Screening of Lactic acid bacteria to produce sustainable fermented whey-based drinks

Marine Penland, Marie-Bernadette Maillard, Fabienne Garnier-Lambrouin, Sandrine Parayre, Nadine Leconte, Florence Valence, Geneviève Gésan-Guiziou, Stéphanie-Marie Deutsch

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Marine PENLAND[1], Marie-Bernadette MAILLARD[1], Fabienne LAMBROUIN[1], Sandrine PARAYRE[1], Nadine LECONTE[1], Olivier COUSIN[2], Florence VALENCE[1], Geneviève GÉSAN-GUIZIOU[1] and Stéphanie-Marie DEUTSCH[1]

[1] *STLO, INRAE, Institut Agro, Rennes, France* – [2] *SODIAAL International*

CONTEXT & OBJECTIVE

Little known by consumers, cheese whey is the watery part obtained after milk curdling in cheese-making. With up to 9 L obtained for 1 kg of cheese produced, whey constitutes a major by-product of dairy industry. It mostly contains water but also lactose, minerals, vitamins (B6, B1, B12) and high-quality proteins. Despite these obvious nutritional qualities, the sour and salty taste of whey makes it not appealing to the consumer and restricts its direct consumption or use in food. For this reason, whey has long been poorly valorized and mainly used as feedstock for pigs or discarded. Nowadays, valorization rely on whey protein extraction/purification by heavy industrial process to obtain value-added food ingredients. However, these processes are only applicable to some cheese wheys, are energy-consuming and mostly concern large-scale dairy plants leaving small ones out of the system. As part of FAIRCHAIN project, we aimed at developing whey-based drinks**,** as alternative whey valorization, using microbial fermentation to preserve whey while also improving its native sensory properties.

This study explores the ability of Lactic Acid Bacteria (LAB) to ferment and acidify different types of whey and obtain the best aroma profiles for the drinks.

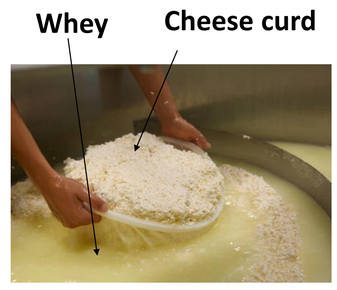


Figure 1. Separation of curd (coagulated milk) and whey during artisanal cheese-making.

EXPERIMENTAL DESIGN

Two types of cheese whey

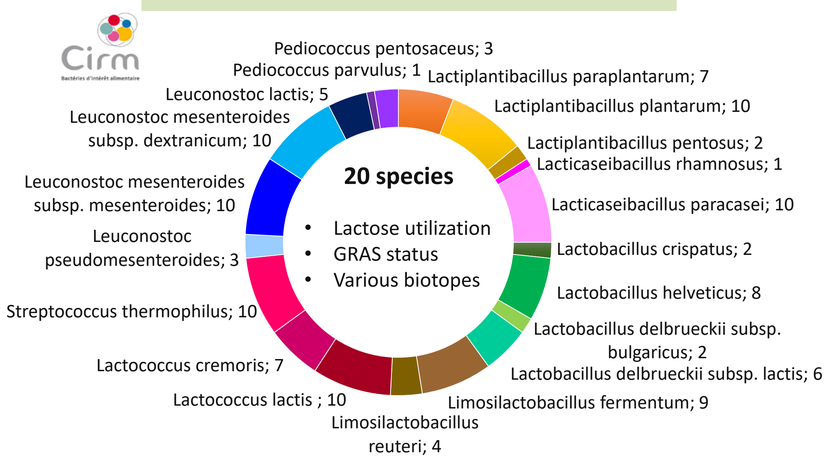
Collected from one dairy-plant

Heat-treated to eliminate native microbial flora

Comté cheese Acid whey
pH = 4.75
[lactose] = 45 g/L
[lactate] = 4.4 g/L
[proteins] = 6.4 g/L
Sensory descriptors: « salty », « cheesy »

