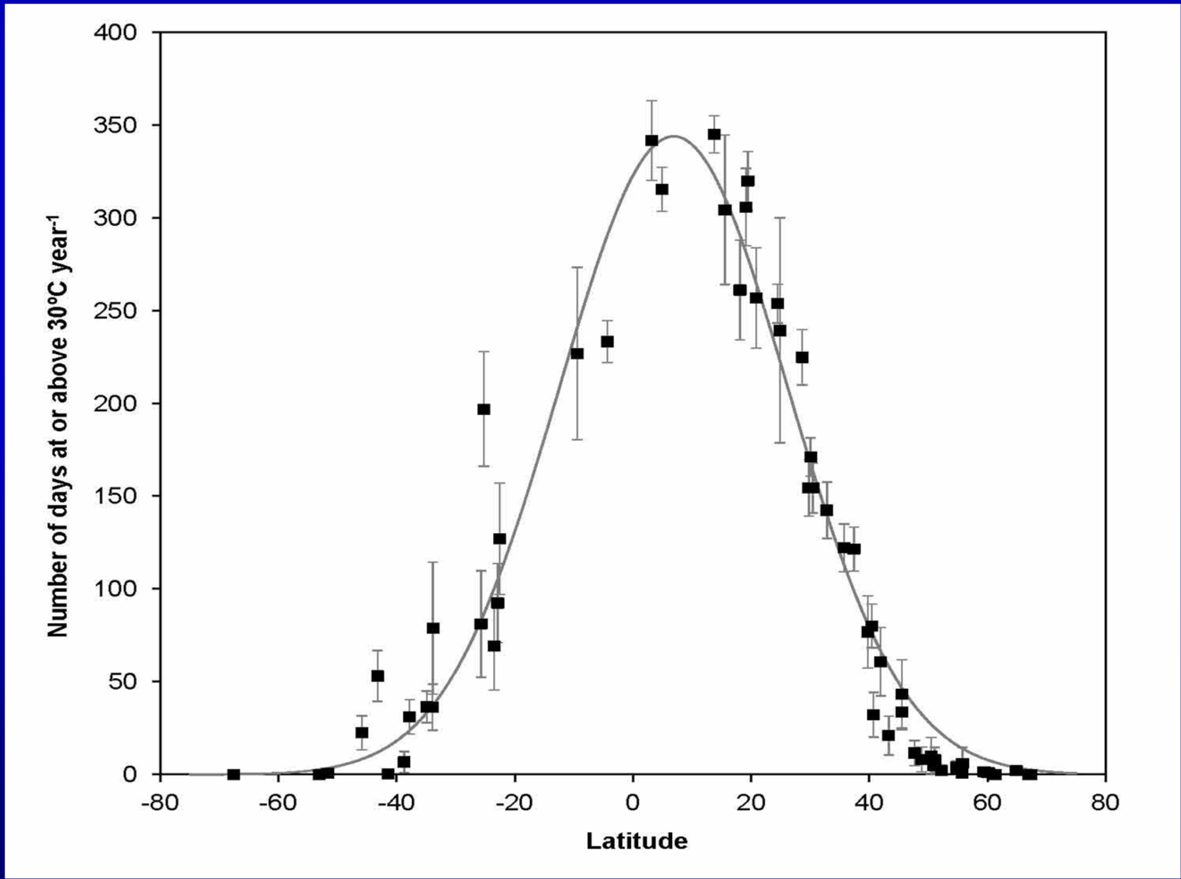
Genomic analysis of soil thermophiles

Pathway leading to sulfate production by thermophilic Firmicutes in soils (Santana et al. 2014)

Thermophiles in temperate environments

Can these thermophiles play a significant role in soils?

