

# Cold-adapted yeasts: a restricted club of extremophilic organisms

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<sup>1</sup> University of Perugia, Perugia, Italy & Industrial yeasts Collection DBVPG ([www.dbvpg.unipg.it](http://www.dbvpg.unipg.it))

<sup>2</sup> Free University of Bozen/Bolzano, Italy

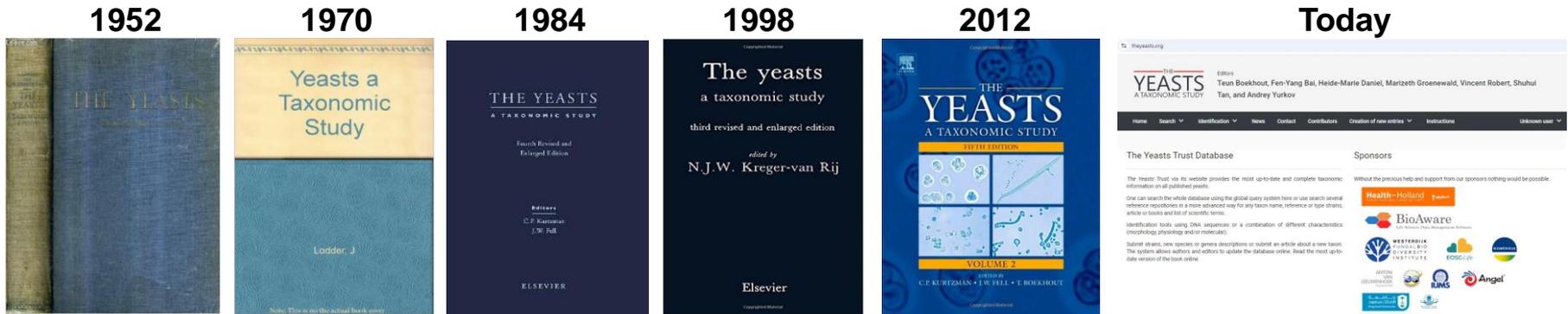


# General summary

1. Yeast diversity – the state of the art
2. Extremophilic yeasts
3. Cold-adapted yeasts
4. Cold environments worldwide
5. Culturable diversity of cold-adapted yeasts in worldwide cold environments
6. 6. Unculturable diversity (NGS) of cold-adapted fungi (including yeasts)
7. Predicting the impact of climate change on Alpine soil fungal community
8. Physiological/molecular adaptation to cold
9. A look to biotechnology of cold-adapted yeasts
10. Take home message
11. Acknowledgements
12. Sampling pictures
13. A bit of humor...



# 1. Yeast diversity – the state of the art



Fungal Diversity  
<https://doi.org/10.1007/s13225-021-00494-6>

## REVIEW

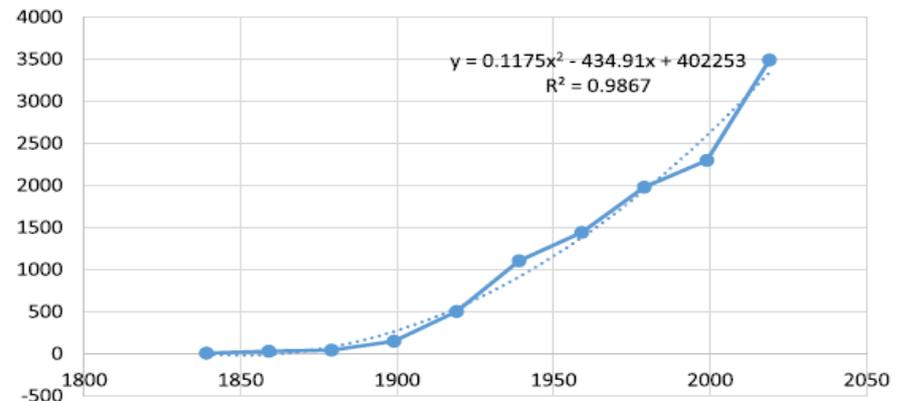


### Trends in yeast diversity discovery

Teun Boekhout<sup>1,2</sup> · Anthony S. Amend<sup>3</sup> · Fouad El Baidouri<sup>3</sup> · Toni Gabaldón<sup>4,5,6</sup> · József Geml<sup>7</sup> · Moritz Mittelbach<sup>8</sup> · Vincent Robert<sup>1</sup> · Chen Shuhui Tan<sup>9</sup> · Benedetta Turchetti<sup>10,11</sup> · Duong Vu<sup>1</sup> · Qi-Ming Wang<sup>12</sup> · Andrey Yurkov<sup>13</sup>

- ✓ Yeasts → probably **the most ancient eukaryotic organisms**
- ✓ <1% → **estimated ratio** between described & existing yeast species
- ✓ **Expected number of yeast species** on Earth → around **150,000**

Number of species described cumulative



# 2. Extremophilic yeasts (A)

Received: 18 December 2017 | Revised: 31 January 2018 | Accepted: 6 March 2018

DOI: 10.1002/yea.3314

**ECOYEAST REVIEW**

WILEY **Yeast**

## Extremophilic yeasts: the toughest yeasts around?

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<sup>2</sup>Leibniz Institute DSMZ – German Collection of Microorganisms and Cell Cultures, Braunschweig, Germany

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Pietro Buzzini, Department of Agricultural, Food and Environmental Sciences, University of Perugia, Italy.

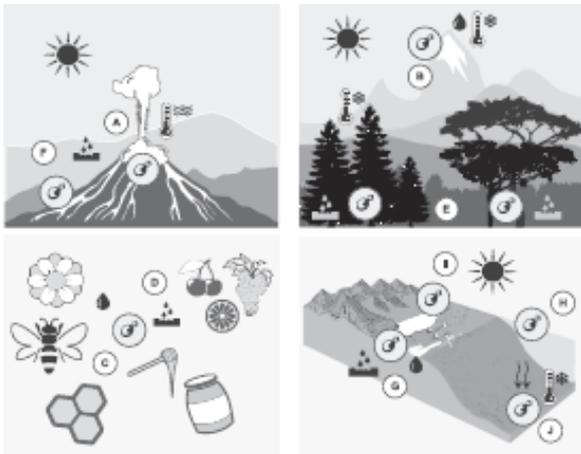
Email: [pietro.buzzini@unipg.it](mailto:pietro.buzzini@unipg.it)

**Abstract**

Microorganisms are widely distributed in a multitude of environments including ecosystems that show challenging features to most life forms. The combination of extreme physical and chemical factors contributes to the definition of extreme habitats although the definition of extreme environments changes depending on one's point of view: anthropocentric, microbial-centric or zymo-centric. Microorganisms that live under conditions that cause hard survival are called extremophiles. In particular organisms that require extreme conditions are called true extremophiles while organisms that tolerate them to some extent are termed extremotolerant. Deviation of temperature, pH, osmotic stress, pressure and radiation from the common range delineates extreme environments. Yeasts are versatile eukaryotic organisms that are not frequently considered the toughest microorganisms in comparison with prokaryotes. Nevertheless extremophilic or extremotolerant species are present also within this group. Here a brief description is provided of the main extreme habitats and the metabolic and physiological modifications adopted by yeasts depending on their adverse conditions. Additionally the main extremophilic and extremotolerant yeast species associated with a few extreme habitats are listed.

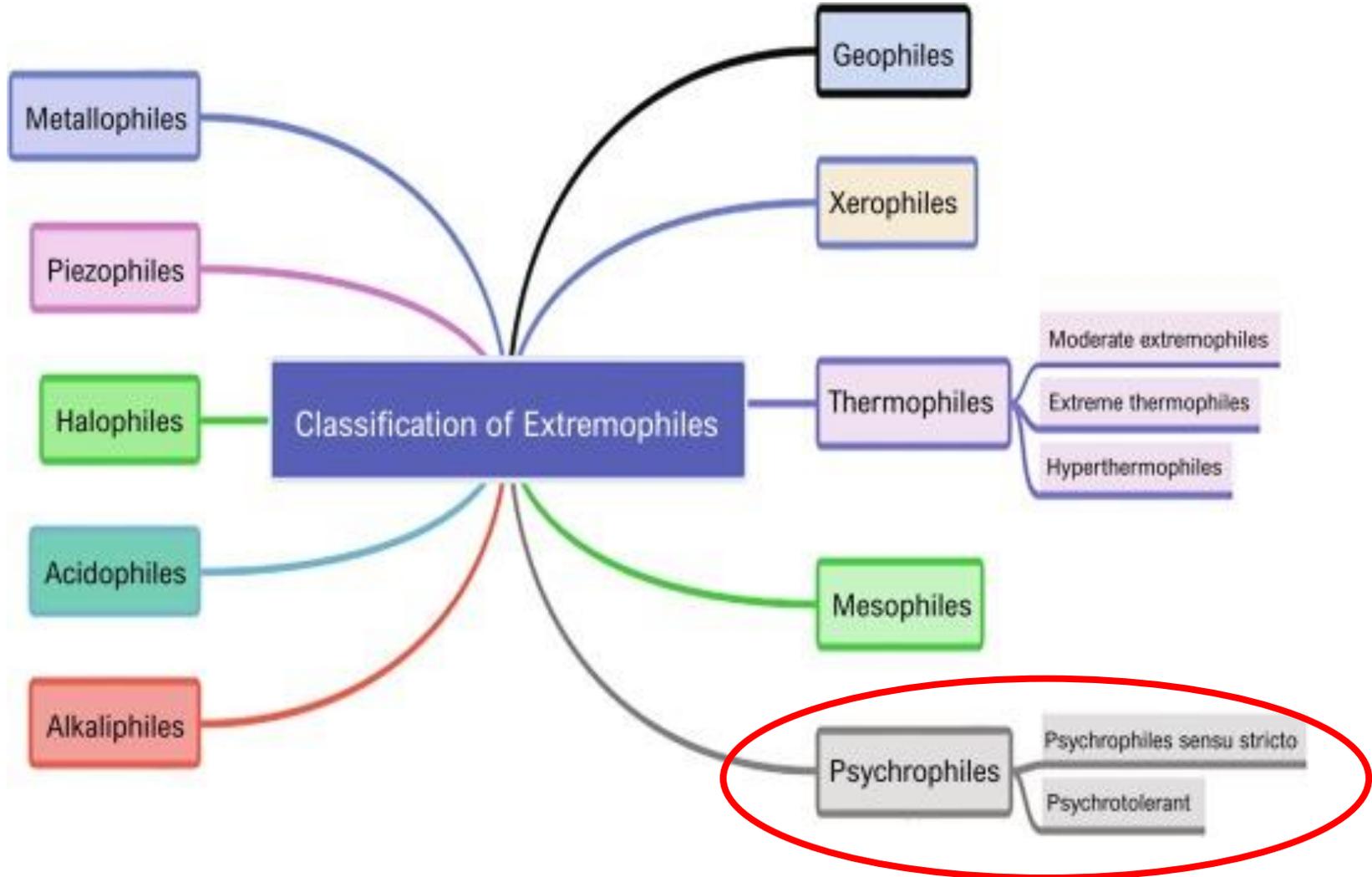
**KEYWORDS**

extremophilic yeasts, extremotolerant yeasts, extreme environments



## 2. Extremophilic yeasts (B)

### 2.1 Some basic definitions



# 3. Cold-adapted yeasts (A)

## 3.1 Some basic definitions

### Obligate (true or *sensu stricto*) psychrophiles

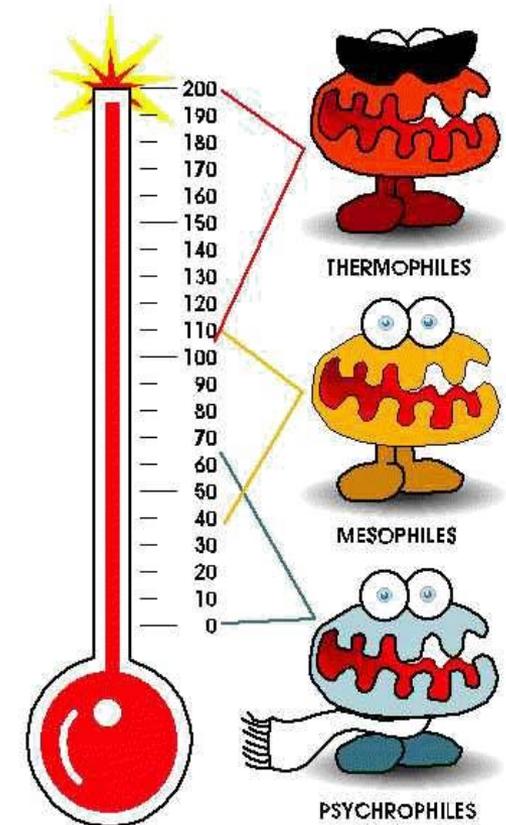


- ◆ optimal growth temperature  $\approx 15^{\circ}\text{C}$  (or below)
- ◆ minimum growth temperature  $\approx 0^{\circ}\text{C}$  (or below)
- ◆ maximum growth temperature  $< 20^{\circ}\text{C}$

