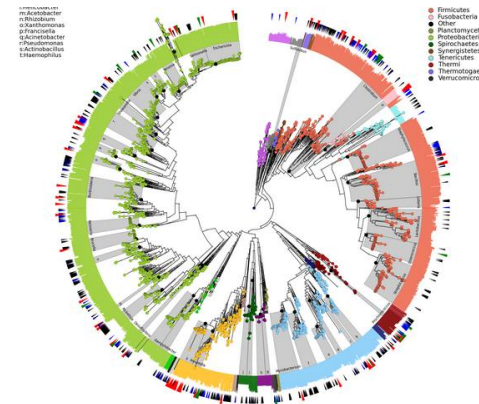




# The apple skin microbiome – implications for postharvest quality

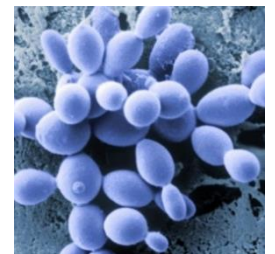


**Andreas Bühlmann**  
**Agroscope**



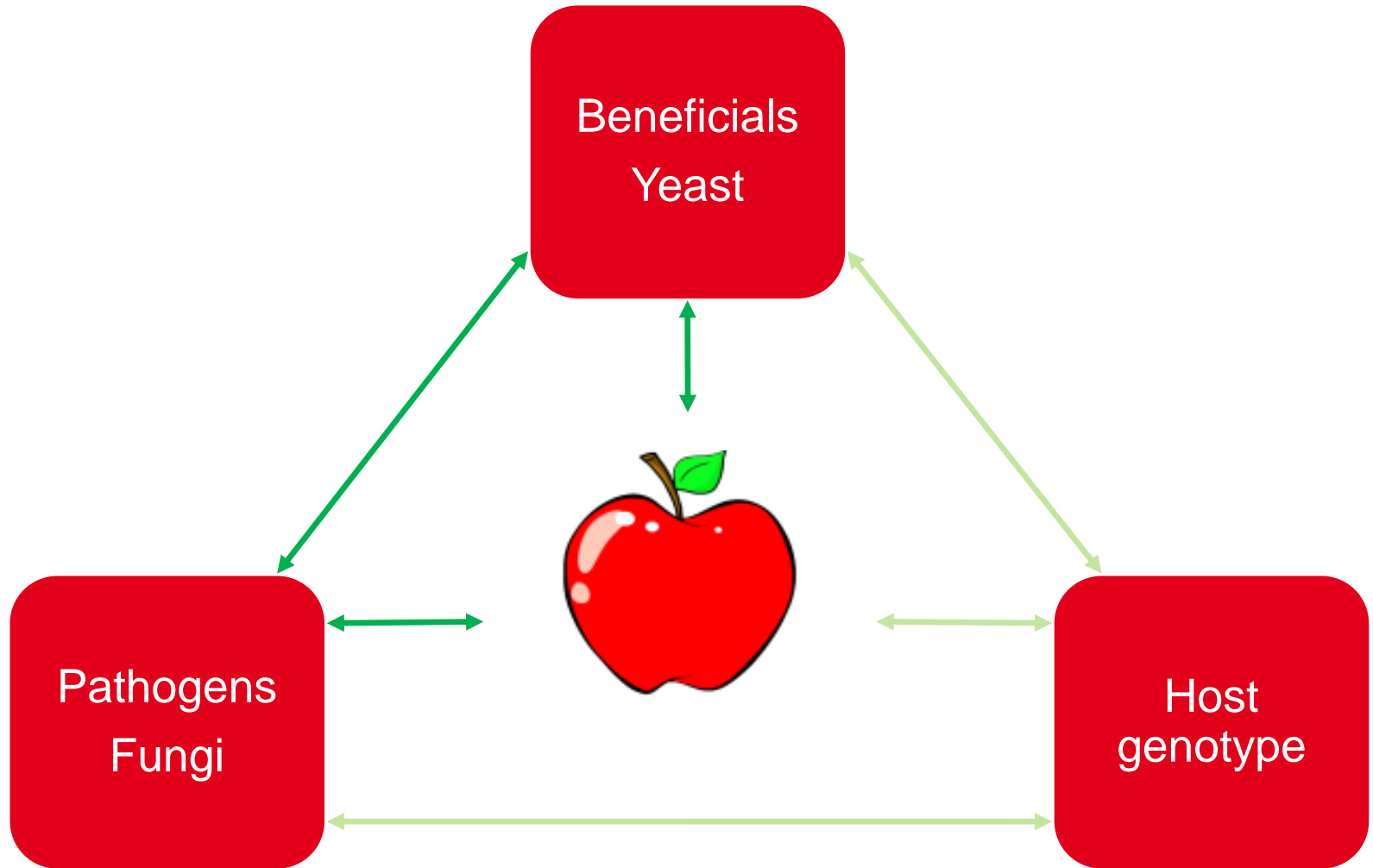
# Microbiome of an apple

- Pathogens are fungi, beneficials are yeast
- Pre- and post harvest differences
- New technologies allow to study communities
- Controlling the microbiome along the value chain
  - There are antagonists, but not widely used
    - formulation, timing
    - mixtures





