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Cold adaptation of the cheese ripening bacterium *Protectionniste freudenreichii*

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► **To cite this version:**

Marion Dalmasso, H el ene Falentin, St ephanie-Marie Deutsch, Gw ena el Jan, Sandrine Parayre-Breton, et al.. Cold adaptation of the cheese ripening bacterium *Protectionniste freudenreichii*. 8 Cheese Symposium, Sep 2011, Cork, Ireland. 91 (6), 124 p., 2011, Dairy Science and Technology. hal-01454331

HAL Id: hal-01454331

<https://hal.science/hal-01454331>

Submitted on 3 Jun 2020

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Cold adaptation of the cheese ripening bacterium *Propionibacterium freudenreichii*

Novel Technologies for Sustainable Dairy Products

Dairy Science and Technology

Dr. Marion Dalmasso

SCIENCE AND TECHNOLOGY
OF MILK AND EGGS RESEARCH UNIT
Rennes, FRANCE

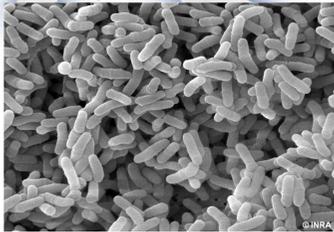


ALIMENTATION
AGRICULTURE
ENVIRONNEMENT





CONTEXT



Propionibacterium freudenreichii is widely used in Swiss-type cheese manufacture and especially in Emmental cheese.

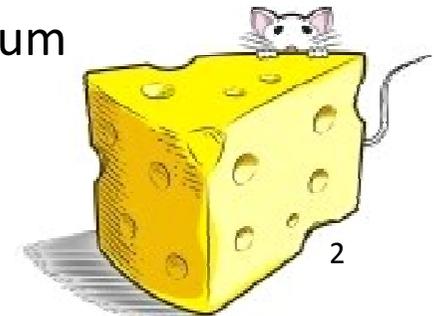


In France:

- Emmental cheese production > 253 000 tones (in 2010)
 - 76% of the production of cow milk hard-cooked cheeses
 - 37% of the total production of cheeses
- Consumption: 4 kg of Emmental cheese/person/year

Emmental cheese ripening is based on different combinations of time and temperature, according to manufacturer's practices.

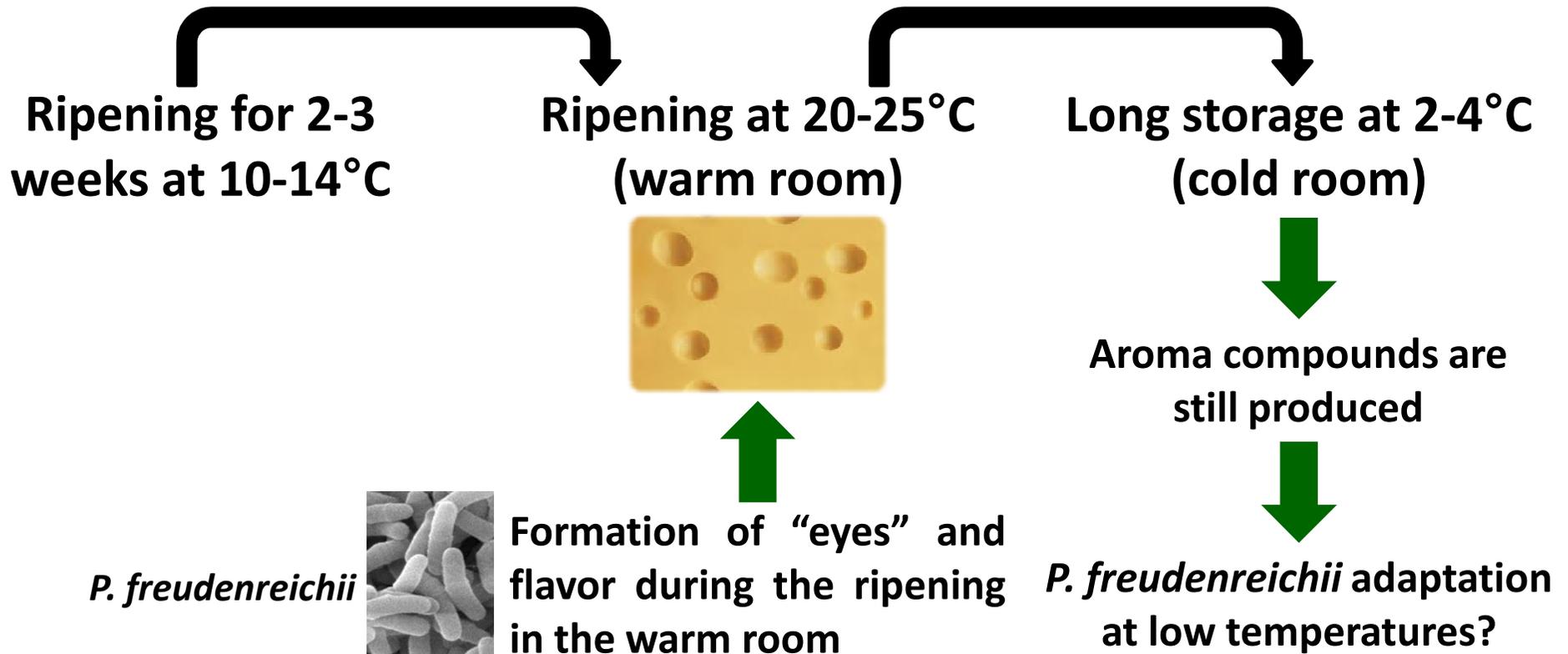
In France, the legal ripening time of Emmental cheese is a minimum of 6 weeks. But, it can be extended to months, like for example Emmental PDO cheese from Switzerland.





CONTEXT

Emmental cheese ripening

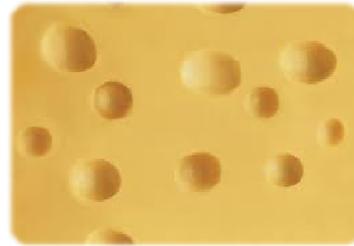




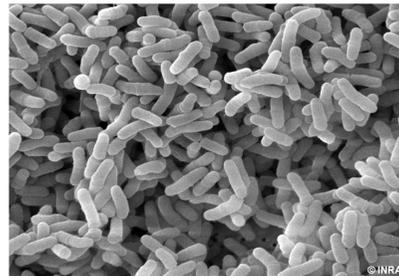
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OBJECTIVE



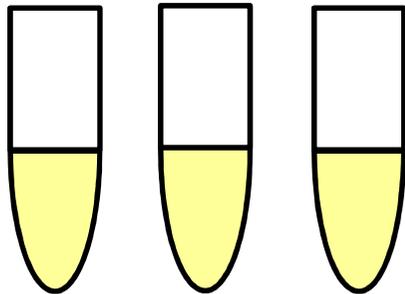
How *P. freudenreichii* copes with the cold-induced stress and remains active when cheeses are transferred to the cold room?