### Facultative psychrophiles (psychrotolerant or psychrotrophic)



- ✗ optimal growth temperature  $\approx 25^{\circ}\text{C}$  (or higher)
- ✗ minimum growth temperature  $\approx 0^{\circ}\text{C}$
- ✗ maximum growth temperature  $\approx 30^{\circ}\text{C}$  (or higher)



# 3. Cold-adapted yeasts (B)

## 3.2 Current literature



ELSEVIER

FEMS Microbiology Ecology 53 (2005) 117–128



www.fems-microbiology.org

### Biodiversity of cryopegs in permafrost

David Gilichinsky <sup>a,\*</sup>, Elizaveta Rivkina <sup>a</sup>, Corien Bakermans <sup>b</sup>, Viktoria Shcherbakova <sup>c</sup>,  
Lada Petrovskaya <sup>d</sup>, Svetlana Ozerskaya <sup>c</sup>, Natalia Ivanushkina <sup>c</sup>, Galina Kochkina <sup>c</sup>,  
Kyastus Laurinavichuis <sup>c</sup>, Svetlana Pecheritsina <sup>c</sup>, Rushania Fattakhova <sup>a</sup>,  
James M. Tiedje <sup>b</sup>



FEMS Microbiology Ecology, 92, 2016, fw018

doi: 10.1093/femsec/fw018

Advance Access Publication Date: 31 January 2016  
Research Article

RESEARCH ARTICLE

### Microbial diversity in European alpine permafrost and active layers

Beat Frey<sup>1</sup>, Thomas Rime<sup>1</sup>, Marcia Phillips<sup>2</sup>, Beat Stierli<sup>1</sup>, Irka Hajdas<sup>3</sup>,  
Franco Widmer<sup>4</sup> and Martin Hartmann<sup>1,\*</sup>

Environmental Microbiology (2009) 11(2), 360–381

doi:10.1111/j.1462-2920.2008.01777.x

### Microbial eukaryotes in the hypersaline anoxic L'Atalante deep-sea basin

Eva Alexander,<sup>1</sup> Alexandra Stock,<sup>1</sup>  
Hans-Werner Breiner,<sup>1</sup> Anke Behnke,<sup>1</sup> John Bunge,<sup>2</sup>  
Michail M. Yakimov<sup>2</sup> and Thorsten Stoeck<sup>1\*</sup>  
<sup>1</sup>University of Kaiserslautern, School of Biology,  
Erwin-Schrodinger-Strasse 14, D-67773,  
Kaiserslautern, Germany.

ing in the evolution of an exceptional and distinctive assemblage of protists. The deep hypersaline anoxic basins in the Mediterranean Sea provide an ideal platform to test for this hypothesis and are promising targets for the discovery of undescribed protists with unknown physiological capabilities.

### Growth kinetics of microorganisms isolated from Alaskan soil and permafrost in solid media frozen down to – 35 °C

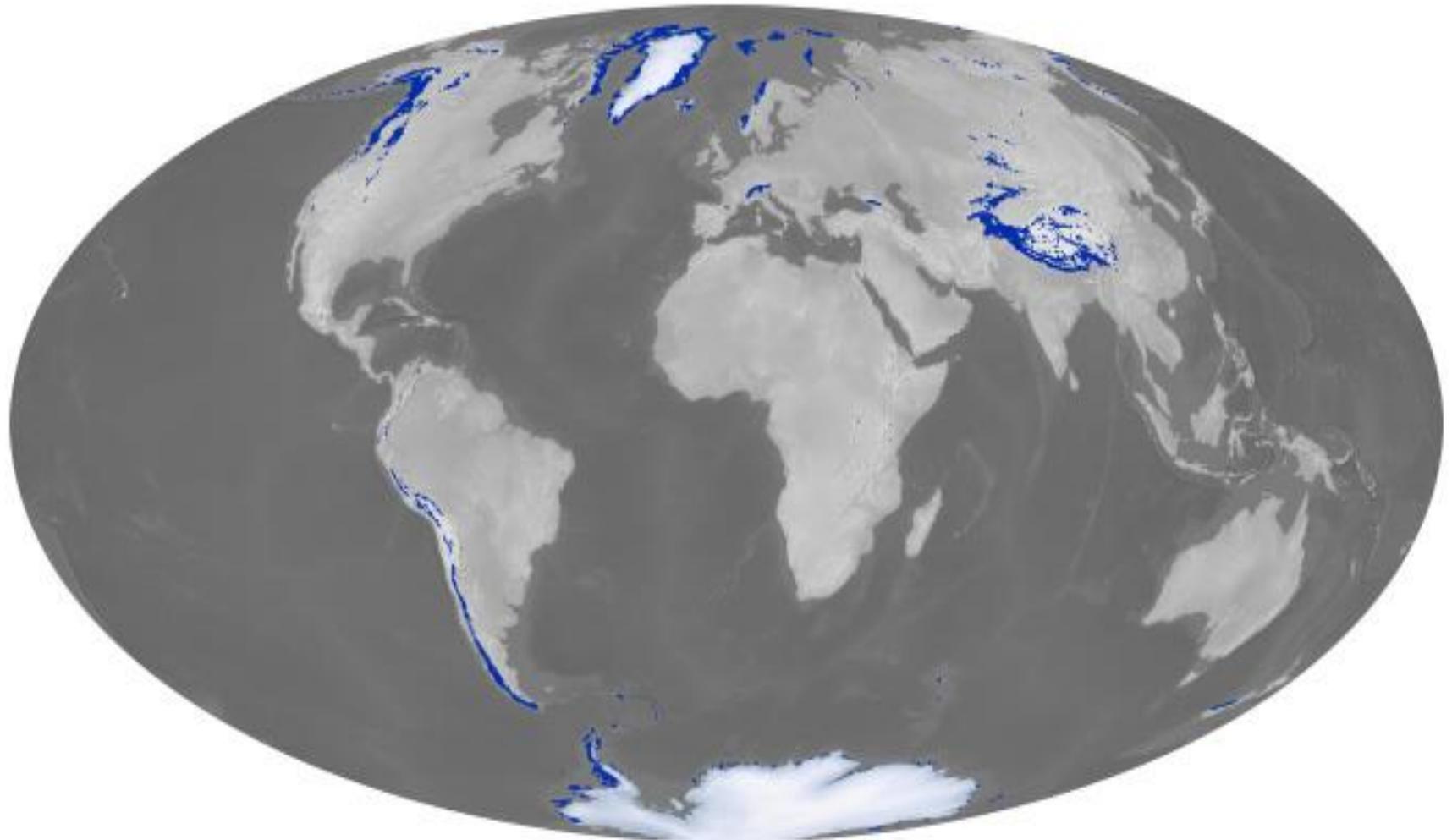
Nicolai S. Panikov & Maria V. Sizova

Department of Chemistry & Chemical Biology, Stevens Institute of Technology, NJ, USA



## 4. Cold environments worldwide (A)

### 4.1 The Randolph Glacier Inventory



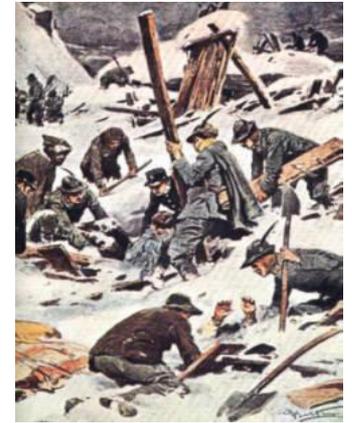
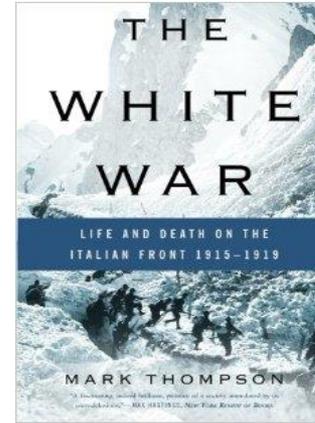
## 4. Cold environments worldwide (B)

### 4.2 Impact of ongoing climate change: “Only 17 years to try to reverse the retreating trend” (IPCC, 2014)



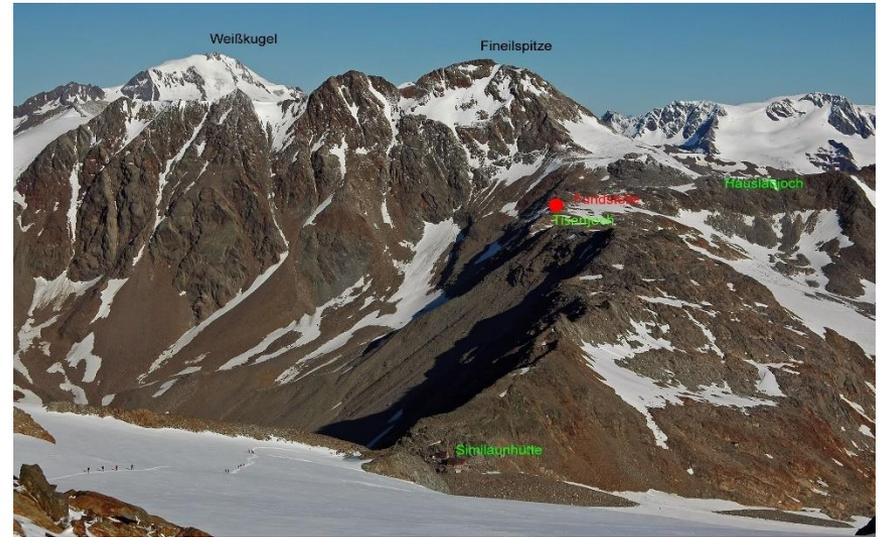
## 4. Cold environments worldwide (C)

### 4.3 A few “collateral” effects of Alpine glacier retreat (1)



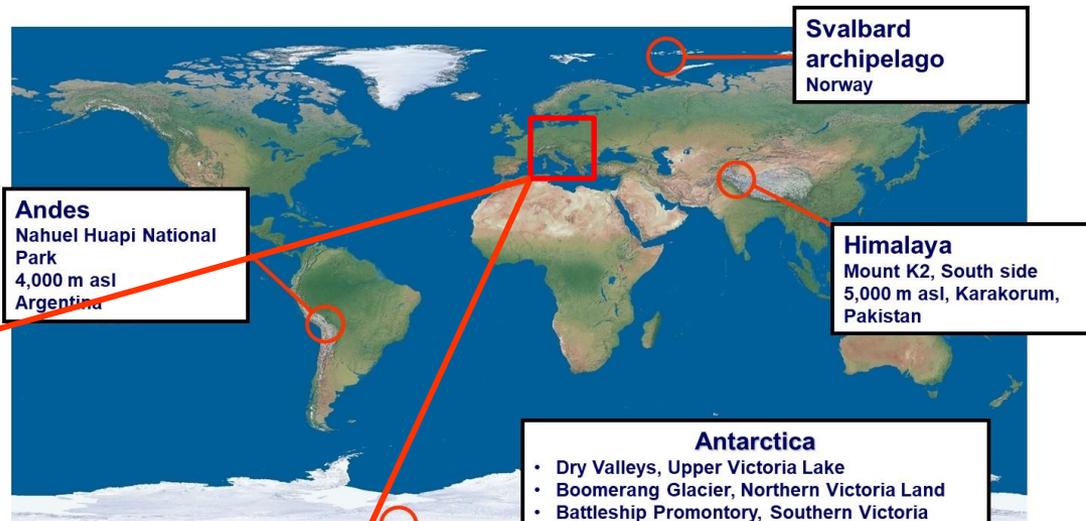
## 4. Cold environments worldwide (D)