Average number of days per year with an air temperature ≥30°C

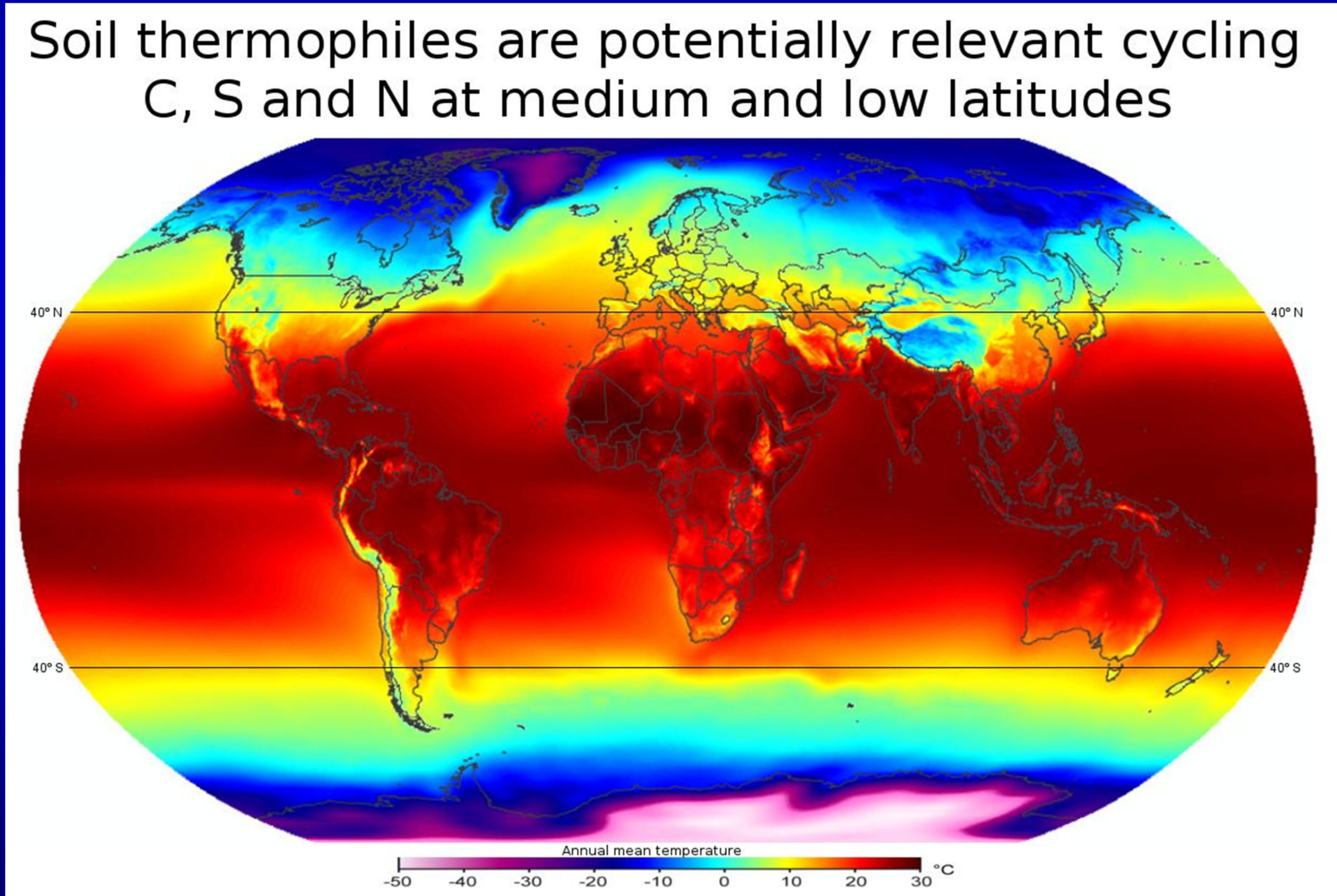


(Santana and Gonzalez, 2015)

Source of data: National Oceanic and Atmospheric Administration (NOAA, USA)