STRATEGY:

An approach combining transcriptomics, proteomics and metabolomics



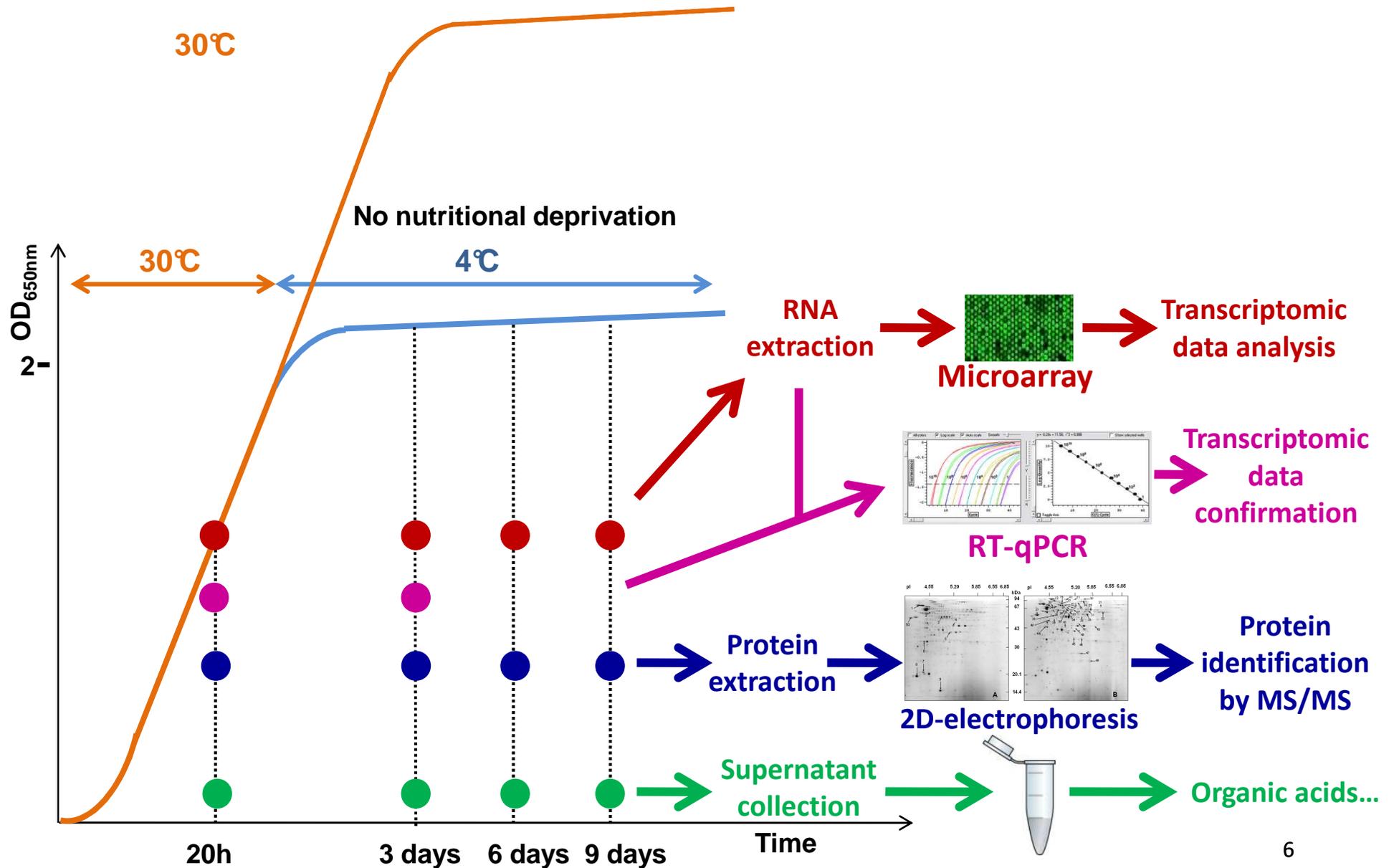
Cultures in triplicate

- ✓ The sequenced strain CIRM-BIA1^T
- ✓ The medium had a composition in complex nitrogen sources and carbon (lactate) close to Swiss cheese aqueous phase

→ Nutritional conditions mimicking cheese environment

STRATEGY:

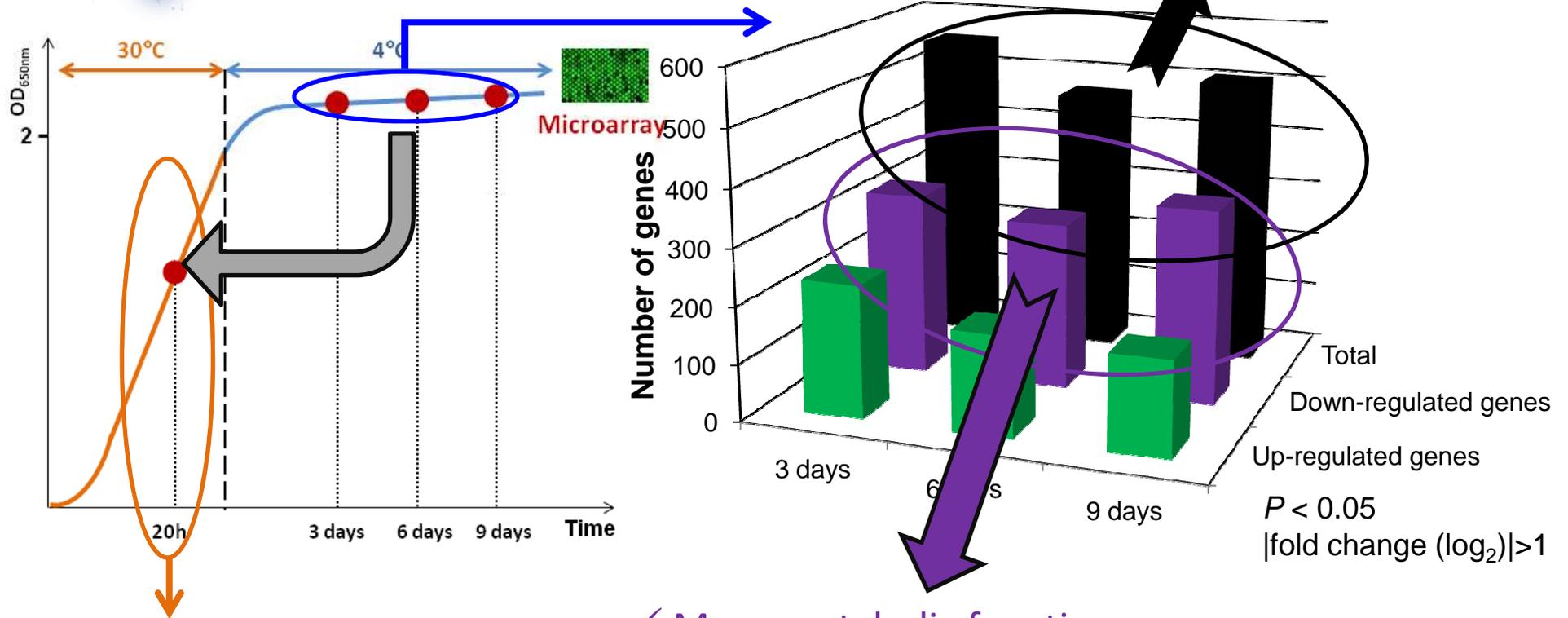
An approach combining transcriptomics, proteomics and metabolomics