### 4.4 A few “collateral” effects of Alpine glacier retreat (2)



# 5. Culturable diversity of cold-adapted yeasts in worldwide cold environments (A)

## 5.1 Sampling sites (2002-2020)



**Svalbard archipelago**  
Norway

**Andes**  
Nahuel Huapi National Park  
4,000 m asl  
Argentina

**Himalaya**  
Mount K2, South side  
5,000 m asl, Karakorum,  
Pakistan

**Antarctica**

- Dry Valleys, Upper Victoria Lake
- Boomerang Glacier, Northern Victoria Land
- Battleship Promontory, Southern Victoria Land, McMurdo Dry Valleys
- Lichen Hills, North Victoria Land
- Morris Basin, North Victoria Land
- Vegetation Island, North Victoria Land

**Western Alps**  
Mount Blanc area  
About 3,600 m a.s.l.

**Central Alps**  
Ortles-Cevedale area  
About 3,400 m a.s.l.

**Apennines**  
Gran Sasso area  
About 3,100 m a.s.l.



# 5. Culturable diversity of cold-adapted yeasts in worldwide cold environments (B)

## 5.2 Samples

Snow



Melt waters



Ice cores



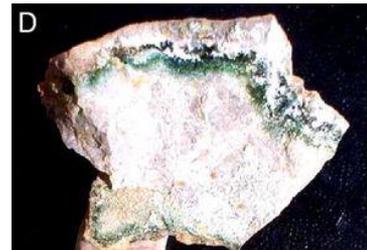
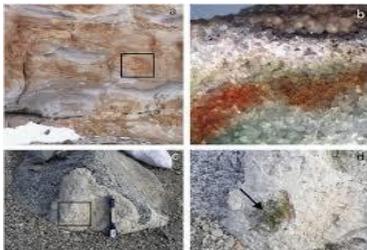
Supra- & sub-glacial debris (1-2°C *in situ*)



Permafrost, ice cores & brines



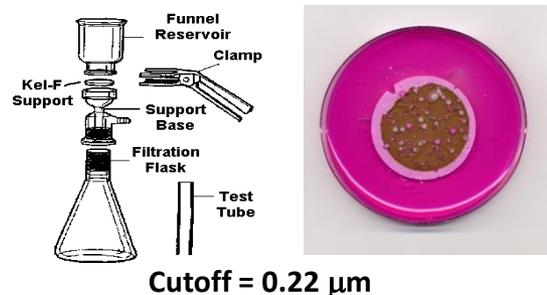
Rocks



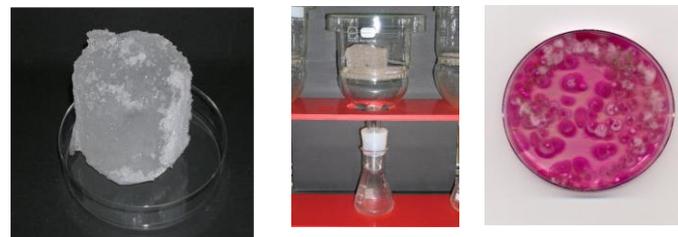
# 5. Culturable diversity of cold-adapted yeasts in worldwide cold environments (C)

## 5.3 Isolation protocols (incubation at 4°C and 20°C for 12 and 4 weeks)

Melt waters, snow (after melting under asepsis) and brines



Lab surface decontamination protocols for ice and permafrost



- Surface washing (20'') with 5%) NaClO
- Series of surface washings with sterile water
- Melting of cores into a sterile funnel and discharge of the first melt aliquots
- Harvesting and streaking of subsequent aliquots of melt cores on Petri dishes
- Incubation: 4°C and 20°C for 12 and 4 weeks

Lab surface decontamination protocols for rocks



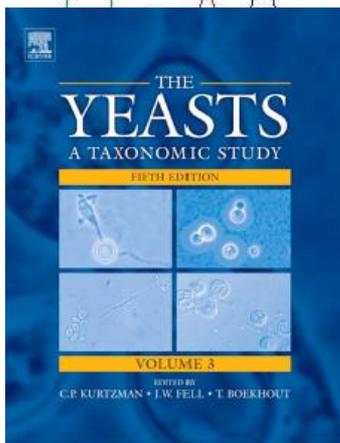
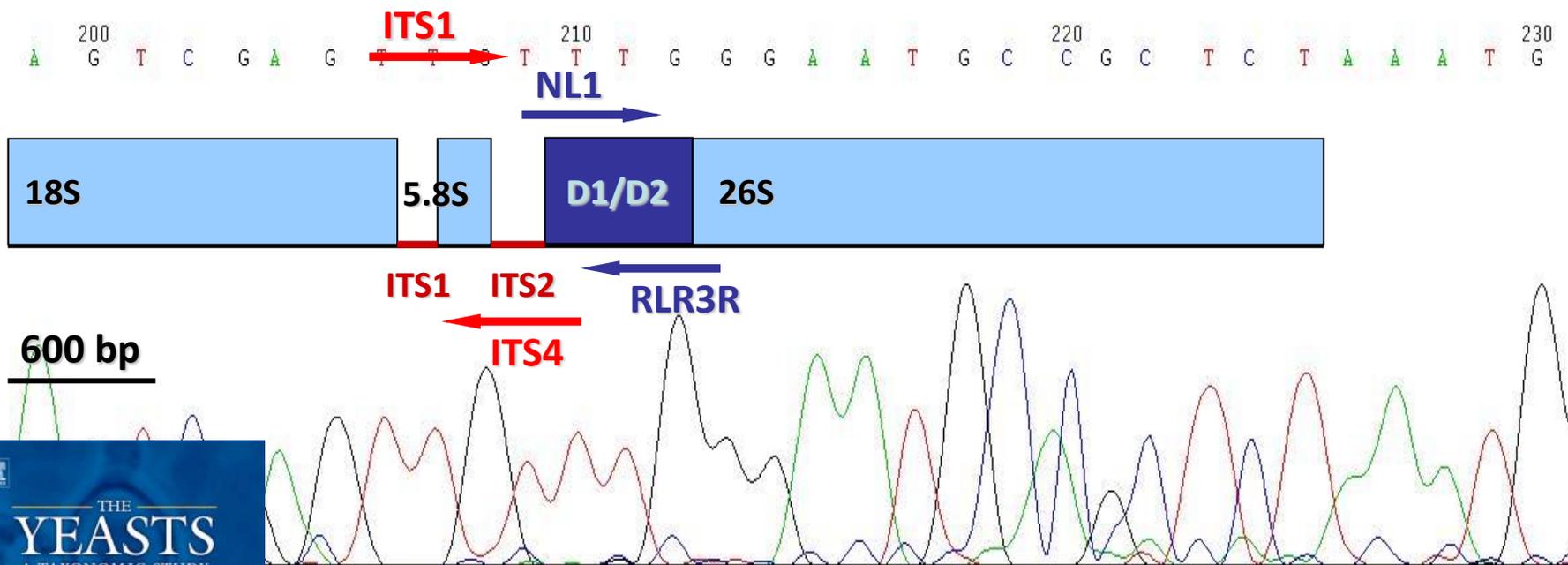
- Surface washing (20'') with 5%) NaClO
- Series of surface washings with sterile water
- Rock crushing and grinding in pestle (under aseptic conditions)
- Preparation of a water suspension 1:10 (v/v) and streaking on Petri dishes

Debris (aseptically collected)



# 5. Culturable diversity of cold-adapted yeasts in worldwide cold environments (D)

## 5.4 Identification by sequencing of D1/D2 of 26S rRNA gene and ITS (1&2)



available online at [www.studiesinmycology.org](http://www.studiesinmycology.org) STUDIES IN MYCOLOGY 81: 85–147.

**Towards an integrated phylogenetic classification of the Tremellomycetes**

X.-Z. Liu<sup>1,2</sup>, Q.-M. Wang<sup>1,2</sup>, M. Göker<sup>3</sup>, M. Groenewald<sup>4</sup>, A.V. Kachalkin<sup>5</sup>, H.T. Lumbsch<sup>6</sup>, A.M. Milanés<sup>7</sup>, M. Wedin<sup>8</sup>, A.M. Yurkov<sup>9</sup>, T. Boekhout<sup>1,2,3\*</sup>, and F.-Y. Bai<sup>1,2</sup>

<sup>1</sup>State Key Laboratory for Mycology, Institute of Microbiology, Chinese Academy of Sciences, Beijing 100101, PR China; <sup>2</sup>CBS Fungal Biodiversity Centre (CBS-KNAW), Uppsalaalan 8, Uppsala, The Netherlands; <sup>3</sup>Leibniz Institute DSMZ-German Collection of Microorganisms and Cell Cultures, Braunschweig 38124, Germany; <sup>4</sup>Faculty of Soil Science, Lomonosov Moscow State University, Moscow 119891, Russia; <sup>5</sup>Science & Education, The Field Museum, 1400 S. Lake Shore Drive, Chicago, IL 60605, USA; <sup>6</sup>Departamento de Biología y Geología, Física y Química Inorgánica, Universidad Rey Juan Carlos, E-28933 Móstoles, Spain; <sup>7</sup>Department of Botany, Swedish Museum of Natural History, P.O. Box 50007, SE-10405 Stockholm, Sweden; <sup>8</sup>Shanghai Key Laboratory of Molecular Medical Mycology, Changzheng Hospital, Second Military Medical University, Shanghai, PR China

\*Correspondence: F.-Y. Bai, [bai@im.ac.cn](mailto:bai@im.ac.cn); T. Boekhout, [tboekhout@cbi.knaw.nl](mailto:tboekhout@cbi.knaw.nl)

available online at [www.studiesinmycology.org](http://www.studiesinmycology.org) STUDIES IN MYCOLOGY 81: 149–189.

**Phylogenetic classification of yeasts and related taxa within Pucciniomycotina**

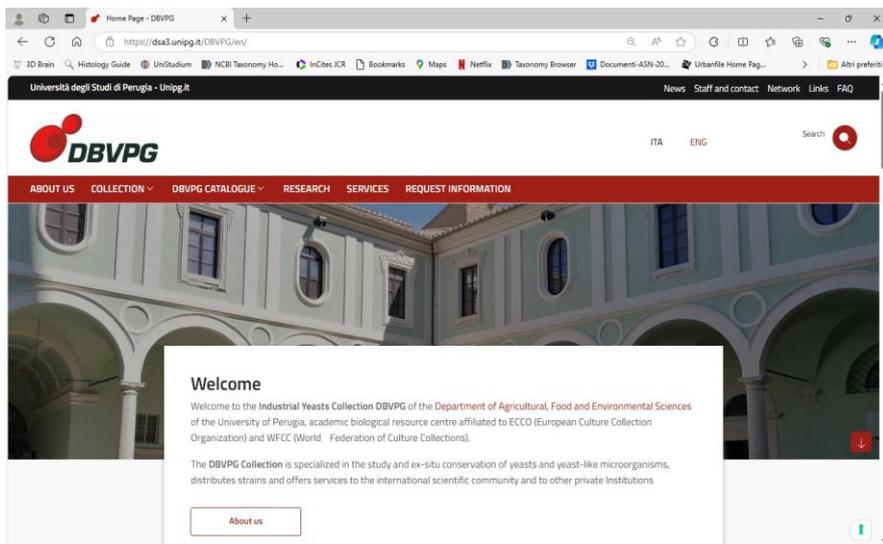
Q.-M. Wang<sup>1</sup>, A.M. Yurkov<sup>2</sup>, M. Göker<sup>3</sup>, H.T. Lumbsch<sup>4</sup>, S.D. Leavitt<sup>5</sup>, M. Groenewald<sup>6</sup>, B. Theelen<sup>7</sup>, X.-Z. Liu<sup>1</sup>, T. Boekhout<sup>1,4,5,6\*</sup>, and F.-Y. Bai<sup>1,4\*</sup>

