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Microbial diversity of Pélardon, an artisanal raw goat milk cheese

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Context

Pélardon is a Protection Designation of Origin (PDO) French artisanal soft rind cheese, produced in mountainous areas in southern France. It is exclusively produced from raw goat's milk using a backslopping method for inoculation. Its manufacture and unique organoleptic properties rely on the autochthonous microbiota and their activities. The aim of this work is thus to study Pélardon microbial (fungal & bacterial) diversity and dynamics during cheese-making and ripening by culture-dependent and -independent approaches.

Strategy

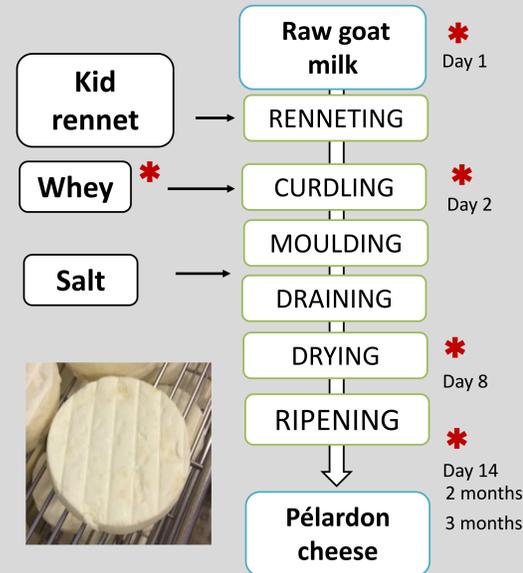
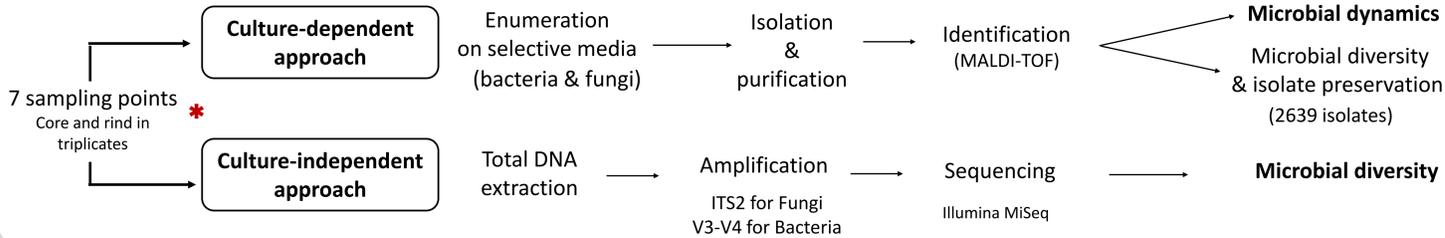


Figure 1. Pélardon cheese-making process. Red stars correspond to the sampling points.

Culture-dependent approach

Microbial diversity and dynamics monitored during Pélardon cheese-making process in both rind and core

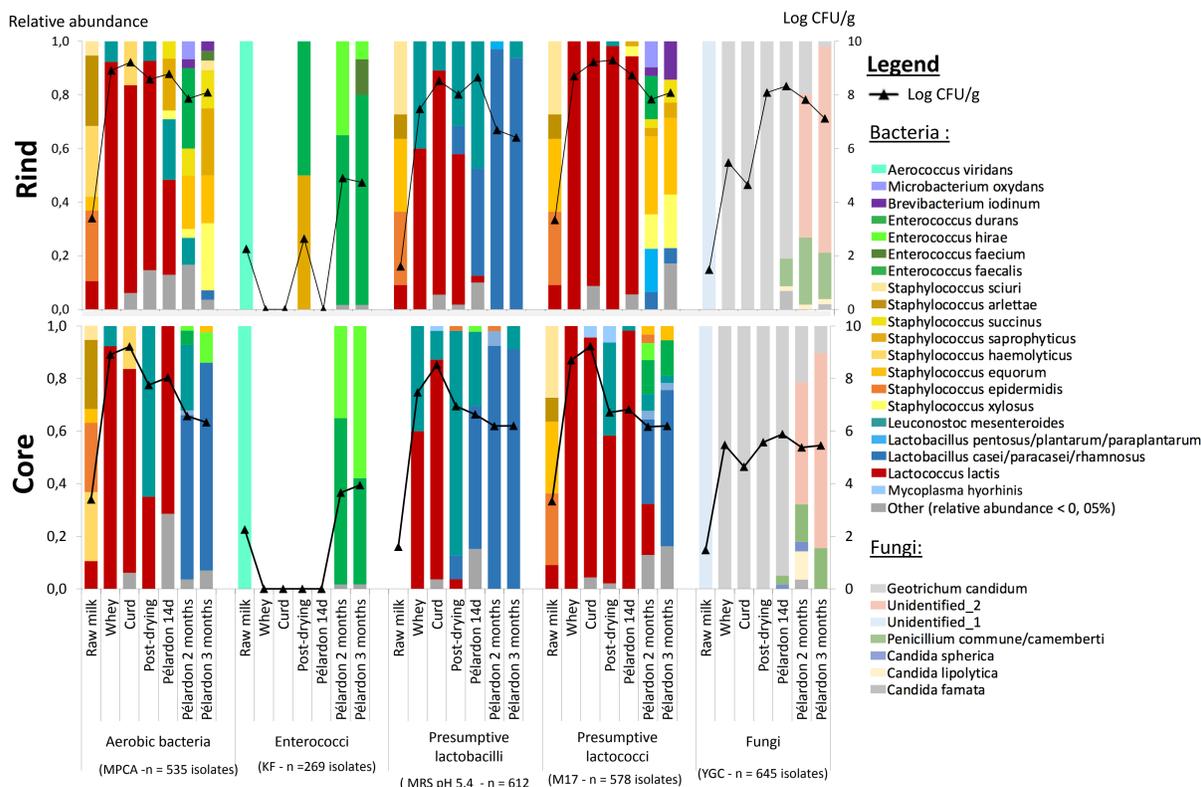


Figure 2. Microbial counts in log CFU/g and diversity encountered on different media. Diversity data and isolate identifications were obtained via MALDI-TOF analysis against a commercial and internal database (identification for each isolate was performed in duplicate)