Morbier cheese Sweet whey
pH = 6.25
[lactose] = 41 g/L
[lactate] = 0.41 g/L
[proteins] = 5.1 g/L
Sensory descriptor: « cardboard flavour »

Pre-selection of 120 LAB strains



Fermentation assays

1 LAB strain + 1 whey type **240 combinaisons tested**

Inoculation rate = 1 ‰
Duration = 24 h
T° = strain optimum

Fermentation → Selection

Assessment criteria:
- Acidification properties
- Sensory properties « Sniffing/tasting sessions »

Best combinaisons → **Biochemical profile analyses of the fermented wheys** (HPLC, IC and GC-MS)

RESULTS

Are LAB strains able to ferment acid and sweet wheys?

Acidification properties of the 120 candidates LAB strains

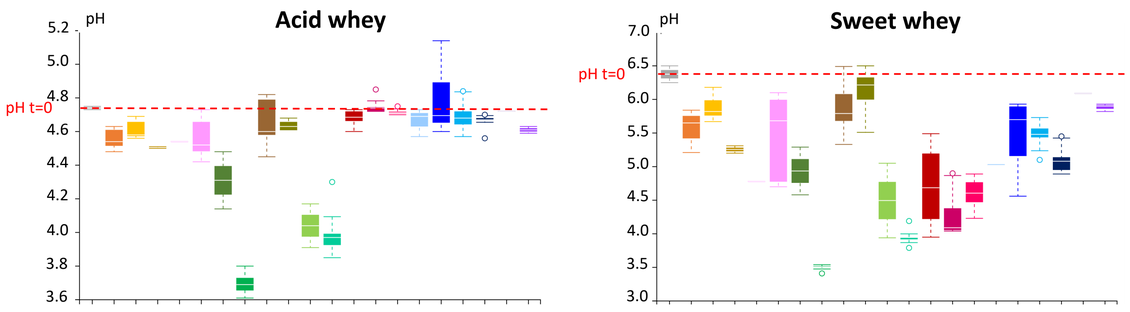


Figure 2. pH values reached by LAB strains by species in Acid whey and Sweet whey after 24h fermentation. Red dotted lines indicate the initial pH value before fermentation.

- Acidification is whey-dependent: most species acidified sweet whey with a pH decrease of up to 2.75 U, while 16 species showed a decrease inferior to 0.5 U in acid whey.
- *Lb. helveticus* (- 1.1 U pH) and *Lb. delbrueckii* (- 0.7 U pH) species were the most effective in acidifying acid whey.
- In sweet whey, highest acidifications were observed with *L. helveticus, Lb. delbrueckii*, *Lc. cremoris* and *S. thermophilus.*