<sup>1</sup>State Key Laboratory of Mycology, Institute of Microbiology, Chinese Academy of Sciences, Beijing 100101, China; <sup>2</sup>Leibniz Institute DSMZ – German Collection of Microorganisms and Cell Cultures, Braunschweig, Germany; <sup>3</sup>Science & Education, The Field Museum, 1400 S. Lake Shore Drive, Chicago, IL 60605, USA; <sup>4</sup>CBS Fungal Biodiversity Centre (CBS-KNAW), Uppsalaalan 8, 3504 CT Utrecht, The Netherlands; <sup>5</sup>Shanghai Key Laboratory of Molecular Medical Mycology, Changzheng Hospital, Second Military Medical University, Shanghai, China

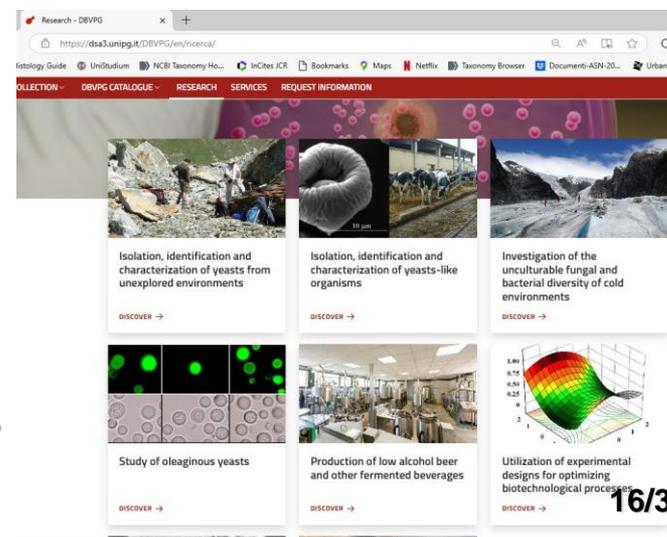
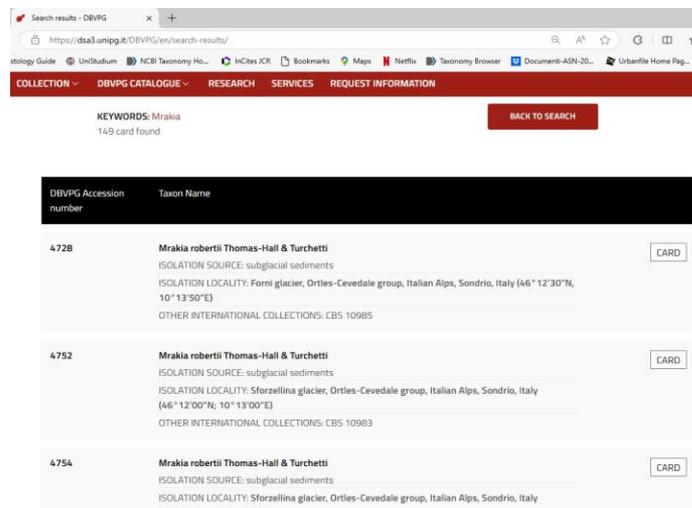
\*Correspondence: T. Boekhout, [tboekhout@cbi.knaw.nl](mailto:tboekhout@cbi.knaw.nl); F.-Y. Bai, [bai@im.ac.cn](mailto:bai@im.ac.cn)

# 5. Culturability diversity of cold-adapted yeasts in worldwide cold environments (E)

## 5.5 Ex-situ conservation of yeast isolates in the Industrial Yeasts Collection DBVPG



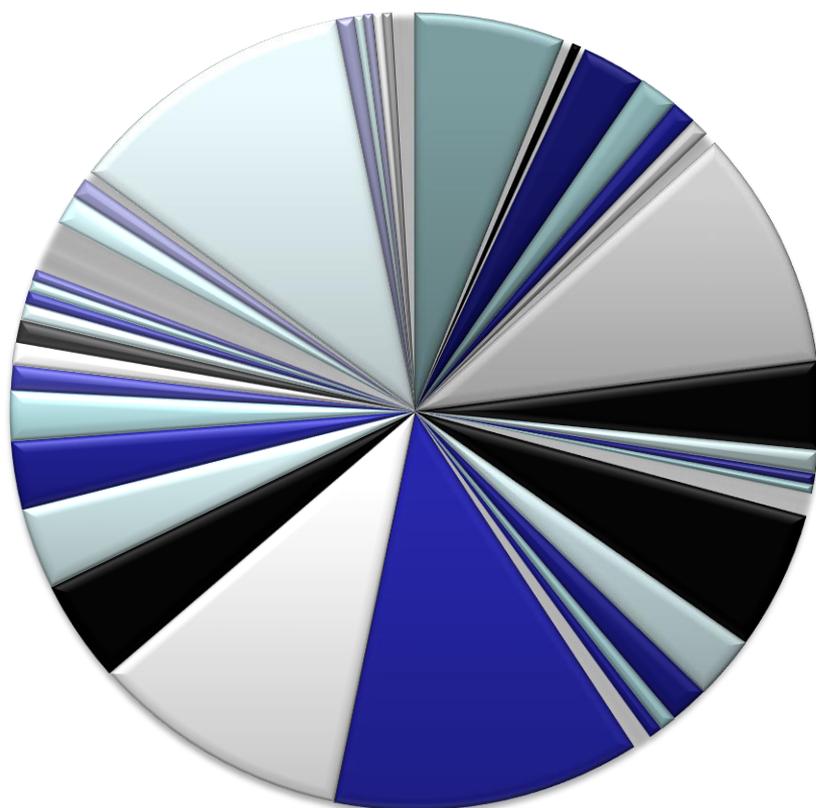
[www.dbvpg.unipg.it](http://www.dbvpg.unipg.it)



# 5. Culturable diversity of cold-adapted yeasts in worldwide cold environments (F)

## 5.5 Results: culturable yeast diversity found

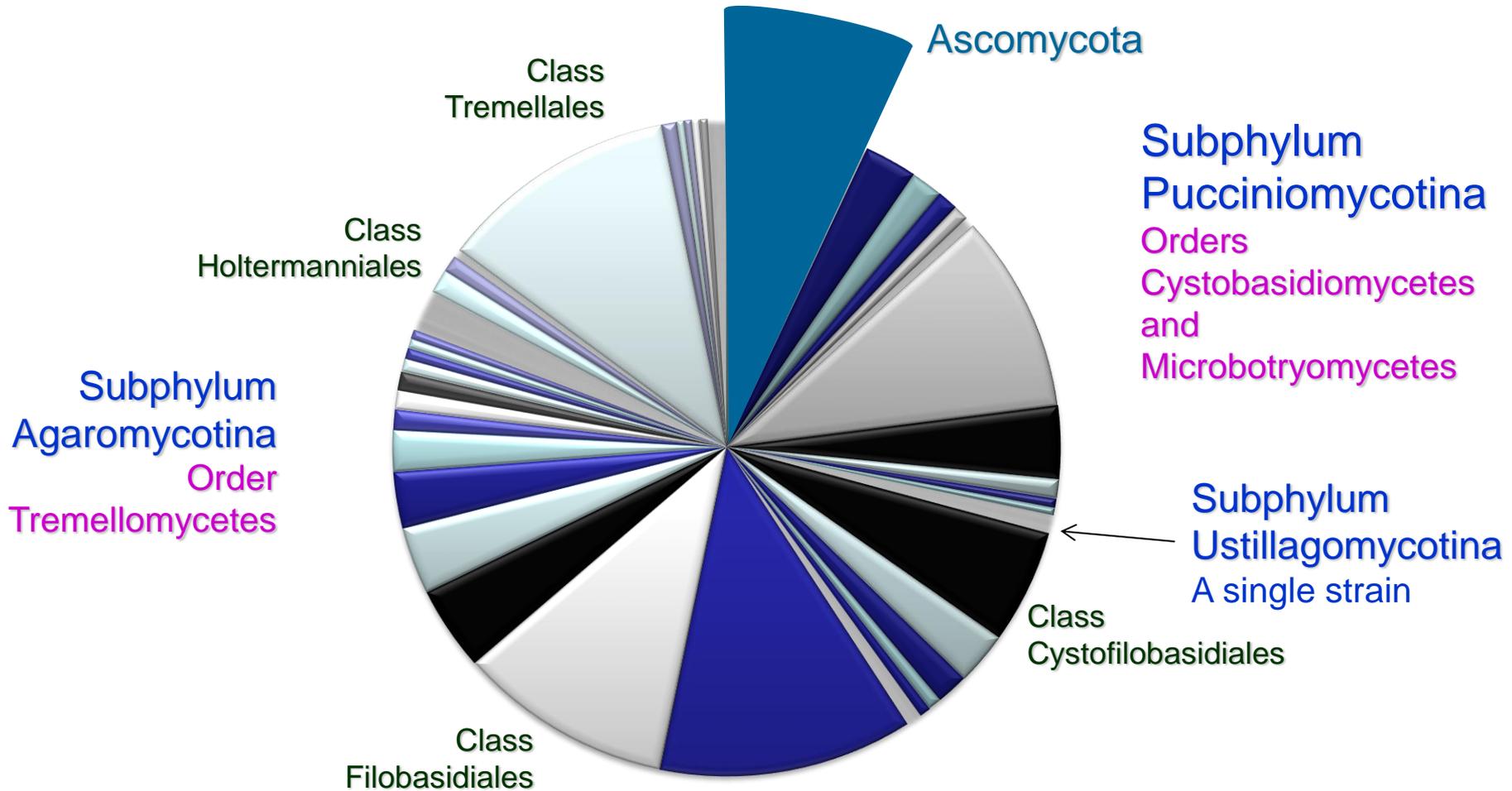
1,160 strains belonging to 78 species



- Aureobasidium pullulans
- Aureobasidium sp.
- Exophiala dermatitidis
- Candida santamariae
- Candida sp.
- Cystofilobasidium macerans
- Cystobasidium laryngis
- Cystofilobasidium capitatum
- Cystobasidium cf. laryngis
- Cystobasidium sp.
- Cystobasidium sp.
- Cystofilobasidium infirmominiatum
- Buckleyzyma aurantiaca
- Erythrobasidium hasegawianum
- Phenoliferia psychropholica
- Phenoliferia glacialis
- Sporobolomyces roseus
- Ustilentyloma graminis
- Glaciozyma watsonii
- Leucosporidium creatinivorum
- Rhodosporidiobolus colostri
- Glaciozyma martinii
- Leucosporidium intermedium
- Leucosporidium sp.
- Phenoliferia sp.
- Sporobolomyces metaroseus
- Rhodotorula bacarum
- Mrakia robertii
- Mrakia gelida
- Tausonia pullulans
- Mrakia aquatica
- Mrakia cryoconiti
- Mrakia cf. gelida
- Mrakia psychrophila
- Itersonilia pannonica
- Mrakia blollopis
- Mrakia niccombsii

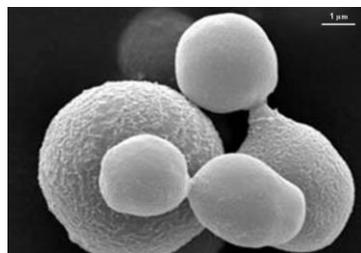
# 5. Culturable diversity of cold-adapted yeasts in worldwide cold environments (F)

## 5.5 Results: Ascomycota vs Basidiomycota

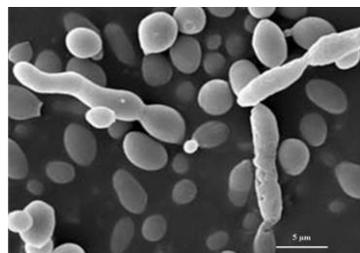


# 5. Culturable diversity of cold-adapted yeasts in worldwide cold environments (G)

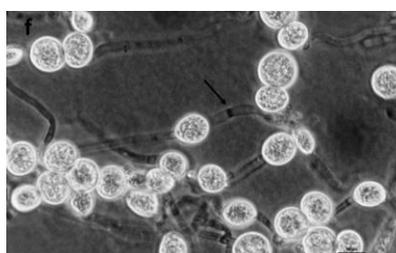
## 5.6 Some new genera and species recently described by DBVPG (1)



*Glaciozyma martinii*



*Glaciozyma watsonii*



*Mrakia stelviica*

Extremophiles (2011) 15:573–586  
DOI 10.1007/s00792-011-0388-x

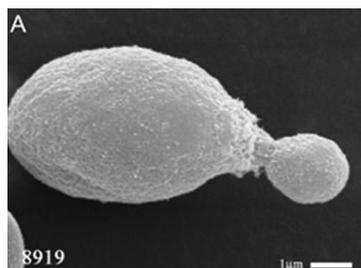
ORIGINAL PAPER

Psychrophilic yeasts from Antarctica and European glaciers: description of *Glaciozyma* gen. nov., *Glaciozyma martinii* sp. nov. and *Glaciozyma watsonii* sp. nov.

