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Agroscope

Influence of the microbiome on the quality and safety of Raclette du Valais AOP cheese: Current findings and a look back at the last 150 years

prepared by Hans-Peter Bachmann (Agroscope)
presented by Monika Lüscher Bertocco (Grangeneuve)

12th European Farmhouse and Artisan Cheese & Dairy Meeting in Bohinj, Slovenia
3rd October 2022

To start with, Hans-Peter's favourite citation about artisan cheese making

«The challenge for the cheesemaker is to strike the right **balance between art and science**.»

The complete citation and the source reference for further reading

"The challenge for the farm-stead cheesemaker is to strike the right **balance between art and science**. The goal should be to achieve the appropriate level of control to ensure safety and consistently high quality while at the same time giving nature enough free rein to encourage the diversity and uniqueness of character that make artisanal cheeses special."

HEATHER PAXSON Massachusetts Institute of Technology

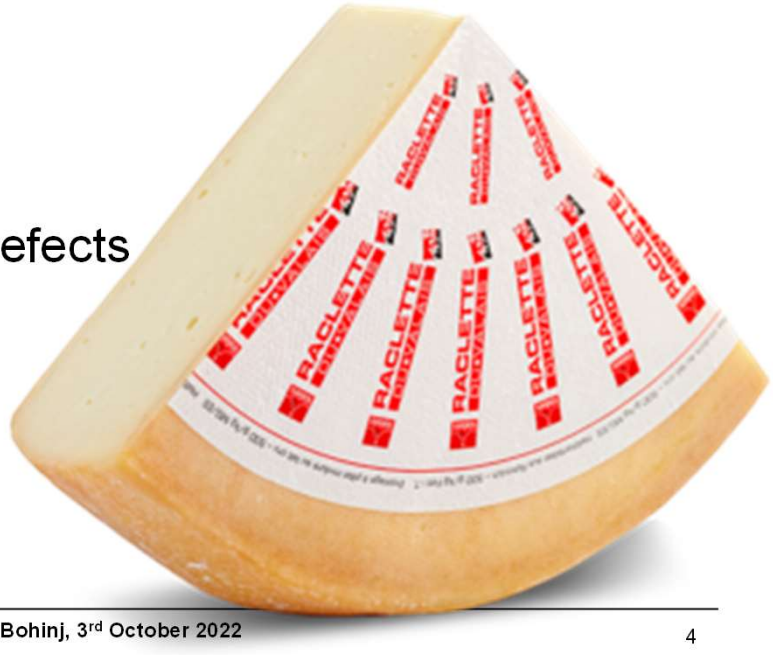
POST-PASTEURIAN CULTURES: The Microbiopolitics of Raw-Milk Cheese in the United States

CULTURAL ANTHROPOLOGY,Vol. 23, Issue 1, pp. 15–47. ISSN 0886-7356, online ISSN 1548-1360. C2008 by the American Anthropological Association. All rights reserved. DOI: 10.1525/can.2008.23.1.15.

https://anthrosource.onlinelibrary.wiley.com/doi/abs/10.1111/j.1548-1360.2008.00002.x

Four aspects will be highlighted

1. Introducing the cheese variety Raclette du Valais AOP
   - History
   - Production areas (lowlands, mountains, alpine pastures)
   - Technology
2. New insights into the microbiota
   - Microbial biodiversity within the cheese and on its surface
   - Discover unknown causes of known cheese defects
3. Historical context: looking back on 150 years
4. Development of new AOP acidification cultures



Agroscope

Origin of the name

- Raclette is derived from the word "racler", which means "to scrape" in the local French dialect.
- The reason for scraping is that after melting over the fire, the cheese is gently scraped off the cheese wheel.


RACLETTE
DU VALAIS
AOP
L'ORIGINAL.
WALLIS

History of Raclette du Valais AOP (slide 1)

- In Valais, cheese-making is documented by archaeological findings, such as a cheese sieve, dating back to the 7th century BC.
- During Roman times, alpine cheese was famous.
- Between the 14th and 19th centuries, cheese was often used as a means of payment, remuneration for work or as an export product.

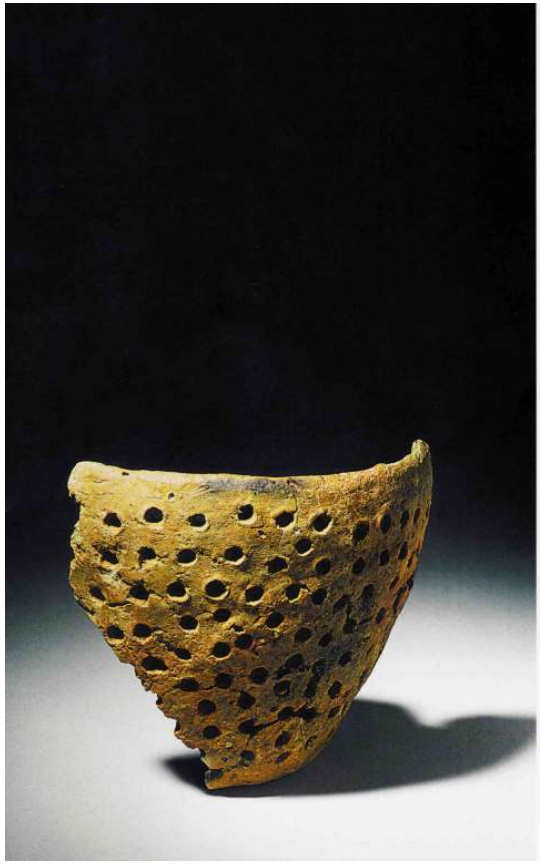


Photo: Canton Museum, J.-Y. Glassey

History of Raclette du Valais AOP (slide 2)

- The Val de Bagnes provides numerous indications of this. A semi-hard cheese was mainly produced there, Fructus or Caseus, which had great similarities with the cheese from the Aosta Valley.
- By 1574, cheese melting was already known in Valais.
- The name "Raclette" was used in written language from 1874 to designate the cheese of the same name, but was already common in local dialect earlier.
- And the history of Raclette is closely linked to a special breed of cows, the "Eringers", which are also known for their cow fights.


6
7
10

Cheese country Switzerland

How can you govern a country which has 246 varieties of cheese?

— Charles de Gaulle —

AZ QUOTES

Raclette du Valais AOP is produced exclusively in the canton of Valais

Agroscope

25 cheese dairies producing all year round in the valley and mountain regions

Raclette du Valais AOP

Käsereien im Wallis
Les fromageries en Valais

1 **«GOMSER 32»**
Käserei Reckingen, 3998 Reckingen
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urs-holzer@bluewin.ch

2 **«BIO GOMSER 11»**
Bio-Bergkäserei Goms, 3998 Gluringen
+41 27 973 20 80
info@biogomser.ch
www.biogomser.ch

3 **«GOMSER 55»**
Käserei Grengiols, 3993 Grengiols
+41 27 927 17 11
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www.landschaftspark-binntal.ch

4 **«GOMSER 1»**
Walker Bergkäserei, 3982 Bitsch
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5 **«HEIDA»**
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www.heidakaese.ch

6 **«SIMPLON»**
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7 **«WALLIS 65, BIO WALLIS 65»**
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8 **«ANNIVIERS»**
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9 **«CRANS-MONTANA»**
Fromagerie de la Ferme des Trontières,
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10 **«AYENT»**
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16 **«CHATEAUNEUF»**
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1951 Sion
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Walliser Raclette – Ein Naturprodukt aus den Walliser Bergen

Raclette du Valais AOP ist ein halbharter, vollfetter Rohmilchkäse aus Kuhmilch. Die Milchproduktion, die Verarbeitung und Reifung des Raclette du Valais AOP erfolgen ausschliesslich im Kanton Wallis. Nach drei Monaten Reifung ist der Käse für den Konsum bereit.

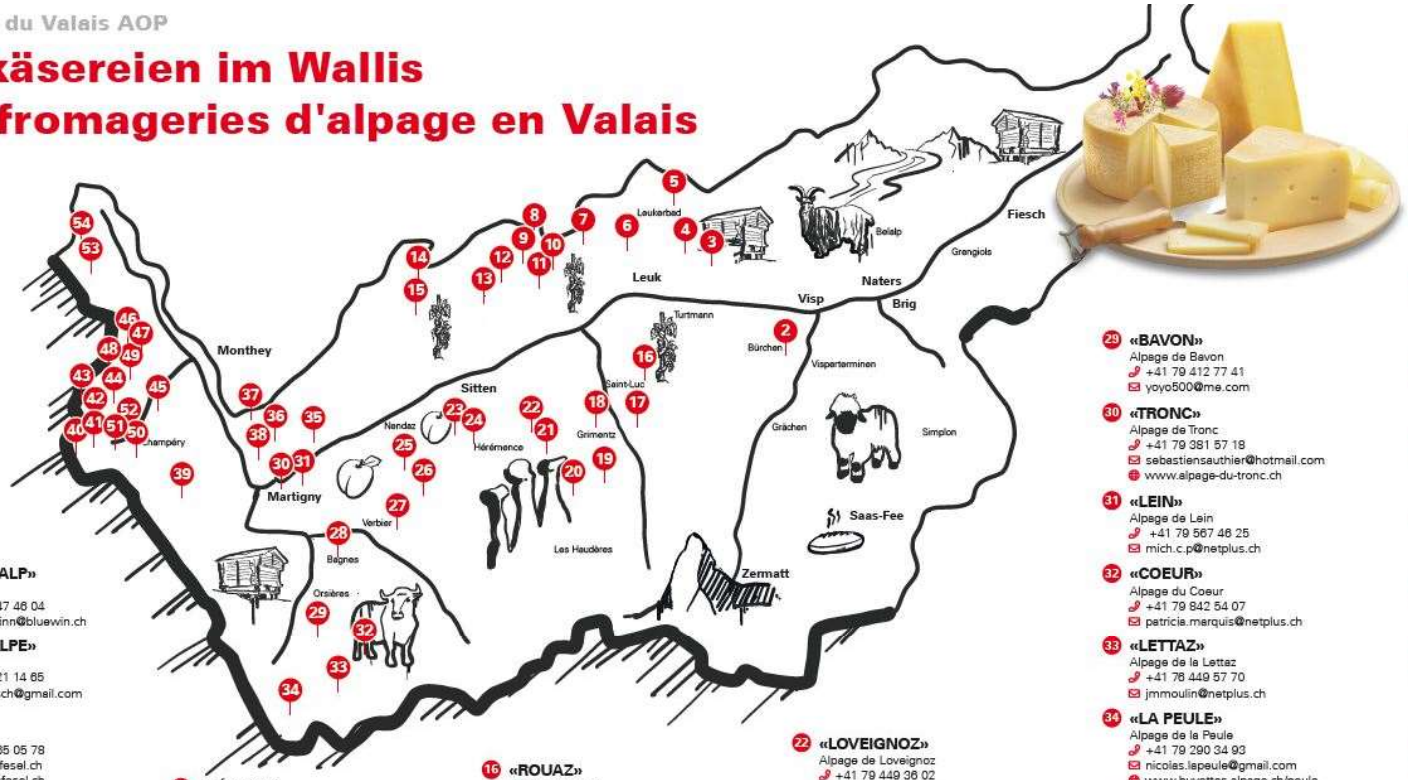
Cremig, wohlduftend, geniesserisch – Raclette du Valais AOP verführt die Feinschmecker durch seinen aussergewöhnlichen Geschmack und die Qualität. Die reichhaltige Flora der Walliser Berg- und Alpregionen, das mediterrane Klima und die traditionelle Verarbeitung verleihen dem Raclette du Valais AOP seinen einzigartigen frischen und würzigen Geschmack.