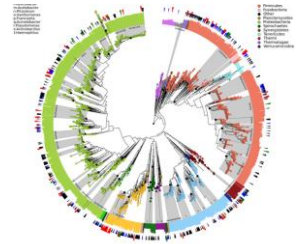
# Microbiome of an apple





# Set up and aim

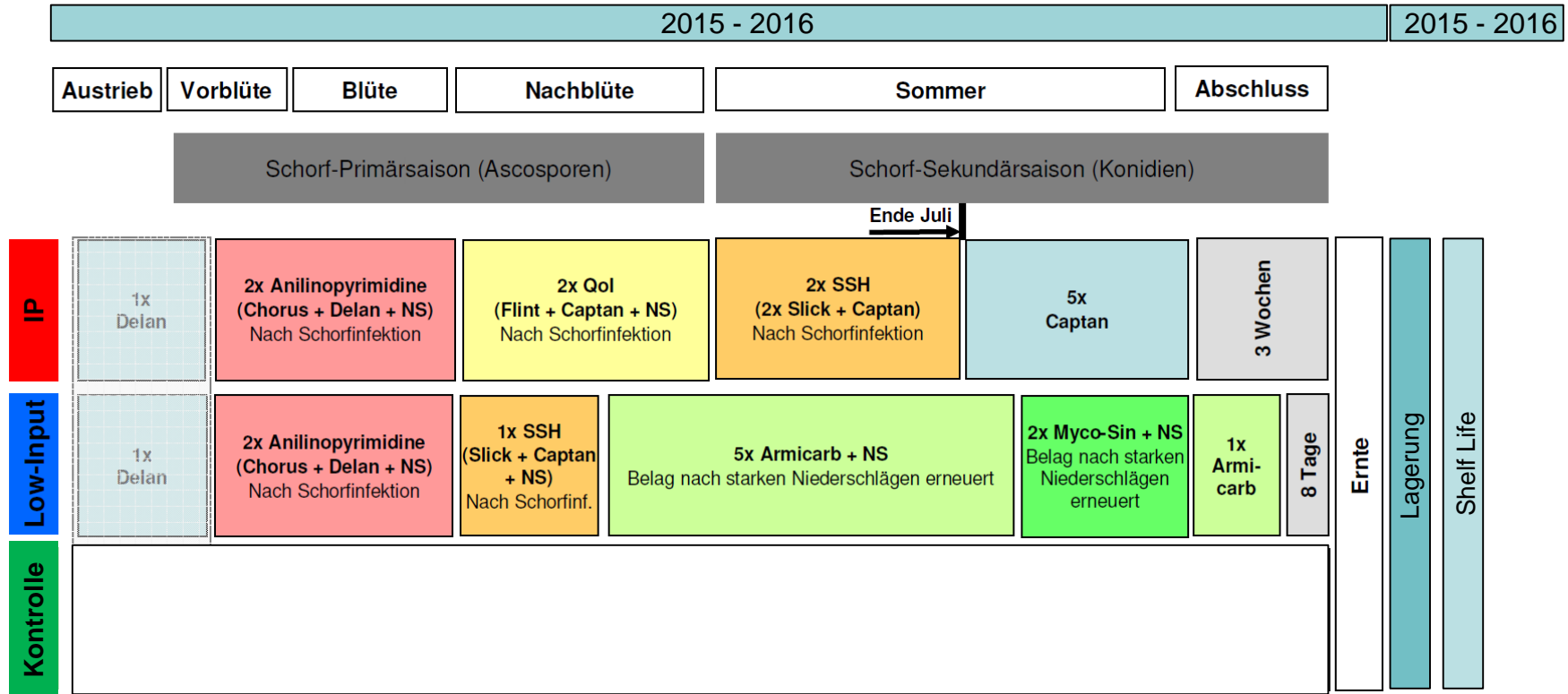
- Existing Low residue trial
- Characterize microbial load on apples from harvest to retail
- Can we predict pathogen damage before symptoms appear – focus on *Neofabraea*
- Can we identify ideal biocontrol mixtures