Thermophiles in temperate environments

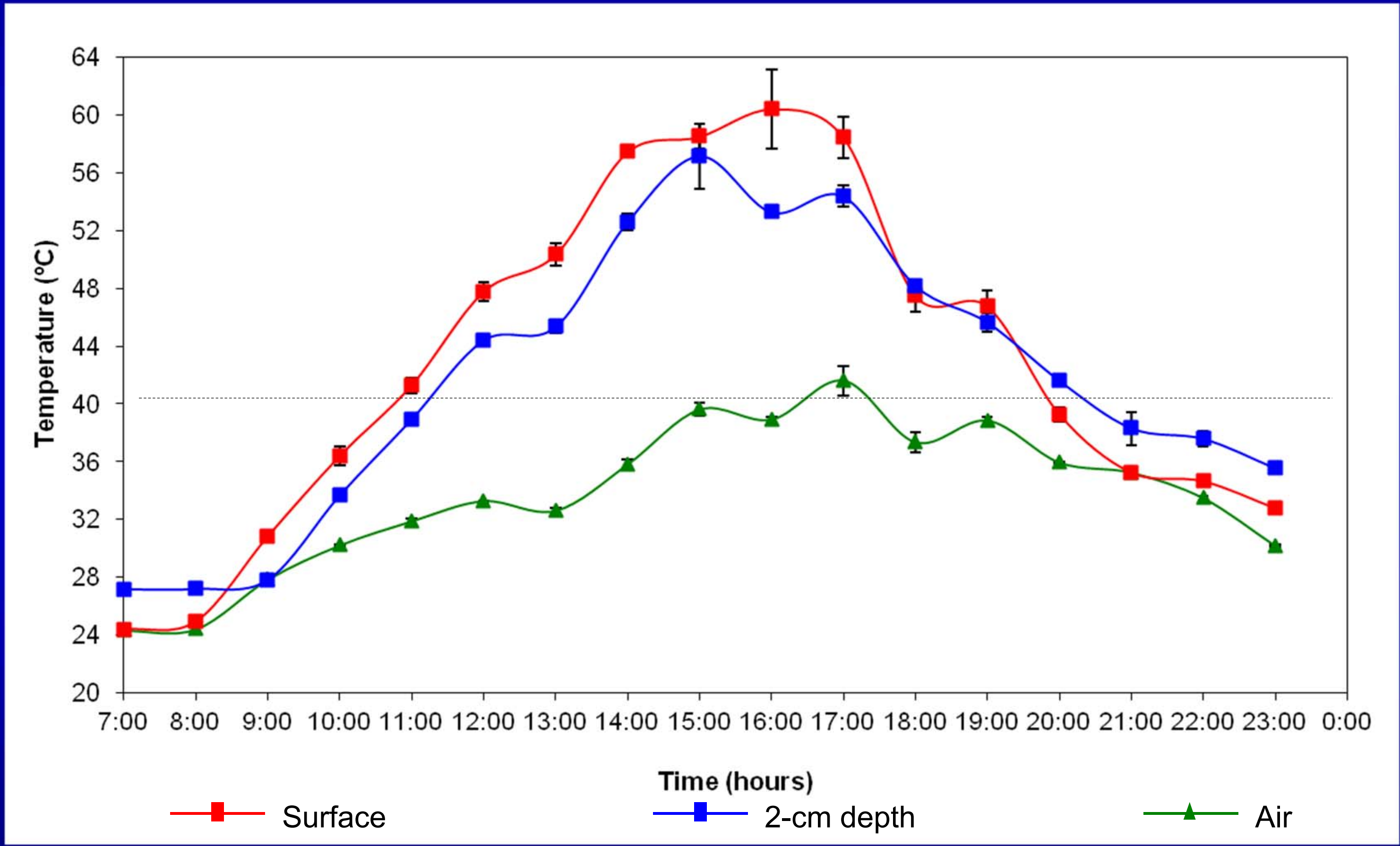
At medium and low latitudes, soil thermophiles could be relevant on C, S, and N cycling



Reports on the significance of soil thermophiles at 56°N (Alberta, Canada)(Wong et al. 2015)

Thermophiles in temperate environments

Can these thermophiles play a significant role in soils?