RESULTS: General slowing down of cell machinery

✓ 25% of *P. freudenreichii* genes

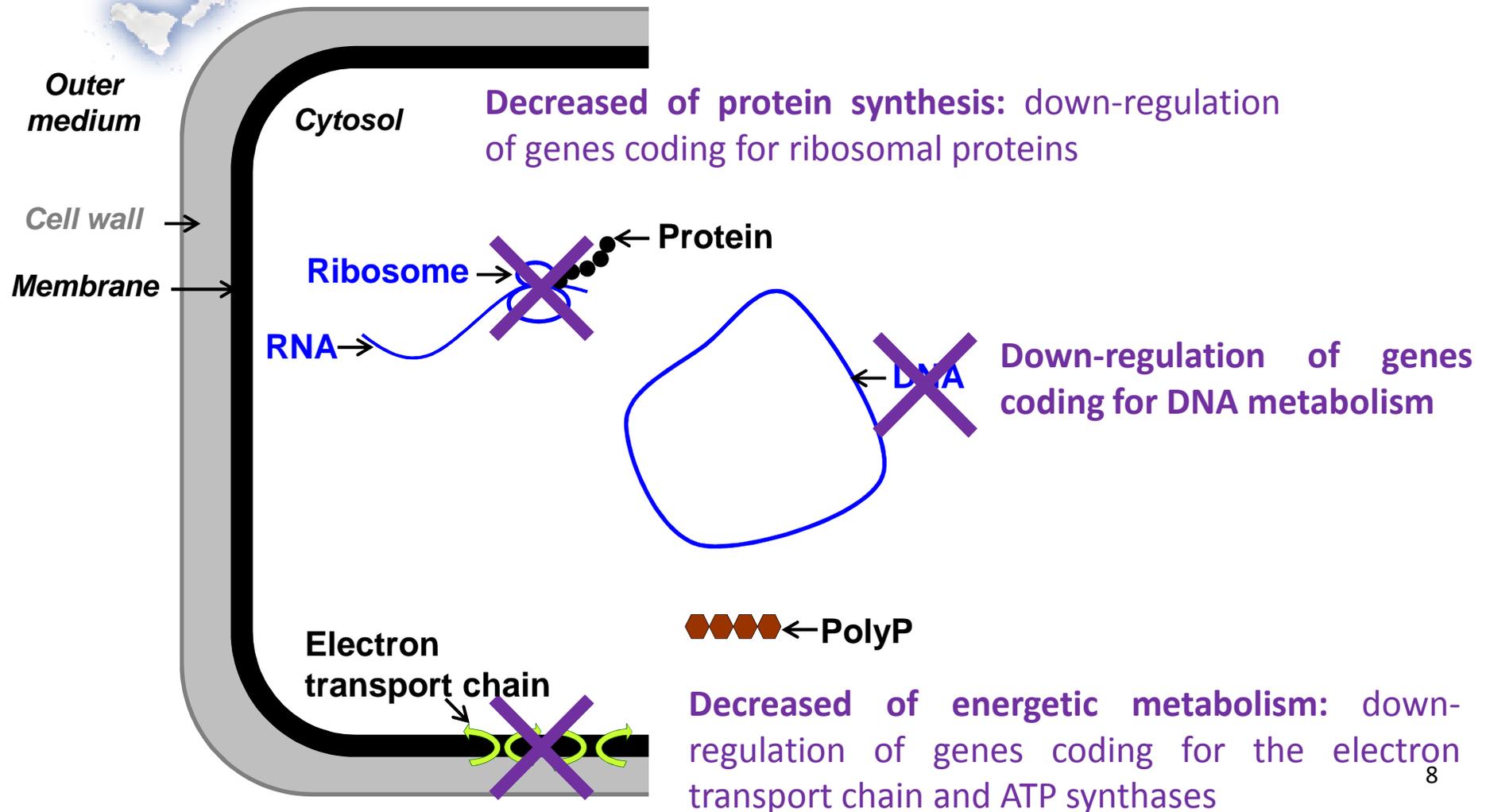


✓ Many metabolic functions were down-regulated in the cold