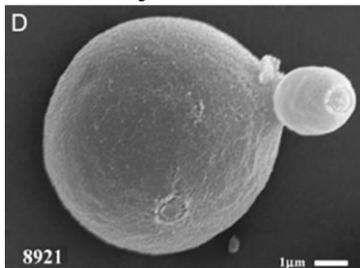
Benedetta Turchetti · Skye R. Thomas Hall · Laurie B. Connell · Eva Branda · Pietro Buzzini · Bart Theelen · Wally H. Müller · Teun Boekhout

Extremophiles (2010) 14:47–59  
DOI 10.1007/s00792-009-0286-7

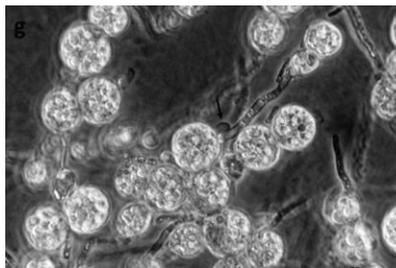
ORIGINAL PAPER



*Mrakia robertii*



*Mrakia blollopis*



*Mrakia montana*

Cold-adapted yeasts from Antarctica and the Italian Alps—description of three novel species: *Mrakia robertii* sp. nov., *Mrakia blollopis* sp. nov. and *Mrakiella niccombsii* sp. nov.

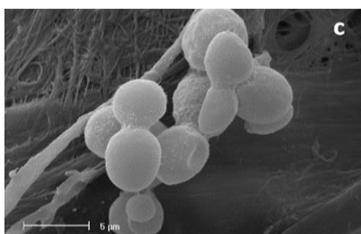
Skye Robin Thomas-Hall · Benedetta Turchetti · Pietro Buzzini · Eva Branda · Teun Boekhout · Bart Theelen · Kenneth Watson

Extremophiles (2015) 19:149–159  
DOI 10.1007/s00792-014-0692-3

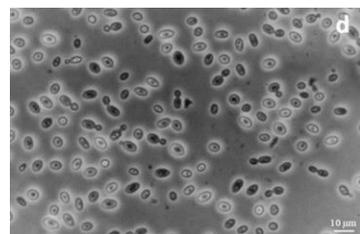
ORIGINAL PAPER

*Cryptococcus vaughanmartinae* sp. nov. and *Cryptococcus onofrii* sp. nov.: two new species isolated from worldwide cold environments

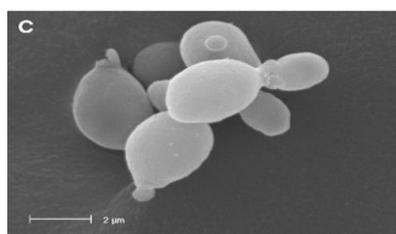
Benedetta Turchetti · Laura Selbmann · Robert A. Blanchette · Simone Di Mauro · Elisabetta Marchegiani · Laura Zucconi · Brett E. Arendz · Pietro Buzzini



*Naganishia vaughanmartinae*



*Naganishia onofrii*



*Taphrina antarctica*

INTERNATIONAL JOURNAL OF SYSTEMATIC AND EVOLUTIONARY MICROBIOLOGY

TAXONOMIC DESCRIPTION  
Turchetti et al., Int. J. Syst. Evol. Microbiol. 2020, 70, 4704–4713  
DOI 10.1099/ijsem.0.004336



*Mrakia stelviica* sp. nov. and *Mrakia montana* sp. nov., two novel basidiomycetous yeast species isolated from cold environments

Benedetta Turchetti<sup>1</sup>\*, Ciro Sannino<sup>1</sup>, Ambra Mezzasoma<sup>1</sup>, Laura Zucconi<sup>1</sup>, Silvano Onofri<sup>2</sup> and Pietro Buzzini<sup>1</sup>

Extremophiles (2014) 18:707–721  
DOI 10.1007/s00792-014-0651-z

ORIGINAL PAPER

Description of *Taphrina antarctica* f.a. sp. nov., a new anamorphic ascomycetous yeast species associated with Antarctic endolithic microbial communities and transfer of four *Lalaria* species in the genus *Taphrina*

Laura Selbmann · Benedetta Turchetti · Andrey Yurkov · Clarissa Cecchini · Laura Zucconi · Daniela Isola · Pietro Buzzini · Silvano Onofri

# 5. Culturable diversity of cold-adapted yeasts in worldwide cold environments (H)

## 5.7 Some new genera and species recently described by DBVPG (2)

Mycological Progress (2019) 18:945–971  
<https://doi.org/10.1007/s11557-019-01491-5>

DGfM

ORIGINAL ARTICLE

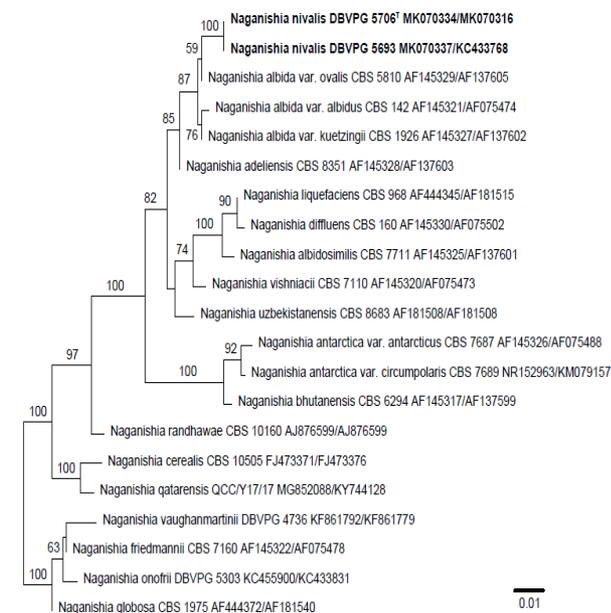


### Rare and undersampled dimorphic basidiomycetes

A. V. Kachalkin<sup>1,2</sup> · B. Turchetti<sup>3</sup> · J. Inácio<sup>4,5</sup> · C. Carvalho<sup>6</sup> · T. Mašínová<sup>7</sup> · A. Pontes<sup>6</sup> · O. Röhl<sup>8</sup> · A. M. Glushakova<sup>1</sup> · A. Akulov<sup>9</sup> · P. Baldrian<sup>7</sup> · D. Begerow<sup>8</sup> · P. Buzzini<sup>3</sup> · J. P. Sampaio<sup>6</sup> · A. M. Yurkov<sup>10</sup>

### New genera and species described

- ✓ *Vustinia terrea*
- ✓ *Udeniomyces* (*U. caspiensis*, *U. orazovii*)
- ✓ *Tausonia rosea*
- ✓ *Itersonia diksonensis*
- ✓ *Krasilnikovozyma fibulata*
- ✓ *Gelidatrema glaciarii*
- ✓ *Kwoniella fici*
- ✓ *Heterocephalacria* (*H. fruticeti*, *H. gelida*, *H. hypogea*, *H. lusitanica*)
- ✓ *Piskurozyma* (*P. arborea*, *P. silvicultrix*, *P. stramentorum*)
- ✓ *Naganishia nivalis*
- ✓ *Yurkovia nerthusi*



0.01

# 5. Culturable diversity of cold-adapted yeasts in worldwide cold environments (I)

## 5.7 Some new genera and species yet to be described by DBVPG

- ✓ ~ 13% of total strains resulted to belong to new species
- ✓ ~ 20% of species found were new species

A few examples

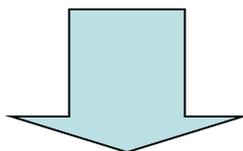


<b>FILOBASIDIUM sp.</b>				<b>DOSZEGIA sp. 1</b>			
<b>No of strains</b>	<b>year of</b>	<b>Locality</b>	<b>isolation sources</b>	<b>No of strains</b>	<b>year of</b>	<b>Locality</b>	<b>isolation sources</b>
13	from 2007 to 2010	Apennines	sediments, ice cores, snow	15	from 2010 to 2014	Alps	snow, sediments, melt water
<b>VISHNIACOZYMA sp.</b>				<b>DOSZEGIA sp. 2</b>			
<b>No of strains</b>	<b>year of</b>	<b>Locality</b>	<b>isolation sources</b>	<b>No of strains</b>	<b>year of</b>	<b>Locality</b>	<b>isolation sources</b>
3	2008, 2009	Alps	sediments,	2	2010, 2011	Alps	snow
<b>GELIDATREMA sp.</b>				<b>DOSZEGIA sp. 3</b>			
<b>No of strains</b>	<b>year of</b>	<b>Locality</b>	<b>isolation sources</b>	<b>No of strains</b>	<b>year of</b>	<b>Locality</b>	<b>isolation sources</b>
1	2008	Alps	snow	1	2011	Alps	snow
<b>CYSTOBASIDIUM sp.</b>				<b>NAGANISHA sp.</b>			
<b>No of strains</b>	<b>year of</b>	<b>Locality</b>	<b>isolation sources</b>	<b>No of strains</b>	<b>year of</b>	<b>Locality</b>	<b>isolation sources</b>
11	from 2008 to 2014	Alps	soils, sediments, snow	23	from 2008 to 2014	Alps	soils, sediments
<b>LEUCOSPORIDIUM sp.</b>				<b>MRAKIA sp.</b>			
<b>No of strains</b>	<b>year of</b>	<b>Locality</b>	<b>isolation sources</b>	<b>No of strains</b>	<b>year of</b>	<b>Locality</b>	<b>isolation sources</b>
1	2008	Alps	soil	3	2009, 2010	Alps	sediments, ice cores,
<b>CRYPTOCOCCUS sp.</b>				<b>AUREOBASIDIUM SP.</b>			
<b>No of strains</b>	<b>year of</b>	<b>Locality</b>	<b>isolation sources</b>	<b>No of strains</b>	<b>year of</b>	<b>Locality</b>	<b>isolation sources</b>
1	2009	Miage	sedimenti	1	2009	Alps	sediments

## 6. Unculturable diversity (NGS) of cold-adapted fungi (including yeasts) (A)

### 6.1 A paradigm shift (2020-today)

- Most microbial taxa (including yeasts) cannot be cultured under laboratory conditions
- As a consequence, culturomic results represent at best only a partial picture of the yeast diversity occurring in worldwide cold ecosystems