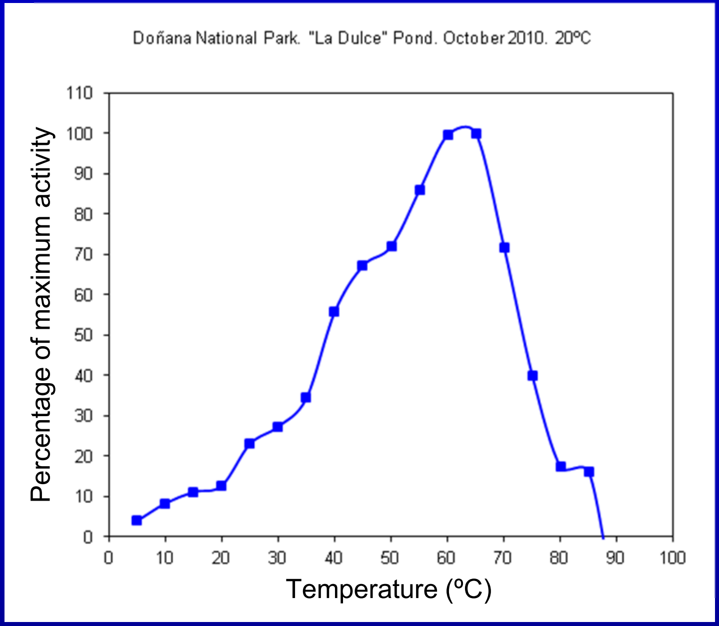


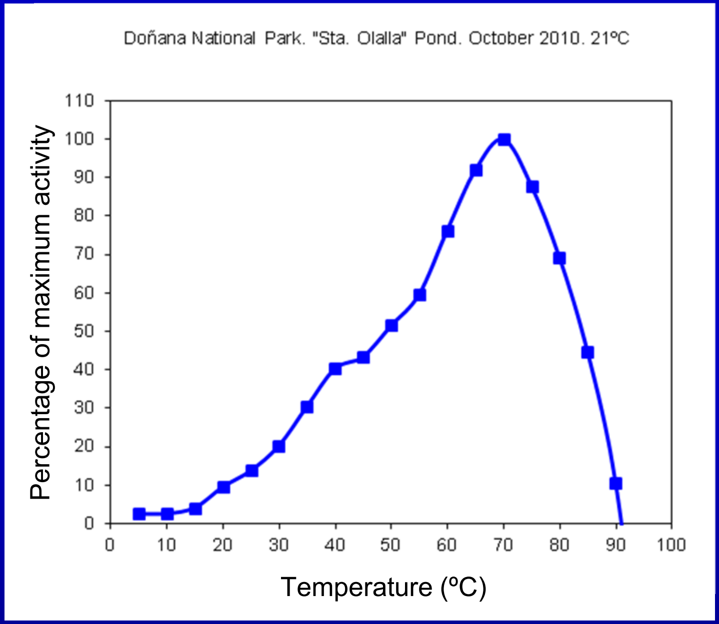
Soils and sediments can reach temperatures high enough for these thermophiles to grow

Global warming can lead to an increasing relevance of thermophiles

Thermophiles in temperate environments

Activity by extracellular enzymes in soils

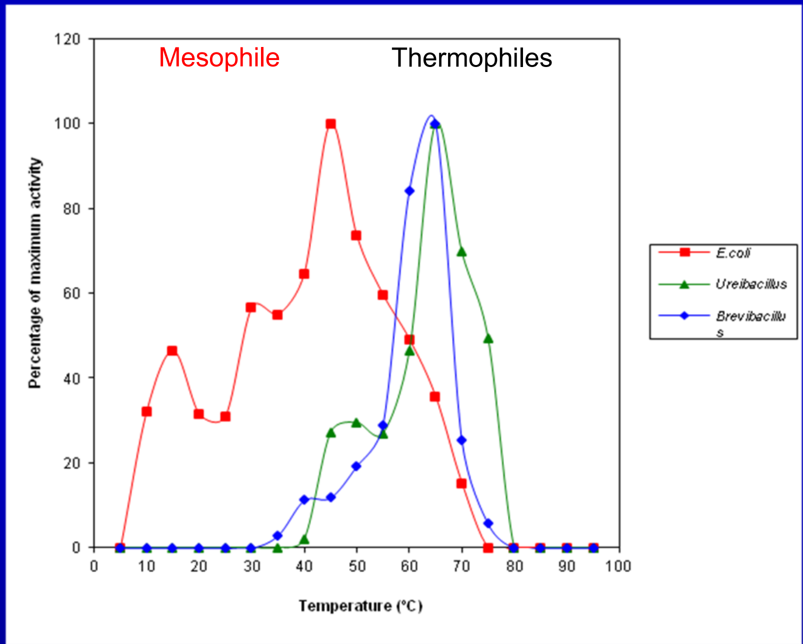




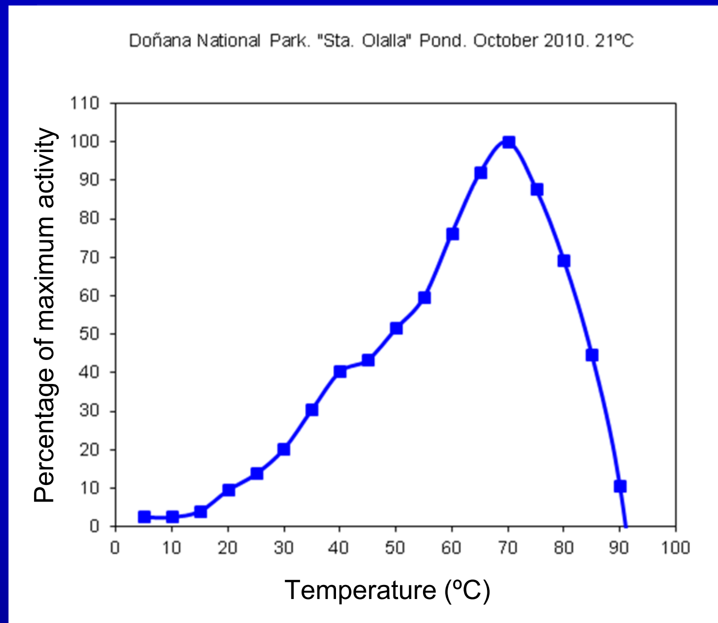
Organic matter decomposition increases at increasing temperatures in soils and sediments