Reference time



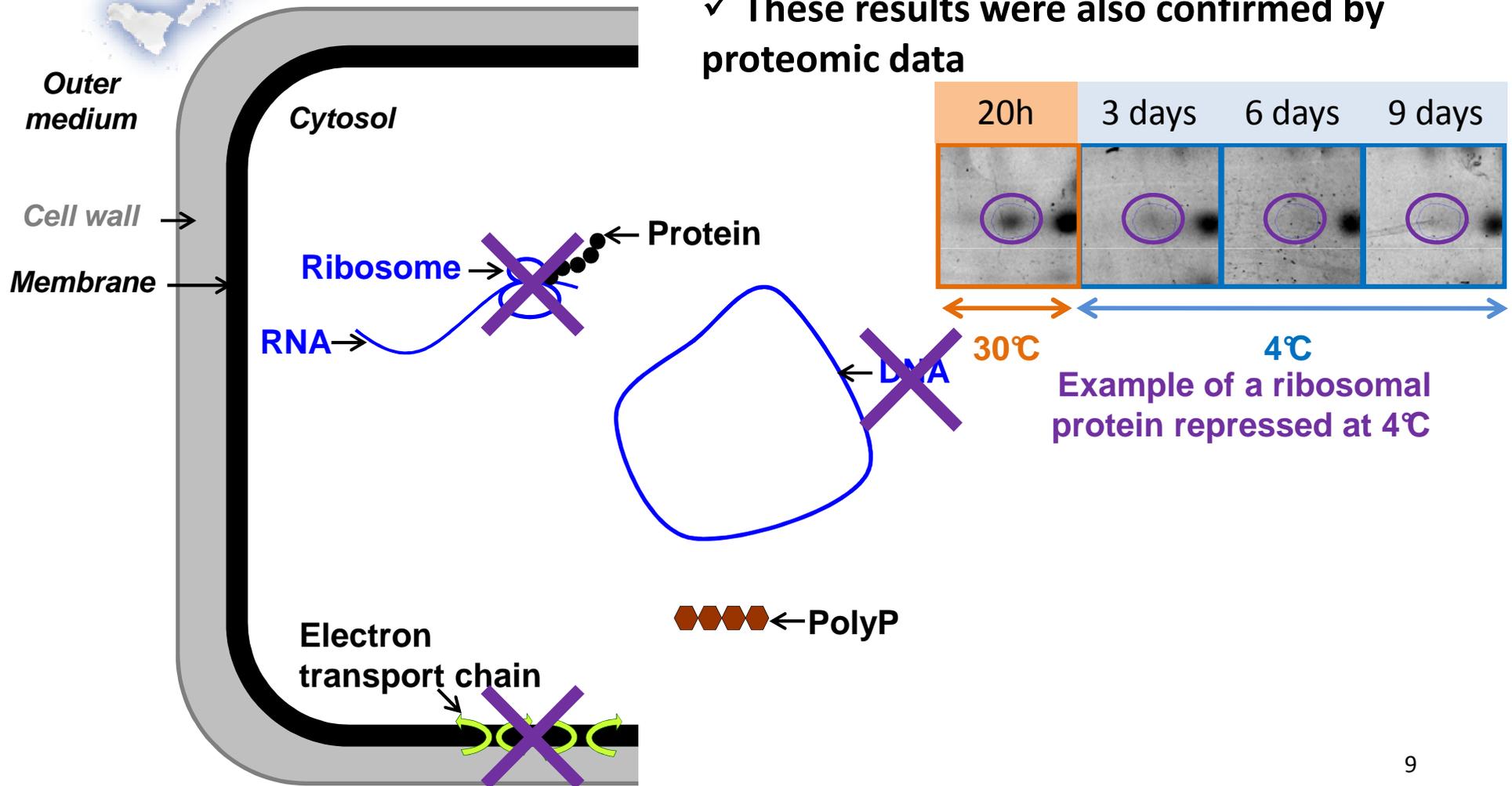
RESULTS: General slowing down of cell machinery





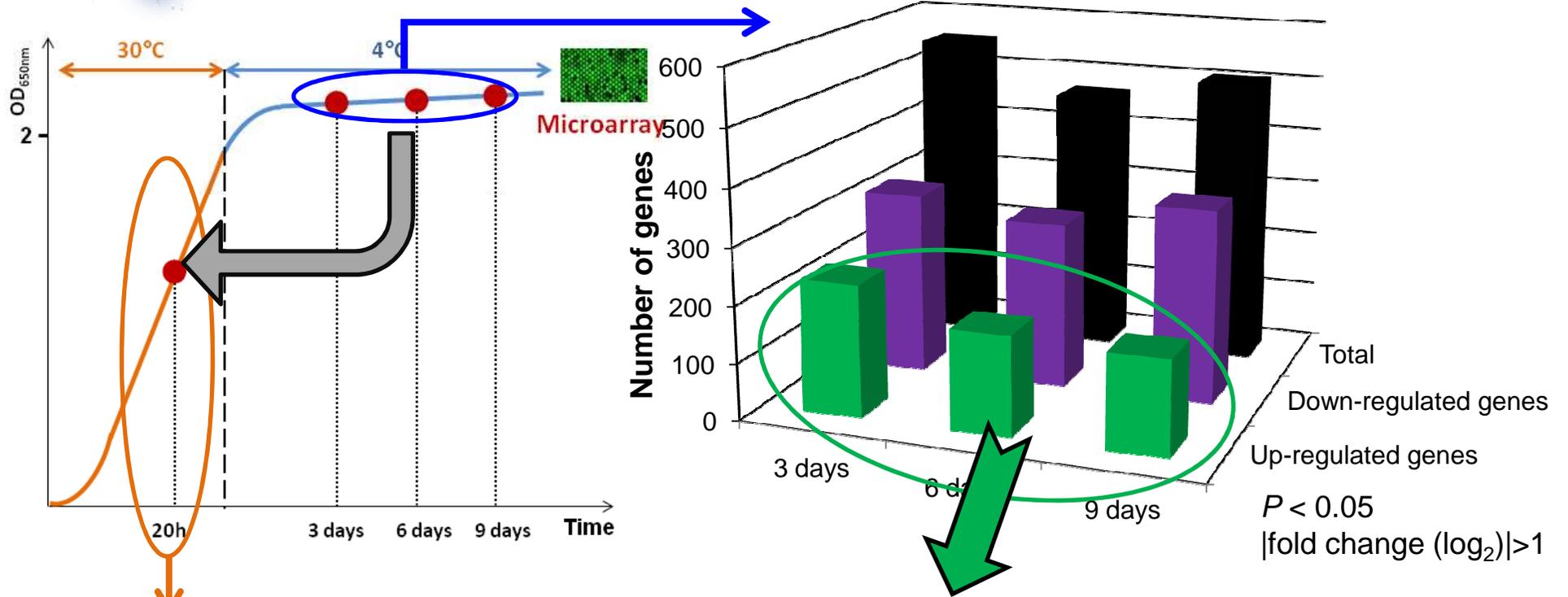
RESULTS: General slowing down of cell machinery