Sensory properties of the wheys fermented with the 120 LAB strains

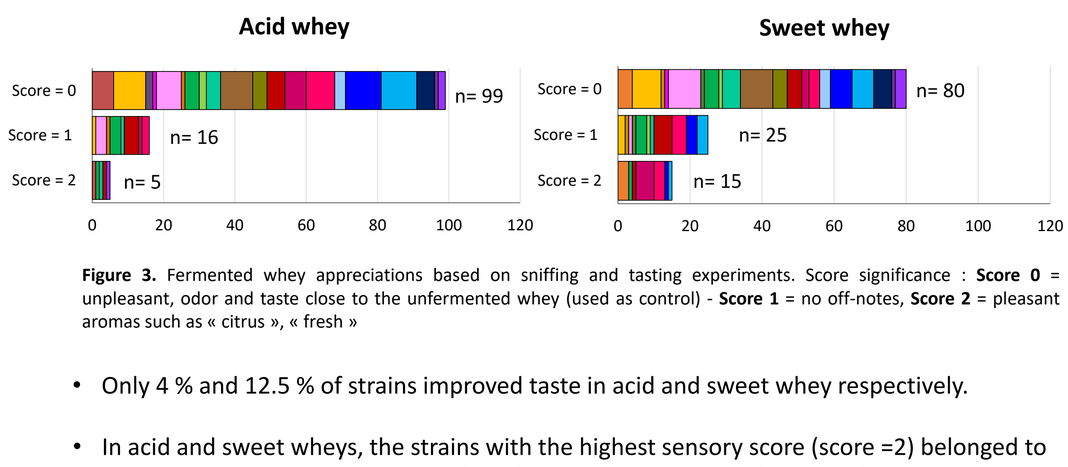


Figure 3. Fermented whey appreciations based on sniffing and tasting experiments. Score significance : **Score 0** = unpleasant, odor and taste close to the unfermented whey (used as control) - **Score 1** = no off-notes, **Score 2** = pleasant aromas such as « citrus », « fresh »

- Only 4 % and 12.5 % of strains improved taste in acid and sweet whey respectively.
- In acid and sweet wheys, the strains with the highest sensory score (score =2) belonged to 5 and 7 species respectively, revealing that sensory profiles of fermented whey are strain-dependent.

Which strains are the best candidates for each whey fermentation?

Selection of LAB strains based on their acidification properties and the fermented whey sensory profiles

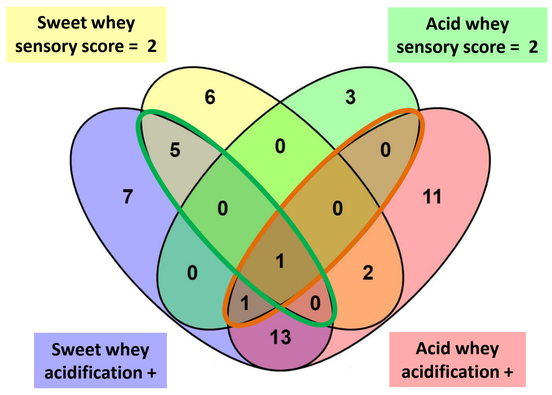


Figure 4. Venn diagram representing the repartition of the strains with the best acidification and sensory properties in acid and sweet whey. Categories: "acidification +" = strains for which fermented whey pH ≤ 4.5 ; "sensory score = 2" = strains bringing pleasant aromas such as « citrus », « fresh » notes during fermentation.

- **6 LAB strains in Sweet whey:**

Lc. lactis strain	LcLac-24
Lc. cremoris strains	LcCre-1
	LcCre-24
	LcCre-9
S. thermophilus strain	StTh-10
Lb. helveticus strain	LbHel-1

- **2 LAB strains in Acid whey:**

Lb. helveticus strain	LbHel-1
Lb. delbrueckii susp. *lactis* strain	LbLac-2

→ One strain (LbHel-1) is common to both whey types

What is the biochemical profile of the fermented wheys?

Lactose and lactic acid concentrations

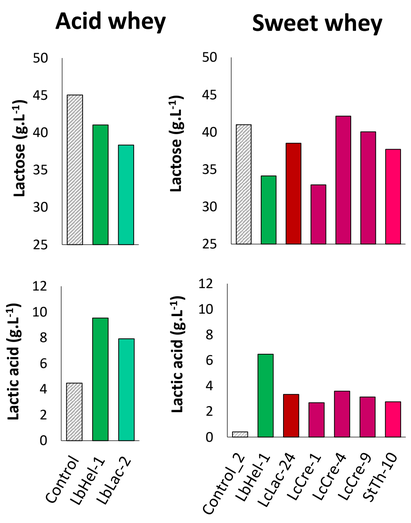


Figure 5. Lactose and lactic acid quantification in acid or sweet whey fermented with the selected strains. Quantification of lactose was performed by Ion Chromatography while lactic acid was quantified using HPLC.