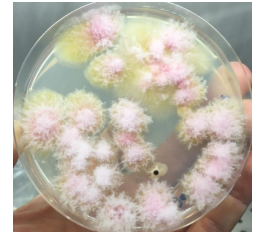
# Methods

## Low residue trial - orchard

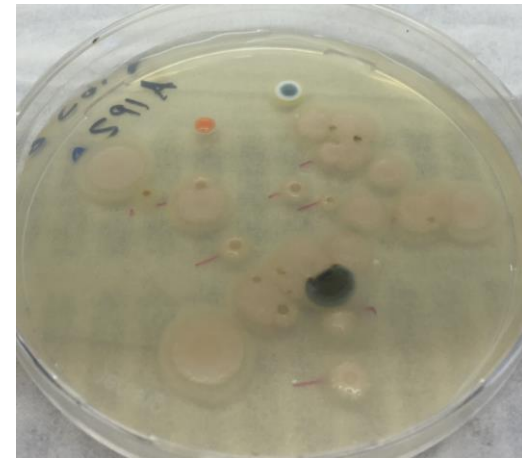




# Methods Microbiology



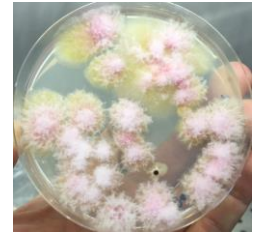
Varieties	Okt 15			Nov 15			Dez 15			Jan 16			Feb 16			Mrz 16			Apr 16			Mai 16			SL 7d		SL14d	
Ariane	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A
Otava	C	C	C	C	C	C	C	C	C	C	C	C	C	C	C	C	C	C	C	C	C	C	C	C	C	C	C	C
Topaz	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D



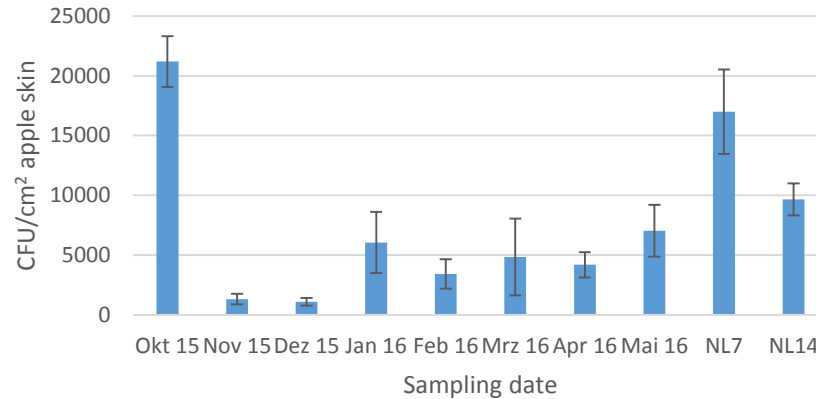
Counting colonies, species identification, sanger sequencing