RACLETTE DU VALAIS AOP

Raclette du Valais AOP

Alpkäsereien im Wallis
Les fromageries d'alpage en Valais

54 Alpine cheese dairies that only produce in summer (alpine season)



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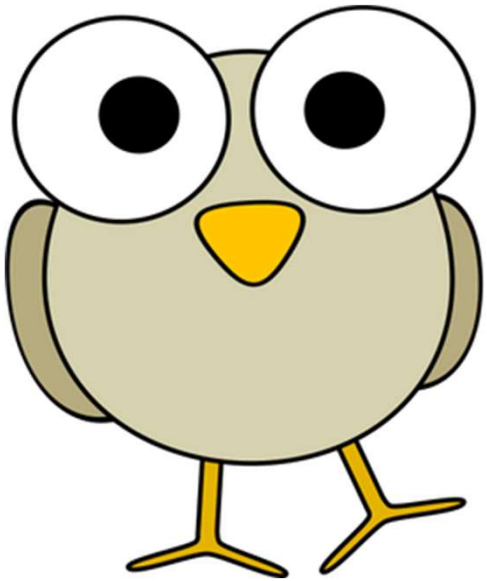
Der Name des Käses ist auf der Järbseite eingeprägt.
Nom incrustés dans le talon du fromage.

Agroscope

Mountain ?

Valley ?

Alp ?



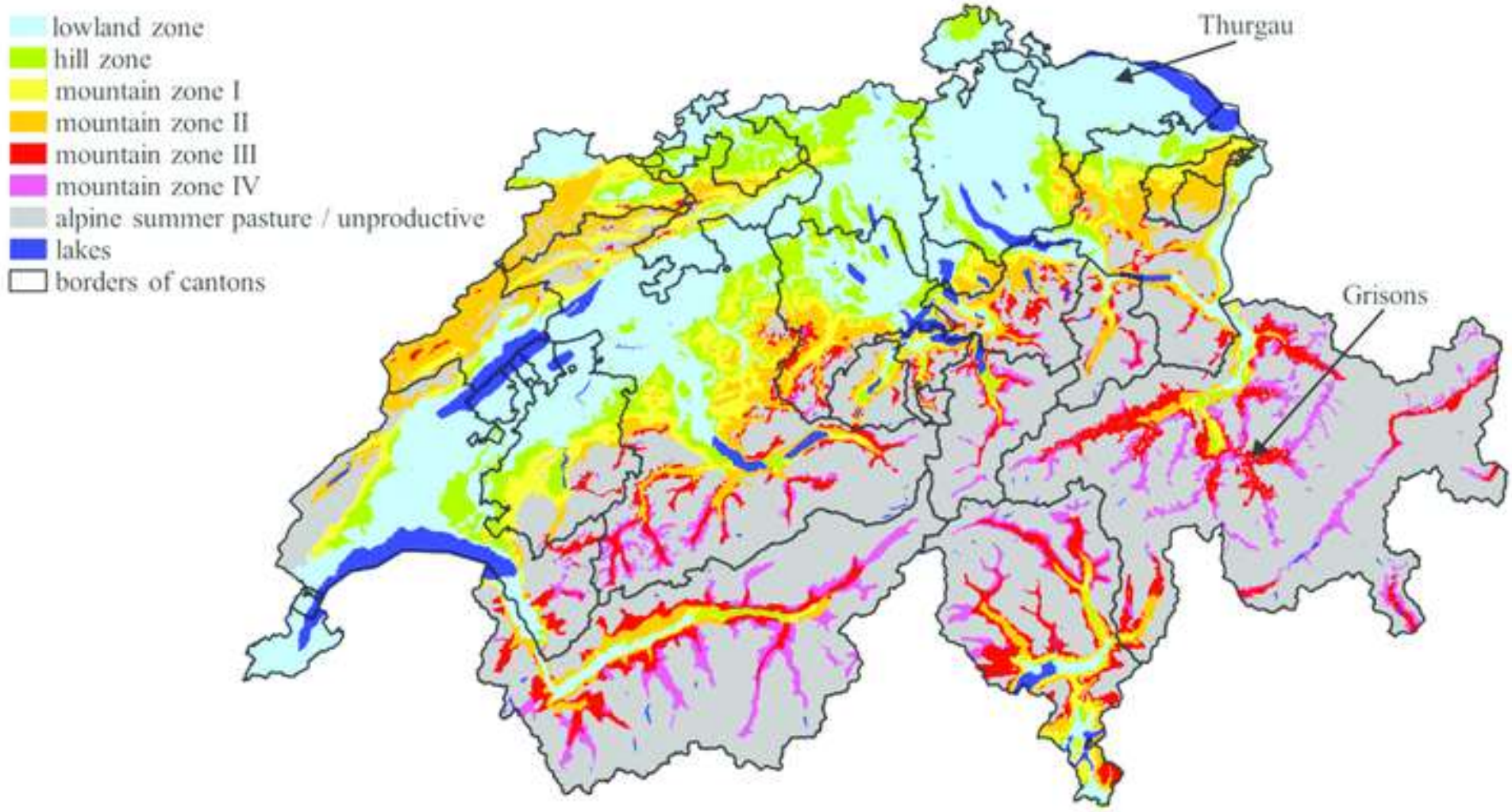
??

Hill ?

Lowland ?

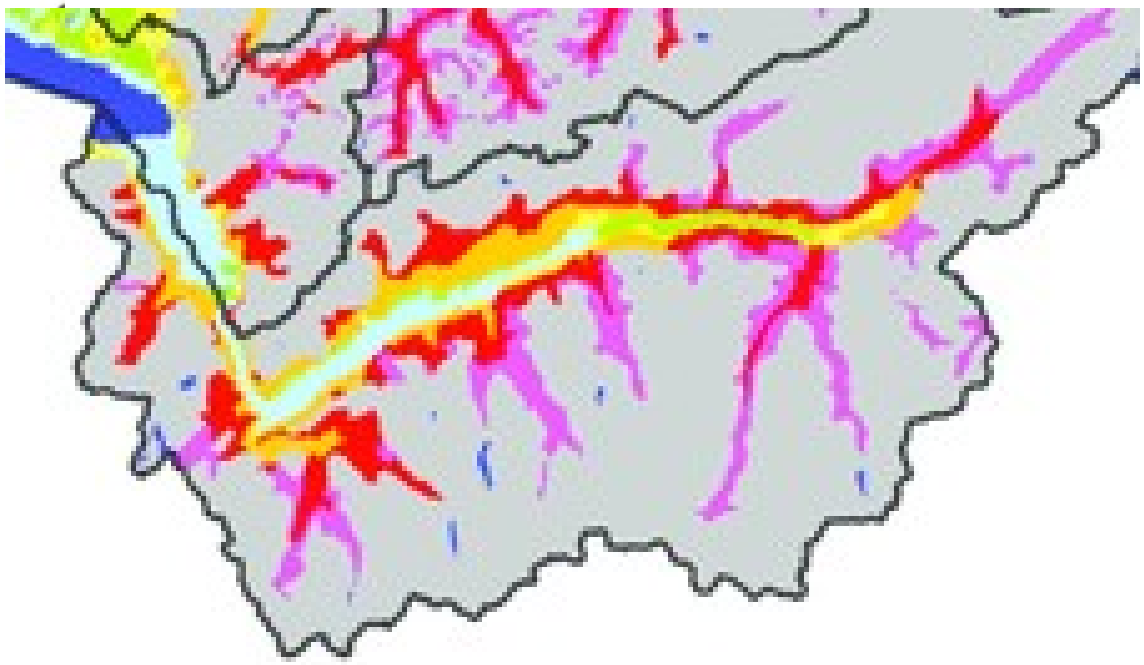
Swiss Definitions Valley - Mountain - Alp

- Altitude above sea level is not enough…
- The Agricultural Zones Ordinance determines to which agricultural zone an area is assigned.
- The following criteria - in descending order of importance - are taken into account for the classification of the mountain area:
  - the climatic situation, in particular the duration of the vegetation period;
  - the geographical situation, in particular the accessibility from the nearest village and the nearest centre;
  - the topography, in particular the proportion of slopes and steep slopes.
- Alpine pastures are not counted as agricultural land because they can only be farmed for a short period of time.
- The alpine dairies are located at the Alpine pastures. These pastures at high altitudes can only be managed in summer. For this purpose, cows are moved up to these higher alps.