- Bacterial populations were higher in the rind compared to those in the core although the same trends were observed. The curdling step led to a high increase of all populations (except for enterococci), followed by a progressive decrease until the end of the ripening.
- Lactococcus lactis* dominated during acidification and drying in both core and rind and was mainly brought by the whey used for backslopping. After prolonged ripening, *Lactobacillus* and *Leuconostoc* became dominant in the core whereas *Corynebacteria* and *Staphylococcus* were dominant in the rind.
- Fungal populations were low in raw milk compared to the whey used for backslopping. Regarding diversity, *Geotrichum candidum* was dominant although other species such as *Penicillium* spp. were found in late ripening stages.

Culture-independent approach

Preliminary metagenetic analyses & comparison with microbial counts on curd and 3-month ripened cheese from a previous batch

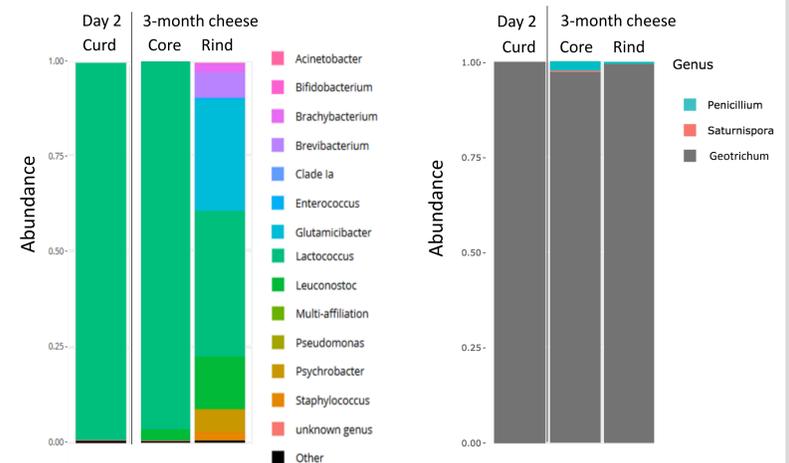


Figure 3. Bacterial diversity in the samples at the genus level. Data were analysed using FROGS pipeline⁽¹⁾ and Silva132 database.

Figure 4. Fungal diversity in the samples at the genus level. Data were analysed using FROGS ITSx pipeline⁽¹⁾ and the UNITE_Fungi database

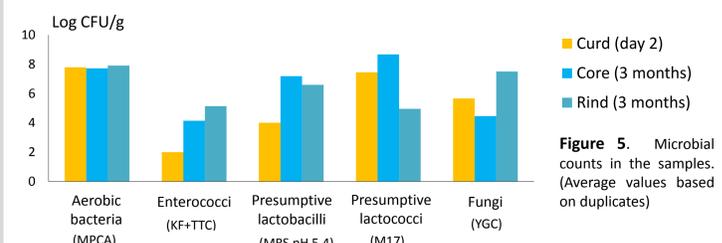


Figure 5. Microbial counts in the samples. (Average values based on duplicates)

- Aerobic bacterial counts were similar in all samples but dynamics in other populations were observed. Lactococci decrease was concomitant with an increase in lactobacilli and enterococci populations.
- Lactococcus lactis* dominance was confirmed by metagenetics. Surprisingly, lactobacilli relative abundance was low despite an increase in their populations in the 3-month cheese.
- Bacterial diversity was higher in rind with up to 6 genera, mainly *Corynebacteria* and Gram-negative bacteria.
- Fungal populations were higher in the rind of the ripened cheese and dominated by *Geotrichum* after curdling step.

Conclusion

Pélardon cheese production was monitored using culture-dependent and -independent methods. Culture-dependent results, based on the analysis of more than 2000 isolates, highlighted the importance of whey as a source of bacteria and fungi. Results also shed light on the dynamics during the process. Diversity increased during ripening for fungi and bacteria and metagenetic analyses corroborated with the previous results. *Lactococcus lactis* was shown to be a key player of the microbial ecosystem. To have a better understanding of the dynamics, metagenetic analyses will be performed for all sampling points. Moreover, volatiles, sugars and organic acids will be monitored at the different process steps in order to link the microbial diversity and dynamics with cheese biochemical changes.

References

(1) Frédéric Escudié, Lucas Auer, Maria Bernard, Mahendra Mariadassou, Laurent Cauquil, Katia Vidal, Sarah Maman, Guillermina Hernandez-Raquet, Sylvie Combes, Géraldine Pascal; FROGS: Find, Rapidly, OTUs with Galaxy Solution, *Bioinformatics*, Volume 34, Issue 8, 15 April 2018, Pages 1287–1294

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