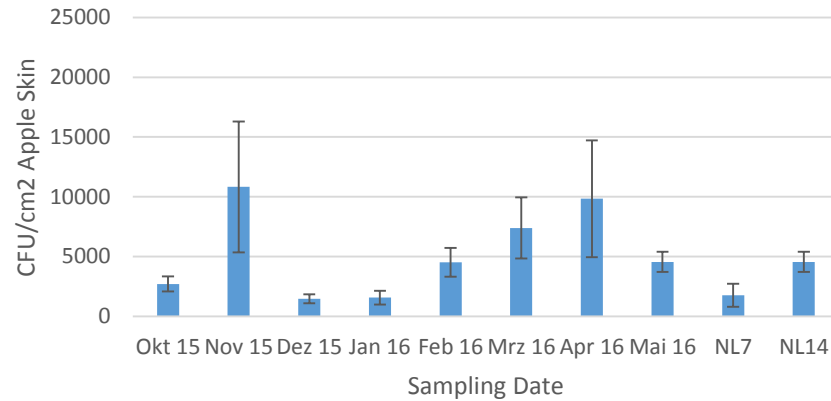
# Results Microbiology



### Microorganisms per month 2016



### Microorganisms per month 2017

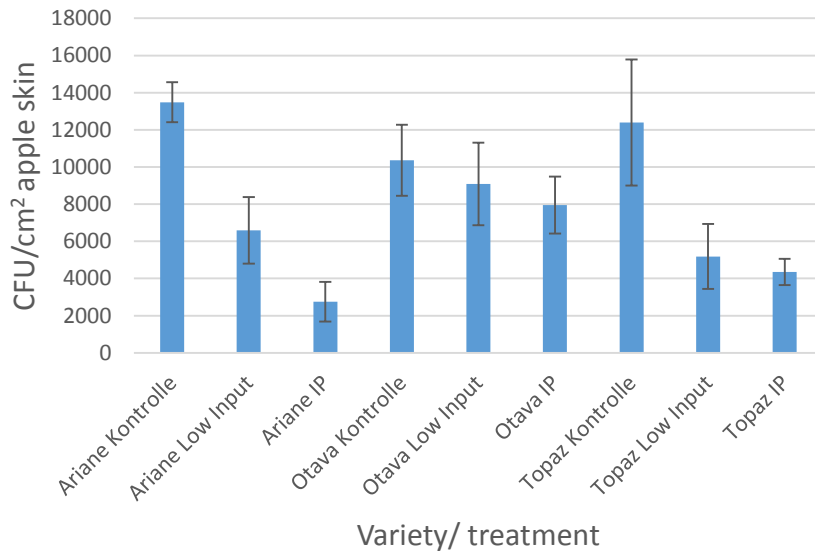




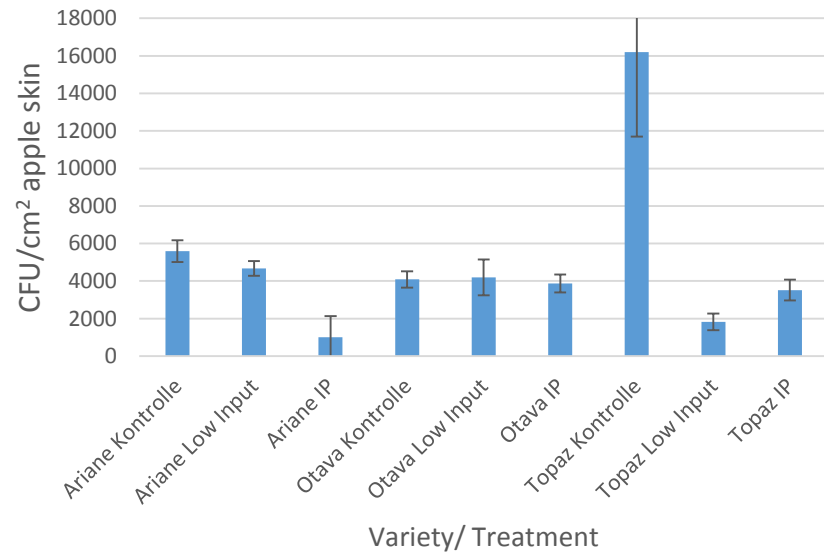
# Results Microbiology



### Microorganisms per variety/ treatment 2016



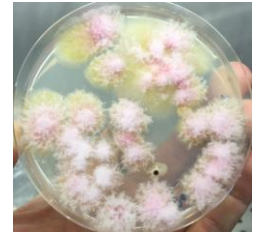
### Microorganisms per variety/ treatment 2017



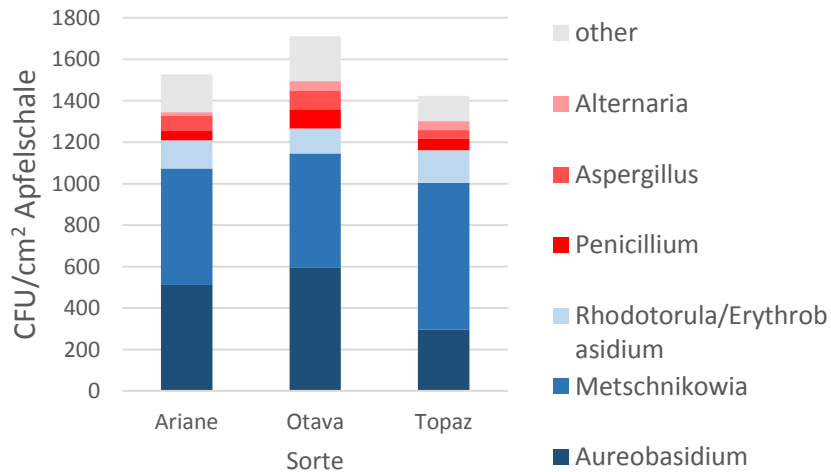




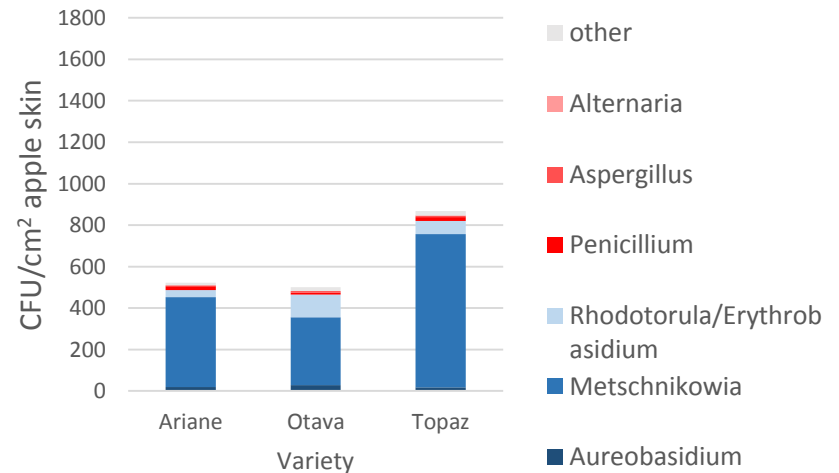
# Results Microbiology



## Species composition per variety 2016



## Species composition per variety 2017





# Methods Metagenomics



Agar  
plates

Varieties	Okt 15	Nov 15	Dez 15	Jan 16	Feb 16	Mrz 16	Apr 16	Mai 16	SL 7d	SL14d
Treatment	C LR IP	C LR IP	C LR IP	C LR IP	C LR IP	C LR IP	C LR IP	C LR IP	C LR IP	C LR IP
Ariane	A A A	A A A	A A A	A A A	A A A	A A A	A A A	A A A	A A A	A A A
Otava	B B B	B B B	B B B	B B B	B B B	B B B	B B B	B B B	B B B	B B B
Topaz	C C C	C C C	C C C	C C C	C C C	C C C	C C C	C C C	C C C	C C C