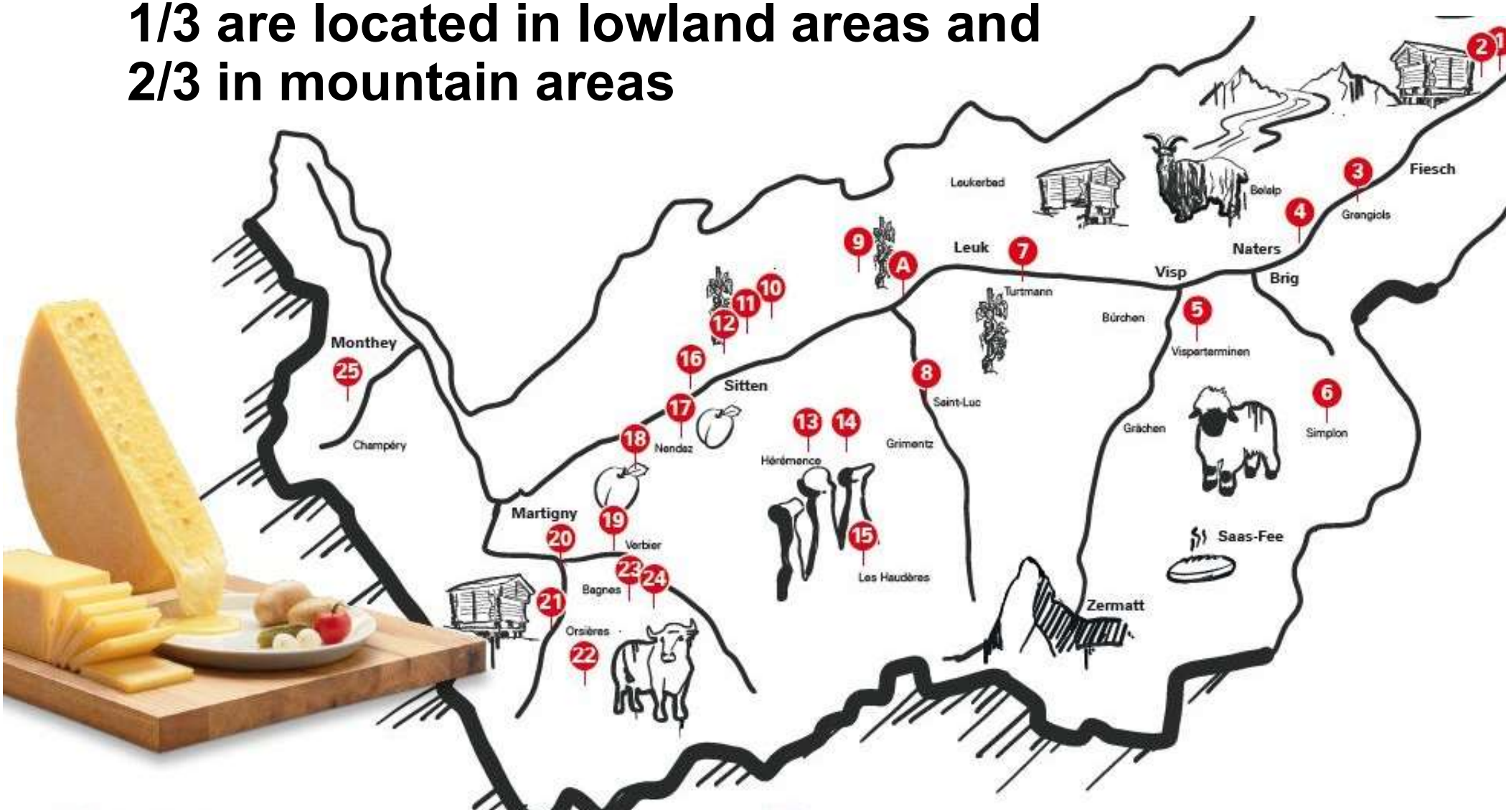

lowland zone
hill zone
mountain zone I
mountain zone II
mountain zone III
mountain zone IV
alpine summer pasture / unproductive
lakes
borders of cantons
Thurgau
Grisons

Agricultural zones in the canton of Valais

lowland zone
hill zone
mountain zone I
mountain zone II
mountain zone III
mountain zone IV
alpine summer pasture / unproductive
lakes
borders of cantons



Of the 25 cheese dairies producing all year round, 1/3 are located in lowland areas and 2/3 in mountain areas



Artisanal Technology

- Full-fat cheese, semi-hard
- Daily processing of fresh raw milk
- Starter culture with mesophilic and thermophilic strains
- Curd is heated in copper vat to 36 – 45 °C
- Important: very rapid acidification (pH below 6.0, 2 hours after filling) and complete degradation of lactose after 1 day (pH 5.0 +/- 0.2)
- 1 day brine salting
- Ripening
  - 10 - 11°C; 90 - 96 % relative humidity
  - Smearing with salt water (approx. 3 %), at the beginning daily, then 2 x weekly
  - Duration 2 – 12 months

Average composition of 120-day-old Raclette du Valais AOP cheeses (n=21)

Content	Unit	Median	Mean value	Standard deviation	Minimum	Maximum
Water	g/kg	414	415	21	377	457
Fat	g/kg	317	315	16	276	344
Water in fatfree cheese	g/kg	608	606	19	574	646
Fat in dry matter	g/kg	539	540	14	508	564
Salt (NaCl)	g/kg	19.6	19.7	1.8	16.4	24.3
Calcium	g/kg	4.9	4.9	0.4	4.2	6.0

Positive for melting properties: more water, less calcium, more intensive proteolysis (lipophilic peptides, activity of chymosin)


HALAG


RACLETTE
VALAISANNE

Four aspects will be highlighted

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   - History
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2. New insights into the microbiota
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   - Discover unknown causes of known cheese defects
3. Historical context: looking back on 150 years
4. Development of new AOP acidification cultures

Agroscope

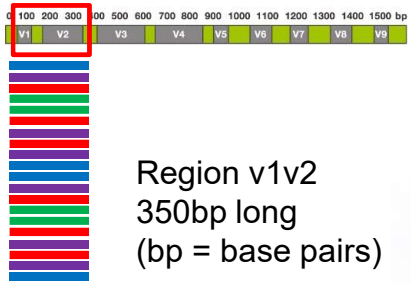
Sequencing of the 16S rRNA gene

1) Sampling and DNA extraction

Sampling of rind and cheese

Enzymatic DNA extraction

2) Amplification of the 16S ribosomal gene

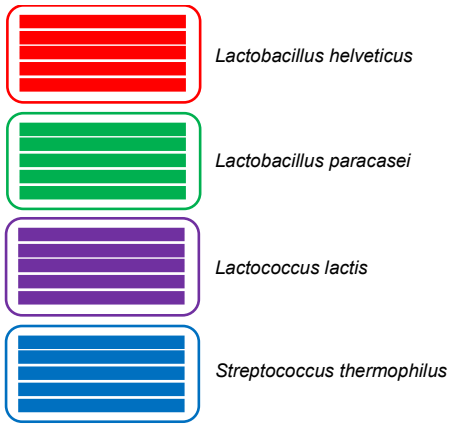


Region v1v2
350bp long
(bp = base pairs)

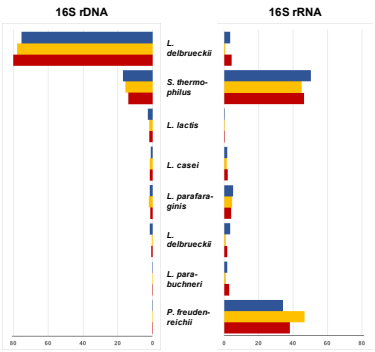
```
>OTU_1
TTGATCCTGGCTCAGGACGAACGCTGGCGGCGTGCCTAATACATGCAAGTCGAGCGAGCTGAATTCAAAGATTCCTTCGGGATGATTTGTTGG
ACGCTAGCGGCGGATGGGTGAGTAACACGTGGGCAATCTGCCCTAAGACGTGGATGACACCTTGGAAACAGGTGCTAATACCGGATAACAACA
TGAATCGCATGATTCAAGTTTGAAAGGCGGCGCAAGCTGTCACTTTAGGATGAGCCCGCGGCGCATTAGCTAGTTGGTGGGGTAAAGGCCTAC
CAAGGCAATGATGCGTAGCCGAGTTGAGAGACTGATCGGCCACATTGGGACTGAGACACGG
```

3) Sequencing of the fragments of the 16S

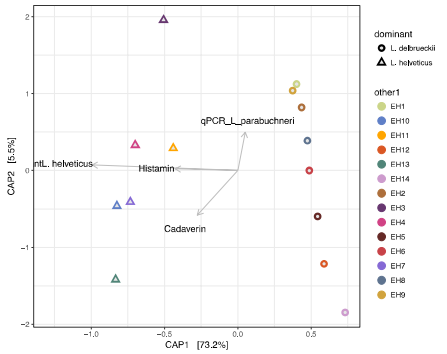
4) Bioinformatic analysis of the sequences