- In acid whey, lactose consumption was higher with LbLac-2 (decrease of 8 g/L).
- Sweet whey fermentation: lactose consumption was lower than acid whey and strain-dependent. LbHel-1 and LcCre-1 were the largest lactose consumers (up to 7g/L).
- In both wheys, highest lactic acid production was observed with LbHel-1: 6 g/L in sweet whey and 5 g/L in acid whey.

Volatile compound profiles

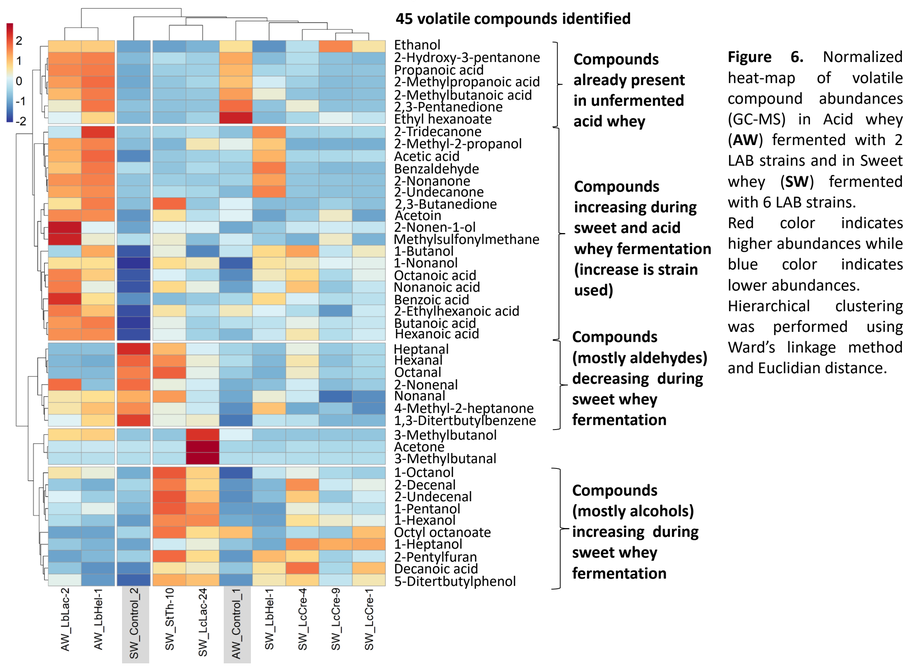


Figure 6. Normalized heat-map of volatile compound abundances (GC-MS) in Acid whey (**AW**) fermented with 2 LAB strains and in Sweet whey (**SW**) fermented with 6 LAB strains. Red color indicates higher abundances while blue color indicates lower abundances. Hierarchical clustering was performed using Ward's linkage method and Euclidian distance.

- For each whey type, fermented wheys with the different strains highly differed from unfermented ones.
- Acid whey fermentation by LbLac-2 and LbHel-1 led to similar profiles with higher abundances of organic acids and ketones.
- Sweet whey fermentation with the 6 strains revealed different profiles even within the same species (*Lc. cremoris*). Most strains degraded aldehydes such as heptanal, nonanal known to be responsible for «cardboard » off-flavour.

CONCLUSION AND PERSPECTIVES

- Lactic Acid Bacteria are able to ferment both acid and sweet whey. However, fermentation outcomes are strain-dependent and due to its low initial pH, fewer strains satisfyingly acidified acid whey.
- Two and six LAB strains were identified as good candidates for drink development in acid and sweet whey, respectively. One *Lb. helveticus* strain, namely LbHel-1, showed promising results on both types of whey.
- Biochemical analyses of the fermented wheys revealed that fermentation considerably changed sugar and aroma profiles. Adequate strain selection thus allowed to obtain a fermented whey with acceptable sensory profile. Aromatization experiments are currently carried out to further improve the taste of the fermented drinks.

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Marine.Penland@inrae.fr
Stephanie.Deutsch.@inrae.fr
fairchain-h2020.eu