✓ These results were also confirmed by proteomic data





RESULTS: Cell adaptation and survival in the cold

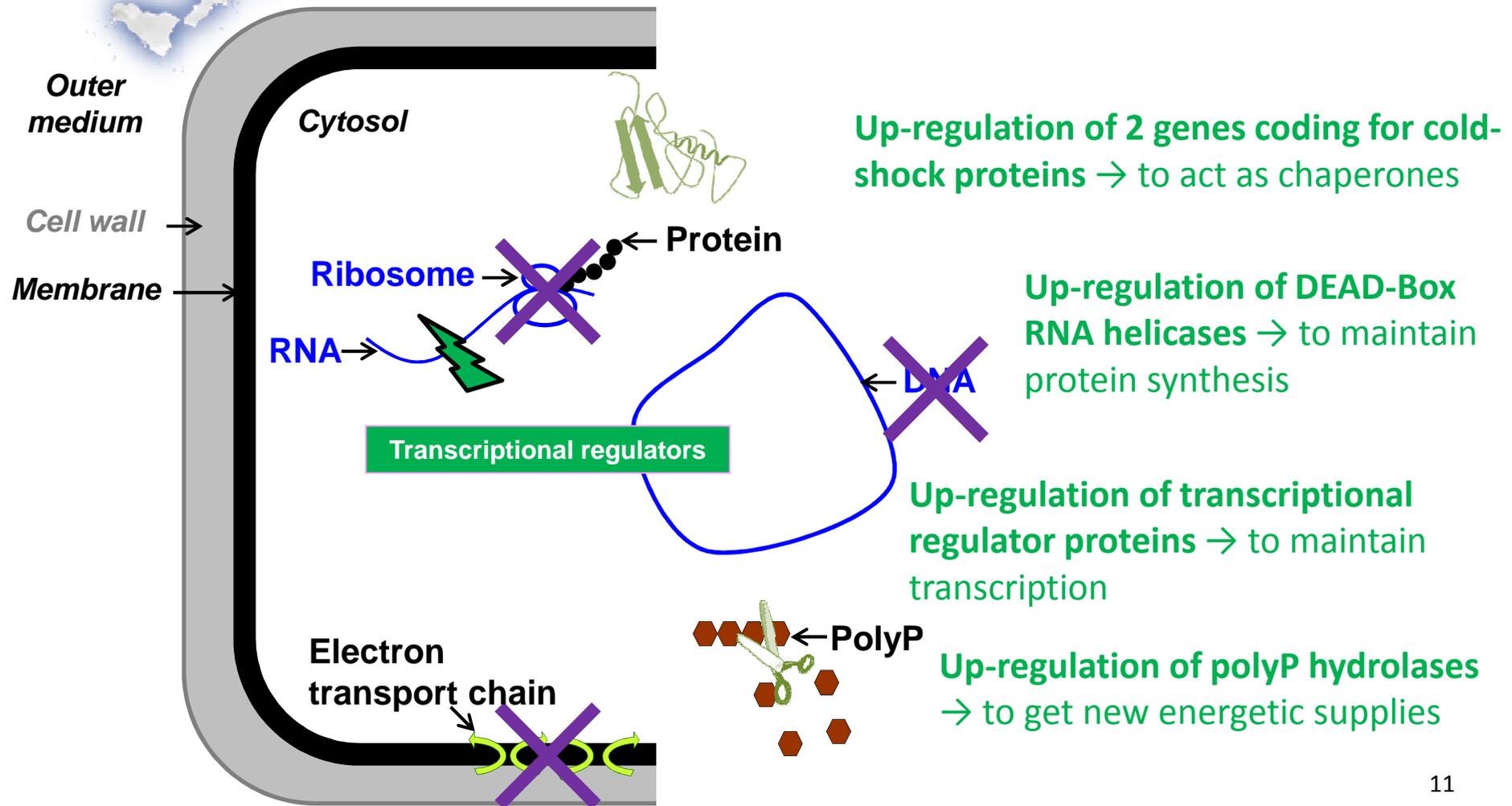


Reference time

✓ Metabolic functions displayed up-regulated genes at 4°C



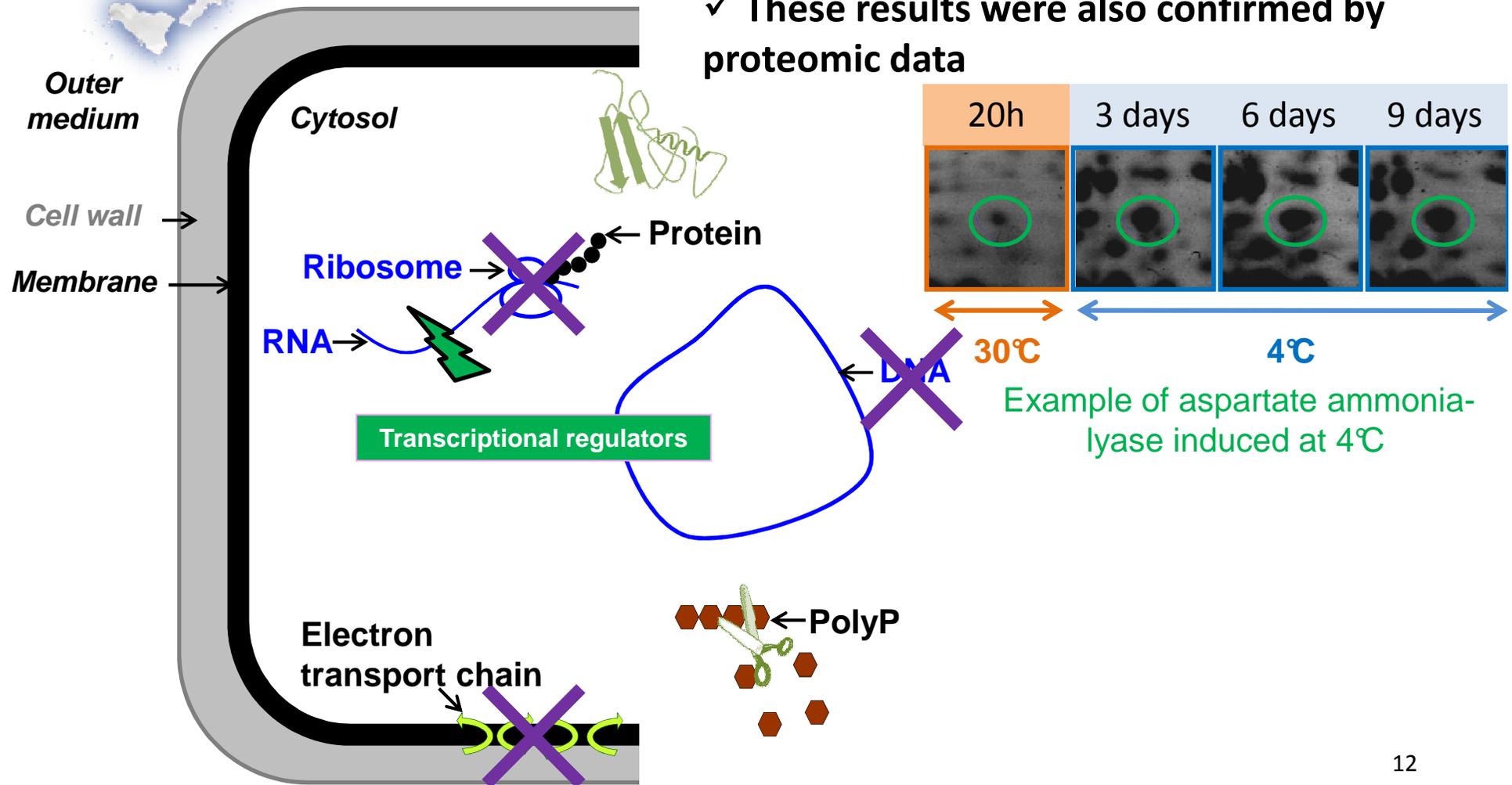
RESULTS: Cell adaptation and survival in the cold