5) Biostatistical analysis of bacterial communities



Composition



Correlation analyses

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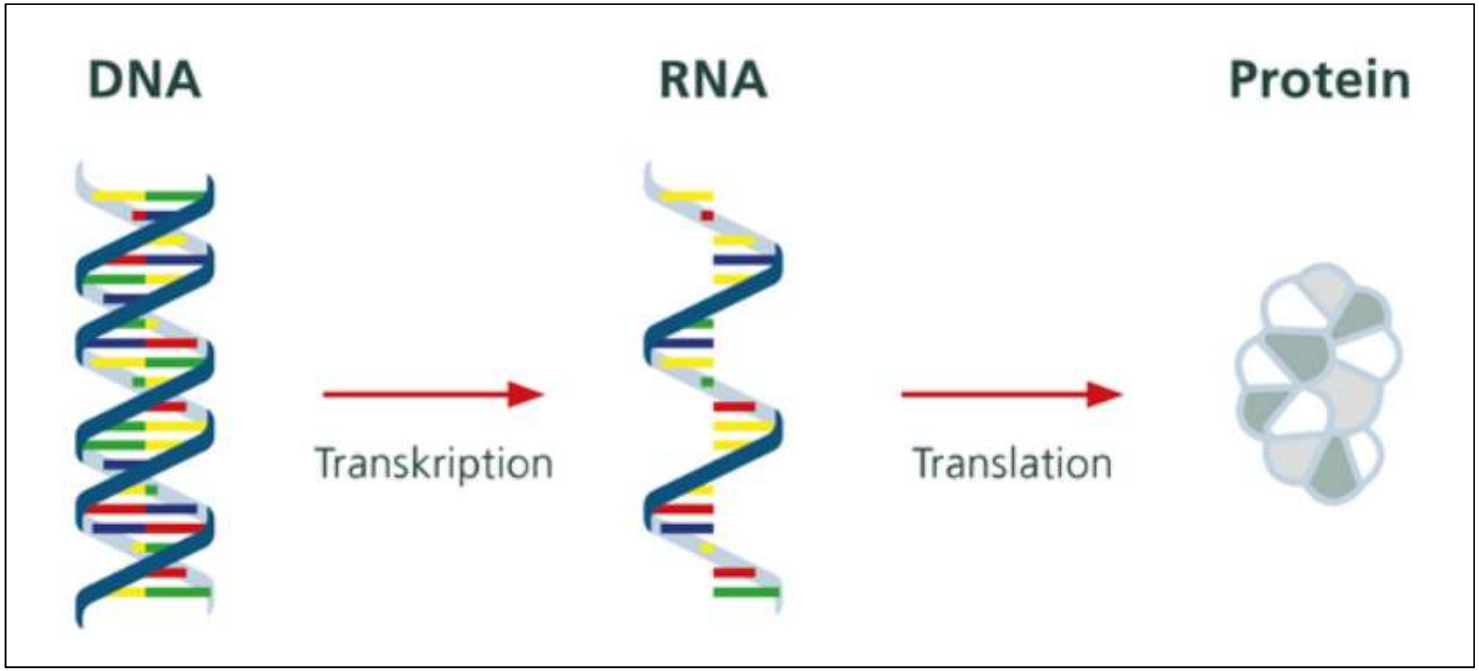
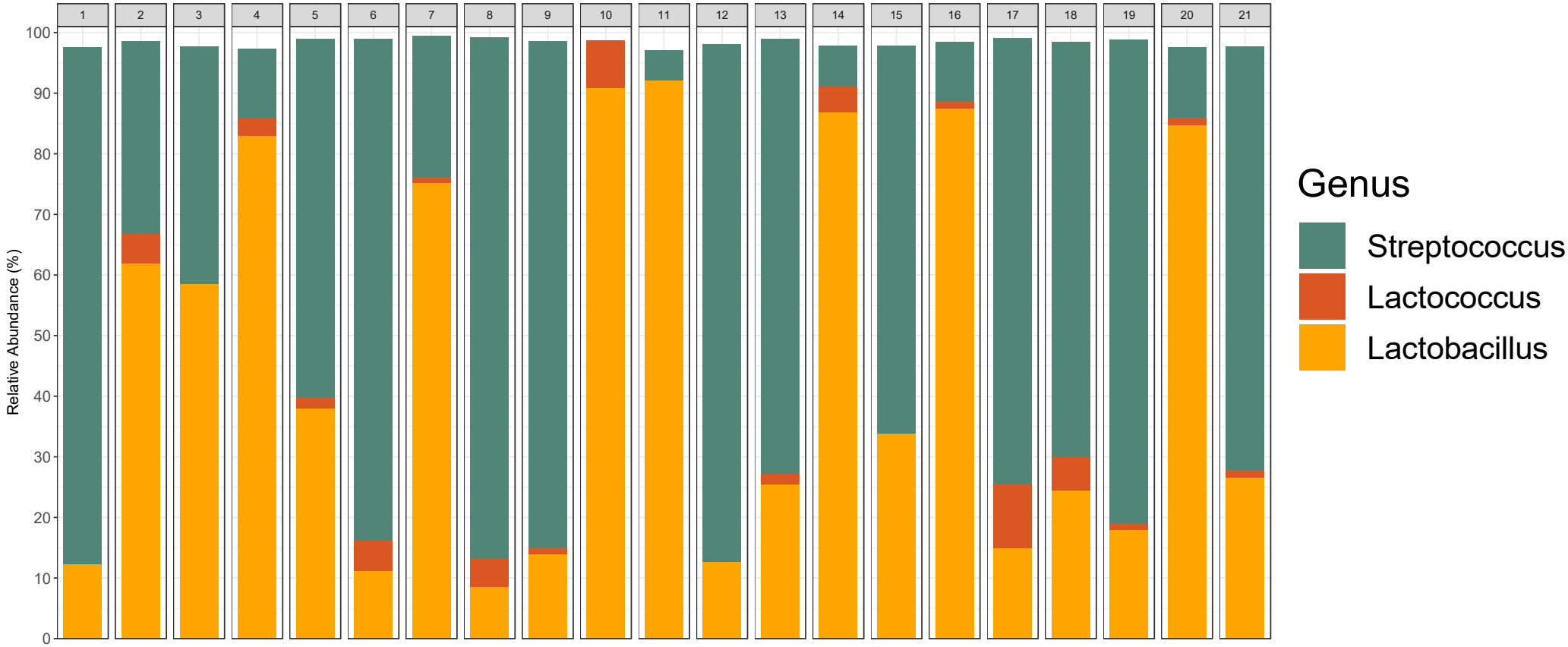
Different interpretation

- DNA (very stable) = Who is (or was) there?
- RNA (rapidly degraded) = Who is active?

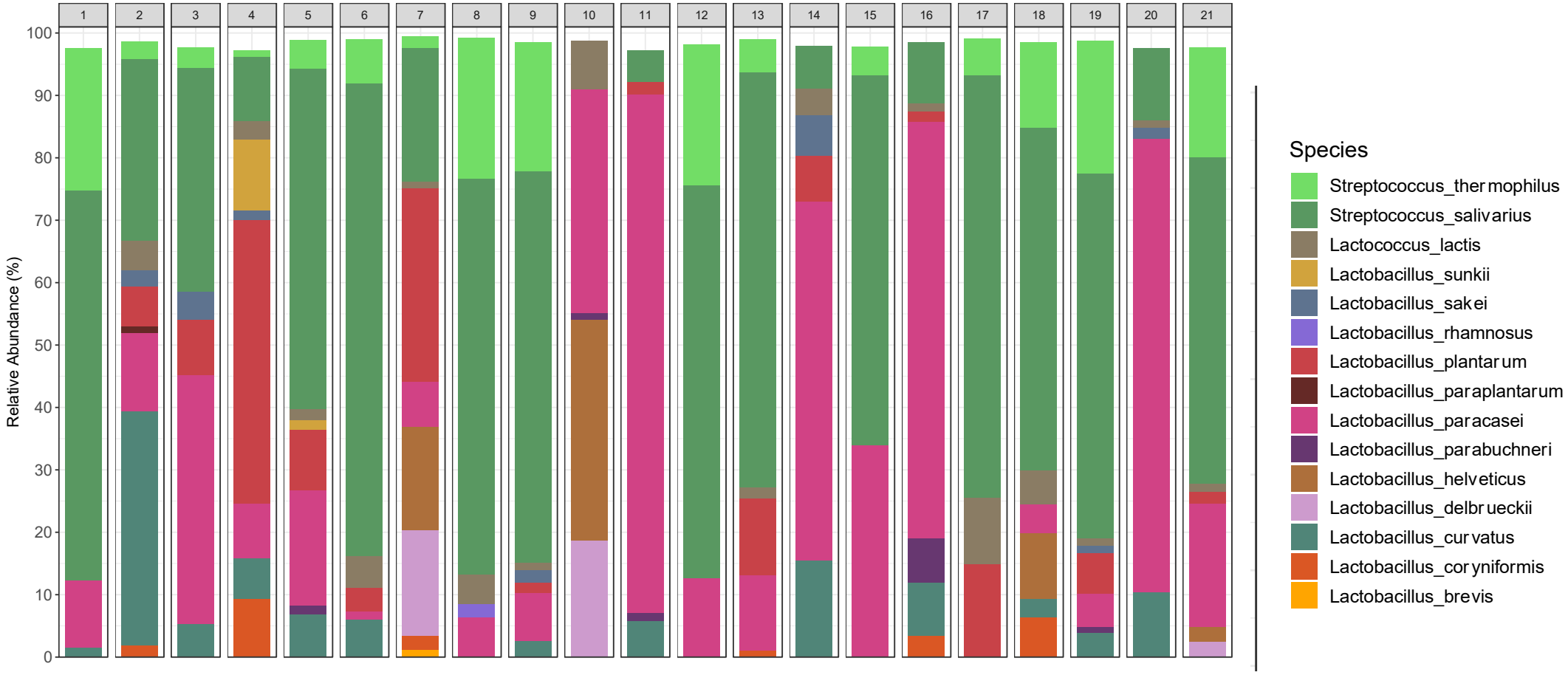


Study with cheeses from 21 different year-round cheese dairies (matured for 4 months)