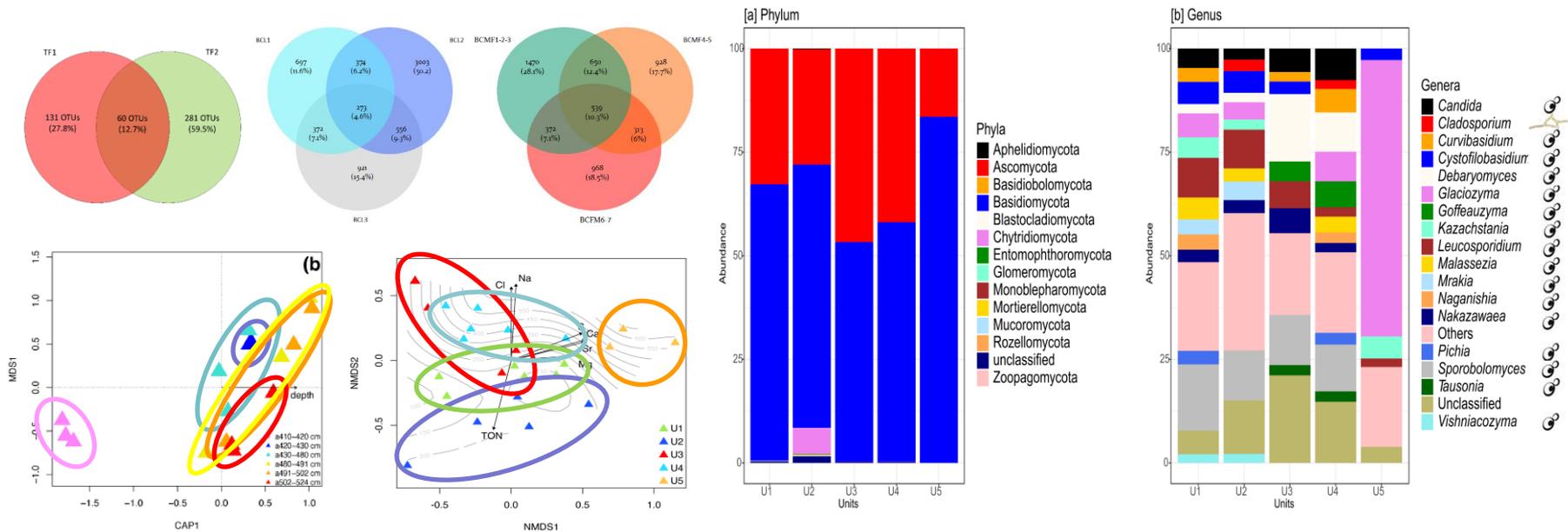


- Non-culturable yeast (fungal) diversity → Next Generation Sequencing of Antarctic and Alpine samples

# 6. Unculturable diversity (NGS) of cold-adapted fungi (including yeasts) (B)

## 6.2 Recent results on fungal biodiversity found on Antarctic and Alpine habitats: a general overview (2020-today)

- ✓ Alpha-diversity and beta-diversity → fungal (including yeasts) communities well adapted to cold ecosystems
- ✓ High differentiation of fungal communities among different sites/layers → predominance of specialist OTUs



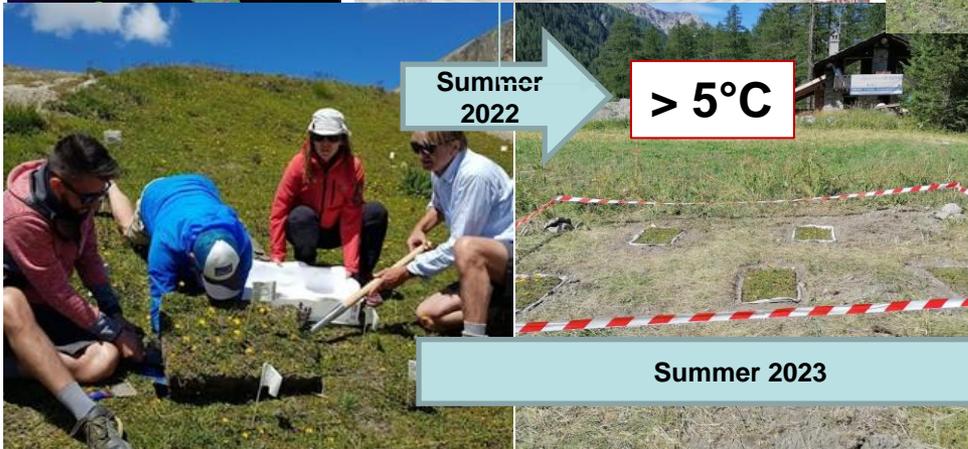
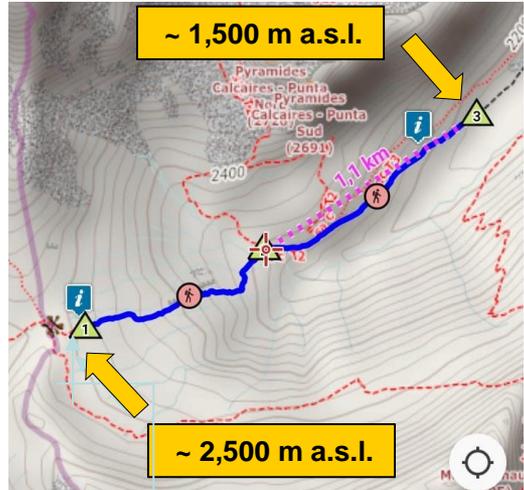
- ✓ Yeasts dominated the fungal biodiversity in almost all studied habitats

# 7. Predicting the impact of climate change on Alpine soil fungal community

## 7.1 Ongoing project (2022-2025): "MICROPLANTALP"

<https://sites.google.com/unitus.it/microplantalp/home>

PRIN 2020



# 8. Physiological/molecular adaptation to cold (A)

## 8.1 Cold-active enzymes

- ✓ Extracellular cold-active enzymes degrading organic polymers → *in-situ* C and N co-mineralizers?

### RESEARCH ARTICLE

#### Psychrophilic yeasts in glacial environments of Alpine glaciers

Benedetta Turchetti<sup>1</sup>, Pietro Buzzini<sup>1</sup>, Marta Goretti<sup>1</sup>, Eva Branda<sup>1</sup>, Guglielmina Diolaiuti<sup>2</sup>, Carlo D'Agata<sup>2</sup>, Claudio Smiraglia<sup>2</sup> & Ann Vaughan-Martini<sup>1</sup>

<sup>1</sup>Dipartimento di Biologia Applicata, Sezione di Microbiologia, University of Perugia, Perugia, Italy; and <sup>2</sup>Dipartimento di Scienze della Terra 'Ardito Desio', University of Milan, Milan, Italy



### RESEARCH ARTICLE

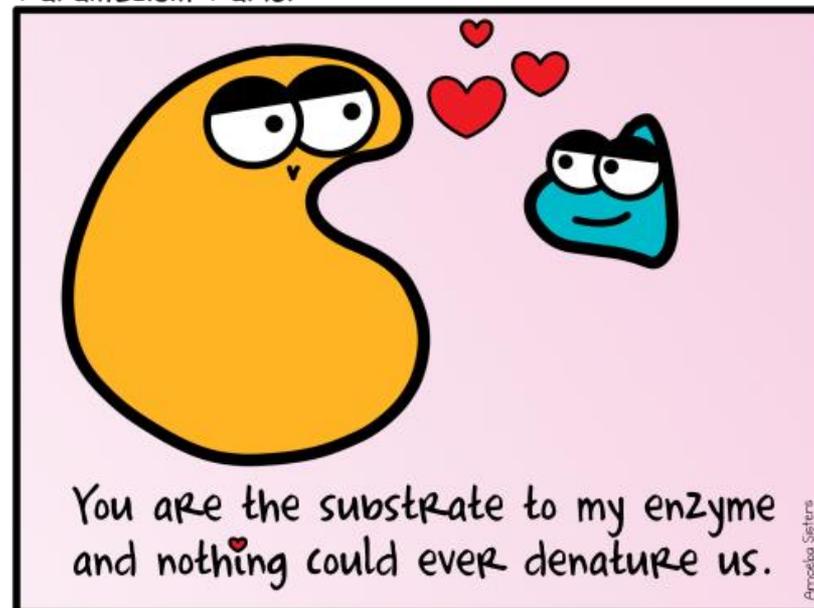
#### Yeast and yeast-like diversity in the southernmost glacier of Europe (Calderone Glacier, Apennines, Italy)

Eva Branda<sup>1</sup>, Benedetta Turchetti<sup>1</sup>, Guglielmina Diolaiuti<sup>2</sup>, Massimo Pecci<sup>3</sup>, Claudio Smiraglia<sup>2</sup> & Pietro Buzzini<sup>1</sup>

<sup>1</sup>Department of Applied Biology and Industrial Yeasts Collection DBVPG, University of Perugia, Italy; <sup>2</sup>Department of Health Sciences 'Ardito Desio', University of Milan, Italy; and <sup>3</sup>Italian Mountain Institute (EIM), Rome, Italy



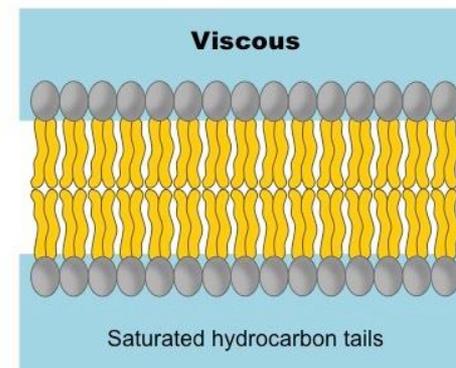
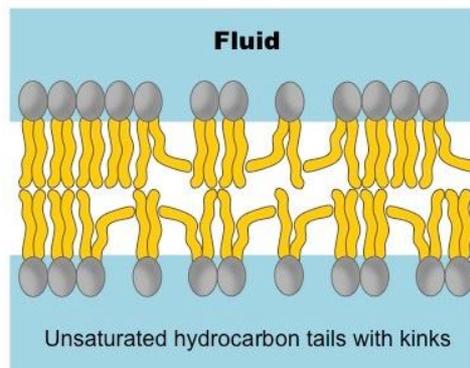
### Paramecium Parlor



# 8. Physiological/molecular adaptation to cold (B)

## 8.2 Lipid composition

- High concentration of intracellular unsaturated lipids (e.g. PUFAs) → increased fluidity of cell cytoplasm and membranes at low temperatures



RESEARCH ARTICLE

### Growth, lipid accumulation, and fatty acid composition in obligate psychrophilic, facultative psychrophilic, and mesophilic yeasts

Maddalena Rossi<sup>1</sup>, Pietro Buzzini<sup>2</sup>, Lisa Cordisco<sup>3</sup>, Alberto Amaretti<sup>1</sup>, Maurizio Sala<sup>1</sup>, Stefano Raimondi<sup>1</sup>, Chiara Ponzoni<sup>1</sup>, Ugo Maria Pagnoni<sup>1</sup> & Diego Matteuzzi<sup>3</sup>

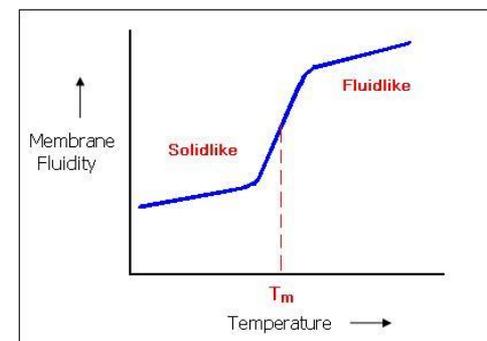
<sup>1</sup>Department of Chemistry, University of Modena and Reggio Emilia, Modena, Italy; <sup>2</sup>Department of Applied Biology, Industrial Yeasts Collection DBVPG, University of Perugia, Perugia, Italy; and <sup>3</sup>Department of Pharmaceutical Sciences, University of Bologna, Bologna, Italy