Metagenomics  
Bacteria/fungi

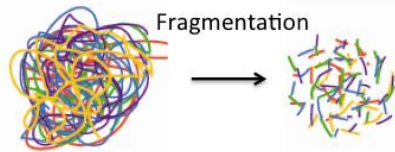
Varieties	Okt 15	Jan 16	Mai 16	SL14d
Treatment	C LR IP	C LR IP	C LR IP	C LR IP
Ariane	A A A	A A A	A A A	A A A
Otava	B B B	B B B	B B B	B B B
Topaz	D D D	D D D	D D D	D D D



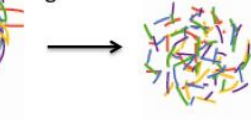
wash



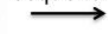
DNA  
extraction



Fragmentation



DNA  
sequencing



```
>seq1
GCCGTAGTCC...
>seq2
TATGCCGGTA...
>seq3
...
```

Database  
comparison

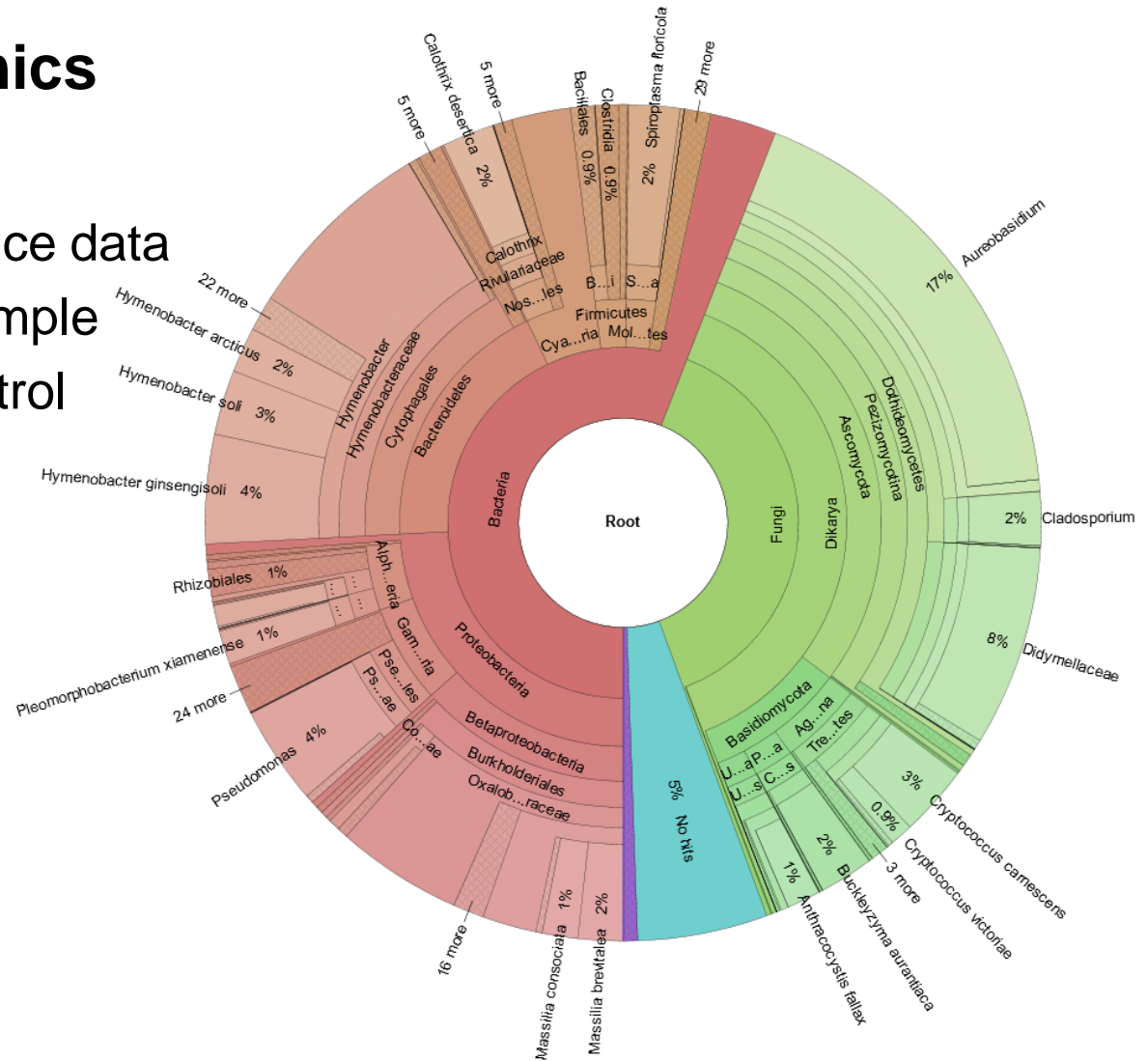


```
>seq1
Aureobasidium
>seq2
Penicillium
>seq3
....
```