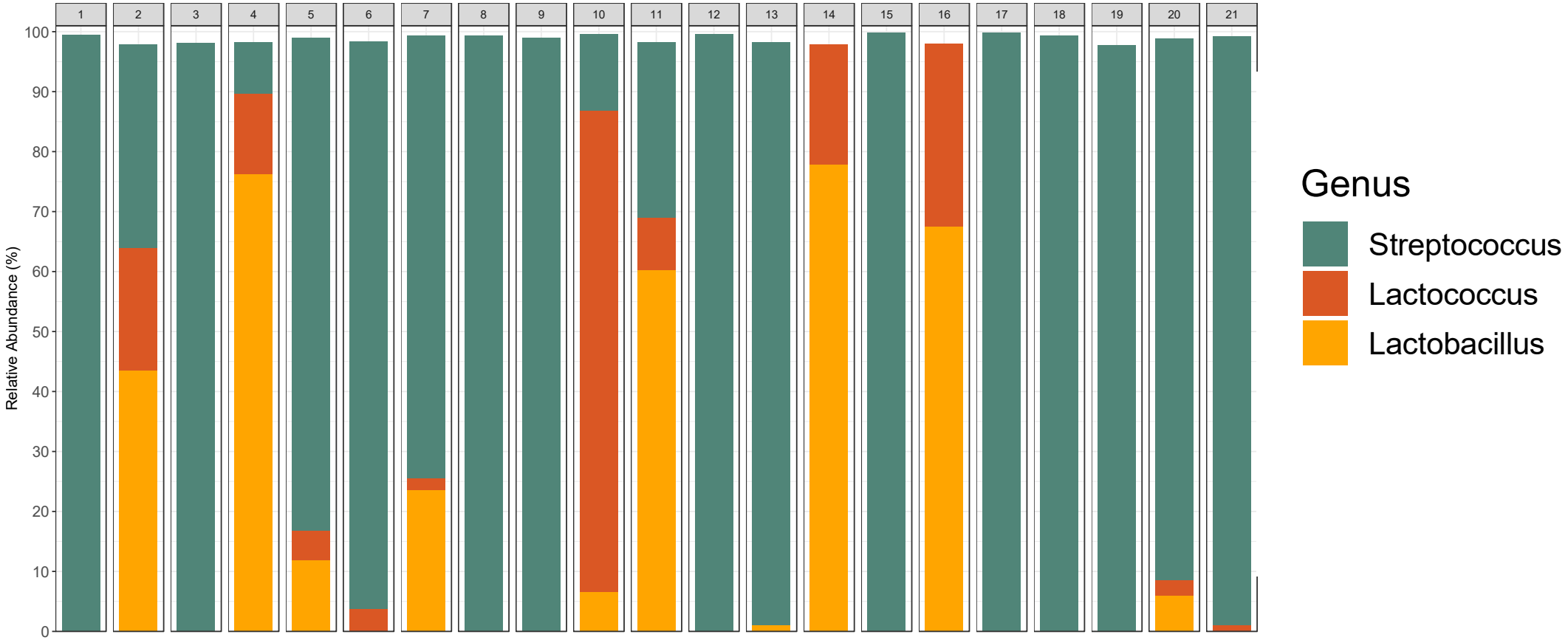
Bacterial community in Raclette cheese (DNA)



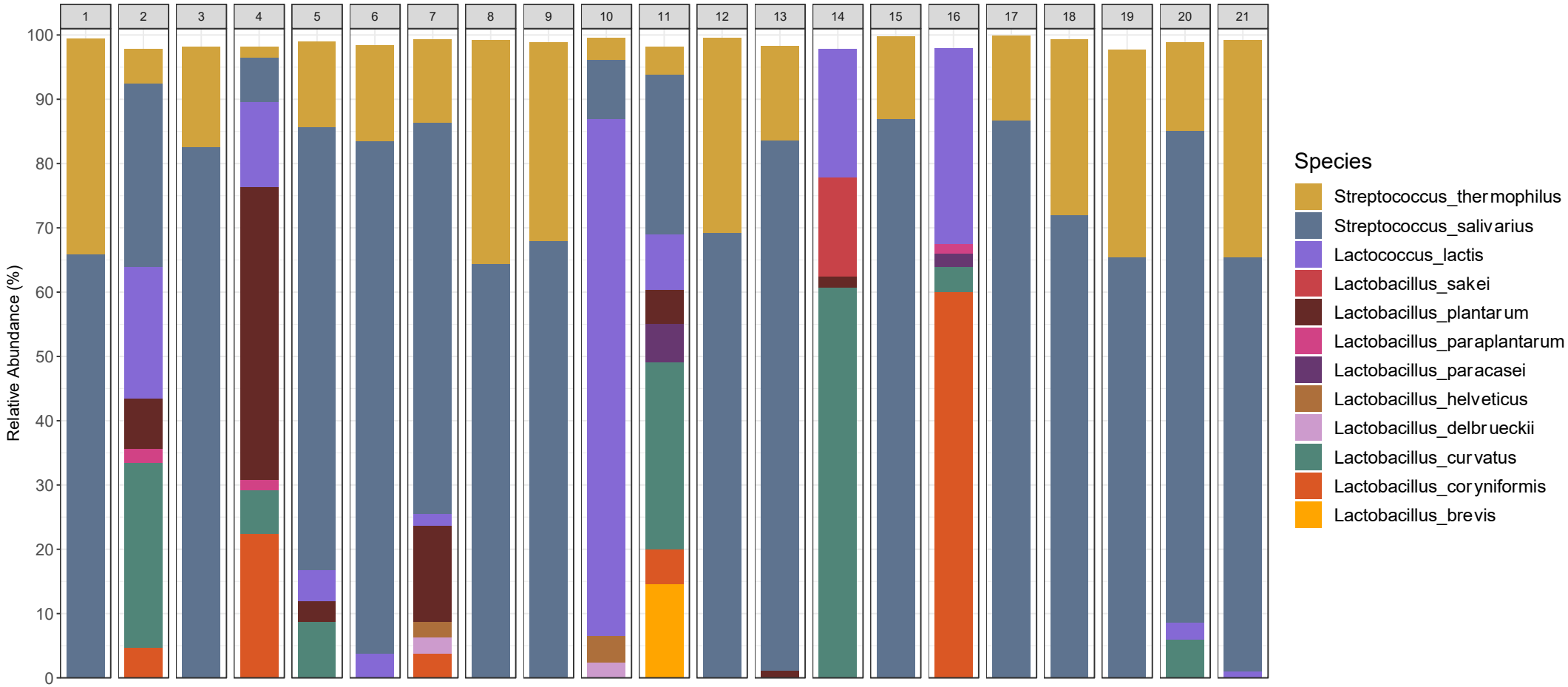
Bacterial community in Raclette cheese (DNA)



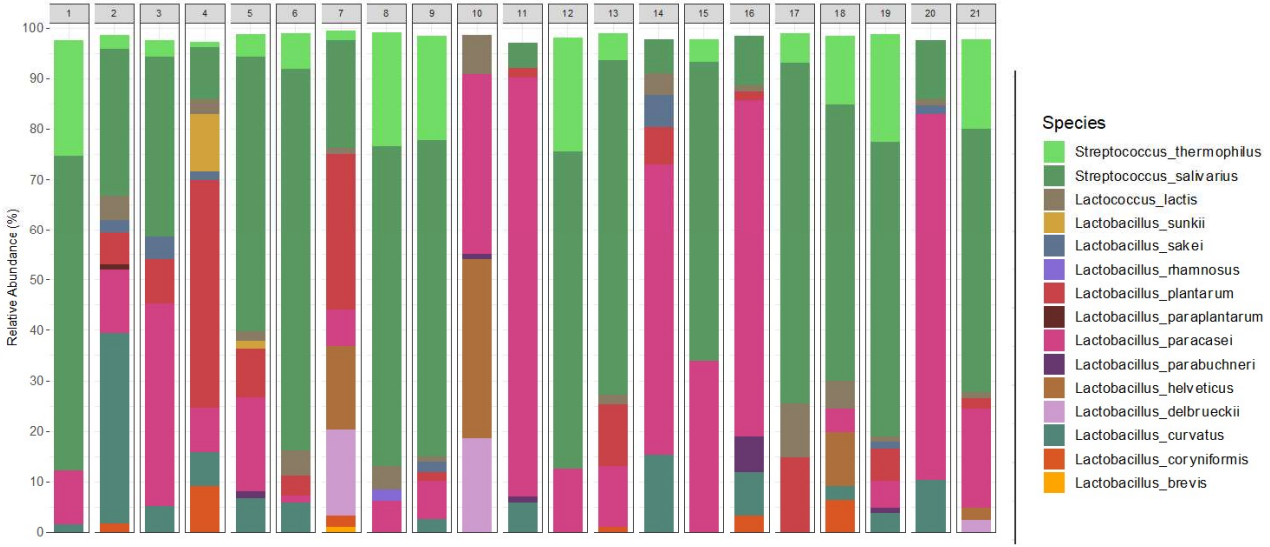
Bacterial community in Raclette cheese (RNA)