Thermophiles in temperate environments

Activity by extracellular enzymes in soils



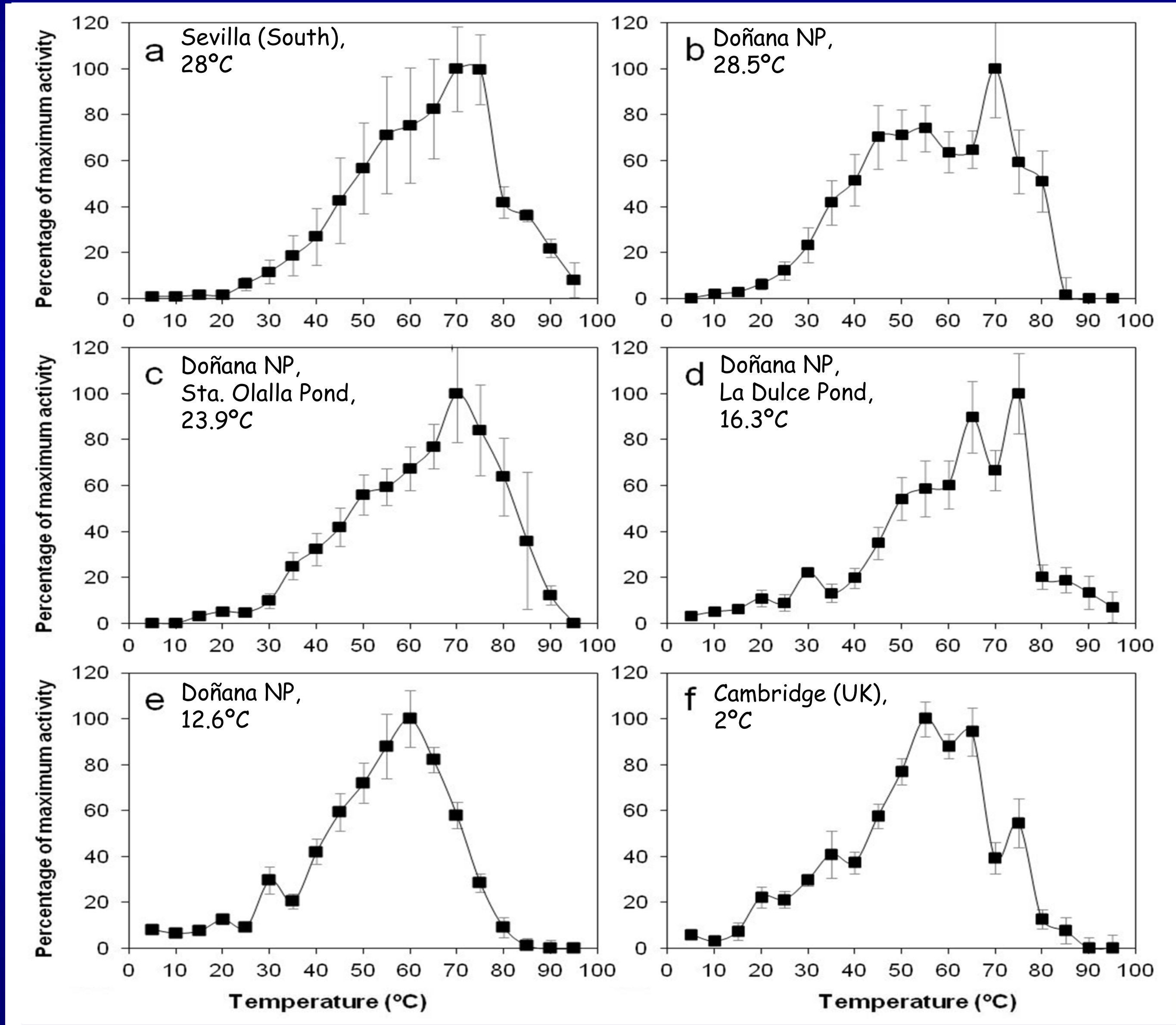
Cultures



Peaks of maximum enzyme activity above 50°C could be due to thermophiles

Thermophiles in temperate environments

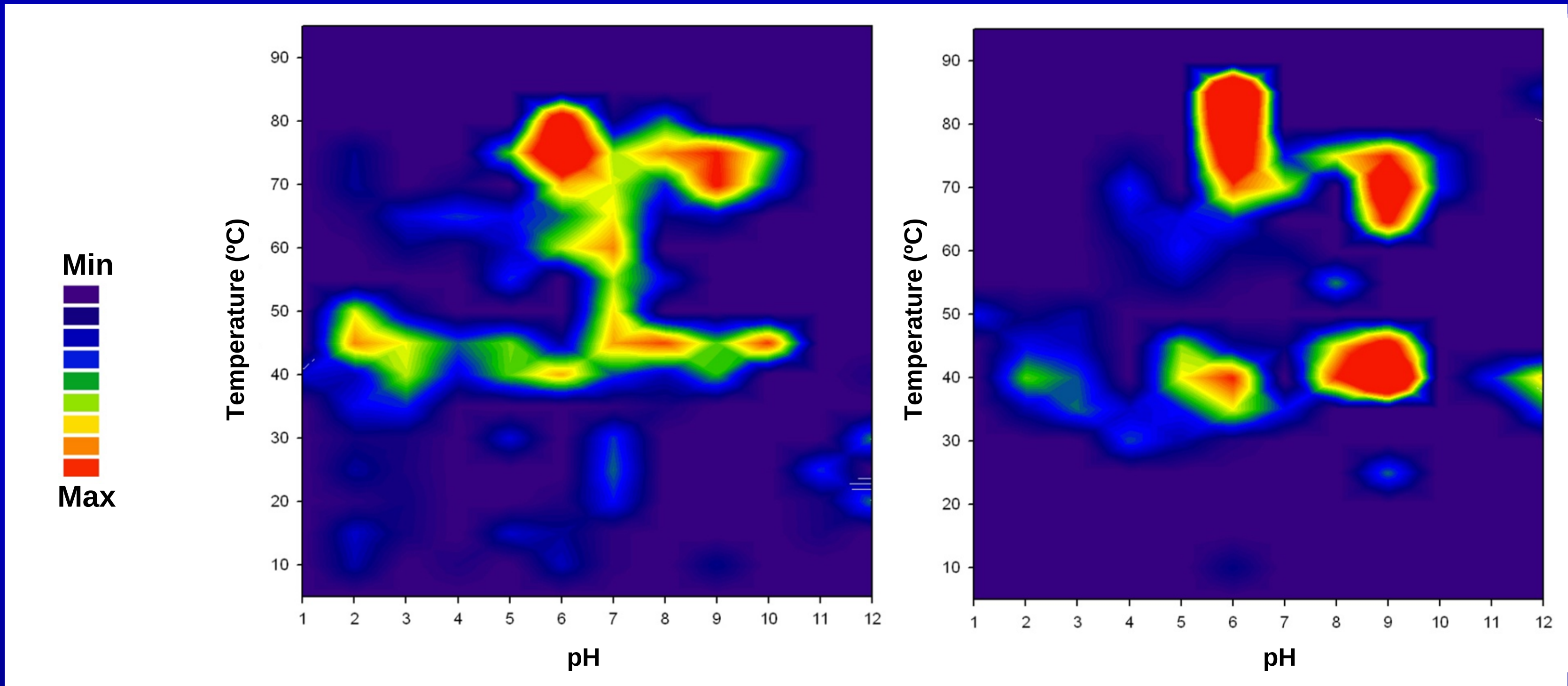
Activity of extracellular enzymes



A similar pattern was found in all soils and sediments

Thermophiles in temperate environments

Activity by extracellular enzymes depends on temperature and pH