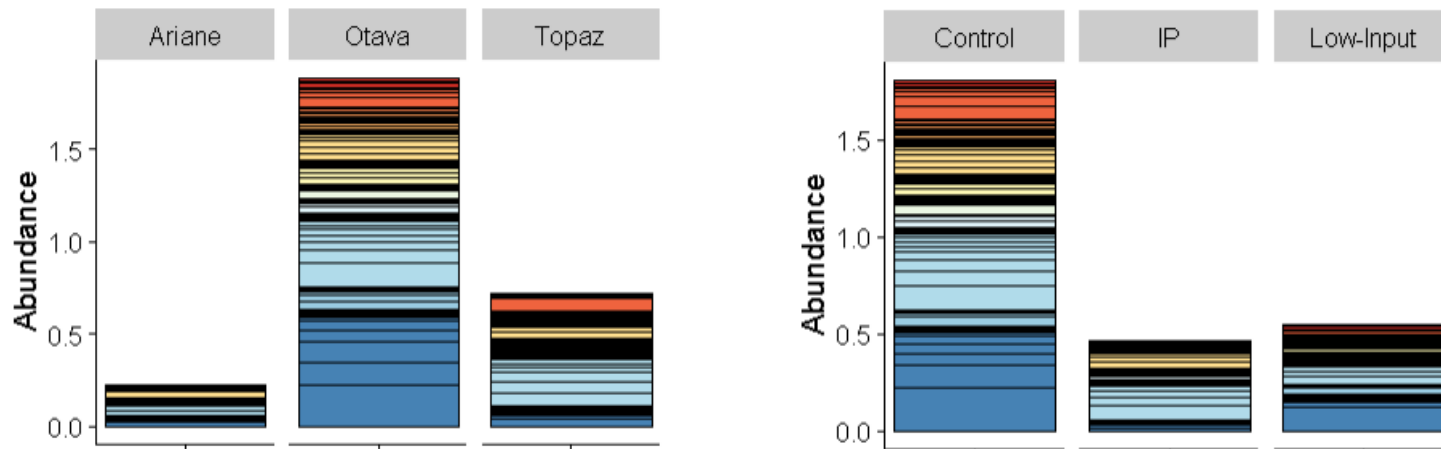
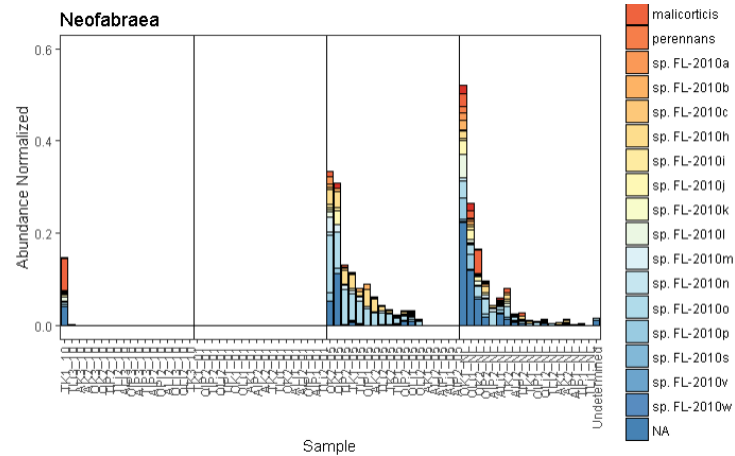
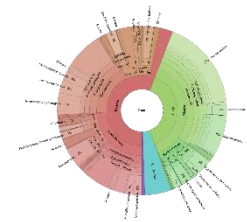
# Results Metagenomics