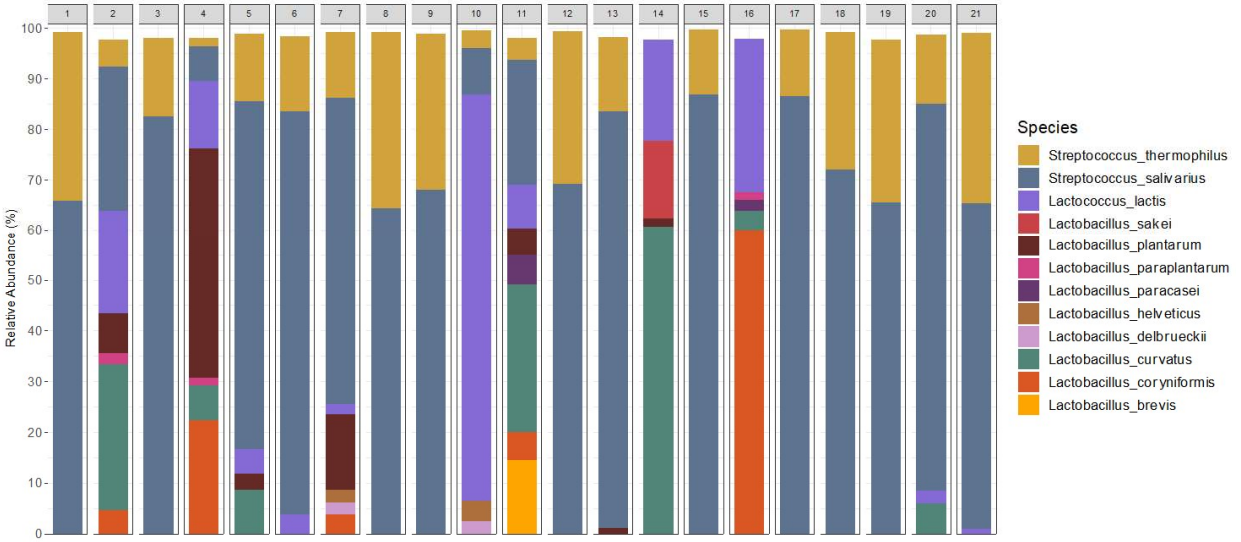
Bacterial community in Raclette cheese (RNA)



Bacterial community in raclette cheese (DNA)



Bacterial community in raclette cheese (RNA)



Observations

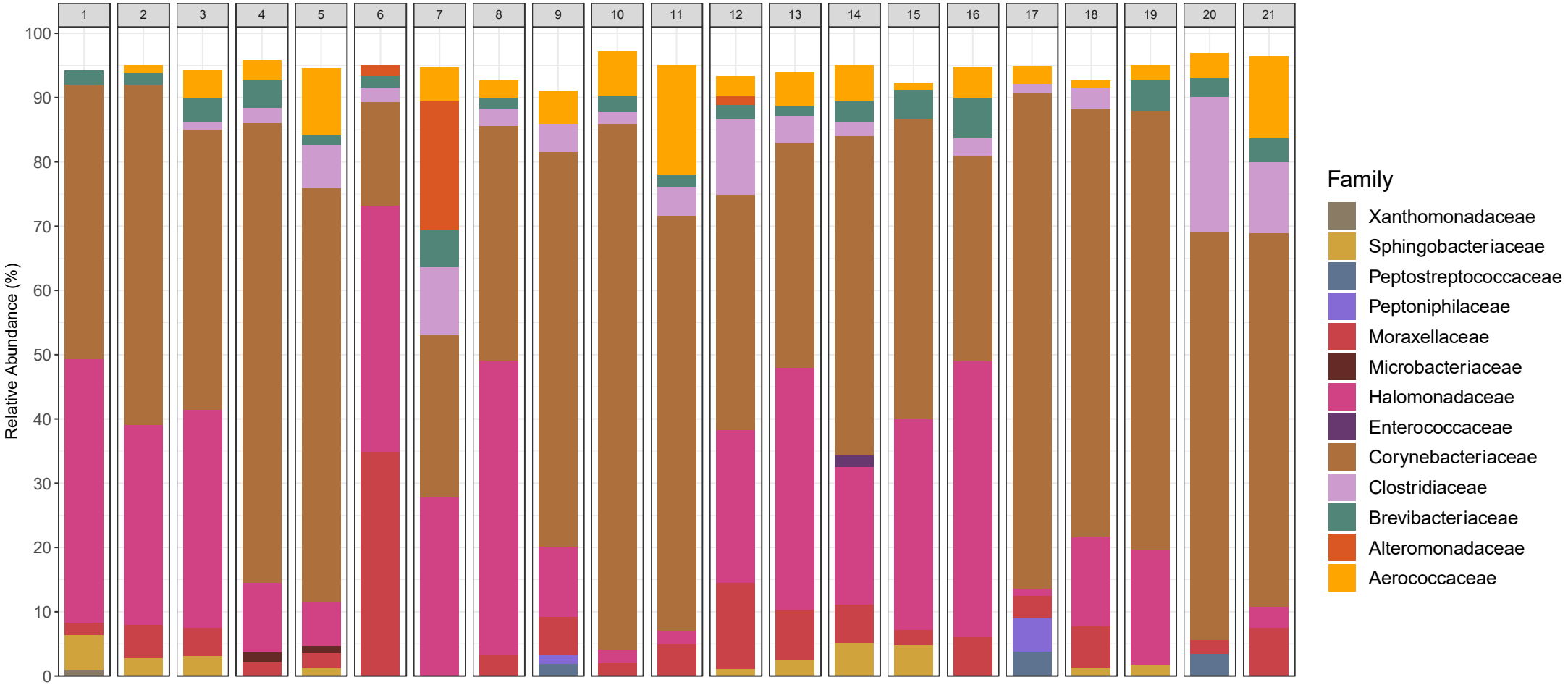
- clear differences between the cheeses
- *Streptococcus thermophilus* dominated in 15 (DNA) and 12 (RNA) cheeses, respectively
- Large diversity of lactobacilli (DNA), but partly little activity (RNA)
- 6 Cheeses with a high activity of lactobacilli (RNA)
- Non-lactic acid bacteria had only a very low abundance (DNA and RNA) in all cheeses

Relative abundance of different bacteria groups

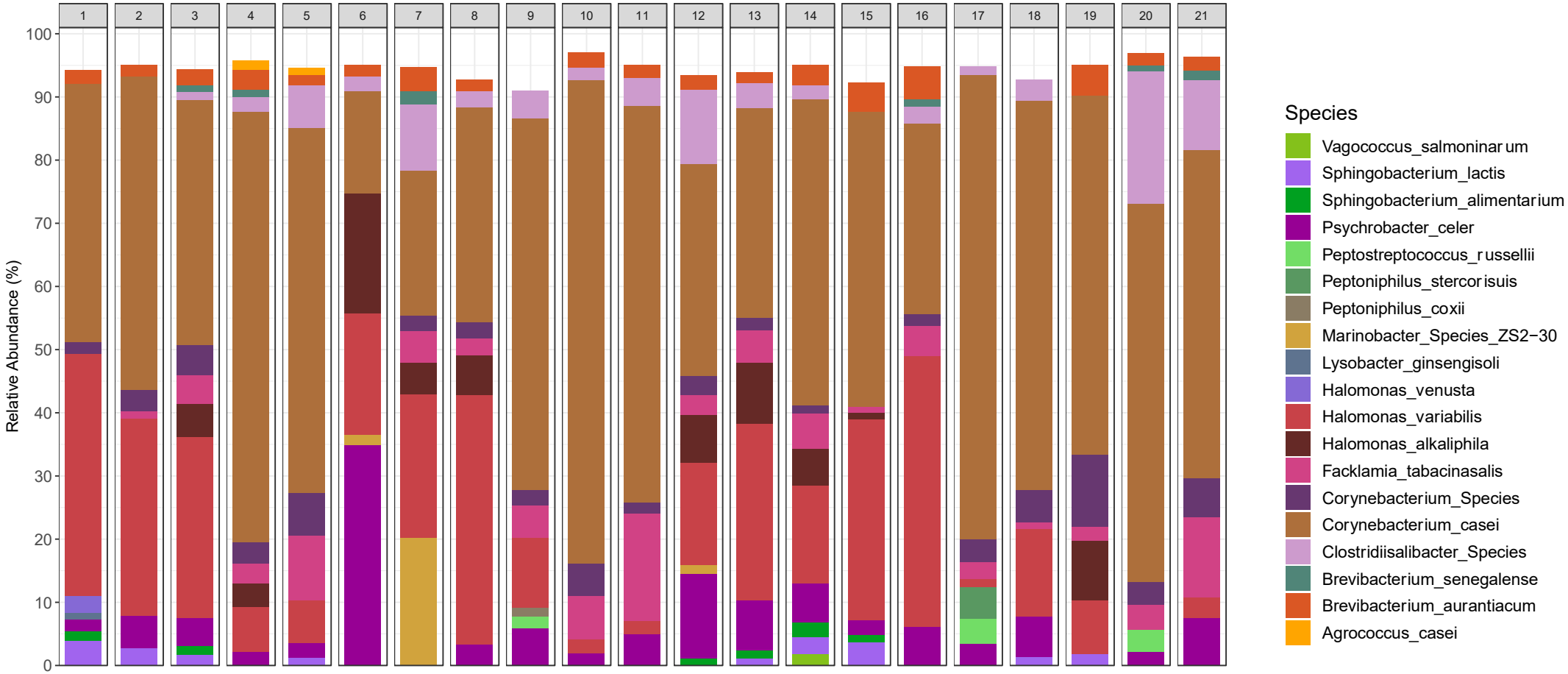
Germ group	Number of species	Relative abundance				
		Mean	s_x	Median	Min	Max
Starter Lactic acid bacteria	5	80.73	18.19	88.47	33.32	97.23
Lactobacillus spp. / pos. 1)	6	15.03	13.57	9.16	1.98	46.95
Lactobacillus spp. / neg. 2)	4	3.00	6.14	0.78	0.08	27.48
Lactobacillus spp. / ? 3)	16	1.27	4.14	0.09	<0.01	19.01
Enterococcus spp.	4	0.03	0.04	0.01	<0.01	0.17
Staphylococcus spp.	3	0.02	0.01	0.02	<0.01	0.04
Clostridiaceae spp.	1	<0.01	<0.01	<0.01	<0.01	<0.01
Propionibacteriaceae spp.		n.d.	n.d.	n.d.	n.d.	n.d.
Enterobacteriaceae spp.		n.d.	n.d.	n.d.	n.d.	n.d.