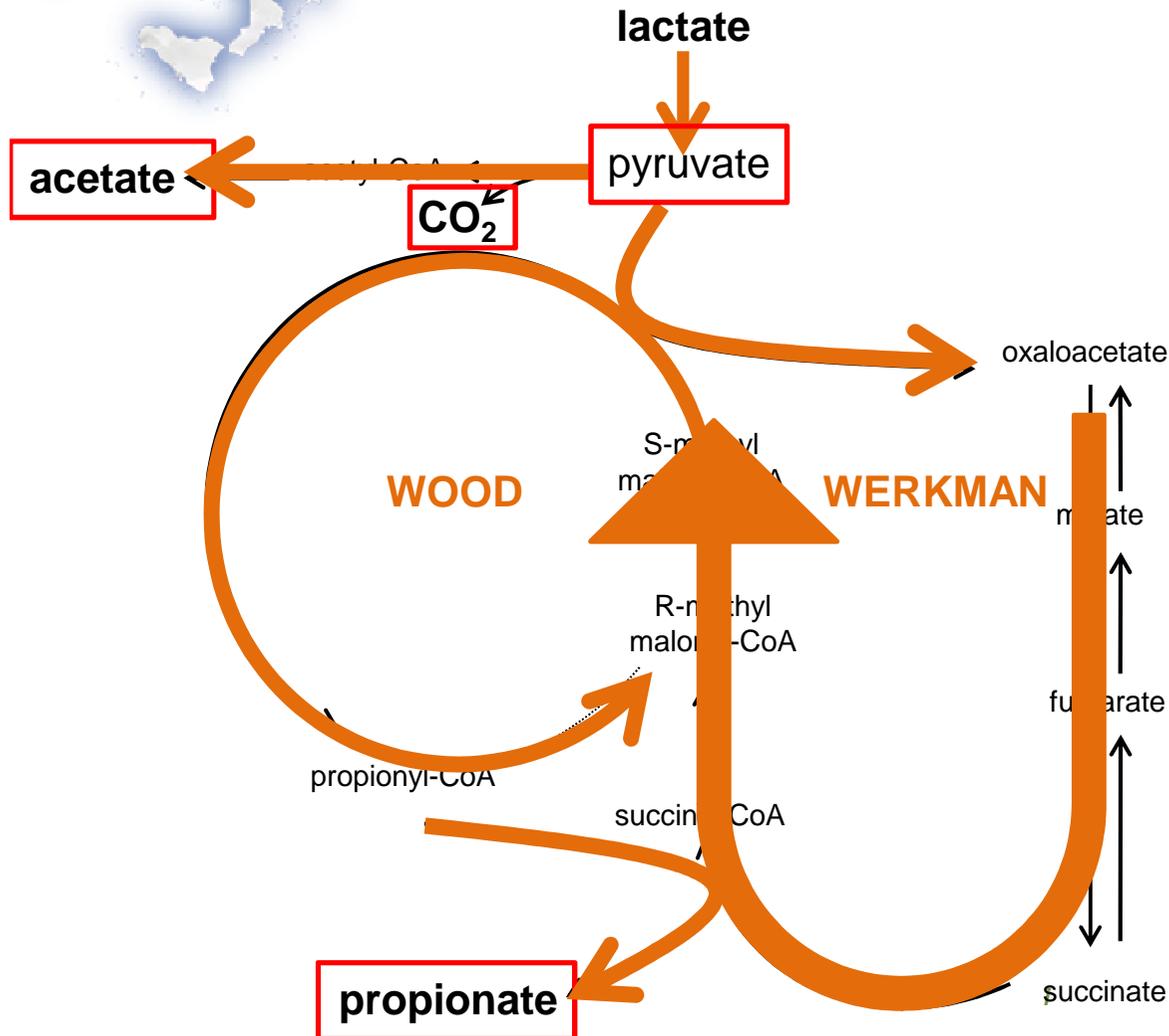
RESULTS: Cell adaptation and survival in the cold