- ~ 36Gbp sequence data
- ~1Mio reads/ sample
- Bsp. Ariane Control



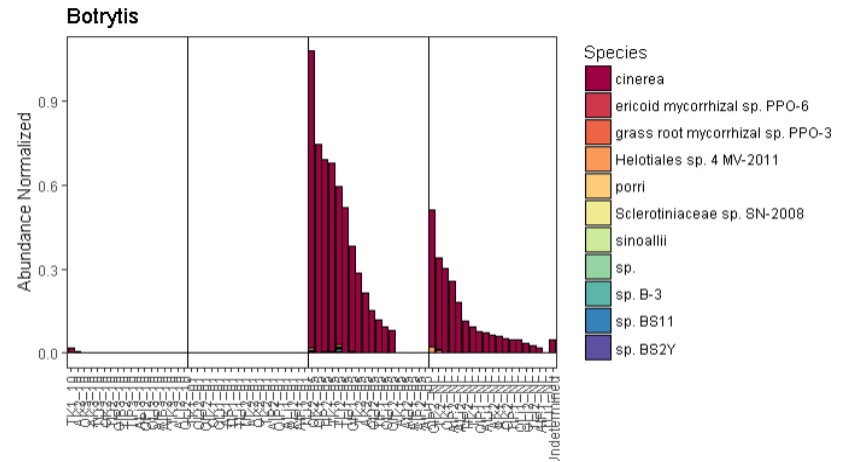
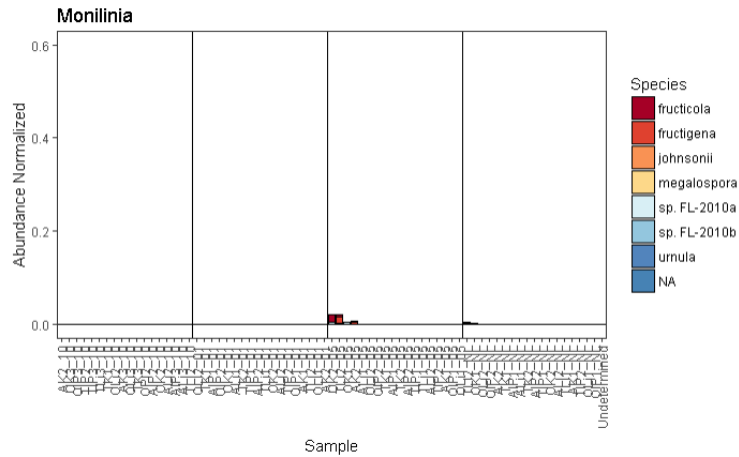
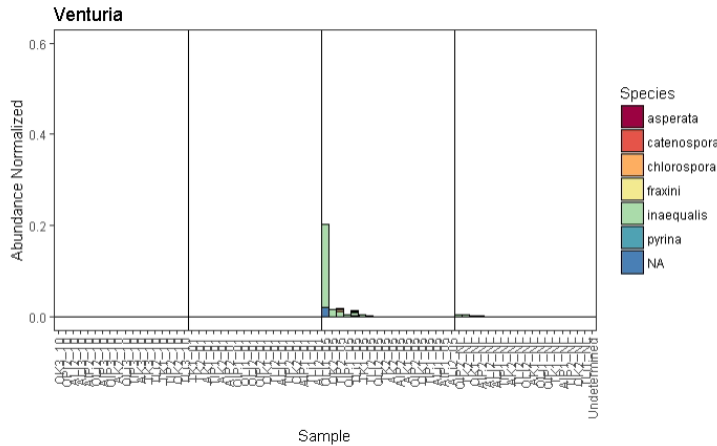