Effects on cheese quality: 1) positive / 2) negative, e.g. due to the formation of biogenic amines / 3) unknown
n.d. = not detectable

Bacterial community on the rind of Raclette cheese (DNA)



Bacterial community on the rind of Raclette cheese (DNA)



Discover unknown causes of known cheese defects: Too high content of histamine

- Cheese defect:
  - Trigeminal perception of "burning" and "pungent" during tasting
  - Undesirable formation of eyes, splits or cracks
  - Increased levels of histamine, which can lead to allergy-like symptoms in sensitive people, such as abdominal cramps, diarrhoea, flatulence, feeling of fever, reddening of the skin, rashes, itching, nausea or even vomiting

- Cause:
  - Continuous degradation of the amino acid histidine during ripening (splitting off the acid group)
- New finding from microbiome analysis:
  - *Lentilactobacillus parabuchneri* is responsible for the formation of histamine in cheese

Recommendations in case of too high histamine content

- Persistent contamination in milking systems is the main cause
  - Check raw milk samples from each dairy farmer for the presence of histamine-forming bacteria (Simple practice method available)
    - Check cleaning parameters: too low cleaning temperatures? Too short cleaning period? Too low detergent dosage or use of unsuitable cleaning agents?
    - Processing environment control at the dairy farm concerned to identify the source: Milking unit? Milk line? Milk tank?
    - Careful cleaning or, if necessary, replacement (e.g. of seals)
  - Contaminated milk should be excluded from processing into raw milk products.

Deposits in the collection piece of the milking unit

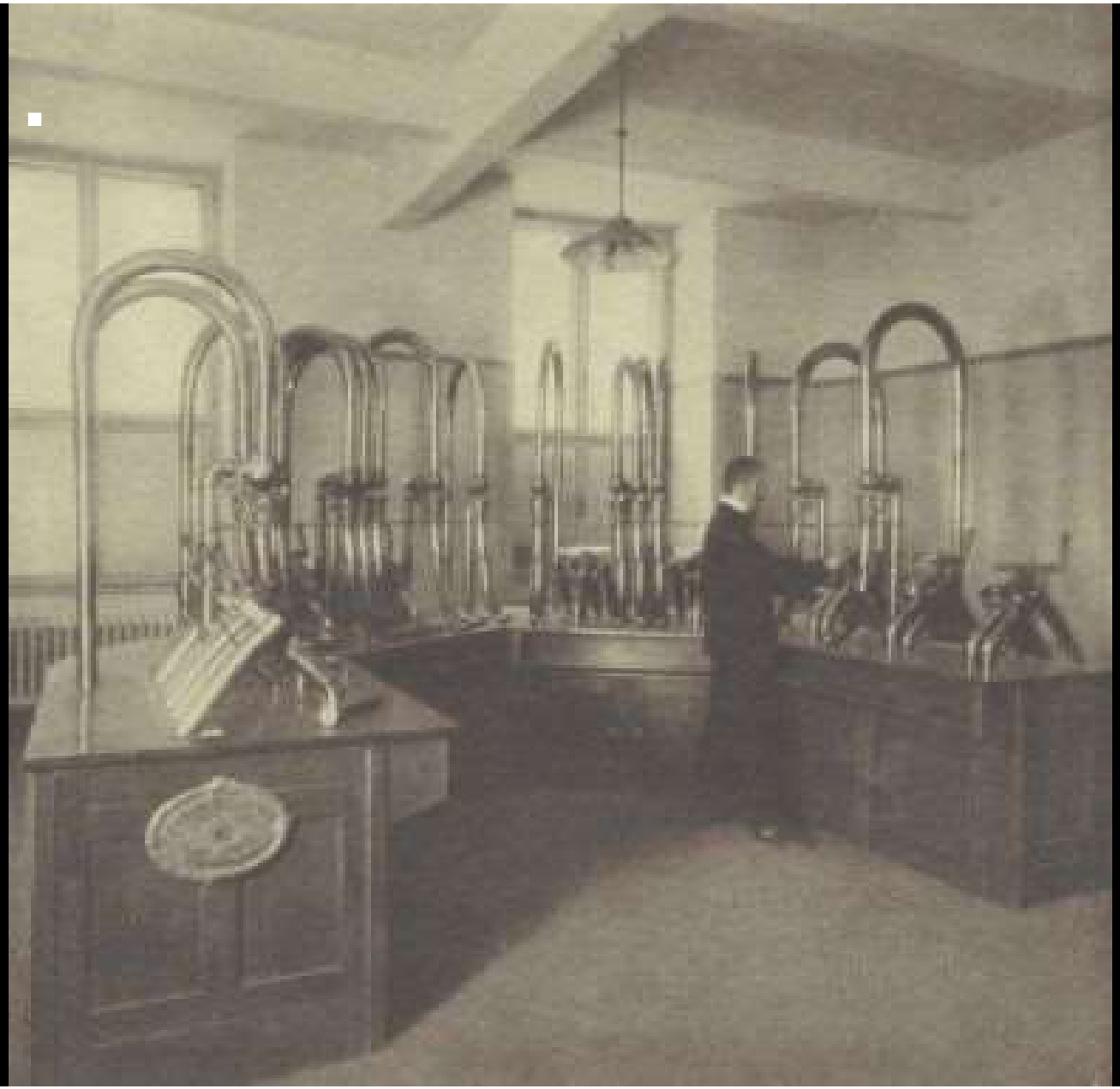
Discover unknown causes of known cheese defects: Atypical butyric and propionic acid formation

- Cheese defect:
  - Increased levels of propionic and/or butyric acid in the cheese without the typical gas formation.
  - Flavour defects: sweetish (propionic acid) or rancid, putrid (butyric acid)
- Cause:
  - Degradation of amino acids by the cheese surface microflora
  - Diffusion of these carboxylic acids from the cheese smear to the inside (Such migration is easily detectable by zonal gradients (rind, under rind, centre))
- New finding from microbiome analysis:
  - *Peptostreptococcus russellii* was present in the smear of cheeses with elevated levels of propionic and butyric acid, but not in the rind of good quality cheeses.

Recommendations in case of atypical butyric and propionic acid formation

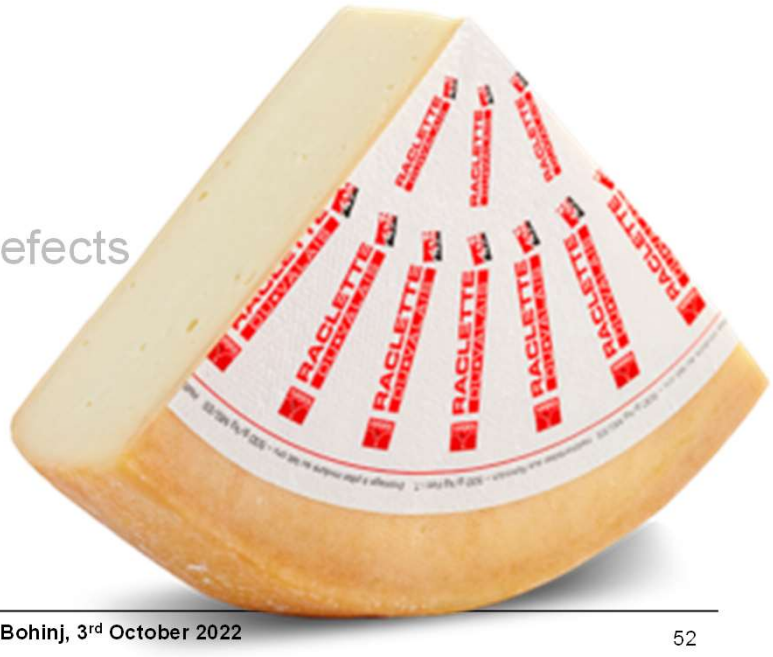
- Complete lactose degradation before brining
- Check salt content in the cheese (note zonal differences)
- Check ripening parameters:
  - Cheese smearing (frequency, intensity, salt content)
  - Temperature and humidity
  - Air circulation
  - Supply of fresh air
- Never stock smearing water for long, and NOT in anaerobic conditions
- Use of surface cultures or smearing water from a cheese dairy without this quality defect
- Sanitise wooden boards if necessary

1875...



Four aspects will be highlighted

1. Introducing the cheese variety Raclette du Valais AOP
   - History
   - Production areas (lowlands, mountains, alpine pastures)
   - Technology
2. New insights into the microbiota
   - Microbial biodiversity within the cheese and on its surface
   - Discover unknown causes of known cheese defects
3. Historical context: looking back on 150 years
4. Development of new AOP acidification cultures



Looking back on 150 years: How the story actually began

A well-kept treasure in Grimentz (Village in the Canton of Valais)

Collection of old cheese wheels (Family Zufferey)

2 cheese wheels from **1875**

Original idea: isolate ancient bacteria

Cheese wheels in their historical context

- Research questions:
  - Microbiological composition of cheese from the 19th century and its development
  - Influence of technological and scientific developments
  - Development of the most important bacteria in cheese over the course of almost 150 years
- Sterile sampling from cheese wheels from the years: 1875, 1944, 1957, 1975, 1985, 1991, 2010, 2017

First preliminary findings (publication in preparation)

- DNA well preserved: Oldest described cheese microbiome
- No living microorganisms could be isolated from the cheese from 1875
- A distinction can be made between 3 time periods:
    1. “Spontaneous" fermentation: high abundance of intestinal bacteria (lactobacilli)
    2. Dominance of mesophilic lactic acid bacteria
    3. In recent years, increasing importance of *Streptococcus thermophilus*