Microbial activity on a broad range of temperature and pH suggests that soils and sediments are complex systems with great functional diversity

Acknowledgements

Collaborators:

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NILS Science and Sustainability (EEA Grants)

Spanish Network of Extremophiles (RedEx)

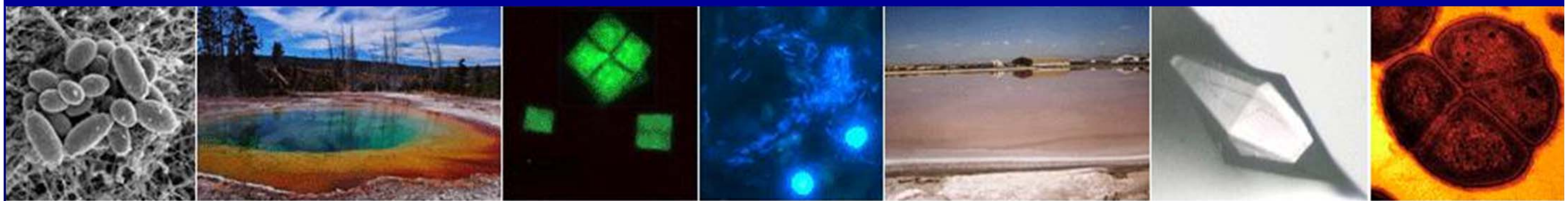
RedEx involves most Spanish researchers working on microorganisms in extreme environments

Meetings are held regularly (each 1-2 years depending on available funds)