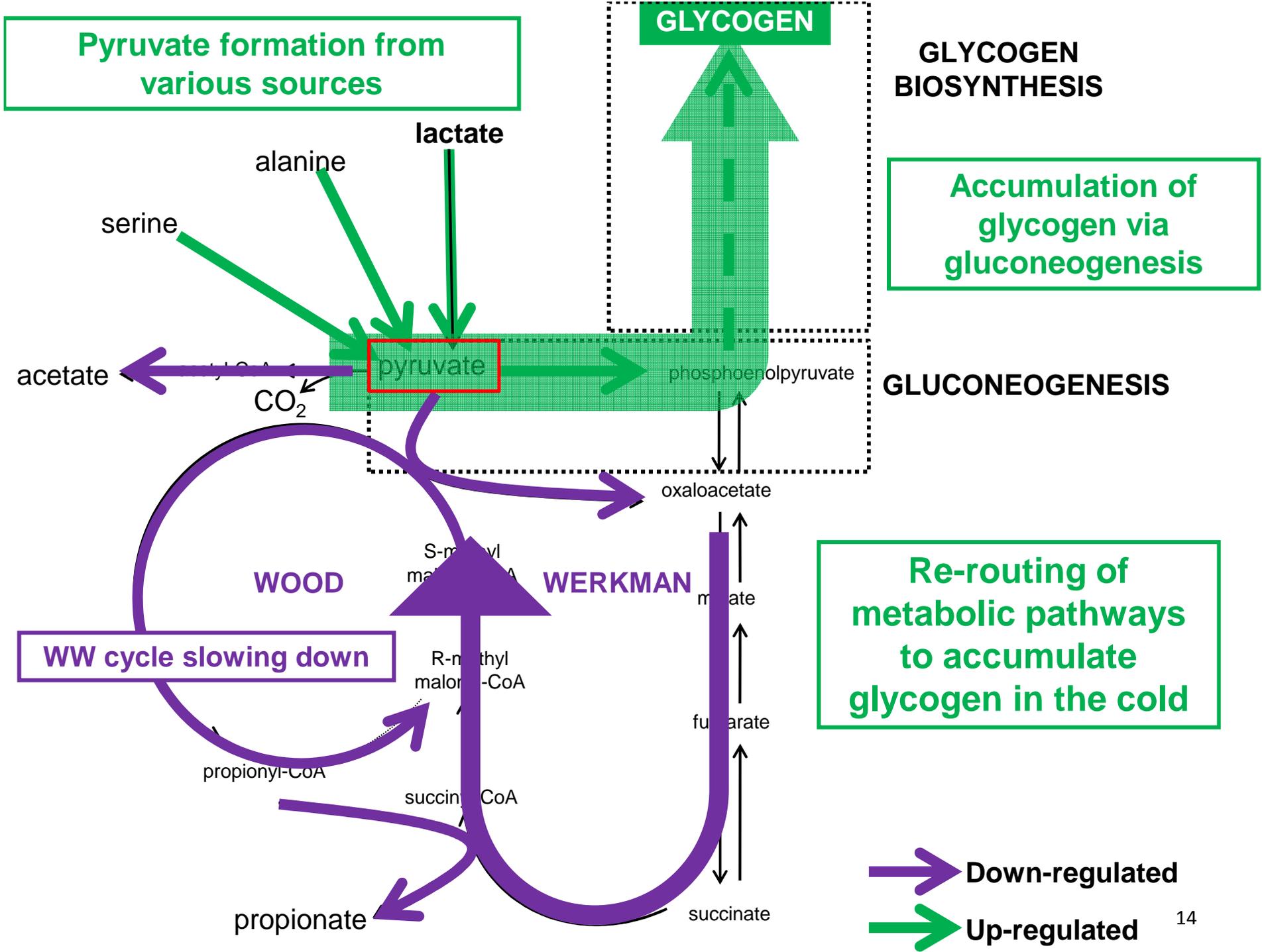
✓ These results were also confirmed by proteomic data





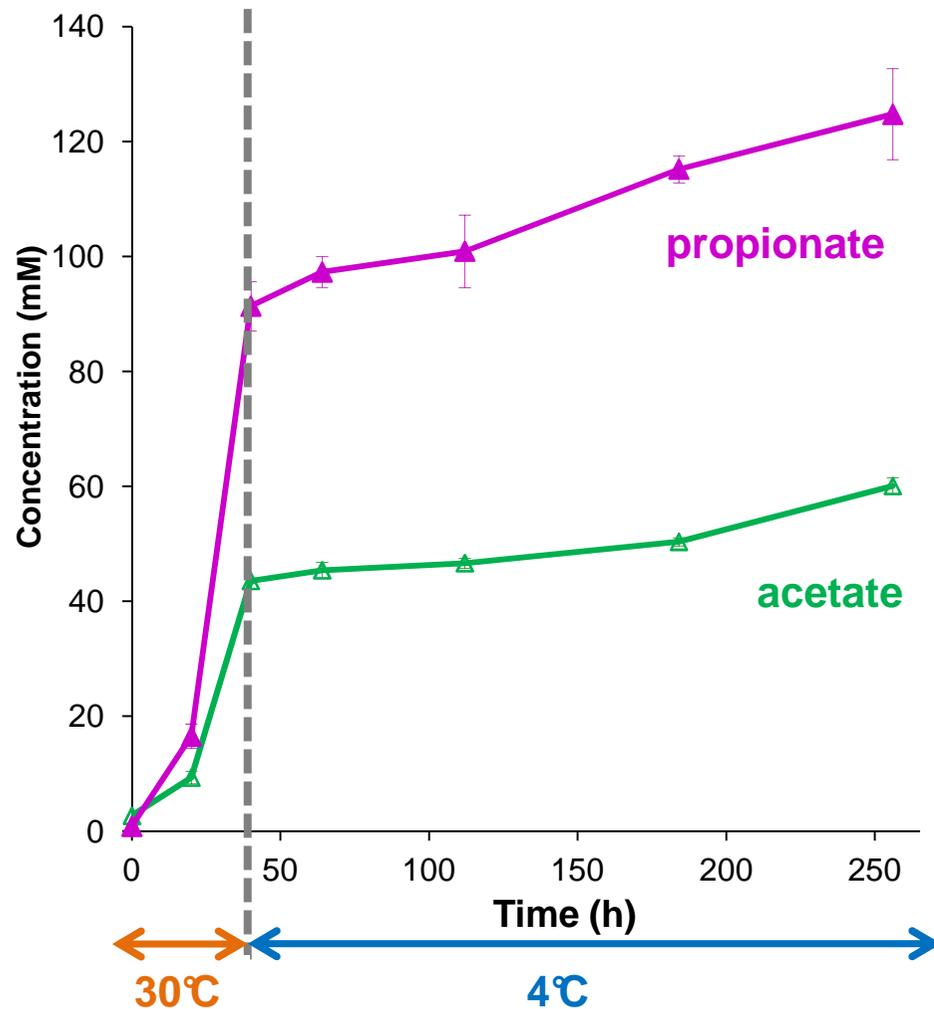
RESULTS: Long-term survival strategies





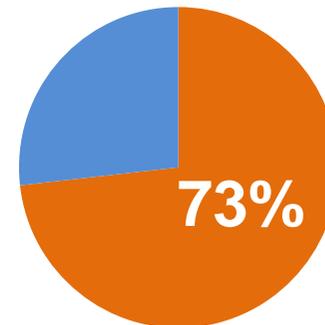


RESULTS: Production of aroma compounds



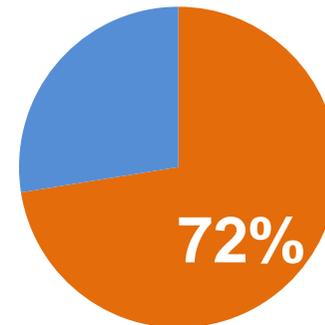
Propionate production decreased by a factor of 25 at 4°C