Four aspects will be highlighted

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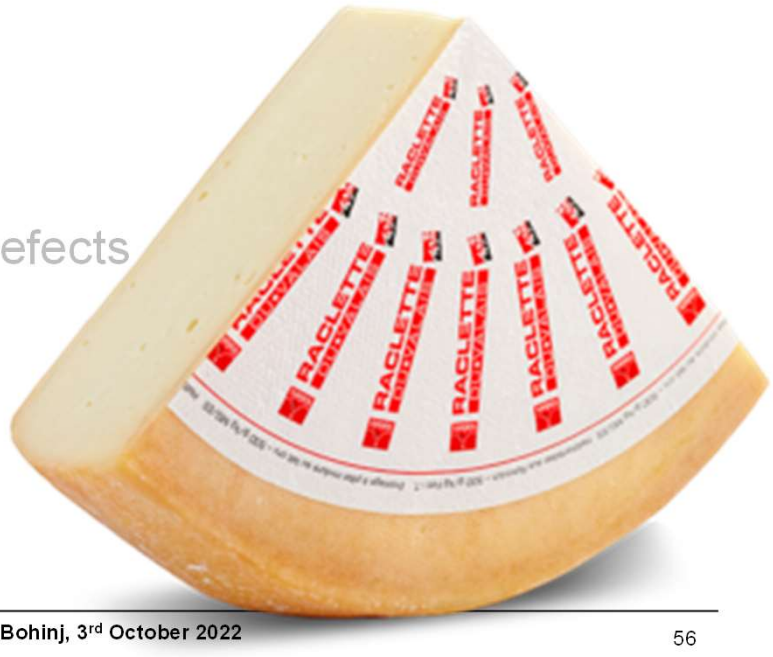


New AOP acidification cultures

- Goals: Broaden microbial diversity, increase authenticity and strengthen the connection to the terroir.
- Nowadays: Mostly commercial direct starter cultures from globally active suppliers.
- Future: exclusive AOP cultures only for cheese dairies producing Raclette du Valais AOP:
  - lyophilised
  - with a selection of mesophilic and thermophilic strains
  - 2 forms of supply with 2 cultures each (different phage spectrum)
    - 2 semi-direct cultures (mainly for year-round cheese dairies)
      - The cheesemaker can use it to produce specific liquid cultures
      - Preserve and maintain traditional knowledge of culture production
    - 2 direct cultures (mainly for alpine dairies)


D4
D5
D6
D7
D8
D9
D10
D11

Labour-intensive and time-consuming development by Agroscope

- Started in 2013
  - more than 500 strains
  - more than 100 experimental cultures
  - 20 trials in pilot plant and 6 trials in practical cheese dairies (lowlands and alpine pastures)
- Planned introduction:
  - semi-direct cultures: 2023
  - direct cultures: 2025

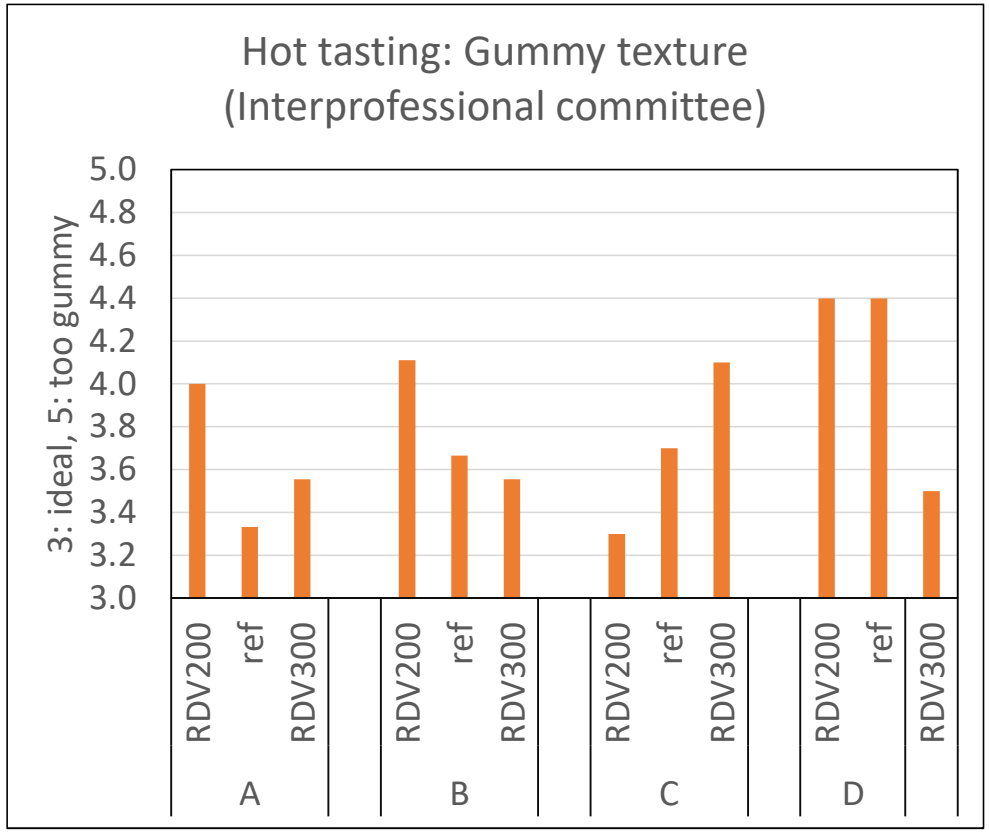
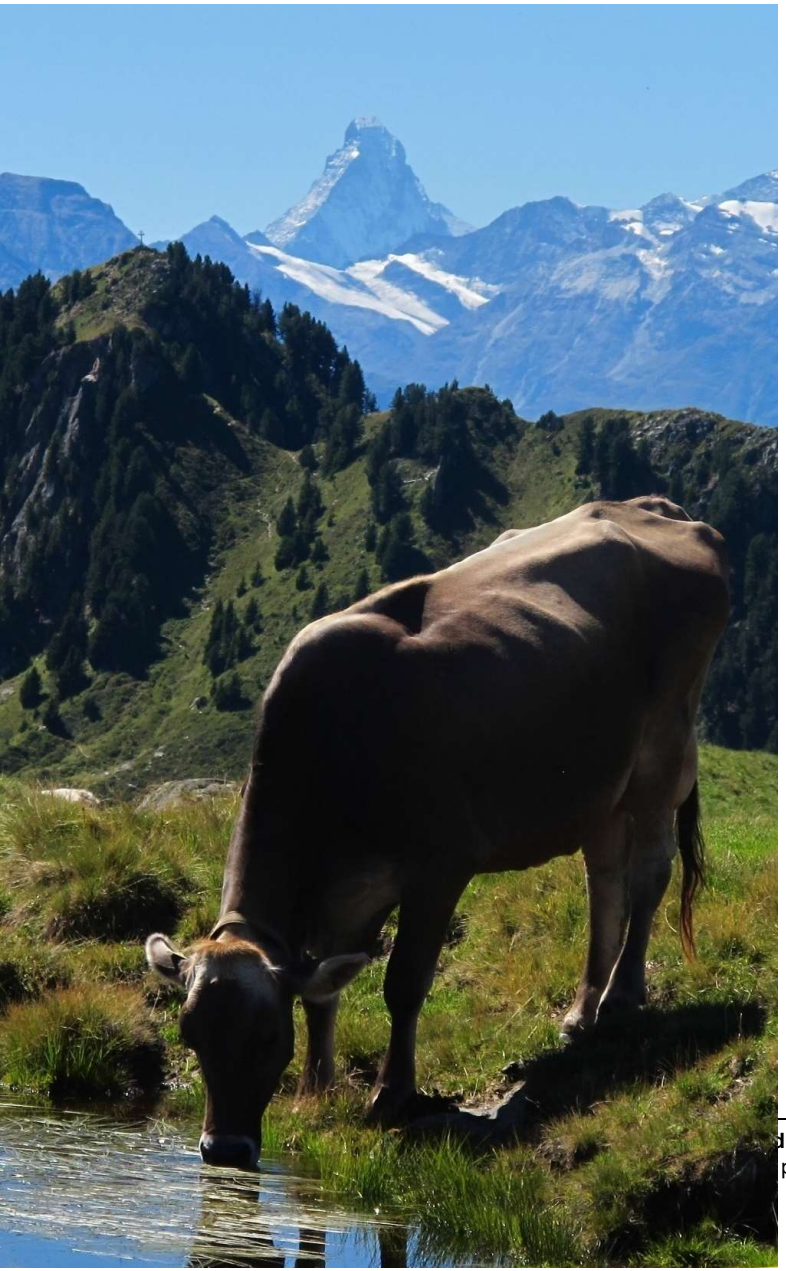


Figure 37: photos de coupe des fromages de Verbier (2: RDV200 ; 3: référence ; 4: RDV300) et Liddes (6: RDV200 ; 7: référence ; 8: RDV300)

Figure 38: photos de coupe des fromages d'Etiez (10: RDV200 ; 11: référence ; 12: RDV300) et Simplon (14: RDV200 ; 15: référence ; 16: RDV300)

Conclusions

1. The knowledge to produce safe and high quality raw milk cheese is available.
2. The microbial biodiversity inside the cheese is mainly determined by the lactic acid bacteria. They come from the raw milk, the cheese dairy and the starter cultures.
3. The ripening process contributes significantly to the diversity of the cheeses.
4. Modern molecular biological methods open up new possibilities for improving the quality and safety of cheeses.



Cheeses from the mountain region are distinguished by...

1. A greater influence of environmental factors, e.g. season, weather, feed quality.
2. Greater diversity in the microbiome
   - proximity to the cowshed
   - greater variety in starter cultures
   - less “hygienic design” e.g. more wood and less stainless steel
   - etc.
3. Greater influence of individual milkings.
4. Much less standardised manufacturing processes.
5. Not to be underestimated: happy cows and happy alpine cheesemakers

I hope to see you all again next year in Switzerland

- **13th FACE Conference**
  - from **11th - 13th October 2023**
  - in Grangeneuve-Fribourg
- Local Host: Swiss Centre of Excellence for Raw Milk Products
- 4 thematic sessions: Sustainability, Safety and quality, Health effects, Microbial biodiversity
- Scientific Posters: exhibition, short presentations, award

Let us work together to make it happen !

«The challenge for the cheesemaker is to strike the right **balance between art and science**.»

Thank you very much...

...to all the colleagues from Agroscope and from the Interprofession Raclette du Valais AOP for the interesting research results and the beautiful pictures,

...the organisers of the FACE Conference for the invitation, and

...to all of you for your attention.
















Thank you for your attention

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