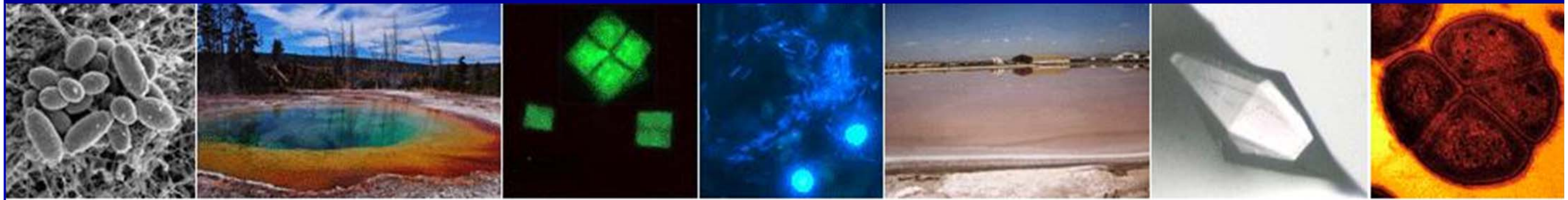
http://web.ua.es/en/rnme/ (English)

http://web.ua.es/es/rnme/

It includes 26 groups involving most techniques and extremophilic microorganisms



Spanish Network of Extremophiles (RedEx)

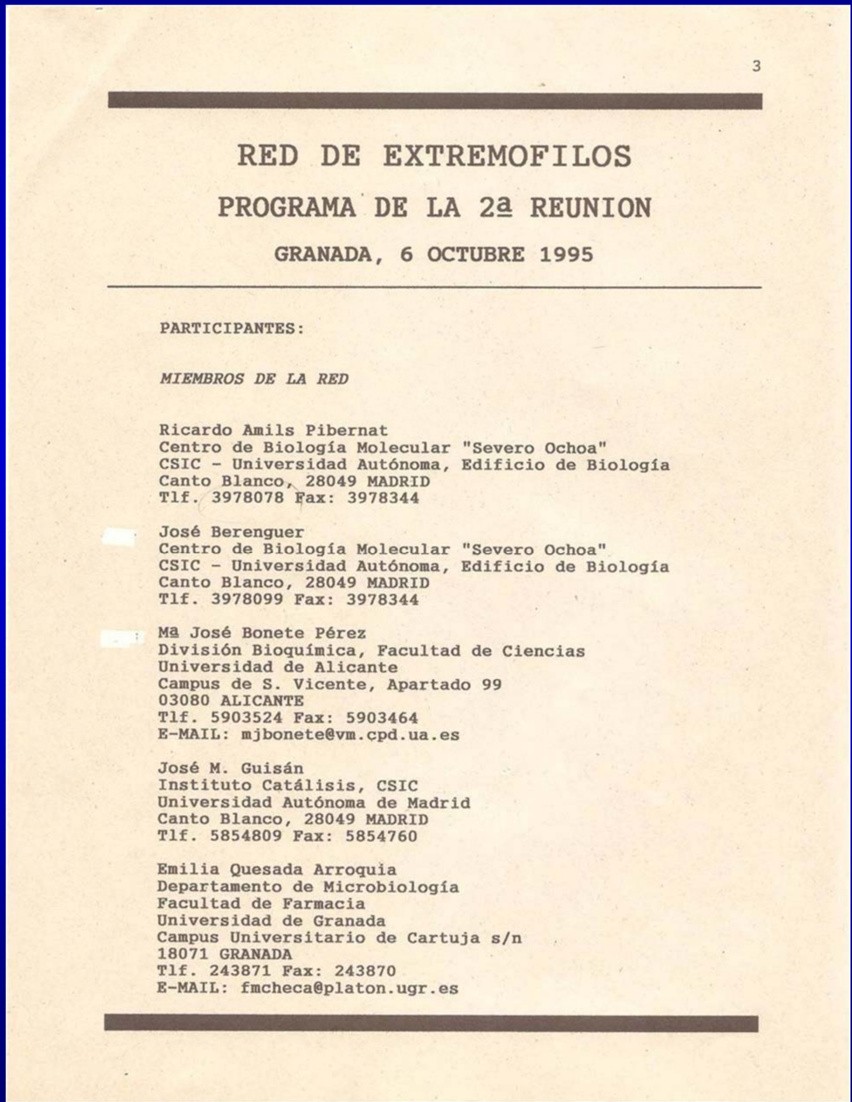


It includes 26 groups involving most techniques and extremophilic microorganisms

Acidophiles, Alkalophiles, Barophiles, Halophiles, Methanogens, Psychrophiles, Thermophiles, Xerophiles, etc.

Biochemistry, Biotechnology, Ecology, Genomics, Microbiology, Molecular Biology, Physiology, Taxonomy, Virology, etc.