# Results Metagenomics





# Results Metagenomics



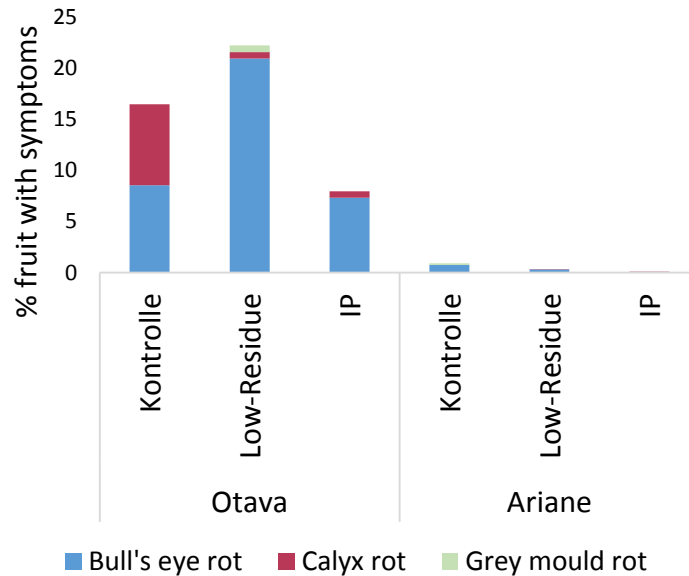


# Results Metagenomics

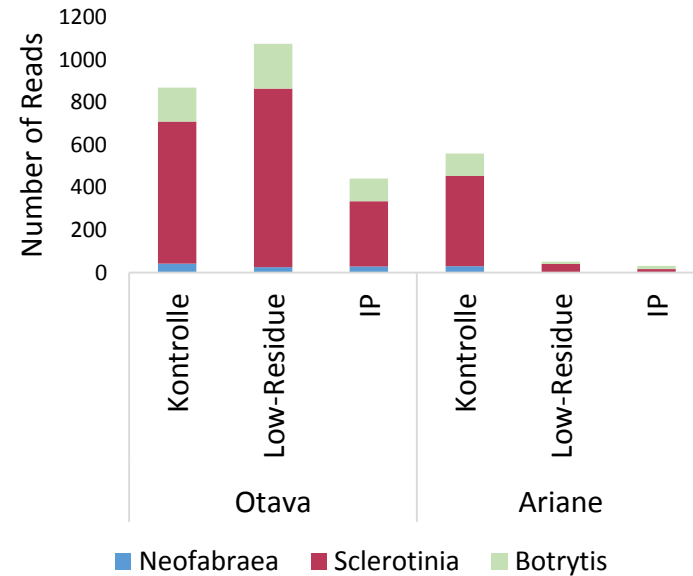


- Correlate metagenomics and disease scoring

Storage Diseases Visual  
Inspection



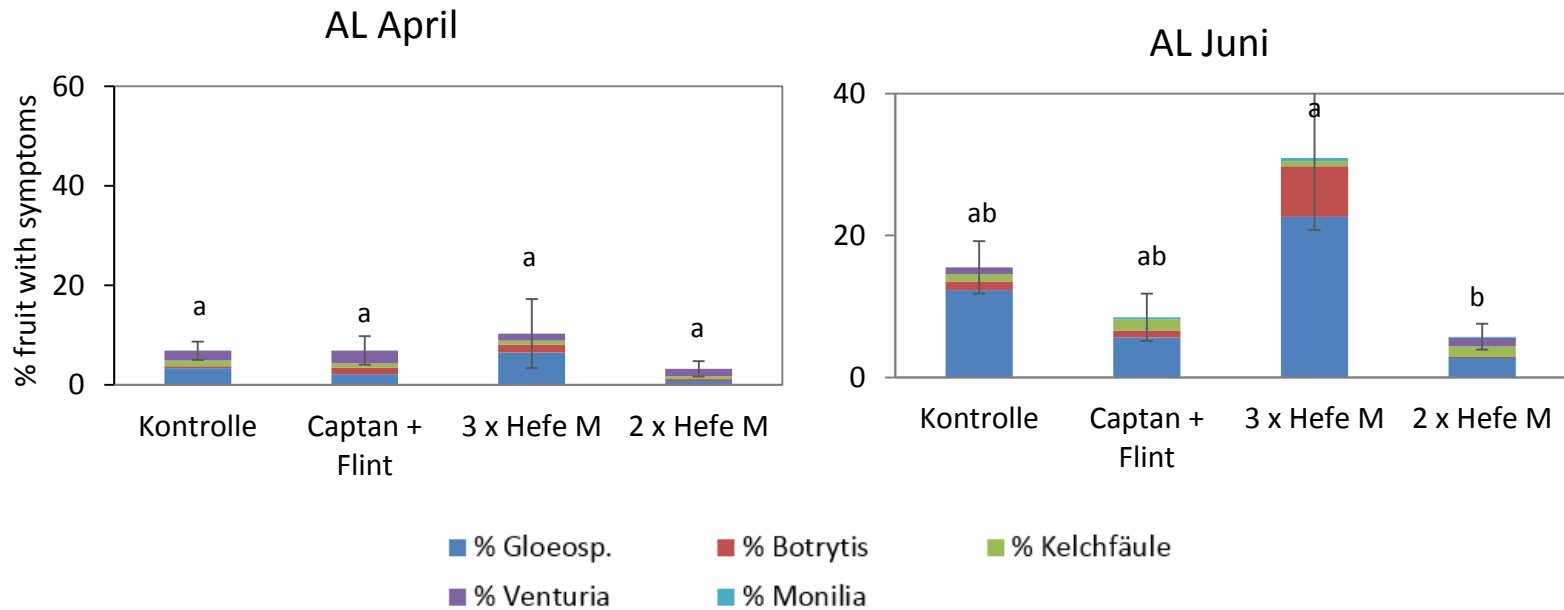
Storage Diseases  
Metagenomics





# Changing the microbiome

- Biocontrol trial with yeasts





# Conclusions/ Outlook

- Pathogens can be detected on symptomless apple
- Narrow down *Neofabrea* growth in storage January – May
- Apply metagenomics in orchard - identify time of infection
- Identify key species driving the composition of the apple skin microbiome in order to identify possible antagonists
- Apply antagonists in the field
- Reduce postharvest losses





# Thanks

- Agroscope
  - Gruppe Petignat
  - Frey/Ahrens – Molecular diagnostics Genomics Bioinformatics
  - Freimoser/Lutz – Phytopathology
  - Näf/Perren – Fruit extension
  - Gasser/Kellerhals – Apple breeding and genetic resources
  - Drissner/Gafner – Food microbiology
  - Christen/ Gabioud - Fruit crops in alpine regions
- External
  - EUFRUIT/ EU Post- harvest people
  - Fenaco
  - EMPA/ Thijs Defraeye