■ At 30°C
■ At 4°C



Acetate production decreased by a factor of 29 at 4°C

■ At 30°C
■ At 4°C



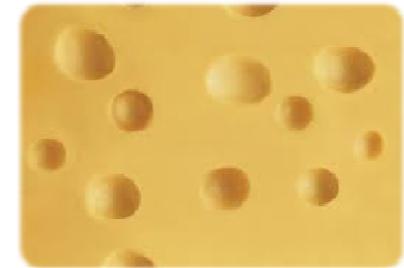


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RESULTS: Production of aroma compounds

P. freudenreichii is also known to produce:



- Free fatty acids and esters
- Methylbutanoic acids

The expression of these genes remained unchanged at 4°C

F1

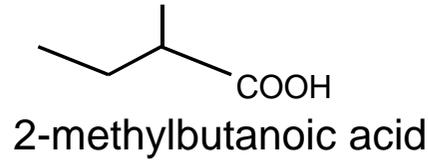


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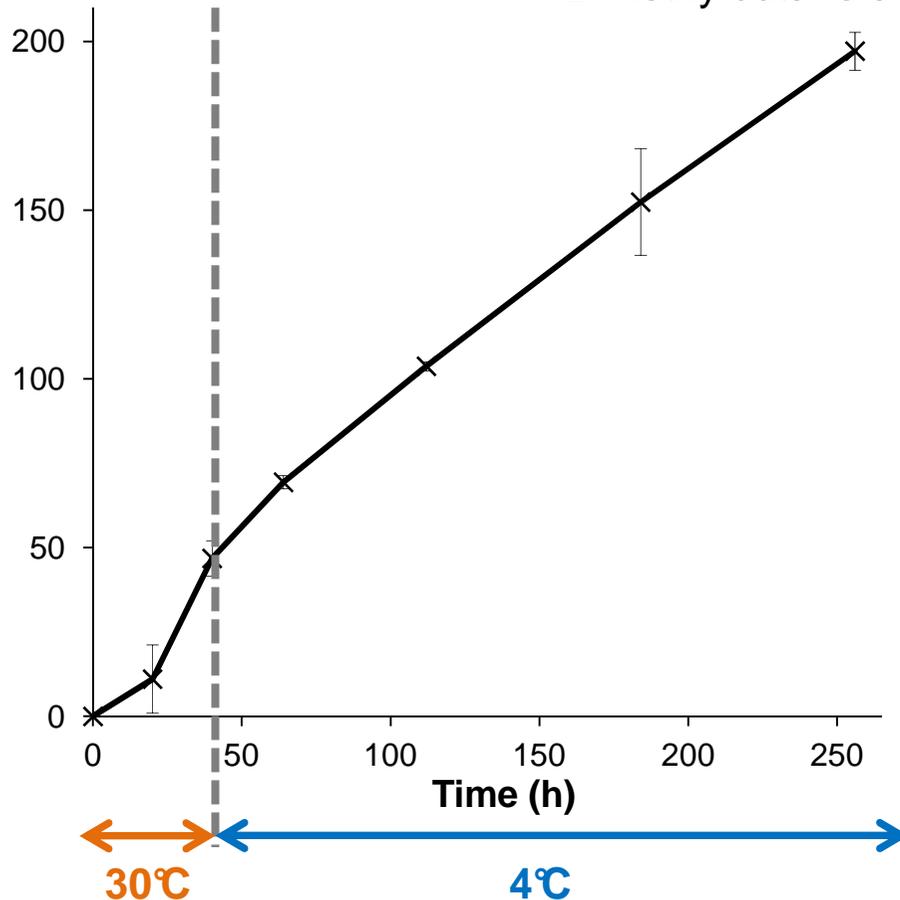
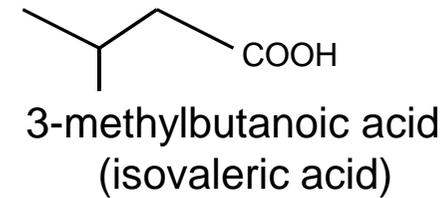


RESULTS: Production of aroma compounds

Methylbutanoic acids (μM)

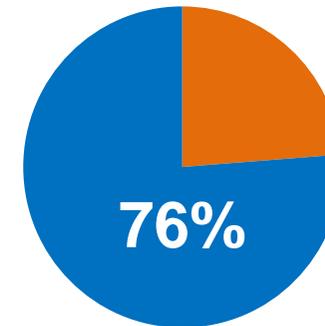


+



The production of methylbutanoic acids decreased by a factor of only 3 at 4°C

■ At 30°C
■ At 4°C



→ *P. freudenreichii* continued to produce aroma compounds at 4°C

Diapositive 17

F1 Echelle de temps : 20h et 3, 6 et 9 days ?

Couleurs ? Pas de bleu et de vert ?

Fabien, 11/10/2011



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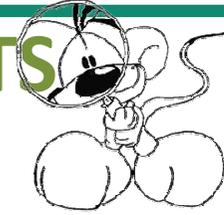


CONCLUSIONS

- ❖ This is the first transcriptomic study for a *P. freudenreichii* strain
- ❖ It brings new insights into the physiology of this bacterium in the cold
 - ✓ Its metabolic activity was slowed down
 - ✓ Its carbon metabolism was re-routed to accumulate glycogen
 - ✓ Its production of aroma compounds remained unchanged
- ❖ These results could explain the contribution of *P. freudenreichii* to cheese ripening even during cold storage

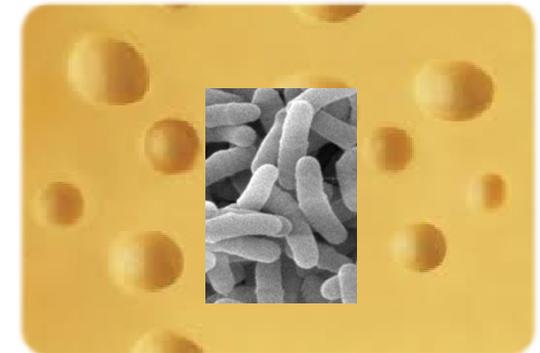


PROSPECTS



- ❖ Other *P. freudenreichii* strains are currently tested using the same -omic approach
- ❖ This work will help in getting global insights into the physiology and the metabolic adaptation of the species during cheese ripening

➔ This work could lead to a better selection of strains and an optimization of Swiss cheese ripening





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ACKNOWLEDGMENTS



FRANCE

FIL-IDF France
Grant 2011



INRA-AGROCAMPUS OUEST (Rennes, France)
UMR1253 *Science and Technology of Milk and Eggs* research unit

“Bacterial Biodiversity and *In Situ* Interactions” team

Thank you for your attention !



Valio Ltd (Helsinki, Finland)
Tekes (the Finish Funding
Agency for Technology and
Innovation)



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