### Chapter 10 Changes in Lipids Composition and Fluidity of Yeast Plasma Membrane as Response to Cold

Nina Gunde-Cimerman, Ana Plemenitaš and Pietro Buzzini

DOI: 10.1007/978-3-642-39681-6\_5, © Springer-Verlag Berlin Heidelberg 2014

Membrane Fluidity vs. Temperature

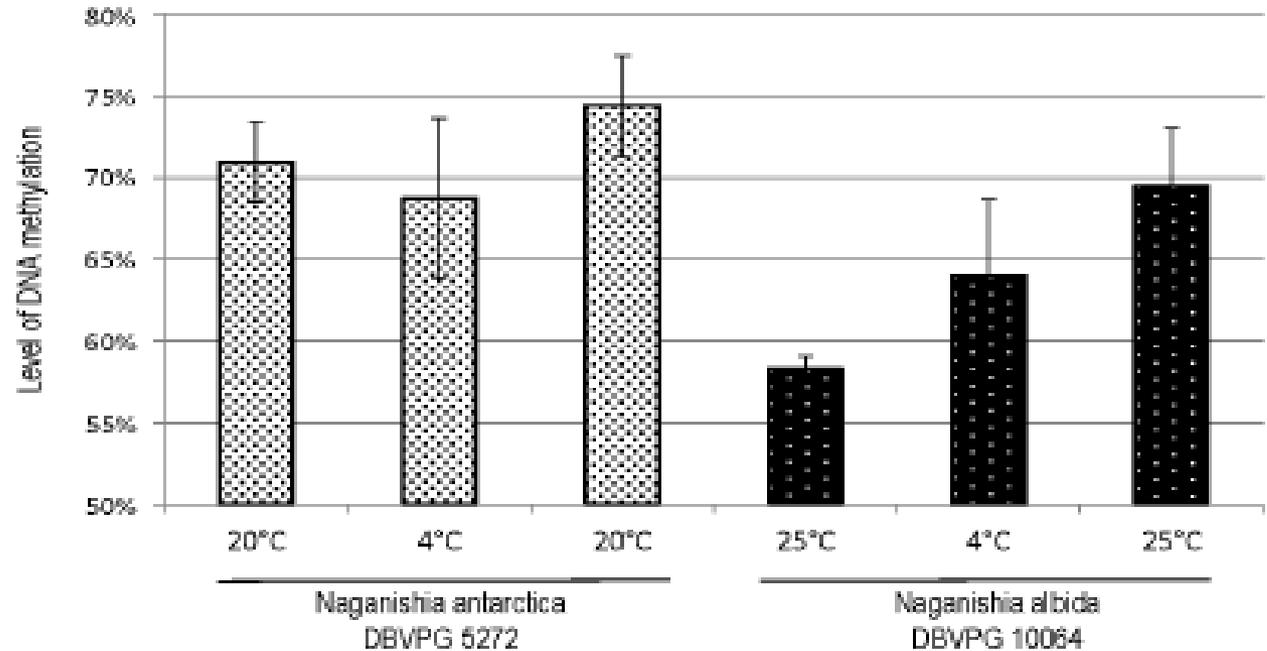
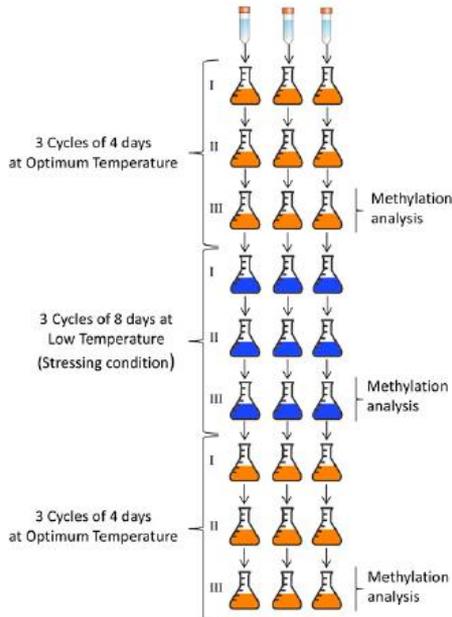


With an increase in temperature, the sharp transition is made from a more rigid membrane to a more fluid one.



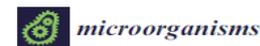
# 8. Physiological/molecular adaptation to cold (C)

## 8.2 DNA methylation



✓ *N. antarctica* → no increase of methylation level after recovery from stress → **reversible** cold-induced physiology and biochemistry

✓ *N. albida* → increase of methylation level after recovery from stress → **not-reversible** cold-induced physiology and biochemistry



Article  
DNA Methylation Changes Induced by Cold in Psychrophilic and Psychrotolerant *Naganishia* Yeast Species

Benedetta Turchetti <sup>\*,†</sup>, Gianpiero Marconi <sup>†</sup>, Ciro Sannino, Pietro Buzzini and Emidio Albertini <sup>✉</sup>

Department of Agricultural, Food and Environmental Sciences, University of Perugia, Borgo XX Giugno, 74, 06121 Perugia, Italy; gianpiero.marconi@unipg.it (G.M.); ciro.sannino@unipg.it (C.S.); pietero.buzzini@unipg.it (P.B.); emidio.albertini@unipg.it (E.A.)

\* Correspondence: benedetta.turchetti@unipg.it; Tel: +39-0755-856-487  
† Contributed equally to this work.

# 9. A look to biotechnology of cold-adapted yeasts (A)

## 9.1 Cold-active enzymes for mild technologies (A)

- ✓ A number of yeasts strains were found to secrete extracellular enzymes active at 4°C

### A few examples:

- *Goffeauzyma gilvescens*: 90% positive for SDA, 100% for esterase activity
- *Mrakia gelida*: 100% positive for SDA\* and protease activity
- *Cystobasidium laryngis*: 100% positive for esterase activity

\* Starch Degrading Activity



#### RESEARCH ARTICLE

#### **Psychrophilic yeasts in glacial environments of Alpine glaciers**

Benedetta Turchetti<sup>1</sup>, Pietro Buzzini<sup>1</sup>, Marta Goretti<sup>1</sup>, Eva Branda<sup>1</sup>, Guglielmina Diolaiuti<sup>2</sup>, Carlo D'Agata<sup>2</sup>, Claudio Smiraglia<sup>2</sup> & Ann Vaughan-Martini<sup>1</sup>

<sup>1</sup>Dipartimento di Biologia Applicata, Sezione di Microbiologia, University of Perugia, Perugia, Italy; and <sup>2</sup>Dipartimento di Scienze della Terra 'Ardito Desio', University of Milan, Milan, Italy



#### RESEARCH ARTICLE

#### **Yeast and yeast-like diversity in the southernmost glacier of Europe (Calderone Glacier, Apennines, Italy)**

Eva Branda<sup>1</sup>, Benedetta Turchetti<sup>1</sup>, Guglielmina Diolaiuti<sup>2</sup>, Massimo Pecci<sup>3</sup>, Claudio Smiraglia<sup>2</sup> & Pietro Buzzini<sup>1</sup>

<sup>1</sup>Department of Applied Biology and Industrial Yeasts Collection DBVPG, University of Perugia, Italy; <sup>2</sup>Department of Health Sciences 'Ardito Desio', University of Milan, Italy; and <sup>3</sup>Italian Mountain Institute (EIM), Rome, Italy

# 9. A look to biotechnology of cold-adapted yeasts (B)

## 9.2 Cold-active enzymes for mild technologies (B)



RESEARCH ARTICLE

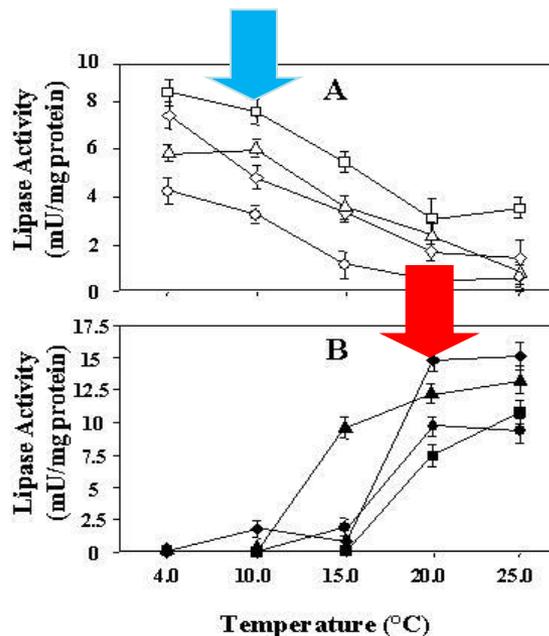
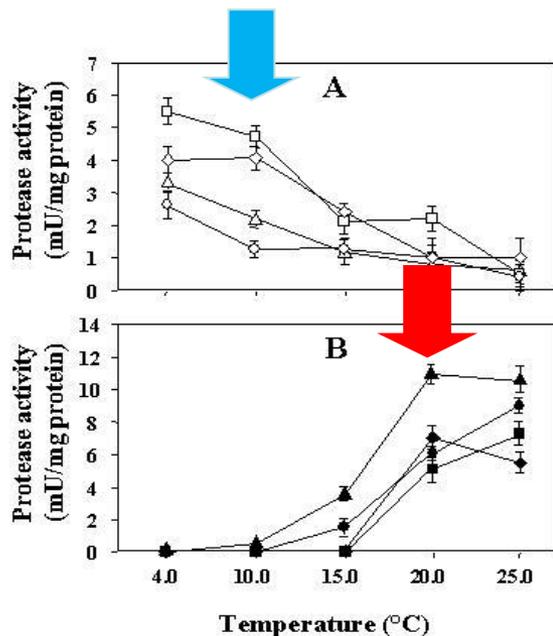
### Yeast and yeast-like diversity in the southernmost glacier of Europe (Calderone Glacier, Apennines, Italy)

Eva Branda<sup>1</sup>, Benedetta Turchetti<sup>1</sup>, Guglielmina Diolaiuti<sup>2</sup>, Massimo Pecci<sup>3</sup>, Claudio Smiraglia<sup>2</sup> & Pietro Buzzini<sup>1</sup>

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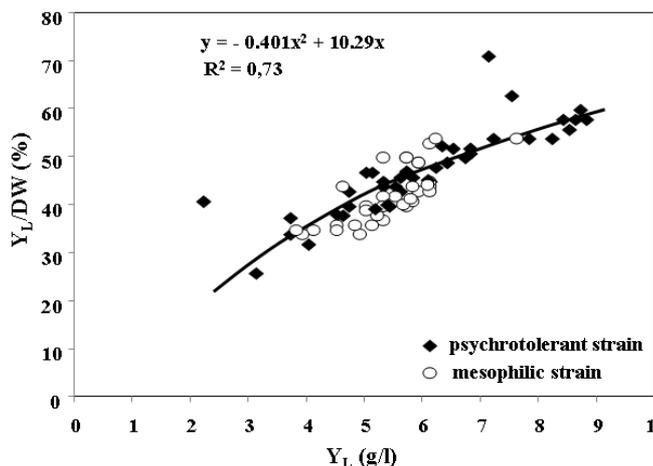
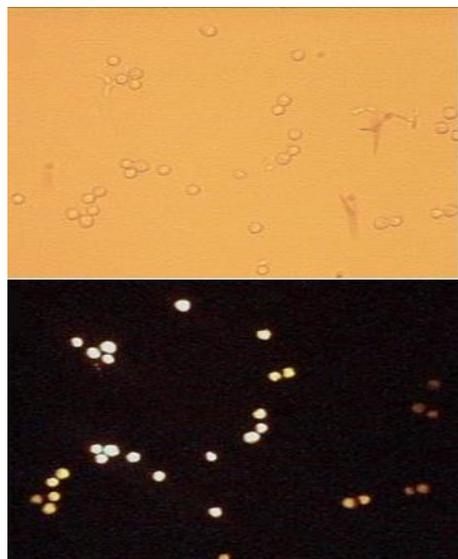
Extracellular enzymatic activity at different temperatures of cell-free extracts of psychrophilic yeasts isolated from:

- glacial (A) habitats
- tropical (B) habitats



# 9. A look to biotechnology of cold-adapted yeasts (C)

## 9.2 Accumulation of PUFAs



Biotechnology for Biofuels  
 DOI 10.1186/s13068-016-0672-1

RESEARCH

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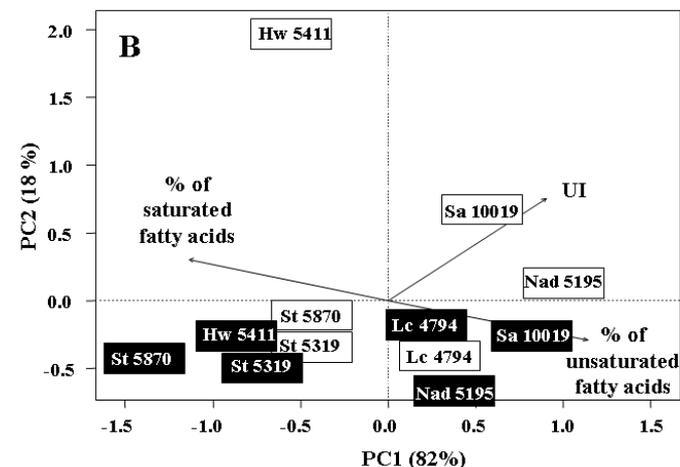
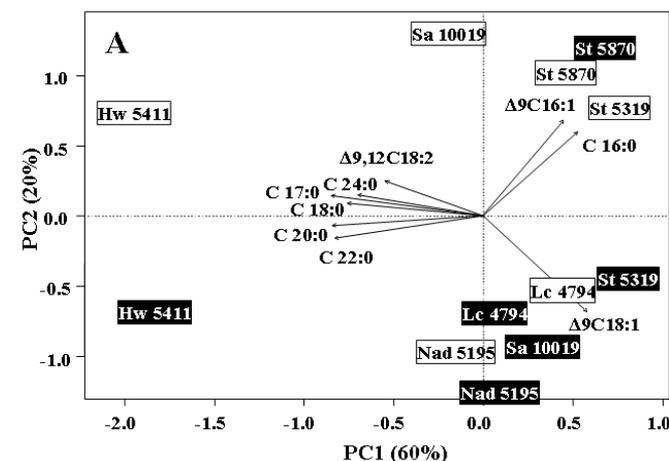


Adaptation strategy to low temperatures:

- increased lipid production
- increased lipid unsaturation

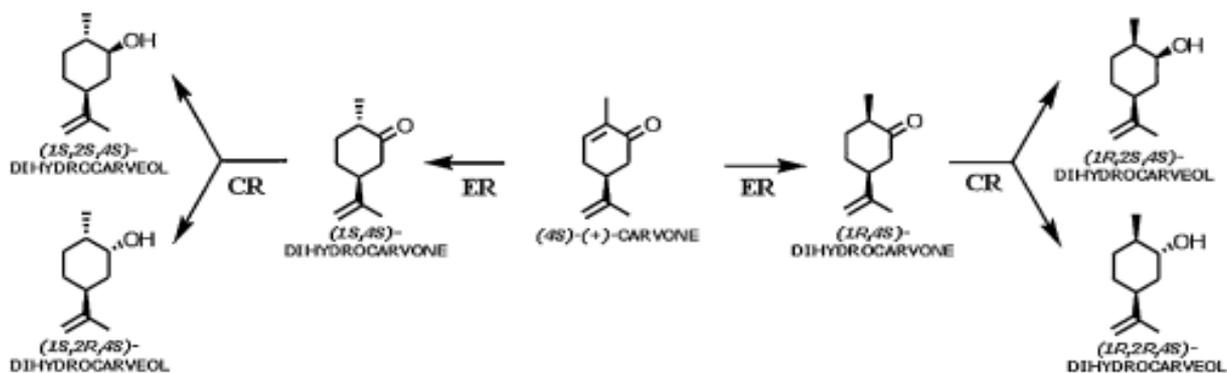
Study of *Holtermanniella wattica*, *Leucosporidium creatinivorum*, *Naganishia adeliensis*, *Solicoccozyma aeria*, and *Solicoccozyma terricola* for their lipogenic aptitude from different carbon sources

Sara Filippucci<sup>1†</sup>, Giorgia Tasselli<sup>1†</sup>, Alessandro Scardua<sup>2</sup>, Simone Di Mauro<sup>1</sup>, Maria Rita Cramarossa<sup>3</sup>, Davide Perini<sup>2</sup>, Benedetta Turchetti<sup>1</sup>, Andrea Onofri<sup>1</sup>, Luca Forti<sup>3</sup> and Pietro Buzzini<sup>1\*</sup>

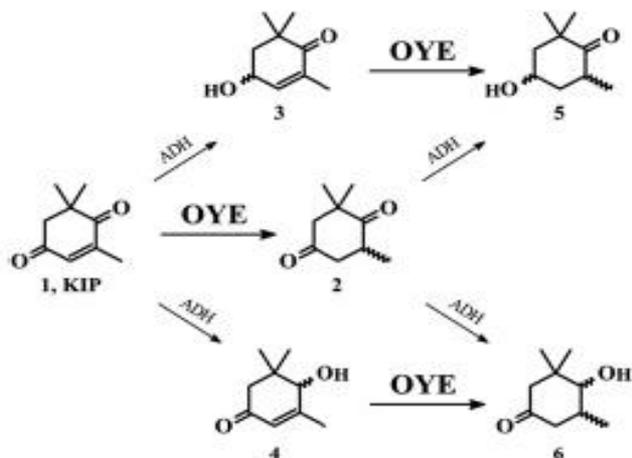


# 9. A look to biotechnology of cold-adapted yeasts (D)

## 9.3 Stereoselective catalysis of NADH/NADPH-mediated redox reactions



- ✓ Ability to reduce  $\alpha,\beta$  electron-poor compounds
- ✓ Synthesis of “building-blocks” molecules of pharmaceutical interest via biocatalysis



Bioresource Technology 102 (2011) 3993–3998



Contents lists available at ScienceDirect

Bioresource Technology

journal homepage: [www.elsevier.com/locate/biortech](http://www.elsevier.com/locate/biortech)



Bioreduction of  $\alpha,\beta$ -unsaturated ketones and aldehydes by non-conventional yeast (NCY) whole-cells

Marta Goretti<sup>a</sup>, Chiara Ponzoni<sup>b</sup>, Elisa Caselli<sup>b</sup>, Elisabetta Marchegiani<sup>a</sup>, Maria Rita Cramarossa<sup>b</sup>, Benedetta Turchetti<sup>a</sup>, Luca Forti<sup>b</sup>, Pietro Buzzini<sup>a,\*</sup>

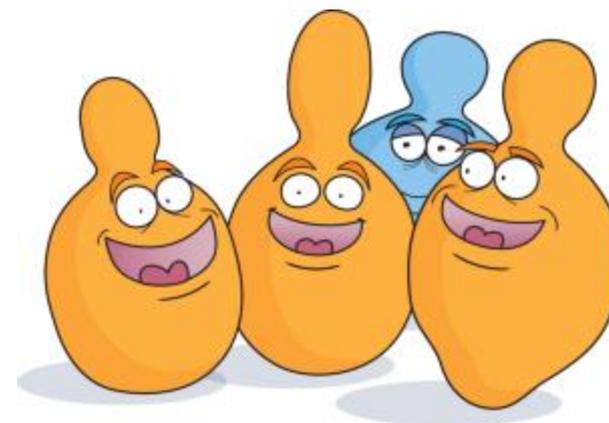
# 10. Take home messages

- Cold-adapted yeasts → extremophilic organisms inhabiting cold habitats worldwide
- A source of new genera/species
- The study of unculturable yeast diversity by NGS → a more complete picture
- Physiological/molecular adaptation to cold → peculiar mechanisms (e.g. cold-active enzymes, lipid composition, DNA methylation, etc.)
- Cold-adapted yeasts → possible biotechnological exploitation

## Take Home Messages



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# 11. Acknowledgements

1. C. Sannino, G. Mugnai, A. Bernetti, D. Andreani, B. Turchetti – University of Perugia & DBVPG, Italy
2. L. Borruso – University of Bozen /Bolzano, Italy



- ✓ A. Yurkov, Leibniz Institute DSMZ-German Collection of Microorganisms and Cell Cultures, Brunswick, Germany
- ✓ T. Boekhout, Westerdijk Fungal Biodiversity Institute, The Netherlands
- ✓ S. R. Thomas-Hall, University of New England, Australia
- ✓ L. B. Connell, University of Maine, USA
- ✓ D. Libkind, University of Comahue, CONICET, Argentina
- ✓ N. Gunde-Cimerman, Biology Department, Biotechnical Faculty, University of Ljubljana, Slovenia
- ✓ R. Margesin, Institute of Microbiology, University of Innsbruck, Austria
- ✓ M. Baeza Cancino, Universidad De Chile, Santiago, Chile



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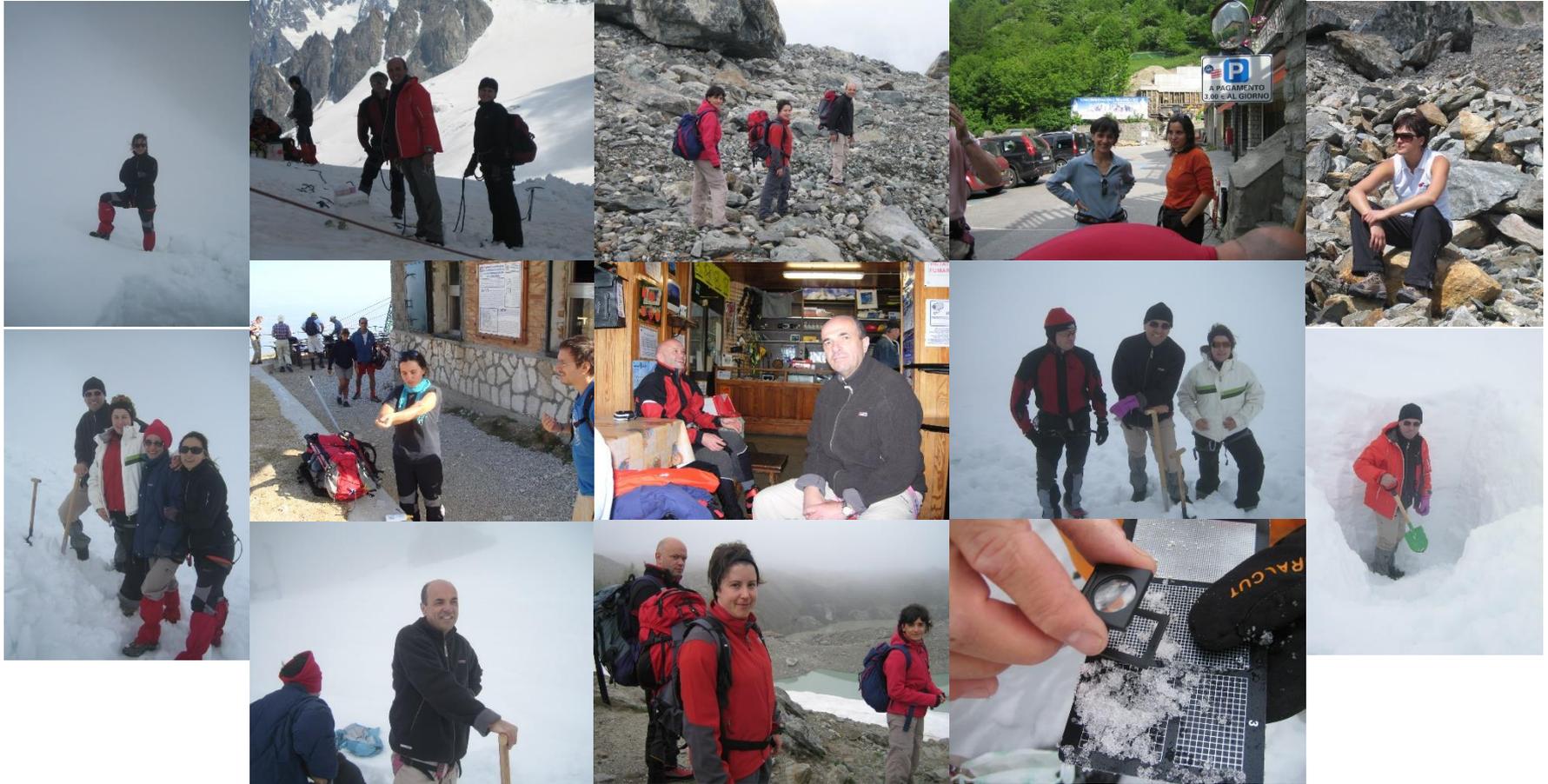


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# 12. Sampling pictures



# 13. A bit of humor

100 *How to Write and Publish a Scientific Paper*



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