Spanish Network of Extremophiles (RedEx)



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RED DE EXTREMOFILOS

PROGRAMA DE LA 2ª REUNION

GRANADA, 6 OCTUBRE 1995

PARTICIPANTES:

MIEMBROS DE LA RED

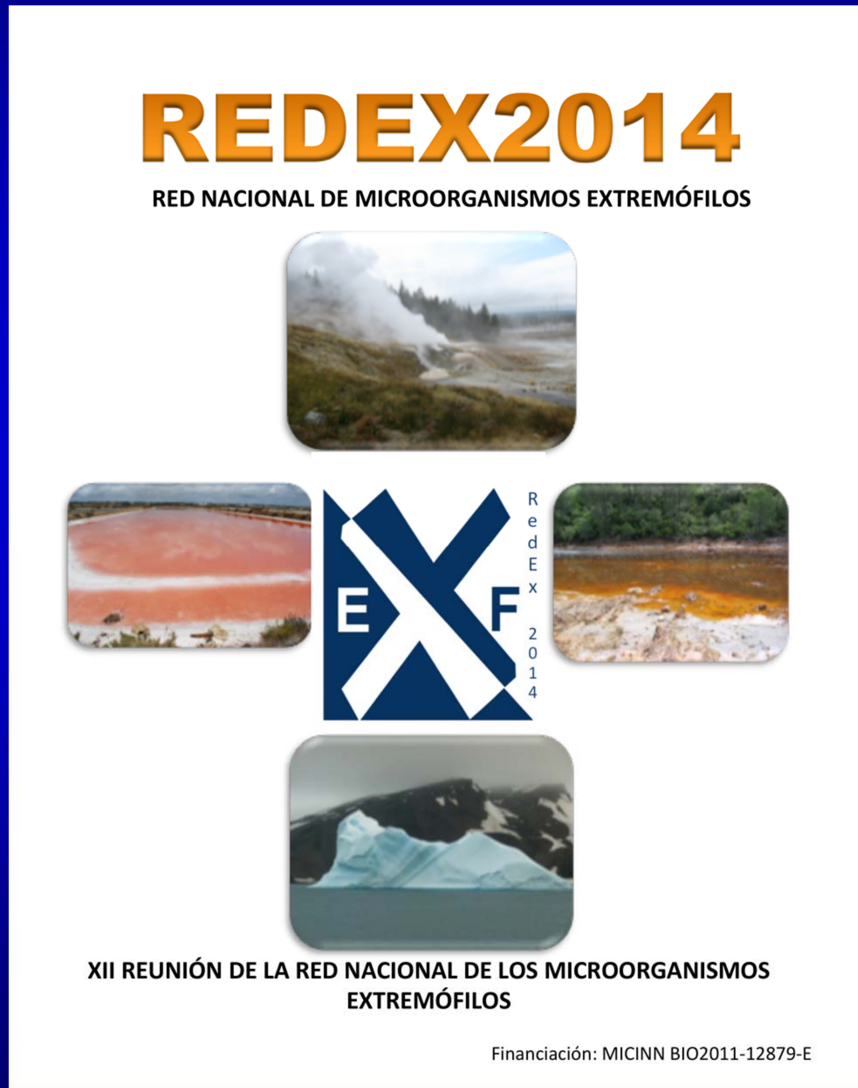
Ricardo Amils Pibernat
Centro de Biología Molecular "Severo Ochoa"
CSIC - Universidad Autónoma, Edificio de Biología
Canto Blanco, 28049 MADRID
Tlf. 3978078 Fax: 3978344

José Berenguer
Centro de Biología Molecular "Severo Ochoa"
CSIC - Universidad Autónoma, Edificio de Biología
Canto Blanco, 28049 MADRID
Tlf. 3978099 Fax: 3978344

Mª José Bonete Pérez
División Bioquímica, Facultad de Ciencias
Universidad de Alicante
Campus de S. Vicente, Apartado 99
03080 ALICANTE
Tlf. 5903524 Fax: 5903464
E-MAIL: mjbonete@vm.cpd.ua.es

José M. Guisán
Instituto Catálisis, CSIC
Universidad Autónoma de Madrid
Canto Blanco, 28049 MADRID
Tlf. 5854809 Fax: 5854760

Emilia Quesada Arroquia
Departamento de Microbiología
Facultad de Farmacia
Universidad de Granada
Campus Universitario de Cartuja s/n
18071 GRANADA
Tlf. 243871 Fax: 243870
E-MAIL: fmcheca@platon.ugr.es



Spanish Network of Extremophiles (RedEx)

XI Meeting of RedEx. 2013